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Nuclear fission safety programme 1990-94

Radiation protection research action 1992-94

Technical description of scientific projects

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Radiation protection research action 1992-94

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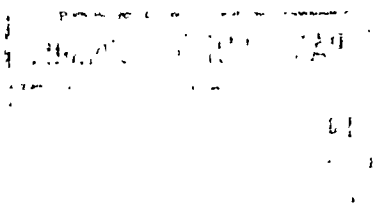

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PREFACE

This technical description of contracts carried out within the Radiation Protection Research Action of the Energy Directorate is designed to disseminate administrative information about the research groups and to identify clearly the scientific basis and scope of the contracts. The information provided is two-fold: first, all administrative data such as names and addresses of contractors as well as the duration and budget of the contract; second, the scientific background and expected developments as well as their possible application in this specific area are briefly set out. This broad multidisciplinary research activity has been divided into three main but interactive themes for management reasons and for the benefit of the reader, namely:

A. Human exposure to radiation and radioactivity;
Measurement of radiation dose and radioecology.

B. Consequences of radiation exposure to man;
Stochastic and non-stochastic effects and effects on the developing organism.

C. Risk and management of radiation exposure;
Assessment of risk and optimization of radiation protection.

Under these very general headings are grouped the following: the management policies for the optimisation of radiological work; exposure of patients and members of the medical profession during diagnostic radiology; behaviour of radionuclides in the environment; the study of the mechanisms of radiation carcinogenesis; the problems of natural radioactivity (including that which is concentrated as a result of human activities) as well as the means of achieving optimal management and communication about the risks associated with radiation and radioactivity.

Descriptions are also provided of the experimental collaborative projects and the joint study projects which have been established under an Agreement between the Commission and the three republics Belarus, Ukraine and the Russian Federation to carry out cooperative work on the consequences of the Chernobyl reactor accident.

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Radiation Protection Research Action

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PART I

Radiation Protection Research Action 1992 - 1994

A) HUMAN EXPOSURE TO RADIATION AND RADIOACTIVITY

A1 Measurement of Radiation Dose and its Interpretation

A1 Measurement of Radiation Dose and its Interpretation

Contract FI3P-CT920001 Collaboration on radiation dosimetry for radiation protection applications (EURADOS).

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Total Contribution by the Commission: 120 KECU
21 months 1/09/92 to 31/05/94

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120 KECU

Description of research work

The European Radiation Dosimetry Group (EURADOS) is a scientific society founded in 1981 to stimulate and improve cooperation of research in radiation dosimetry and related topics within Europe. It now includes about 180 scientists from more than 60 laboratories.

The main objectives of EURADOS within the contract are:

1. The stimulation of collaboration in research and technical developments concerning the measurement and evaluation of exposures by ionizing radiation.

2. The harmonization of methods for assessing radiation exposures by means of intercomparisons, workshops and active collaboration between laboratories in Europe.

3. The collection and evaluation of physical data relevant to radiation dosimetry and to the assessment of occupational, accidental and environmental exposures.

4. The support of transfer of expertise in radiation measurement and dosimetry from well experienced laboratories to other sites and young scientists in Europe by organizing seminars and tutorials. The dissemination of information regarding radiation dosimetry and EURADOS, EULEP and EUROMET activities is also an important task.

EURADOS is operated by scientists who are experts in the field of radiation research working with a minimum of bureaucracy and administration which results in an effective use of the available funding.

The work is mainly carried out by EURADOS Working Groups (WG), each of which consists of about 10 to 15 scientists from different laboratories of various European countries. Generally a Working Group will be set up to meet defined objectives and once these are achieved the Group will be disbanded.

The formation and scope of work is agreed by the EURADOS Council. EURADOS is able to react to further needs and new developments very quickly by setting up new Working Groups when appropriate. Seven Working Groups dealing with various topics are running at this moment.

The Working Groups will deal with the following tasks:

WG 2: Skin Dosimetry

As a result of the work during the previous contract period 1990/91 the Working Group organized a Skin Dosimetry Workshop (Dublin, May 1991). Following this workshop new objectives have been proposed and the group will now concentrate their efforts on the following topics:

1. HotParticles

The measurement and dosimetry of hot particles will be investigated in more detail. Collaboration in theoretical and experimental studies will be initiated in order to estimate the absorbed dose in the skin in the vincinity of those particles.

2. ß-Dosimetry

The dosimetry of low-energy ß-rays from a Pm-147 source has been studied by intercomparison measurements and it is aimed to produce a detailed report describing the measurement procedures, the results of the intercomparison and the implication for ß-dosimetry protocols in 1992/1993.

3. Collaboration with EULEP

Collaboration with EULEP on studies of biological effectiveness of different radiations and depth of sensitive layers in the skin will be continued during the programme period. In accidental dosimetry with very high doses of low-penetrating radiation the doses at depths of 2 to 3 mm become important. Consideration will be given to the need for additional physical measurements in individual dosimetry to assess doses at this depth.

WG 4: Numerical Dosimetry

In radiation dosimetry computational work becomes more and more important. Measurements of doses are often very difficult in phantoms and mostly impossible in human bodies. Hence dose distributions or conversion coefficients fixing the relation between radiation field and dose quantities are often determined by calculations using the Monte Carlo technique. Another important application of numerical calculations is in the development of new radiation detectors and dosemeters which can be more effectively designed, if their response and other properties are simulated by theoretical calculations. The intention of the Working Group is to evaluate and test computer codes developed for these purposes and to coordinate the application of those programs.

The special tasks for the contract period are:

1. Collection and distribution of information on the use of computer programs for dosimetric problems. This will be achieved by the evaluation of a questionnaire already sent to laboratories in Europe. A report will be presented in 1993.

2. Test of the reliability of results obtained by different codes. The estimation of a total uncertainty of results from a Monte Carlo calculation is a difficult task. On the basis of the questionaire the Working Group intends to organize a bench mark study and to perform intercomparisons with the aim of reducing the variance of the results obtained by different codes.

3. Collaboration in the study and intercomparison of electron transport codes. Such codes are becoming more generally available and because of the increasing power of personal computers they are more widely used by non-specialists who are not aware of the approximations included in those codes. The Working Group will study this situation and give recommendations.

4. The revision of ICRP Publication 51 which is under way by an ICRU/ICRP task group, will be supported by the EURADOS Working Group. The group will coordinate new calculations of conversion coefficients and help in evaluating new data sets especially for neutrons and electrons. This work is planned for the years 1992/93.

WG 6: Assessment of Internal Dose

The determination of doses after an intake of radioactive material is an important task in radiation dosimetry. The Working Group continues to improve collaboration in internal dosimetry within Europe with the following objectives for 1992-94.

1. The Working Group will support the work on stable isotopes (Sr and Te have been chosen for first studies) for use in metabolic studies.

2. The intercomparison studies of dose assessments following the intake of radioactive material will be continued in order to reduce the variation between results from different laboratories in Europe obtained during former intercomparisons.

3. The Working Group will support the collaboration in setting up European Registries for Internal Dosimetry which include assessment data, autopsy data and models.

4. The cooperation with EULEP in the Human Lung Task Group will be continued. The implications of the proposed ICRP lung model for dose assessments will be investigated and the sensitivity of predicted doses to variations in model parameters will be assessed.

5. A proposal has been made to use a person accidently contaminated with 60Co to visit European laboratories for body monitoring and excretion analysis. The Working Group has offered to contribute to this task.

WG 7: Radiation Spectrometry at Working Environments

Sufficient accuracy in the determination of dose equivalent in mixed neutron-photon fields can be achieved, if instruments with spectrometric properties are used. Different systems have now been developed in Europe and the Working Group will

- discuss the properties of various spectrometers,
- intercompare the different spectrometer systems by measurements in various neutron-photon radiation fields and
- investigate the use of spectrometric measurements in the interpretation of individual dosemeter readings for the assessment of organ and effective doses.

The following programme is planned for the contract period:

1. The study of spectrometers for area monitoring. This task will be performed in 1992/93.

2. The properties of portable neutron-photon spectrometers will be intercompared.

3. An intercomparison of spectrometers in realistic mixed radiation fields is planned for 1993/94.

4. It is aimed to prepare recommendations for the use of spectrometers in working environments.

WG 8: Development of Individual Dosemeters for External Penetrating Radiation

Individual monitoring in mixed neutron/photon fields becomes an increasing problem, when the new ICRP recommendations are introduced into practice, because the sensitivities of existing individual dosemeters are mostly not sufficient. The Working Group will deal with the following tasks:

1. Various groups in Europe are using track-etch detectors in neutron dose measurements. In this context they have developed many different procedures in the evaluation of irradiated polycarbonate foils. A detailed intercomparison of methods and materials will be organized in 1993/94 in order to find an optimal procedure and finally to formulate recommendations for future use.

2. With respect to sensitivity the development of new detectors, like e.g. bubble detectors, looks promising. It is planned to analyse the advantages and disadvantages of new detector designs by collaboration and to include newly available detectors in future intercomparisons.

3. The calibration procedures in individual monitoring are still under discussion. The Working Group will discuss and investigate the methods proposed by the ICRU and offer recommendations to improve harmonisation within the European Communities.

4. The Working Group will organize a Workshop on "Individual Monitoring of Penetrating Radiation-The Impact of the Recent Recommendations of the ICRU and ICRP". (Villigen, 5-7 May 1993). The aims of the Workshop are to stimulate the discussion on these topics, to support the revision of ICRP Publication 51 and to consider the need for formal guidance notes to personal dosimetry services.

WG 9: Criticality Accident Dosimetry

The Working Group coordinates investigations on the quality of the criticality accident dosimetry. The group will organize an international intercomparison of criticality accident dosimetry systems.

It will be performed at the SILENE reactor at Valduc/France. Beforehand, some work has already been done with measurements of spectral distributions of leakage neutrons from the lead shielded SILENE reactor. Additional neutron spectrum and dose measurements must be performed near the unshielded SILENE reactor in order to obtain a complete description of the radiation field. A Workshop on Criticality Accident Dosimetry is planned in cooperation with CEA und IAEA, where results and implications will be discussed.

WG 10: Basic Physical Data and Characteristics of Gas Ionization Devices

The general objective is the assessment of basic physical data relevant to the biological effect of ionising radiation and the development of instrumentation for dosimetry in radiation protection and radiobiology, with special emphasis on gas ionization devices, e.g. proportional counters (PC) and ionization chambers. The following tasks are planned for the contract period 1992-1994:

1. Basic data for ionization in gases

It is intended to update the average W-values for neutrons, Wn, which has not been done since the recommendations of Goodman and Coyne (1980). The Working Group will critically check and evaluate new data and prepare a report. The programme proposed includes also the evaluation of electron collision cross section data necessary to describe the secondary ionization production in charge collection processes.

2. Basic characteristics of ion chambers or proportional counters

The goal is to increase the collaboration between groups involved in theoretical and experimental research with gas devices. It is aimed to improve the performance of detectors by better knowledge of their basic properties, like electrical discharge mechanism, gas gain and time distribution of charge pulses.

3. Proportional Counters

An advisory report documenting the technical and scientific information required to construct low pressure proportional counters will be prepared. This is an important task, because commercially available devices are often unreliable and there exists an increasing demand for better detectors. The report is planned for 1993/94.

Training Courses

EURADOS will increase efforts in the training of young scientists in the fields of radiation physics, radiation dosimetry and radiation protection in order to transfer expertise and to distribute the knowledge on modern methods in these fields. In cooperation with other partners EURADOS will organize and actively participate in seminars or tutorials. A first seminar has been organized in cooperation with the GSF, Neuherberg, in November 1992 ("Modern Methods in Radiation Measurement and Dosimetry"). A similar course will be performed in 1994. A course on "Internal Dosimetry" is also planned.

A1 Measurement of Radiation Dose and its Interpretation

Contract FI3P-CT920054 Radiation quantities units and measurement techniques for radiation protection.

Coordinator ICRU
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Total Contribution by the Commission: 95 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Any determination of human exposure to radiation and radioactivity requires a fundamental set of quantities and units with which exposure can be specified and the means and ability to make measurements which yield results in terms of these quantities and units. Radiation protection, then, requires the capability to accurately quantify the characteristics and extent of radiation exposure so that appropriate and useful assessments of the potential health consequences and risks can be formulated. The work to be carried out seeks to meet these needs.

Specific Objectives

The development of internationally accepted recommendations on:

(1) quantities, units and concepts applicable to the practical determination of radiation exposure

(2) conversion factors for translation from calibration quantities to operational quantities for radiation protection purposes

(3) interaction data relevant to the fundamental interaction of radiation with body tissues

(4) measurement of quantities relevant to radiation protection

(5) phantoms and computational models for use in radiation protection

(6) utilization of in situ gamma spectrometry to assess radionuclides in the environment

(7) measurement of beta radiation for radiation protection purposes

(8) determination of body burdens for radionuclides

(9) fundamentals of particle counting applied to radioactivity measurements

(10) stopping powers for protons and alpha particles

(11) elements of assessment of exposure in medical applications relevant to dose reduction including performance assessment for diagnostic modalities and prescribing, recording and reporting doses in therapy procedures.

A11 Development and implementation of standards and procedures linked to the concepts of dose equivalent quantities for both external and internal exposure.

Contract FI3P-CT920040 Accident dosimetry in populated areas: the use of solid-state dosimetry techniques with ceramics and other natural materials.

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Total Contribution by the Commission: 230 KECU
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Description of research work

As part of the European Programme on Nuclear Fission Safety, the four contractors involved in this proposal will undertake a structured and integrated programme of investigation into the use of solid-state dosimetry techniques for the measurement of dose in populated areas arising from radiation accidents. The study will focus on current problems in Europe but has wide applicability to other areas. This document outlines the objectives of the work, the plan and output of the project and highlights the coherence and complementary nature of the work of the contractors.

OBJECTIVES

The objectives of the programme are as follows:

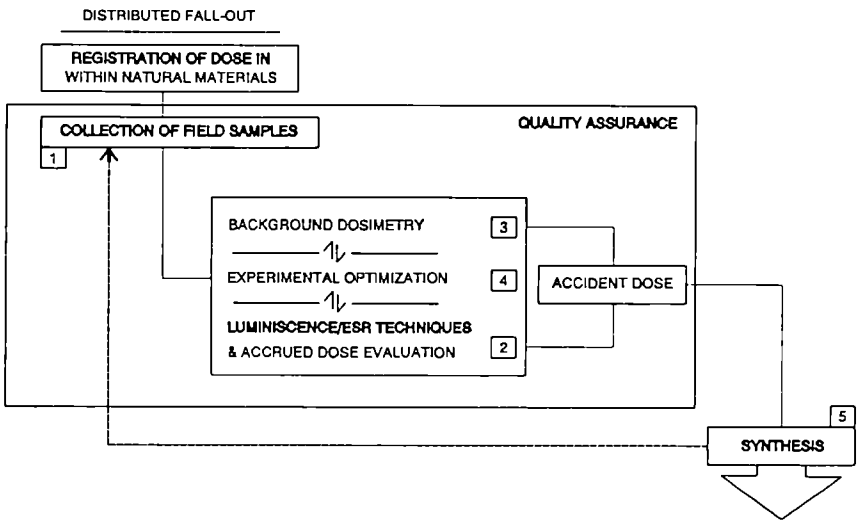
1. To identify ceramic and other natural materials (such as teeth) which are suitable for retrospective accident dosimetry using solid-state techniques.

2. To ascertain appropriate experimental procedures for dose evaluation.

3. To evaluate transient fall-out dose in the sampled study areas down-wind of Chernobyl fall-out in the Ukraine and southern Byeloruss. To derive shielding factors by comparing dose determinations for interior and exterior samples.

4. To identify factors which affect minimum resolvable fall-out dose.

5. Integration of the results obtained in 1) - 4) above to evaluate the capability of solid-state dosimetry techniques for retrospective accident dosimetry in populated areas These objectives will be achieved by completion of investigations in five modules as shown in the figure below.



MODULES OF THE PROGRAMME

The project represents the first stage of an investigation of the potential of the method and will comprise two strands:

1) an investigation of the methods and procedures used in the laboratory (see Annexe 1) and in the field followed by,

2) testing of these methods using a case study for which preliminary fieldwork has been completed. The case study will focus on the populated areas comprising selected areas of Pripyat, Ukraine and the environs of Kranopolye, southern Byeloruss, both of which which were reported to have received significant fall-out following the Chernobyl accident and which enable evaluation of the method in a proximal urban and a distal rural environment.

The work to be performed within each module is as follows:

1. Fieldwork

Review of circumstances of accident. Survey of study area, including previous monitoring, nature of buildings and population movements. Identification of samples, sample collection, recording and description. Measurement of ambient radiation levels.

2. Luminescence and ESR Techniques

Laboratory analyses of field samples. Mineral extraction; characterization of luminescent properties; determination of dose response; accrued dose estimates for samples (including dose-depth in the case of building fabric samples, eg bricks). ESR techniques will be employed with biogenic materials such as teeth and their potential explored in a pilot programme.

3. Background dosimetry

Assessment of current ambient dose-rate. Dose to selected luminescent minerals from natural sources of radioactivity.

4. Experimental Optimization and Standards

Optimization of experimental systems; the use of semi-automated control of luminescence measurements. Quality assurance and inter-laboratory standards for dose measurements (using field and other selected samples). Protocols for establishing checks and standards for experimental procedures, including comparisons with traceable dose standards.

5. Synthesis

Comparison of calculated and measured dose distribution within ceramic samples for isotope inventories appropriate to Chernobyl fall-out.

Assessment of pilot ESR/biogenic dosemeters programme.

Evaluation of shielding factors.

Examination of area dose distribution.

Error assessment.

DISTRIBUTION OF TASKS

Areas of primary responsibility and research contribution provided by participants are outlined below (see also Annexe 1).

01: Durham University

Investigation of luminescence properties of natural dosemeter materials including thermal and optical excitation; including field samples and selected materials [Module 2]. The use of tunable laser sources for stimulation of samples at elevated and low temperatures. Measurement of spectral emission.

02: GSF, Neuherberg

Background dosimetry including that within ceramic samples and the surrounding medium [Module 3]; coordination, with Risø, of laboratory source calibrations and intercomparisons; evaluation of potential of electron spin resonance (ESR, or EPR) for use with field samples, in particular biogenic materials such as teeth.

03: Oxford University

Optimization of experimental routines including luminescence dose evaluation and sample preparation [Module 4]; adaptation of porcelain testing procedures; coordination of an inter-laboratory comparison of internal sample dose-rate; maintenance a database of samples in circulation and serve as main repository where required.

04 Risø National Laboratory*

The use of semi-automated instrumentation and the use of non-laser sources; approaches to Quality Assurance, including reference dose measurements; deconvolution of doses to minerals for further use in studies of dose to humans;

* with sub-contractor University of Helsinki

METHODOLOGY

The project will be conducted in three stages comprising:

Stage 1. Evaluation of materials and methods

Assessment of currently available experimental procedures and evaluation using samples obtained from the study area. ESR Pilot study. Commencement of quality assurance programme designed to minimize and assess uncertainties in the measurement (in collaboration with the University of Utah) involving an interlaboratory calibration of sources, a blind intercomparison of artificially irradiated samples. Coordination with other dosimetry efforts including soil sampling and meteorological modelling with common points to be selected to allow comparison of results. Field collection of further samples, as dictated by outcome of assessments. A mid-term review will be held; agreed procedures will be adopted and form the basis of the Case Study.

Stage 2. Execution of case study

Completion of experimental work for study area following Adopted Procedures.

Stage 3. Synthesis

Assessment of experimental fall-out dose determinations and pilot ESR programme for natural biogenic dosemeters. Luminescence evaluations compared against modelling estimates in the case of Pripyat (there are data from over 26 monitoring sites) and against monitoring data available for the Southern Byeloruss sites. Shielding factors from indoor locations at ground and elevated levels (Pripyat). Evaluation of fitness for purpose. A workshop is also planned.

FUNCTIONING OF GROUP

The modules interlock to provide the output of the project. Each participant, although assigned a primary area of responsibility as described in the Distrubution of Tasks, will contribute some of their effort to all the topics according to need and ability. There will be, for example, tasks set such as inter-comparisons which will be common by necessity to two or more participants. Thus each participant has an essential role in the progression of the project towards its final objectives.

As a result the structure is highly complementary and, because of the inter-dependence of the modules in yielding the output of the project, will stimulate exchange and development of ideas between laboratories. It is also designed to provide the necessary mechanisms of primary collation before dissemination by the Coordinator.

Group interaction:

The group will use electronic mail for exchange of information; text documents will be prepared and transmitted in Rich Text Format, including diagrams where software support is available.

The exchange of workers between the laboratories is planned and visits to laboratories by the Coordinator during the project will promote effective operational balance.

The group has three scheduled group meetings at the beginning (Project Coordination meeting), at end of Stage 1 (Mid-term Review) and during Stage 3 (Final Report and Workshop). Opportunites for the group to meet and discuss progress (ie conferences) have also been identified.

The Coordinator will visit participants' laboratories during Stages 1 and Stage 3 in order to provide a reportage for the group.

FURTHER DETAILS OF PARTICIPANTS CONTRIBUTION

01: Durham University (Coordinator)

The main role of the laboratory will be dealing with fundamental aspects of the luminescence techniques used in the project, particularly those identified during Stage1. Thus those laboratories engaged in examining materials for dose evaluation, including Durham, would be reporting aspects requiring deeper investigation. Other research projects now in progress at Durham are highly complementary (investigation of the role of defects associated with luminescence) and are based on the study of both thermally and optically stimulated luminescence.

Discussions with the Univ. of Utah are in progress regarding various aspects of measurements (high resolution measurement of spectral emission and automated system control) and methodology (protocols for inter-laboratory comparisons) which would be of value to the project and from whom Durham would be keen to seek external assistance.

TASK PLAN

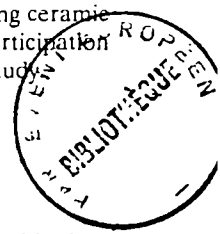
Stage 1. Planning and start up, including coordination tasks; Preparation of samples in the forms required, including inspection of mineralogy; Examination of the luminescence properties of mineral extracts (quartz, feldspar and mixed-mineral) for assessment of suitability for dose measurement using existing procedures.
Measurement of luminescence emission spectra; Optimization of detection systems and indentification of modifications; The use of tunable laser sources for stimulation of samples at elevated and low temperatures.
Mid-term report and (Coordinator) arrangements for mid-term review.

Stage 2. Determination of accrued dose estimates for selected field samples and reference irradiated ceramics, including dose-depth studies; Measurement of internal dose-rate using different methods (beta TLD, thick-source alpha-counting and EDXRF).

Stage 3. Reports and synthesis of final report (Coordinator).

External Assistance

External assistance may be sought from the University of Utah concerning: Deconvolution of dose to humans from luminescence/ESR determinations using ceramic materials; Software development for instrument control and data processing; Participation with laboratory inter-comparisons and initiating a US NIH regulated blind study.



Execution

The experimental work will be performed by a Research Assistant who will be hired on a short-term contract. The arrangements for the appointment of the RA will commence after the exchange of contracts.

02: GSF, Neuherberg

The role of the laboratory includes: background dosimetry including that within ceramic samples and the surrounding medium [Module 3]; coordination, with Risø of laboratory, of source calibrations and intercomparisons; evaluation of potential of electron paramagnetic resonance for use with field samples, in particular biogenic materials such as teeth.

1. ESR Investigations

Material collected from Pripyat and also similar commercially available products will be investigated in terms of measurable dose range and suitability for dose assessment using ESR techniques (see table below). Fifteen different samples collected from Pripyat and as many samples from manufacturers will be tested. Samples such as teeth and glass will also be examined, requiring a more extended period of testing. The studies are complicated in the case of teeth due to the complexity of the sample preparation and in case of glass because of the need to measure at low temperatures; also internal dose determinations are complex in the case of teeth. Determinations of the natural radioactive content of materials will also be determined.

During the second phase of the project, materials found to be suitable will be further investigated to develop procedures for measurements at low doses. GSF can provide samples irradiated at secondary standard laboratory facilities for the purpose of interlaboratory source calibration according to the demands of the other laboratories.

GSF will participate in elements of the second phase dose evaluation measurements (luminescence) using samples from the study area. Teeth from the study area will be sought for ESR measurements.

Table. Range of materials available for investigation

Sample	Method
Aspirin	ESR
Cough medicine($NaHCa0_3$)	ESR
Chalk	ESR+TL
Na_3PO_4	ESR
$Pb(CH_3C00)$	ESR
Metyl Orange	ESR
$NaCO_3$	ESR+TL
$CaCO_3$	ESR+TL
Penicillin	ESR
Glass	ESR
Sugar	ESR
Butadioni	ESR
Pot scale	ESR+TL
Teeth	ESR
Intestolan	ESR

03: Oxford University

The role of the laboratory will include: the optimization of experimental routines including luminescence dose evaluation and sample preparation [Module 4]; adaptation of porcelain testing procedures; coordination of an inter-laboratory comparison of internal sample dose-rate; maintenance a database of samples in circulation and serve as main repository where required.

The laboratory is well equipped for luminescence analysis using thermoluminescence and various optically stimulated methods. The laboratory has three Riso automated TL readers, an argon ion laser with appropriate software for automated luminescence measurements and automated infra-red stimulation equipment. In addition the laboratory is well-equipped for sample preparation for fine-grain and inclusion techniques and has a Buehler low speed diamond saw for sectioning stoneware and porcelain.

Samples will be prepared and tested using using high temperature fine-grain and quartz inclusion TL techniques and the pre-dose technique on stoneware and porcelain to evaluate absorbed radiation dose. If necessary mineral separation techniques will be used. Each sample will be measured using one or more techniques and assessed for the best method to use for dose evaluation in terms of sensitivity and reproducibility. The laboratory is prepared to offer training in the relevant techniques to members of other laboratories.

The optimized procedures will be evaluated in the second phase of the project where dose evaluations will be obtained with selected samples from the study area by each participant.

The application of standard methods for dose evaluation and further investigations using both the fine-grain technique and pre-dose measurements on porcelain and stoneware will include:

Fine-grain method

i) Use of fine-grains in dose-depth measurements; to give information regarding the energy of the incident photons.

ii) Investigation of firing temperatures and the effect of low (<500 °C) firing on the fine-grain signal.

iii) Trials with infra-red stimulated luminescence of fine-grains in cases where the TL signal is weak.

iv) Identification of mineral composition under the SEM or by other microscopic means.

High-fired ceramics

Samples of irradiated stoneware and porcelain (tiles and insulators) will be cut and measured using various techniques to evaluate the dose. If necessary, measurements will be carried out on artificially irradiated samples to compare the different methods and attempt to assess which is the optimum approach.

04 Risø National Laboratory (with sub-contractor University of Helsinki)

Risø National Laboratory will contribute to the project using its expertise within solid state dosimetry for assessing personal and environmental doses and dating archaeological and geological samples. Risø will thus in addition to thermoluminescence (TL) apply newly developed measuring techniques based on Optically Stimulated Luminescence (OSL) for determining the accumulated doses in brick and tile materials collected from the Chernobyl area.

OSL equipment based on a halogen lamp for measuring quarts samples using a filtered green wavelength band has recently been developed at Risø. The new OSL system, that also includes an excellent stability and sensitivity and is thus an attractive alternative to the commonly used more expensive laser systems. Preliminary OSL measurements of quartz samples using a green wavelength band from 420 to 550 nm, filtered from a simple incandescent low-power halogen lamp, has shown that accumulated doses in the order of 10 mGy give bright signals and thus OSL measurements of doses of 1 mGy and lower are achievable with sufficient uncertainty. This dose level is comparable to the total annual dose received by e.g. building materials from the natural background radiation. Other features of the new system is that combined TL/OSL measurements can be made in sequences and that quartz samples can be screened for feldspar contaminants (inclusions) prior to illumination with green light.

It is proposed to measure single aliquots i.e. very small samples to enable the determination of dose-depth profiles in bricks and tiles from buildings. Such data are valuable for assessing the total absorbed dose to man from the ambient radiation. The feature of being able to measure OSL from both quartz and feldspar minerals included in the same sample provides an excellent possibility for cross checking the results.

Risø will undertake the following tasks over 19 months (1st November 1992 to 31st May 1994):

1) Planning and start-up of project
2) Selection and preparation of samples (coarse grains) from different brick and tile materials including old samples with known age (dose) and new samples from the Chernobyl area.
3) Investigation of TL/OSL techniques and methods with special regard to measuring accumulated doses to natural quartz and feldspars included in brick and tile materials. The experimental work will comprise:
   a) Determination of the lowest detectable dose
   b) Determination of precision and reproducibility at different dose levels.
   c) Determination of the effect on calibration procedures of the sensitivity change of TL and OSL signals as functions of preheat, annealing and optical bleaching.
4) Dose evaluations including the determination of the natural background dose (using neutron activation analysis for dose-rate determination) and superimposed man made doses based on calibrations using reference Co-60 and Cs-137 gamma radiation fields at Risø. It is suggested to perform dose evaluation intercomparisons between the different partners using different methods on identical samples.
5) Applications for accident dosimetry. Attempts will be made to measure depth dose profiles in bricks using the single aliquot method (single grains). Such data are valuable in the estimation of the absorbed dose to man.
6) Preparation of reports. A progress report presenting applied methods and preliminary results and a final report containing the results and conclusions of the above described work will be completed around 1st September 1993 and 1st May 1994, respectively, as required by the CEC.

ANNEXE 1: A summary of the luminescence techniques to be employed

The property of luminescence enable certain natural crystalline minerals - such as quartz and feldspar - when exposed to ionizing radiation, to register the absorbed dose and, at some later time, to yield a quantitative measure of that dose. It arises from the trapping of charge carriers, freed during irradiation, at defect sites which provide metastable states within the forbidden gap, the trapped charge carrier population being proportional to dose. When previously irradiated crystals are stimulated, the trapped charge carriers are released and recombine with charge carriers of opposite sign at other trapping sites, resulting in the release of light.

Until recently the stimulation was achieved using heat, resulting in thermoluminescence (TL); research has shown that optical stimulation may also be used, giving rise to optically stimulated luminescence (OSL). If freshly irradiated materials is stored at ambient temperatures the latent signal can be retained without loss (depending on the mineral concerned) for many thousands of years, thus providing an extremely stable dosemeter. Electron Spin Resonance (ESR) is a related technique for the measurement of trapped charge in insulating materials and has the advantage of being suitable for use with organic materials such as teeth or bone. For brevity we will confine our discussion of the principle of the method to the use of TL.

Measurement of absorbed dose

In its simplest form the TL method may be used to evaluate an unknown dose (comprising the accrued dose due to all sources of ionizing radiation) delivered to TL crystals by heating them to record the first glow curve (a record of luminescence intensity vs sample temperature) and comparing this curve with the second glow curve obtained using a calibrated laboratory radiation source (either beta or gamma emitting). TL minerals found in ceramic materials exhibit complex behaviour which require carefully developed experimental procedures - they are described in various papers cited elsewhere in this application. Common to all techniques is an investigation of the linearity of TL response with dose (referred to as the growth characteristic). In this project we propose to use the quartz pre-dose and inclusion techniques; the former being considerably more sensitive than the latter and capable of registering doses of ca 10 mGy.

The dose absorbed by crystalline minerals, D_x, from a transient accident dose, is given by,

$$D_x = D_{TL} - A.(R_\alpha + R_\beta + R_\gamma + R_\chi)$$

where,

R_α , R_β , R_γ and R_χ refer to the annual dose due to alpha, beta, gamma and cosmic from natural sources of radiation. A = sample age and D_{TL} = accrued dose determined using TL (or OSL).

The annual dose arises from the decay of uranium, thorium and potassium within the sample and the surrounding medium. Two approaches are used routinely; one where the alpha contribution is fully registered (by selecting grains of diameter ca 2-10 Ám) and the other where the alpha contribution is negligible (by selecting grains of diameter ca 100 Ám). The annual dose is determined by a variety of procedures; for typical brick structures the accrued dose due to natural sources of radiation is expected to be in the region of 2.5 - 3.5 mGy/a with contributions of approximately 60%, 25% and 5% for beta, gamma and cosmic components respectively (using the larger grains, assumed to be of negligible internal radioactivity). Thus for such a structure, the accrued dose due to natural sources is expected to be be roughly 30 mGy per decade - this forms the baseline onto which further transient dose contributions are made. Hence assessment of the uncertainty associated with the determination of the age and dose-rate is of some importance where the additional transient dose approaches the naturally accrued dose. Analysis of the errors associated with the required measurements have shown that for brick of age 10 ±1 years and associated uncertainties of ±5% (ß) and ±10% ($\gamma + \chi$) for annual dose components, and ±10% for the accrued dose, the minimum detectable transient dose is currently circa 10 mGy. These estimates are probably reasonable to assume for the proposed study.

A11 Development and implementation of standards and procedures linked to the concepts of dose equivalent quantities for both external and internal exposure.

Contract FI3P-CT920064h The measurement of the spectral and angular distribution of external radiations in the workplace and implications for personal dosimetry.

Coordinator NRPB

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Total Contribution by the Commission: 140 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Overall objectives

The main objectives of the collaborative project are to develop methods for measuring the spectral distribution of external radiation in the workplace, particularly photon and neutron radiations, and to examine calibration methods for personal dosimetry. The results of the measurement programme will be used to investigate the use of operational dose equivalent quantities in the workplace, and to make estimates of effective dose in known radiation fields. Theoretical calculations will be used to support experimental measurements and, where appropriate, recommendations will be made for the calibration of personal dosemeters. At present there are methods available to measure the energy spectra of neutron radiation in the workplace, but surprisingly little work has been done to measure energy and angular spectra and photon radiation. A method has been developed at NRPB to measure photon spectra in the workplace using Geiger-Muller detectors, and current work is aimed at developing a novel experimental method using sodium iodide detectors. The results of this work can then be used to give practical advice on calibration methods for personal dosemeters, and the suitability of operational dose equivalent quantities.

Description and specific objectives

1. To develop and compare techniques of measuring spectra and the directional distribution of x and γ radiation in the workplace (NRPB/AEA).

   To refine equipment based on Geiger-Muller and Sodium Iodide detectors and carry out measurements in the workplace of energy and angular distribution.

   To examine unfolding techniques developed for x-ray and neutron spectroscopy, and use them to obtain energy and angular distribution from the results of measurements in the workplace.

   To examine the results of measurements and make calculations of effective dose and operational quantities in the workplace.

2. Development of methods to measure the spectra of neutron radiations in the workplace (AEA/ENEA).

   To refine existing measurement techniques.

   To make measurements of the spectra of neutron radiations in the workplace.

   To catalogue the results of measurements.

3. Calculations of experimental measurements to examine the performance of personal dosemeters (ENEA/CEA).

   Calculation of the fluence and energy of backscatter from the ICRU sphere and other phantoms in ISO beams of X and γ radiation.

   To carry out measurements to verify results of calculations.

   To examine calibration methods for new quantities and make recommendations for appropriate methods.

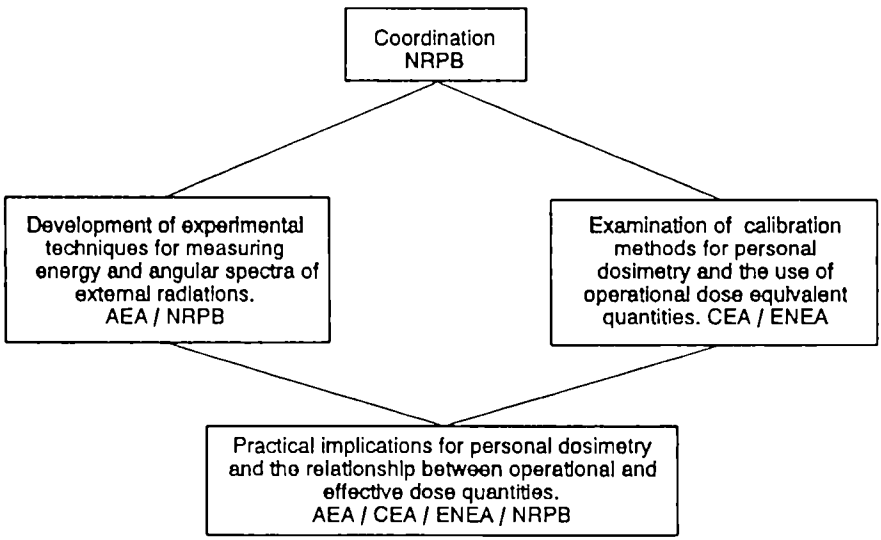
4. To assess the implications of spectral and spatial distribution measurements on personal dosimetry in the workplace and on methods of calibration (NRPB/AEA/ENEA/CEA).

   To assemble a catalogue of the results of measurements of the energy spectra and angular distribution of photon radiations in the workplace.

   To recommend the best method of measuring the energy and spatial distribution of photon radiations in the workplace.

   To recommend methods of calibration, personal dosemeters given a knowledge of the energy and angular distributions in the workplace.

Overall schematic of project management and roles.



Contribution of the NRPB, Chilton, UK

NRPB will coordinate the project, and provide exposure and facilities to develop reliable methods for measuring the energy and angular distribution of photon radiations in the workplace. In collaboration with AEA Harwell, the design of a spectrometer based on a shielded sodium iodide detector will be optimised and a catalogue of measurements in typical workplaces will be obtained. The implications of such measurements for personal dosimetry in the workplace will be examined, including the use of operational dose equivalent quantities for estimating effective doses to workers. This latter work will be carried out in collaboration with AEA, CEA and ENEA.

The Dosimetry and Instrumentation Department at NRPB has considerable experience in developing new methods for measuring spectral characteristics of external radiation. The Department has the necessary expertise and facilities to complete the project and the work is consistent with the Board's corporate policy to do research and development in support of improvements in radiation dosimetry. The work will be given a high priority because of the need to improve dosimetry in the workplace following recent recommendations from ICRP and ICRU. The use of dose constraints in the workplace will lead to pressure for greater accuracy in assessments of worker dose, which can be achieved by spectral measurements and improvements in personal dosimetry.

Contribution of the AEA, Harwell, UK

Harwell Laboratory and the NRPB have put together a combined experimental and theoretical programme with the aim of producing instrument for the operational measurement of the energy and angular distribution of photon spectra. In addition, Harwell Laboratory have made operational improvements to their neutron spectrometry system and measured neutron spectra at a number of locations in Europe.
In close collaboration with the NRPB, Harwell Laboratory will provide theoretical and computational input to:

a) to compare the theoretical and experimental evaluations of the portable gamma-ray spectrometer and then refine its design to improve its angular and energy response;

b) use of portable gamma-ray spectrometer in operational conditions to provide a catalogue of information for EURADOS-CENDOS working group 7 to enable better design of dosemeters and their interpretation.

c) evaluate the use of the unfolding programs STAY'SL and RADAK for unfolding neutron energy spectra; and

d) perform an intercomparison of neutron spectrometers in conjunction with the CEA.

Contribution of the ENEA, BOLOGNA, ITALY

The Metrology and Radiation Protection Division of ENEA, include a Primary Dosimetry Laboratory, a Secondary Standard Dosimetry Laboratory and a Personal Dosimetry Service. The division is also deeply involved in the development of computational dosimetry models. In this particular framework dosimetric and safety assessments for radioactive material transport flasks have been carried out in the past years.
Extensive theoretical studies and experiments on the characterization of radiation fields related to the ICRU new quantity determinations have been done in the frame of CEC Contract No. B16347(UK)H. This activity, which was successfully connected with research programmes of other European Laboratories, obtained some important results but it is necessary to extend the investigations in order to complete the initial aim of the project. Taking into account also the ICRP 60 and ICRU 47 Recommendations, the main topics to be developed will therefore be the following:

1. Dosimetric calculations on photon radiation fields in the presence of an ICRU reference phantom, namely the tissue sphere. These calculations will be devoted to the evaluation of H' at 0,07, 3 and 10 mm for different incident angles and various x and gamma reference radiation beams. The results will be validated through a comparison with measurements on the RS-1 tissue equivalent sphere to be performed in our SSDL.

2. Further efforts will be devoted to implementation of complex anthropomorphic phantoms (eg. Adam and Eva) in the Monte Carlo code MCNP, in view of the evaluations of the various dosimetric quantities in realistic conditions.

3. In connection with point 2, the workspace photon spectra as determined by NRPB laboratories will be used in the Monte Carlo simulations.

The feasibility of applying Monte Carlo techniques to worker movements will also be investigated.

Contribution of CEA, FONTENAY AUX ROSES, FRANCE

Taking into account the ICRU proposals about the dose equivalent operational quantities for radiation protection (ICRU Reports 39 and 43) and the latest recommendations of ICRP Publication 60, defining new maximum primary dose limits with a different distribution of weighting factors for organs, it is necessary, for the individual monitoring of exposed persons to have a better accuracy in the assessment of personal doses. The CEA-SDOS contribution rests on the study and elaboration of calibration procedures for personal dosemeters in agreement with those requirements. It represents an extension of the work initiated during the previous period (1990-92), stressing the evaluation of the uncertainties on results provided by usual dosimetric systems used by Dosimetry Services.

In the project, it is aimed at determining the calibration factor of individual dosemeters, assuming the knowledge of the photon spectrum (AEA Harwell and NRPB contributions) and the determination of their characteristics (energy and angular responses). The simulation of "realistic" conditions of irradiation will be progressively obtained by duplicating, as close as possible, the energy and angular distributions in the workplace,

the uncertainties in position of the dosemeters at the body surface and the movement of persons in the workplace area. Different types of dosemeters will be involved in an extensive irradiation programme, partially performed on the ENEA facilities. In this way, it is intended to take into account the specific types of dosemeters and derive information as a function of the chosen calibration quantity. The use of the operational dose equivalent quantities requires the use of calibration phantoms as recommended by ICRU Report 47. Work will be carried out to evaluate the possibility of using other calibration quantities (free in air), compatible with the needs of routine dosimetry services. Where large numbers of dosemeters have to be irradiated and the use of calibration phantoms is inconsistent. This approach will also rely on calibrations and actions coordinated with the ENEA participation.

A12 Radiation measurement and instrumentation for individual and area dosimetry.

Contract FI3P-CT920002 Realistic neutron calibration fields and related dosimetric quantities.

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Total Contribution by the Commission: 310 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The main objective of this collaborative project is to provide in the laboratory a few well-characterised neutron fields that replicate typical spectral neutron fluence distributions met in radiation protection practice. These fields are needed for the calibration of neutron area and personnel dosemeters since these do not generally have the energy response required to determine dose equivalent quantities.

The project consists of four distinct parts, namely

- the measurement of the spectral neutron fluence typically encountered in practical situations,
- the preparation of a catalogue of all measured spectra in an agreed format including the calculation of relevant dose equivalent quantities and the response of generally used neutron dosemeters,
- the inspection of this catalogue in order to extract a few representative spectra and their expansion in terms of already available or, if necessary, newly defined calibration spectra, and finally
- the computational prediction of configurations consisting of the usual neutron sources (accelerator, reactor or radionuclide based) and appropriate moderators in order to produce these reference fields in the laboratory.

The second part includes in addition:

- the calculation of fluence-to-dose equivalent conversion functions for the operational and the primary limiting quantities (effective (body) dose (equivalent)) according to the recently revised ICRP recommendations (ICRP60) and
- the setting-up of an expert system with access to those parts of the catalogue needed to estimate in advance the readings of neutron monitors and dosemeters in specific neutron fields. This program package will be made available to experts responsible for providing a radiation protection service in laboratories and plants.

Allocation of the tasks:

The research groups at CEA, GSF, NPL and PTB collaborating in the project have considerable experience in the field of neutron metrology and dosimetry. The tasks are allocated as follows:

I. CEA, NPL artd PTB will continue to make measurements at working places and to improve spectrometry techniques and the analysis of measured data.

2. GSF, NPL and PTB will cooperate in updating the catalogue, refining the program for data handling and analysis, and designing and setting-up a user guided expert system.

3. GSF will concentrate on the calculation of conversion functions on the basis of the recently revised ICRP recommendation, partially overlapping with activities at PTB.

4. CEA, PTB and NPL will numerically simulate "realistic" neutron fields. Optimised moderator assemblies will be realised and experimentally investigated at the accelerator facilities of CEA, NPL and PTB. The most suitable configurations may then be used for intercomparison measurements and also be duplicated at the accelerator of GSF.

Contribution of the Participants

1. Physikalisch-Technische Bundesanstalt (PTB), FRG

The PTB group will contribute to all parts of the project.

Measurements of the spectral fluence at working places will be performed wherever necessary and possible, e.g. at accelerators used for isotope production, cancer therapy or fundamantal research, in the environs of containers for the transport of fuel elements, at calibration facilities etc. In connection with these investigations improved response functions will be applied and various unfolding procedures which are available will be compared (partially in cooperation with CEA).

The program for handling the catalogue will be refined and the catalogue will be continuously updated. Recently determined conversion and detector response functions will also be included. A special concern is to search for a few basic spectra suitable for describing most of the data measured at working places. The expert system may be extracted from the program package and the data library.

Monte Carlo simulations have already shown that very different neutron fields can be produced with accelerator-based (monoenergetic) neutron sources using rather simple shielding and moderator assemblies. The configuration of various materials must be optimised computationally in order to replicate the few typical neutron spectra. The most promising neutron fields will then be realised at the accelerators available at PTB and experimentally be characterized.

2. National Physical Laboratory (NPL), UK

NPL has already made a number of spectrum measurements in typical protection level fields, and will continue to make measurements requested by customers in the UK and elsewhere. In addition work will proceed to produce an integrated spectrometry system where the results of three separate spectrometers (a Bonner sphere set, an NE213 scintillator, and a ^{3}He spectrometer) are combined to provide a single spectral distribution incorporating all the information from the three devices.

The main task of the contract is to utilize the measured spectral distributions and use the data base of spectra in working areas to answer a number of questions, i.e.:

- What are the typical under or over readings of dosemeters (area monitors, personal dosemeters, and albedo dosemeters) in working environments?
- To what extent are there similarities in the fields measured at similar facilities (reactors, source fabrication plants, nuclear fuel reprocessing plants, the environs of fuel transport flasks etc.)?

- Are the spectra at a particular type of facility sufficiently alike for a single calibration field to be viable for that type of facility?
- Can valid calibration spectra be reproduced in the laboratory using radionuclide sources, or accelerator produced neutrons, and appropriate moderation?

If the answer to the last two questions is yes, the latter part of the contract will involve work to construct and characterise more appropriate calibration facilities. If the answer is no, the investigation will concentrate on identifying the most suitable combination of presently available calibration fields to provide calibrations for particular dosemeters which will minimise the degree of over- or under-read in the types of field in which they will be used.

3. Commissariat a l'Energie Atomique (CEA/SDOS)

Being convinced that the Bonner-sphere technique and proton-recoil spectrometry must be combined to produce reliable results in the energy range O.01 eV - 20 MeV, the methodology used for the measurements and the evaluation of numerical results must be improved. Work will concentrate on the use of different readily available codes, a better knowledge of the response functions of the various detectors used and on estimation of the most evident parasitic effects (if possible). A simplified approach to evaluating uncertainties of results should be included.

A facility simulating a few examples of "wide" neutron spectra has been realised in the SDOS-Cadarache Laboratory (14.6 MeV facility). Calibration procedures will be elaborated. In particular, the question of the traceability to primary references must be considered (neutron fluence, monitoring).

A 2nd configuration of this accelerator-based facility (2.8 MeV via the reaction $D(d,n)He^3$), previously studied by means of Monte Carlo calculations in terms of neutron spectra and dose equivalent rate, will be developed. In particular, a specific line of work must be undertaken for the optimisation and realisation of the monitoring system (associated particle technique). The spectrometric and dosimetric characterisation of the field will be performed and compared with the calculated spectra.

Finally, the well specified "14.6 MeV" and "2.8 MeV" fields will be made available for spectrometric intercomparison experiments organised in the framework of the contract (or by EURADOS WG 7).

There will be a continuing programme of measurements at French installations where spectrometric measurements at working places need to be performed provided that official administrative or legal authorisations are available.

4. GSF - Forschungszentrum für Umwelt und Gesundheit FRG

The activities of GSF will concentrate on the determination of conversion functions. This includes the calculation of the absorbed dose in the organs of the heterogen man phantom (MIRD phantom) and the application of the revised quality factors and revised tissue weights as given by ICRP60, to determine the dose equivalent.

Evaluation of the impact of the introduction of the proposed radiation weighting factors in comparison with the ICRP/ICRU approach, adopted up-to-now and using the Q(L) dependence for calculation of the tissue doses, is of particular concern.

It is evident that any kind of operational quantity which is chosen to be conservative at high neutron energies will greatly overestimate the effective (body) dose (equivalent) at lower neutron energies. This is unavoidable and is due to the strong energy dependence of the neutron depth dose characteristic on the geometry of the various exposure conditions. This means that the actual integral differences between the operational quantity and effective (body) dose (equivalent) depends essentially on the respective energy distribution of the specific neutron field.

The idea is now to establish an expert system which enables the responsible experts on site to estimate in advance the integral readings and conversion factors for the specific neutron field. This system will make use of the catalogue compiled in the frame work of the project.

The following steps are envisaged:

1. Calculation of effective dose equivalent using the recent ICRP proposals for quality factors and tissue weights.

2. Calculation of the ratio "effective dose" to effective dose equivalent for all available spectral data sets.

3. Design and setting-up of a user-guided expert system.

4. Trial to establish a simple slowing-down calibration field taking into account the restricted possibilities of the GSF neutron laboratory.

A12 Radiation measurement and instrumentation for individual and area dosimetry.

Contract FI3P-CT920018 The measurement of environmental radiation doses and dose rates.

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Total Contribution by the Commission: 185 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Based on the effort put on the evaluation of practical calibration methods for environmental monitoring systems, including integrating TL dosemeters, and the determinations of a variety of detector characteristics carried out as a successful cooperation between RISØ, PTB and CIEMAT in 1990/91 (under CEC contract Bi7-027) it is the intention to continue this work by studying further demanding aspects of environmental monitoring. In view of the increasing use of electronic dosemeters for dosimetry applications, such devices will be involved in the environmental programme to see how they respond to ambient doses.

Special emphasis will be put into the evaluation of the contribution of radon daughters to the total detector responses when measuring the natural radiation. Experiences with several early warning gamma monitoring systems installed in many European countries as a consequence of the Chernobyl accident have shown that varying detector responses from radon daughters have caused problems with the alarm setting of the environmental monitoring systems. It is thus important to initiate long term studies of detector responses to radon daughters to enable the evaluation and recommendation of suitable alarm criteria.

Experiments will be undertaken to determine the terrestrial and cosmic radiation responses of different new detectors used for assessing the doses received by people from environmental photon radiation

It is furthermore intended to investigate new commercially available TL materials and to study self-dose and dose rate dependence effects for a variety of TL materials suitable for measuring the natural background radiation.

Distribution of tasks and collaborate links

It is proposed to establish a Natural Environmental Monitoring Station at a selected field site at RISØ with the aim of being able to offer to CEC member states reference measurements at a well determined natural environmental photon spectrum. A reference Cosmic Measurement Station at Roskilde Fjord is also planned. Long-term measurements at the Natural Environmental Stations using different detectors, including TLDs, will be intensively studied with the aim of evaluating the influence of radon daughters, air pressure, rainfall and snow shielding. The desirability of establishing such European standard monitoring stations (terrestrial and cosmic radiations) is being clearly demonstrated by the present intercomparison of different detector types at the Hinkley Point Power Station (CEC contract Bi7-027). This work clearly shows that inconsistent and inaccurate measurements of environmental releases from nuclear facilities can be made unless such field stations are used to accurately determine the response to terrestrial and cosmic radiations.

At present European countries who wish to assess and intercompare their environmental TLDs have to send these to the USA. The establishment of a European field station at RISØ would provide a much cheaper and more convenient way of doing this work.

It would eliminate the large errors that can arise from the significant transit doses of two Atlantic flights. Continuity with the American international intercomparison programme would be achieved by direct comparison measurements between RISØ and the USA Chester Field Station.

The RISØ free-field and shadow-shield calibration facilities will be used for checking the participants' instrument calibration following transit and prior to the exposure of their monitoring equipment at the field stations.

The newly established UDO low-level measurement laboratory in the Asse saltmine under the management of PTB, Braunschweig will play an important role in future activities e.g. for the determination of sensitivity, linearity and inherent background of new types of detectors and electronic dosemeters. Experience has further shown that it is very important to investigate and determine the dose rate dependence of TL materials at ultra-low dose rates taking into account the self-dose contribution and the fading characteristics. The UDO laboratory appears to be a unique facility for experimental investigations of such parameters.

An intercomparison of four different detectors used over a prolonged time to determine the ambient radiation at the Hinkley Point Power Station (project Bi7-027) also showed inconsistant results due to the high energy component (N-16). To facilitate monitoring around nuclear installations PTB will provide their 6 MeV photon calibration facility to determine the responses of different new detectors, including TL dosemeters, to high energy photons.

The TL glow curve analysis techniques introduced by CIEMAT will be further developed to assess the dose rate dependence and self-dose properties of a variety of TL materials used for environmental monitoring. This technique will be tested against experimental measurements carried out in the ultra-low radiation environment in the Asse facility. The extremely sensitive but complex LiF:Mg,Cu,P TL material (Chinese GR-200) is an interesting alternative to TL materials normally used for measuring the ambient radiation. It is thus intended at CIEMAT to develop a glow curve analysis computer programme with the aim of using this particular phosphor for routine environmental measurements. The performance of the programme will be tested against measurements carried out at the natural environmental test station at RISØ. CIEMAT will further assist in implementing a numerical glow curve analysis computer programme with the RISØ TL environmental monitoring system that is based on special non-linear N2 heating.

To facilitate the intercomparison of measuring results of the environmental radiation within the European countries it is important to provide guidance based on the experience gained on studies of calibration methods, measurement procedures and the handling of TL materials.

All the proposed activities deal with subjects that are highly relevant to the participating laboratories and also all the participants are experienced and actively involved with the topics. The work will thus be undertaken both theoretically and experimentally in close cooperation between RISØ (including I.M.G. Thompson), PTB, CIEMAT and NPL.

Project 1: Risø National Laboratory

Based on the effort put into the evaluation of practical field and laboratory calibration methods for environmental monitoring systems (Monte Carlo calculations of a variety of source-detector geometries) carried out under the CEC contract Bi7-027, it is the intention to continue this work by studying new demanding aspects of environmental monitoring. In 1992/94 Risø National Laboratory will undertake the following work:

1) Commissioning of established free-field and shadow-shield calibration techniques and facilities at RISØ, including Monte Carlo calculations of scatter contributions from calibration geometries using different gamma sources, for the benefit of European environmental measurement laboratories. (RISØ-PTB).

2) Establishment of an European Standard Natural Environmental Monitoring Station at RISØ, including both a terrestrial and a cosmic site, for long term tests of environmental monitors and dosemeters. Parameters to be studied are the influence on the natural background dose rate of radon daughters, air pressure, rainfall and snow shielding. The monitoring stations will provide a standardised natural environment for terrestrial and cosmic radiation at which CEC member countries would evaluate and intercompare their dose rate monitoring equipment and dosemeters. (RISØ-PTB-CIEMAT).

3) Long-term studies of the effect of radon daughters as measured with different environmental gamma monitors with the aim of being able to recommend reliable alarm criteria for early warning environmental total gamma monitoring systems installed in CEC member states. (RISØ-PTB).

4) Free-field calibration studies of a variety of recently developed electronic dosemeters, which might replace TL dosemeters in future dosimetry applications, with the aim of investigating the possibilities of using such devices for environmental monitoring. (RISØ-PTB-CIEMAT).

5) Implementation, testing and commissioning of numerical glow curve analysis techniques developed at CIEMAT with the routine TLD environmental monitoring system (non-linear N2 heating) at RISØ. (RISØ-CIEMAT).

6) Enter into a **subcontract** with I.M.G. Thompson (Consultant), U.K. who will assist RISØ in establishing their standard environmental dose rate stations.

Project 2: P.T.B. Braunschweig

The intercomparisons of environmental gamma dose rate meters performed in recent years and which were supported by the CEC have shown that measurements of the environmental photon radiation can be only correctly interpreted and intercompared if the properties of the instruments used are well known.

Several kinds of instruments have a significant inherent background of the order of 10% of the normal natural environmental radiation level. To evaluate dose rate measurements it is important to take into account the inherent background.

In addition the linearity between the reading and the dose rate for low radiation levels must be known. For continuation of the work in the frame of a CEC contract the following proposed tasks will be undertaken by PTB:

1) Investigation of the inherent background in the UDO ultra low background facility of new types of dosemeters e.g. electronic dosemeters. In the future this kind of dosemeter is foreseen to be an alternative to other kinds of passive dosemeters in environmental monitoring.

2) In cooperation with the CIEMAT continuation of the work in the field of TL dosemeters:

- dose rate dependence at low rates
- self-dose contribution
- fading and sensitivity changes.

3. Determination at low dose rates of the response of dose rate meters and integrating dosemeters (including electronic dosemeters) on the dose rate (linearity).

4) In cooperation with the Risø National Laboratory experimentally verify their theoretical calculations of dose rates in different field calibration geometries using the Monte Carlo programme developed at RISØ which takes into account the geometrical arrangement of the irradiation facility. The RISØ Monte Carlo code will further be used to calculate the photon fields in the UDO laboratory.

5) Providing a 6 MeV photon calibration facility to determine detector responses with the aim of measuring doses from high energy photon fields.

Project 3: CIEMAT

The contribution of CIEMAT to the project involves studying the important properties of passive TLD detectors which are relevant to environmental monitoring. Such properties are not very well established, either because they are still the subject of debate between different authors or because there is very little published data available. In particular the CIEMAT group will pay special attention to new promising TL materials, such as LiF: Mg, Cu, P, that are thought to possess important advantages over other conventional materials.

For this project the CIEMAT group propose to cover the following topics for the different TLD's currently employed in environmental monitoring:

- Temperature effects during long exposures (CIEMAT-RISØ)
- Linearity at very low dose rates. Dose rate effects (CIEMAT-PTB/UDO-RISØ)
- Self-dosing and non-radiation spurious signals affecting low dose measurements (CIEMAT-PTB/UDO)
- Development of computer aided procedures for TL evaluation at very low doses (glow curve analysis) (CIEMAT-RISØ)
- Development of TLD badge especially suited for the measurement of very high energy photons (6 MeV) (CIEMAT-PTB)

In addition CIEMAT will provide TLD's for other common experimental activities at RISØ and PTB.

The collective experience of the groups participating in the project and the facilities they provide creates a very favourable opportunity for CIEMAT to develop its proposed research on TLD detectors. The UDO Laboratory in the Asse mine will provide a unique facility in which to undertake the self-dosing and low dose rate linearity experiments, since it is essential to undertake such work in an environment having a natural dose rate as low as possible. The proposed RISØ monitoring field station will be a suitable place for performing the long term exposures necessary for studying temperature effects under real field conditions. It will also enable CIEMAT to check the adequacy of the new procedures they have developed for taking account of possible TLD sensitivity changes resulting from fluctuating ambient temperatures during exposure.

For these studies CIEMAT will develop computer codes for the analysis of glow curves. The programmes perform either a simplified analysis of the glow curves which is adequate for normal TL dose evaluations, or a more complex analysis which resolves the individual TL peaks and enables numerical values to be estimated for different peak parameters (activation energy, frequency factor and peak maximum temperature). Using this method of analysis a more detailed study of the TL signals is possible, permitting separate information to be obtained on the properties and features of each of the individual TL processes contributing to the full glow curve. This approach has clear and objective advantages over conventional TLD evaluation techniques which merely integrate all the light emitted by the dosemeters, mixing the contribution of the different TL processes. These advantages have been clearly demonstrated by some of the results obtained during the former CEC project Bi7-027 "Measurement of Environmental Gamma Doses".

A12 Radiation measurement and instrumentation for individual and area dosimetry.

Contract FI3P-CT920026 Detection and dosimetry of neutrons and charged particles at aviation altitudes in the earths's atmosphere.

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Total Contribution by the Commission: 260 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

It is known that the dose to aircrew on long-haul civil aircraft may exceed the dose limits now recommended for members of the public according to the recent Publication 60 of the International Commission for Radiological Protection.

The objectives of this project are:

(1) To obtain detailed information on the flux and energy spectrum of neutrons at aviation altitudes.
(2) To obtain charge spectra and flux for high Z particles at aviation altitudes.
(3) To use the measured data to calculate the doses received by aircrew as a result of their occupation.
(4) To validate and modify computer programs for the computation of doses on the basis of the impinging external radiation fields.
(5) To suggest procedures to minimise doses.

The basis of the project will be the development of a multidetector package using etched track detectors and superheated bubble dosimeters combined with a modified double moderated Anderson-Braun counter to measure neutrons up to energies of 400 MeV. A tissue equivalent proportional counter will also be used for direct assessment of doses relevant to the project in in-flight measurements. These will be calibrated at existing facilities and then flown in aircraft at a sufficient range of latitudes and altitudes to enable calculation of the charged particle and neutron flux. These data will then be used for calculation of equivalent doses and hence for determination of organ dose response functions for the instruments used.

The data sets and assumptions relating to the incident cosmic ray field will be analysed and existing computer codes for the transport of the incoming galactic and solar radiation fields in the atmosphere will be implemented and tested. This should enable predictions to be made of energy spectra for the different cosmic ray components in the atmosphere as a function of co-ordinates and altitude. These predictions can then be compared to the experimental results enabling further refinement of the computational procedures.

It is expected that this project will enable a validated and documented procedure to be produced for the calculation of organ equivalent doses as a function of longitude, latitude, and altitude. This could be in the form of a computer program for computation of passenger and air-crew exposures for any specified flight co-ordinates by line integration over the three dimensional dose rate space traversed by the aircraft.

Statements from each of the partners.

1. University of Dublin. Department of Pure and Applied Physics. Trinity College

When experimental results have been obtained and numerical estimates of doses are available on various routes used in civil aviation, it will be possible to make an assessment of the implications for health of these exposures.

Following this, guidelines can be suggested for civil aviation authorities to ensure that the highest standards of protection are available for exposed passengers and crew.

As part of the co-ordination of this contract, it is intended to maintain close liaison with the EURADOS Committee and with other Community and national authorities involved in regulatory aspects of radiological protection.

2. ENEA - Divisione Metodologie e Misure.

In-Flight Neutron Dosimetry

Measurements of cosmic ray neutrons require different types of detectors to cover the entire energy range from 0.1 MeV up to tens of GeV.

The multidetector system proposed in this project will make it possible to obtain short-term and long-term neutron measurements and will be formed by:

- Bubble detectors,
- Electrochemically-etched track detectors of polycarbonate derivatives,
- Spark counter of damage tracks of neutron-induced fission fragments in bismuth, gold and tantalum,
- Lead-shielded Rem counter (provided by Prof M Pelliccioni from INFN, Frascati, Rome).

3. GSF - Institut für Strahlenschutz.

Calculation of Individual Exposures in Aircrafts and of Response Functions for Radiation Measurement Devices for Validation Experiments

Galactic cosmic rays and energetic particles from solar particle events produce elevated radiation exposure levels in aircrafts. The recent ICRP-Publication 60 recommends to consider also such elevated levels of natural radiation in the assessment of occupational exposure, eg of air crews. Frequently flying members of critical groups of the general public might also receive doses of concern in radiation protection. The study aims at calculating organ dose equivalents at aircraft altitudes from these radiation fields and at providing the theoretical basis for experimental measurements for validation of these calculations for selected flights.

The work will be separated into three parts:

(a) the calculation of the impinging external radiation fields at flight altitudes for all important components,

(b) the calculation of the resulting organ dose equivalents for all these components, and

(c) The calculation of the energy response of selected instruments to be used by the experimental partners (in particular for TEPC) for comparisons of measurements with calculations for selected flights.

To this purpose the data sets and assumptions on the incident cosmic ray fields (differential in direction and energy) will be analysed and existing computer codes for the transport of the incoming galactic and solar radiation fields in the atmosphere will be implemented and tested for this purpose in cooperation with the authors of these codes.

Other neutral and charged particle transport codes will then be used to calculate the response of tissue equivalent proportional counters to these components (for the validation studies foreseen). The organ dose equivalents in an antropormorphic phantom will be derived.

Particular emphasis will be given to the contributions of fast neutrons and heavy ions. The results will be compared to those obtained by the experimental partners in this project and those from the integrated American code (Friedberg et al). In addition, the usefulness of the concept 'dose' for the description of health hazards from rare events in biological cells as those from cosmic rays will be considered.

The final result of this work should be a validated, documented program for the calculation of organ dose equivalents as a function of longitude, latitude and altitude. This programme could be used for computation of passenger and/or air crew exposures for any specified flight coordinates by line integration over the three dimensional dose rate space to be derived in this project.

To make optimum use of different previous expertises needed in this study, it is intended to carry it out in cooperation with partners in the USA (K O'Brien and M D Wilson), at CERN (G R Stevenson) and University of Siegen (W Heinrich). Close cooperation with the experimental partners of the whole project is also essential.

4. DIAS - Cosmic Ray Section.

The investigation of the flux and composition of cosmic ray nuclei at high altitudes in the Earth's atmosphere will be undertaken by the Cosmic Ray Group at the Dublin Institute for Advanced Studies. This group will prepare detectors for flight and carry out processing and analysis in the post-flight period.

The measurements associated with particle identification will be performed on high powered optical microscopes suitable for observation in nuclear track detectors.

Calibration of the solid state nuclear track detectors will be carried out at specialised accelerator facilities in Europe and the United States and the exposed detector stacks returned to Dublin for analysis.

The information obtained from the analysis of detectors exposed to in-flight radiation will be combined with that from other partners in the project and used in the development of appropriate models.

5. Univ. Saarlandes - Institut für Biophysik.

Summary of the investigation under civil aviation exposure contract

The determination of absorbed dose and dose equivalent received in aircraft will be performed with the HANDI (Homburg Area Neutron Dosemeter) -System at altitudes of about 10-13 km and at different geomagnetic latitudes. The HANDI has been constructed as a portable radiation protection instrument for mixed neutron and photon radiation fields. It is based on a low pressure tissue equivalent proportional counter (TEPC) including the electronics for the pulse height analysis and related data processing. Using the methods of microdosimetry, the HANDI offers the possibility to determine the quality factor by measuring the energy deposition events and lineal energy spectra (LET). The knowledge of the lineal energy spectra provides a quantitative assessment for radiation quality of the field accounting for the relative photon and neutron dose components. In contrast to a conventional laboratory equipment used in irradiation experiments the HANDI is easy to operate, battery powered and of light weight.

In order to get further information on the cosmic radiation field on board measurements are planned with proportional counters made of tissue equivalent material, graphite and, possibly, metal (aluminium and iron). In addition the influence of wall thickness and simulated diameter (gas pressure) on the response of the detectors will be investigated.

For detailed analysis and comparison with calculations performed within this contract some measurements will be made with our laboratory measurement system providing a much high resolution of the lineal energy spectra (360 channels instead of 16 of the HANDI-system).

6. CERN - Radioprotection.

Radiation field measurements

Following two irradiations in 1991 and 1992 of various active and passive detectors in the stray radiation field around the beam facility meant also to be used in the EC project the group will analyse the problems that came up during these two runs and require improving. These relate in particular to the beam monitoring and the knowledge of the field distribution in the place where dosemeters are to be exposed. As far as beam monitoring is concerned we shall acquire the necessary equipment to assure a pulse by pulse monitoring of the intensity for normalising the results of all participants during the runs. For measuring the space distribution of the radiation we shall buy a scanner that can map the field. It is hoped that all the equipment will be prepared and tested ready for the run in June 1993. We are also looking into a rearrangement of the beam target and shielding geometry such that it should become possible to calculate the radiation field.

A12 Radiation measurement and instrumentation for individual and area dosimetry

Contract FI3P-CT920032 Dosimetry of beta and low-energy photon radiations.

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Total Contribution by the Commission: 240 KECU
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Description of research work

1. General scope.

Ultimate objectives of the project are to develop standard calibration facilities and to establish standardised measurement and calculation procedures for dosimetry of weakly penetrating radiations. This will lead to a more consistent and reproducible dosimetric practice for determining exposures to individuals throughout the EEC countries.
The proposed research programme is expected to bring about new knowledge and developments in the following main areas:

- identification and evaluation of influencing parameters important for the dosimetric characteristics of the radiation fields in designing standard beta calibration beams, e.g. source construction, surrounding shieldings, and beam-flattening filters.

- characterisation of beta radiation and the accompanying photon radiation (bremsstrahlung and characteristic X rays) fields in terms of dosimetric quantities as well as in terms of energy and angular spectra.

- investigation and development of measurement techniques and procedures for characterising beta radiation (with the accompanying photon radiations) fields and for individual monitoring for doses from these fields.

- establishment and characterisation of low-energy photon radiation calibration beams and characterisation of individual dosemeters for dosimetry of these fields.

The results of this study will lead to the realisation and characterisation of beta calibration facilities which conform to the specifications of ISO Series 2 references as well as low-energy photon calibration facilities and thereby facilitate calibration of personal dosemeters and monitoring instruments for weakly penetrating radiations. Expected results of the project are furthermore development of improved dosimetry techniques and procedures for characterising weakly penetrating radiations and for personal dosimetry in such fields. The study will comprise different irradiation conditions including exposure at some distance from the source as well as exposure from particulate sources in contact with the skin (hot particle dosimetry).

2. Description of the project

2.1. Development of beta calibration facilities

Under the previous contract beta calibration facilities have been established at laboratories of the participants and the radiation fields have been characterised in terms of directional dose equivalent, $H'(d;\alpha^{o})$, using extrapolation chambers as standard instrument. Together, these facilities constitute a broad spectrum of beta calibration fields which are available for all participants for further joint studies of the characteristics of beta radiation beams and of the responses of dosemeters and instruments to beta radiation exposures. A more specific knowledge of energy and

angular spectra of the radiation fields would enable a better understanding of responses of dosemeters and dose ratemeters. In addition to experimental analyses computations using a Monte Carlo code will therefore be applied for obtaining data on energy and angular spectra and for evaluating response data of dosemeters and dose ratemeters. In particular, computations are expected to be a helpful tool for the design of radiation fields with optimal dose homogeneity for calibration purposes. The data obtained from computations will be validated by experimental analyses of spectra and determination of dosemeter and instrument responses.

The project is aimed at identifying and evaluating influencing parameters important for the dosimetric characteristics of the beta radiation fields and using this information to define optimal designs of the irradiation facilities. Influencing parameters to be studied are:

- source construction: e.g. dimension, cover and backscatter material of the source. Three standard dimensions are used for this study: square, 40 mm x 40 mm, circular, 42 mm in diameter, and 10 mm in diameter "point sources", all commercially available from the Amersham company. Other sizes will also be included in the study, e.g. a ^{14}C source consisting of a 1 mm thick acrylic sheet with the the ^{14}C radioisotope incorporated in the plastic material.

- shielding: the influence of back scattering from surrounding shieldings will be studied, in particular for high-energy beta radiations.

- beam-flattening filters: e.g. dimension, material and position. Results of dose homogeneity studies of beta radiation beams indicate that the use of beam-flattening filters for large-area beta sources may be advisable for cases where high accuracy of the measurement is required.

- emergent spectra (both of beta radiations and the accompanying photon radiations): the influence of these spectra and their modifications by external absorbers on the measured dose rates and on the response characteristics of detectors used for monitoring weakly penetrating radiations.

- nuclide: In the previous contract project beta sources with maximum energies from 0.156 to 3.54 MeV have been involved. Since radiation protection also involves lower energies the nuclide ^{63}Ni with E_{Max}= 0.067 MeV will be included in the study, too.

2.2. Development and study of techniques and methods for characterising beta radiation fields and for individual monitoring for beta radiation doses.

Work will continue in developing and studying different dosimetry techniques for use for analyses of beta radiation fields and for use as dosemeters for beta dosimetry. The extrapolation chamber measurement method will be further refined and studied for absolute determination of beta dose rates. Solid state dosemeters will be studied for application as beta dosemeters for a number of different dosimetry purposes. Spectrometric methods will be used and studied for analyses of energy and angular distributions of beta radiation fields. Computation models will be developed and calculated data will be validated by experimental data.

Extrapolation chamber.

During the previous contract work important uncertainty sources of the extrapolation chamber measurement method have been identified and improvements of the evaluation procedure have been achieved. Further studies will be made aiming at optimising the evaluation procedure and the accuracy of measurement of the method. Results will be obtained through comparative measurements of $H'(d,\alpha^{\circ})$ among participants using different types of extrapolation chambers. The importance of dimension of collecting electrode and thickness of entrance window for the extrapolation chamber measurement method will be further studied.

Thin solid state detectors.

Solid state dosemeters intended for characterisation of beta radiation fields and for individual monitoring for skin doses from beta radiation should contain detectors with a small effective thickness capable of measuring $H'(d;\alpha^{\circ})$ nearly independently of energy and angle of incidence of the radiation. If photon radiation is involved the dosemeters should ideally show a response that is tissue-equivalent also for photon doses. Response data of thin TL detectors and TSEE detectors for exposures to beta radiations obtained under the previous contract show that it is possible to obtain solid state detectors with satisfactory response characteristics. Work will continue in developing and studying thin solid state detectors (e.g. TL, TSEE, Radiochromic dye film detectors) for facilitating the practical application of the detectors for a number of dosimetry purposes (e.g. dose homogeneity studies, hot particle dosimetry, individual monitoring).

Beta spectrometry.

A beta spectrometer based on a silicon semiconductor diode will be established and used for obtaining experimental analyses of energy and angular distributions of beta radiation fields. Information on beta energy spectra will furthermore be obtained from analyses of transmission curves. The experimental data will be supplemented by calculations.

2.3. Hot particle dosimetry.

Methods for the measurement and calculation of doses from radioactive particulates, particularly "hot particles" will be developed and validated. Measurement techniques will include extrapolation chamber, radiochromic dye films and TL dosimetry. Calculational methods will be Monte Carlo and semi-impirical computer codes.

Results obtained under the previous contract work using 100 μm ^{60}Co spheres showed some disagreement between calculations and measurements. This might be explained by the use of non-ideal source geometries which were not accounted for in the calculations. Future measurements will be done on a more strictly controlled ^{60}Co source design and using a higher energy beta emitter, ^{170}Tm. The project is aiming at recommending suitable systems for dosimetry measurements and calculations of small sources.

2.4. Low-energy photon dosimetry.

An irradiation facility, established at CEA Fontenay-aux-Roses to produce low-energy photons of energies in the range from 5 to 15 keV will be characterised in terms of $H'(d;\alpha^{o})$ using extrapolation chamber . The facility will be used by other participants for studying responses of dosemeters to low-energy photons.

The possibility of using the ^{55}Fe radionuclide for providing a 6 keV photon calibration beam will be investigated.

3. Contribution by each participant.

Under the previous contract a variety of beta calibration beams have been established at laboratories of the participants. These facilities will be used in the present project for joint studies of dosemeter responses and comparison of dosimetry techniques. An extensive intercomparison of extrapolation chambers for the measurement of dose rates from a low energy beta source will take place by circulating an extended ^{147}Pm area source between five laboratories of the participants.

Risø National Laboratory:

Objectives of this work are to establish and characterise standard calibration fields for weakly penetrating radiations, analyse and refine the extrapolation chamber measurement method and develop and characterise thin solid state detectors for dosimetry of weakly penetrating radiations.

Risø National Laboratory will coordinate the joint project.

Beta calibration beams from $^{106}Ru/^{106}Rh$, ^{147}Pm, ^{14}C, and ^{63}Ni extended area sources and the PTB/Buchler ($^{90}Sr/^{90}Y$, ^{204}Tl, ^{147}Pm) standard "point" sources will be available for the joint project. The study of beta calibration beams will concentrate on $^{106}Ru/^{106}Rh$, ^{14}C, and ^{63}Ni sources. Results from analyses of a $^{106}Ru/^{106}Rh$ source under the previous contract showed that the source contained a significant amount of a radioactivity with energies lower than that of $^{106}Ru/^{106}Rh$. The problem has been addressed to the producer of the source with the intention to aquire a new pure $^{106}Ru/^{106}Rh$ source. Studies of the new source will include characterisation in terms of $H'(d;\alpha^{o})$, dose homogeneity measurements and analyses of the influence of scattering from the surrounding walls for different source-to-wall distances. The radiation field from a ^{14}C area source consisting of a 1 mm thick acrylic sheet with the ^{14}C radioisotope incorporated in the material will be characterised in terms of $H'(d;\alpha^{o})$ and the dose homogeneity will be studied for different source sizes. The results obtained for this type of source ("infinitely thick") will be compared with data obtained from a thin source. Dose data will be obtained for a ^{63}Ni source by use of extrapolation chamber and the results will be compared with data obtained with TSEE detectors in collaboration with University of Giessen.

The laboratory will continue the study of the extrapolation chamber measurement method. The importance of using thin entrance windows of extrapolation chambers for the measurement of dose rates from low-energy beta emitters (e.g. ^{14}C and ^{63}Ni) will be studied by using different thicknesses.

The development and study of thin TL detectors with the aim of obtaining a highly sensitive thin tissue-equivalent detector suitable for skin dosimetry will continue. The work will concentrate on LiF:Mg,Cu,P. In particular the possibility of improving the annealing characteristics of the material will be investigated.
The possibility of using ^{55}Fe for providing a 6-keV photon radiation reference beam for calibration purposes will be investigated and responses of thin TL detectors/dosemeters to low-energy photon radiations will be studied by using this beam as well the low-energy photon calibration facility established in CEA-Fontenay-aux-Roses.

CEA, Fontenay-aux-Roses:

Objectives of the work are to establish and characterise standard calibration beams of beta and low-energy photon radiations, to study and refine the extrapolation chamber measurement method, and to establish and operate a beta spectrometer.

Beta calibration beams from ^{204}Tl, and ^{147}Pm extended area sources will be available for the joint project. Work will continue in characterising beta radiation fields in terms of $H'(d;\alpha^o)$ using extrapolation chamber.

The extrapolation chamber measurement method will be further studied aiming at drawing conclusions as to methods of evaluation of results and evaluation of uncertainties.

A beta spectrometer based on a silicon semiconductor diode will be established and used for obtaining experimental analyses of energy and angular distributions of beta radiation fields. In particular, the method might be helpful for studies of dose homogeneities of beta radiation fields and for analyses of the influence of beam-flattening filters. The experimental data will be validated by calculations by other members of the group.

Work will continue in establishing an automated computer-controlled facility for producing low-energy (5-15 keV) photon standard calibration fields. The beams will be characterised in terms of K_{air} and $H'(0.07;\alpha^o)$. The established calibration beams will be available for other participants for characterising solid state dosemeters for dosimetry of low-energy photon radiations.

NRPB, Chilton:

The overall objective of the project is to characterise absorbed dose to air and/or tissue in terms of emergent spectra from practical beta ray sources.

Beta sources that are used in practice emit complex spectra containing components made up of beta radiation, inherent photon radiation, and bremsstrahlung and characteristic X-rays generated in the source and covering materials. All these spectra

undergo changes as they travers intervening air between the source and measuring medium and throughout the depth of the medium itself. Furthermore, spectral distributions can be quite different depending on the orientation of the beam with respect to the measurement plane. Consequently, a knowledge of these spectra and their influence om the absorbed dose is of great importance for completely defining the radiation field used as a metrological standard. In addition, it is useful for making appropriate corrections and interpreting the results of investigations using such fields.

Experimental data will be provided which will supplement those already acquired during the development of Series 2 beta ray secondary standard ($^{90}Sr/^{90}Y$ extended area source in a specially designed holder) under the previous contract. The programme will be designed to complement any computational model that will be developed by other participants in the group.

The project will contribute: (1) to investigation of the emergent spectra (both of beta radiations and photon radiations) and their modifications by external absorbers including air and tissue and at interfaces such as tissue/bone, and to the determination of the effect due to different orientations of the radiation beam with respect to the measurement plane;(2) to the development of a methodology for computing absorbed dose to a medium such as tissue (on the surface, at a given depth and at interfaces) and for estimating the influence of the changing spectral distributions on absorbed dose.

Calibration beams established at the laboratory.i.e. $^{90}Sr/^{90}Y$, ^{204}Tl, ^{147}Pm, and ^{63}Ni extended area sources will be available for the joint project.

The laboratory will continue its work of characterisation and refinement of the extrapolation chamber measurement method.

CEA, Grenoble:

Objectives of the work of CEA Grenoble are to establish and characterise standard beta calibration fields, to study and refine the extrapolation chamber measurement method and to characterise thin TL detectors for application for beta dosimetry.

Beta calibration beams from $^{90}Sr/^{90}Y$, ^{204}Tl, and ^{147}Pm extended area sources and the PTB/Buchler standard "point" sources will be available for the joint project. Work will continue in characterising beta calibration fields in terms of $H'(d;\alpha^{o})$. Results will be obtained for a ^{204}Tl source using two different holder designs. Furthermore, determination of the residual maximum beta energy of the spectrum will be made for different calibration distances of the established beta sources.

The laboratory operates three different types of extrapolation chambers of which one can use different electrode sizes. Comparative studies will be made of the different chamber types and the influence of diameter of the collecting electrode will be further investigated.

TL detectors with small effective detector thicknesses will be studied for application for characterisation of beta radiation fields and for individual monitoring for beta radiation doses. The study will concentrate on thin, ultra-thin, and carbon-loaded TL detectors.

AEA Technology, Harwell:

The objective of the project is to simulate the beta-ray fields near standard sources, using the Monte Carlo electron-photon transport code EGS4. This will complement the experimental investigations by other members of the group.

The code will be used to calculate the dose rate at different distances and in different directions for large-area beta sources conforming to ISO Series 2 specifications. The dose rate will be calculated as a function of depth in tissue. The variation in energy deposition in a small volume will be calculated as a function of the displacement of this volume from the central axis. If the uniformity is found to be poor, the code will be used to design beam flattening filters for the sources. The nuclides to be considered are ^{63}Ni, ^{14}C, ^{147}Pm, ^{204}Tl, $^{90}Sr/^{90}Y$, and $^{106}Ru/^{106}Rh$. The code will be used to examine the extent to which the beta radiation fields may be perturbed by scatter from the surrounding shieldings.

Calculated responses of simulated extrapolation chambers of the dimensions used experimentally by other participants will be compared with the experimentally determined values.
Calculations will be made of the energy and angular distribution of electrons at different radii from the central axis and of the effect of these distributions on the sensitivity of detectors such as TLD and TSEE. This will enable the prediction of the beta sensitivity of these detectors under various experimental conditions.

University of Montpellier.

The overall objectives of the project are to study, develop and apply the TLD laser-heating technique for dosimetry of weakly penetrating radiations. The laser heating technique offers the possibility of using detectors with a thin effective thickness which is important for dosimetry of weakly penetrating radiations. Furthermore the technique operates with a fast reading which is valuable for dosimetry applications where many measurements are involved, e.g. dose mapping of radiation fields and individual monitoring. A fully automated computer-controlled laser-heated TLD reader will be used for this study.

The accuracy of large-size (200 mm x 200 mm) dosemeter plates for determining the dose homogeneity of beta radiation fields of different energies will be analysed. If satisfactory measurement accuracy can be obtained by the method it will useful for analysing dose homogeneities of the different beta radiation fields applied in the joint CEC project.

Thin film TL dosemeters based on $CaSO_4$ and Al_2O_3 deposited on kapton and suited for laser heating will be developed and studied for application for characterising beta radiation fields in terms of $H'(d;\alpha^o)$. Dosemeter responses will be measured for different beta energies and irradiation angles.

The possibility of using laser-heated thin film detectors for "hot particle" dosimetry will be investigated in collaboration with Nuclear Electric plc, Berkeley.
For application of the TLD laser-heating technique for individual monitoring appropriate dosemeter constructions will be developed and investigated. The development work will mainly be concentrated on the $CaSO_4$ and Al_2O_3 TL materials. However the possibility of using the technique for thin LiF dosemeters will also be studied. Important dosimetric characteristics of the dosemeters for application for individual monitoring will be investigated.

The response of various TL detectors to low-energy photons will be studied by using the irradiation facility established at Fontenay-aux-Roses to produce low-energy photons.

I. Physikalisches Institut, University of Giessen.

The overall objectives of the project are to study, develop and apply the TSEE dosimetry technique for dosimetry of weakly penetrating radiations. The combination of an extremely thin effective detector thickness and a high sensitivity of TSEE detectors make them attractive not only for individual routine monitoring but also for solving a number of specific dosimetry problems occurring in radiation protection, in particular where low-penetrating radiations are involved.

BeO thin film detectors produced by the Battelle Institute in Frankfurt have previously been studied for application for beta dosimetry. The characteristics of BeO thin film detecters produced by Staatliches Materialprüfungsamt in Dortmund will be thoroughly analysed to test whether they offer the same high reliability as the TSEE detectors previously produced by the Battelle Institute in Frankfurt. Parameters to be tested are: dose sensitivity, short and long term reproducibility of detectors stored in different atmospheres (normal laboratory air, dry air, saturated water vapour), and chemical surface composition (determined by means of SIMS = secondary ion mass spectroscopy).

In collaboration with other participants of the joint CEC project TSEE detectors will be used for analysing dose homogenities of beta radiation fields as well as for characterising low-energy beta radiation fields (e.g. ^{14}C and ^{63}Ni).

Work will continue on optimisation of badge constructions of TSEE detectors for individual monitoring in mixed beta/gamma radiation fields. In particular, influences of different cover materials on the dose response will be analysed; but also the importance of the back-up material of the BeO film will be studied. The under-response of the thin film detectors to photons observed around 100 keV may be caused by the graphite backup of the detectors and improvements might be obtainable by reducing the thickness of this substrate or by using additional backup by a material of higher atomic number.

The response of BeO thin film TSEE detectors to low-energy photons will be studied by using the irradiation facility established at Fontenay-aux-Roses to produce low-energy photons.

Nuclear Electric plc, Berkeley Technology Centre.

The objective of this project is to develop and validate methods for the measurement and calculation of doses from radioactive particulates, particularly "hot particles". Measurement techniques will include extrapolation chamber, radiochromic dye films and thermoluminescence dosimetry. Calculational methods will be Monte Carlo and semi-empirical computer codes such as VARSKIN.

The Berkeley dosimetry group will produce neutron activated radioactive particulate sources with spherical and cylindrical geometries of dimensions 0.1 -1 mm (hot particles). Dosimetry evaluations will be carried out using (i) and automated small-electrode scanning extrapolation chamber and (ii) image analysis read-out of radiochromic dye films. The radiochromic dye film analysis will be done in collaboration with Dr C. Soares at NIST, Washington DC using a scanning laser system. Low activity sources will be used for dosimetry inter-comparisons and high activity sources will also be available for in-vivo pig skin studies as part of separate CEC contract. The results of the dosimetry studies will be compared with the results of Monte-Carlo beta dosimetry codes and with other recently developed semi-empirical codes(e.g.VARSKIN 2) in order to provide advice on a suitable validated method of "hot particle" dose calculations.

Previous work on ^{60}Co sources will be repeated using a source holder with a strictly defined geometry. The studies will be extended to include ^{170}Tm. The results of these studies should clarify the origin of the current disagreement between measurements and calculations for ^{60}Co sources which are thought to arise from self absorption in the material surrounding the source. This should enable a methodology to be developed for the calculation of "hot particle" doses from various geometry sources using either a semi-empirical or Monte-Carlo code, whichever is found to be most appropriate.

The non-linear response of the extrapolation chamber to small "hot particle" sources will be further analysed in terms of a recently developed theoretical model in order to provide a defensible methodology for the continued use of this absolute dosimetry system.

Dosimetry studies will be extended to include laser readout of TL films at Montpellier (Gasiot). Calculations will be extended to include the particular geometries of the Grenoble extrapolation chamber and the radiochromic and TL film systems.

An extensive inter-comparison will be provided between the extrapolation chamber and radiochromic dye and TL film techniques in order to provide guidance on the most suitable dosimetry system for routine and research applications in "hot particle" monitoring.

The dosimetry studies described above will be carried out with the lowest activity sources possible in order to reduce personnel doses. Higher activity sources will be produced by prolonging neutron activation times and these will be available for in-vivo radiobiology studies; e.g.^{60}Co particles with dimensions form ~ 100 μm to 1 mm with activities up to about 10^7 Bq (~ 300 μCi).

A12 Radiation measurement and instrumentation for individual and area dosimetry.

Contract FI3P-CT920039 Development of instruments and methods for radiation protection dosimetry with the variance-covariance method.

Coordinator Univ. München
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Total Contribution by the Commission: 120 KECU
21 months 1/09/92 to 31/05/94

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Description of research work:

The recent reductions of dose limits in radiation protection require increased accuracy and practicability of dosimetric measurements and methods. Beyond the determination of absorbed dose and dose rate there is an added need for determinations of radiation quality. Microdosimetric measurements are required to determine dose averaged lineal energy, which is the measurable parameter to derive the quality factor in radiation protection.

In previous projects the theoretical basis and the practical implementation of the variance-covariance method have been developed. This method has two major advantages. It is neither restricted to the very low dose rates that are required for measurements of single event spectra nor is it limited to constant dose rates, as they must prevail for the conventional variance method.

Tissue equivalent twin proportional counters and ionization chambers have been developed for application in variance-covariance measurements. These instruments (using electrometers in the integrating or in the current measuring mode) have demonstrated the advantages of the method and they are now ready to be adapted for radiation protection practice.

While the variance covariance method admits high dose rates, there are frequent situations where, in personal dosimetry or in area monitoring, especially in neutron fields detectors are insufficiently sensitive. Rossi multi-element proportional counters will be developed for routine applications with the variance-covariance method to meet this difficulty.

The variance-covariance method will be used for quality assurance and dose optimization in diagnostic and therapeutic beams. Twin ionization chambers for this special purpose will be developed and will be integrated into portable instrumentation. The variance-covariance method will be used to improve the dosimetry protocol for high energy photons.

Universität München (D)

Development of a Multi-Element Tissue Equivalent Proportional Counter for Variance-Covariance Measurements in Radiation Protection

The reduction of the dose limits in radiation protection and the revision of the quality factors necessitates more stringent measurements. Proportional counters are well suited for the purpose, but for low dose rate, especially of neutrons, they are insufficiently sensitive.

The aim of this project is the development of improved detectors that cover a wide range of dose rates. The use of the variance-covariance method permits operation at high dose rates, to make the instrument suitable for low dose rates one needs to employ a multi-element proportional counter. Rossi and coworkers have developed the prototype of such a detector in 1988. Their detector consisted of 296 small counter elements enclosed in a common housing, sized 1 inch in diameter and 2 inch

in height. The detector exhibited excellent performance but was too sophisticated in design for the production in substantial numbers.

The first objective of the project is the simplification of the multi-element counter, to permit its introduction into radiation protection practice. The increase in practicability may lessen somewhat the performance characteristics; but an adequate compromise will be sought.

The first task in the project is the testing of the counter characteristics of individual elements to optimize their size, the gas pressure, wire diameter and related factors. The second step will be concerned with a 37 channel detector in a hexagonal structure, and with the analogous design with subdivided channels. A third step will be aimed at the development of procedures for injection moulding to reduce element sizes and to permit ready production of the detectors.

The channels of the multi-element detector will be separated into two interdispersed groups, to form a twin detector. This avoids possible artefacts when the detector moves through non-uniform radiation fields. The signal processing according to the variance-covariance method will permit the evaluation of dose rate and, through the dose averaged lineal energy, the quality factor in difficult situations, e.g. in the vicinity of accelerators, or in radiation protection measurements for air travel and for manned space activities.

Computational studies will accompany the detector development and will support the interpretation of experimental data.

Univ. Århus-Hospital

Application of the Variance-Covariance Method to characterize the Quality of X-Ray Beams in Diagnostic Radiology.

In previous research sponsored by the European Community a cylindrical tissue equivalent detector pair has been developed suitable for variance-covariance measurements in diagnostic x-ray beams of different qualities. The variance-covariance method is a measuring technique designed especially for measurements of the dose averaged lineal energy, $\bar{y}_D$, in fluctuating radiation beams. The detector pair has been used in the ionization mode, in two separate integrators during simultaneous intervals the charge integration is performed in two programmable electrometers connected to a computer controlled circuit with relay switches. The integration and handling of the collected data are computer controlled. The application of the ionization mode is preferable in high dose rate beams where there is no need for gas multiplication.

The twin detector system will be improved, in order to avoid recognized shortcomings and to extend their applicability. The collecting electrodes will be made of tissue equivalent plastics, and the aluminum vacuum housing will be removed, to reduce the size and to make measurements possible in the low kV-range (used in mammography), in tissue equivalent phantoms, and in special geometrics (computed tomography). For such measurements a redesign of the detector pair is necessary that

will comprise all of the experience gained in the earlier phase of the project. Phantom measurements in therapeutic electron and x-ray beams will be performed as well, and will be compared to other conventional dosimetric methods.

Further simplification of the equipment by developing a portable system with compact electronics for practical use will be performed. This is regarded as an important step for the practical use of microdosimetric techniques in the three areas, characterisation of therapeutic and diagnostic beams, quality assurance in radiology, and dose optimization in the medical use of ionizing radiations.

A12 Radiation measurement and instrumentation for individual and area dosimetry.

Contract FI3P-CT920045 The use of microdosimetric methods for determination of dose equivalent quantities and of basic data for dosimetry.

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Total Contribution by the Commission: 410 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

1. State of the art

Combined dose-LET measurements remain a basic approach for determination of the dose equivalent particularly in radiation environments with mixed fields. Dose LET spectra, or a suitable approximation, allow a discrimination to be made between the photon and neutron components and provide information on the energy components of the spectra, their contribution to the absorbed dose and the mean quality factor. The tissue equivalent proportional counters (TEPC) is a typical application of this approach. Inherent limitations due to the approximation of the LET by the microdosimetric quantity lineal energy and the field perturbation by the detector may be overcome with sufficient accuracy by means of appropriate computational techniques and calibration procedures. This allows the dose-LET approach to be extended to personal dosimetry for which no satisfactory technique exists in mixed fields.

The TEPC is an established means for area monitoring but requires improvement since its response is still considerably too low for neutrons with low and intermediate energies. Different methods have been shown to improve the TEPC response but, especially around 100 keV, their use is limited and further improvements require lower pressures to be used compared with the common technique. Furthermore, the regular production of reliable TEPC sensors remains a major problem for their implementation in practical radiation protection, especially if low pressures are desired.

The extension of the TEPC technique to personal dosimeters is possible in principle but requires a different counter design due to the requirements of high sensitivity and small size. With this aim, a multi-cellular geometry has recently been developed in CEA/S.DOS. Modelling studies for TEPCs have significantly progressed and are currently used to support such developments. For proportional counters at low pressures in general, particularly using complex geometries, the determination of the conditions where the detector is operating in the true proportional mode with optimum gas gain characteristics is indeed very important and calculations are required to solve this problem.

As an alternative for electronic personal dosimeters, the development of a semi-conductor dosimeter as a microdosimetric device is promising. Preliminary investigations are being performed within the previous contract to determine the technical specification of the chips which can be used for this application, i.e. with sufficiently small sensitive volumes (μm dimensions). First prototypes are expected to be produced soon and tested in neutron and photon fields.

In order to assess the applicability of the two personal dosimeters, a comparison with the TEPC as transfer instrument will be particularly useful. Computer codes combining radiation transport and energy deposition calculations have been developed, and can be used to simulate the response of the different dosimeters taking into account the recommended operational quantities.

Due to new demands for survey dosimetry in environments with high-energy neutron components (space, civil aviation, high energy accelerators), the characterisation of high energy neutron fields is becoming necessary. Few facilities are available for such investigations. Experience was gained in neutron fields from 20 to 70 MeV at Villigen (Ch) showing the need for investigating neutron fluence measuring techniques at these energies. Required basic data such as kerma factors for carbon and TE plastic, were determined. Comparable data for oxygen are still missing. Furthermore neutron fields with reduced low energy contamination are desired. Preliminary investigations have shown the suitability of the neutron facility at Louvain-la-Neuve.

2. Objectives and expected achievement:

The main aim of this project is to develop detector systems for radiation protection dosimetry in photon-neutron fields. The techniques investigated are based on the measurement of absorbed dose and LET to determine the dose equivalent. The response of the dosimeters will be assessed with regard to the operational dose equivalent quantities. Problems related to the optimisation of the dosimeters and their calibration in reference and realistic fields will be investigated, intending to improve various methods for radiation detection and instrumentation modelling.

Concerning low pressure TEPC, as the conventional dose-LET approach, the response will be studied and improved for neutrons in the low and intermediate energy range by operation at lower pressures and improvement of the counter design. The project includes the application of a TEPC especially designed and built in an appropriate phantom to be used as a transfer instrument for the calibration of different dose equivalent meters.

New techniques will be developed using the dose-LET approach, particularly for their application to personal dosimetry. The methods, i.e. a semi-conductor detector as microdosimetric device (SCD) and a multi-cellular low pressure tissue equivalent proportional counter (MCPC), are proposed as electronic personal dosimeters with the advantages of combining dose and radiation quality information being closely related to the well established TEPC method.

The calibration of the dosimeters and their implementation for radiation protection is required to investigate their response and specifications taking into account the new recommendations by the ICRP. The project is aimed at investigating these problems with respect to ambient and personal dosimetry using appropriate computational methods and comparing the response of the dosimeters in well characterised fields. To meet the requirements for radiation protection dosimetry in environments with high energy radiation, the programme includes the characterisation of reference neutron fields in the high energy range.

Expected achievements are:

TEPC: to achieve a dose equivalent response of ±30 % for neutrons with low and intermediate energies, to construct reliable TEPC sensors operating at low pressure, to specify the TEPC in realistic fields, to design and achieve a prototype of the transfer instrument

SCD, MCPC: to complete the feasibility studies of the instruments and to compare prototypes with the TEPC in the same conditions, to assess their applicability as personal dosimeters

Modelling of detector response: to improve electrical discharge modelling calculations (basic data, numerical methods) for different types of proportional counters (geometry, filling gas) and to perform computer simulations for the operational detectors under development, to improve protocols for using combined radiation transport and energy deposition computer codes

Dose equivalent response: to calibrate and inter compare the dosimeters in reference and realistic radiation fields, to assess the response of the instruments and their technical specifications with regard to the new recommendations by the ICRP

High energy neutron fields: to extend characterisation methods (fluence determination, monitoring, dosimetry) to neutron energies up to 90 MeV, to provide high energy neutron facilities for dosimetry investigations, to complete kerma factor measurements above 20MeV for oxygen.

3. Distribution of tasks and collaborative links between participants:

1. Toulouse:

-to complete and improve the determination of the electron collision cross sections and swarm parameters required for the various organic vapours and gas mixtures used in gas ionisation devices employed for radiation dosimetry (5)(6)
- to determine the electrical and gas gain characteristics of low pressure proportional chambers with different geometries, filled with various gases, and at different pressures in order to support optimisation studies for the gas detectors developed within the project for radiation protection dosimetry (5)(6), reference and research instruments (2)

2. PTB:

- to investigate the response of dose equivalent meters as function of neutron energy relative to the operational quantities used in radiation protection (5)(3)(6)
- to optimise a dose equivalent transfer instrument based on a TEPC for neutron energies below 20MeV (1)(5)
- to characterise neutron beams between 20 and 90 MeV to be used as reference fields to investigate the response of operational dose equivalent meters (5) to be used in environments with high energy neutron components, to determine the required basic physical data (oxygen kerma, charged particle cross sections) (5)

3. TNO:

- calibration of mixed fields using ionisation chamber techniques (2)(6) - assessment of operational dose equivalent quantities with regard to the detection methods used within the project for ambient and individual dosimetry in mixed fields (1)(6)

4. KFA:

- to develop semiconductor detectors (SCDs) as microdosimetric devices for the dosimetry of mixed fields and to optimise the SCD for its application as personal dosimeter, in particular to relate its response to that of the TEPC (5)(6) using combined radiation transport and energy deposition calculations (2)(3)
- to investigate the effect of the new ICRP recommendations on operational quantities in radiation protection using radiation transport calculations in humanoid phantoms (2)(3)
- to investigate biological response functions as a contribution to the judgement of quality factors

5. Homburg:

- to improve low-pressure proportional counter (TEPC) techniques (2), in particular to provide reliable sensors for radiation protection and research instruments
- to improve the dose equivalent response of TEPCs for neutrons around 100 keV by implementing counters operating at low pressures (1)
- to investigate the response of TEPCs in realistic radiation fields (6) and to provide LET-spectra information to allow, in combination with spectrometry information, the comparison between the TEPC and other dosimeters (3)(6)

6. CEA/S.DOS:

- to investigate the applicability of an individual electronic dosimeter (4) based on a multicellular low-pressure proportional counter (MCPC) (1) in comparison with the other detectors (TEPC, SCD) (5)(3)(4)
- to provide well characterised simulated fields with realistic neutron spectra to investigate the response of the dosimeters developed within the project (5)(3)(4)

Finally, all participants belong to the Working Group 10 of Eurados.

4. Relation to other RTD projects:

The work within the project is directly related to the following programmes:

- Eurados Working Group 10 on "Basic Data and Characteristics of Gas Ionisation Devices"
- Contract FI3P-CT930072 on "Individual electronic neutron dosemeter"
- Contract FI3P-CT920039 on "Development of instruments and methods for radiation protection dosimetry with the variance-covariance method"

The calibration of the dosimeters in qualified fields will be performed in collaboration with :

- Eurados Working Group 7 on "Radiation spectrometry in working environments"
- Contract FI3P-CT920002 on "Realistic neutron calibration fields and related dosimetric quantities"

Information of interest will be provided to contribute to the progress of the research programmes:

- Contract FI3P-CT920041 on "Specification of radiation quality at the nanometer level"
- Contract Bi7 0051 F on "Dosimetry and spectrometry measurements of the leakage radiation fields from the Silene reactor with various shields".

Contribution of Toulouse and interaction with other partners

The conception of many gaseous proportional counters for radiation research has been for a long time mainly based on empirical considerations. One reason was that these detectors were known to operate rather well in most usual situations and that a fuller understanding of charge collection processes inside the cavity accounting for the specific structure and geometry of the detector was thought to be neither required nor important. This situation was in particular true for the PCs used in microdosimetry. However, the increasing applications of microdosimetry techniques and the demand for more performant PCs has strongly contributed to modifying this point of view and a better understanding of the multiplication process inside these detectors appeared to be necessary. In this aim, numerical modelling may give very important information.

However, for modelling to be possible the knowledge of some basic data such as electron-molecule cross-sections is necessary. Unfortunately, information about these cross-sections is very poor in the organic vapours of interest for microdosimetric detectors.

First, we will determine the electron-molecule cross-section data for propane and isobutane and we will begin to investigate other organic vapours of interest in dosimetry and spectrometry. Secondly, we will use these cross-sections to study the behaviour of gaseous devices for low pressures and complex geometries.

Our work is intended to contribute towards an accurate and quantitative assessment of the characteristics of PCs operated at low gas pressures compared to those working in the conventional mode. Indeed at present, PCs used for most current applications in microdosimetry operate at the pressures corresponding to simulated sites of one to several micrometers in size. To reduce the effect of short -ange particles for low-energy radiation, lower pressures (at sites of several hundred nanometers) are desirable. For such low gas pressures, it is likely that the counters will no longer have to work in conventional proportional mode or at least will reach the limit of operation. To solve this critical problem the calculations will accurately determine the conditions for which the counters still operate in true proportional mode. Thus information is of direct interest for the optimisation of TEPCs for radiation protection dosimetry.

The calculations will be applied to different types of PCs used or developed by the other partners (Homburg, KFA, CEA/S.DOS). The work is carried out in close collaboration with working group 10 of Eurados.

Contribution of PTB and interaction with other partners

Within this CEC contract PTB has concentrated its activities in two different fields.

First, we are trying to push forward the set-up of a transfer device for the dose equivalent quantity H*(10), the quantity defined in a 30 cm diameter sphere at 10 mm depth. Calculational studies will be performed in order to find a combination of an especially designed tissue equivalent proportional counter (TEPC) and a phantom suited to measure the dose equivalent quantities for neutrons of thermal energies of up to 20 MeV. In particular, this transfer instrument should be useful to improve the calibration of individual dosimeters according to the new recommendations of the ICRP. The transfer device will be developed within a collaboration of the Universities of Homburg and Toulouse. The energy response of the transfer device will be studied in monoenergetic reference fields and later on in calibration fields with broad energy distributions such as from D2O moderated 252 Cf sources.

Secondly, due to the increasing number of people who might be exposed to high-energy neutrons (En > 20 MeV), calibration facilities and basic data will be required for radiation protection studies. The investigation and specification of high-energy neutron fields produced at the Paul Scherrer Institute (PSI), Switzerland, and at the University Louvain-la-Neuve (UCL), Belgium, will be extended up to neutron energies of 80 MeV. This work includes the improvement of various measuring techniques such as time-of-flight techniques with proportional counters and scintillation detectors, fluence determination with a proton recoil telescope and neutron monitoring with the 107, 109Ag(n,3/5n)105Ag activation cross section. PTB will make these reference fields available for the partners and other external user groups. The scientific program will be focused on the determination of fluence-to-kerma conversion factors for oxygen, zirconium and aluminium with proportional counters in close co-operation with the Universities of Homburg, Birmingham, U.K., and Madison, Wisconsin, USA. Especially the kerma ratios of carbon and oxygen are needed to reduce a major source of errors in the determination of dose equivalent quantities.

Contribution of TNO and interaction with other partners

The TNO Medical Biological laboratory (previously Institute of Applied Radio biology and Immunology) will be involved in dosimetry in mixed fields using ionisation chamber techniques, in radiation transport calculations using a Monte-Carlo code (MCNP), and in energy deposition/ion yield calculations using the Caswell/Coyne code.

An increasing number of persons might be exposed to neutrons with energies in excess of 20 MeV, e.g., in the vicinity of high energy accelerators used for physics studies or radiotherapy or inside aircraft at high altitudes. Therefore, information on basic data and the characteristics of dosimetry systems for this energy region will be required for radiation protection purposes. The irradiation facilities at the University of Louvain-la-Neuve are essential to explore basic data and detector characteristics of neutron dosimetry systems at energies in excess of 20 MeV. TNO can contribute to the characterisation of these neutron fields as planned for by PTB through measurements with ionisation chambers and Geiger-Müller counters to establish total absorbed doses and relative photon contributions to the dose at reference positions. These measurements will be carried out in close collaboration with PTB and UCL.

In addition to the experimental studies by other participants in the response of detectors inside various radiation fields in terms of recommended operational dose equivalent quantities, calculations will be performed by PTB, TNO and KFA. At TNO both the Monte Carlo neutron and photon transport code (MCNP-4) and the Caswell/Coyne (NIST) analytical code for calculation of energy deposition and ion yield in gaseous detectors are available. These codes can be used to improve the responses of ambient and personal dosimeters by simulating detectors used by other participants. The calculations will be performed complementary to the experimental investigations carried out with different detectors at the various reference fields. Finally, TNO will contribute to the activities of EURADOS Working Group 10, collaborating on improvement of basic data for neutron dosimetry and detector construction to which all participants in the project contribute. Emphasis will be placed on improvement of W-values for neutron dosimetry.

Contribution of KFA and interaction with other partners

The central topic of the contribution is the development of a semiconductor detector which is able to measure microdosimetric energy deposition spectra in mixed neutron and photon radiation fields. This requires the sensitive detector sites to have μm dimensions. In addition, the device must be able to handle charge impulses with a dynamic range of up to six decades. Two types of semiconductor device are taken into account. a) Commercial memory chips (Static Random Access Memory, SRAM), which consist of an array of transistor structures. Charge generated in the collector base junction of a transistor can be measured as a current pulse at the power terminals of the chip. b) A specially developed charge-coupled device (CCD). This detector consists of an array of charge-sensitive elements, called pixels. Charge generated in a pixel by radiation is stored until the pixel is read by external electronics. The inactive top layer in particular has to be very thin (about 30 nm), so that electrons and low-energy heavier charged particles, which are produced in a tissue equivalent cover, reach the sensitive volume. This is necessary for reasonable photon sensitivity and ensures that the neutron energy range which will be covered is sufficiently broad. The photon sensitivity is increased in addition by decreasing the noise level through the implantation of necessary preamplifiers on the detector chip.

The semiconductor chip will be embedded into tissue-equivalent material thickness to ensure charged particle equilibrium at the inner surface. Additional covers of tissue-equivalent material to moderate the radiation field may be necessary to modify and optimise the response of the detector.

The influence of the tissue-silicon interface and the interpretation of the measured spectra should be aided by calculations. For this purpose, neutron cross-sections in a silicon and silicon dioxide will be obtained from suitable nuclear data libraries (e.g. ENDF/B VI) and prepared for neutron transport calculations. For charged particle transport and energy deposition calculations suitable stopping powers must be provided. Existing programs like the transport code ANISN and the microdosimetric NESLES code will be initially adapted. The data libraries will, however, also be prepared for MCNP and possible extensions provided for charged particle transport.

Of major importance for all partners in the proposed co-ordinated project, as well as for regulatory bodies in the EC, is a knowledge of the consequences of the new ICRP 60 recommendation on effective dose and the operational quantity ambient dose equivalent H*(d) and individual dose equivalent H'(d). Using the transport code MCNP and a humanoid phantom, which was adapted for this code in Jülich, values for equivalent dose using the concept of radiation weighting factors, as well as the quality factor concept will be provided and discussed. The ambient dose equivalent will be calculated in the ICRU-sphere using the new quality factor proposed by ICRP.

Contribution of Homburg and interaction with other partners

The work of Homburg is concentrated on improving:

- the performance of TEPC based devices for radiation protection dosimetry

- the methods used to analyse lineal energy spectra measured by TEPCs with respect to the physical properties of radiation fields and in relation with response of the dosimeters developed within the present program.

Although the use of a TEPC is a well established method for dose measurements, there are still large difficulties to construct reliable detectors which meet the requirements of radiation protection work. Based on the experience gained in constructing prototypes, the aim of the work is the optimisation of the geometry, the electrical and mechanical properties of TEPCs in order to achieve an appropriate sensitivity, stability and robustness.

The dose response of TEPCs is nearly independent of neutron energy over a large range of energies. However, for intermediate neutron energies (about 10 keV up to several hundred keV) TEPCs underestimate the dose due to the effect of short range particles. Several methods to improve this situation are known but a significant amelioration without changing the good response for higher neutron energies requires the simulated diameter to be reduced to several hundred nanometers instead of the 2mm commonly used. The project is to assess the operational characteristics of TEPCs operated at such low pressures.

Due to their spectral capabilities, TEPCs are particularly well suited to investigate complex fields encountered in practice. However, the interpretation of the measurements is in general limited by insufficient knowledge of neutron fluence data. Therefore, a part of the work in Homburg consists in combining neutron-photon spectrometry studies (in collaboration with Working Group 7 of Eurados) with TEPC measurements in mixed field simulating practical environments.

Beside these activities Homburg will continue to give expertises of radiation fields used for research work based on TEPC measurements. Furthermore, the work will contribute towards a better knowledge of cavity principles and supply basic physical data required.

Interaction with other groups of the contract

Toulouse

The optimisation of the design and the electrical properties of the TEPC is attended by calculations simulating the electrical field distribution and the electrical discharge processes using the parameters of real detectors.

PTB

Calculations of the dose equivalent response in dependence on different parameters (e.g. simulated diameter, wall thickness) and measurements in monoenergetic reference neutron fields.

CEA

Simultaneous measurements of neutron fluence and microdosimetric spectra in radiation fields simulating realistic neutron spectra.

CEA/KFA

Comparison of measurements under the same conditions with newly developed personal dosimeters with such of "classical" TEPCs.

Contribution of CEA/S.DOS and interaction with other partners

The CEA/SDOS will contribute to the project by proposing its expert advice in operational radiation protection. The emphasis will be placed on the comparison of the operational characteristics of the instruments developed within the contract for ambient and individual dosimetry taking into account the incidence of new operational quantities and recommendations in radiation protection.

The CEA/S.DOS is developing a proportional counter to be used as personal dosimeter. The application of TEPCs to personal dosimeters met large difficulties mainly due to too low a sensitivity. This led the CEA/S.DOS to investigate completely new designs based a multi-cellular geometry to increase the radiating surface of the cathode. Conceptually, the detector is made of a drift zone constituted of cylindrical holes in the tissue-equivalent material. The counter "walls" are made of parallel layers of A-150 tissue-equivalent plastic separated by a thickness of polarised resistive tissue-equivalent plastic which defines the drift regions. The walls are separated by glass plates on which are etched parallel anodic wires to determine the gas amplification region. The design of the detector was first made using modelling calculations in close collaboration with Toulouse. A prototype with a single channel and a needle as anode was built and successfully tested. The current work consists successively in using a single wire as anode with one channel, a linear row of channels with a single wire and to finally achieve the first complete prototype within the first project year to be tested in various radiation fields. The present project will give the opportunity to test the multi-cellular counter prototype of personal dosimeter in comparison with the TEPC and the SCD to allow judgement of the applicability of such a technique for radiation protection use.

The CEA/S.DOS will contribute towards the organisation of inter comparison experiments insofar as sufficient progress has been achieved in building the new instruments. Such inter comparison be organised in particular in mixed fields which simulate practical situations to investigate the response and capabilities of the dosimeters in work conditions.

A12 Radiation measurement and instrumentation for individual and area dosimetry

Contract FI3P-CT930072 Individual electronic neutron dosemeter.

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Total Contribution by the Commission: 160 KECU
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Description of research work

This project of contract deals with individual neutron dosimetry in accordance with ICRP 60 recommandations, with the aim to study then compare two dosemeters which are different of the classical ones used thanks to etched track detectors:

- electronic device
- superheated drops (bubble) detector.

Two parts can be considered:

- a separate study of theoritical conceiving and realization of the two devices in order to improve them
- an experimental comparizon of the two devices thanks to joint irradiations. The aim of this comparizon is to determine the possibilities of dosemeters taking into account ICRP 60 recommandations and realistic conditions of personal dosimetry.

A - PRESENTATION OF THE DOSEMETERS

1 - Electronic dosemeter (figure nº 1)

It is composed of:

a - A Sensor with 2 electronic detectors with and without a front neutron converter. The converter gives α, Li particles and protons. The characteristics of the converter and electronic detector are separately determined, then together studied in the final configuration.

b - An electronic device which counts pulses from the sensor, applies differential method, calculates dose equivalent and/or dose equivalent rate.

2 - Bubble detector (figure nº 2)

The superheated drop (bubble) detector consists of a uniform dispersion of superheated halocarbon drops suspended in an immiscible tissue equivalent gel.

Its operation relies on the same principle as the bubble chamber: upon exposure to neutrons, the high LET recoils from nuclear interactions nucleate the boiling of the superheated drops, generating macroscopic vapor bubbles. Contrary to the bubble chamber, which works in pulsed mode, undergoing rapid superheating and repressurizing cycles, the drops may be kept in their superheated state for long periods of time since they are trapped inside a gel, a container with perfectly smooth walls.

Each drop is therefore a miniature, continously sensitive bubble chamber, and its boiling does not initiate that of adjacent drops.

The volume of vapor or the pressure pulses generated when drops vaporize may be recorded and serve as a measure of neutron irradiation.

The characteristics of these acoustic pulses have been studied and electronic circuity for their detection has been designed.

B - OBJECTIVES

1 - Realization of a real time dosemeter by the means of the electronic device.

Thanks to electronic possibilities this dosemeter will be able to calculate and give the "EXPOSURE STORY" of any people, then compare it to the ICRP recommandations.

In these conditions it is possible to use the "principle of optimization in radioprotection", thanks to this real time dosemeter.

2 - Superheated drop detectors for dosemeter with immediate reading.

An exhaustive analysis of the combined temperature (pressure) and energy dependance is necessary. This should lead to the identification of the best combination of superheated material (or mixture of materials) and thermodynamic conditions in order to realize an accurate dosemeter.

3 - Optimization of the characteristics of the two types of dosemeter.

The dosemeter will be characterized through:

* the response to direct neutron beams
* the neutron energy fields in which the response is sufficient to give valuable informations
* the limits of detection
  - lowest dose equivalent which is measurable
  - lowest dose equivalent rate which is measurable
* the on phantom response
* uncertainties on the measured dose equivalent and rate.

4 - Comparisons of the two types of dosemeters.

It is of great interest to test in the same conditions the various devices in order to give performances of each one and compare each other.

C - DESCRIPTION OF THE CONTRACT WORK

Various tasks are planed to realize the proposed objectives. We can summarize them as follows:

1 - New converter used with improved electronic device

THESSALONIKI and LIMOGES will mainly contribute to this task.

Results on converter (^{6}LiF evaporated on a thick polyehtylene foil) and on electronic device (PIPS diodes and differential method) already obtained during the CEC contract BI 700 20 C are used in order to improve by calculations and experiments the dosemeter.

2 - Electronic detector

BARCELONA and CEA-FAR will mainly contribute to this task.

The realization of a new electronic detector for the dosemeter is proposed: two diodes in back to back position on the same wafer in order to improve the differential method on electronic device.

3 - Superheated drops (bubble) detector

DCMN-PISA is concerned with this task.

This new method needs to fully identify and explore its possibilities in order to develop dosemeters providing accurate and immediate reading of ambient and individual dose equivalent over a wide range of neutron energies. Expected achievements is the realisation of prototype instrumentation based on this technology.

4 - Calculation: γ contribution, phantom influence

CEA-FAR, BARCELONA, LIMOGES will mainly contribute to this task.

Previous studies have shown that the γ contribution must be reduced. Furthermore the phantom influence is very important. For these two objectives calculations are necessary and will be realize thanks EGS 4 code (for γ) and PNEDIOD code (for neutrons).

5 - A first approach of multiaera dosemeter

LIMOGES is concerned with this task.

In order to well know the γ contribution and the contribution of neutrons within different energy ranges a multi aera dosemeter will be considered thanks to microelectronic technics.

6 - Tests and comparizons in various neutron-γ fields

The five laboratories are concerned with this task.

It will be realized thanks to joint irradiations (monoenergetic beams in Bruyêres-Le-Chatel) using a phantom. In these conditions some informations (theoretical and experimental results) will be obtained and will allow us to discuss and compare the properties of each dosemeter and finally prospect on the use of the devices in the frame of personal dosimetry.

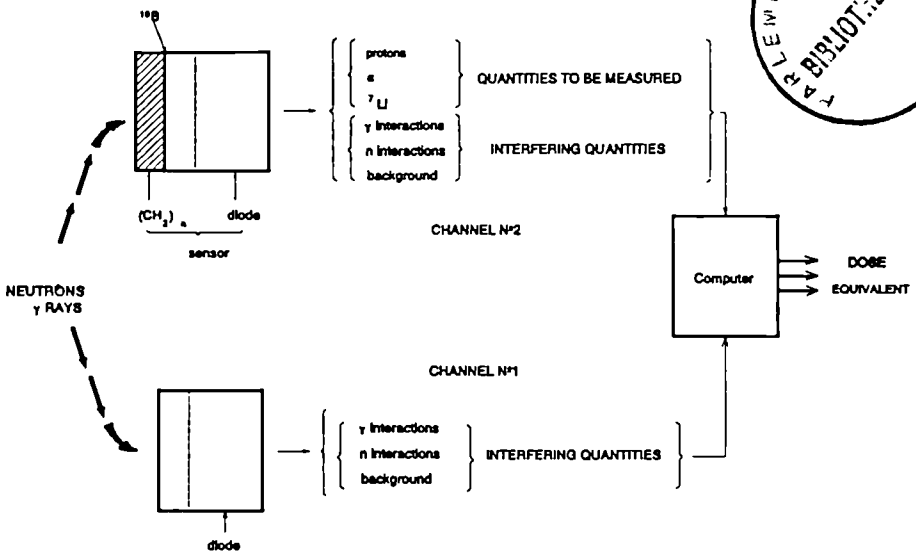

Figure N°1: Schematic diagram of an electronic dosemeter

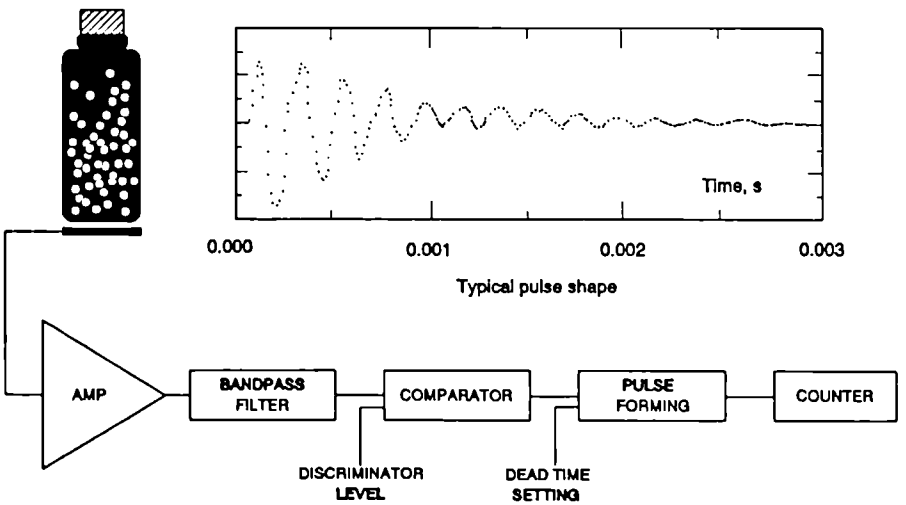

Figure N°2: Electronic circuity for superheat drop (bubble) detectors

Contribution of LEPOFI - LIMOGES

The 1990-1992 contract has already given and will give informations on the diode based neutron dosemeter. The structure of the sensor {implanted Boron 10 polyethylene + PIPS diode} has to be fully tested especially in mixed fields on a phantom. Thanks to a computer code already developped at LEPOFI, simulations on phantom of the response will be done and experimental tests realized on a device taking into account the results obtained on the γ contribution (metal parts). The limitations already pointed out for energy (intermediary neutrons), sensitivity (low dose equivalent and dose equivalent rate) will be measured in various fields on a phantom during joint irradiations.

In accordance with new ICRP 60 recommandations an increase of the sensitivity is possible by different ways:

- electronic improvements (choice of electronic components with optimized characteristics),

- converter improvements which will be possible thanks to Thessaloniki staff collaboration.

The limitations already mentionned, the need to realize a personal dosemeter, the necessity to obtain a reproducible sensor at low cost lead the LEPOFI staff to propose new sensors based on multidetector system (some hundreds or thousands of integrated diodes or transistors) with or without integrated converters. Thanks to Monte-Carlo and EGS4 computer code and microelectronic CAO tools, conceiving and simulating will begin at LEPOFI in collaboration with LAAS (Toulouse): this microelectronic technology laboratory agrees to collaborate with LEPOFI. The main goal for this device is to improve the response through various factors: discrimination of the γ response, sensitivity for low energy neutrons (intermediary neutrons) in order to reduce as much as possible the lowest detectable dose equivalent and improve the response in low dose equivalent rate beams.

In the beginnning of this contract an investigator of UAB-Barcelona will stay at LEPOFI in order to prepare the work on new diodes which will take place in Barcelona taking into account conclusions obtained on the present device. Role and contribution of

Contribution of the AUT (Thessaloniki)

The research on the development of an individual neutron dosemeter is one of the activities of the laboratory of AUT. During CEC contract BI 700 20C this laboratory had the opportunity of improving its experience on neutron dosimetry based on SSNTD technique. The main improvement in this topic was the use of a new radiator system. This system developed in AUT consist of a polyethylene (n, p) radiator and a ^{6}LiF (n, a) converter. The ^{6}LiF is evaporated on a thick polyethylen radiator (0.5 - 1.0 cm) which acts as a proton recoil source as well as a neutron moderator, shifting a part of the neutron spectrum toward lower energies.

The main advantages of the proposed radiator system are: the alpha particles from the reaction ^{6}Li (n, a) ^{3}H have energies $\geq$2 MeV and the ^{3}H $\geq$2.7 MeV. These particles can be easily measured with a suitable detector. The proton recoils from PE radiator can also be counted at the same time with the alpha particles and ^{3}H.

An important property is the increase of the response especialy for the low energy neutrons (from thermal up to 2 MeV) because of the contribution of the PE moderator. At higher energies the response is also good as we have seen using SSNTD.

Our calculations based on the kinematics of the reactions with fast neutron, as well as experimental results using SSNTD, show that the angular dependence could be improved.

During the BI-7-0020C contract the laboraotry has also had the opprotunity to collaborate with LEPOFI on the development of the electronic neutron dosemeter. One member of AUT, Dr. Elias SAVVIDIS, spent 6 months in studying the system and developping an original experiment with it at Limoges.

During the first part of the 1992-94 contract a microcomputer based pulse analysis system will be implemented. The experience of Dr. SAVVIDIS thanks to his stay at LEPOFI, will allow a fast calibration and test of the system.

Our radiator will be used with the electronic analyser. We propose in a first step to test the radiator with PIPS diodes (now used in LEPOFI and in SDOS CEA), to measure neutrons of several sources and in mixed fields (neutrons and γ). The measurements will be done mainly on a phantom, and the properties of the device compared with the results obtained during the contract which is now in progress.

In a second step the converter, modified if necessary, could be used with other electronic detectors studied in the frame of this contract.

Contribution of SDOS/CEA

The results, obtained up to now, are very encouraging but some difficulties have occured particularly concerning the neutron response at low energy. Indeed, for low energy neutrons, protons, coming from the converter have a very low probability to be detected by the diode. The overall sensitivity of the dosemeter decreases with neutron energy and background signal, due to intrinsic photon-diode interactions, increases proportionately. This effect becomes very significant between a few keV and 200 keV. One, of the objectives proposed by the SDOS, is to improve the current situation in the framework of the new ICRP recomendations.

The underestimation of neutron doses at low energies is likely to be the greastest difficulty encountered. This factor is a crucial point in an effective use: the dosemeter response diminishes by a factor 3 between 500 keV and 2 MeV. This effect is due to the fact that recoil protons of low energies, generated in the converter layer, are absorbed inside the collecting electrode and the diode dead zone before reaching the depleted zone.

The SDOS contribution to the research project will relate mainly to the following items:

1- to perform irradiation tests at different neutron energies to avoid or to minimize undesirable effects ; several methods are envisaged:

- to reduce the thickness of both the dead zone and the collecting electrode,
- to design specific specially shaped converters in order to increase the probability of generating low energy recoil protons, based for example, on the use of microchanelled polypropylene converting layer.

2- calculations on the γ-contribution thanks to the ESG 4 code which is implanted in our laboratory.

3- to perform intercomparisons between the dosemeter being studied and different usual or routine neutron dosemeters in will known neutron radiation fields. These intercomparisons will be performed successively with and without a fantom taking into account all constraints imposed by radiation protection practice.

Radiation fields used for intercomparison will be successively monochromatic neutron beams from an accelerator provided with specific targets.

Contribution of the UAB (BARCELONA)

Our group will contribute to the project by participating in the intercomparison of the electronic dosemeter proposed in the project with the track etch dosemeters set up by our group, irradiated under the same conditions.

The intercomparisons will allow us, in the first place, to develop a personal neutron dosemeter with a known energy and angle response based on the plastic track detector and its associated reading device.

These intercomparisons will also allow, at the same time, the verification by our group of a Monte Carlo simulation of the neutron interaction. This simulation is already working (CENDOS 91) and is very easily adaptable to different dosemeter configurations and to various exposure and etching conditions.

At the same time, the transfer to our laboratory of the achievements of the other two groups that use electronic dosemeters would be of great importance and utility for our country. In order to facilitate this transfer we plan to send an investigator to stay in the Limoges Laboratory.

I want to emphasize that the Spanish National Microelectronics Center is located in our University. This fact is of a great importance as it will allow to develop new aspects of this project, for instance: the search of other electronic detectors, as new diodes. We propose to work on this way.

Furthermore the EGS 4 code used in our laboratory allows us to work in collaboration with SDOS CEA in order to study γ contribution on new devices using a phantom.

Contribution of DCMN - PISA

The approach DCMN-Pisa suggests is based on the use of dispersions of superheated liquide, a technique we have contributed to develop through a long standing collaboration with its inventor (Prof. R.E. Apfel of Yale University).

The proposed research aims at the analysis of several superheated materials, which appear to be interesting potential detectors, in terms of their pressure and temperature dependent energy response, in order to create both a device with flat dose equivalent response and a spectrometer based on variable energy thresholds. In fact, while some materials are sensitive to thermal neutrons, others present a sharp threshold response at various energies. In both cases the response may be modified, either to achieve a better agreement with the recommended fluence to dose equivalent conversion factor or to vary the response threshold and thus sweep and analyse neutron spectra.

In either case, the data acquisition is based on the acoustic counting of the nucleation events and the accurate control of temperature and pressure conditions of the sensitive material.

The approach is new and problematic. When the pressure is varied, for instance, great care must be taken not to increase the concentration of gas dissolved in the detector, since this would decrease the energy threshold by increasing the effective degree of superheat. Our past experience is broad, though, and we are confident it would help us to proceed effectively. Upon completion of the prototype instrumentation, measurements will be first performed with monoenergetic neutrons and high energy γ rays. Irradiations will mainly take place at PTB - Braunschweig and INFN - Legnaro and with joint irradiations in the framework of this contract.

Once the characteristics of each material are tested, a study will be first performed on their suitability for dosimetry applications through a Monte-Carlo simulation of the response at those energies which are not experimentally available, and a microdosimetric analysis for a better understanding of the role each chemical element plays in the detection mechanism.

Next, unfolding techniques will be studied for the use in neutron spectrometry of the materials with a suitable threshold response.

Again, the expertise provided by INFN in the field of microdosimetry and that of PTB in the field of spectrometry will be fundamental.

A14 Assessment of internal exposure.

Contract FI3P-CT920048 Assessment of internal dose from plutonium and other radionuclides using stable isotope tracer techniques in man.

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Total Contribution by the Commission: 250 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

General:

The global objective of this project is to evaluate the use of stable isotopes as tracers in metabolic investigations in humans to meet the accepted and continuing need for more realistic biokinetic data of radionuclides of relevance in radiation protection. The transfer of radionuclides into the human body via the food chain will be investigated by experimental studies in man, with particular attention to the reliability and variability of transfer parameters under realistic conditions. Additional investigations on the internal distribution and excretion patterns will improve the metabolic and dosimetric models and consequently the dose assessments of internal exposure.

The metabolic behaviour of some radionuclides will be studied by substituting the radioactive isotopes by stable isotopes of the same element as tracers. For plutonium and other highly toxic radioelements, where no stable isotopes exist, stable analogues will be used in metabolic studies. The use of selected elements of the lanthanide series as surrogates for actinides in stable element tracer studies in human volunteers could provide valuable information which is not otherwise obtainable by planned systematic investigations.

Therefore, as a prerequisite step of applying this technique to the actinides, comparative biochemical and animal studies must prove that the stable element exhibits essentially the same kinetics in vivo as the actinide it is to model. Animal studies with radioactive hafnium isotopes as tracers have shown that hafnium is a promising candidate as an analogue for plutonium. The results obtained so far show that the protein binding and the metabolic behaviour of hafnium are very similar to that of plutonium. This suggests that under carefully defined conditions hafnium can be used as an analogue for plutonium in metabolic studies and in the investigation of interactions with biochemical ligands in cells and tissues. Moreover, the use of stable hafnium isotopes may permit metabolic studies in humans. Similarly, lanthanide series elements may also act as analogues for gastrointestinal absorption of actinides. Stable isotopes of barium might be sucessfully employed to mimic the metabolic behaviour of radium in man, as well as to provide additional information on the radioisotopes of barium in humans.

The proposed work involves four major tasks:

1. Development and validation of methods.

- Development of techniques for determination of stable isotopes of hafnium, neodynium, zirconium, and molybdenum in biological samples (Harwell, Milan, Frankfurt).
- Exchange of biological samples for the optimization of analytical methods and of samples from in vivo studies to evaluate the reliability of biokinetic parameters as determined by different measuring techniques (Harwell, Milan, Frankfurt).

- Use of computer-based and other methods of speciation analysis to provide detailed validation of the use of hafnium, europium and gadolinium as surrogates for plutonium, americium and curium for studies of humans (Cardiff).
- Evaluation of interactions of the lanthanide elements with the iron-transport protein transferrin and estimation of the formation constants for the relevant actinide/lanthanide-protein complex (Cardiff).

2. Metabolic investigations.

- Assessment of the metabolism of barium and neodynium in man by ad-ministration of stable isotopes by ingestion and by intravenous injection as analogues of radium and actinides respectively (Harwell).
- Evaluation of the effect of speciation and fasting on fractional gut uptake of barium and neodynium in man as analogues of radium and the actinides respectively (Harwell).
- Animal studies on the intestinal uptake of hafnium, zirconium and cerium (Milan, Frankfurt).
- Double isotope studies in animals involving injection and ingestion of radioactive plutonium and hafnium isotopes and simultaneous administration of stable isotopes of hafnium and neodynium (Harwell).
- Pilot studies on the intestinal uptake of hafnium in humans (Milan, Frankfurt).
- Assessment of intestinal uptake of molybdenum in man and evaluation of the possibility of developing counter measures designed to reduce intestinal molybdenum absorption in emergency situations (Frankfurt, Milan).

3. Improvement of metabolic models.

- Review of the available data on the biodistribution and biokinetics of transuranium and lanthanide elements from the point of view of their speciation and fractionation will help to assess the importance of any differences with respect to the interpretation of human data or the extrapolation of animal data to humans (Cardiff).
- The biokinetic data obtained in humans will be used to improve the metabolic and dosimetric models for the elements investigated (All).

4. Dose assessments.

- On the basis of the obtained data and models, new calculations of internal dose will be performed for these elements (Frankfurt).

Contribution of GSF - Frankfurt

The group of GSF at Frankfurt will provide biokinetic data in humans for several radionuclides which are of relevance in radiation protection and for which the current status of knowledge is poor. This will be achieved by metabolic studies in man with stable isotopes as tracers. The data obtained will be used to prove and modify the metabolic

and dosimetric models for these elements and to perform new dose calculations for the incorporation of the respective radionuclides.

The transfer of molybdenum into the human body via the food chain will be investigated using stable molybdenum isotopes as tracers to evaluate the reliability and variability of transfer parameters under realistic conditions. Particular efforts will be made to evaluate the possibility of developing counter measures designed to reduce the intestinal absorption of radioisotopes of molybdenum in emergency situations. The metabolic behaviour of plutonium in the human body will be investigated using stable hafnium as surrogate. Analytical techniques for the determination of hafnium concentrations and of isotope ratios in biological samples will be developed. Biological materials with defined hafnium contents and suitable for metabolic investigations must be developed before they can be administered to humans. After validation of the methods in animal studies, this technique will be applied to evaluate intestinal uptake of hafnium in human volunteers. On the basis of the obtained data and improved metabolic models, new calculations of internal dose will be performed for these elements.

Contribution of the University of Milan - Physics Laboratory

The group at the University of Milan will develop and optimize methods for the quantitative determination of hafnium and zirconium in biological samples by means of nuclear activation induced by charged particles and/or thermal neutrons. The main facilities employed will be the cyclotron of the JRC-Ispra and the LENA Triga2 reactor of the Universita di Pavia. For each element under study a series of preliminary measurements is necessary: yield functions, linearity response, limits of detection, control of interferences etc. A new irradiation chamber to activate up to 40 samples with a cyclotron is under completion: this device will allow the measurement of short living radioisotopes.

Applying this technique the intestinal uptake of cerium, zirconium, and hafnium in animals will be studied by means of radioactive and stable isotopes as tracers. The results of such investigations carried out for molybdenum have shown the validity of the whole procedure. Therefore, intestinal uptake of molybdenum in man will be evaluated by means of stable isotopes. Additionally, the possibilities of reduction of the intestinal absorption of molybdenum in man by adequate counter measures will be investigated.

Contribution of the University of Wales - College of Cardiff

The assessment of human health risks following environmental or accidental exposure to radionuclides requires a sound knowledge of the age-dependent biokinetics of the elements in the whole population. While some limited, direct studies of actinide biokinetics are possible in human volunteers, for ethical and legal reasons they are not permissible in children or in general members of the adult population. The use of selected elements of the lanthanide series as surrogates for actinides in stable element tracer studies in human volunteers could provide valuable information which is not otherwise obtainable by planned systematic investgations. Previous studies have suggested that hafnium may be an acceptable analogue for plutonium and that the lanthanides europium and gadolinium could act as models for americium and curium. However, the

use of such surrogates requires careful validation. The purpose of the proposed studies is to evaluate, using computer, and other methods for speciation analysis, the interactions of the actinide-lanthanide pairs, hafnium-plutonium, europium-americium and gadolinium-curium with proteins and other biological important ligands in mammalian blood and tissues, in order to provide a detailed validation of the use of lanthanides as surrogates for actinides. Special importance will be attached to interactions with the iron transport protein transferrin, which functions as the principal carrier of plutonium, and may be trivalent actinides and lanthanides, in plasma, and attempts will be made to extend and to refine the estimations of the binding constants for these metals to transferrin.

The published data for the biodistribution of the actinide and lanthanide elements in animals and humans, will be re-evaluated from the viewpoint of their chemical speciation and fractionation in order to assess the extent to which any differences in organ uptake or retention may reflect inter-element or inter-species variations which could be important in relation to the interpretation of human data, or the extrapolation of animal data to humans.

Contribution of UKAEA - Harwell Laboratory

Harwell Laboratory has many decades of collective experience in the field of metal uptake and metabolism studies in humans and animals using radioactive or stable tracers. These tracers have been used to assess the uptake of heavy metals and radionuclides, and to study the metabolism of essential trace elements. Data from these studies have been applied in the fields of radiological and environmental protection and food safety, as well as in biomedical applications.

In this project the metabolism of barium and neodynium in man will be assessed by administration of stable isotopes by ingestion and by intravenous injection as analogues of radium and the actinides respectively. The work on the effect of speciation and fasting on fractional gut uptake of these elements in man will be continued. Analytical methods are developed for determination of hafnium concentrations and isotope ratios in biological samples using ICP-MS. This will include measurements of hafnium in blood, urine, faeces and diet from normal UK residents. Additionally, animal studies are conducted involving injection and ingestion of radioactive ^{237}Pu and ^{175}Hf and of stable isotopes ^{176}Hf and ^{143}Nd at the same time. This will allow the validation of analytical techniques for hafnium measurements, and yield information on the metabolic behaviour of neodynium and hafnium, relative to plutonium.

A14 Assessment of internal exposure.

Contract FI3P-CT920060 Inhalation and ingestion of radionuclides.

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Total Contribution by the Commission: 150 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The objectives of the project are to develop and implement more comprehensive and realistic models for the evaluation of organ and effective doses due to intakes of radionuclides by workers and the general public including doses to the embryo and fetus due to intakes of radionuclides by the mother, and to address the more important uncertainties in the biokinetic and dosimetric models used in radiation protection for evaluating doses to organs from intakes of radionuclides.

The biokinetic and dosimetric models used in ICRP Publication 30 are in need of updating, and extending in scope. Consideration has to be given to providing not only either "conservative" or "realistic" estimates of doses, but also uncertainties in the models and their parameters, and measures of variability. In recognition of this, the ICRP has several Task Groups working in this area, developing revised models for the respiratory tract; developing age-dependent models, including the embryo and fetus; and revising reference man. Account has also to be taken of the revised recommendations of ICRP Publication 60. This programme will eventually lead to a full revision of ICRP Publication 30.

The ICRP Task Group on Age-Dependent Doses is reviewing biokinetic models for about 30 elements and assistance will continue to be given on reviewing the literature and carrying out dose calculations. Biokinetic models for the remaining approximately 50 elements will be re-evaluated. It is planned to develop generic models for chemically similar elements such as the lanthanides as it has been done for the alkaline earths and the actinides in ICRP Publication 56.

A thorough review of the literature is an essential part of the development of internal dosimetry models as well as the determination of the reliability of models. This becomes increasing demanding as both the scope of the problem and the database expand. A novel systematic approach is used within this project, based on a fulltext retrieval system.

In a similar way, the ICRP GI tract model will be reviewed. The present model is independent of age, and, apart from the f_1 value, of chemical form. The ICRP Publication 60 recommendations assign higher tissue weighting factors to GI tract components, and hence the GI tract model has a greater influence on doses and ALIs. For these reasons, and for improved bioassay modelling, a more realistic model is required. A review of the literature relating to GI tract biokinetics will be conducted, and alternative models implemented. The sensitivity of doses and ALIs to model structure and parameters, including age dependence, will be investigated, and an appropriate model structure and parameters selected.

Additionally to the biokinetic models, work has to be done concerning the dosimetric models. Due to the new weighting factors of ICRP Publication 60, SEE values are needed for the oesophagus. SEE values for additional organs are needed in nuclear medicine for dose assessments of radiopharmaceuticals. Such values will be developed within this project.

Further work will be carried out on the calculation of doses to the embryo and fetus. This presents special problems because of the rapid growth and differentiation of the embryo/fetus and because the placenta acts as a potentially selective barrier between it and the mother's circulation. For this purpose, new biokinetic and dosimetric models will be developed which will take into account those problems.

As these new biokinetic and dosimetric models are developed, new software is required to implement them. This may well require different approaches to be taken, because of the greater complexity of the proposed models. The new respiratory tract model specifies the calculation of doses to target cells in each region, rather than mean lung dose. Models for other organs, particularly skeleton, are becoming more physiologically realistic, in order to provide a generic approach that can be extended to other elements, and other ages, for which there is little information. This involves recyling of activity from source organs to blood. Account has also to be taken of advances in computer technology, and the consequent expectations of users. There are thus requirements both for programs able to calculate doses rapidly for a very large number of radionuclides, and/or variations in parameters, for example for assessments of the consequences of environmental releases or uncertainty analyses; and also for user-friendly PC-compatible programs for health physicists and scientists to investigate specific situations. Quality assurance of such software is becoming increasingly demanding.

Work will continue on the implementation of new biokinetic and dosimetric models. Software will be written in conjunction with the development of the models to test their implications and applicability. As the models are adopted, programs will be produced to apply them to the wide range of radionuclides and subject types required in practice. This will include quantifying uncertainty and variability in dose per unit intake values.

Programs will be written for both mainframe computers, to produce compilations of dose coefficients for a very wide range of radionuclides and parameters such as age, and for personal computers with a user-friendly menue-driven interface. The development of complementary codes will enable intercomparisons to be conducted for essential quality assurance.

Much of the work is closely linked to the development of ICRP models and recommendations relating to the dosimetry of internally deposited radionuclides. This is a particularly active area at present and will continue to be for the next several years, with the revision of the respiratory tract model, reference man, biokinetic models for individual elements (in conjunction with age-dependent models), and possible new models for bone and GI tract. These will lead to a full revision of ICRP Publication 30. Several of the participants are full or corresponding members of the Task Groups of ICRP Committee 2, and their contributions (and that of their colleagues) to the work of these Task Groups are

are part of the work within this project: Drs. Kendall and Noßke to the ICRP Task Group on Age-Dependent Dosimetry and Dose per Unit Intake for Members of the Public, AGDOS and to the ICRP Task Group on Dose Calculations, and Professor Taylor, as a member of ICRP Committee 2 itself, contributes to several Task Groups, especially to AGDOS.

There are, of course, complementary efforts elsewhere, especially in the United States of America, largely funded through the US Department of Energy (USDOE). The participants liase with their counterparts through a variety of mechanisms, such as the ICRP Task Groups mentioned above, direct individual contacts and visits, and participation at international conferences.

Direct collaborative links have existed between various participating organisations for some time, and these will continue. For example, BfS and NRPB have carried out intercomparisons of their compilations of dose coefficients. This will be also done with the ITRI-TNO group. Also there is a cooperation of BfS and NRPB with UWCC concerning the revision of ICRP Pulication 30.

The benefits to be gained from collaboration are to promote the interactions noted above required for model development; to avoid unnecessary duplication; to conduct intercomparisons where useful for quality control, notably in results of programs to calculate dose coefficients.

Contribution of the Federal Office for Radiation Protection (BfS)
Institute for Radiation Hygiene (ISH)

The work of the preceding research project had been the collection, compilation in a data base, and the assessment of biokinetic data for cobalt and thorium according to uniform criteria, together with the identification of those biokinetic parameters which are of primary importance for the calculation of doses or ALIs. This work will be continued with further elements (e.g. polonium). This work will be done in close collaboration with the ICRP Task Group on Age-dependent Doses.

Besides this the techniques developed during the preceding project will be applied to the examination of the gastrointestinal tract model. Compared to ICRP Publication 26 the new recommendations of ICRP Publication 60 result in a greater influence of the doses to the gastrointestinal tract on the effective dose. Additionally, the effective dose will be the only quantity determining the ALI according to the new dose limitation concept of ICRP Publication 60. The current ICRP model of the gastrointestinal tract is a four-compartment-model which is independent of age, the element and the chemical form considered (except the f_1 value of the gastrointestinal absorption). The influence of alternative models and element- and age-specific data on doses and ALIs will be determined.

The current dose calculation program of the ISH will be changed to allow age-dependent dose calculations for more general biokinetic models. The results will be checked with those of the former program as well as with results of NRPB and ITRI-TNO.

Contribution of the National Radiological Protection Board (NRPB)

The work planned is largely in support of ICRP Committee 2. It is carried out in particularly close collaboration with Drs. Noßke (ISH) and Taylor (UWCC) who are also directly involved with the work of Committee 2 but also with ITRI-TNO.

A major part of the work involves age dependent internal dosimetry calculations. This covers both age dependent biokinetics and also age dependent specific effective energies. The work is largely in support of Committee 2's development of the ICRP Publication 56 series, for which intercomparisons between the participating laboratories are planned. Attention will be concentrated on nuclides of practical importance.

Different laboratories have developed different methodologies for calculating doses from internal emitters. It is proposed that the ICRP Task Group on Dose Calculations should review the various methods and assumptions with a view to harmonising the different approaches. This will require careful discussion between the participating organisations since, on the one hand, a common approach will make far more readily interpreted results but, on the other hand, a diversity of approach allows for independent quality control.

It is becoming increasingly clear that investigations should be carried out into the uncertainty and variability in dose coefficients. This frequently calls for very careful analysis of rather fragmentary data and must be carried out on a case by case basis. Investigations of uncertainty and variability are especially relevant to a number of recently proposed biokinetic models which attempt to follow detailed physiological processes. Such models tend to involve many more parameters than the traditional ICRP 30 type equivalent and it is the rule rather than the exception that experimental data do not allow all of the parameters to be firmly established.

The calculation of doses to the fetus is a topic of growing importance. This involves both a review and refinement of methodology and also the application of established methods to new nuclides. Particular areas which require attention are:

a) discrimination by the placenta against specific nuclides at different stages of fetal development
b) uptake by fetal organs of different materials as a function of time
c) the relative weight to attach to irradiation of fetal organs at the various stages of differentiation and development.

Contribution of the University of Wales College of Cardiff
School of Chemistry and Applied Chemistry (UWCC)

The 1990 Recommendations of the International Commission on Radiological Protection [ICRP Publication 60] recommended changes in tissue weighting factors, new approaches to dose calculation and a reduction in the dose limits for workers.

These necessitate the recalculation of the Annual Limits on Intake and other secondary limits for a very wide range of radionuclides. For these new calculations the biokinetic models for every element must be reviewed to ensure that they are based on the best available information.

The work will be carried out in close collaboration with ICRP Committee 2; the Bundesamt für Strahlenschutz (DE) and the National Radiological Protection Board (GB). It involves a critical review of the biokinetic models for the individual elements presented in ICRP Publication 30 and their revision in the light of new, especially human, information. Approximately 30 elements are being considered by a Task Group of the ICRP which is concerned with age-dependent dosimetry, leaving some fifty elements to be re-evaluated in the present contract. All the biokinetic models will be reviewed and where necessary revised and improved in the light of new information and/or a better understanding of the biological, biochemical and chemical considerations. For chemically related elements, for example the lanthanides, attempts will be made to develop generic models.

The new, or newly re-confirmed biokinetic models, will be used in the revision of ICRP Publication 30 and in the updating of dose calculation programmes such as the EULEP Programme DOSELIB.

Contribution of TNO-Institute of Applied Radiobiology and Immunology (ITRI-TNO)

Compartmental models for the biokinetic behaviour of inhaled, ingested or injected radionuclides can be characterised as large and stiff systems (i.e. number of compartments in the order of magnitude of 100 with parameter values of widely different magnitudes). These systems are not easily evaluated numerically. It is therefore proposed to develop a new mathematical method based on the reformulation of the biokinetic processes in terms of difference equations of a discrete or stochastic nature. The methods will be tested by recalculation of various accumulated activities after inhalation, ingestion or injection of some typical radionuclides published by the ICRP and the ISH for both instantaneous and time varying intakes.

Cumulated activities in organs depend on various factors such as retention and coupling to other organs. These factors can be considered to be age-dependent. In order to calculate doses and dose limits for persons different from the adult male, it is therefore important to have the means to introduce different biokinetic parameters into the compartmental models. This has to be performed in a well documented and easy way and can best be implemented by means of menu-oriented interactive codes, such as previously developed at our institute for the ICRP-30 computations in the form of PCDOSE. By means of such codes it will, e.g., be possible to study the influence of newly obtained experimental data on the behaviour of respiratory tract tissues and to evaluate the effect of different gastro-intestinal absorption fractions. Close collaboration with the NRPB and other participants in the contract will be of major importance in this respect.

It might be of interest for the future to extend the modelling in such a way that also non-linear behaviour such as, for instance, saturation could be simulated. In this way, it will eventually be possible to gain insight into phenomena which are, until

now poorly understood (e.g., the saturation of the thyroid with iodide).

An interactive software system for ICRP-30 computations which can be implemented on a personal computer has been developed and is currently being tested. By means of this software system it is possible to compute dose and intake limits for persons with a physique different from reference man and to study the influence of newly proposed weighting and risk factors. However, the system relies heavily on the ICRP-30 models and the computation of dose after injection of a radiopharmaceutical cannot be readily performed. A more general software system will be developed by which various intake routes can be studied. It has to be noted that for such a system new specific effective energy (SEE) data have to be obtained. To that end the Monte Carlo Neutron Photon transport code (MCNP) will be used to obtain SEE data for an extended set of source and target organs.

Organ doses, equivalent doses and intake limits obtained by means of the proposed models and techniques have to be compared with data reported by the ICRP and ISH. In this way, the influence of improved biokinetic models on dose and dose limits can be evaluated.

A14 Assessment of internal exposure.

Contract FI3P-CT920064a Inhalation and ingestion of radionuclides.

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Total Contribution by the Commission: 280 KECU
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Description of research work

Introduction

Inhalation is the main route of intake of radionuclides for workers and, through the inhalation of radon decay products, for the general public. Ingestion is often the main route of intake for the public following environmental releases of radioactivity. Knowledge of the doses to different body organs following intakes of radionuclides is therefore an essential component in the control of occupational exposures. Similar data are needed in assessing the consequences of accidental and routine releases from nuclear installations, the estimation of doses from weapons fall-out, and in nuclear medicine. There is thus a continuing requirement for more realistic internal dosimetry models to improve radiation protection standards, ensure that measures taken are cost-effective, allay public fears, and answer informed criticism. These needs are particularly relevant to the European Community because of the extent of its existing nuclear power industry, including nuclear fuel reprocessing; future involvement in decommissioning; its requirement that committed doses from workers' intakes be recorded; and the growth in public concern since the Chernobyl accident.

This project is an extension and development of part of the previous co-ordinated project "Inhalation and Ingestion of Radionuclides" (Bi6-0347b) carried out within the CEC/NRPB Association Agreement, by four of the organisations listed above (01-04). The new project focuses more closely on the inhalation route of intake. Two new partners have joined it, extending its scope to the application of models in particular situations, and including the determination of relevant site-specific parameters.

Objectives

The objectives of the project are to address the more important uncertainties in the models used in radiation protection for evaluating doses to internal organs from intakes of radionuclides, by inhalation and ingestion; to develop and implement more comprehensive and realistic models for relating exposures and intakes to organ doses and monitoring results for workers, and for calculating the distribution of doses among the general population. The main emphasis is on inhalation, and on experimental studies aimed at providing basic data required to improve models.

The project also includes studies on the characteristics of the environmental radioactivity resulting from the nuclear weapon accident at Palomares, Spain in 1966. Major objectives of this part of the project are to provide an experimental basis for improving the assessment of internal dose from this contamination, and a rational basis for assessing internal doses from contaminated dusts present in other accidentally contaminated environments.

State of the art

The biokinetic and dosimetric models used in ICRP Publication 30 are in need of updating, and extending in scope. There is a need to provide not only either "conservative" or "realistic" estimates of doses, but also uncertainties in the models and their parameters, measures of variability, and modifying factors for sub-groups of

the population. In recognition of this, the ICRP has several Task Groups working in this area, developing revised, and age-dependent, models for the respiratory tract, bone, and GI tract; models for the biokinetics of individual elements; and a revision of reference man. This programme will lead to a full revision of ICRP Publication 30.

As these models are developed, new software is required to implement them. This may well require different approaches to be taken, because of the greater complexity of the proposed models. The new respiratory tract model specifies the calculation of doses to target cells in each region, rather than mean lung dose. Models for other organs, particularly skeleton, are becoming more physiologically realistic, involving remodelling and recycling. Account has also to be taken of advances in computer technology, and the consequent expectations of users. There are thus requirements for user-friendly personal computer (PC) compatible programs for health physicists and scientists to investigate specific situations. Similarly there are requirements for more sophisticated models for interpreting bioassay data. These need to be consistent with, and preferably unified with the models used to calculate dose coefficients.

Because of the importance of the inhalation route for the radiological protection of workers and the general public, following both controlled and accidental releases of radioactivity, the deposition and clearance of inhaled materials have been extensively studied. Significant advances have been made, resulting in the current revision of the ICRP respiratory tract model, but important gaps in knowledge remain, some of which have been highlighted by the ICRP Task Group. Most human inhalation studies have been conducted on small groups of healthy men. There are few data on submicron or hygroscopic particles. There are major uncertainties about clearance kinetics in each region of the respiratory tract, but most notably in the bronchial airways, which have also been identified as being of particular radiosensitivity.

Scientific and Technical Description

Much of the work is closely linked to the development of ICRP models and recommendations relating to the dosimetry of internally deposited radionuclides. Several of the participants are members of the Task Groups of ICRP Committee 2, and their contributions (and that of their colleagues) to the work of these Task Groups are included in this project.

Work will continue on the implementation of new biokinetic and dosimetric models. Software will be written in conjunction with the development of the models to test their implications and applicability. As the models are adopted, programs will be produced to apply them to the wide range of radionuclides and subject types required in practice. Further consideration will be given to the treatment of radioactive decay products produced within the body: the selection of appropriate biokinetic parameters and the feasibility of using different parameters for each member of the decay chain.

In the previous project considerable effort went towards development of the new ICRP respiratory tract model. In the next period the emphasis will shift towards its implementation, and the development of software to apply it more widely. Work will continue on the specification of parameters describing the rate of absorption to blood for a range of compounds of practical importance. The PC software developed previously will be modified to take account of further changes to the model, and

extended to treat age-dependence, radioactive decay products, and in particular inhalation of radon and thoron and their decay products.

As in the previous project, considerable effort will go into volunteer studies to quantify parameters for the human respiratory tract.

Further measurements will be made of the physiological parameters needed for dosimetric modelling, and their variations between individuals. Since the nasal passage is generally a more effective particle filter than the mouth, the distribution of air between the two will be measured, at graded levels of exercise and in both adults and children. Respiratory function parameters will be compared between Caucasians, Africans and Asiatics.

Deposition studies will focus on submicron particles, providing data needed to assess doses from inhalation of radon daughters and environmental particles; and on intersubject differences due to age, sex, race, and health status. Measurements of regional deposition made using the ultrafine, monodisperse, inert ^{111}In-oxide particles recently developed will be extended to smokers and subjects with lung disease. Total respiratory tract deposition of particles will be compared between Caucasians, Africans and Asiatics, using techniques established in the previous project.

Mechanisms that determine the patterns of flow and deposition in human airway structures will be investigated, principally by means of bolus dispersion: analysis of the concentration profile in exhaled air, following inhalation of aerosol as a narrow 'bolus'. Measurements will be extended to physical models of airways and to subjects with various lung diseases, and compared with theoretical predictions.

Particle clearance and radionuclide uptake from each part of the respiratory tract will be quantified, with emphasis on the sites and duration of long-term retention. A study will be conducted to determine the effects of particle size and breathing pattern on the clearance of particles from both the anterior and posterior nasal passages. Alveolar clearance studies will continue, with magnetometric measurements of Fe_3O_4 particles inhaled by healthy subjects (smokers and non-smokers), to obtain standard data for long-term retention, magnetic relaxation, intracellular viscosity, and macrophage mobility as indicators of the status of the defence system of the human lungs. The investigations will be extended to subjects with lung diseases associated with macrophage dysfunction.

Investigations of the clearance of particles from the human tracheo-bronchial region will continue, with emphasis on clarifying the extent and duration of the slow phase of clearance, and its dependence on factors such as particle size and site of deposition within the tracheo-bronchial tree. Subjects will inhale radiolabelled particles as a small bolus at a selected point in the breathing cycle. Aerosols of ^{99m}Tc-labelled Fe_2O_3 particles of various sizes will be used, and regional deposition and retention followed using both lung scintillation counting and gamma camera imaging. Sub-micron ^{111}In-oxide particles will be used to complement previous studies with larger particles. Particles will be labelled with a longer-lived radionuclide such as ^{51}Cr to measure the retention time of the slow phase. When these studies enable the site of deposition of the bolus to be predicted with confidence, the technique will be used to measure clearance rates at different lung depths.

To complement these phenomenological human studies, investigations of the mechanisms involved in the delayed tracheo-bronchial clearance of particles will continue. The fraction of the airway surface covered by ciliated cells will be measured: studies on rat trachea will be extended to other species. It has been proposed that the retained particles may be taken up by macrophages resident on the airway surfaces. These cells will be harvested and their properties compared with those of alveolar macrophages.

Studies will continue in which, following inhalation or intra-tracheal injection, the amounts of material on the tracheal surface and in the tracheal wall are measured. Further studies will utilise micro-injection into alveoli to address the key issue of the extent of delayed bronchial clearance of particles deposited in alveoli.

The biokinetic behaviour of radioactive materials taken into the body by inhalation or ingestion in practical situations may well differ substantially from that predicted using the default parameters specified in general models such as those recommended by the ICRP. They may also differ from the behaviour of the simple compounds normally used in laboratory studies, both *in vivo* and *in vitro*. It is recommended by the ICRP Task Group revising the respiratory tract model that material-specific data should be used where available, and the model is structured to facilitate the incorporation of such data.

Two of the partners in this project are collaborating in a study to investigate the biokinetics of radionuclides associated with environmental contamination. Few opportunities exist for studying the behaviour of actinides after contamination of the environment. As a consequence of the aeroplane accident over Palomares in 1966, however, persons living in the area were contaminated with ^{239}Pu and ^{241}Am, and the soil is still contaminated with these actinides.

One aim of this study is to provide an experimental basis for improving the assessment of internal dose from environmental radioactivity in the Palomares area. Initially, *in vitro* dissolution measurements will be used to identify the likely variation in the characteristics of absorption from lungs to blood of ^{239}Pu and ^{241}Am in the respirable fraction derived from different soil types and particle size distributions. The biokinetics of these actinides in those fractions showing the greatest differences in solubility will then be investigated in rats after their deposition in the lungs or gastrointestinal tract.

The information gained will be used to assess the appropriate doses per unit intake and to predict the behaviour of ^{239}Pu and ^{241}Am in humans using the protocols associated with the new ICRP respiratory tract model. These predictions will be of value for improving interpretation of chest monitoring and bioassay data and for assessing the potential risks to individuals when combined with previous measurements of ^{239}Pu and ^{241}Am in the air, foodstuffs and soil. Other aims of the study will be to investigate the relationship between the absorption characteristics of the actinides *in vivo* and *in vitro* and between the absorption characteristics and the mineralogical composition of the dusts. These data should provide a rational basis for assessing the radiological impact of inhaling or ingesting contaminated dusts present in other accidentally contaminated environments.

CONTRIBUTION OF THE NATIONAL RADIOLOGICAL PROTECTION BOARD, NRPB (Dr. M.R. Bailey)

The NRPB will co-ordinate the project, and carry out work in two distinct but related areas. Experimental inhalation studies will be carried out to provide quantitative information on the deposition, clearance and uptake of materials in the human respiratory tract. Theoretical modelling studies will use information from these and other studies to develop more comprehensive and realistic respiratory tract models.

Work carried out under the current contract relating to inhalation studies has concentrated on the development of the techniques needed, in particular on the generation of labelled aerosols, the measurement of inhaled and exhaled aerosol concentrations and particle size distributions, and the use of *in vivo* measurement techniques for quantifying clearance from the respiratory tract. This development work will be completed. Experimental studies using human volunteers will be carried out to investigate deposition, clearance and radionuclide uptake for different regions of the respiratory tract following inhalation of monodisperse aerosols. Initially, emphasis will be placed on determination of the effect of particle size and breathing pattern on the deposition and clearance of particles in the nasal airways. The work will be extended to investigate clearance kinetics in the bronchial tree. Because of the impact that any slow bronchial clearance will have on equivalent doses calculated using the lung model proposed by the ICRP Task Group, experiments will be carried out to complement those at GSF. Initial objectives will be to provide independent confirmation of the findings, and to measure the retention time of the slow phase, using a radionuclide of longer half-life, such as ^{51}Cr. Uptake of radionuclides into blood from regions of the respiratory tract will be measured, with initial emphasis on uptake from the nasal airways.

Under the current contract, modelling studies have concentrated on development and implementation of the respiratory tract model currently proposed by the ICRP Task Group. Work will continue on the specification of parameters describing absorption into blood for a range of compounds of radiologically significant radionuclides, and will be extended to the following areas:

- Treatment of radioactive decay products
- Implementation of age dependence in the respiratory tract model
- Application of the proposed ICRP respiratory tract model to the dosimetry of radon and its decay products
- Development of analytical and numerical simulation methods for implementation of models containing large numbers of compartments.

As a test case, the ICRP Task Group model's predictions for the levels of actinides found in the general public as a result of weapons test fallout will be compared with autopsy data.

CONTRIBUTION OF FORSCHUNGSZENTRUM FÜR UMWELT UND GESUNDHEIT GMBH, GSF

(i) Bolus dispersion will be studied in models representing various airway geometries (tubes and systems of tubes; models of bifurcations, of the tracheobronchial tree, and of the pulmonary region). Dispersion mechanisms will be investigated by means of a statistical analysis of bolus dispersion data from humans. Both results will be compared with model calculations. Additionally, bolus dispersion will be investigated in diseased subjects suffering from bronchial obstruction, emphysema, and fibrosis. These data will be correlated to other lung function data and disease symptoms.

(ii) Aerosols of ultrafine, monodisperse, inert ^{111}In oxide particles will be inhaled both at steady state breathing and as a bolus. Clearance will be studied by scintillation counting. This allows intercomparisons between clearance data which traditionally have been associated with regional deposition, and information on the actual site of deposition of the particles given by the bolus data. The actual regional deposition can then be determined more successfully. These studies will be carried out with both smokers and nonsmokers, and with patients suffering from various kinds of lung disease. Furthermore, deposition will be studied in airway models and compared with model calculations.

(iii) Aerosols of ^{99m}Tc-labelled Fe_2O_3 particles of various sizes inhaled as a bolus will be used to investigate the regional distribution of the deposited particles, and the clearance mechanisms in varying volumetric depths of the lungs. This will be done by γ-camera imaging and by scintillation counting. Furthermore, effort will be devoted to developing simpler production methods of inhalable radioaerosols.

(iv) Magnetometric measurements of inhaled Fe_3O_4 particles will be conducted on healthy subjects to obtain standard data for long-term retention, magnetic relaxation, intracellular viscosity, and macrophage mobility as indicators of the status of the defense system of the human lungs. Both smokers and nonsmokers will be studied since retention and relaxation have been shown to depend on smoking habit. When standard values are available, the investigations will be extended to subjects with specific lung diseases associated with alveolar macrophage dysfunction. This is expected to provide new information on the mechanisms of these diseases. Also, studies will be performed on cultured macrophages, which permit the alteration of certain properties of the macrophages.

CONTRIBUTION OF COMMISSARIAT À L'ENERGIE ATOMIQUE, CEA

Dosimetry of inhaled substances requires the use of lung physiological parameters which may vary from one subject to another; especially important are those related to the growing pattern during childhood and also those of significant peculiarities in human ethnic groups.

The proposed contribution to dosimetry of inhaled substances is to conduct human physiological measurements, on the occasion of clinical investigations, within a hospital

department and with the approval of its Ethical Committee.

1. Oronasal distribution of respiratory airflow

The variations of respiratory airflow between the oral and nasal routes has been studied experimentally by Niinimaa *et al.*, in 1980 and 1981, in 30 adults during incrementally graded exercise. This study is important because total and regional depositions are different depending on whether gases and aerosols are inhaled through the nose, considered to be a very efficient natural filter or directly through the mouth. We propose to measure these airflows in children and in adults for comparisons, during incrementally graded exercise, in relationship to airway resistances.

2. Deposition of inhaled particles in the airways of subjects from various ethnic groups

Comparisons made between individuals of the same body size have shown that Caucasians have lung gas volumes higher by 10 to 15% than those of Africans and Asiatics; this has been observed in adults and in children. In order to determine the importance of these variabilities upon airway deposition of radionuclides that could lead to different exposures, we propose to compare this deposition in healthy subjects of those three ethnic groups.

3. Methods

Measurements of lung volumes will be done by standard spirometry, helium technique and plethysmography. Air flows during incremental exercise will be controlled by respiratory masks, Fleisch pneumotachographs and pressure transducers and eventually hot wires, with monitoring from a bicycle ergometer.

Particle deposition in the airways will be measured by comparing inhaled and expired aerosol concentrations by laser velocimetry, using inert monodisperse polystyrene particles and during spontaneous breathing with controlled tidal volumes and respiratory frequencies.

CONTRIBUTION OF THE MEDICAL RESEARCH COUNCIL, MRC

The following experimental investigations are of importance concerning the modelling of delayed tracheo-bronchial clearance in man: for the validation and further development of this aspect of the lung model, it is necessary that the physiological and cellular aspects are properly understood. Hence the mechanism of delayed tracheo-bronchial clearance of particles will be the major focus of this part of the project.

Studies will be continued in which, following inhalation or intra-tracheal instillation of insoluble particles, the amount of material on the tracheal surface is determined at sacrifice. From this it is possible to analyse the tracheal content into fast-moving and stationary (or slow-moving) particles on the airway surface, as well as material incorporated into the tissue. With inhaled fused alumino-silicate particles it has already been shown that the fraction which is stationary on the epithelial surface is an order of magnitude greater than the rapidly cleared fraction after 30 and 120 days.

The study of the functional morphology of the large airways will be continued, to explore the possibility that the delayed clearance of particles is related to discontinuities in the muco-ciliary clearance process. Morphometric studies have commenced on the rat trachea, in which the percentage of the surface of the airway epithelium which is not made up of ciliated cells is being estimated. These will be completed and extended to other strains and species.

Preliminary evidence suggests that particles residing on the surface of the ciliated epithelium for extended periods are within macrophages. These cells promise to be the object of considerable interest in the next few years: they may be monitoring inhaled particles too large to reach the alveoli and be detected by alveolar macrophages. Efforts will be made to develop methods for harvesting these resident airway macrophages so that their properties can be compared with alveolar macrophages.

Using alveolar microinjection it has been shown that particles may be found on the surface of small airways for up to 15 months after deposition specifically in the alveoli, as distinct from deposition on the airways themselves. Further studies will be made with this technique, to clarify to what extent and under what conditions delayed bronchial clearance can result from particles deposited in alveolar tissue. These studies would also be expected to improve our understanding of certain aspects of alveolar clearance, such as differential particle clearance rates from different micro-anatomical regions of the lung, and to address some of the outstanding problems of particle phagocytsosis *in vivo*.

CONTRIBUTION OF THE NATIONAL RADIOLOGICAL PROTECTION BOARD, NRPB (Dr.G.N. Stradling)

As a consequence of the aeroplane accident over Palomares, Spain in 1966, persons living in the area were contaminated with ^{239}Pu and ^{241}Am. The soil is still contaminated with these actinides. NRPB and CIEMAT are collaborating in a study, the main aim of which is to provide an experimental basis for improving the assessment of internal doses from environmental radioactivity in the Palomares area. Initially, *in vitro* solubility measurements will be used to identify the likely variation in the absorption characteristics of ^{239}Pu and ^{241}Am in the respirable fraction derived from different soil types and particle size distributions.

The biokinetics of these actinides in those fractions showing the greatest differences in solubility will then be investigated in rats after their deposition in the lungs or gastrointestinal tract. The information gained will be used to assess the appropriate doses per unit intake and to predict the behaviour of ^{239}Pu and ^{241}Am in humans using the protocols associated with the new ICRP respiratory tract model. These predictions will be of value for improving interpretation of chest monitoring and bioassay data and for assessing the potential risks to individuals when combined with previous measurements of ^{239}Pu and ^{241}Am in the air, foodstuffs and soil.

The NRPB will undertake all the animal experiments designed to investigate the biokinetics of Pu and Am after inhalation, instillation or ingestion of defined dust samples. In the inhalation/instillation experiments, each dust or residue of known isotopic

composition and mineralogical composition ascertained by CIEMAT, will be administered to groups of 35 animals. Groups of five animals will be killed at intervals up to 1 y after exposure, to determine the tissue distribution of Pu and Am and assess their transfer rates to the blood. The experimental data will be used to predict the biokinetics of Pu and Am in humans and to provide guidance for the interpretation of personal monitoring data. In support of these studies the NRPB will investigate the biokinetic behaviour of Pu and Am in the presence of Fe_3O_4, $CaSO_4$, $SrSO_4$ and $CaCO_3$ with which they are known to be associated in an accidentally contaminated environment.

In the ingestion studies, the dusts or residues will be mixed with the normal diet and fed to groups of five rats over appropriate intervals. The animals will be killed for tissue analysis after a further week on their normal diet. On the basis of the results obtained, materials specific gut transfer factors will be recommended.

CONTRIBUTION OF CENTRO DE INVESTIGACIONES MEDIOAMBIENTALES Y TECHNOLOGICAS, CIEMAT

As a consequence of the aeroplane accident over Palomares, Spain in 1966, persons living in the area were contaminated with ^{239}Pu and ^{241}Am. The soil is still contaminated with these actinides. NRPB and CIEMAT are collaborating in a study, the main aim of which is to provide an experimental basis for improving the assessment of internal doses from environmental radioactivity in the Palomares area. Initially, *in vitro* solubility measurements will be used to identify the likely variation in the absorption characteristics of ^{239}Pu and ^{241}Am in the respirable fraction derived from different soil types and particle size distributions. The biokinetics of these actinides in those fractions showing the greatest differences in solubility will then be investigated in rats after their deposition in the lungs or gastrointestinal tract.

The information gained will be used to assess the appropriate doses per unit intake and to predict the behaviour of ^{239}Pu and ^{241}Am in humans using the protocols associated with the new ICRP respiratory tract model. These predictions will be of value for improving interpretation of chest monitoring and bioassay data and for assessing the potential risks to individuals when combined with previous measurements of ^{239}Pu and ^{241}Am in the air, foodstuffs and soil.

CIEMAT's contribution will involve particle size analysis of various dust fractions, their chemical and mineralogical composition and correlation of these characteristics with the physico-chemical forms of Pu and Am. *In vitro* dissolution studies will be undertaken on the respirable fraction of a variety of different dusts in order to identify those containing the least and most mobile forms of these actinides. These particular dusts will be used for animal experiments conducted at NRPB. CIEMAT will also provide advice on the mineral forms most likely to be associated with Pu and Am, surrogates of which will be used in inhalation/instillation and ingestion experiments conducted at NRPB for more closely defining the factors affecting the absorption characteristics of the actinides.

Thus the work conducted at CIEMAT will include - (1) Particle size analysis of dust fractions obtained from different locations within the contaminated environment,

(2) Separation of the respirable fraction (AMAD <5 μm) for inhalation/instillation experiments and the fraction <250 μm for the ingestion experiments, both using rats, (3) Determination of the chemical and mineralogical composition of each particle size fraction, (4) Determination of the Pu and Am content of each particle size fraction, (5) Short term (28 d) *in vitro* experiments to identify those dusts displaying the extremes of Pu and Am dissolution, (6) Long term (0.5-1 y) *in vitro* experiments with those dusts used in inhalation/instillation experiments in order to compare the dissolution rates *in vitro* with the transfer rates to blood *in vivo*, (7) Joint assessment with NRPB of the doses per unit intake and limits on intake for the dusts present in a contaminated environment, (8) Review, in collaboration with NRPB, of the parameters to assess the internal doses to individuals living or working in a defined area, based on the biokinetics of Pu and Am associated with the dust, personal monitoring data and measurements of these actinides in air, foodstuffs and soils.

A2 Transfer and behaviour of radionuclides in the environment

Contract FI3P-CT920003 Promotion of formation, knowledge and exchange of information in radioecology.

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Total Contribution by the Commission: 200 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

I. GENERAL FRAMEWORK OF IUR PROGRAMME AND OBJECTIVES.

Recently the Board of Council of IUR has reevaluated the priorities, taking into account the expertise of its members and the flexibility of the organization. It is important to remember that IUR in its various activities should be concerned primarily with Radioecology in both basic and applied areas. Where appropriate, such activities can involve matters relating to Radiation Protection, especially in the context of Ecology. The IUR will not become involved in proper scientific research and in questions that are best handled by the appropriate scientific disciplines such as, for example, Meteorology, Botany, Physiology etc...

The programme 92-94 aims to promote:

1) the cooperation and the exchange of information between radioecologists, in particular from Eastern European countries and other countries outside EC, in order to stimulate interactions that would increase our understanding of Radioecology problems; the Newsletter publications play an important role in this context;

2) the formation and training of young scientists; training courses on specialized radioecological topics are also foreseen, as well as the development of a curriculum for a textbook on Radioecology;

3) the fulfilment of the objectives of several specified Task-Forces, dealing respectively with: a) the priorities in Radioecology, b) the relative contribution of various exposure pathways in different environmental conditions , c) the assessment of the relative importance of various parameters for the contamination of freshwater fish, d) the behaviour of less commonly considered radionuclides.

II. IMPLEMENTATION OF THE PROGRAMME

A. STEERING COMMITTEES (SC)

- SC 1: **Training**
  (Chairman C. Myttenaere)

One of the best methods of promoting interest in radioecology, among young scientists, is through the organisation of SUMMER SCHOOLS.

- SC 2: **Publications**
  (Chairman N. Pattenden)

The first priority will focus on the NEWSLETTERS which represent a major method of diffusion information among all scientists involved in radioecological studies.

The next priority will be the production of a TEXTBOOK on Radioecology which will serve as background material for students and scientists who are not specialized in this field.

Finally, methods of contributing to information for the general public about radioecology and nuclear energy, will be evaluated.

- SC 3: Cooperation with Central and Eastern Europe countries
(Chairman R. Kirchmann)

The objectives of SC3 are to develop scientific contacts with Central Europe countries.

The cooperation with scientists from CIS will be pursued, particularly from the point of view of exchange of scientific informations of mutual interest.

B. TASK-FORCES (TF)

- TF 1: Priorities in Radioecology
(Chairman : C. Myttenaere)

Research in radioecology has been the subject of a varying interest in time, for example the recent demonstration of the necessity to acquire data in order to improve our knowledge on the behaviour of released radionuclides in the environment.

Many data were accumulated on the transfer of some of the long lived radionuclides from studies on the fallout of nuclear weapon tests, and for some time after, by studies of the impact of routine releases from nuclear installations.

It is inevitable that new problems will arise given the programmed radioactive discharges, development of new technologies (fusion) and ageing (decommissioning) and comparatively good working conditions of some European installations (e.g. in Eastern countries). Some of the potential hazards that could arise have not yet been considered with respect to the possible transfer of radioactivity to man in the Community.

The relevant transfer models have thus to be improved or modified to incorporate source term characteristics, ecological properties of ecosystems and the variability and reliability of transfer parameters used (i.e. influence of the environmental characteristics on derived transfer factors) A comparative study of their relative merits and defaults is also needed to improve the accuracy of model predictions.

Observations and research connected with the consequences of major nuclear accidents has allowed some observations and allied research (now in collaboration with our CIS colleagues) which indicate that the radioecological approaches to such releases needs to be improved in various fields and that some new problems need to be tackled (consequences of the speciation; behaviour of long-lived radionuclides in natural ecosystems, countermeasures ...).

This will be essential to enable improved dynamics models to be produced, particularly with regard to the kinetics of processes which are responsible for the environmental transfer of radionuclides to man.

The aim of the proposed task force is to produce an updated assessment of the current "state" of radioecology and to identify those areas which require more knowledge.

- TF 2: **Assessment of the Relative Dose Contribution of Various Pathway under Different Environmental Conditions, Including Semi-Natural Environments.**
(Chairman : C. Bunnenberg)

Post-Chernobyl investigations of contamination levels in the different compartments of affected areas and estimations of the total dose to members of the population have revealed considerable variation in the relative importance of different pathways. The results also differ considerably from previous evaluations of the possible radiation dose arising from weapons fallout, at least with respect to the ratios of external to internal exposure. More recent studies show the comparative importance of pathways in semi-natural environments, which had received little attention previously.

A number of parameters have been identified which cause differences in the relative importance of exposure pathways within and between countries and with the time elapsed after deposition of the Chernobyl fallout. Important factors identified include (i) the heterogeneity of the fallout, both with respect to levels and relative abundance of radionuclides, (ii) environment-specific characteristics (affecting, eg, deposition velocities, transfer rates into the food chain and the ecological half-lives of radionuclides, (iii) seasonal and general climatic variations, (iv) differences in land use (urban, agricultural, seminatural with quite different contamination dynamics), (v) regional and national characteristics with respect to dietary and recreational habits (eg, the great importance of food products originating from semi-natural environments for critical groups and (vi) the greatly differing efficiencies of countermeasures, which have been initiated at different times and intervention levels in each country.

The purpose of this project is to collate the available information to evaluate and quantify the relative contributions of different pathways to the total dose of the population and of critical groups and to define research activities to fill knowledge gaps. Therefore, the project represents an important step towards the improvement of dose predictions for a variety of different population groups and towards a more comprehensive understanding to enable decisions to be made on the use of countermeasures to optimize benefit-to-cost ratios.

- TF 3: **Assessment of the relative importance of various freshwater exposure pathways and the major parameters influencing pathways.**
(Co-chairmen : L. Foulquier and C.V.Voytsekhovitch)

The important freshwater exposure pathways are: internal exposure via drinking water, freshwater fish and irrigation of agricultural products; and external exposure due to swimming, boating, contact with fishing utensils and use of beach areas.

The content of radioactive materials in a water body will consist of materials deposited directly upon the water surface and run-off of deposited material in the catchment area. The latter component can in some cases be dominant. It is also strongly weather and/or season-dependent. The seasonal dependence is pronounced in areas where there is significant snow cover in winter, where there will be a peak in the activity concentration in the lake during spring and also peaks during winter, coinciding with periods with above-freezing temperatures. Significant parts of the activity brought into the lake in such a manner may be incorporated in dead organic material (leaves, twigs etc.), and may be more bioavailable than material transferred into the lake with soil.

Rivers and lakes contain radioactive materials in solution or fixed on solid particles. In the latter case the particles may be in suspension or settle down to the bottom, which is a continuous process. A typical velocity of sediment buildup is about 1 cm per year. Concentration factors linking concentration in fish to that in water have frequently been used, though the appropriateness of such an approach has been questioned after Chernobyl, since uptake does not take place directly from water, but indirectly via numerous nutrition pathways. Post-Chernobyl measurements indicate that radiocaesium concentrations in fish are closely linked to content in sediment of the same lake, modified by various sensitivity parameters used to characterize the lake. The sensitivity parameters include height above sea level, area of the lake, residence time of water in the lake and potassium concentration in lake water. The concentration of radiocaesium in fish also depends upon the feeding habits of the fish, which vary for different species, but which also changes with age and size of fish of the same species. Another observed correlation is that in the same catchment the concentration in fish is higher in smaller lakes. The concentration is also higher in lakes where the residence time of the water is longer. Activity concentrations in fish are also lower in hard water or water with high concentrations of phosphorus or potassium.

It is expected that the relative importance of the freshwater pathways and parameters may vary significantly between different areas in Europe, and the purpose of the proposed task is to evaluate these pathways and parameters for several typical types of areas.

- TF 4: **The Behaviour of less commonly considered Radionuclides**
(chairman : A. Aarkrog)

From a radiological point of view Cs-137 is the most important radionuclide released by the Chernobyl reactor accident. Cs-137 is also the major contributor to the collective dose to the world population from global fallout and also resulting from the former discharges from Sellafield.

High resolution gamma-spectrometry makes determination of low levels of Cs-137 and most other gamma-emitters comparatively easy. Most Chernobyl data consequently deals primarily with gamma-emitters, in particular radiocaesium. However, the Chernobyl accident also involved other radionuclides and some of these may be studied in the highly contaminated areas in Ukraine, Belarus and Russia. These other radionuclides often comprise pure beta- and/or alpha-emitters e.g. Ni-63, Tc-99, Sr-90, Pu, (Am), Cm. However, more exotic long-lived radionuclides, such as Se-79, Zr-93, Nb-94 and Np-237 may also be detectable in the Chernobyl debris. The former USSR contains other highly contaminated areas where the behaviour of less commonly radionuclides may be studied, particularly the Chelyabinsk region in the southern Urals. There are however also localities within the EC which may be examined in this respect, such as the Irish Sea.

It should be borne in mind that determination of beta and alpha-emitters involves radiochemical analysis which is much more expensive, both in consumables and labour, than gamma-spectroscopy. Hence the amounts of data produced for pure alpha or beta emitters are typically one to two orders of magnitude less than those obtained from an equivalent effort spent on gamma-emitting radionuclides.

Nevertheless it is important to obtain information on less commonly considered radionuclides, particularly when they may still be detectable in the biosphere. It is thus proposed that the task force should concentrate on alpha and beta-emitting radionuclides rather than on gamma-emitters.

- TF 5: **Effects of *ionizing* radiations on plants and animals.**
(Chairman : D. Woodhead)

The general objective is to identify those areas which require more knowledge in order to be able to calculate the dose to plants and animals living in contaminated environments.

In this context, it has become clear that there is an enormous amount of information available from CIS sources concerning the impact of the release at Kyshtym and work is continuing on the impact of the Chernobyl accident.

The Task-force will as a first step, try to collect and translate all relevant literature in Russian. As a next step a report should be produced based upon this literature review together with available Western information.

A21 Environmetal behaviour of radionuclides in situations meriting particular attention for long-term behaviour or post-acident conditions.

Contract FI3P-CT920029 Towards a functional model of radionuclide transport in freshwaters.

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Total Contribution by the Commission: 400 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Contribution of The Institute of Freshwater Ecology

a) A survey of natural fresh waters of different chemical types will be carried out to measure the stable Cs : K ratio and the stable Sr : Ca+Mg ratio in order to ascertain the natural range of variability. Samples will be supplied by most of the other members of the group for comparison.

b) ammonia concentrations will be measured over the productive season in the epilimnium and hypolimnum of a eutrophic, monomictic lake. These data, in conjunction with laboratory measurements of the availability of radiocaesium from particulate material made by the Catholic University of Leuven, will be used to assess the relative contribution of catchment sources and the sediments to radiocaesium build up in the hypolimnium after a deposition event.

c) Ammonia concentrations in cores from the sediments of a eutrophic lake will be measured over a year to assess the local variability in the surface layers. These data will be used, in conjunction with in situ K_Ds, to estimate local variability in-situ pore water caesium concentrations. Using measured boundary layers the probable flux of radiocaesium into the sediments by direct diffusion will be calculated and the results used to test the hypothesis that local differences in interstitial water ammonia concentrations are sufficient to vary diffusional fluxes of radiocaesium, thus creating the local variations in sedimentary radiocaesium deposition observed after Chernobyl. This work will be carried out in collaboration with ECN, the Netherlands.

INETI contribution.

OBJECTIVES

1) Sorption-desorption processes of radionuclides in sediments.

Continuation of on-going work concerning the immobilization dynamics of radiocaesium dynamics of radiocaesium in Tejo River sediments.

Sorption of radiostrontium in Tejo River sediments to study the selective and sequential separation of radiocaesium and radiostrontium using ion exchange techniques.

2) Biological processes

To improve the evaluation of the reliability of transfer factors in freshwater trophic chains, mainly through the knowledge of the parameters affecting their variability.

Uptake and loss experiments with freshwater fish, under different Cs/K ratios (artificial water) using radiocaesium, will be carried out.

The study of the rate of transfer of radiocaesium from microalgae to zooplankton will be implemented.

COLLABORATION

The studies in sediments are carried out in close collaboration with the University of Leuven (where Maria Jose Madruga is preparing a doctorate).

The studies concerning the biological processes will proceed with the collaboration of the Institute of Freshwater Ecology, the University of Malaga and the Service d'Etudes et Recherches sur l'Environnement (CEA-Cadarache).

Contribution of K.U.LEUVEN

The contribution of K.U.Leuven will focus on the following objectives.

a) A systematic and comparative study of the reversibility, the desorption dynamics and aging effects of radiocaesium in river, lacustrine and estuarine sediments. The study on river sediments includes a range of substrates originating from various large river basins in the E.C. The study on lacustrine sediments covers a range of substrates of varying mineralogy, including those from the Cumbrian lake district, (studied by IFE and ECN) and The Netherlands (studied by ECN); the study on estuarine sediments will be limited to the Loire, Po and Tejo.

b) The desorption dynamics of radiocaesium of freshwater sediments deposited in a marine environment. These studies include sediments originating from the large river basins specified in the section a)

c) The development of diagnostic tools which may allow to identify sediments characterized by a very rapid fixation of radiocaesium and to predict such behaviour on the basis of readily measurable properties of the system.

d) Desorption behaviour of radiostrontium in sediments; this study concentrates on a selective and sequential displacement methodology of radiostrontium and radiocaesium.

COLLABORATIVE LINKS

-IFE-ECN

All systems originating from the Cumbrian lake district and The Netherlands are to be included in our experimental protocol. In situ measurements and desorption fluxes from sediments will be complemented by our desorption studies, thus providing the necessary input for interpretation of field behaviour.

-CEA, Cadarache

The study of the dynamics of caesium in river basins will be carried out in collaboration with CEA. Sampling and general characterization will be made by CEA.

-ENEA

The systems studied by ENEA are included in our program. These studies should enable to rationalize field behaviour on the basis of laboratory studies. ENEA will carry out the field sampling in the Po river and in the Chernobyl area. These systems will be made available to K.U.Leuven, thus providing an additional link with ECP-3 (CHECIR).

-LNETI

Direct links with the Tejo river study via a current Ph.D. program by M.J Madruga.

Contribution of IPSN-CEA/SERE/URSED

1) The concentration of caesium-137 in French river sediments

The accident of Chernobyl confirmed the importance of radiocaesium in the aquatic ecosystems and specially its fixation in the sediments.

Considering the impact of different source terms as the liquid effluents from the power stations and fuel cycle plants or fall out, a review will be made on the fixation of caesium-137 in the sediments of the french rivers sediments in relation with different parameters like the particle size, and mineralogical structure of these sediments.

The data concern the seven french rivers equipped with nuclear facilities, which are: Rhone, Seine, Loire, Garonne, Moselle, Meuse, Rhin.

2) The transfer of radioruthenium in aquatic ecosystems.

The gamma measurements of field samples show the importance of radioruthenium downstream from the fuel reprocessing plant of Marcoule. This nuclide represents between 70 to 90% of the liquid effluent radioactivity. Ruthenium 103 and 106 were detected in the aquatic compartments after the accident of Chernobyl.

There is a rather complete absence of studies concerning the modalities of the transfer of this radionuclide in freshwater ecosystems. We know that in the Rhone river, downstream from Marcoule we noted a rather high concentration (of about 100 Bq/kg dry weight) in sediments or aquatic vegetation and a lower concentration in fish <10 Bq/kg wet weight).

To be able to answer questions about it, an experimental work is being done (as it was done in Cadarache for silver 110m, caesium 137, and 60 cobalt).

The experimental ecosystem is composed of several compartments (water, sediment, three benthic organisms, a zooplanctonic species, two algae and a fish).

The Physical-chemical form of the ruthenium is $RuCl_3$ which is very simple and rather stable during time.

One experimentation will be done in the field conditions downstream from Marcoule.

The experimentation will give several transfer factors and kinetic equations which will be used to build the transfer model.

This mathematical tool will give a synthetic view of the different ruthenium flow between the compartments of the aquatic ecosystem, in order to interpret field data.

Interaction with other partners

The first theme will bring field information to other people of the working group making laboratory research on the fixation of caesium-137 in the sediment.
The experimental works done during last period contract on silver 110m, allow us to give to Oscar Vanderborght (University of Antwerpen) the methodology used for 110m-Ag speciation.

Contribution of ENEA-DISP

The aim of this research is to study the radiocaesium transfer processes, between dissolved and particulate phases, in water bodies.

Studies in the earlier CEC contract were focused on the influence of salinity and suspended particle size on adsorption-desorption processes in 2 rivers located in the north-eastern part of Italy (Stella and Tagliamento rivers).

The results confirmed that radiocaesium is mainly associated with particle in the size range 5 - 0.1 μm. A high variability was found in the Kd values in these fine particles which suggests that the grain composition plays a significant role on radiocaesium adsorption-desorption processes.

For this reason, the effort of ENEA-DISP studies within the current programme in the next 2 years will concentrate principally on the study of the role of organic and inorganic composition of suspended particles on the caesium sorption-desorption processes.

Water and the different fractions of suspended particles will be taken seasonally along the above mentioned rivers at locations of different salinity, using devices capable of performing size fractionation of suspended solids and with a set of columns containing ion-exchange resins to fix Cs-137 dissolved in the water.

The ENEA-DISP sampling system is designed to gather different fractions of suspended particles, using 4 different cylinder cartridges (from 105 to 0.45 μm)

The samples of suspended particles will be analyzed for grain size; mineral constitution by X-ray diffraction; radiocaesium content by gamma spectrometry; cation exchange capacity; NH4, K, Cs, Ca, Mg, Sr concentrations in the water, salinity, temperature, Ph.

In order to evaluate the effects of parameters influencing the solid-liquid partitioning of radiocaesium and stable elements, Kd values will be determined for the different fractions of the suspended solids sampled at locations of different salinities. The specific site capacity and aging factors will be characterised in cooperation of Leuven University (Belgium).

The Programme should enable us to rationalise field behaviour in the terms of laboratory characterisation.

Contribution of the Department of Biology, University of Antwerp (RUCA).

Transport of radionuclides across exchange surfaces in freshwaters in relation to environmental conditions.

The uptake and accumulation of radionuclides in fish and other aquatic organisms depends on the biological availability of the radionuclides in the environment. The biological availability of a radionuclide is determined by the speciation of the element in the environment and the physiological organisation of the epithelial exchange surfaces. The translocation of a radionuclide from the environment across the epithelium to the blood is the first critical step in the accumulation process. Important aspects of this process are: 1) the fixation of the radionuclide at the water-body interface, 2) the facilitated transport across the apical membranes of the epithelium, 3) the intracellular transport by metal-binding proteins and 4) the facilitated transport across the baso-lateral membranes towards the blood. A basic understanding of the processes that are involved in the uptake of radionuclides is an essential and important step in the building of a functional model for the transfer of radionuclides. Within the framework of the project we aim to characterise the transport of radiocobalt across the epithelium of the gills and digestive system in the common carp (Cyprinus carpio) by studying:

- The effect of the chemical speciation of the radionuclide on the uptake of the radionuclide.
- The effect of ionic strength and temperature on the uptake of the radionuclide.
- The effect of protons, alkaline, alkaline-earth and transition metals on the uptake of the radionuclide.
- The effect of modulators and inhibitors of passive and active transport processes on the uptake of the radionuclide.

Together these different experiments will provide the necessary data to build a model for radionuclide transport across exchange surfaces that can be used to predict and explain the effect of changes in environmental conditions on radionuclide uptake. The model will be developed in collaboration with the group of the University of Malaga (Fernandez-Serrano) who is studying the effect of environmental conditions on the uptake of radionuclides in aquatic plants. The work will also provide the fundamental understanding of radionuclide uptake processes required by the groups of the CEA Cadarache (Lambrechts-Foulquier) and the INETI Lisbon (Vaz Carreiro-Galvao) who are studying the effects of environmental conditions on the bioconcentration and biotransfer of radionuclides in freshwater environments. In addition the model will provide the basis for relating changes in radionuclide sediment-water distribution coefficients, studied by the

groups of the NERC (Hilton), the LNETI Lisbon (Vaz Carreiro-Galvao), the ENEA (Belli-Sansone), the University of Leuven (Cremers) and the ECN (Comans), to changes in the biological availability of the radionuclides.

Contribution of the University of Málaga.

Mechanisms of radionuclide (Cs^{134}) accumulation in freshwater plants.

Our proposal is to investigate the mechanisms of radionuclide accumulation in freshwater plants at two levels,

A. Membrane level, the working hypothesis is that radionuclides are absorbed into the cells and adsorbed to the external surface in the periplasmic space and in the outer part of the cell membrane.

It will be assumed, for the case of absorption, that radionuclides go into the cells through channels, they are driven by an electrochemical gradient across the plasmalemma ($\Delta\mu_i/F$) that is dependent on the charge of the radionuclide (z), the membrane potential of the cell (Em) and the Nernst potential of the radionuclide in equilibrium with the amount of its respective natural isotope present inside and outside the cells. In the equilibrium, i.e. when there is no net flow of radionuclide across the membrane, E_N^i may be computed from the concentration factor for "i".

Radionuclides will be adsorbed to the cells mainly by binding to the free hydroxyl groups of the cellulose matrix of cell wall. This fraction is the amount accumulated when the channels are closed or when the electrochemical gradient across the plasmalemma is zero.

The study of radionuclide accumulation at the membrane level will be carried out in the aquatic liverwort Riccia fluitans.

Whole organism level. The goal of this part will be investigate the effect of growth rate and major metabolic variables (photosynthesis and respiration) in the accumulation of radionuclides. The concomitant increase of cell content leads to the accumulation of metabolizable and non metabolizable ions and by addition to the accumulation of radionuclides.

Our intention is to share concepts and some experiments on uptake and availability with Dr. Blust (University of Antwerp, Belgium). The experiments with micro algae will be performed in collaboration with Dr. Vaz Carreiro (LNETI, Lisbon).

ECN contribution

Significant progress has been made in laboratory studies of radiocaesium mobility, yet in-situ, environmental studies are hampered by the very low activities in aqueous compartments soon after the accident. Because it is our ultimate goal to predict the transport of radiocaesium through the environment, in-situ measurements of radiocaesium and its mobility-controlling parameters are necessary to verify predictions and to indicate additional factors affecting its transport that should be considered in our models.

The contribution of ECN focuses on the geochemical processes controlling the interaction of radiocaesium with freshwater particles. Following a combined field and laboratory approach, the research emphasizes the determination of reliable (in-situ- and laboratory predicted-) sediment/water distribution coefficients (K_D's). This parameter quantifies the interaction of the radionuclide with the solid compartment in sediments and soils and controls, therefore, its mobility.

The K_D-value for radiocaesium varies between different sediments and soils, because of differences in the availability of illitic frayed edges, but is also subject to chemical changes within these environments. ECN research to date has shown that post-depositional changes in the pore water chemistry of freshwater sediments may result in a partial remobilization of sediment-bound radiocaesium. The extent of such remobilization depends on the reversibility of the caesium sediment interaction and is of great importance for the establishment of the radiological consequences of nuclear accidents. ECN work includes, therefore, an in-situ investigation of radiocaesium mobility in freshwater sediments, as well as a laboratory and field investigation of the mechanisms affecting caesium sorption reversibility.

Laboratory and modelling work at ECN has highlighted a strong interrelationship between the kinetics of radiocaesium sorption and its reversibility. A slow uptake process is believed to represent radiocaesium migration to illitic interlayer sites, from which the radionuclide is not easily released. The rate of migration depends strongly on the major competing cation and is a key parameter in the evaluation of radiocaesium mobility in aquatic environments and for estimating remobilization risks from soils or sediments. Present work emphasizes reversibility, its effect on K_D's and its application to transport models. ECN works closely together on these subjects with the Institute of Freshwater Ecology (IFE, Windermere, UK) and the Colloid Chemistry Laboratory, Faculty of Agronomy, K.U Leuven (Belgium).

Contribution of the Norwegian Institute for Nature Research

Radiocaesium turnover in freshwater fishes and invertebrates

The radioactivity in fish depends upon the balance between intake and excretion of the isotopes. The intake is determined by the feeding rate, the radioactivity in food and the absorption efficiency. We quantify the absorption of radiocaesium from different prey types by ecologically different fish species in feeding experiments.

The excretion of radionuclides varies largely between nuclides and fish species, and depends on both fish size and ambient temperature. The excretion rate is a major determinant for radioactivity in fishes, and experiments on radionuclide excretion in ecologically different species is thus important in the model work. The excretion rate in different fish species, and its temperature and size dependency, will be examined by administrating a radioactive jelly to the fish.

The rate of radiocaesium accumulation from water is important for the radioactivity of freshwater invertebrates especially during the initial phase after a fallout. Thereafter the dominating determinants for invertebrate radioactivity are their feeding and excretion rate.

The accumulation of radiocaesium by freshwater invertebrates will be examined in aquarium with radiolabelled water. Radiocaesium retention will be studied by keeping labelled organisms in label-free water.

This project provide input data on radiocaesium turnover in fish and its prey to the joint EC model of radionuclide transport in freshwater environment. The experiments, however, will be carried out in Norway.

A21 Environmetal behaviour of radionuclides in situations meriting particular attention for long-term behaviour or post-acident conditions.

Contract FI3P-CT920046 Mechanisms governing the behaviour and transport of transuranics (analogues) and other radionuclides in marine ecosystems.

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Total Contribution by the Commission: 310 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

OBJECTIVES/ OVERVIEW

The purpose of this programme is to define the fundamental parameters governing the transport and behaviour of transuranic (including analogue elements) and other radionuclides in the marine environment, with particular emphasis on:

(a) the characterization of the source term,
(b) the influence of physical and chemical speciation including the role of colloids,
(c) the role of inorganic and organic complexes,
(d) hot particles,
(e) chemical modification after release,
(f) behaviour in estuarine and deep environments,
(g) transfer via particles in shallow environments,
(h) transfer from shelf waters to deep-sea,
(i) transfer from land to sea, and
(j) transfer through the primary producer route into the food chain leading to man.

In essence, this programme seeks to identify basic parameters and provide data of a universal character, suitable for use in real, predictive models based on fundamental mechanisms, rather than on the earlier and inadequate box-model approach.

The programme addresses a number of diverse environments where radionuclides become concentrated and pose potential problems in environmental protection. This is the first time a comprehensive and integrated marine ecosystem programme has been initiated simultaneously in the following regions: open ocean, enclosed seas, estuaries and continental shelf areas. The seven participating laboratories, representing six countries throughout Western Europe, provide a diverse and proven expertise in the several areas relevant to sampling, physical and chemical speciation, sedimentation, physical transport and ocean currents, as well as laboratory-based radiochemical, radiometric and sequential extraction analyses.

The detailed studies now underway should provide valuable information which can be utilized in the assessment of human exposure and risks associated with the transfer of radionuclides to man. This vital radiological protection goal provides impetus to increase our understanding of the environment into which man has introduced contaminants and trace pollutants. It is well known that the incorporation of radionuclides in the food chain leading to man begins with the biological process of uptake by phytoplankton and algae. In the case of lipid-soluble compounds, the biological magnification often exceeds a factor of 10^5. On the other hand, the uptake of many inorganic elements is relatively low and often does not exceed a factor of 10^2. Thus, the nature of the chemical form of a radionuclide is a very important aspect which should be understood for the purposes of optimization of radiation protection. It is for this reason that the physico-chemical speciation of transuranics and other radionuclides must be determined experimentally and then evaluated in the framework of risk identification. Moreover, the nature of the organic complexation of these radionuclides must also be evaluated throughout the food chain (sediment - water - plankton - crustacea - fish - man). For this reason, a significant

biological component has been included which should enable a preliminary evaluation to be made of the effect organic compounds and complexes have on food-chain concentration processes. Such studies, in combination with the other components of the programme, provide the essential ingredients for a better understanding of the marine bio-geo-chemical ecosystem. This, in turn, can be applied at a broader level to the development of guidelines for remedial action programmes involving the release of radionuclides and other trace contaminants into the environment.

It is anticipated that the data bases and the improved understanding of fundamental mechanisms which should be derived from this collaborative programme of research, will be of considerable value in the context of other CEC projects involving a number of the present participants.

SCIENTIFIC/TECHNICAL DESCRIPTION

The programme of work to be carried out will include both field- and laboratory-based studies in the north-west European seas, The Mediterranean Sea and Baltic estuaries. Sampling platforms necessary for the study will include research vessels such as the *Cirolana*, the *Noroit*, the *Urania* and others, as well as smaller vessels for inshore sampling. In the Irish Sea, the physico-chemical speciation of transuranic nuclides (plutonium, americium and curium) and other nuclides (cobalt and thorium) will be examined using state-of-the-art techniques including dual tracer oxidation state identification, sequential sorption, ultrafiltration, activation analysis, electron microscopy and sequential leaching techniques (IPSN-CEA, MAFF, ISEM-NLH, and UCD). Emphasis will be given to the characterization of the source term and changes in the physico-chemical forms of discharged nuclides, due to radical modifications in operational procedures, will be investigated. The significance of colloids, and inorganic and organic complexes, in modifying the behaviour of the radionuclides studied, will be assessed in detail. The distribution of colloids and other complexes in the sediments close to the outfall and through the water column in the eastern Irish Sea will also be examined, and a follow-up study carried out to establish whether the introduction of the new Enhanced Actinide Removal Plant (EARP) at Sellafield in 1993 (expected, though not yet certain) alters the significance of such species. Ultrafiltration techniques will be employed to determine the distribution of radionuclides on the basis of particle size, while aluminium oxide beds will provide a means of identifying colloidal species on the basis of sorptive properties. Novel techniques to quantify inorganically and organically complexed radionuclides will be tested and applied in the field (UCD). The properties of certain colloids will be determined using electron microscopy (IPSN-CEA).

In the Channel, the short- and long-term behaviour of plutonium isotopes bound to various sediment types and sedimentological fractions of various origins, will be investigated in terms of suspension and bottom-drifting, transit times and sinks, if any (IPSN-CEA and UCD). Remobilization of transuranics and other radionuclides including ^{144}Ce, from bottom sediments in fined-grained sedimentation areas will also be studied. Dating, using isotopic ratios, will be employed extensively and a selection will be analyzed for ^{241}Pu ($T_{1/2}$ = 14.4 y) by UCD. The speciation of transuranics will be studied, using the several techniques referred to above, in the main sedimentation zone of the English Channel namely, the Gulf of Normandy (in this context, it is proposed that a post doctoral fellow from UCD assist in the work at the IPSN-CEA laboratory). In addition, an existing and extensive data base on the nuclide, ^{144}Ce, will be examined from

the hydrological point of view, and should provide valuable information on particulate mobility in this zone over the short term.The behaviour of plutonium nuclides in the Rhône Delta and their dispersion in the Gulf of Lyons will be elucidated using a combination of experimental studies and *in situ* observations.

The behaviour of long-lived radionuclides (radiocaesium, radiostrontium, radium, thorium, ^{239,240}Pu and ^{241}Am) in the interphase between fresh and brackish waters (spiked by Chernobyl) will be examined in the Kalix River estuary system by the DRP-LUND. Many of the above mentioned novel techniques of speciation of transuranics will be used and new parameters, specific to estuaries, will be examined. In particular, studies will be conducted on:

(a) the formation of organic and inorganic complexes of radionuclides in fresh water,

(b) flocculation and desorption processes at the salinity wedge,

(c) the effects of seasonal variations (flood, neap tide), and

(d) radionuclide balance *via* the analysis of sediment cores and sediment traps.

These estuaries represent ideal locations in which to compare the behaviour of lanthanides (as published in the literature) and transuranics. Similar studies will be carried out in the Seine estuary (IPSN-CEA and UCD). The conditions in the latter are very different, it being a mega-tidal estuary where the source (term) of transuranics is the sea.

To improve our understanding of the chemical and bio-geo-chemical behaviour of transuranics and other nuclides released from the loss of a nuclear device and, in particular, to study the long-term behaviour of plutonium and americium in sediments and benthic animals from the vicinity of such an environmental insult, a study will be carried out on sediment and biota samples from the Palomares zone (CIEMAT and SFR-UAB). The differential behaviour of plutonium and americium in sediments will be examined and evidence for remobilization sought. Hot particles will also be examined and their biological availability studied, while ecological half-lives for plutonium and americium in the sediment - benthic-organism system will be determined in order to estimate reliable transfer factors.

The physical transport of transuranics and other radionuclides (^{90}Sr and ^{137}Cs) will be studied throughout the western Mediterranean and in the Palomares off-shore zone (CIEMAT, ENEA, IPSN-CEA, SFR-UAB and UCD). The radionuclides transported by rivers or present in the surface waters of the NW Mediterranean Sea are transported through the basin by two different mechanisms namely, horizontal transport controlled by the prevailing surface currents and seasonal vertical transport by dense water masses formed during the winter in the Gulf of Lyons and in the Ligurian Sea. Moreover, studies carried out in 1989 by the IAEA-MEL, together with preliminary results obtained within the framework of the CEC Marine Radioecology Programme (1990-92), show that the Levantine Intermediate Water, found in the eastern Mediterranean basin, carries small quantities of Chernobyl deposition to the western Mediterranean.

A detailed study is proposed for the characterization of the physical transport of the above radionuclides through the western Mediterranean Basin, in the open sea, the Gulf of Lyons, the Catalan - Balearic Sea and along the coast of Spain (CIEMAT, ENEA and SFR-UAB). The vertical profiles of ^{137}Cs, $^{239,240}Pu$ and ^{90}Sr will be determined in the water column and correlated to the hydrological characteristics of the water masses and to their circulation in the Basin. The physico-chemical speciation of plutonium in both deep and shallow waters will also be examined and the fractions of plutonium and americium in colloidal form determined.

Furthermore, the role of submarine canyons (Palomares and Gulf of Taranto) in the transport of transuranics from the continental shelf to deeper waters and the distribution of transuranics in marine sediment in this area of enhanced activity, will be examined intensively (CIEMAT, ENEA and SFR-UAB). The terrigenous input to the sea from the Almanzora River will be compared with other Mediterranean inputs such as from the Ebro and the Rhône. With regard to the remobilization of radionuclides and their uptake into marine organisms, a detailed investigation will also be made of the transfer of transuranics and other radionuclides through the primary producer route into the food chain leading to man (UAB).

The studies conducted in the Mediterranean Sea, together with previous work, should provide a sound basis for making decisions or formulating countermeasures in case of significant accidental releases of radionuclides into this environment.

Conclusions of general applicability will be drawn and location specific differences identified from the above-mentioned studies. A number of the techniques which will be applied in the field are complementary to one another, and it should prove to be extremely valuable to compare and contrast the results of the different techniques, particularly those used to study the association of long-lived radionuclides with marine colloids. Some of the more obvious benefits likely to accrue from the collaboration include the significant improvement expected in our understanding of the fundamental mechanisms governing radionuclide behaviour in marine ecosystems, the resulting improvement in the scientific foundation for that aspect of Radiation Protection relating to source definition and pathway identification, and the direct dissemination of practical expertise via collaborative interaction in the field.

CONTRIBUTION OF NUCLEAR RADIATION RESEARCH LABORATORY, DEPARTMENT OF EXPERIMENTAL PHYSICS, UNIVERSITY COLLEGE DUBLIN (UCD)

OVERALL OBJECTIVE

To examine the physical and chemical speciation of transuranic and other radionuclides in diverse marine ecosystems, with particular emphasis on the influence of colloids, and organic and inorganic complexes.

The UCD contribution to the programme will concentrate on studying (i) the physical and chemical speciation of plutonium, americium and other radionuclides in the water column and (ii) the role of colloids and, in particular, the competition between suspended

particulate and colloidal/pseudo-colloidal matter for reduced plutonium and americium in three distinct marine/estuarine environments. The envisaged zones of study include (i) a shallow, semi-enclosed shelf sea (the Irish Sea), (ii) a deep enclosed sea (the western Mediterranean) and (iii) a region of particularly high sedimentation (the Gulf of Normandy). The analytical techniques to be employed will include dual tracer chemical speciation analyses of plutonium and americium using the neodymium fluoride and bismuth phosphate co-precipitation methods, ultrafiltration, sequential sorption, counter-flow or continuous extraction using a recyclable organic solvent and surface-microlayer sampling.

DETAILED OBJECTIVES

(a) Extend the application of the dual isotopic speciation technique for measuring reduced and oxidized plutonium to the western Mediterranean Sea and the English Channel. In particular, analyses will be completed on deep water samples already collected (*N/O Bannock* cruise 7/91 and *N/O Urania* cruise 8/92) in the western Mediterranean, and the technique will also be applied to the Continental Shelf close to Palomares and in the Gulf of Lyons, in collaboration with CIEMAT, ENEA and SFR-UAB, and in the Gulf of Normandy in collaboration with IPSN-CEA.

(b) Refine, calibrate and apply state-of-the-art techniques for the assessment of colloidally associated plutonium, americium and other radionuclides in the following marine environments:

- Irish Sea (with MAFF and ISEM-NLH)
- western Mediterranean (with CIEMAT, ENEA, IPSN-CEA and SFR-UAB)
- English Channel (with IPSN-CEA).

In this context, both ultrafiltration and sequential sorption techniques will be employed and the results from both compared. With regard to sorption, a new model (recently developed in our laboratory) based on the differential sorption of reduced and oxidized plutonium on aluminium oxide, will be refined and calibrated.

(c) Utilize a sequential ion-exchange system for the assessment of charged molecular (organic or inorganic) complexes of plutonium, americium and other radionuclides.

(d) Apply the standard chemical engineering technique of counter-flow or continuous extraction, using a suitable recyclable organic solvent, adapted for use in the marine environment, to assess the component of transuranics and other radionuclides in the form of organic complexes in sea water. The method will be developed for field use but, initially, will be studied and optimized in laboratory experiments. Stable organic zinc and ^{65}Zn radiotracer may be employed in these studies.

(e) Examine the physico-chemical speciation of plutonium and americium in the sea surface microlayer using precision sampling technology.

The programme is consistent with previous research projects of a marine radioecological character carried out at UCD. In particular, it both complements and has evolved from the programme of work on the chemical speciation of transuranic nuclides in the Irish Sea, recently completed within the framework of the Commission's Action "Radiation Protection Research" Programme (Contract: Bi7-042).

CONTRIBUTION OF CENTRO DE INVESTIGACIONES ENERGETICAS, MEDIOAMBIENTALES Y TECNOLOGICAS (CIEMAT)

The CIEMAT programme of work, within the framework of the Radiation Protection Research Action, will be carried out in the coastal zone off Palomares. This ecosystem offers an excellent opportunity to identify the pathways of transuranic migration as previous studies have demonstrated occasional transfer from land to sea in its neighbourhood.

One of the main objectives will be to acquire a clear understanding of the mechanisms governing the transport of radionuclides through canyons. The topography of the continental shelf in this area shows a group of canyons which are tributaries of the main canyon. The possibility of long-term transfer to the deeper sea-floor will also be examined.

A second objective will be to define the preferential pathways to man of plutonium and americium via the food-chain by studying the biogeochemical recycling of transuranics in the area.

The experimental work will be carried out in cooperation with the Department of Physics at Universidad Autónoma de Barcelona (SFR-UAB) and with the Spanish Institute of Oceanography (IEO). Regular interaction and exchange of scientific data with the other participants in the collaboration will greatly assist in the interpretation of the observations made in this zone.

Specifically, the following actions will be undertaken:

(a) The role of submarine canyons in the transport of radionuclides and the influence of the topography in their redistribution will be studied. The Palomares submarine canyon system will be compared with Italian coastal areas having similar characteristics and where evidence of transfer through canyons has been found (this will involve collaboration with the following institutions: ENEA, IEO and UAB-SFR).

(b) The mobility of plutonium and its possible post-depositional migration in Palomares marine sediments will be examined using sequential leaching techniques.

(c) The Almanzora River terrigenous input to the sea will be compared with other Mediterranean river inputs, e.g., the coastal area off Barcelona, the Ebro estuary, Valencia and Murcia.

(d) Radionuclide intercomparisons among the participants will be coordinated by CIEMAT using Mediterranean sediment as the sample matrix.

(e) The physico-chemical speciation of plutonium (and other radionuclides) in the Palomares marine ecosystem will be examined in order to evaluate their potential for uptake into the food-chain leading to man in collaboration with UCD and SFR-UAB. Emphasis will be given to the chemical speciation of plutonium in filtered sea water, colloidal associations of both plutonium and americium, and marine productivity in the primary food-chain (plankton). The uptake of radionuclides in this zone by benthic and pelagic feeders and crustaceans will also be studied.

Finally it may be added that the participation of some of the Spanish researchers involved in this project with the Group studying the radiological impact of non-nuclear industries (Phosphogypsum) is expected to enhance our collective understanding of the behaviour of natural and artificial radionuclides in the estuarine environment.

CONTRIBUTION OF CEA - INSTITUT DE PROTECTION ET DE SURETE NUCLEAIRE, UNITE DE RADIOECOLOGIE ESTUAIRES ET MILIEU MARIN (UREMM)

The programme of work to be undertaken at UREMM is as follows:

(a) Studies of particulate and dissolved plutonium in the Channel. In particular, studies will be conducted with a view to the following:

  (i) Estimating the fluxes of dissolved and particulate plutonium in the Straits of Dover,
  (ii) Measuring variations in the sediment-water distribution coefficient (K_D) as a function of various environmental parameters in the megatidal Seine estuary, and
  (iii) Estimating the transit time of particulate matter by measuring the $^{238}Pu/^{239,240}Pu$ activity ratio.

(b) Behaviour of plutonium nuclides in the Rhône Delta by *in situ* observations and experimental studies, and their dispersion in the Gulf of Lyons.

(c) Extrapolation of the measurements to ^{241}Am and ^{241}Pu and, in the future, to the rare earths.

(d) Participation in Group discussions on the physico-chemical speciation of the transuranics in open waters and estuaries. In this regard, links with other laboratories in the collaboration will be vigorously activated with a view to:

  (i) The implementation in the Cherbourg laboratory of the techniques (aluminium oxide sorption beds, ultrafiltration, chemical speciation) presently used at the UCD laboratory.
  (ii) Improvements to various analytical methods by experimental studies at Cherbourg (the ideas expressed by other laboratories will be tested in this way).

(iii) Preparation by UREMM laboratory of a future campaign of plutonium measurements in the Channel, the North Sea and the Arctic Ocean.

CONTRIBUTION OF ENEA - CENTRO RICERCHE AMBIENTE MARINO

The purpose of the ENEA participation in the programme is to define the fundamental mechanisms governing the distribution and behaviour of transuranics and other long-lived radionuclides in selected areas of the Mediterranean Sea.

The role of submarine canyons in the transport of transuranics and other long-lived radionuclides from the continental shelf to the slope and the deep sea will be extensively studied in the Gulf of Taranto (Ionian Sea) and off Palomares. The vertical distribution and inventories of these radionuclides will be determined in sediments from the two sites and from selected deep sea areas in the western Mediterranean Sea, characterized by pelagic sedimentation only or influenced by contributions of terrigenous material (Ebro and Rhône fans).

The study of a complex canyon system will be carried out in cooperation with CIEMAT, while the data on the western Mediterranean deep sea environments will complement CEA-ISPN-UREMM programmes on the Gulf of Lyons.

Studies on the role of different water masses in the physical transport of radionuclides present in surface waters or transported by rivers will also be carried out in the western Mediterranean Sea. The vertical profiles of long-lived radionuclides in the water column will be determined in selected areas of the basin, characterized by the presence of water masses of different origin (dense waters formed during the winter in the Gulf of Lyons and Levantine Intermediate Water originated in the eastern Mediterranean Basin). The areas of interest will be the Gulf of Lyons, the Balearic Sea and the main path of the Levantine Intermediate Water, close to the coasts of Sardinia and Sicily.

Sampling campaigns will be carried out in the western Mediterranean Sea at least once a year. For sampling in open sea, an Italian research vessel will be made available to all the participants in the present programme.

CONTRIBUTION OF THE MINISTRY OF AGRICULTURE, FISHERIES AND FOOD, DIRECTORATE OF FISHERIES RESEARCH (MAFF)

The MAFF contribution will be to provide information concerning the physico-chemical speciation of some transuranic nuclides (e.g., plutonium, americium and curium) and other radionuclides (e.g., cobalt) discharged into the eastern Irish Sea. In recent years discharges from Sellafield have decreased significantly as a result of improvements in waste treatment. Further, the introduction of the Enhanced Actinide Removal Plant (EARP), in 1993, will assist the reduction of α-emitting radionuclides discharged to sea. As a consequence of changes in operational procedures, it is likely that the physico-chemical forms of radionuclides may change, possibly causing some nuclides to become more mobile then before.

A preliminary study, using MAFF's research vessels, will be carried out to determine the physico-chemical forms by size distribution of present radionuclides discharges (prior to 1993) around the Sellafield outfall which will allow assessment of the significance of colloids toward the overall behaviour of radionuclides. Similarly, the impact of previous discharges will be evaluated by considering the colloid distribution of radionuclides found in the vicinity of the mud patch located in the eastern Irish Sea. Because of the decreased discharges, over a number of years, this area is now becoming a contributing source-term for those radionuclides which are particle-reactive in nature. A follow-up study (in 1993-94) will be carried out to determine the significance of radionuclides associated with colloids following the introduction of EARP. In conjunction with field studies, laboratory experiments will be undertaken to characterise the physico-chemical forms of radionuclides of effluents prior to disposal and to study the transformation of species following dilution of effluent into sea water.

Innovative techniques, recently developed by MAFF and UCD, will be used to independently characterise the different physico-chemical forms in the Irish Sea. Because of the diversity of these methods, it will be extremely valuable to compare and contrast the results of the different techniques. From the scheme of work outlined, and complementary work carried out by UCD, it will be possible to assess the extent by which colloid associations affect the overall transport, behaviour and bio-availability of radionuclides.

CONTRIBUTION OF UNIVERSIDAD AUTÓNOMA DE BARCELONA - SERVICIO DE FÍSICA DE LAS RADIACIONES (SFR-UAB)

It is known that the physico-chemical speciation of elements can change with biological and geochemical activity and the measurements proposed here should provide complementary information on the biochemical processes responsible for the observed changes in such speciation. Studies will be carried out by SFR-UAB on the transfer of transuranic and other radionuclides through the primary producer route into the food-chain leading to man (for example: sediment-water-plankton-fish-man). Zooplankton, which act as carriers of radionuclides to depth, may also bring radionuclides from depth to the surface. Moreover, the rate at which certain microbial and food-chain uptake processes occur may be influenced strongly by different temperature and salinity conditions. It should, therefore, prove valuable to compare this pathway in different marine environments (western Mediterranean, Gulf of Vera, Catalan - Balearic Sea and the Irish Sea) as it has not been sufficiently well studied in the past and is clearly of importance from the radiological protection point of view.

Programme of work

The tasks to be accomplished include:

(a) Measurements will be made on the concentrations of transuranics and other radionuclides in zooplankton and phytoplankton from surface waters of the western Mediterranean and the Irish Sea. In this way the concentration process in the primary food-chain will be determined.

(b) The colloidal state of various long-lived radionuclides will be investigated in the Catalan - Balearic Sea zone. Samples will be collected and fractionated using different particle-sizing equipment, including microfiltration and ultrafiltration. In addition, the magnitude of the colloidal fraction will be determined by sequential sorption.

(c) The vertical transport of transuranics in the Catalan - Balearic Sea will be investigated by (i) measuring the concentration versus depth profile and (ii) determining vertical fluxes using sediment traps. The results will be correlated with microbial species and number in similar samples.

(d) A selection of sediment cores, taken in the Catalan - Balearic Sea and the Palomares zone, will be analyzed for plutonium and americium to determine representative inventories for these radionuclides in sediments (the latter play an important role in the transfer of plutonium and americium to man *via* the food-chain).

Interaction with other participants

Extensive data on transuranic physico-chemical speciation and colloidal association, gathered in the course of joint research cruises in the western Mediterranean in 1991 and 1992 and presently being analyzed at UCD, will be utilised to refine the interpretation of the results derived above.

Collaborative sampling campaigns involving the SFR-UAB and other participants will be undertaken in the western Mediterranean and the Irish Sea in the course of 1993. Joint research cruises are foreseen for the Catalan - Balearic Sea (June 1993) and the Irish Sea (September 1993).

A22 Natural radioactivity in the environment and its pathways to man.

Contract FI3P-CT920035 Pathways of radionuclides emmited by non nuclear industries.

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Total Contribution by the Commission: 270 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

In contrast to the nuclear industry little is known about radionuclides emitted by the non nuclear industry. During 1990-1992 the CEC contract Bi7-006 "Behaviour of Polonium-210 and Lead-210 in European environments. Application of bioindicators" was executed. It provided a first reconnaissance of Po-210 and Pb-210 in four European estuaries and their enhancement by the Phosphorous industry. Based on its results the "Pathways" project has been started as a follow-up, and has been expanded with participants from Spain, Ireland and Denmark. The "Pathways" project comprises:

- Study, characterisation and quantification of the Po-210 and Pb-210 emitted by the Phosphorous industries into estuaries, either directly as effluent (NL, ES) or indirectly via erosion of phosphogypsum stockpiled on riverbanks (ES and PT).

- Additional field studies of abiotic and biotic distribution of radionuclides emitted by phosphate industries into estuaries to expand and verify the first findings and to look into annual fluctuations (NL, FR, ES, PT, IE).

- Reconnaissance study of the residual effects in the marine environment after closing-down of a wet process Phosphorous plant (DK, PT). This will be of general interest to other sites and situations in the CEC.

- Field studies of the uptake by mussels of Po and in a few cases of Ra-226 from effluents and/or natural background in different waters, geology, seasons (FR, ES, PT, IE, DK). It will give insight into the natural local, seasonal and geographic variation of Po-210 in mussels and the enhancement by different Phosphorous industries.

- Studies of soils, crustacea and fish of an inter-tidal marsh area flooded by estuary waters, on which effluents from Phosphorous industries are emitted (ES).

- Laboratory studies under controlled conditions of the actual uptake by mussels of Po-210 from effluents (NL). The mussels will be studied both in a season when they are in poor and when they are in good conditions. The studies aim to identify the bio-availability of Po-210 in: effluents of wet (i.e. phosphogypsum) and of thermal Phosphorous plants; the dissolved and the particulate fraction in the phosphogypsum.

- The effects of food preparation practices on Po-210 levels in the parts consumed by man of molluscs, crustaceans and fish (PT).

- Detailed dose assessments of Po-210 by consumption of fishery produce from natural marine waters and from estuarine waters exposed to inputs from stockpiled phosphogypsum (PT).

- General dose assessments of Po-210 by consumption of fishery produce either or not affected by enhancements from the Phosphorous industry (NL, FR, ES, PT, IE, DK).

- Modelling of the abiotic distribution of radionuclides in the Westerscheldt estuary into which effluents from wet and thermal process Phosphorous industries are emitted. Coupled with strategic field studies for model validation (NL). Intercomparison and checking of models on different estuaries (NL, PT). It will give insight to which extend the complex distribution processes in estuaries can be captured by modelling, when supported with parameter values from the dataset obtained by the project.

- Exchange and study by all contractors of experience, results, conclusions and further questions with respect to Po-210 and Pb-210 in natural and enhanced environments, in effluents from the Phosphorous industries, and other aspects as mentioned above. In order to obtain a better understanding of their general applicability, their complementarity, and into consistent geographical or other differences and their causes (NL, FR, ES, PT, IE, DK).

A22 Natural radioactivity in the environment and its pathways to man.

Contract FI3P-CT930075 Investigation on exposure to natural radionuclides in selected areas affected by U-processing.

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Total Contribution by the Commission: 90 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Solid wastes from uranium mining and milling have the potential to release radionuclides into air, groundwater, rivers or lakes. These radionuclides contribute to the radiation dose that can be delivered to the population living in the vicinity of the disposal site. The most recent radiation safety recommendations of ICRP raise the pressure towards determining the impact of the wastes from uranium industry and investigating appropriate countermeasures. In the past, measurements have been carried out around many disposal sites to determine the concentrations of the radionuclides being released to the environment and the resulting doses. But the data obtained in such measurements cannot be easily used to predict the long-term impact of the disposal sites, or to determine the consequences of a remedial action. These objectives can only be reached by a modelling approach based on a knowledge of the mechanisms that drive the transport of radionuclides to the environment. The development of realistic models on the dispersion of radon from the soil to the atmosphere was hindered by the difficulties encountered in treating the problem of large-area sources situated on uneven ground.

The modelling of the geochemical migration of the natural radionuclides was also delayed by a relatively poor knowledge of the behaviour of trace elements in the pore water of the residues and ground material. An effort has to be made to improve knowledge and develop new models in order to be able to predict in a realistic way the radiological impact of the mining and milling residues.

Given the international interest in these topics, a cooperative approach to future work was estimated to be beneficial. Possible opportunities for collaborative research were discussed between IPSN, France; BfS and GRS, Germany. The three parties decided to join their efforts in a multidisciplinary approach of the above issues, based on modelling and experimentation on typical test sites situated in France and Germany. It was agreed necessary to improve the evaluation of the radiological impact of uranium tailings and waste rocks disposal sites and to try to obtain a realistic assessment of such sites. The essential objectives of the planned research programme were to develop advanced models that describe in a realistic way the environmental dynamics of radionuclides released from waste materials; provide by experimentation on test sites the data that are required to drive the models and validate them by comparing experimental data and theoretical predictions; determine the kind and extension of measurements that will be necessary to evaluate other sites where such models can be applied.

The development of advanced models for the transport of natural radionuclides through the atmospheric and aquatic pathways will be undertaken by GRS. Such models will consider characteristics of the disposal site, nature and distribution of waste materials, radon fluence rate pattern, terrain orography, weather and hydrology conditions and the efficiency of soil or geotechnical barriers in preventing the migration of radionuclides. Considering the duration of the contract and the funds attributed, the effort will be in a first step mainly focused on the atmospheric pathway modelling.

The experimentations on test sites will be carried out by BfS and IPSN in Germany (Legenfeld) and in France (Jouac). There will be first a collection of data on site history, nature and amount of residues, main features of regional geology, hydrology and climatology. Then, both teams will acquire the data required to drive the models and validate them. In particular, the distribution of radon fluence rates, and the air concentrations of radon around the disposal site on study, will be determined for short periods of time (a few hours). During these periods, the weather will be characterized by monitoring the vertical profiles of wind speed and air temperature, and the radiative balance of the soil-atmosphere system. These data will be used to drive and validate the GRS model.

An exploration of the soil and aquatic pathways will also be initiated during the time available for the study in order to prepare the data necessary for a future validation of an hydrological model. The exercise will give also the opportunity to test and compare sampling techniques and analytical methods, and to establish monitoring programmes.

Contribution of IPSN / DPEI, Fontenay-aux-Roses, France

The IPSN team will acquire field data on the tests fields of Legenfeld (Germany) and Jouac (France), during two campaigns of 2-week duration each. These data are intended to be used primarily to drive and validate the models that will be developed by GRS.

At ground surface, the radon fluence rate will be determined at the nodes of a sampling grid designed to characterize the source term and the regional background. The method used to measure the fluence rate is based on the accumulation of radon in open-bottom containers resting on the ground surface. After a certain accumulation time, a sample from each container is taken in an evacuated alpha-scintillation flask. The samples are counted on a radon-counting system after three to four hours have elapsed to insure daughter product equilibrium. The fluence rate is calculated from the change in concentration of radon inside the container. The mesh of the mapping grid is fixed in function of the spatial variability of the fluence rate.

In the atmosphere, the average ambient concentrations of radon will be measured during small periods of time (one hour for instance) at specified points chosen in function of wind direction and terrain configuration. The concentration of radon will be obtained by using large-volume ion chambers. Meanwhile, average micrometeorological parameters will be obtained to characterize the weather conditions during the period of reference and be able to feed the theoretical model. There will be a determination of vertical profiles of wind speed and temperature from 0.1 m to 7 m above ground level, and also a measurement of thermal radiation fluxes. These measurements will allow the calculation of the Monin-Obukhov length that characterizes the stability of the atmosphere and hence the conditions of radon dispersion in the atmosphere.

Moreover, in collaboration with BfS, samples will be taken at different depths from the waste material and the substrate, in order to begin with an exploration of the geochemical environment, and a first approach of radionuclide migration through terrestrial and aquatic pathways.

Contribution of BfS, Berlin, Germany

Radionuclides from solid wastes from uranium mining and milling contribute to the radiation dose that can be delivered to the population living in the vicinity of the disposal site. In order to improve current methods for evaluation of the radiological impact of uranium tailings and waste rock disposal sites, suitable exposure models should be developed and examined at two tests sites in Germany and France.

BfS has the first task to collect data on site history, nature and amount of residues and main features of regional geology, hydrology and climatology at the test site Legenfeld. The second task is to carry out a first series of measurements of radon activity and alpha potential energy concentration in air, concentration of resuspended long-lived radon daughters in air, radon exhalation rate, radon concentration in soil air, concentration of uranium, radium-226 and lead-220 in samples from soil / rocks, plants, water and sediments as well as dose rate measurements and in situ gamma spectrometry at the site and its environment. Samples of the waste material are taken at different depths from bore holes.

These measurements are conducted in collaboration with IPSN. The results are compared to evaluate the accuracy of the measurements and to form the basis for some kind of quality assurance. In the same way, joint measurements are carried out at the french test site as to be agreed upon. All measurements are aimed at driving the exposure models developed by GRS and at validating the model predictions on dispersion of radon and radon progeny in air and migration of natural radionuclides through aquatic and terrestrial pathways.

Contribution of GRS, Köln, Germany

GRS will concentrate on the atmospheric pathway modelling. The data obtained by the two other teams will be used to drive and validate the models.

The mining and milling residues are often located on uneven ground. In order to make predictions of population exposure in complex terrain, a detailed modelling of flow and turbulence fields in the atmosphere is necessary. Coupled flow and dispersion models have been implemented at GRS either on a mainframe computer or on a microcomputer. The models enable the calculation of atmospheric dispersion under the influence of uneven terrain and inhomogeneous land use. They will be modified The atmospheric models will be validated from the measurements of IPSN and BfS. The measurements will consist in determining the meteorological parameters, and mapping the radon fluxes and air concentration around the disposal site during relatively short periods of time. The meteorological data and the flux pattern will be used as input to the models. The concentration data will serve to validate the theoretical predictions issued from the GRS models. The differences between the results of simple gaussian plume models and those of advances modelling techniques described above will be evaluated. The consequences of the dispersion and deposition of radon and its daughters in terms of potential human exposures will be evaluated by using a radioecological model.

The consideration of radionuclide migration in geosphere and hydrosphere, originally planned, has been postponed, because of CEC financial support reduction.

A23 Influence of speciation, chemical modification, changes in physico-chemical properties and biological conversion.

Contract FI3P-CT920010 The bio-availability of long-lived radionuclides in retion to their physico-chemical form in soil sytems.

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Total Contribution by the Commission: 220 KECU
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Description of research work

The participating institutes will study the relation between chemical speciation of radionuclides in soil systems and their bio-availability. The parameters which mainly define the relations between the compartments soil, plant and soil (micro)organisms will be investigated for a number of elements, caesium and strontium being the most important ones. The aspects which influence solid / liquid distribution and the aspects which affect uptake and accumulation will be studied separately and in combination, under controlled conditions. These data will be evaluated from available field observations. The final goal will be a generalized description of the relationship between the solid phase, the liquid phase and biota.

The results will benefit in judging the sensitivity of soils with respect to a potential contamination and the effectiveness of soil treatments and crop selection to inhibit contamination of the foodchain.

Objectives and expected achievement

Environmental impact of toxic substances and radionuclides is not only dependent on the total concentration but also on their chemical form. Identification of chemical species and of relevant variables influencing speciation makes predictions possible on movement through the biosphere and on effects of natural or artificial changes in ecosystem structure of functioning. In the ongoing project speciation of Cs, Sr and transuranic elements is studied in relation to their transfer along the soil-soil solution-plant pathway.

Objectives of this proposal are:

- Study the uptake of nuclides (mainly Cs and Sr) from the soil liquid, in relation to major characteristics of the soil.
- Study the efficiency of the uptake process (characteristics of the organism which affect uptake).
- Evaluate the effects of common soil treatments on solid / liquid equilibria in soils in order to select the most appropriate corrective measure to use following contamination of agricultural land with radiocaesium and radiostrontium.
- Rapid methods may be developed to predict the effect of countermeasures in the event of an accident.
- Assessment of the value of fractionation techniques meant to quantify the available fraction by comparing obtained results with uptake patterns.
- Study and model the role of soil microorganisms in the leaching and retention of caesium in soils.
- Develop a dynamic mechanistic model to simulate the time dependent soil - soil solution - plant root distribution of radiocaesium; sensitivity analysis of the model as a tool for efficient experimental design.

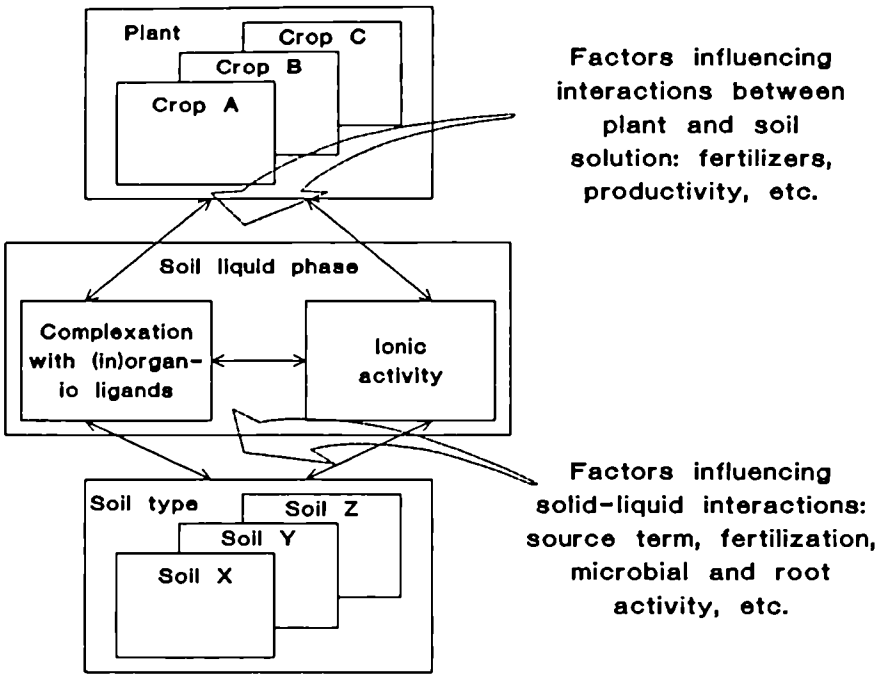

Figure 1: Studied interactions between soil, soil solution and plant

Scientific and technical description

Introduction

The transnational research project started in 1990, on "The bio-availability of long-lived radionuclides in relation to their physico-chemical form in soil system", focused on two of the major aspects affecting the behaviour of radionuclides in soil/plant systems : 1) availability in the soil liquid in relation to major characteristics of the soil and 2) the efficiency of the uptake process (figure 1). The nuclide of main interest was ^{137}Cs, although some experiments on transuranics have been done as well.

The general structure of the research items of this follow-up project will be comparable to those of the previous project. Whereas field measurements were an important part of the previous study, the follow-up project will mainly focus on the study of interactions between soil, plant and soil (micro)organism under controlled conditions. Indeed, the participants being new to this project will mainly contribute to this fundamental laboratory study.

Soil-related research items

* *Chemical speciation in soil and soil liquid in relation to major characteristics of the soil*

The speciation study will concern both naturally labelled soils (well-characterised sites in Cumbria) and artificially labelled soils (lysimeters and potted soil). It will accentuate the quantification of chemical forms in the soil and soil solution and the effect of microorganisms on the chemical behaviour.

Amongst others ultrafiltration and chromatographic techniques will be used to investigate the association of radionuclides with different molecular size fractions in solution and to monitor time dependent changes in speciation.

Further attention will be given to reversibility of the sorption of caesium from soils for which the specific interception potential was quantitatively characterized: desorption yields will be studied as a function of contact time (aging), desorption time and desorbing agent. A stepwise approach will be adopted to study the sorption of caesium, increasing the complexity of the system from simple mixtures to soil modified by root exudation and absorption.

* *Effectiveness of countermeasures*

The transfer of radionuclides from soils to plants depends on the activity concentrations in the soil solution of radionuclides and of other ions that might compete with radionuclides for uptake. Uptake can thus be reduced by: 1) reducing the radionuclide concentration in the soil solution, or 2) by increasing the concentration of competing ions in the soil solution.

By conducting batch equilibrium experiments with soils and their associated soil liquid the effects of various treatments on sorption of Cs and Sr are determined under laboratory conditions. Treatments will be those readily applicable to large areas, i.e. K, Ca, NH_4 and various forms of organic matter applied at about standard rates. In complementary laboratory procedures the potassium release characteristics and Sr/Ca selectivity in a range of different soil types will be investigated as a possible basis for setting optimum levels of K and Ca fertilization. The soils will include three diverse agricultural soil types (loam, peat and sand) currently under investigation at NRPB, in addition to peaty soils from Cumbria.

Plant-related research items

The study on the soil characteristics which define the solid/liquid distribution is complemented with experiments on bioaccumulation. The comparison between uptake from soils and uptake from nutrient solutions is further elaborated.

* *The efficiency of the uptake process*

Conclusions drawn from the current EC contract on the competition of nutrients and caesium for uptake will be checked for other crops. This serves a double purpose: 1) the differences between crops in nutrient uptake rate (i.e. the yield per unit of accumulated

nutrient) on their contamination level will be quantified and 2) it will facilitate an explanation and comparison of results on soil-to-plant transfer studies using different crops.

To further enlighten the relationship between the solid phase of the soil, the soil liquid and uptake by plants attention will also be given to Sr and to a nuclide showing a more complex chemical behaviour in the soil system, i.e. ^{237}Np.

- *Uptake from soil by plants*

Transfer along the soil / soil solution / plant pathway will be studied under controlled conditions in phytotrons, and validated with reference to available field observations on upland soils from Cumbria and data to be collected in the forthcoming growing season.

The time trend in uptake and im- and remobilization of radiocontaminants will be studied throughout the growing period in order to explain changes in transfer and to extend the information derived from the detailed soil studies. Attention will mainly be given to sandy and peaty soils. Effects of application of fertilizers (K, N and Ca) on uptake will be studied in relation with their effect on the solid / liquid distribution of the nuclides. The effect of plant roots in the surrounding soil (i.e. induced changes in pH and concentration of organic acids), and thus on the phytoavailability of caesium will be simulated. Factors such as porosity and soil moisture content will be taken into account.

Generalized description of relations between compartments

The bio-availability of radiocaesium depends not only on the chemical speciation of the isotope but also on its diffusive flux to plant roots and root uptake. It is impossible to isolate the individual weight of each of the many, often interrelated determinant factors from *in situ* measurements. Thus the extrapolation from experimental data obtained under controlled conditions, to field conditions is poor. Empirical models have been used to fit observed data, but are less successful at predicting the fate of radiocaesium in a particular soil.

As a result a dynamic mechanistic model of the relation between the solid phase of the soil, the liquid phase and biota will be developed taking into account the kinetics of caesium adsorption, desorption and fixation on soil constituents, diffusion to the solution-root interface and uptake characteristics of plant roots.

Distribution of major tasks between participating organisations

Although each of the participating organisations will work on different aspects of soil-to-plant transfer, they will accentuate specific topics and will have specific responsibilities, because of their experience, equipment and facilities. A schematic overview is given in the subsequent figure.

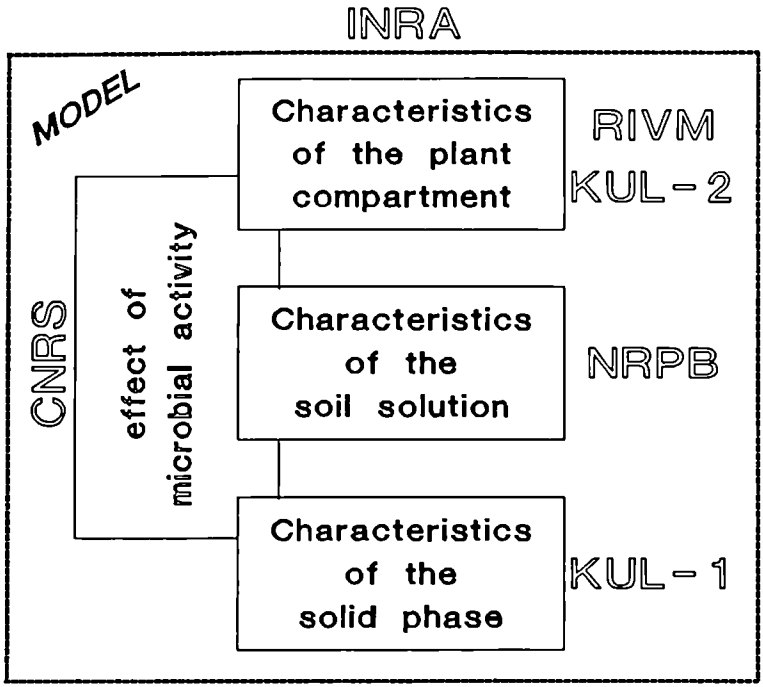

Figure 2: Specific expertise to be contributed by each of the participants

1, Rijksinstituut voor Volksgezondheid en Milieuhygiëne, Laboratory for Radiation Research

RIVM/LSO will act as coordinator of the project and will be responsible for the management of the project and editing of progress reports and final report.

Up to now RIVM/LSO has done experiments on bio-accumulation of ^{134}Cs by lettuce (*Lactuca sativa L.*) and earthworms (*Eisenia* sp.) under controlled conditions. The dual effect of changes in soil structural and chemical composition was investigated: these changes affect the concentration of the nuclide in the soil liquid phase and the efficiency of the uptake process.

The two effects were studied separately: 1) the effect of various mixtures of nutrients on uptake of nuclides was studied by growing crops on nutrient solutions and 2) the effect of e.g. various levels of fertilization on availability in soils was studied by following re- and immobilization of nuclides in soils prior to and during cropping of plants and by measuring radionuclide levels in the plant throughout the growth period. Effects of K, Ca and NH_4 on uptake and of K and Ca on availability have been studied. Relevant parameters (such as K- and Ca-concentration) of soil, soil liquid phase and plant material have been monitored intensively.

Two parts can be distinguished in the contribution of RIVM/LSO: 1) experiments will be done to further elucidate transfer of Cs from soil to plant and earthworm and 2) transfer of another radioelement will be studied in order to more thoroughly test the relationship between accumulation by plants and soil solution chemical composition.

1) The effect of some nutrients on the uptake of caesium by roots of lettuce was described in the previous project. Further attention will be given to the behaviour of Cs in the soil-plant system in order to allow a better comparison of results of experiments with plants growing on soils and results of experiments with hydroponic systems. Experiments will be done with soils investigated by other participants of the project in order to link results obtained with the different experimental systems. The study on factors affecting the accumulation of radiocaesium by earthworms will be further elaborated.

2) To further enlighten the relationship between the solid phase of the soil, the soil liquid and uptake by plants attention will also be given to a nuclide showing a more complex chemical behaviour in the soil system, i.e. ^{237}Np. Distinct chemical species will be present in the soil liquid, the bio-availability of which is thought to be different. RIVM/LSO will study these fractions and their availability for a limited number of systems. The results of a fractionation will be compared with the observed accumulation pattern. The crop, soil type and other experimental conditions used for these experiments will be the same as those used in the study on caesium.

2, Katholieke Universiteit Leuven, Laboratory on Colloid Chemistry

The objectives of KUL in the context of this project are:

1) The quantification of the kinetics of immobilization of radiocaesium in a broad range of soils (sandy, loam, clay, forest). Such study will be carried out on laboratory- and field-contaminated soils. The aging process will be monitored as a function of time using drying-wetting cycles. *In situ* solid-liquid distribution measurements and measurements of fixation levels are the key experimental approaches in this part of the project.

2) The development of diagnostic criteria which will allow to predict the caesium fixation capacity of soils. This aspect deals with the problem of quantitatively identifying problem soils and represents a first step in setting up a vulnerability chart of soil groups.

3) The study of the soil-to-plant transfer process of radiocaesium in two types of problem soils (podzol, peat) which have been thoroughly characterised in terms of radiocaesium interception properties and desorption dynamics.

The various soils used in field experiments or phytotron uptake studies by the other partners (RIVM, NRPB and AEA) will all be included in our experimental protocol (1). A set of representative soils which are studied by NLH will also be included in this protocol. In addition a range of forest soils, as supplied by ENEA and originating from the Chernobyl area will be included in our studies, thus providing a link with ECP-5 (CHECIR).

3, National Radiological Protection Board, Environmental Investigation Group

The objectives of NRPB in the context of this project are:

1) To evaluate the effects of common soil treatments on solid-liquid equilibria in soils in order to select the most appropriate corrective measure to use in a range of different circumstances, following contamination of agricultural land with Cs and Sr.

2) to develop rapid methods that can be used in the event of any future accident to predict what the effects of these different countermeasures might be.

3) To contribute to collaborative studies on the relationship between the uptake of radionuclides by plants and their behaviour in soil solutions.

In the event of a large accident, intervention in agricultural and semi-natural ecosystems might be required over relatively large areas in order to return this land to useful agricultural production as soon as possible. One possible remedial measure is the application of soil treatments to reduce radionuclide uptake by plant roots.

The transfer of radionuclides from soils to plants depends on the activity concentrations of their different chemical forms in soil solution and the concentration of other ions competing with radionuclides for uptake. Uptake to plants can thus be reduced by one of two means: i) reducing the radionuclide concentration in soil solution, and ii) increasing the concentration of competing ions in soil solution.

An objective of NRPB's work is to evaluate the effects of common soil treatments on these two parameters. Emphasis will be placed on developing a better understanding of solid-liquid equilibria in soils in order to predict more reliably the longer term outcome of potentially corrective measures. This is directly relevant to the work of the other collaborators, on the dynamics of radiocaesium release from the solid phase, and complementary to studies on the effect of ion competition at the plant root surface.

K is the most effective countermeasure for radiocaesium contaminated soils, whilst Ca and to a lesser extent organic matter are good ameliorants for radiostrontium. Laboratory procedures to determine K release characteristics in a range of different soils will be investigated as a possible basis for setting optimum levels of K fertilization, following their accidental contamination with radiocaesium. Similarly for radiostrontium, procedures to determine Sr/Ca selectivity coefficients will be followed for a range of soil types in order to identify those soils that will respond most favourable to lime.

Complementary studies involving a modified batch equilibrium technique will be used to provide a small scale screening procedure for those treatments, and treatment rates that seem to be most appropriate. This will provide supplementary information on positive or negative effects of each treatment on the distribution of radionuclides and their stable nutrient analogues between the solid and liquid phases.

The project will be confined to those treatments that could readily be applied to large areas, i.e. K, Ca, NH_4, organic matter, applied at standard and greater than standard rates. The soils to be investigated will include a loam, peat and sand that have already been well characterised as part of a previous five year lysimeter study conducted at

NRPB. These agricultural soils are typical of those found in Northern Europe. Several equally well characterised organic soils from an upland site in Cumbria will also be studied because of the sustained bio-availability of radiocaesium in semi-natural ecosystems such as this. This laboratory based study will involve the contamination of fresh samples of soil with radiocaesium and -strontium. The effectiveness of various soil treatments will then be tested after delays of up to one year. Soil solution samples required for the batch equilibrium experiments will be obtained *in situ*, using porous ceramic cups installed on land from which the soil samples were taken originally.

Although NRPB's effort will be principally on the countermeasures investigation described above, lysimeter soils and soil solutions will be available for collaborative studies on ultrafiltration, sequential extraction and desorption kinetics. Similarly porous pots installed in Cumbria will be maintained, and soil solution made available.

4, Katholieke Universiteit Leuven, Lab. of Soil Fertility and Soil Biology

Radionuclide uptake as a soil-to-plant transfer phenomenon is conceptually identical to nutrient uptake processes and should therefore be treated by similar formalisms. It is the main objective of this proposal to develop and test these general principles to be in a position to propose measures to reduce contamination levels in the food-chain through a judicious selection of crops.

In general, the radionuclide uptake by plants is either limited by its supply from the soil or the uptake itself is limited by the assimilation potential of the plant. In the first case, genotypic differences in uptake must have their origin in differences in root morphology or growth rate. In the second case different *radionuclide uptake rates* would be a result of different *nutrient uptake rates*. Herein "nutrient" stands for an element, chemically most closely related to the radionuclide. Essentially, this means accepting the existence of a constant K, relating nutrient/radionuclide concentrations in the plant to those in the surrounding medium (the soil solution) under equilibrium conditions. The relation is as follows:

$$K = \frac{(X)\text{plant} \;/\; (Y)\text{plant}}{(X)\text{soil solution} \;/\; (Y)\text{soil solution}}$$

wherein (X) are the concentrations of the radionuclide and (Y) those of the resembling nutrient. There are some well known examples of X/Y pairs such as Sr/Ca or Cs/K.

A second assumption is that K is a constant for different species, grown at equal conditions. Genotypic variation in $(X)_{plant}$ can thus be reduced to genotypic variation in $(Y)_{plant}$. Plants with high potassium productivities (= high yields per unit of K taken up) and low (Y) values will thence have lower ^{137}Cs concentrations.

The validity of the "K" concept and the assumed links between nutrient and radionuclide concentrations will be tested using different plant species and growing them under similar conditions with different X/Y ratios in the surrounding medium. In particular the Cs/K and Sr/Ca pairs will be investigated in detail. Experiments will be conducted using spinach (low productivity) and barley (high productivity). In order to mimic soil solution concentrations and to have easy access to nutrient/radionuclide concentrations, these experiments will be conducted in controlled environments.

5, Centre National de la Recherche Scientifique, Lab. de géomicrobiologie

The influence of microorganisms on the functioning of the biogeochemical cycle of radiocaesium will be studied to understand its mobility and transfer.

The purpose of the study is to analyze, in laboratory and field experimental models, the role of soil microorganisms and most particularly of cellulolytic microorganisms in the leaching and retention of caesium in soils significantly present around nuclear sites. Typical large exchange capacity clay minerals will be used as model of contaminated material.

In the field the *in situ* bag method will allow to determine the mobility of caesium provided by contaminated clays and the possible retention by cellulolytic microflora comparatively to soil constituents. The main experiments will be done in the laboratory by using semi-continuous flow devices with soil columns and with batch incubations to study the leaching, the complexation, the biosorption and the bioaccumulation of caesium by selected cellulolytic microorganisms.

The experiments will not only provide information on the influence of microorganisms on mobility and transfer of radiocaesium to the soil solution, but also on the ability of soil microorganisms to degrade soil organic matter and to produce acid and complexing agents.

6, Institut National de la Recherche Agronomique, Station de Science du Sol

A dynamic mechanistic model will be developed to simulate the time dependent soil - soil solution - plant root distribution of radiocaesium. The kinetics of caesium adsorption, desorption and fixation on soil constituents, diffusion to the solution-root interface and the uptake characteristics of plant roots will be taken into account. Input data will be taken from the literature and from the investigations carried out in the context of this project.

The model will be tested using the available experimental and *in situ* data. Experimental measurements will be made to elucidate the mechanisms of adsorption and fixation of caesium on clay minerals in association with soil organic matter.

Sensitivity analysis will enable the model to be used (i) as a tool for efficient and cost effective experimental design and (ii) to determine the most appropriate choice of plant species and soil treatments following the contamination of a given soil to minimize bio-availability and hence entry in the human food chain.

The model will be kept as simple as possible to facilitate its insertion into models taking into account a larger ecosystem and modification of the boundary conditions to allow for deposition modes other than rapid wet deposition.

A23 Influence of speciation, chemical modification, changes in physico-chemical properties and biological conversion.

Contract FI3P-CT920022 Investigations and modelling of the dynamics of environmental HT/HTO/OBT levels resulting from tritium releases.

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Total Contribution by the Commission: 120 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Introduction:

Progress in the technical realization of fusion involving the handling and storage of kilograms of tritium within a single facility and the potential of accidental releases of those quantities, stresses the necessity to predict time courses and maximum levels of tritium concentrations in environmental compartments especially after short-term emissions and to provide reliable analytical methods for the quantification of dose-relevant tritium compounds in environmental samples.

The urgency to improve the understanding of tritium behaviour in the environment is stressed by the fact that in November 1991 tritium fuelling and D-T plasma reactions have been performed at JET initiating an operation phase of substantially different radiation protection qualities with respect to tritium than the preceding phases. Besides the enhancement of precision in dose predictions from tritium releases the proposed research is also meant to affect the design of Next Step Devices in the sense of possibly limiting tritium inventories and potential release amounts to levels which meet radiation protection regulations, in order to incorporate a high degree of passive safety. In this context radiation protection is directly depending on the certainty and accuracy of dose predictions.

Objectives, state of the art and expected achievement:

At the OECD/IEA Tritium Workshops it was agreed necessary to reduce the uncertainty factor of dose predictions below three, and several environmental tritium processes were identified, which need further investigations, in order to achieve this goal. Two essential topics of this list have been selected for intensive studies within this research proposal:

a) Parameterization of tritium re-emission after HT or HTO deposition to soils representing a secondary source of air-HTO and

b) Plant-specific variations and dynamics of tritium uptake, OBT formation and translocation in biological systems.

In case of HT releases, tritium re-emission from soil in the HTO form was found to dominate the inhalation and skin absorption dose contributions and to be the source of immediate appearance of tritium in vegetation. From the knowledge on water cycling in the atmosphere-soil system it is expected that the re-emission rate is depending on a number of meteorological and soil physical parameters. Consequently, it will be largely site and weather-specific and will vary considerably with respect to time of day and season of the release occurrence. All those variations are superimposed by the general tendency of a decreasing re-emission rate with time, as the availability of soil-HTO is reduced. Evaluations of data obtained in the HT field experiments performed in France and Canada have yielded re-emission rates with low time resolution, and they refer to the given conditions of the experiments, so that no parameter dependencies could be derived.

In order to establish single-parameter dependencies of the re-emission rate, laboratory experiments will be performed at NIR with help of a wind-tunnel/soil column arrangement under controlled conditions. The information will be used to develop a re-emission model. Small-scale field experiments will be carried out at CEA-IPSN under natural conditions using a special air-HTO collecting device, which is installed in the downwind direction of a small tritium labelled soil plot. NIR findings and the derived model will be validated against the field results.

For this purpose it is planned to use the field soil in the laboratory device and to exchange scientists for participation in joint experiments and evaluations. As the radiotoxicity of organically-bound tritium (OBT) is higher and its transfer behaviour along the food-chain is different compared to that of tissue water tritium (TWT), "enrichments" of OBT in biological samples, expressed as an OBT/TWT-ratio (R-value) exceeding unity, which are frequently reported in the literature, have raised special attention. However, there is no unique method yet, to reliably determine OBT (exchangeable and non-exchangeable), and a considerable number of high R-values may be attributed to analytical artifacts. They easily occur in the analytical procedure, as the extraction of one tritium species shifts the original partitioning of the different compounds in the biological sample. Therefore, TUM is planning to develop an accurate method to determine OBT and to use this method in evaluating R-values in biological samples of the food chain under steady state exposure conditions.

The OBT analysis will be adopted by KfK in investigating plant-specific parameters for tritium uptake, OBT formation and translocation into diet-relevant plant organs. It can be concluded from estimations that neglecting environmental and physiological conditions for tritium uptake and OBT formation rates in vegetation and using mean values instead, may mispredict ingestion doses up to one order of magnitude. Therefore, research in this contribution will focus on the parameterization of the dynamics of the processes, in order to develop a refined submodel of tritium in vegetation under short-term exposure conditions.

Distribution of tasks, collaborative links and overall benefits: The two main topics of tritium re-emission and tritium speciation in vegetation are distributed among two participants each. This concept ensures the complete coverage of all associated aspects and the promotion of close cooperation and diversity of the scientific and technical approaches. The link between the two topics is given by the fact that the source terms for HTO uptake through the aerial and subsoil organs of the investigated plants will be supplied by the results on re-emission and the complimentary soil-HTO contents. Both submodels will be designed for incorporation into existing tritium codes (eg. UFOTRI, ETMOD).

The improved knowledge on environmental tritium behaviour and dose consequences of tritium releases will contribute to the lowering of the uncertainty factor in dose predictions, and it will help in the design of Next Step Fusion Devices to incorporate a high degree of passive safety by possibly limiting tritium inventories and potential release amounts. The proposed research should also contribute to the information necessary for the development and timing of effective counter-measures in case of accidental releases of large amounts of tritium.

1. Investigations on Tritium Re-emission from Soils: Laboratory Studies and Modelling (NIR)

Re-emission of tritium from soils has been identified a major contribution to the radiation exposure from tritium releases. Estimations performed with data of the tritium field experiments in France and Canada have shown that in the case of HT releases inhalation and skin absorption doses from re-emission are 2 to 3 orders of magnitude higher than those from the primary plume, and in case of HTO releases the direct dose from the primary and re-emission plumes are of the same order of magnitude. Furthermore, re-emission is the source of immediate appearance of tritium in vegetation after HT releases. As from the results of the field experiments no basic relationship between the re-emission rates and environmental parameters could be derived, a soil-column/wind-tunnel laboratory device has been installed, to perform single-parameter studies under controlled conditions. The experiments conducted so far show a two-phase time behaviour of the re-emission rate and a pronounced air/soil-temperature dependency. A notable finding is that with initially tritium-free air the re-emission rate of HTO is higher than the evaporation rate of H_2O. This agrees with the respective observation from the reverse process of HTO deposition to soil. During the 92/94 contract period research will focus on dependencies of the re-emission rate on air-HTO and H2O contents, considering pre-contamination of the upwind air, as well as on soil physical parameters, like soil type, moisture regime and HTO distributions within the soil. For comparison and verification of the laboratory and field results, also soils from the small-scale field experiments of CEA-IPSN will be used and respective environmental conditions will be simulated in the lab device for re-emission rate evaluations. The model derived from the lab experiments will be validated against the field results, and joint experiments and evaluations will be performed both in the lab and in the field.

2. Study of the Re-emission of Tritium from Soils: Small-Scale Field Experiments and Model Improvements (CEA-IPSN)

Tritium re-emission in small-scale field experiments

A new technique is being developed to study the re-emission of tritium from a small area (0.1 m2) of contaminated soil, under field conditions prevailing in the open air. The small area of soil surface is labelled by introducing a small amount of tritium (10 MBq) into a field chamber placed onto the soil surface. When the tritium is completely deposited, the chamber is removed and a determination is made of the amount of tritiated water that escapes from the contaminated area during a given period of time. This is achieved by measuring wind speeds and concentrations in a cross-section of the plume and calculating from the measured values the horizontal flux of HTO in the downwind direction. In parallel, measurements are made of soil and micrometeorological parameters to characterize the conditions of tritium transport in the soil column and the atmosphere. Such a determination can be repeated during consecutive periods of time, thus allowing the evaluation of the re-emission flux at different times after exposure.
This technique will be applied to soils of the Cadarache site. One of the main objectives will be to determine the evolution of the re-emission flux in the hours following the deposition to the soil of a small amount of tritium gas or tritiated water vapour. This will be done for typical soil and weather conditions. The results obtained will be compared to those derived from laboratory experiments (see NIR-programme) and from theoretical models that describe the tritium deposition and re-emission.

Improvement of models

The existing models are based on a simplified description of the transfer of tritium and water in the soil-plant-atmosphere system. The models assume that the soil is uniformly wet ant that the soil humidity is constant during the re-emission period. Unfortunately this situation is not the most frequent, diurnal fluctuations in soil heat flux are responsible for the common observation that after rain, even though the soil mass is wet, the surface frequently dries out each day and becomes wet again at night. In this case, the exchange of tritiated water cannot be dissociated from the evolution of the soil humidity profile at ground surface. The existing simplified model will be tentatively improved to account for the effect of water inhomogeneity in the soil boundary layer. This improvement will be made in conjunction with the experimental efforts made in the field (see above) and in the laboratory (see NIR-programme).

3. Chemical and Biological Behaviour of Tritium in Tissue Cell Systems of the Food Chain and Accurate Determination of Organically Bound Tritium (TUM)

The radiotoxicity of tritium increases by conversion from tissue water tritium (TWT) to organically bound tritium (OBT), which is retained much longer in the biological system. This increase might even be enhanced and OBT accumulated along the food chain by eventual enrichment processes resulting from isotope effects in enzyme reactions. Biochemical tritium conversion is quantified by a fractionation factor, expressed as the ratio (R) of OBT and TWT. Our previous work indicates that R values exceeding unity can arise from analytical artifacts through the tritium enrichment involved automatically in the separation of TWT and OBT through vaporization. There are two kinds of OBT, i.e. exchangeable (-OT) and non-exchangeable (-CT) and several kinds of TWT, i.e. free HTO, solvated HTO to salts in tissue water and absorbed HTO to the cell walls. The differentiation of OBT from TWT by experiment accompanies, according to physical nature, a tritium enrichment in bound tissue water which subsequently enriches tritium into exchangeable OBT. Such a process involved in analytical procedures may lead to a wrong interpretation of the biological uptake of tritium and consequently to an unreliable assessment of its radiological effect. Therefore, TUM first intends to develop an accurate analytical method for the determination of OBT and TWT. The method to be developed comprises the measurement of well differentiated fractions of non-exchangeable and exchangeable OBT in a given biological sample without analytical artifacts. This includes an evaluation of several overlapping isotope effects involved in separation of tissue water from bioorganics, which might enhance the amount of exchangeable OBT and hence increase the total OBT. The analytical fractionation processes to be analyzed require basic investigations as well as quantification of isotope effects at solvation, adsorption and at vaporization under the laboratory conditions. The method must further comprise an accurate assessment of hydrated water bound onto salts in tissue water and cell walls, which eventually influences the OBT measurement.

The method will be directly applied for the accurate measurement of biological tritium fractionation through the accurate determination of R-values of biological systems during a full growth period under steady state tritium exposure. The KfK group will adopt the method for OBT measurement in the investigation of OBT translocation in diet relevant

plants. The analytical problems connected with the previously mentioned isotope effects at the separation of free water from exchangeably bound water will be investigated also for the soil/water system by analogy with the biosystem, which will be of benefit to the NIR and CEA groups to consider the mobility of tritium associated with soil humus and mineral lattices.

4. Elaboration of Plant Specific Data Sets for the Production and Translocation of OBT in Diet Relevant Plants (KfK)

When tritium releases into the environment occur during the vegetation period the formation of organically bound tritium (OBT) in crop plants has to be considered in dose estimations due to the ingestion pathway. The formation of OBT in green leaves depends on weather conditions, light intensity and plant physiological parameters during the tritium release. Depending on the developmental stage of the plant, OBT is partly translocated into fruits or other storage organs.

The objective of the project is to investigate by experiments, how much of the OBT formed in the leaves is translocated into edible parts of nutriment plants and how much remains there until harvest. Additionally to the translocation experiments, the relevant physiological parameters of the plants are recorded under laboratory conditions as well as under natural conditions. These data sets allow the calculation of HTO uptake and formation of OBT in a given stand of vegetation in dependence of the day time, the actual weather conditions and the developmental stage of the plants.
Translocation of OBT:

Wheat and potatoes were selected as experimental plants because their storage organs belong to the basic food-stuffs of man. Plants in different developmental stage will be exposed to atmospheric HTO for several hours in a controlled environment. Then they remain in an uncontaminated atmosphere until maturity of grains or tubers. During the analytical procedure of the plant samples the exchangeable OBT will be removed. An accurate method for the determination of OBT, which will be developed by another contribution to the coordinated project (TUM), is of great importance for this project.

Elaboration of plant specific data sets:

The physiological parameters of the leaves, like stomatal resistance, transpiration and net-photosynthetic rates, are recorded with a gas exchange measuring device. The parameters are calculated from the measured climatic data, the differences of the H2O- and CO2-concentrations in the atmosphere, and the area of the enclosed leaf. It is necessary to elaborate diurnal and seasonal trends of these parameters, in order to assess the theoretical tritium uptake into tissue water and OBT at a given HTO-concentration in the atmosphere. The results may be compared with experimental observations.

A24 The behaviour of accidentally released radionuclides, evaluation of the reliability of transfer parameters and experimental studies.

Contract FI3P-CT920006 Transfer of radionuclides in animal production systems.

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Description of research work

BACKGROUND

The studies performed by the group within the previous CEC DGXII-F-6 programme tried to identify and quantify some of the most important factors responsible for determining the levels of radiocaesium in animal products. The importance of radiocaesium bio-availability was highlighted. The studies attempted to identify the underlying mechanisms governing radionuclide transfer, rather than using empirical measurements such as concentration ratios, and successfully isolated the transfer from feed to blood plasma. This has been done by establishing methods to determine true absorption of radiocaesium which indicate that the true absorption coefficient (A_t) should be used as the gastrointestinal absorption factor (f_1) in predictive models. A_t did not change with age or between lactating and non-lactating sheep (but F_m, the transfer coefficient, did change over the lactation period), and similar values were estimated for sheep of differing breeds and dairy cattle.

The current programme primarily concentrates on three mobile radionuclides: radiocaesium, radiostrontium and radioiodine. Studies on radiocaesium within this programme concentrate on areas not covered in the previous contract or highlighted during the previous programme as needing further work, for instance in trying to explain anomalous results in the soil adhesion studies. Previous studies of radiostrontium have often used the OR ratio (Sr/Ca in animal tissue: Sr/Ca in feed). Although useful, this gives little information on the mechanisms governing the behaviour of radiostrontium in animals. A similar approach will be adopted to that used previously for radiocaesium, to study dietary and physiological factors which may influence radiostrontium levels in animals. A number of parameters which are known to influence stable iodine metabolism will be investigated with regard to radioiodine transfer, again using approaches adopted in the previous programme. The aim is to determine whether the range of environmental conditions prevailing throughout Europe significantly affect radioiodine transfer to animals.

Additionally the group will attempt to identify effective chemical binders to reduce radiostrontium transfer to animals and also to develop a combined countermeasure to reduce both radiostrontium and radiocaesium simultaneously.

OBJECTIVES

1. To provide improved information on the behaviour of radionuclides in animals to enhance the ability to predict contamination levels in foodstuffs.
2. To measure A_t for radiostrontium and radioiodine with the aim of substituting these physiological parameters for the more mechanistic transfer factors in predictive models.
3. To investigate factors affecting plasma to tissue transfer of radiocaesium.
4. To improve the range of available countermeasures, especially for radiostrontium.
5. To identify important factors which may affect the behaviour of radioiodine in the environmental and agriculture conditions prevailing throughout Europe.

6. To investigate the characteristics of soil particles adhered onto vegetation surfaces, and its effect on soil ingestion by ruminants.

1. ABSORPTION

The true absorption coefficient (A_t) can be used to describe the transfer of radiocaesium across the gut wall and provides a useful parameter that should be used as an f_1 in predictive models. We intend to extend this measurement to other elements. In particular to make direct measurements of interactions of radiostrontium-dietary Ca at the gut level to give detailed physiological information than is given by the observed ratio (OR).

(a) Gastrointestinal absorption of radiostrontium

The proposed studies on radiostrontium will compare the relative importance of stable element (Ca and Sr) levels in feed with differences in the bio-availability of radiostrontium in determining levels within tissues and milk. This will complement, internally-funded studies to determine the bio-availability of radiostrontium from different sources.

(i) Effects of dietary Ca and stable Sr on the absorption, milk secretion and tissue deposition of radiostrontium.

Whilst much attention has been paid to dietary radiostrontium:Ca ratios and their influence on radiostrontium transfer, little is known of how environmental and physiological factors may affect the absorption of radiostrontium, or the reasons for the observed fluctuations in OR values for milk of 0.04 to 0.2. The effects of dietary Ca and Sr levels on absorption, milk secretion and tissue deposition will be studied.

(ii) Bio-availability of radiostrontium

We have found that the bio-availability of radiocaesium from a range of sources varies over two orders of magnitude. Similar measurements for the availability of radiostrontium are not available. Therefore the availability for absorption of different sources of radiostrontium will be measured. The bio-availability determined by measuring A_t of radiostrontium incorporated into clover will be compared with that of ionic radiostrontium. The clover diet will be offered to sheep as silage and in a dry state. The availability of ^{90}Sr from a sandy soil from within the 10km Chernobyl exclusion zone for transfer to milk will be determined.

(b) True absorption of radioiodine

Preliminary studies to devise suitable methods to measure the true absorption coefficient for radioiodine will be conducted.

2. METABOLIC BEHAVIOUR OF RADIOCAESIUM

a) Radiocaesium metabolism

Parameters describing blood plasma-tissue transfer will be evaluated as a logical extension of the use of the true absorption coefficient in prediction models. Measurements will be made in ewes of differing physiological status in an extension of a study funded by other sources.

It is unclear if order of magnitude differences in transfer coefficients between cattle and sheep/goats are determined by metabolic differences or related to differences in pool size. The models we have developed for adult sheep suggest that by altering body size as a parameter, realistic estimates of transfer coefficients can be derived for sheep and cattle. We will try and differentiate between "body size" and other factors in the transfer of radiocaesium to ruminant tissues. It may then be possible to extend body size: radiocaesium transfer relationship to other ruminants.

b) Effect of stable caesium

Recent studies on the interaction between stable ^{133}Cs and radiocaesium have shown that radiocaesium transfer coefficients were not valid when animals were given rations which had a comparatively high content of stable caesium. Therefore, the range of ^{133}Cs intakes for which radiocaesium behaves as a true tracer needs to be established. The effect on F_m, F_f, A_l and A_s in lactating goats of varying the level of ^{133}Cs intake over a range of 2 orders of magnitude from 5-500 mg ^{133}Cs will be measured. The study should help to improve understanding of the potential use of ^{133}Cs as a countermeasure.

3. EFFECT OF ENVIRONMENTAL FACTORS

(a) Environmental factors affecting radiocaesium transfer

(i) Grazing effects

Modelling studies within the current programme and independent field studies have suggested that the grazing pressure on a pasture may affect the radiocaesium activity of grazing animals. Therefore it is possible that changing management of grazed pasture after an accidental release could be used as a 'farm management countermeasure'. Combined field and lysimeter studies will investigate the effect of grazing regimes on radionuclide levels in grazing animals. Rates of soil adhesion under varying grazing regimes will also be studied.

(ii) Factors affecting soil adhesion onto vegetation

Previous studies within the programme suggest that Ti may not be a good marker for Cs on plant surfaces. A study of the type of soil particle found on plant surfaces will be carried out to try and quantify the importance of soil adhesion in a range of different circumstances.

(b) Environmental factors affecting radioiodine metabolism

The effect of various environmental factors which may influence the transfer of radioiodine to milk are not fully understood. The following topics will be investigated to enable us more fully to understand the transfer of radioiodine in the environmental conditions prevailing throughout Europe.

(i) Effect of feed type

The presence of goitrogens, present in various forage species (eg brassicas and legumes) can have a considerable effect on stable I metabolism by ruminants. However, although they are often important feedstuffs for dairy animals (for instance brassicas in the winter) it is currently difficult to assess their effect on radioiodine transfer. We intend to study the effect of feeding different goitrogenic containing feedstuffs to dairy goats on the transfer of radioiodine (and Sr) to milk and the thyroid.

ii) Environmental temperature

Environmental temperature can affect the stable I metabolism of animals (since it influences the rate of thyroid hormone secretion). It is possible that within the range of environmental temperatures found within EEC countries significant differences in stable I and hence radioiodine metabolism may be expected. We will measure the transfer of radioiodine to the milk and thyroid of dairy goats under two different controlled temperatures. Consideration will be given to the possibility of extending this study to other radionuclides (eg Cs and Sr).

(iii) Stable Iodine

Measurements of the transfer of radioiodine to cow milk vary from 0.001 to 0.19 d^{-1}. Some of this variation may be due to differences in environmental stable I levels found in different European regions. Additionally dosing cattle with stable I may block thyroid uptake of radioiodine, but increase the activity concentration in milk. Depending on the progress made in developing methods of measuring the A_1 of radioiodine the influence of different dietary I levels, typical of those occurring in Europe, on the true absorption of radioiodine may be addressed.

4. COUNTERMEASURES

Hexacyano-ferrate compounds are effective radiocaesium binders reducing absorption in the gut. Practical application of these binders, in combination with other countermeasures developed since the Chernobyl accident, have considerably improved the range of available countermeasures that can be used to minimize radiocaesium levels in grazing animals. However fewer effective countermeasures are available for the other radiologically significant isotopes (radiostrontium and radioiodine). Although the use of Ca to reduce the transfer of radiostrontium has been shown to be effective, the usefulness of some of the other suggested countermeasures is very limited. High affinity binders which can be used in small daily quantities are particularly lacking.

We intend to try to identify effective chemical binders/countermeasures for radiostrontium using a combination of laboratory studies of binding capacity and animal feeding trials. Additionally we will attempt to develop a single chemical countermeasure which will be effective against more than one radioisotope. The potential use of modified smectites with a high binding capacity for both radiostrontium and radiocaesium, or the inclusion of a radiostrontium binder in the AFCF boli will be considered.

5. MODELLING

The models developed during the radiocaesium studies have been used to identify areas which require further research (such as grazing pressure and body size). These suggestions have been incorporated into the above proposals, and the resulting data will be used to further improve the model. Data on the behaviour of radiocaesium within the ruminant gut, from the final stages of the previous contract will also be incorporated into the model.

The types of factors which may affect radionuclide transfer outlined above are complex and difficult to separate without a clear framework of understanding. As in the previous programme, this will be achieved by parallel modelling studies which provides a control for the experimental work.

It is intended to develop models for radiostrontium and radioiodine which can be used to identify areas where our understanding is inadequate. Experiments within the programme will provide data on the dynamic behaviour of both radionuclides. These data will be used, in combination with previously published information, to develop and evaluate these models.

SUMMARY OF STUDIES AT ITE, MERLEWOOD

ITE Merlewood co-ordinates the programme; organizing meetings, synchronizing experimental protocols, arranging joint publications, preparing interim and final reports and financial administration. The group has full yearly meetings and sub-group meetings to discuss individual topic areas.

Experimental work is conducted in close collaboration with MLURI and includes studies (i) on factors affecting radiostrontium absorption in sheep, (ii) providing data on Sr dynamics in lactating goats for Nottingham Univ. to develop a radiostrontium model, (iii) determining the influence of environmental temperature and goitrogenic feedstuffs on radioiodine transfer to milk, and (iv) establishing parameters describing the transfer of radiocaesium from blood plasma to tissues. Further details of the planned experiments relevant to the strontium studies are given here whilst those for radiocaesium and radioiodine are presented in the MLURI summary.

1. Radiostrontium absorption and metabolism

The effects of dietary Ca and stable Sr levels on absorption, milk secretion and tissue deposition of radiostrontium in dairy goats will be examined. Similar multi-isotope

techniques to those developed in the previous CEC contract for determining the true absorption coefficient (A_t) for radiocaesium will be employed. The radiostrontium sources used will be ^{85}Sr and ^{89}Sr. Concurrent estimates of A_t of Ca will be made by using intravenous administration of ^{45}Ca.

Additionally, pulse doses of radiostrontium will be applied both intravenously and orally to lactating goats to obtain a time series of radiostrontium measurements for the major compartments of the animal which will then be used by Nottingham Univ. to produce a ruminant radiostrontium model.

SUMMARY OF STUDIES AT NPL, IOANNINA

1. Plasma-tissue transfer of Cs in ruminants

According to NPL's, and Nottingham Univ's models, which assume that the transfer of a contaminant to any compartment of an animal takes place through the animal's blood, transfer coefficients (f(k)) to any compartment (k) of the animal are related to the transfer coefficient to its blood (f(blood)) through a constant (a(k)) ie: f(k) = a(k) f(blood). Therefore if f(blood) is known, f(k) for any compartment can be calculated. Our experiments have verified the relationship for sheep. As defined the parameters are independent of animal size and they should be similar for physiologically similar animals. We intend to test the validity of this for goats and cows, complementing studies by ITE/MLURI. The experimental animals (n=4-5) will be fed contaminated hay until equilibrium is attained. Changes with time in radiocaesium concentrations in blood will be monitored. Upon equilibrium, the radiocaesium concentration will be measured in each compartment.

2. Countermeasures for Sr and Cs

Bentonite, which reduces radiocaesium absorption in ruminants, consists mainly of montomorillonite, a clay mineral with a CEC of about 80 mEq/100 g and an effective surface area (ESA) of around 25m^2/g. Recently the NPL (& Material Science Laboratory) have been trying to modify this, and other smectites, to develop a microporous structure with an ESA of 100-500 m^2/g. The binding efficiency of montmorillonite and its modified forms will be assessed as combined radiostrontium and radiocaesium binders for administering to animals. *In-vitro* experimentation will be followed by *in-vivo* studies, in collaboration with UAN.

3. Sr transfer through soil ingestion

Within the first programme, a sandy soil from the Chernobyl 10 km zone was fed to sheep to determine the availability of radiocaesium. The transfer of ^{90}Sr from this soil to sheep milk will be determined through further analysis. Bio-availability results will be compared with those measured by SCK/CEN with contaminated clover.

SUMMARY OF STUDIES AT NOTTINGHAM UNIV.

1. Strontium model

In the previous project a detailed model of radiocaesium behaviour in ruminants was developed - this has proved an invaluable aid in planning and evaluating experimental work. We now intend to develop a similar model for radiostrontium. This will involve collaboration with experimental groups at ITE and MLURI applying pulse doses of radiostrontium both intravenously and orally to obtain a time series of radiostrontium measurements for the major compartments of lactating goats. These observations can then be fitted to the model by an optimisation process, enabling rate constants to be evaluated for the various transfers within the animal. As for radiocaesium this will provide a framework for developing a more physiologically based model, which for Sr will have to carefully consider the animals calcium status.

2. Radiocaesium metabolism in the gut

A number of experiments conducted by MLURI and TEAGASC within the previous contract have looked in detail at the distribution of radiocaesium within the gastro-intestinal tract of both sheep and cattle. This is an important area for improved understanding, as many countermeasures operate within the gut. We will therefore extend our current models to include a more detailed description of the transport and absorption of radiocaesium within the gut using the results from MLURI/TEAGASC's experiments. This would be an early objective within the project.

3. Iodine model

The development of a mechanistic model of radioiodine behaviour in animals will be considered. This would pay particular attention to the dietary intake of stable iodine and will link closely with experimental work planned by ITE/MLURI and GSF.

SUMMARY OF STUDIES AT MLURI, ABERDEEN

The MLURI, in collaboration with ITE, will conduct research on physiological factors affecting uptake of radiocaesium in sheep, factors affecting the true absorption of radiostrontium in lactating goats, Sr dynamics in lactating goats, and environmental factors influencing the metabolism of radioiodine by lactating goats. Details of the radiocaesium and radioiodine studies are given below whilst those for radiostrontium are presented in the ITE summary.

1. Radiocaesium metabolism

Parameters describing blood plasma-tissue transfer will be evaluated as a logical extension of the use of the true absorption coefficient in prediction models. Measurements will be made in ewes of differing physiological status in an extension of a study funded by other sources.

2. Effect of temperature and goitrogens on radioiodine transfer to milk

Iodine metabolism in ruminants has been shown to be influenced by environmental temperature and by the presence of goitrogenic substances in the diet. It is therefore likely that in lactating ruminants, transfers of radioiodine to the thyroid and to milk will be influenced by such factors. Using ^{125}I and ^{131}I, we will examine the effect of ambient temperature and of the diet on radioiodine transfers to the thyroid and to milk in lactating goats. The animals will be housed in environmentally-controlled rooms and some of the experimental treatments will be administered goitrogenic compounds to simulate the effect of feed-types such as brassicas and legumes in the diet. Radioiodine uptake by the thyroid will be measured by live-monitoring.

SUMMARY OF STUDIES AT GSF

1 Simulation of grazing intensity-lysimeter studies

The root-grass uptake of two soils with differing nutritional status (low and high exchangeable K) will be investigated using soil with a high Chernobyl radiocaesium content. In addition, soils will be artificially contaminated with a range of other gamma-emitting radionuclides of radioecological significance (possibly including ^{134}Cs, ^{85}Sr, ^{110m}Ag, ^{57}Co, ^{139}Ce, ^{65}Zn) - *Lolium perenne* will be seeded as a representative species of pasture grass in Germany (and other EEC countries) and will be maintained for sampling between April - October 1993 at three different sward heights (3, 6 and 9 cm) by weekly clipping. The tiller densities, biomass changes and the relationship between stem to laminae will be measured over the growing season. Bio-availability of the different radiocaesium isotopes will be determined using the CsCl extraction method developed by ITE/MLURI.

2 Field experiments to determine radiocaesium transfer from vegetation to milk under different grazing regimes

These studies will be carried out at two farms. Farm A carries out a continuous grazing regime, Farm B carries out rotational grazing, which is more commonly used in Bavaria. Bulk milk and vegetation samples (3 replicates) will be collected weekly on Farm A and daily on Farm B. Soil characteristics, biomass and species composition changes will be determined at both farms. Additionally at 3 separate time periods (Spring, Summer and Autumn) the activity concentrations in milk from individual cows (up to 10 replicates) will be followed in order to compare individual with bulk milk samples. If possible herbage intake and composition will be determined using the alkane method: Alkane analysis will be carried out at MLURI.

3 True absorption of I in lactating cows

Possible experimental designs to look at the effect of stable iodine on the true absorption of radioiodine will be postponed until later in the contract period in order to benefit from the experience and initial results obtained by MLURI/ITE using sheep and goats.

SUMMARY OF STUDIES AT SCK/CEN

1 Bio-availability of radiostrontium from different feedstuffs to ruminants

The bio-availability of radiostrontium bio-accumulated in clover, fed dry and as silage, will be compared with that of ionic radiostrontium, using the true absorption method developed by MLURI/ITE. The highly contaminated plant material will be given together with a diet consisting of uncontaminated clover prepared in the same way (ie dried or silage respectively). Stable Ca contents of the various diets will be measured. Strontium, like calcium, is strongly bound and complexed with organic matter in the plant. Therefore, unlike radiocaesium, where absorption was similar when bio-incorporated into different plant species, strontium is expected to have different availability depending on its chemical speciation.

2 Transfer of radioiodine to milk throughout a complete lactation period in dairy cows

Because dairy cows represent the main source of milk for human consumption, the data obtained for smaller ruminants (sheep and goats) should be validated for dairy cows. In parallel with the studies which will be carried out by MLURI/ITE on the factors influencing the metabolism and transfer of radioiodine in goats, SCK/CEN will investigate the changes with time in the transfer of radioiodine to milk throughout a complete lactation period in dairy cows. Contamination will commence about ten days prior to calving. Detailed measurements will be made at intervals to compare radioiodine levels in plasma, urine and faeces to enable some comparison with the behaviour of radioiodine measured during the more detailed studies on goats. Such comparisons are important as sheep/goats have different plasma/milk ratios to cattle.

Contribution of the UAN

Norway is an EFTA country. The contribution towards the Agricultural University of Norway will be published in an addendum after signature of the Association Agreement.

Contribution of the PSI

Switzerland is an EFTA country. The contribution towards the Paul Scherrer Institute will be published in an addendum after signature of the Association Agreement.

A25 The role of retention and release of radionuclides in natural ecosystems and in marginal agricultural areas.

Contract FI3P-CT920016 Deposition of radionuclides on tree canopies and their subsequent fate in forest ecosystems - Further studies.

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Description of research work

i) Background and General Description of Study

During the previous contractual period (1990 - '92) the participating groups addressed the capture of radioactive aerosols by forest canopies and followed their subsequent physical fate. Due to a lack of information on these topics following the Chernobyl accident a knowledge of these processes is of major importance both for the prediction of contamination immediately after the accident and for the design and implementation of suitable short- and long-term countermeasures in the forest environment. Data currently available from Russia, Ukraine and Belarus confirm this statement.

During 1990 - '92 successful links were forged between the three currently participating groups and a fourth, led by Dr. Belot at CEA, Fontenay-aux-Roses, France. Research focused on the ability of individual trees and 'model' forest canopies to intercept aerosols depositing from the atmosphere under dry conditions and to subsequently retain these deposits over time. In addition, the Louvain-la-Neuve and Barcelona groups extended their interests to an examination of the physical migration of caesium in multi-layered forest soils typical of both northern and southern Europe.

During the current contractual period several lines of investigation are being pursued as a result of both experimental findings and methodological development during the first two years of the study. Dry deposition of radioactive aerosols is of relevance to both the near- and far-field situations. The complexity of dry deposition results in 1990 - '92 was greater than initially expected and has led to a detailed elucidation of 'model' canopy morphology and related micro-meteorological conditions during wind tunnel studies in 1993. This will lead to a better understanding of small scale turbulent diffusion and impaction processes contributing to aerosol deposition in tree canopies. Professor Ronneau' s group pioneered a high temperature generation technique for uranium aerosols during 1990 -'92;this is currently being adapted for use with delayed neutron counting by Miss Minski' s group. These two techniques in tandem will be used for high sensitivity studies of aerosol interception by sub-components of trees.

The problem of radiocontamination of forest soils in the near-field situation is also being addressed. The thermo-generation technique developed at Louvain-la-Neuve allows synthesis of vitrified hot particles which are being used by Dr Rauret' s and Professor Ronneau's groups in studies of fuel fragment behaviour in soils typical of Mediterranean and northern European forests, respectively.

ii) Imperial College (UK) Activities, 1992 - 1994

Activities at the Imperial College' s Centre for Analytical Research in the Environment (CARE) are based around the wind tunnel and reactor facilities at Silwood Park, Ascot. New techniques for both the thermal generation and thermal conditioning of monodispersed, uranium-based aerosols are currently being developed. A delayed neutron counting (DNC) facility already exists at CARE which allows sub-ppb determinations of uranium; this is to be used as a means of detecting aerosols deposited on tree tissues at low concentration with great sensitivity. The ultimate aim

of this work is to give high resolution data on the three dimensional distribution of aerosol deposits within small scale tree canopies.

Deposition experiments are carried using 'model' canopies comprising regularly spaced tree saplings. Hot wire and hot film anemometry are used to characterise canopy friction velocities and mean wind speed and turbulence distributions both within and above test canopies. These can then be related to deposition rates to different canopy sub-components. Studies during 1992 - '94 are centred on understanding deposition to tree canopies under conditions of steady wind flow and also gusty conditions. The latter can be recreated within the wind tunnel using a pneumatically operated flap. Data generated by these studies will be used to help validate the model developed by Belot's CEA group during the previous contractual period. Following deposition, resuspension rates of aerosols will be followed within discrete horizontal strata of the tree canopies.

Finally, studies at CARE will address the role of the forest edge in possibly enhancing deposition rates at the boundaries of open fields. This will be achieved using reduced scale tree canopies as well as artificial canopy elements.

iii) Catholic University of Louvain Activities, 1992 - 1994

The current activities of Professor Ronneau's group are directed at two distinct problems: first, the physical interception of thermo-generated aerosols by spruce trees and secondly, the physical mobility within multi-layered forest soils of radionuclides bound into UO2 fuel particles.

With respect to canopy interception processes, emphasis during the 1992 - '94 contractual period is being placed on small scale processes of aerosol capture related to the surface structure and roughness of spruce trees. Experiments are being carried out in a small wind tunnel at Louvain-la-Neuve.

Experiments on the mobility of synthetic fuel particles are addressing two groups of processes: first, the mechanical mobility of particles and secondly, the chemical leaching of radionuclides from both synthetic (laboratory-derived) and Chernobyl-derived 'hot particles'. The mechanical redistribution of fuel particles following their deposition to soil surfaces is most likely to be controlled by the movement of water following precipitation events and this factor is being taken into account as part of studies at Louvain-la-Neuve. However, other physico-chemical factors, such as pH, redox conditions, temperature, the presence of complexing agents and the activity of soil microorganisms, are likely to exert an important influence on the chemical leaching of radionuclides from fuel particle matrices. All of these factors are being taken into account in studies which are comparing the behaviour of laboratory synthesised particles with that of fuel particles collected from the Chernobyl near-field region.

iv) University of Barcelona Activities, 1992 - 1994

During the 1990 - '92 contractual period a novel field method for the assessment of radionuclide migration in forest soils was developed which involves the placement of ion exchange resin in discrete soil layers. This is currently being used at field study

sites in the Prades mountains, Catalonia, to elucidate the movement of 134Cs and 85Sr derived from leaf material contaminated using thermo-generated aerosols typical of the 'far-field'. Solvent extraction techniques are being used to determine the specific locations of these radionuclides with respect to the leaf surface in order to gain an understanding of the mechanisms involved in their release from contaminated forest floor litter. Extensive instrumentation of the study sites is providing supplementary data which enables model descriptions of the radionuclide release processes using the 'SOIL' and 'DECO' models.

'Near-field' radiocontamination is also being considered in studies at Barcelona. Thermally synthesised 'hot particles' are currently being obtained from the Louvain-la-Neuve group in Belgium for use in investigations of their physical migration in forest soils in northern Spain. Similar methods to those developed during the previous contractual period are being employed in this study and will give much information on the behaviour of radionuclides introduced into Mediterranean forest ecosystems as a result of a very different scenario to that which followed the Chernobyl accident in 1986.

A25 The role of retention and release of radionuclides in natural ecosystems and in marginal agricultural areas.

Contract FI3P-CT920050 Cycling of cesium 137 and strontium 90 in natural ecosystems.

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Description of research work

Radionuclides behave differently in natural ecosystems than in agricultural areas, which is expressed by the significantly higher activities in forest's plants and animals, compared to agricultural ones at same deposition rates. As forest ecosystems are important to man, since they provide wood, paper, wild berries, mushrooms, game and recreation, it is of interest to analyze the behaviour of radionuclides and to estimate the potential radiation exposure from their extensive use by the population.

The project deals with the cycling of radiocaesium and -strontium within and between abiotic and biotic compartments of forests and with the radiation exposure to man by the use of these systems. For the investigations different coniferous and deciduous forests were chosen from different climate zones in Sweden, Belgium, Italy and Germany. The experimental strategy emphasizes on parameters and mechanisms to understand and to quantify transfer processes of ^{137}Cs and ^{90}Sr. The investigations of the participating groups cover the following criteria:

- influence of soil characteristics on the retention and vertical distribution of caesium and strontium in the different layers of the soil,
- plant availability and biological fixation of these radionuclides, especially in the organic horizons,
- influence of rooting depth and nutrient levels in soil on the transfer of ^{137}Cs and ^{90}Sr into plants,
- radionuclide concentration patterns in vascular plants within a natural forest ecosystem,
- dependency of the accumulation rates on the living habit of fungi species,
- transfer of ^{137}Cs and ^{90}Sr from plants into mammals,
- distribution of ^{137}Cs and ^{90}Sr between the different forest compartments,
- influence of forest litter production and decomposition on radiocaesium availability to the leaching along multilayered soil profiles,
- development of a mathematical model for ^{137}Cs and ^{40}K transfer, uptake and removal in a forest ecosystem.

Based on measurement results, radioecological modelling will be carried out on three levels: different equations describing the transfer of ^{137}Cs and ^{90}Sr between two compartments will be developed and their reliability statistically analyzed; models describing long term cycling will be suggested, considering the distribution of the including radionuclides and the loss from the systems. Finally the potential radiation exposure to man originating from the use of the natural forest environment will be assessed.

Contribution of Bundesamt für Strahlenschutz

During the last two years the cycling of ^{137}Cs and ^{90}Sr has been investigated in three different coniferous forests. The emphasis of these investigations was put on the behaviour of radionuclides in soil and their transfer into mushrooms and green plants, which primarily take up their nutrients from organic horizons. It could be demonstrated that the uptake of ^{137}Cs by saprophytic and parasitic mushrooms is not exceptional, since

green plants show similar ^{137}Cs activities as well. Only symbiontic fungi accumulate radiocaesium to a higher degree. However, the transfer is not completely understood and the present transfer equations we developed are not satisfying for two major reasons:

While on mineral soils, ^{137}Cs is strongly absorbed in silicates and therefore hardly plant available, our preliminary results indicate that ^{137}Cs is biologically fixed in the organic layers of forest soils. After ^{137}Cs is released by decomposition of organic material, it might be immediately taken up again, i.e. the plant availability of caesium in soil might be short and therefore the migration rate is low. This raises the question, how important are soil parameters for the transfer soil/plant. Therefore, the amount of radiocaesium that is biologically fixed in the organic horizons, will be experimentally estimated.

The same problem exists also for ^{90}Sr. Our measurements indicate that most of ^{90}Sr, deposited during the time of weapon test fallout, is found in the organic horizons. Considering the low K_D-values for radiostrontium, biological fixation of ^{90}Sr should prevent its migration. Therefore, the biologically fixed amount of ^{90}S will be analyzed as well.

The uncertainty of the transfer equation for ^{137}Cs is partly due to the differing rooting depths of the plant species. The question is the amount of nutrient uptake from each horizon. This problem will be investigated in different plant types and discussed with respect to the reliability of transfer equations.

A general transfer equation soil/mushroom is difficult, as the activities in different species vary by more than three orders of magnitude at the same site. Preliminary results indicate that each species accumulates an individual amount of ^{137}Cs. This is observed from the ratio of the ^{137}Cs activities between Xerocomos badius and other species. There seems to be an individual constant ratio factor for a number of species, independent of site and year. Constant ratios provide the opportunity to estimate the ^{137}Cs levels in mushrooms by comparing their activities. Our data base for statistical analyses is presently not large enough, especially because only a few mushroom species could be sampled in 1991.

To complete the investigations on the transfer soil/plant, and to calculate the distribution of ^{137}Cs in coniferous sites, the uptake of ^{137}Cs by trees will be included into the future investigations.

For one selected site, similar measurements of ^{90}Sr have been carried out. The preliminary results show that ^{90}Sr behaves similarly as Ca. Therefore, the ^{90}Sr activities in mushrooms are very low in accordance with the low Ca content in these species. Significantly higher ^{90}Sr activities are found in green plants, which generally show higher Ca levels. Within the next two years, it will be analyzed to which extent ^{90}Sr cycling can be described with the help of Ca cycling.

The investigations were carried out in coniferous forests. It is of interest to analyze whether the same transfer mechanisms and parameters regulate ^{137}Cs and ^{90}Sr cycling in different systems. Therefore, an additional deciduous forest will be included in this programme.

The results will be the basis for developing radioecological models, which describe the long term cycling of ^{137}Cs and ^{90}Sr in the forest soil-plant systems.

Contribution of Faculté des Sciences Agronomiques - U.R. de Radioécologie

During the earlier CEC programme, research was founded on those ecological factors explaining the actual or apparent variations in soil-plant and soil-fungus radionuclide transfers in similar forest types, leaving aside the variable level of radioactive deposition in the soil. These studies were repeated on various soil types and in different kinds of forests in Belgium and Luxembourg, as well as in Sweden, with our colleague, Judith Melin. The preliminary findings, which are in agreement with the research conducted by our colleague, P. L. Nimis, suggest that the depth of the root system in plants or the mycelium in fungi (and, of course, all the ecological factors which influence this depth) is the prime factor to consider in explaining variations in inter-individual and interspecific contamination levels. In our temperate climatic conditions, we have demonstrated the vital role played by the type of humus (in conjunction with "rooting") in radionuclide transfers. During the new programme, we shall continue to take soil and plant samples from our experimental plots in order to pinpoint the effect of the litter on radionuclide migration in particular, as well as on the composition of the vegetation stratum and microfungus and therefore on competition at root absorption level.

In order to gain greater understanding of transfer mechanisms, we initiated a preliminary experiment in a controlled chamber on undisturbed forest soil. This experiment studied the effect of the humus microflora on the retention and release of radiocaesium. During the next programme, these controlled chamber experiments will be developed and extended to a study on the role of ectomycorrhizae in radionuclide transfers to young forest plants.

Contribution of the University of Trieste

The Trieste group was mainly concerned with two topics during the first phase of the research financed by the CEC:

a) Study of radiocontamination patterns in vascular plants within a natural forest ecosystem.
b) Individuation and use of bioindicators of radioactive deposition (mainly mushrooms and bryophytes).

The results obtained during the first research period allow to focus our future attention to two further main topics:

1. Studies on the relations between contamination patterns of vascular plants and the behaviour of radionuclides in the forest soils. The first results showed that it is possible to simplify the main radioecologial features of a forest ecosystem, subdividing it into a few compartments, by grouping the species according to their ecological requirements. This renders more easy the construction of quantitative models on radiocaesium transfer in natural ecosystems. The future research will try to explain the patterns evidenced by the first results, focusing the attention to the relations between the soil rhizosphere interactions and the radiocontamination of vascular plants. This research will be carried out at the same pilot station used in the first phase of research (Paso Pura, Carnic Alps, Italy). Part of the research, and especially some special topics related to the soils, will be carried out by joint activity with the group coordinated by E. Wirth (Munich, Germany).

2. Studies on the role of bryophytes in the retention of radiocaesium in forest ecosystems. The previous results revealed that bryophyte mats are able to retain and immobilize important quantities of radiocaesium, slowing down their transfer to the soil. This point will be the object of a special study. Furthermore, since bryophytes have proved to be reliable indicators of radiocontamination, we plan the construction of a radiocontamination map of the Carnic Alps, based on bryophyte data as a case study for mapping radiodeposition in mountain areas.

Furthermore, the Trieste group will carry out a general elaboration of the data concerning the radiocontamination of mushrooms, relative to several stations throughout northeastern Italy, that have been collected every year, from 1986 to 1991. We expect to construct a model on the time-dependent development of radiocontamination of edible species that could be of direct relevance of radioprotection measures.

Contribution of ENEA-DISP (ITALY)

The aim of this research programme is to study the influence of forest litter production and decomposition on radiocaesium availability to the leaching along the multilayered soil profile.

The following topics will be investigated:

1) the seasonal production of leaf litter and the evaluation of radiocaesium input into the soil system:

   a) the forest-seasonal production of the litter will be assessed using traps and nets, placed on a representative area;
   b) the main components of the litter-fall (leaves, twigs, etc.) will be assessed;
   c) radiocaesium content will be determined on litter samples.

2) the role of litter decomposition on caesium availability to the transfer between different soil compartments:

   a) the rate of decomposition will be assessed using litter bags. The bags, once filled with a known weight of fresh litter, will be sealed and relocated at the position in the canopy (above ground) or in the horizon (below ground) where the material occurs naturally. The bags will be positioned according a randomized block design. To assess the decomposition rate, the bags will then be removed at regular time intervals, at which the change in weight and the decomposition rate will be estimated;
   b) the rate of loss of radiocaesium and potassium from litter (during its breakdown) will be assessed;
   c) pH, C and N contents will be determined.

In the frame of the project the experimental activities of ENEA-DISP will be strongly linked with the modelling activities of CETA (Italy).

Contribution of the International Center for Theoretical and Applied Ecology (CETA), ITALY

The main aim of CETA contribution within the Project will be the development of a mathematical model of the radionuclides (^{137}Cs, ^{40}K) transfer, uptake and removal in a mountain mixed forest ecosystem (beech and spruce trees). The choice of this ecosystem is due to its high diffusion in the mountain areas of Europe.

The activity will be defined according to the following steps:

1) Development and calibration of a mathematical model estimating the litter decomposition and degradation, in function of the main ecological factors, based on literature data.

2) The radionuclides field data collected by ENEA-DISP in the frame of this project will be used to develop and calibrate the radioecological part of the above mentioned model, to assess the radionuclides removal and cycling through the ecosystem.

Contributions of SSI, Dep. of NBC Defense (FOA) and SUAS

Sweden is an EFTA country. The contributions towards the National Institute of Radiation Protection, the Department of NBC Defense and the Swedish University of Agricultural Science will be published in an addendum after signature of the Association Agreement.

A25 The role of retention and release of radionuclides in natural ecosystems and in marginal agricultural areas.

Contract FI3P-CT920058 Radiation doses and pathways to man from semi-natural ecosystems.

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Description of research work

Background.

Consumption of foodstuffs produced from semi-natural ecosystems contaminated with man-made radioactivity can significantly contribute to radiation dose to the general population and to critical groups in particular. Because of the long ecological half-life of artificial radioactivity in the natural vegetation and in the animal species which characterise such ecosystems, this dose can be delivered over a period of several decades. Review of published data and experience gained since the Chernobyl accident shows that factors controlling radionuclide uptake are not yet properly understood. This restricts the degree to which radiation doses from semi-natural ecosystems can be evaluated and allows limited predictive capability for future nuclear accidents.

One of the most important pathways to man is via the consumption of animal products contaminated with radiocaesium. In particular, the consumption of moose in Sweden during 1991 resulted in the transfer of 2,000 MBq caesium-137 (500 times ALI value) to the population. Current work has shown that fungi are an important part of the diet of many farmed and wild animals grazing semi-natural ecosystems and are responsible for much of the seasonal and year-to-year variability in the radiocaesium body burden of sheep, reindeer and roe-deer. While the Scandinavian countries are particularly at risk in this regard, increasing afforestation of upland areas in Ireland and the United Kingdom, all of which are extensively grazed by sheep, makes similar studies in these ecosystems equally important. The controlling influences on the production rate of fungal fruitbodies should also be evaluated.

While much of the radiological data generated since the Chernobyl accident relates to radiocaesium, it must not be forgotten that Sr-90 is a radionuclide which, in severe accident conditions, may result in a significant radiation dose to man. The uptake of strontium from agricultural soils, particularly in terms of Sr/Ca ratios, has been extensively studied. However, much less data have been produced for high organic acidic soils with a low calcium content which are often typical of semi-natural ecosystems.

Some observations suggest that Sr may be more mobile than Cs in low pH soils, while others have reported that organic matter retains Sr in a way that makes it unavailable for root uptake. There is clearly a need to resolve the possible competing influences of these parameters. Given the diet selectivity of many herbivores, the Sr-90 uptake by individual plant species also needs to be evaluated. Potential pathways, which may not be the same as those for Cs-137, should therefore be identified.

The transfer of radionuclides from soil is often defined in terms of transfer factors or transfer coefficients. These terms were originally developed for studies of agricultural ecosystems and they may be less appropriate for semi-natural ecosystems due to the absence of regular mixing (ploughing) of the upper soil layers. The limits of applicability of these transfer parameters in connection with semi-natural ecosystems should be investigated. Specifically, account may need to be taken of parameters such as radionuclide extractability from soils, vertical distribution within the soil profile,

seasonal variability and year-to-year variations in plant uptake. It is known that radiocaesium in soil exists in a number of fractions of different availability. Once the soil/soil water equilibria are established for the mobile fractions, additions to the more immobile fractions are unimportant from the point of view of soil-plant transfer although they have large effects when transfer factors are calculated. Better methods need to be developed if reliable estimates of the radiological hazards from semi-natural ecosystems are to be made.

Because of the importance of animal products as a source of radiation exposure to man, radionuclide levels in vegetation are often of more direct relevance than those found in the underlying soils. In semi-natural ecosystems, Calluna vulgaris normally contains the highest concentration of radiocaesium and is therefore a potential useful bioindicator. Its value as a predictor of radiocaesium concentrations in other plant species and at other sites needs further investigation. The identification of a suitable bioindicator for Sr-90 would also be desirable.

Work Programme

In order to address these deficiencies in our present knowledge and to provide better estimates of radiation doses to man from semi-natural ecosystems, the following programme of research will be undertaken:

Studies will be performed in Sweden and Norway on the seasonal variation in caesium-137 content and production of fungi. Regular collection of all fruit bodies within existing research sites will be undertaken for species determination and caesium-137 measurement. This will provide a good estimate of the importance of fungi in caesium-137 transfer to man and the knowledge on seasonal and year-to-year variability will allow a better estimate of ecological half-life to be made. Soil temperature and soil moisture content will be continuously measured and the effects of these climatic factors on the production of fruitbodies will be evaluated. Data on caesium-137 concentrations in fungi collected at research sites in Denmark, Greece, Ireland and UK will be made available as a basis for assessing the similarities and differences between all ecosystems. It is anticipated that these studies will provide a basis for deciding on the most appropriate countermeasures which should be applied in order to reduce radiocaesium levels in animal tissues, thereby also limiting collective and critical group doses.

As a first step in evaluating the important pathways to man from Sr-90 deposited in semi-natural ecosystems, Sr/Ca ratios and the Sr-90 content of several vegetation species and fungi common to all ecosystems will be measured and compared with those in underlying soils. This will assist in the identification of suitable bioindicator species and will allow an evaluation of the extent to which results, obtained for one ecosystem, can be applied to another. In the second year, a disused Sr mine site in an upland area of Scotland will also be investigated and the levels of stable strontium in soils and in a number of plant species common to the site will be measured.

In investigating various transfer parameters, work on soils, vegetation and foodstuffs will be necessary. ITE will examine the bulk radiocaesium in the soil/soil water system and, using methods already developed, divide this into different fractions. Each fraction will be correlated with plant uptake to investigate which fraction, or combination of fractions, provides the best estimate of plant concentration.

In Ireland, existing work on Calluna vulgaris will continue through studies of site variability and relationships between radionuclide concentrations in different plant species. Replicate sampling of a number of upland vegetation species and of soil will be carried out on several occasions during the grazing season on 3-4 sites. Between farm and within farm predictions will then be carried out using plant-to-plant and soil-to-plant ratios and the two systems compared statistically. In support of these specific investigations, Risø will evaluate the time series of radiocaesium and of radiostrontium in food products taken from the Faroese ecosystem in order to improve the estimate of transfer factors, ecological half-lives and committed doses.

By means of laboratory experiments, Cs/K and Sr/Ca relationships will be investigated using a range of plant species typical of semi-natural ecosystems. The effects of nutrient status, physico-chemical characteristics of the soil and the root distribution of plant species on Cs and Sr uptake will be evaluated. These data will provide valuable information of the dynamics of uptake and are complementary to the studies of all other participating groups.

Table 1
Division of Tasks Between Participants

	RPII	ITE	AUT	SLU	RISØ	AUN
Fungi	✓	✓	✓	✓*	✓	✓
Sr-90	✓	✓	✓	✓	✓*	✓
Sr/Ca Ratios	✓	✓	✓*	✓	✓	✓
Transfer Factors	✓	✓*			✓	
Laboratory Studies			✓			
Stable Sr Site Study		✓				

✓ indicates participation
* indicates laboratory with primary responsibility to co-ordinate this area of work.

Programme Management

The proposed work programme has been designed to maximise the input of participants in areas where they have expertise over-and-above that available in the other laboratories. Work on soils will be concentrated at ITE, who will also provide an analytical service on behalf of other participants. Measurements of stable isotopes will be made at AUT, and all laboratory experiments relevant to other studies will also be undertaken in Greece. The long-standing expertise of RISØ in Sr-90 measurement will be available to the other participants and work on fungi will be concentrated in Scandinavia. This allows the research programme to proceed in a way which would not be possible without the high degree of co-ordination and co-operation which has now been agreed.

Within the overall co-ordination of the RPII, other participants have been given responsibility to manage specific areas of the programme. These are indicated on Table 1. As part of this management a series of intercomparison studies on determination on stable and artificial nuclides will be undertaken to ensure uniformity of measurement and comparability of results. A joint site sampling exercise is also planned. In finalising this

proposal, consideration has been given to avoiding unnecessary duplication of work presently in hand within the CEC Radiation Protection Research Programme or planned as part of the CHECIR studies in the Ukraine. However, a high degree of contact is desirable and will be maintained with other research groups, particularly those studying forest ecosystems and animal metabolism.

Of the six participant laboratories, five have already worked together as part of the 1990-1991 research programme. The addition of a Norwegian laboratory brings additional expertise on fungi through the participation of Dr. Olsen. The ability of this group to undertake high quality research in a collaborative manner has already been proven.

Contribution of the Radiological Protection Institute of Ireland

Data produced over the past two years have been used to develop a plant - plant ratio predictive system for ^{137}Cs in vegetation found in semi-natural ecosystems in Ireland. The concept of using soil-plant transfer factors and concentration ratios was developed for radionuclides studies in agricultural ecosystems. Such calculations are, for many reasons, inappropriate for semi-natural ecosystems. These reasons include the presence of a litter layer, absence of ploughing and differences in bulk density and in ^{137}Cs distribution in soil profiles in unmanaged soils. The importance of these factors is fully discussed elsewhere (Ref. 1). Earlier studies, using Calluna vulgaris as the bioindicator species, demonstrated that a plant - plant ratio system can give consistent results and is also of use for between site predictions.

In order to further test and refine such a predictive system, four peatland sites of different environmental characteristics will be chosen. Plant species to be sampled will include C. vulgaris, J. squarrosus, E. tetralix and M. caerulea. Replicate vegetation and soil sampling will allow plant - plant ratios to be compared with the traditional soil - plant transfer factor. Consistency, reliability and predictive accuracy of both within and between site predictions will be evaluated.

Ongoing work to obtain a more accurate estimate of the ecological and effective half-life of ^{137}Cs in semi-natural ecosystems will be continued. Data on several vegetation species and on mountain sheep will be made available to Riso for individual, collective and committed dose calculations and a predictive model will be developed. An existing model to predict ^{137}Cs levels in sheep from levels in faecal fragments will be further refined and the model extended to red deer grazing a montane peatland site.

The RPII will co-ordinate the work of the group and will participate in all field and laboratory intercomparisons. Pilots studies on the critical pathways for ^{90}Sr transfer to peatland vegetation and foodstuffs produced in semi-natural ecosystems will also be initiated as part of the programme.

Reference

1. Mc Gee, E.J., Colgan, P.A. and Synott, H.J. (1993). A new method for prediction of radiocaesium levels in vegetation : evidence from Irish uplands. Journal of Environmental Radioactivity 18: 53-70

Contribution of the Institute of Terrestrial Ecology

From previous studies we have found that measuring total radiocaesium in our semi-natural ecosystems and calculating transfers in the conventional manner is unsatisfactory. This means that predictions of the movement of radiocaesium through the plant/animal pathway

to man are unreliable. Better methods need to be developed if good estimates of the radiological hazard from semi-natural systems are to be made. We suspect that the radiocaesium in soils exists in a number of fractions of differing availability. Once the soil/soil water equilibria are established for the mobile fractions additions to the more immobile fractions are unimportant from the point of view of soil/plant transfers although they have large effects when transfer factors are calculated.

We will therefore examine the bulk radiocaesium in the systems already studied and, using methods already developed, divide this into a number of pools or fractions. We shall then attempt to correlate each of these fractions with plant uptake and investigate which fraction, or combination of fractions, provide the best estimator of plant concentration.

During the above work it will be convenient to collect sufficient samples to enable radiostrontium analyses to be carried out on all components of the ecosystems under consideration. Using an old mining area in Scotland the distribution of stable strontium will be investigated in species common to this area and the main experimental site.

Co-operation with other members of the group will continue particularly in the intercomparison of analytical techniques for radiostrontium, the provision of a soil analytical service for the group and in collecting fungi for radiochemical analysis.

Advantage of this approach

1) We can use an extensive data bank of physical, chemical and biological information collected in previous work.

2) We can extend the existing data into a much longer time sequence.

3) We rapidly get a model for radiostrontium as most of the physical and chemical measurements exist.

4) From a radiological viewpoint a much more reliable estimate of the risk to man from pathways in semi-natural systems will be attainable.

Contribution of the Risø National Laboratory

Risø will undertake the following work programme:

1. Four species of fungi (available in UK, IE, SE & DK) will be collected in autumn 1992 along with corresponding soil and will be analyzed for ^{90}Sr, Ca, ^{134}Cs, ^{137}Cs and ^{40}K.

2. The Irish soil and the UK heather and bracken samples, earlier used for radiocaesium intercomparison, will be analyzed for ^{90}Sr and Ca. Furthermore, a milk powder sample provided by RPII will be analyzed for ^{90}Sr and Ca.

3. The samples collected in the Faroes in 1991 of Polytrichum commune, Eriophorum angustifolium, Calluna vulgaris and Vaccinium myrtillus will be analyzed for ^{90}Sr & Ca along with their corresponding soil samples.

4. The time series of radiocaesium and ^{90}Sr Faraose lamb and milk will be continued in order to improve the estimate of transfer factors: Bq $kg^{-1}yr$ per Bq m^{-2}.

5. Risø volunteers to receive guest scientists for training in Sr-90 analysis.

6. Dose assessment based on data supplied by all group participants will be undertaken.

Contribution of the Aristotlean University of Thessaloniki

AUT will test the hypothesis that there is an interaction between plant species ranking and location in Cs (or possibly Sr) concentration in plant biomass (supported by the Riso group, while the alternative hypothesis is supported by RPII). To implement this we are going to carry out an experiment in which 10 plant species (grasses-legumes-other forbs) in mixtures will be grown in pots (volume of 50 L) filled with soils taken from different locations in north Greece. These soils will differ in the various physico-chemical characteristics (organic matter proportion, pH, nutrient concentrations, especially in K and Ca, etc.) in this experiment we also hope to test the following hypotheses:

a. Is there any correlation between Cs (or Sr) and K (or Ca) concentration in the above ground biomass in the plant species in the different soils used? and

b. Do the physico-chemical parameters of the soil and the root distribution of plant species affect the Cs (or Sr) uptake by plant species?

These tests will give information on whether plant and soil indexes can serve as predictors for Cs and Sr plant uptake. Also, an effort will be made to verify how well these indexes predict Cs and Sr concentration in natural ecosystems.

Medium scale field experiments will be conducted to investigate the fate of ^{137}Cs when applied to soil covered with mixtures of pasture plant species. In this experiment the ^{137}Cs uptake by various plant species will be related not only to total amount of ^{137}Cs applied in the soil but also to extractable ^{137}Cs in the soil which is expected to be reduced with time.

Since the kinetics of Cs and Sr in the soil seem to affect the Cs and Sr plant uptake, a batch experiment will be conducted to investigate the rate of absorption and release of the above elements in the soil.

Contribution of SUAS

Sweden is an EFTA country. The contribution towards the Swedish University of Agricultural Sciences will be published in an addendum after signature of the Association Agreement.

Contribution of the Agricultural University of Norway

Norway is an EFTA country. The contribution towards the Agricultural University of Norway will be published in an addendum after signature of the Association Agreement.

A26 Development of countermeasures to reduce the contamination in the environment and to impede its transfer to man.

Contract FI3P-CT920013a Studies of methods for the rehabilitation of soils and surfaces after a nuclear accident (RESSAC).

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Total Contribution by the Commission: 730 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The RESSAC programme (REhabilitation of Soils and Surfaces after an ACcident) is aimed at providing data to be used for the management of the emergency response to a nuclear accident, and to try effective agricultural countermeasure techniques rendering the decontaminated zone habitable.

Studies on lysimeters.

By the end of 1992, a special facility will be put in service for the simulation of fall-out radionuclide deposition on lysimeters with a special device (the POLYR furnace) able to simulate the core melting and the subsequent hot-particle emission. The lysimeters are aimed at simulating natural conditions of (i) scale factor, minimizing edge effects, (ii) soil profile moisture, which governs radionuclide bio-availability, (iii) climatic conditions, air-soil temperature, air moisture with an effect on soil biochemistry and plant physiology. In complement to French soil types, the main typical soils from the European Community will be represented (E, B, D, UK and F). Soil-to-plant transfer factors experiments will be carried out within an integrated experimental programme. Hence, in order to standardise experimental cultures on the different European soils, the first cycle of experiments will be focused on wheat which has been selected to be more representative European crop. According to the results of the first cycle of experiments the subsequent cycles will be focused to crops or specific experiments of national interest.

Studies on the physico-chemical properties of hot-particles

Global experiments with lysimeters are necessary to integrate parameters like scale factor, climatic conditions, soil biology (worms, insects, terrestrial arthropods) or biogeochemistry, but they are not favourable to analytical experiments on physico-chemical properties of hot-particles where replicates (not possible with 12 t lysimeters) are necessary to study the effect of isolated parameters (composition of the charge, temperature cycles, atmospheric composition of the experimental box, soil interaction). Therefore, in complement to global experiments, analytical studies on the physico-chemistry of hot-particle are essential to improve soil-to-plant transfer models and to define a common European assessment on the bio-availability of radionuclides.

Studies on Countermeasures.

Countermeasures involving radioactive tracers studying the effect of different agricultural practices on the plant transfer (fertilizers, deep-ploughing, defoliation, etc.) will be made in the lysimeters according to the experimental opportunity.

Some countermeasure techniques such as soil scraping together within the Decontaminating Vegetal Network (D.V.N.), use of polysaccharides, trials of chemical defoliants, etc., necessitate in-situ experiments.

Non-lethal defoliation.

Previous investigations on the non-lethal defoliation have shown that some non-zootoxic chemical defoliants can be used on forest trees to burn the leaves within 3 to 20 days, depending of the tree species. Preliminary experiments on the translocation rate of radionuclides from the leaves to internal ligneous tissues indicate that the internal contamination reach about 2% of the initial deposition on the leaves surfaces within 150 d. Therefore, if further observations can prove that the detriment caused by the defoliation is not prohibitive, non-lethal defoliation might also be used for perennial crops like vine of fruit trees which are of high economical values, especially for Mediterranean countries. A joint proposal together with Greece, Athens University of Agriculture, is aimed to study the possible use of non-lethal defoliation to minimize the foliar uptake of radionuclides by the vine. Other Mediterranean crops like olive, peach, etc., may also be studied. Simulation exercises of a nuclear accident carried out in France have shown that in most of the cases, the foliar transfer to the vine is poorly understood, with very few valuable transfer coefficients. A special effort seems to be needed on this topic.

A26 Development of countermeasures to reduce the contamination in the environment and to impede its transfer to man.

Contract FI3P-CT920049 Transfer of accidentally released radionuclides in agricultural systems.

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Description of research work

The experiences following the Chernobyl accident in 1986 have emphasized the need to improve the quantification of several processes and parameters in the transfer of radionuclides along the food chain. The use of reliable parameters and accurate descriptions of processes are essential for pre-accident assessment modelling, and for the implementation of protective measures designed to mitigate the radiological consequences of an eventual accident.

The aim of this project is to contribute to the reliability of radiological assessment parameters and to establish sound scientific bases to be used in the design of postaccidental countermeasures.

The objectives of the project deal with the following aspects: i) the characterization of an accidental aerosol release containing Sr, Cs, and Ag isotopes that simulate an accidental source term of a PWR reactor; ii) the study of the physico-chemical processes involved in their deposition on some important and common European soil-crops systems.

Some experimental designs are proposed in order to describe these processes and to determine the parameters associated with them. The same soil types as in the previous phase, will be used: a French one (sandy soil) and a Spanish one (sandy-loam soil).

As in the previous phase the lettuce (*Lactuca sativa*), a leafy vegetable, was studied, another one has been selected for this period, peas (*Pisum sativum*), fruit vegetable.

In the previous TARRAS contract, the generation of the aerosol was carried out by a modified atomic absorption model that made possible to produce an aerosol with comparable physico-chemical characteristics to the theoretical aerosols given by safety experts (DAS) in case of a nuclear accident of a PWR reactor. For this contract, it is proposed to generate the radioactive aerosol with the system used in the EURO-SOIL RESSAC programme, the POLYR model.

The use of this larger POLYR model will allow the contamination of 6 m^2 surface with homogeneous deposit. The differences in physico-chemical behaviour, due to differences in their characteristics (e.g. moisture content), of the emitted particles by both systems will be compared. For this reason, it is proposed to use mature lettuces in one of the shoots.

Three aerosol contaminations will be carried out in the Cadarache Laboratory with the same three radionuclides previously used, Cs-134, Ag-110m and Sr-85. In each shoot, the deposition will be measured on some filters and this will provide a test of the homogeneity of the deposition.

The fractions intercepted by plants, adhered to leaves, directly absorbed, as well as the dynamics of leaf washout, root uptake, soil migration and radionuclides speciation are studied in different growth stages of the plants.

The soil dynamics, the activity transfer to mature plant will be described in soil contaminated aerosol deposits. The variability of these parameters with the type and growth stages of the plants, type of soil and irrigation conditions is going to be demonstrated.

Three shoots are planned, the experimental design is summarized in the following table:

Soil 1: Sandy-loam soil
Soil 2: Sandy soil

SHOOT	CROP STAGE	SOIL
First	Sowed peas	Soil 1 and Soil2
Second	Young pea plants	Soil 1 (covered and without the cover)
Third	Mature pea plant Mature lettuce	Covered soil 1 (with and without irrigation)

Some different aspects are going to be considered:

(a) The Influence of the plant growth phase on the radionuclide retention.
Three growth stages of the plant will be contaminated in order to follow the soil-plant dynamics. Special attention will be paid to the influence of the root length and soil retention in the radionuclide uptake.

(b) The Influence of the soil characteristics in soil migration.
Soil migration and speciation in the two kinds of soils will be compared taking into account the differences in physico-chemical soil characteristics e.g. the interchange capacity of the soil, the organic matter present in them, the clay content, etc.

(c) Root uptake of radionuclides.
The root uptake by the selected crop, peas, with each plant growth stage will be followed, comparing two experiences: the first one with covered soil and the second one without the cover. Speciation studies will be done in order to evaluate the radionuclides availability.

(d) Radionuclide translocation, plant interception factors and washout.
After radionuclides deposition on leaves, the plant interception factors will be analyzed, besides the washout effect of irrigation. Analysis of radionuclides speciation in leaves will be carried out to study the different penetration degree of the isotopes.

ROLE AND CONTRIBUTION OF IMA/CIEMAT.

IMA/CIEMAT will carry out the coordination of the whole project, organize periodical meetings and workshops where the progress of the project will be reviewed and the results discussed.

Furthermore, IMA/CIEMAT will collaborate with the other two institutions carrying out the analysis of the obtained experimental measures, deriving parameters that quantify the behaviour of the radionuclides in the soil-crops systems and in a way that can be applied to dynamic models.

It is expected that the following parameters will be obtained:

- Soil to plant transfer factors, considering both types of soils and three different growth stages of the peas. The temporal variations of these parameters as a function of the plant growth will be considered. These results will be compared with the results obtained in the previous phase of this project in which leaf vegetables, such as lettuce were studied.
- Translocation, interception factor and washout of the activity from the plants are going to be quantified.
- Chemical speciation in the soil and in leaves, in order to know the availability for plant uptake of the radionuclides in the soil and the degree of association of the aerosol with vegetal tissues.
- Radionuclides soil migration: The behaviour of radionuclides in the soil will be studied taking into account the different physico-chemical characteristics of the selected soils and the different watering systems.

All these parameters will be compared with the ones that are currently used in dynamic models and with the parameters obtained in the previous phase of this project, taking into account the differences in the source generator.

ROLE AND CONTRIBUTION OF CEA/IPSN/LETVA-CADARACHE.

As in the previous phase of TARRAS, the role of the LETVA is to produce the radioactive aerosol, evaluate its composition, prepare the crops, transfer the aerosols to the studied plants and care of the crops until the finalisation of the experimental stage.

For the present project, the aerosol will be generated with the same system as the one that is used in the EURO-SOIL RESSAC program, the POLYR oven. In the preceding phase of the TARRAS project, the experimental aerosol was produced using a modified atomic absorption oven able to produce aerosols with chemical and physical characteristics that have proven to be similar to the ones given by DAS as theoretical result of a serious accident on a PWR reactor.

The reasons of this change is, on one hand, to harmonize the work carried out in RESSAC and TARRAS, and the availability to compare the obtained results with the ones in the RESSAC program; on the other hand, a larger surface can be contaminated in a single shoot.

It will also be possible to compare the behaviour of the previous system as an experiment has been designed with mature lettuces.

In each experiment the deposition will be measured using filters that will make possible to control the distribution of the deposited activity.

ROLE AND CONTRIBUTION OF THE UNIVERSITY OF BARCELONA.

The Barcelona University will carry out the following aspects:

- The sampling of soils and the crops at different stages of growth at Cadarache facilities.
- The behaviour of the new source term in the foliar radionuclide uptake. The obtained differences will permit to know about the differences in the kinetics of the penetration through the leaves between the new aerosol and the one used before.
- The radionuclides migration in the two types of soil used. The migration of the radionuclides in both types of soil after a short irrigation period (few months) and after a longer one (one year) will permit to get more information about the migration of the radionuclides deposited from the termogenerated aerosols.
- The radionuclides speciation using sequential extraction schemes. In order to obtain information on the binding mechanisms and kinetics of the studied forms of the radionuclides with soil components, sequential extraction schemes will be applied. In a first step, the optimum methodology will be established using statistical optimization procedures. The variables studied for each fraction will be the ratio extractant solution/mass of soil, and the extraction time. A procedure will be applied to soil samples collected immediately after the deposition the synthetic aerosol and another to the ones collected after a period of irrigation.
- The foliar radionuclide uptake by peas, plant and fruit, by sequential extraction: to study the foliar and the fruit uptake by peas, a sequential extraction scheme which leads to five fraction will be applied. With this procedure it should be possible to establish the degree of penetration into the leaf by the different radionuclides.
- The radionuclide absorption by radicular path. To study the radicular absorption by peas deposition experiments in pea seeds sowed in a bare soil will be carried out.
- The influence of growth stage on the retention of radionuclides by the different parts of the plant. Deposition experiments will be carried out with plants at different growth stages.
- To study retranslocation in peas deposition experiments with and without covering the soil by a waterproof layer will be performed.

All the work will be carried out by quadruplicate.

A26 Development of countermeasures to reduce the contamination in the environment and to impede its transfer to man.

Contract FI3P-CT930071 Influence of the food-processing techniques on the level of radionuclides in foodstuffs.

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Total Contribution by the Commission: 85 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

The objectives are to determine the extent of radionuclide transfer through the food chain to man as modified by food processing methods applied to vegetables and cereals. The results will improve the reliability of radiological assessments and will identify processing methods and possible modifications that result in maximum removal of radionuclides from food products. Thus the data obtained will influence countermeasures, including application of derived intervention levels, following an accidental release of radioactivity.

An initial collaborative research program between HARWELL LABORATORY, the UNIVERSITY OF READING and IPSN/DPEI/SERE/CADARACHE has been made since 1990, during two years within the CEC "TARRAS" project. It has demonstrated very variable food processing retention factors (derived from radioactivity in processed food relative to the raw material), according to both processing method and type of crops. It has also demonstrated that the reduction of radioisotopes due to processing leads to a simultaneous increase in the by-products and wastes that may be recycled in the food chain by animal feed. More detailed examination of basic processes is now required to explore the ways to reduce radionuclide concentrations in foodstuffs: the main topics to be examined are the influence of different processing technologies and of additives on the radionuclide contents of vegetables. It is also intended to assess the fate of some radionuclides that enter factory by-products for animal feed.

The proposed programme of work comprises the following investigations:

A The influence of processing methods and additives on radionuclide concentrations in foods:

The objective is to carry out more detailed examination of some treatments of vegetables and of the most common additives used during canning of vegetables, to identify procedures that will enhance losses of activity from the final products intended for human consumption.

B The influence of food processing on removal of radionuclides deposited to foliage of mature leafy vegetables.

Certain radionuclides which possess relatively low soil-to plant transfer factors are nevertheless important contaminants of foliage by direct deposition from the atmosphere and/or resuspension of soil following a nuclear accident. Therefore, it is proposed by the British participants to quantify the effect of food processing on the removal of these radionuclides when contaminate leafy vegetables that are close to maturity.

C The fate of radionuclides in by-products and wastes from food processing:

Research recently carried out within the CEC "TARRAS" programme has shown that substantial quantities of radioisotopes that are removed by processing will be found in by-products and/or factory wastes. Some of these products are recycled mainly by

their uses for animal feed. It is proposed that each participant investigates the importance of this transfer pathway to animals and to man by liaison with the food processing industry.

The proposed study involves three organisations with experience in research programs. The teams are multi-disciplinary, therefore the resulting scientific-technical information exchanges will enhance the links among them.

HARWELL AEA-Technology and IPSN/CADARACHE have expertise in radioecological and nuclear environment research (measurements, assessment, modelling...). The project takes advantage of the contributions of the READING UNIVERSITY expertise in food processing studies. It is clear that the different tasks are complementary and that they will expand the capabilities of the participating groups in the field of Radiation Protection. The research program will contribute to assurance of the Community population that studies on consequences of the nuclear power program and the ability to apply effective countermeasures in the event of an accidental release continue to be made in detail. The proposed programme will provide additional information of practical value concerning food chain transfer of radionuclides to man, for the benefit of Community Members. Moreover, the project offers a broad and instructive field of investigation including some of the major facets of radiation protection research for efficient training of new scientists who will constitute the European research community.

CONTRIBUTION OF IPSN/DPEI/SERE/CADARACHE

IPSN/DPEI/SERE/CADARACHE will coordinate the whole project and contribute to topics A and C. Discussion of results will be made through regular contacts or meetings with the participants in order to carry out the scientific and technical coordination of the different parts included in the project, organizing periodical workshops where the progress of the project will be reviewed, the experimental procedures standardized and the results discussed.

IPSN/DPEI/SERE/CADARACHE will review specific papers concerning additives. Different parameters may be systematically studied for one or two canned vegetables. The effect of the more commons additives (such as sodium chloride, citric acid..) will be tested, as well as the influence of different kind of processing before canning (washing with pure water or with additives, blanching in vapour or in boiling water..). This study may be carried out either by measuring stable elements in industrial products, or if necessary, by tracer experiments with contaminated crops grown and treated in the laboratory.

A close collaboration with Harwell Laboratory and Reading University will determine the choice of the material and processing methods which are studied. Frequent contacts between the participants will ensure the standardization of operating methods and analytical procedures. Concerning recycled by-products, investigations will be carried out on the radioactivity redistribution between foodstuffs that are directly commercialized and by-products which may be recycled in other food chains and wastes, notably in the case of cereals processing.

CONTRIBUTION OF AEA/HARWELL

AEA/HARWELL will contribute to topics A, and B and study the following points:

Concerning topic A, the effect of different treatments (blanching and/or canning) on the content of K, Sr, Cs of processed vegetables will be examined. Crops to be studied are peas, potatoes and carrots. AEA-TECHNOLOGY / HARWELL LABORATORY will provide suitable vegetable crops for processing studies by Reading University. The produce will be obtained from West Cumbria, near to Sellafield Works. Stable and radioactive isotopes measurements will be made at Harwell before and after processing.

Concerning topic B, soluble radiotracers (representing the worst possible situation) will be applied to cauliflower, cabbage, spinach and oilseed rape and losses of radioactivity will be recorded after processing. A close collaboration with the other participants will determine the choice of the material and processing methods which are studied.

CONTRIBUTION OF DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY / READING UNIVERSITY

DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY / READING UNIVERSITY will contribute to topics A and C.

The appropriate processing (blanching, canning) of the raw material will be carried out in the pilot plant. The samples processed by DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY / READING UNIVERSITY will then be analyzed for radioactive and stable elements by Harwell Laboratory.

In addition, the University will undertake a study of the processing wastes produced by the UK food processing industry with particular reference to their use in animal feed. This will involve liaison with food processing and animal feed companies. By products intended for animal feed will be collected from factories and supplied to Harwell for analysis of stable Cs and Sr. Exchange of information concerning this point will be made through informal working meetings with other participants.

B) CONSEQUENCES OF RADIATION EXPOSURE TO MAN; THEIR ASSESSMENT, PREVENTION AND TREATMENT

B1 Stochastics effects of radiation

B1 Stochastics effects of radiation

Contract FI3P-CT920030 Co-operative research on late somatic effects of ionizing radiation in the mammalian organism.

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Total Contribution by the Commission: 300 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Objectives

The object of the European Late Effects Project Group (EULEP) is to plan, promote, execute and analyse, within the field of science and education, all co-operative research relevant to the understanding of the biological effects of exposure of living organisms to ionizing radiations and other agents, in particular to those that may result in late effects. The co-ordination of research within this field is brought about by standardization and development of methodology in the member institutions of EULEP, the promotion of co-operative research by means of problem-oriented task groups, the establishment of a European radiobiology archive, and the organisation of training activities, workshops and symposia.

Means of Executing the Programme of Work

The means by which EULEP fosters co-operation between laboratories engaged in the study of late effects is, and will continue to be, innovative and flexible. A review will be undertaken of how the structure of EULEP can be adapted to meet future requirements. Since the main function of EULEP is the co-ordination of research projects at the European level, it is well placed to assess the importance of new co-operative work and to provide the necessary stimulus for its initiation.

EULEP comprises a unique body of expertise within the European Community. As such, it is well placed to act as a permanent reference source for the public, and is readily available to provide scientific advice to the appropriate authorities at national and Community level.

EULEP has direct links to research programmes outside its own member institutions at two levels. Firstly, it has individual corresponding members who belong to different universities or other institutes whose research interests include, or are related to, those of EULEP itself. Secondly, active collaboration has been recently developed with three other bodies with related scientific interests relevant to radiation protection: the European Radiation Dosimetry Group (EURADOS), the United States Department of Energy (DOE) and the European Bone Marrow Transplantation Group (EBMT).

Scientific Programme

The programme covers standardisation activities, research and training. For research activities a medium-term programme has been developed which outlines the problem areas to which greatest importance should be attached. In this regard, the present two-year period, in which some of these problems will be tackled and others will begin to be studied, is seen as the start of a new phase in radiation protection research. In general, it is recognised that on the one hand, problems in radiation protection remain, and new practical problems will arise, which can be addressed with existing methodologies. On the other hand, there are more fundamental problems calling for

the latest approaches in modern biology. In all its research activities, EULEP will seek to bring about new inter-laboratory collaborations and to strengthen on-going collaborative effort which is not already supported by multi-national contracts; this could include collaboration with Central and East European countries.

The medium-term research programme referred to above may be summarized as follows:

- *Sources of Radiation.*
  Studies of great practical importance will be required concerning indoor radon, other alpha-emitters, neutrons, beta-emitters in relation to skin exposure, and high *z*-particles. These studies will include further work on the dosimetry of radionuclides, especially inhaled alpha-emitters and aspects of fetal dosimetry.

- *Low Dose Studies.*
  Here the most important aspects are low dose rate, basic mechanisms leading to carcinogenesis including an evaluation of the threshold problem, differences in individual radiosensitivity, radiosensitivity as a function of age including studies on the embryo and fetus, the development of new *in vitro* and *in vivo* model systems, the development of biological markers for the diagnosis of radiation-related disease, and the irradiation of reproductive cells.

- *Radiation Accidents and the Treatment of Radiation Damage.*
  Particular importance is attached to the discovery of ways to intervene in the course of development of late effects following high-dose irradiation; priority will also be given to studies on the basic mechanisms of, and on early markers for, the development of non-neoplastic damage.

- *Interspecies Comparisons for Extrapolation to Man.*
  These have been, and will continue to be, a distinctive contribution from inter-laboratory collaborative studies.

For the present, co-operative research among EULEP laboratories will be carried out by means of the problem-orientated task groups. Their range of activities will however be reviewed. There are currently 14 groups, evenly divided between (a) the biological effects of radiation and (b) the dosimetry and effects of incorporated radionuclides.

The co-ordinated research programme will reflect the research carried out in the different Voting Member laboratories and in the laboratories of corresponding members. The contribution of EULEP aims to enhance what could be achieved by separate institutes working alone, and thereby to increase the scientific value of the overall programme.

One of the most important ways in which EULEP will bring about this enhancement of total research output is by standardisation between laboratories. This will entail in-depth standardisation activities by committees working in the areas of external radiation dosimetry, internal radiation dosimetry, pathology, and cell and molecular biology. A need can be foreseen in future activities to standardise *in vitro* as well as *in vivo* models used in late effects studies.

Training activities will receive special emphasis in the coming period, recognising the need to replace a whole generation of radiobiologists which will shortly have passed into retirement, and also the need to attract young scientists into the field, including those with a background in cell and molecular biology. A range of advanced training activities is envisaged, including scientific exchange visits to acquire technical expertise, formal specialised training courses in new methodologies, lectures for research scientists on the basis and requirements of radiation protection, and the provision of course material for teaching radiobiology. These activities will be developed in association with the Commission's European Radiation Protection Education and Training programme (ERPET).

Work will continue on the EULEP *Pathology Atlas*, which is to be revised and republished so that it can be made available to a wider readership.

A recent and very significant initiative by EULEP has been the decision to establish a European *Radiobiology Archive*. The twin aims are (i) to create a centralized database for large long-term animal radiation experiments (which are unlikely to be repeated on such a scale in the future); and (ii) to set up a decentralized archive of pathological specimens from these experiments, which may be invaluable for future studies utilizing modern techniques. The EULEP Radiobiology Archive will in many ways match a similar venture being undertaken by the US DOE.

B11 Interpretation of low dose and low dose rate effects with the help of microdosimetry.

Contract FI3P-CT920027 Biophysical models for the effectiveness of different radiations.

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Total Contribution by the Commission: 490 KECU
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Description of research work

OBJECTIVES

In this proposal, experimental, computational, and system analytical work is foreseen to improve the present knowledge on the shape of the dose-time-effect curves especially in the low dose regime of radiation protection and its dependence on the time dependence of dose delivery (rate and fractionation), the type of radiation (X, gamma, alpha, neutrons and mixed fields) and on the influence of environmental factors.

The scientific approach to develop such an integral mechanistic (i.e. not empirical) model for radiation carcinogenesis is structured in this project into the development of several separate, serial models for the different levels of biological complexity, namely for:

a) the induction by radiation action of primary and secondary physical and early chemical changes in the DNA (by Terrisol et al., O'Neill et al., v. Sonntag et al.),

b) the subsequent formation of mutations (e.g. $HPRT^-$ and chromosome aberrations (Paretzke et al., Goodhead et al.),

c) the induction of cellular changes and late somatic effects in experimental animals and man by such early cell effects including intercomparison with the mechanistic action of UV-light
(Leenhouts et al., Paretzke et al.).

Particular emphasis will be given to the mechanistic interpretation of the $HPRT^-$ mutation and the induction of lung tumors by alpha emitters. The details of the contributions foreseen by the six partner groups are outlined below. Close contacts are also planned with groups of EURADOS improving the measurement techniques for relevant, risk related quantities, with groups improving the experimental biological data base for this topic, and with similar research activities in the USA.

Participation of GSF - Germany

In the previous contract period GSF has developed the track structure simulation code PARTRAC taking into account the space and time dependent transport of photon radiation and their secondary electrons in structured (i.e. heterogeneous) cells and tissues (with complex geometry routines and microscopic voxel 'phantoms') and the subsequent reactions of new chemical species with the DNA. This tool has been used to calculate the yields of single and double strand breaks of DNA, and of dicentric chromosomes for different photon energies. Usind the density functional theory the required inelastic electron scattling cross sections for condensed water were calculated.

In this contract period this work will be extended in three closely related aspects:

(i) Physico-chemical models:

The regime of incoming radiation fields considered in PARTRAC will be extended to p, α,γ,n,C,N,O, HZE particles from 1 keV to 200 MeV/u to be able to simulate tracks for all those radiation qualities for which radiobiological experimental data exist for the induction of the $HPRT^-$ mutation and of chromosome aberrations. Track structures will be made available to other partners for their specific analyses.

(ii) Biological, cellular model:

The computer simulation of the DNA in chromosomes and their dynamic behaviour in the cell during and after an irradiation will be improved (in co-operation with Noolandi et al., Xerox-Comp.) to obtain more realistic, mechanistic models for the later steps after DNA-ssb and dsb-production in the formation of mutations and chromosome aberrations.

(iii) Multi-step cancer model:

The results of these simulation calculations will be used towards the development of a mechanistic model for the induction of lung tumours in members of the public by the inhalation of Rn/Tn-daughter products. This work includes the intercomparison with epidemiological data on this endpoint and will be studied in close co-operation with other contract partners (e.g. RIVM).

Participation of MRC (Phys) - United Kingdom

MRC (Phys) will use radiation track structure calculations for analyses, perform *in vitro* experimental studies and review the literature to develop quantitative mechanistic descriptions of the effectiveness of radiations on mammalian cells, especially for mutational damage.

(i) During the previous contract period an extensive and consistent database has been built up of frequencies of energy deposition over dimensions of 1-100 nm for a wide variety of different radiations, simulated with Monte Carlo codes. DNA strand breakage was also evaluated from these codes by assuming a simple volume model of DNA. These data now allow comparisons of effectiveness between radiations. The robustness of these results will be explored against variations in the methods of simulation and scoring, and also compared with codes for other transport media when available. The database will also be extended to selected further particle energies and types. More advanced descriptions of DNA and early chemical processes will now be incorporated to assess more precisely the initial yields of DNA damage, including complexities of clustered breaks and base damage.

(ii) The experimentally observed effectiveness of different radiations in inducing genetic damage in mammalian cells will be assessed. Particular emphasis will be put on $HPRT^-$ mutations as a prototype system for mechanistic understanding and modelling because of the extensive information that already exists for this locus

(including much data on induction *in vitro* by different radiations and molecular characterization of radiation mutants). A compilation will be prepared of pertinent data in the literature, with emphasis on those studies revealing dependence on quality of radiation, dose or time. Selected *in vitro* experiments will be carried out where there may be important gaps or contradictions in the literature. This will include irradiations with α-particles, and possibly ultrasoft X-rays, with variations in gassing and dose rate. Studies will include assessment of the abilities of cells of different types to survive the passage of a single a-particle and hence to express α-particle-induced genetic changes.

(iii) Comparisons will be made of the radiation track properties (theoretical, part i) with mutagenic effectiveness (experimental, part ii) to seek those features that may be of critical importance in determining mutagenicity. Features at both DNA and cellular levels will be considered. Such analyses aid in the development of realistic mechanistic models to describe the induction of mutations by radiations, including at low dose and low dose rate, where the effectiveness of single tracks are crucial. Studies during the previous contract have pointed to the prime importance of highly localized clusters of damage in determining cell lethality and have led to quantitative hypotheses of different doses of initial damage with differing severity (or probability of not being repaired). Preliminary suggestions are that the probability of $HPRT^-$ mutations are further weighted towards the more severe types of damange. Properties to be explored include concentrations of local energy deposition over dimensions of DNA, or larger, and efficiencies of strand breakage, especially for breaks with greater complexity of associated damage.

Participation of ADPA CPA-France

CPA will continue to develop codes simulating physical interactions for low energy electrons and photons and of the evolution of chemical species as a function of space and time in the biological environment. The codes developed in the previous contract period work for liquid water medium, and it is possible to add solutes and/or scavengers. During this contract period we shall aim to develop codes for more realistic condensed biological media and for more complex structures like a cell with membranes, cellular liquid, DNA and nucleosomes. For simulation of primary interactions one needs cross sections for biological condensed media; CPA will use, for instance, data published from synchrotron experiments to derive such cross sections.

In a cell, chemical reactions take place continuously and are important. In an irradiated cell, we must modelize the perturbation due to the increase of chemical species created by radiolysis. To achieve between them; they will be extracted from the literature and improved before introduction in the codes. The structure and position of cell elements will be dynamic in order to obtain more realistic simulation.

These calculations will be complementary to the work of MRC: the possibility to add scavengers will be used in parallel with their experiments to measure the 'scavengeable' yield of radiation damage as well as to begin the explanation of the role of oxygen and the role of diffusible water radicals produced around DNA and the different chemistry associated.

These track structures and species distributions in space and time in a cell will be used by other partners in their models for radiation action, e.g. to make sensitivity analysis of model parameters and to analyse their relative significance.

Participation of RIVM - The Netherlands

The RIVM will continue the development of a comprehensive model for the analysis and interpretation of radiation effects at cellular, organ and animal level on the basis of specific radiation damage in the DNA. The model is intended to provide a framework for the effectiveness of different radiations on cellular effects and the influence of radiation on the development of tumours and cancer. The ultimate goal is to provide insight into the dose-time-effect relationships for late radiation effects and in particular the extrapolation of radiation effects in animal and man at high doses to the estimated radiation risks at low doses and dose rates.

The work will consist of both a theoretical and experimental facet:

1. In the theoretical approach, firstly, the track model developed in the previous contract period will be used to interpret sets of cellular radiobiological data on the basis of specific damage in the DNA, DNA double strand break. The model is based on first physical principles of the interaction of radiation with matter and accounts for the influence of physico-chemical effects using suitable parameters. The model will be applied to the cytotoxic and mutagenic (e.g. $HPRT^-$ mutation) action of radiation. Special attention will be paid to the different effectiveness of gamma- and X-rays. The results will be compared with the analysis of their partners in the contract.

Secondly, the influence of radiation in the development of tumours and cancer will be studied. A two-mutation model for carcinogenesis will be used and the knowledge of the respnse of cells to radiation gained from the radiation biological model developed in the previous contracts and the biology of cancer will be combined to interpret the effectiveness of radiation with e.g. dose, exposure time and age of the animal. Special attention will be paid to the relation of effects after acute high doses with those after chronic and lifetime exposures. The approach will provide a framework for the influence of biological factors, such as natural incidence of tumours and other factors on the radiation effect, especially at low doses. If possible, the results will also be used to analyse animal data found in other CEC contracts in order to define future research priorities.

2. The experimental approach will be a continuation of experiments from the previous contract on the comparison of cellular dose-time-relationships of ionizing and ultra-violet (UV) radiation. The differences of the influence of repair on the dose-effect relationships of cytotoxicity for the two types of radiation will also be studied for the $HPRT^-$ mutation. The results will be of importance to support and test the interpretation of tumour development after exposure to ionizing and UV radiation in the theoretical approach. Further, the experimental work will be used to investigate the different effectiveness of gamma rays and X-rays.

Participation of MPI - Germany

Radiolytic damage to the DNA in the living cell may be subdivided into two components, the direct effect and the indirect effect. In the former the energy of the ionizing radiation is absorbed by the DNA itself, in the latter by its environment, mainly water.

The radilysis of water produces OH-radicals, H-atoms and solvated electrons, all of whcih react very rapidly with DNA. These reactions have been studied by irradiating DNA in aqueous solution. On the other hand, firm experimental data on the direct effect are scarce. For such studies 'dry' DNA (i.e. DNA that still contains its water of hydration) must be used. In order to investigate the chemical reactions induced by the direct effect we will γ-irradiate solid DNA under controlled conditions of moisture content. The intention is to investigate some overall parameters such as strand breakage and release of unaltered bases, but more specifically we willtry to identify the alterations at the sugar moiety. In the case of γ-irradiated aqueous solutions of DNA, their identification allowed us to infer mechanistic details regarding DNA strand breakage caused by the indirect effect, and it is hoped that this will be possible now for the direct effect as well.

Non-coherent excimer UV-sources emlitting photons at 193 nm (KrF) and 172 nm (Xe_2) will soon be available. At these wavelengths the nucleobases are photoionized. Thus some aspects of the direct effect can be mimicked using these high-energy photons, with the aim to determine and quantify the products of nucleobase photoionisation. Specially tailored oligodeoxynucleotides will be synthesized to prove/disprove the hypotheses that adenine base radicals can induce strand breakage in DNA.

Our present knowledge of purine free-radical chemistry is still very limited. Radiolytic experiments in aqueous solutions with adenine, "'-deoxyadenosine, adenosine and poly(A) will be carried out in the absence and presence of oxygen with the goal to identify the products and elucidate the mechanism of their formation. Special attention will be paid to the role of the superoxide radical in the case of oxygenated solutions.

The DNA products will be identified by gas chromatography combined with mass spectrometry (GC-MS) after acid or enzymatic DNA hydrolysis and adequate derivatisation of the low-molecular-weight material, or, when possible, directly by high performance liquid chromatography (IIPLC). In the case of DNA model studies preparative HPLC will be used to accumulate enough material for further identification by NMR. In addition, we will assess the validity of a theoretical model which has been developed and successfully applied at our laboratory for the treatment of radiation damage in DNA in the presence of radical scavengers. The predictions derived from the model will be compared with numerical results of Monte Carlo calculations. The modelling of the direct effect and of the Schuler correction for the inhomogeneous production of OH radicals will be improved by taking into account definite track structures which will be made available by the GSF group.

A close collaboration with this group and the group at MCR is envisaged.

Participation of Queen Mary and Westfield College, Physics Dept. - United Kingdom

Work Done

The first 2-3 months have been spent acquiring the skills and expertise necessary for the manufacture of good quality multilayer Langmuir-Blodgett (LB) films. Overall film thicknesses between 3 and 120 nm of ω-tricosenoic acid and 12-8 diacetylene have been produced on solid glass substrates using a water sub-phase. Work has so far concentrated on studying the polymerization of 12-8 diacetylene and the real-time electronic response of ω-tricosenoic acid to a variety of radiation types.

Polymerization of 12-8 diacetylene

Films of 10 layers (30nm) have been exposed to broad band UV radiation (250-360nm), ^{241}Am-α particles (5.48 MeV) and 20 keV electrons. In addition to the energy differences, these radiations also had different fluxes which gave widely different energy deposition rates in the films. The degrees of polymerization were assessed using ir absorption at 585 and 638 nm and Raman scattering at 1455 cm^{-1}. Most measurements have been made using the UV and α-particle sources.

Electronic response of ω-tricosenoic acid

Multilayer films of ω-tricosenoic acid (thickness 120 nm) were sandwiched between aluminium electrodes (thickness 30 nm) and exposed to 5.48 MeV α-particles. Instantaneous currents of ~0.3 nA were produced, which quickly reduced to zero after quite modest total exposures.

Although these preliminary measurements must be repeated, they may suggest that highly ionizing radiation modifies the films in a way which alters their expected behaviour. In the two cases studied, the normal polymerization of 12-8 diacetylene under UV irradiation, and the highresistance of the insulatingt w-tricosenoic acid both appear to have been adversely affected by α-particle irradiation.

Work planned up to May 1994

Progress has been made on the production of multilayer films of 12-8 polydiacetylene (PDA) mounted on this metallic mesh. The PDA films are the fully polymerized form of the diacetylene films and are much more robust. It is proposed to use these as variable thickness samples for energy-loss transmission measurements, initially on the low energy proton beam.

For these measurements it is proposed to use thin metallic foils to further energy-degrade a low-current 50 keV proton beam, which will be collimated and electrostatically energy analysed before reaching the sample multilayer films of PDA. The energy-loss spectrum of the transmitted protons will then be determined using further energy-analysis. A comparison between the energy distributions of the incident and transmitted protons will then give an indication of the energy absorbed in nanometre thicknesses of film. Measurements will be made as a function of film thickness and incident proton energies down to 1 keV.

Complementary Monte Carlo calculations will also be performed in order to model the interactions and energy deposition events which take place along the radiation tracks in the thin films. A comparison between the calculated and experimental transmitted proton energies will give an indication of the reliability of the theoretical cross sections used in the code.

Work will continue on the use of nanometre-thick LB films as dosimeter material Effortwill be concentrated on the real-time response to low fluxes of low LET radiation.

Participation of MRC (Chem) - United Kingdom

MRC (Chem) will characterise the chemical stages in the development of DNA damage and its relationship to cellular consequences. This information will be used to develop more sophisticated models of radiation action through interfacing track structure approaches and the subsequent chemical pathways leading to DNA damage. Following the physical events, various chemical processes lead to localisation and modification of reactive chemical entities on DNA and ultimately to a variety of molecular lesions. An important facet of this proposal is the analysis of the clustering of DNA lesions for different radiation qualities.

There will be three inter-related approaches:

i) **Characterisation of radiation-induced DNA damage:**

The induction and repair (extent and kinetics) of cellular DNA lesions produced by radiations of different qualities (e.g. α-particles, γ-rays, soft X-rays) will be investigated. an important aspect of these studies is the modification of the chemical environment of the cell as a mechanistic aid. They will include a radical scavenger to measure the 'scavengeable' yield of radiation damage in order to assess the various chemical stages of radiation action. The techniques of sucrose sedimentation, neutral elution and pulsed field gel electrophoresis (PFGE) will be used to assess DNA damage. With PFGE, it is now possible to approach radiation doses of biological relevance. These studies will provide additional, detailed information on the complexities of DNA damage and repairability together with the role of the chemical environment in their relationship to the biological consequences of radiation action.

ii) **In vitro DNA damage mechanistic studies:**

The *in vitro* DNA studies will concentrate on the role of clustered damage and of radiation quality. Plasmid DNA strand breaks and S1 nuclease sensitive sites (clustered damage) will be determined arising from different radiation qualitites. The radiations will be carried out on DNA solutions in which scavengers are used to mimic the cellular environment through limiting diffusion of water radicals. At low LET, the role of water radicals produced within the water environment associated with the DNA has been identified as being involved in DNA damage induction. DNA will be irradiated at various degrees of hydration to assess the role of the structured water of DNA in strand break induction. The information obtained will enhance our understanding of the role of clustered damage and the hydration of DNA.

iii) Input of Chemical Information into Model Development:

The first generation model used to calculate initial yields of single- and double-strand breaks was based solely on track structure considerations. This model approximated to experimental data for the dependence of DNA damage on LET, however, it could not explain the role of oxygen and/or chemical repair. In order to produce the next generation of models, initially the following aspects will be considered: (a) The role of diffusible water radicals produced in the water associated with DNA using reaction radii based upon known rate constants and diffusion coefficients, (b) The different chemistry associated with inner hydration shell of water compared to the subsequent two to three water layers.

B11 Interpretation of low dose and low dose rate effects with the help of microdosimetry.

Contract FI3P-CT920041 Specification of radiation quality at nanometre level.

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Description of research work

OBJECTIVES AND EXPECTED ACHIEVEMENT

The common aim of this international research group is "research concerning the best possible way of specifying radiation quality for radiobiology and radiation protection by a single physical parameter expressing the track structure". The quoted definition of the group's aim, taken from the scientific purposes of the actual programme (1990-1992), can be easily recognized as an old but urgent target because the application fields of radiation protection and radiotherapy require a physical parameter which can predict its biological efficiency. That aim is difficult because the parameters studied in the past, especially the unrestricted LET and the lineal energy y for a reference volume size of about 1μm, have been proven to be too "rough" to play a role as indicators of track structure at nanometre level. But nanometer is the scale of the critical target of radiation effects, as both molecular biology and radiobiological analysis suggest.

The renewal of this old aim was motivated by new ideas which appeared to describe more adequately the meaningful track parameters at nanometre level (restricted LET and linear primary ionization) The research on the gas detector physics allowed to design microdosimetric instruments able to measure ionization distributions near the particle track with nanometre resolution and the recent advances in technology suggested the possibility to develop a nano-dosimeter in condensed phase. Modern Monte-Carlo codes, are able to simulate the interaction of particles with the matter at nanometre level. This supports both experimental and theoretical research.

The research group, been enlarged in the meantime by the experimental experience of ESR local radical concentration measurements of Roma group, with the new electron measurements calculations by the Darmstadt group, is going to renew that aim in the light of the achieved results.

STATE OF THE ART

The new analyses of the biological data has revealed the important unifying property of the mean free path ionization (λ), and led to the conclusion that the dominant radiosensitive sites are the double-stranded segments of DNA in cell nuclei. Good progress has been made in the development of a unified model for the direct action of radiation on mammalian cells. Confidence that the mean free path is indeed a key parameter for specification of radiation effect is obtained in terms of the new interpretation that emerges for e.g. the magnitude of the saturation effect cross-section; the inverse dose-rate effect; the apparently anomalously large effect of incorporated radionuclides; the occurrence for all radiation types of a common maximum in the RBE when mean free path for interaction is about 2 nm.

The inverse parameter "linear primary ionization density" (I_ρ) and the "restricted linear energy transfer " (L_Δ) have been shown to be closely interrelated. Each of these parameters,which are arithmetic mean values, fully characterizes the statistical distribution of the primary ionization along the path of the primary particle because the latter is a Poisson distribution, and far-reaching δ-rays are in this parametrisation

considered as quasi independent components of the radiation field, contributing on their own to the spectrum of I_ρ or L_Δ values at a given point in the irradiated medium. With regard to the low-energy δ-rays which, when originated from the path of an ionizing particle across a target of nanometre dimension, remain "insiders" of this region their initial spectrum has been postulated to be approximately invariant for all types and energies of ionizing particles as so their pattern of energy deposition. This "invariance theorem" is the base of the close correlation between L_Δ (with Δ = 100 eV) and I_ρ, Theoretical and experimental demonstration of its general validity is un important aim of this group.

The invariance theorem has been demonstrated valid for electron radiations between 100 eV and 150 keV. Moreover the evaluation of the Monte-Carlo results of Charlton et al. for protons and alpha particles and of Nikjoo et al. for photons have shown a good consistence with the invariance theorem.

Cross section data set for Monte-Carlo simulations have been used to calculate the energy deposition of proton tracks in the energy range from 10 keV up to 15 MeV traversing spherical target with diameters from 1 to 100 nm. The invariance theorem was analyzed as a function of proton energy. As indicator the dose mean lineal energy $\overline{y_D}$ for single secondary electron tracks was used. For nanometre targets only a variation of $\leq$10% was found.

To facilitate interpretation of radiation effects in terms of physical parameters of special interest to those in our collaborating group, calculations have been performed, in csda approximation, for a wide range of electron and photon energies and for some commonly used radioisotopes. Quantities calculated are: track and dose average LET, restricted LET_Δ (Δ=100 eV), relative variances, the mean linear primary ionisation and the corresponding mean free path, csda ranges, the mean energies required to produce a primary ion pair and kerma factors for electrons and photon radiations interacting in water. The minimum cut-off energy is 30 eV. Results have been tabulated for monoenergetic electrons (50 eV to 30 MeV), characteristic K_α x-ray line spectra (carbon to uranium), some commonly used bremsstrahlung (50 KV to 300 KV) and γ-spectra (241Am, 137Cs, 60Co, 125I and 131I) and for some typical radionuclides that decay by β emission or electron capture accompanied by Auger electron cascades (^{3}H, ^{14}C, ^{32}P, ^{125}I, and ^{131}I).

The multiplication characteristics of the cylindrical proportional counter equipped with a 10 μm anode have been measured in the propane based T.E. mixture at different pressures down to 22.7 Pa (0.17 torr) by using low velocity ions as probe. The results show that a drift region exists even at very low pressures. Findings of this experiment were used to design a gas track detector to measure the radial ionization distribution of an α-particle track. The detector has been able to measure the mean electron yields as close as 5 nm to the track with a resolution of few nanometres. A maximum of ionizations has been measured between 10 and 15 nm far from the track; similarly the relative variance of the number of ionizations shows a maximum at about 25 nm. These results are still under discussion because of the secondary electron emission from the walls and the avalanche statistics in the proportional counter which are introducing systematic uncertainties difficult to evaluate. The results show however that a track detector made up of a small drift region, where the particle travels, and a relative large multiplication region, to get a gain high enough at

low pressure, is able to perform microdosimetric measurements at nanometre level and to give experimental informations about the track structure and the general validity of the invariance theorem.

Nano-dosimetry in condensed phase has been explored with MOSFET-type devices and thin film scintillators. Initial experiments have been performed in n-type MOSFET in a specially designed configuration. With an intrinsic efficiency close to 100% and a sensitive region of about 15 nm, this device is a good candidate ,but further progress in this promising area will be expensive and requires specialist expertise.

Plastic scintillators can be prepared in nanometre layers, but they are unsuitable for use for the present dosimetry purposes because of their limiting threshold energy of 1 keV for a usable light output. Nevertheless a preliminary work has pointed out the new technique to suspend small spheres of scintillant of micron dimensions in a matrix of non-scintillanting plastic. In such a manner the cumulative light output from multiple spheres can be analysed in order to deduce, with the help of the proximity function theory, experimental dosimetric informations at nanometre level. In order to study the relevance of the track structure on the radiation's indirect effects, the electron spin resonance method has been optimized to measure the spatial distribution of the radical structures (spurs, blobs and other clusters) at nanometre level. ^{60}Co γ-rays and 12 MeV bremsstrahlung X-rays have been used to measure the absolute values of local (i.e. within the cluster or track) adical desities created by radiations with low LET. The technique used was the ESR continuous saturation method employed with an X-band ESR spectrometer fitted with additional components to assure a precise measure of power level in the microwave cavity.

1992-1994 PROJECT

As indicated in the EC research programme for radiation protection, radiation quality is one of the factors which characterizes radiation exposure, modifies the biological consequences and therefore needs to be considered in the assessment of radiation risk.

The important and straightforward practical aim of the coordinated project, its interactive mode of operation, underlined by Padova, St.Andrews, and Neuherberg meetings, the success of the first phase (1990-1992) and the achieved results which have confirmed the validity both of the theoretical and experimental approach, have convinced the coordinated groups to continue the collaboration for the next two years. A new participant, the group of Darmstadt directed by dr. G.Kraft, will enrich the collaboration with an experimental and theoretical study on the very first stage of the energy transfer process from heavy ions as projectile to the target material. This contribution will complete the outlook on the microscopic interaction of radiations with matter to investigate the best possible way of specifying the radiation quality by a single physical parameter expressing the track structure.

The physical part of the project aims at completing the establishment of the "invariance theorem" by experimental and theoretical studies. As result λ or its inverse I_ρ and L_Δ will get fundamental ground to become the key parameters of the radiation quality.

The radiobiological and radiochemical part aims to continue the correlation of existing radiochemical and radiobiological data with I_p and L_Δ to establish biophysical modelling and their relationships.

Detailed distribution of tasks amongst participant and collaborating links are the following.

LEGNARO

The track detector will be modified to allow for calibration with a single electron source produced with UV light. Electron diffusion in the drift region will be evaluated by using even "cold"gases like dimetylether. The amplification unit will be substituted with a multiwire multistep proportional counter able to count single electrons, so the track measurements will get rid of the avalanche statistics of the single wire proportional counter.

Ionization radial distributions with nanometre resolution will be performed with light ions by the CN Van de Graaff Legnaro accelerator in order to validate the "invariance theorem". This task will be achieved in collaboration with Neuherberg and Göttingen. The measurements will continue by the GSI heavy ion accelerator with the help of the Darmstadt group.

The experimental study on the physics of low-pressure cylindrical proportional counter will continue to conclude the feasibility study for a microdosimeter able to measure meaningful track structure parameters at nanometre level.

ST.ANDREWS

The data base will be extended to include results for transformations, incorporated radionuclides and double -strand breaks in DNA in mammalian cells.

The modelling work will continue to quantify the indirect radiation action in collaboration with the ESR studies by the Roma group.

The compilations of track structure quantities will be revised and compared with the Neuherberg and Darmstadt data in order to identify, in collaboration with Götttingen, the best track parameter which represents the radiation quality.

Research on novel detector for dosimetry in condensed phase will continue in collaboration with Legnaro.

GÖTTINGEN

Monte-Carlo studies of track structure and its regularities will be completed and compared with Neuherberg and Darmstadt data.

Theoretical and general aspects of track structure parameters and their interrelationships will be studied in collaboration with St. Andrews.

Systematic correlation of existing yield data for radiation induced radicals, molecular lesions and products, cell inactivation and transformation, chromosome aberrations and mutations, will be accomplished towards L_A in collaboration with St.Antrews and Darmstadt.

Experimental study of delayed fluorescence in liquid scintillators, due to triplet-triplet interaction, will be done in collaboration with Darmstadt to test the significance of L_A.

NEUHERBERG

Extension of proton cross section data set up to 100 MeV to test the "invariance theorem" in the energy range interesting for flight dosimetry.

Extension of cross section data to light ions; this task will be performed in collaboration with Darmstadt.

Performance of critical comparison among different track parameters to provide original calculations at nanometre scale for Göttingen and St.Andrews modelling.

Monte-Carlo simulations of the track measurements of Legnaro to compare with experimental data with light ions.

ROMA

The ESR methods of "continuous saturation" and "line broadening" will be improved both in respect of hardware as well as software to measure the local radical densities on a nanometre scale. Another window into the nanometre and subnanometre scale of radiative events are expected from observation of radical geminate pairs and from half-field observations coupled with the use Eatons' formulae. Irradiations at Legnaro and Darmstadt laboratories, with light and heavy ions, will enable to correlate the spatial distributions of radiative interactions with L_A, λ and other track parameters.

In collaboration with Göttingen, Neuherberg and St.Andrews the dependence of radical spacing (density) on the track parameters will be compared with the similar relationships found for molecular lesions, cell inactivation and transformation, chromosome aberrations, etc.

DARMSTADT

Measured double differential cross-sections (in angle and in energy) of electron emission from gas targets traversed by heavy ions will be used to implement a Monte-Carlo code of the electron transport to study light and heavy ion track structure at nanometre level. The results will be compared with the Neuherberg results which will be obtained with different code and input data.

The electron measurements will be extended to solid target.

Radial ionization distributions will be calculated to compare them with the experimental results of Legnaro.

Radial dose distributions will be a new input to test the radiobiological modelling of St.Andrews and Göttingen.

The project in its entirety falls within the framework of research topics II.1.1 and II.1.3 of the EC radiation protection programme and it is of fundamental interest for the research topics I.1. and II.2. Moreover it is of fundamental interest for all the applications in which mixed radiation fields and their variability make difficult to assess the RBE. For example neutron therapy and light ion therapy (EULIMA and future projects) will take benefit from the results of this coordinated project, as well as space dosimetry which has to deal with cosmic radiation in which are present all kind of atom nuclei (from hydrogen to iron) with energies varying from keV/amu to TeV/amu.

The project will be coordinated by Legnaro. The participants will organize yearly meetings to discuss the results achieved and to define in detail the common tasks. The meetings will produce written reports to be send to the Radiation Programme reference persons in Brussels.

CONTRIBUTION OF INFN - LEGNARO

In order to perform experimental microdosimetry at nanometre level with gas proportional counters (CPC), it is necessary to employ low pressures and thin anodes. CPC works in these conditions at very high reduced electrical fields and its gradient is very high., consequently the electron swarm is not in equilibrium with the electrical field and his behaviour cannot be predicted by the classical Townsend theory. The Legnaro research group of radiation physics has developed an experimental set-up to study the work characteristics of a cylindrical proportional gas counter (CPC) at low pressure (0.2-6.0 torr). Low-energy nitrogen beam has been used as a probe inside the gas detector. The beam scans almost all the sensitive volume between the cathode and the anode, parallel to the equipotential lines. The first ionization coefficient can be extracted from the gain data at different radial distances.

The experimental findings have been employed to design a gas track detector to measure the radial ionization profile around a charged particle track with nanometre resolution. The detector is composed of two parts: a small drift region in which the particle travels producing ionizations in the counting gas, and a multiplication region in which the electrons are multiplied to give a detectable signal. The two regions are connected through a narrow slit which determines the detector sensitive volume: only the electrons under the slit are measured. The variation of the beam-slit distance allows to measure the radial ionization yield.

First results point out the necessity i) to use UV laser beam as a probe to produce ionization lines moveable inside the detector to check the shape of the sensitive region; ii) to measure the electron diffusion contribution to the experimental data

uncertainty; iii) the opportunity to substitute the CPC with a multiwire multistep proportional counter (MMPC)which is able to detect single electrons one by one.

A new measurement chamber will be designed to allow for all the new tasks. The detector will be modified to measure single electrons. This task will be achieved making the electron to drift in the multiplication region after the slit, for a distance long enough to separate them in time. The MMPC is a fast detector ($\leq$1ns), then the avalanches produced by the electrons will be detected separately by a fast digitizer. In such a manner it should be possible really "to count" the electrons produced at a given distance from the particle track. The MMPC will be supplied by the Weizmann Institute (Israel) which has developed it.

Measurements will continue using alpha and electron sources to test and optimize the experimental set-up. Further light ion beams produced with the 7 MV Legnaro Van de Graaff accelerator and heavy ion beams produced at the GSI facility of Darmstadt will be employed. The data will be compared with Monte Carlo calculations of Neuherberg and Darmstadt groups. The results will be discussed in the frame of the "invariance theorem" proposed by the Göttingen group and in collaboration with it.

The efforts do design a tissue-equivalent proportional counter (TEPC), able to simulate nanometre dimensions, will continue. Measurements with a Rossi-like counter pushed the lower simulable size down to almost 100 nm. The data collected with a ion beam like a probe inside a TEPC suggest that is possible to decrease further the simulated volumes on the condition that the electron avalanche is properly confined. Low-pressure TEPC physics is not known because of the far-from-equilibrium conditions in which the detector works. In order to push on our knowledges in low-pressure gas detector physics, our experimental set-up will use the nitrogen beam supplied by the 7 MV Legnaro Van de Graaff accelerator to collect experimental data on the first ionization coefficient in low-pressure argon gas. The results will be compared with calculations in progress inside the EC project on TEPCs-based radiation protection instruments.

The project responsible is dr. Paolo Colautti, leader of the research group of radiation physics at the Legnaro Laboratories. The group has experience in dosimetry, microdosimetry, radiation physics,applied nuclear physics.

CONTRIBUTION UNIVERSITY OF ST. ANDREWS

During 1992-94 it is envisaged that work on biophysical modelling will be completed and that a clear indication will be obtained on type of instrumentation required for the measurement of absolute biological effectiveness. This final part of the programme is expected to be achieved by:

(i) extension of the biological data base to include results for transformations, incorporated radionuclides (Auger electron and alpha particle emitters); and for the measured yields of double-strand breaks in DNA in mammalian cells. The latter is believed to be a key test of the validity of the model and quality parameters used.

(ii) Revised track structure parameters for interpretations of effects. The compilation of the track structure quantities including restricted LET and mean free paths for ionization will be revised in the light of the new stopping power values published by ICRU, to include deuterons, tritons and boron to uranium ions at energies from 1 keV/amu to 1000 MeV/amu. Data will obtained for the equilibrium spectra as well as at istantaneous energies. Similar informations will be tabulated for neutrons and their equilibrium recoil spectra. This data will be compared wherever possible with Monte-Carlo calculations being pursued by our collaborators at Neuherberg and Darmastadt. The availability of agreed set of information will facilitate the collaborative efforts of St. Andrews and Göttingen to specify quality at the nanometre level.

(iii) Quantification of indirect radiation action for biophysical modelling. It is proposed to continue work aimed at accounting for the contribution from indirect effects to the radiation action. The philosophy applied in based on the assumption that since the DNA strand breaks are of the dominant lesion then if inactivation of single hit targets (e.g. enzymes) can be adequately well quantified for model purposes, it will be a simple process to apply statistical arguments to determine the probability for induction of double strand breaks by indirect action in mammalian cells. Consequently work will proceed to find methods of unifying inactivation studies on enzymes and other single hit targets for varying particle LET; varying concentration of enzyme in solution; varying scavenger concentration etc. Preliminary studies already indicate that there are good prospect of success. The work would be supplemented by the information on the effects of track inhomogeneities of radical concentrations, coming from the ESR studies by the Rome group. Transposition of the results to the conditions of the mammalian cells should be straightforward to give the final model which can be tested against published data on the induction of double-strand breaks in DNA, by a range of different radiation types. From the combined theory for direct and indirect action it is expected to be able to define the requirements of the instrument response for absolute dosimetry.

(iv) Novel detectors for absolute dosimetry.

Work already in progress, on MOSFET-type devices and thin film scintillators to evaluate the feasibility of dosimeters operational at nanometre levels will be continued. See the general introductory statement above. This work will be complement at the proceeding at INFN, Legnaro.

CONTRIBUTION OF UNIVERSITY OF GÖTTINGEN

In the proposed second period of this research project, the abilities of L_Δ to characterize radiation quality shall be investigated in full depth. Using the possibilities for cooperation in the coordinated group, and aiming at a broad acknowledgment of this simple and effective way of parametrization. The following partial tasks shall be performed under the supervision of prof. Harder:

- review of the quantum mechanical aspects of glancing collisions which cause the phenomenon of the invariant low-energy δ-ray spectrum (in consultation with dr. Leuthold).

- Collection of the further Monte-Carlo proofs L_A uniquely determines the fluctuations of energy deposition in a nanometre target, including heavy ions (in consultation with drs. Leuthold and Kraft).
- Consultation with dr. Colautti (Legnaro) regarding experimental tests of the "invariance theorem" with a single-electron counter.
- Completion of the biophysical model of "pairwise lesion interaction" to comprise intra-track interaction. This analysis will serve to identify which of spectra mean, e.g. the dose mean of L_A as to be used in various topological situations.
- Test of the claimed L_A dependence of biomolecular interaction between radiation-induced species at the physico-chemical level. The well-known LET-dependent effect in two-component liquid scintillators, known as "quenching" and "delayed fluorescence" of singlet and triplet states shall be reviewed and investigated in the light of the proposed parametrization of radiation quality. We want to extend the experimental basis for this phenomena towards heavy ions in cooperation with prof. Schumacher (Göttingen) and dr. Kraft (GSI Darmstadt). Consultations with drs. Ettinger and Izzo (Rome) are intended.
- Completion of our collection of correlation between bench-mark radiobiological results (molecular and cellular levels) with the appropriate mean values of L_A in cooperation with dr. Watt (St. Andrews).

In the final stage, the merits of parameter L_A to characterize tissue reactions, mutagenesis and carcinogenesis shall be investigated. In the case of successful tests, the coordinated group will propose international recommendations of utilizing quality L_A as a physical parameter of radiation quality.

CONTRIBUTION OF GSF-NEUHERBERG

Monte-Carlo simulation of charged particle track structure is an important method for the analysis of the spatial distribution of primary physical events at nanometre scale.

The basis of the computer codes is a cross-section data set which takes into account all relevant interaction processes and covers the full energy range. From that calculation of different proposed parameters describing radiation quality will be performed.

1. Extension of the proton cross-section data set up to 100 MeV. This energy range is not only interesting in testing the invariant straggling contribution but also of relevance for the high energy neutron component in flight dosimetry.

2. Extension of cross-section input data to light ions. In the low energy range (< 100 keV/amu) the Rudd cross-section modelling can be used to derive an effective charge of the ion from the stopping power cross-section by which the proton cross-sections can be scaled. Additionally double differential cross-sections measured by the Darmstadt group shall be included.

3. Performance of critical comparison among different quality parameters. The comparison strongly depends on a complete input data set for all ions of dosimetric interest in order to establish the restricted LET or primary ionization as single parameter of radiation quality.

4. Comparison of Monte-Carlo calculations with experimental electrons yields for the shape and absolute scale (e.g. gain factor of the experimental devices) measured at Legnaro for various ions.

COLLABORATIONS

1 and 3. Göttingen - Neuherberg
2. Darmstadt - Neuherberg
4. Legnaro - Neuherberg

CONTRIBUTION OF UNIVERSITY OF ROMA

The participating group of the Second University of Rome is concerned principally with the application of the techniques of Electron Spin Resonance to the problems of track structure on a nanometre scale. The particular focus of research is relation between the Radiation Quality and the local spin (radical) densities. The ESR techniques complement and supplement the computational and experimental techniques used by other members of the group. Using techniques different from those of the other teams, the ESR group performs a "post mortem" analysis on an irradiated specimen, when fast processes, immediately following the acts of deposition of energy, have run their courses. The ESR investigations need to be compared and correlated with the efforts of Monte-Carlo computational teams in verifying the spatial distributions of radicals as discrete events of high energy deposition (Darmstadt, Neuherberg). It is important to see if the dependence of radical spacing on the restricted energy loss follows the same law as was found for molecular lesions, cell inactivation and transformation, chromosome aberration etc. (teams at Göttingen, Darmstadt and St. Andrews). As some parameters, measurable by ESR, depend upon initial energy of δ-rays, there is already a partial evidence that the energy deposition fluctuations due to these δ-rays are indeed invariant.

Finally the information on linear free radical density on a local scale gives an estimate of the indirect action of radiation, of interest to St. Andrews team in their study of absolute biological effectiveness of radiation.

The present application is both an extension and a continuation of the previous one, as far as the methods, materials and characteristics of radiation studied are concerned. The irradiation will include heavy ions from accelerators in Legnaro and Darmstadt, with corresponding high and extra high LETs, needed to provide a meaningful comparison with the other techniques and to cover the range of radiations of interest to radiobiology and radiation protection.

The methods of measuring the local radical density will continue to employ the technique of continuous saturation, which has been improved in Rome laboratory and the line widening technique which offers a potentially higher precision of measurements, will be used as well. Furthermore, a new method has been recently described (Eaton and Eaton, J. Am. Chem. Soc. 104, 5002. 1982; Coffman and Pezhesk, J. Mag. Res. 70, 21. 1986) which may serve for measurements of mean spacing between the radicals on a subnanometre and nanometre scale. This technique will be try, where applicable.

The measurement of the mean cluster size and, possibly, the cluster size distribution will be done by technique of growth (build-up) curves, when applicable. The samples will include those of water, water equivalent gels, few pure organic liquids, samples of animal muscles, fat and bone tissue, aminoacids, some polymers and biopolymers. The samples will be irradiated, transported and measured at 77 °K, to avoid transformations and decay of radicals included by radiations. Where possible comparisons will be performed with samples at 300 °K and also with samples irradiated by low LET radiations. the results will be communicated to other participants in the project and published.

CONTRIBUTION OF GSI-DARMSTADT

Measurements of the δ-electron emission in heavy ion atom collisions and Monte-Carlo calculations of the track structure of heavy ions.

The track structure model of Katz and the microdosimetry approach by Rossi and Kellerer cannot explain the observed dependence of the action cross-section from LET or energy quantitatively. Therefore an experimental and theoretical program to study track structure was started at GSI. It is the goal of these studies to re-examine the complete reaction chain starting from the energy deposition electron emission and transport up to biological response. Measurement and calculation of the LET did not explain the measured LET dependencies even when restricted LETs were used. Experiment of the electron emission however found large discrepancies between experiment and theory.

In the experiments performed in close collaboration with H. Schmidt-Böcking and is atomic physics group at the University of Frankfurt/M the electron emission process is studied for heavy ion atom collision. In the experimental set-up the heavy ion beam traverses diluted gas jet. The energy spectrum of the electron created in heavy ion collisions in analysed with two electrostatic sector spectrometers. They cover an energy range between 100 eV and 4 keV and an angular range 25° 165°. Spectrometers for the analysis around 0° are presently developed. Double differential in energy and angle cross sections for the electron emission in collision in heavy ions with light gas atoms have been measured over wide range of projectiles. First results show significant deviations of the energy and angular distributions from the theoretical models. The overwhelming part of the electrons is emitted in forward direction. This is in contradiction to the assumption of an enhanced emission perpendicular to the particle trajectory, on which most model calculations in radiobiology are based. These experiments will be completed in order to understand the dependence of the electron emission on atomic number and particle energy. Up to now all experiments are performed using a gas target. However the question arises, whether the electron emission characteristics change when the target is in condensed phase (liquid or solid) which is more adequate for radiobiology comparison.

In parallel to the electron experiments a Monte-Carlo code for the electron transport in water target was developed, which follows each electron individually. The ionization and energy deposition as function of the radial distance from the centre of the track can be simulated with nanometre resolution. First calculations show that the computer code describes the electron transport in the track in an adequate manner for H_2O vapour. The input data for these calculations are double differential cross sections for electron emission by heavy ion impact. Up to now the Binary Encounter Approximation (BEA)

has been used. However, the experiments on electron emission after heavy ion impact showed that the BEA is a poor approximation of the physical reality.

In order to improve our track structure calculations, the measured cross sections as well as more realistic theoretical approaches for the electron emission like the CTMC (Classical Trajectory Monte-Carlo) method will be used. Refined radial dose distributions will be calculated and the Legnaro experiment be simulated.

B12 Repair and modification of genetic damage and individual radiosensitivity.

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Description of research work

Objectives

The project is a comprehensive multi-disciplinary approach to understand the molecular basis of radiation sensitivity in humans. Such knowledge is essential for assessing relative radiation risk between individuals. Increased radiosensitivity may in many cases be found in individuals who are heterozygous for genes controlling defects in DNA repair.

Scientific description

To investigate the various aspects of the project 7 laboratories are involved: 1. Department of Radiation Genetics and Chemical Mutagenesis, State University of Leiden, The Netherlands (RGL). 2. MRC Cell Mutation Unit, University of Sussex, Brighton, United Kingdom (CMU). 3. Department of Cellbiology and Genetics, Erasmus University, Rotterdam, The Netherlands (EUR). 4. Institut Curie, Paris, France (ICP). 5. MRC Radiobiology Unit, Chilton, United Kingdom (RBU). 6. Department of Molecular Genetics, State University of Leiden, The Netherlands (MGL). 7. Institut für Strahlenbiologie, GSF, Neuherberg, Germany (GSF).

The project contains three separate parts which are best presented as one overall programme in order to highlight the comprehensive nature of it. The work is a detailed study at the cellular and molecular level of type and initial frequency of radiation damage, the repair and the ultimate biological effects.

1. Identification of radiosensitive individuals both in the normal population and in individuals with known radiosensitive genetic disorders.

A battery of complementary techniques, developed in the different participating laboratories, will be used to address this problem. The assay systems will be at the molecular, subcellular and cellular levels. We will continue to measure the radiation sensitivity of fibroblasts and T-lymphocytes using clonal assays to expand the database of normal individuals (CMU). There is now firm evidence to suggest that about 10% of breast cancer patients are heterozygous for the ataxia-telangiectasia (A-T) gene. We have developed a procedure for identifying A-T heterozygotes based on the inability of their fibroblast cultures to repair radiation induced clastogenic damage. We will extend these studies to lymphocytes, as these cells would be the only candidates for possible future screening programmes. We shall measure the repair of clastogenic damage in a series of breast cancer patients to see if a proportion of these patients are deficient in this repair. Detailed investigations of A-T variants and other radiation-sensitive individuals will be pursued as a source of additional human radiation sensitivity genes. Single and double strand breakage repair will be measured in these cells by single cell microgel assay which we have now established as an efficient reliable technique (CMU, ICP).
Our collection of repair deficient rodent cell lines will be expanded. New mutants will be characterized with regard to known human and hamster complementation groups (RGL).

2. Understanding of the cellular response to radiation.

Different and complementary strategies will be used in the different laboratories.

Cloning of DNA repair genes.

It has now been unequivocally demonstrated that basic molecular processes are conserved through the eukaryotic ladder, and that genes involved are conserved between yeast and man. This proven value of the easily manipulated yeast genes as stepping stones to obtain the homologous human genes will be vigorously pursued. The cloning of genes is a vital step towards identifying their function in repairing radiation damage. In the previous period we have cloned the human DNA repair genes ERCC1,3,6 and HHR6A and 6B and purified the XPA protein (EUR). We will attempt to map and clone the human genes complementing the radiosensitive *xrs* and *irs1* Chinese hamster cell lines which we have isolated and characterized (CMU, RBU). It has been shown that the human gene correcting one of the radiosensitive hamster *xrs* mutants is located on chromosome 2, and using radiation reduction hybrids we will further localise the gene to subfragments of chromosome 2. These hamster/human hybrids containing a small fragment of human DNA which includes the *xrs* gene, will be used to clone this gene, using positional cloning techniques (CMU).

Three X-ray sensitive hamster mutants belonging to the same complementation group show phenotypically homology with cells derived from patients with A-T. These cell lines will be used to clone a human gene that confers resistance to X-irradiation (RGL).

We have cloned 12 DNA repair genes from the fission yeast *Schizosaccharomyces pombe*, and are in the process of analyzing these genes. Some of them have homologues in the evolutionary diverged budding yeast, *Saccharomyces cerevisiae*. We will used the sequences conserved between the two yeasts to clone the corresponding human genes using PCR techniques. The *S. pombe* genes will also be studied as a model system for human DNA repair genes, in particular in relation to their involvement with the cell cycle (CMU).

In vitro DNA repair assays.

Human cell free assays will be used to analyze the enzymatic basis of repair and misrepair of relevant types of DNA damage at defined sites. The engineering of DNA molecules to model complex types of damage, similar to those induced by ionizing radiations, and to analyze the specificity of misrepair, is an important aim. We will also investigate the defects in A-T, using such molecular assays (RBU).

The role of chromatin structure in repair of radiation damage will be investigated by using isotonic extraction procedures of cells, encapsulated in agarose beads, to obtain intact chromatin as a template to study DNA repair in an *in vitro* system. Such a system is based on extraction of the majority of proteins from encapsulated cells while maintaining vital cellular functions such as replication, transcription and repair (RGL).

Functional analysis of proteins overproduced by cloned repair genes will be carried out using *in vitro* cell free excision repair assays or by micro-needle injection into repair deficient human cells (EUR).

Topography of DNA repair

Repair studies at the level of the genes will concentrate on investigating different types of radiation induced DNA lesions including UV-induced photolesions (RGL) and X-ray induced lesions such as DNA breaks and S1 sensitive sites. These studies will be carried out using mammalian cells (RGL) as well as yeast (GSF and MGL).

Certain types of DNA damage other then strand breaks induced by ionizing radiation under anoxic conditions are substrate for the S1 endonuclease: they are processed to DNA double strand breaks (DSB). S1 sensitive sites (SSS) are induced 1.5 to 2 times more frequently than DSB. SSS are repaired under non-growth conditions in diploid yeast. SSS repair, like DSB repair, involves recombination events but unlike DSB repair, additional gene products are required (*e.g.* the Rad18 and Rev2 protein in yeast). The research aims to elucidate the biological relevance of SSS. Some preliminary evidence suggests that SSS are induced non-randomly in yeast chromosomes possibly depending on the chromatin structure. This question will also be investigated in mammalian cells (GSF).

Molecular analysis of mutations

To understand molecular mechanisms of radiation-induced mutations, especially the formation of large deletions and rearrangements in human cells, we will characterize both X-ray and ∝-particle-induced mutations at the sequence and genomic levels (RBU). Additionally we will employ methods to rapidly define mutation spectra at different doses, dose rates (RGL) and with different radiation qualities (RBU).

Spectra of genetic changes at the DNA level will be determined in endogenous genes in normal and repair deficient cell lines. Spectra will be correlated to the kinetics and the extent of repair of specific types of DNA damage in the same gene (RGL, ICP).

3. Tissue and organ specificity of the response to radiation.

Use of transgenic animals

Cloned human genes will be used to disrupt the corresponding genes in mouse embryonal cells. These can than be used to create transgenic animals containing mutations in genes known to affect the radiation response. The effects of these mutations on the systemic effects of radiation can then be studied, for example radiation induced carcinogenesis, effects on the immune system etc. (EUR). Transgenic animals containing a marker mutation target gene will also be used to study mutation at the molecular level in whole animals with regard to tissue and organ-specificity (RGL). It is proposed to begin to exploit these new (and expensive) techniques, although this project is likely to extend well beyond the contracting period.

Interference of radiation with gene expression in human keratinocytes.

Exposure of living cells to radiation results in rapid alterations in gene expression. The finding that many of the affected genes are involved in signal transduction (*e.g.* proto-oncogenes and tumour suppressor genes) strongly suggests that such alterations might play an important role during the onset of carcinogenesis. In order to study interference of radiation with normal gene expression we use cultured human keratinocytes and a family of radiation responsive genes (spr genes) which are also regulated during the normal process of keratinocyte differentiation *in vitro* and *in vivo*. The research presented here is confined to *in vitro* cultures of human keratinocytes (submerged and organotypic) in order to identify the molecular factors involved in radiation induced deregulation of normal gene expression. At later stages, when the responsible factors have been identified, the same techniques can be applied to skin biopsies in order to monitor radiation exposed individuals (MGL).

Detailed description of the contribution of MGC - Department of Radiation Genetics and Chemical Mutagenesis, University of Leiden, Leiden, The Netherlands (RGL)

Topography of DNA repair:

We will investigate the role of chromatin structure (nuclear matrix, active and inactive chromatin) and transcription in repair of radiation induced DNA damage. Isotonic extraction procedures of cells, encapsulated in agarose beads, will be employed to obtain intact chromatin as a template to study DNA repair in an *in vitro* system. Such a system is based on extraction of the majority of proteins from encapsulated cells while maintaining vital cellular functions (replication, transcription repair). The encapsulated chromatin is accessible to molecular probes such as inhibitors, enzymes, antibodies and nucleic acids. Repair synthesis, measured by incorporation of radioactive or biotinylated precursors of DNA synthesis, can be studied at the level of the nuclear matrix and defined chromatin regions by enzymatic digestion of chromatin loops and immunological purification of repaired DNA respectively.

Repair studies at the level of genes will concentrate on investigating different types of radiation induced DNA lesions including UV-induced photolesions (cyclobutane pyrimidine dimers, 6-4 photoproducts) and DNA strand breaks (nuclease S_1 sensitive sites) in different chromatin regions in repair proficient and deficient mammalian cells. Repair studies will include the target genes for analysis of mutations i.e. the HPRT and APRT genes.

Molecular analysis of mutations:

Spectra of genetic changes at the DNA level will be determined in the endogeneous HPRT gene in UV-irradiated normal and repair deficient cell lines. Spectra will be correlated to the kinetics and the extent of repair of specific adducts at the HPRT gene. We are planning to extend these types of studies to the APRT gene. The preferential induction of mutations in the transcribed strand of the HPRT gene in repair deficient UV-irradiated hamster cells has pointed to different fidelities of leading and lagging strand synthesis on UV-damaged DNA templates. Further studies aimed to elucidate the

precise role of replication in mutagenesis will focus on the types and distribution of mutations at the APRT locus in repair deficient cells.

Isolation and characterization of repair deficient mutants:

Our collection of repair deficient rodent cell lines will be expanded. New mutants will be characterized with regard to known complementation groups and to mechanisms of DNA damage processing employing biochemical methodologies.

Cloning of repair genes:

Three X-ray sensitive hamster mutants belonging to the same complementation group show phenotypic homology with cells derived from patients with ataxia telangiectasia. These cell lines will be used to clone a human gene that confers resistance to X-irradiation.

It has been shown recently by the work of the EUR group that human repair genes can be successfully cloned from yeast repair genes employing strong sequence homology. This strategy will also be used to clone repair genes in Drosophila. For further characterization of cloned repair genes several repair deficient Drosophila mutants are available.

Detailed description of the contribution of MRC Cell Mutation Unit, Brighton, U.K.

Identification of radiosensitive individuals

In an extension and exploitation of sub-project 1 of our current programme, we will continue to measure the radiation sensitivity of fibroblasts and T-lymphocytes using clonal assays to expand the database for normal individuals. There is now firm evidence to suggest that about 10% of breast cancer patients are heterozygous for the A-T gene. We have developed a procedure for identifying A-T heterozygotes based on the inability of their fibroblast cultures to repair radiation induced clastogenic damage. We will extend these studies to lymphocytes, as these cells would be the only candidates for possible future screening programmes. We shall measure the repair of clastogenic damage in a series of breast cancer patients to see if a proportion of these patients are deficient in this repair. Detailed investigations of A-T variants and other radiation-sensitive individuals will be pursued as a source of additional human radiation sensitivity genes. Single and double strand breakage and repair will be measured in these cells by the single cell microgel assay which we have now established as an efficient and reliable technique.

Cloning of human DNA repair genes

We have shown that the human gene correcting the radiosensitive hamster xrs mutants is located on chromosome 2, and using radiation reduction hybrids we will further localise the gene to subfragments of chromosome 2. These hamster/human hybrids containing a small fragment of human DNA which includes the xrs gene will be used to clone the xrs gene, using both transfection and other molecular techniques.

We have cloned 12 DNA repair genes from the fission yeast *Schizosaccharomyces pombe*, and we are in the process of analysing these genes. Some of them have homologues in the evolutionarily diverged budding yeast, *Saccharomyces cerevisiae*. We will use the sequences conserved between the two yeasts to clone the corresponding human genes using PCR techniques. The *S-pombe* genes will also be studied as a model system for human DNA repair genes, in particular in relation to their involvement in the cell cycle.

Detailed description of the contribution of Department of Cell Biology and Genetics, Medical Genetics Centre, Erasmus University, Rotterdam, The Netherlands

Molecular, Genetic and Biological Analysis of Mammalian Excision Repair.

In the previous grant periods we have cloned the human DNA repair genes ERCC1,3,6, and HHR6A and 6B and purified the XPA protein. From their analysis the following general conclusions can be drawn: (i) A striking sequence conservation exists between human and yeast repair proteins that does not extend to *E. coli*. This suggests that the repair pathways in which these proteins operate are strongly conserved at least within eucaryotes. (ii) Several of the cloned genes are responsible for human repair disorders: ERCC3 for XP complementation group B and ERCC6 for CS group B. (iii) With respect to the function: the XPA gene product is shown to be a DNA-binding protein, ERCC3 and 6 are likely to encode DNA helicases. ERCC6 appears to be specifically implicated in the preferential repair of active genes. The HHR6A and B proteins are - in analogy to their yeast RAD6 homolog - ubiquitin-conjugating enzymes presumably involved in repair associated chromatin remodelling.

The long term goal of the research is to unravel the molecular intricacies of the mammalian excision repair pathway, its involvement in (the prevention of) carcinogenesis and to elucidate the genetic defect in the complex repair syndromes XP and CS.

For the next period we would like to concentrate on the following main topics: (i) Cloning and characterization of additional human repair genes via genomic or cDNA transfection to repair deficient rodent or human mutants or by sequence homology to cloned yeast repair genes. (ii) Functional analysis of the encoded proteins, involving generation of monospecific antibodies, purification of the (overexpressed) proteins, assay for activity using a cell-free *in vitro* excision repair system or microneedle injection into repair-deficient XP cells, analysis of specific functions (such as: lesion-specific DNA binding and incision, DNA unwinding or other DNA metabolizing activities) and interaction with other polypeptides of the DNA repair, recombination or replication machinery. (iii) Generation of a mouse model for XP/CS and other repair deficiencies based on recently developed techniques for targetted genereplacement in totipotent embryo-derived stem cells (ES-cells) from which mouse germline chimera's and eventually homozygous mouse mutants can be derived. Such repair-deficient mouse strains will be invaluable for assessing the biological role of repair and other factors in carcinogen-induced tumorigenesis, as an experimental model for XP and CS (etiology, treatment and prevention of the disease), for the sensitive screening of radiation and chemical agents for their carcinogenic, mutagenic and cytotoxic potential *in vivo*, and for risk assessment of heterozygotes, carrying a recessive repair defect.

Detailed description of the contribution of Institut Curie, Section de Biologie, Paris, France.

Our group essentially contributes to projects 1 and 2 of the overall proposal.

1. Project 1 aims at the identification of radiosensitive individuals. Breast cancer patients of the Institut Curie were examined for the frequencies of mutations at the HPRT locus in lymphocytes before and after radiotherapy and/or chemotherapy. The follow up of these patients will be pursued. In order to understand the molecular nature of the events taking place, mutant clones will be analysed for gene rearrangements and point mutations. Moreover the "comet" assay which allows rapid detection of strand breaks and their repair will be applied in collaboration with the CMU group to our patients in parallel to normal donors. We demonstrated an adaptation to the mutagenic effect of radiations at the HPRT locus by preexposure to low doses of X-rays. The molecular analysis of the induced mutants will be completed by pulse field electrophoresis studies.

The role of activated oncogenes on the modulation of the response of mammary epithelial cells to ionizing radiations (Ref. 2) in relation to repair has been examined for the *ras* oncogene. This approach will be extended to other cellular and genetic context relevant to the *in vivo* situation.

Among the genetic defects in the processing of DNA lesions studied, our group is in charge of the Fanconi anemia (FA) syndrome. We demonstrated that FA cells are hypomutable at the HPRT and Na^+/K^+ ATPase loci. A high frequency of deletions is produced in FA lymphoblasts in comparison to normal cells (Ref. 3). Preliminary data on erythroblastic cells show that, in FA, the glycophorin A marker is lost with a much higher frequency than in normal donors. In view of our results, FA cells offer a good model system to analyse the mechanism of deletions production which is one of the major genetic event induced by ionizing radiations. This will be followed up using multiplex PCR.

2. Project 2 of the programme deals with the understanding of the cellular response to radiation by cloning of human DNA repair genes and by complementary approaches. The molecular cloning of a DNA fragment which complements the FA genetic complementation group B leads to the dissociation of different phenotypic features (Ref. 4) indicating that at least two domains are involved. The molecular analysis of the DNA fragment demonstrates a high degree of conservation among species. It has been localized by *in situ* hybridization. The fragment is expressed at a constitutive level and the size of the mRNA has not been established. The cDNA, although still uncomplete, is sequenced. From the data bank, it does not seem to correspond to a known gene. It remains to determine if FA cell lines contain mutations and to establish the segregation of the probe in FA families.

In the course of the study of the complementation of the FA defect by cocultivation with normal cells, we discovered a new feature of the FA syndrome which might explain the defect in differentiation of the hematopoïetic system. Anomalies in production of two cytokines, interleukine-6 and tumor necrosis factor α, are indeed observed both *in vitro* and *in vivo*. This analysis will be extended to other chromosomal instability syndromes associated to hypersensitivity to radiations. We propose a model implying a network of

genes (including growth factors and repair genes) which would be under the control of common genetic element. This model will be explored.

Detailed description of the contribution of MRC Radiobiology Unit, Chilton, Didcot, U.K.

Isolation of the human gene complementing the irs1 radiosensitive mutant.

Our cloning strategy will be based on the availability of an *irs1* subline carrying small fragments of human chromosome 7, complementing the radiation sensitivity of irs1. We will first derive lines with reduced amounts of human DNA by irradiation, and then screen these lines for the resistance phenotype. Thereafter polymerase chain reaction (PCR)-based techniques or cosmid cloning will be used to find human sequences, followed by screening of human DNA libraries to identify candidate genes.

Molecular analysis of repair/misrepair of defined types of DNA damage at specific sites in DNA molecules

Our studies will use an assay based on cell-free extracts and defined DNA substrates, with the aim of isolating the proteins involved in radiation-damage repair, and to reconstruct this process *in vitro*. Candidate proteins known to have an effect on DNA metabolism (e.g., replication, recombination) will also be tested in this assay. The modelling of radiation damage at specific sites will be extended with the creation of complex types of damage, such as breaks and base damage in close proximity. We will also investigate the defects in radiosensitive cell lines, especially the human disorder ataxia-telangiectasia, using this molecular assay.

Our finding that specific sequences are involved in misrejoining suggests that we have devised for the first time an *in vitro* model for the analysis of deletion formation. These studies will be taken in at least two directions to study mechanisms of misrejoining: (i) the engineering of DNA molecules to create sites which would be predicted, from our data, to promote or reduce the incidence of deletions; (ii) the assay of enzymes considered relevant to the misrejoin process and isolation of these where they can be shown as important to the mechanism of deletion formation.

Studies on the nature of radiation-induced mutations under different irradiation conditions

Molecular analysis of the nature of deletions in the hprt gene of primary human cells will be based largely on PCR techniques. We will continue the analysis of a set of independent mutants induced by X-rays, where 4 large deletion breakpoints have already been sequenced, and extend the study to α-particle-induced mutants. The size of deletions will be analysed by pulsed-field gel electrophoresis (PFGE), by probing with X-linked anonymous sequences, and using yeast artificial chromosomes containing the genomic region around hprt. Where very large deletions are suspected, we will use high-resolution chromosome banding to look for a visible change. We will also use multiplex PCR for several sites in the hprt gene, combined with rapid DNA preparation methods, to define mutation spectra at different radiation doses and with different radiation qualities.

Detailed description of the contribution of Department of Molecular Genetics, University of Leiden, Leiden, The Netherlands (RGL)

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The experimental approach to analyse radiation induced alteration of gene expression consists of using human keratinocytes and a family of radiation responsive genes (spr genes) which were shown to be regulated also during the normal differentiation process of human skin. The advantage of using cultured keratinocytes resides in the fact that these cells retain in culture their natural ability to differentiate. By using spr genes as molecular probes, both the molecular mechanisms involved in the normal regulation of these genes during keratinocyte differentiation and the factors responsible for alteration of gene expression after radiation exposure can be identified. So far we were able to show that the homeodomain protein oct-1, the proto-oncogenes c-fos and c-jun and an as yet unidentified transcription factor play an important role in modulating gene expression after radiation injury. However the molecular mechanisms by which these different regulatory factors interfere remains to be established. Interestingly, human keratinocytes appear to respond differently to radiation, depending on their differentiation status at the moment of their exposure to radiation. This is an important finding as it indicates that in a same organ (skin) different cells of the same lineage (keratinocytes) can respond completely differently to radiation exposure. The molecular reason for this differential response is not yet known and remains to be elucidated. The present project intends to determine (some of) the cellular factors and molecular mecanisms involved in radiation induced alteration of gene expression by using cultured human skin cells as a test system. At later stages, when these factors and mechanisms have been identified, the possibility exists to use similar techniques to monitor skin biopsies from individuals after radiation exposure. Such methods might become an important tool to assess and quantify the immediate and long-term biological effects of human exposure to radiation.

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Objectives

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Within a 2-year period the following topics will be investigated:

1. We plan to analyse SSS in chromatin differing in structure and function and, for control, DSB as well which presumably are distributed randomly. We want to find answers to the question whether the induction, distribution and/or repair of SSS and DSB, respectively, are influenced by the chromatin structure. Individual yeast chromosomes and defined restriction fragments of human chromosomes are separated by PFGE and the chromatin regions of concern are identified by suitable gene probes.

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Detailed description of the contribution of GSF - Forschungszentrum für Umwelt und Gesundheit, Institut für Srahlenbiologie, Neuherberg, Germany.

Objectives

The long-ranging aim of our studies is analysing the induction and repair of S1 nuclease sensitive sites (SSS) by ionizing radiation in the genome of human cells and of the yeast *Saccharomyces cerevisiae*. We will quantify the induction and repair of SSS in chromosomal regions differing in chromatin structure and function. Identifying regions in the chromatin exhibiting an increased sensitivity towards radiation-induced DNA damage is expected to improve the understanding of effects caused by low doses of radiation. Second, we plan to search for mutants deficient in SSS repair. These mutants will be useful to analyse genetic consequences of unrepaired SSS, approaching the question whether SSS-specific repair processes exist and contribute to the protection against radiation effects including cancer. Third, we will continue to clone and characterize the *REV2* gene involved in the genetic control of SSS repair in *S. cerevisiae*.

Within a 2-year period the following topics will be investigated:

1. We plan to analyse SSS in chromatin differing in structure and function and, for control, DSB as well which presumably are distributed randomly. We want to find answers to the question whether the induction, distribution and/or repair of SSS and DSB, respectively, are influenced by the chromatin structure. Individual yeast chromosomes and defined restriction fragments of human chromosomes are separated by PFGE and the chromatin regions of concern are identified by suitable gene probes.

2. The capability for repair of DSB and SSS will be investigated in X-ray-sensitive mutants of yeast and mammalian cells, including cell lines of the human disorder Ataxia telangiectasia (A-T) applying the PFGE technique. We search for mutants which are capable for DSB repair and incapable for the repair of SSS. In future, these mutants will be used to investigate whether SSS play a role for cell death or mutation.

3. We are searching for the human gene homologous to the *REV2* gene from yeast which was cloned and characterized in our lab. The *REV2* gene controls processing of DNA damage induced by radiation and chemicals; it seems to be required for the repair of gamma ray-induced SSS (but not for DSB) and for fixation of premutagenic lesions. The analysis of the regulation of the yeast *REV2* gene expression will be extended. For cloning the human *REV2* gene we shall rely on the experience of the CMU searching human genes homologous to the genes of *Schizosaccharomyces pombe*.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920011 Cytogenetic and molecular mechanisms of radiation myeloid leukaemogenesis in the mouse.

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Description of research work

Aim and objectives

The principal objective of this proposal is to gain a detailed understanding of the complex mechanisms that underly the radiation induction of acute myeloid leukaemia (AML) in the mouse. Animal models of neoplasia have contributed much to our understanding of early events in tumorigenesis and the CBA/H mouse offers a well studied, non-viral model of AML that shows a close histopathological relationship to its human counterpart. Some information on the cytogenetic and molecular nature of early radiation-induced events in CBA/H leukaemogenesis has already been obtained but the picture is far from complete. It is anticipated that the proposal will succeed in extending this knowledge to the involvement of specific genomic damage in haemopoietic cells, the crucial role of AML-predisposing germ line mutations in the induction process and the immediate post-irradiation cellular consequences of early events in leukaemogenic development.

State of the Art

Many human leukaemias are characterised by specific chromosomal rearrangements and deletions. In most cases however the temporal position of these events in the multistep process leading to malignancy is obscure and, although some leukaemia subtypes are radiogenic, in no cases is it possible to comment directly upon the specific inducibility of a leukemia-initiating event by radiation. Similarly while there are some indications of human genetic susceptibility to leukaemia the evidence is far from compelling. In this respect, the involvement of heritable chromosomal fragile sites in leukaemogenesis has been suggested but remains a contentious issue. For the induction of mouse AML by radiation the situation is becoming progressively more clear. More than 90% of induced AMLs in a number of mouse strains show a characteristic rearrangement/deletion of chromosome (ch)2; loss of an interstitial region of this chromosome was the most consistent feature of these events but site-specific ch2 breakage was also evident. Since similar clonal ch2 events have been recorded in irradiated and repopulating normal mouse marrrow cells it has been concluded that ch2 rearrangement/deletion probably represents an induced intitiating event for AML. Further to this, published cytogenetic studies suggest that specific breakage of ch2 at putative fragile sites might involve telomerelike repeat (TLR)DNA sequences containing the motif $(TTAGGG)_n$; also that in some AMLs there is DNA sequence loss and/or modification in the interleukin (IL)-1 haemopoietic gene cluster region (mapping to 2F). It was also concluded from these studies that the CBA/H mouse might carry a germ line mutation predisposing to induced AML. More recently, unpublished fluorescence in situ hybridization (FISH) analysis at NRPB indicate that murine ch2 contains a number of extensive TLR sequence arrays and that the location of two of these cytogenetically corresponds with two of the fragile radiation sensitive sites noted above. Parallel molecular studies also indicate that these TLR arrays may take the form of dispersed inverted repeats of the motif and tht the ch2 F region array may be closely linked to the IL-1 gene cluster where AML gene loss and/or activation has previously been reported. Most importantly, however, evidence has been obtained that a specific TLR sequence polymorphism (TLRp) present in 20-25% of mice in the CBA/H colony identifies 17/18 host animals presenting with induced

AML; this quite unexpected finding in a highly inbred animal strain implies both genetic heterogeneity in the colony and the presence of a germ line mutation predisposing to induced AML that is genetically linked to this TLRp. This conclusion also accords with a) previous cytogenetic observations, noted above, and b) the 20-25% maxiumum incidence of induced AML in CBA/H and c) with the loss of leukaemogenic radiosensitivity recorded in some sub-colonies of CBA/H. These data highlight the need to further explore the involvement of TLR sequences and chromosomal fragile sites in radiation-induced breakage of ch2, the nature of the AML-predisposing mutation and associated gene losses together with the cellular consequences of such events.

Scientific and technical description of the proposed programme

On the basis of the research needs identified above II it is proposed to extend knowledge on mechanisms of murine radiation leukaemogenenesis through studies on a) the involvement of TLR sequences in induced ch2 rearrangement in AMLs, b) the nature of the putative AML-predisposing mutation, c) the specificity of gene losses from ch2 and d) the cellular consequences of the AML-predisposing mutation to cells of the myeloid lineage.

A. Molecular cloning and sequencing of TLR sequences

Using radiolabelled $(TTAGGG)_{2-3}$ and degenerate forms of this motif, plasmid and bacteriophage libraries of murine genomic DNA will be screened for clones containing TLR sequences. Particular emphasis will be placed on obtaining TLR clones from microdissected ch2 libraries (see B). TLR-containing recombinant clones will be sequenced and TLR and unique sequences sub-cloned using conventional techniques; these will be used as molecular probes for fluorescence in situ hybridisation (FISH) mapping and for conventional gel electrophoretic analysis of AMLs with a view to establishing their relationship with the specific ch2 breakpoints and loss events in AMLs (see E).

B. Chromosome 2 microdissection end FISH painting

A PCR technique for the production of ch2 specific DNA libraries from microdissected metaphase DNA has been established (with C. Tease, MRC, Chilton) and a pUC19 library of ± 2500 clones is already available. This will be extended and used for ch2 painting by FISH in order to quantify and characterise induced ch2 events in AMLs and irradiated marrow cell populations in vivo (with M. Ellender and J. Harrison, NRPB, Chilton). These libraries will also act as a source of ch2 encoded TLR and linked unique sequence probes for subsequent analysis of ch2 breakpoints and gene losses (see A and E).

C. Molecular cloning, sequencing and mapping of the TLR sequence polymorphism (TLRp)

TLRp may be closely linked to a putative AML-predisposing mutation and the cloning of TLRp is an important step in the identification of the mutation. Appropriate TLRp restriction fragments already identified in mice presenting with AML will be gel purified, cloned into a λ vector, subcloned and sequenced. TLRp subclones containing non-repeat

suquences will intially be used for FISH chromosome mapping. Further mapping of the polymorphism will involve the screening of DNA from recombinant inbred (RI) mice which will establish linkage with existing mouse loci of known location (with J. Peters, MRC, Chilton). Collaboration (with S. Brown and F. Chartier, St. Mary's Hospital, London) has already been established in order to screen a mouse-yeast artificial chromosome (YAC) library for the isolation of large chromosome fragments (± 500 kb) encoding TLRp and also the TLR array linked to the IL-1 haemopoietic gene cluster. Once obtained such TLRp clones might be expected to also encode the linked AML-predisposing locus and λ subclones from the relevant YAC clone will then be screened for candidate haemopoietic genes; this latter screening procedure is unlikely to be undertaken within the contract period and would demand further collaboration.

D. TLR polymorphisms in AML-sensitive and -insensitive mouse strains

The existing association between TLRp and AML-predisposition in the CBA/H mouse strain will be extended to the similarly AML-sensitive strains SJL, CBA/Ca (with M. Coppola, ENEA, Roma), RFM (with R.J.M. Fry, Oak Ridge, USA) and also the AML-intensitive strains C57Bl and C3H (with I. Hayata, Chiba Japan). DNA obtained from 50 animals chosen randomly from these colonies will be screened with appropriate TLR probes for the frequency of the TLR polymorphism already recorded in CBA/H. In the case of CBA/Ca and RFM it will also be possible to analyse induced AMLs in order to provide a direct comparison with the TLRp data already available for CBA/H.

E. The induction and analysis of AMLs induced in hybrid mice

Genetically divergent mice such as CBA and Mus spretus show a high level of DNA sequence polymorphism which may be used to distinguish parental genetic contributions in their F1 hybrid offspring. Such DNA heterozygosity in F1 hybrids may be determined by molecular analysis of gene coding or polymorphic DNA repeat sequences. Thus, specific DNA deletions in F1 hybrid neoplasms may be characterised by loss of heterozygosity (LOH) for a set of linked markers; this is a well established technique for the identification of tumour suppressor gene loss in human tumours and will be used here to define ch2 deletions in AMLs induced in hybrid mice. A major advantage for this study is the ready identification of AML-sensitive CBA/H variants within the colony using existing probes for TLR sequence polymorphisms. Accordingly, ♂ and ♀ CBA/H TLRp mice will be identified using DNA purified from tail tips. The crosses currently being considered are : CBA x Muss spretus, CBA x C57Bl, and CBA x C3H; litters will be split equally yielding F1 hybrids for 3 Gy X-irradiation and controls. In order to determine the patterns of inheritance and expression of the TLRp-linked mutation, AML-radiosensitivy will also be determined in F2 hybrids ie. in the offspring of F1 hybrids backcrossed to CBA/H TLRp mice. Similarly, crosses between ♀ and ♂ CBA/H TLRp mice will be performed to derive a true-breeding TLRp sub-strain in order to confirm the anticipated increase in leukaemogenic radiosensitivey associated with TLRp. The yield of AMLs from all parts of the above study will be determined; DNA of primary AMLs and normal host tissue from hybrid animals will be screened with ch2-specific probes in order to determine the extent and specificity of LOH. Cytogenetic analyses of selected AMLs will also be performed. These AMLs will not be available until year 2 and, consequently, molecular analyses will run beyond the contract period.

F. Characterisation of AMLs induced by low dose-rate X-rays and fission neutrons

It is not known whether dose-rate and radiation quality influence the cellular and genetic characteristics of induced AMLs. To address this problem, male mice chosen randomly from the CBA/H colony will be irradiated with either 2.0 Gy X-rays at 3 mGy/min (n=200) and 600 mGY/min (n=90) or 0.4 Gy fission neutrons at 0.5 mGy/min (n=125) and 300 mGy/min (n=125). The expected AML yield will be 18% and 11% for X-rays and fission neutrons respectively. These will be histopathologically characterised and their malignant characteristics determined using in vivo passage procedures. DNA from primary AMLs and normal host tissues will be analysed for TLR sequence polymorphisms to determine whether AML-predisposition is influenced by dose-rate and radiation quality. Representative AMLs from this study will also be cytogenetically characterised. In addition, by keeping the irradiated mice during their whole life span, the proposed experiment will yield valuable information concerning the effect of dose protraction with fission neutrons on the induction of tumours other than myeloid leukaemia.

G. Myeloid differentiation and mutagenesis of CBA/H embryonic stem (ES) cell lines

Multipotential ES cell lines allow myeloid cell differentiation to be studied both in vitro and in vivo and will provide the means of studying the cellular consequences of specific leukaemogenic events. ES cell lines will be established from early CBA/H embryos carrying the different TLR sequence polymorphisms that distinguish AML-sensitive and -insensitive animals. The in vitro patterns of myeloid differentiation in these cells will be studied using growth-factor supplemented medium in order to determine whether the AML-sensitive genotype is associated with altered cellular differentiation. Also, attempts will be made to observe conversion from the pre-leukaemic to the leukaemic state in vitro, with the help of exogenous growth factors. Indeed, Haran-Ghera (personal communication) has been able to accelerate leukaemia development and to increase the yield of myeloid leukaemia (up to 100%) by injecting myeloid growth factors (IL-3, GM--CSF). The post-irradiation response of these different cell lines will also be investigated using cytogenetic and clonogenic techniques. Provided that these cell lines are shown to be able to reconstitute in vivo the myeloid lineage of marrow-ablated CBA/H mice, studies to investigate in vitro mutagenesis and myeloid leukaemogenesis will be initiated. Cells will be mutagenised in vitro with X-rays, retroviral vectors (with F. Pedersen, IMV, Aarhus) or homologous recombination and then transplanted to host animals. Leukaemias arising in these animals will be characterised using histopathological, cytogenetic and molecular techniques. In the case of retroviral insertion mutagenesis and homologous recombination the location of the integrated vector in leukeamia cell DNA provides a means to identifying target genes for leukaemogenic initiation; such a study would extend beyond the contract period.

In conclusion, the initial phase of collaboration centres on the already established use of ch2 and TLR sequence probes for the characterisation of ch2 rearrangement in AMLs and AML-predisposition in variant mice. Studies on AML induction in hybrid mice and the isolation of ES cell lines initiated in this early phase will, however, broaden the experimental approach and are essential for longer-term investigations. It is anticipated that availability of F1/F2 AMLs and indeed other induced murine neoplasms will provide a unique resource for future EC studies on the links between radiation damage, gene-loss events and neoplastic initiation.

NRPB (GB)

Studies at NRPB will focus on the cytogenetic and molecular analysis of the chromosome (ch)2 deletions and rearrangements that are believed to initiate murine AML.

Cytogenetic studies

Cytogenetic studies on CBA AMLs induced at Petten will be undertaken but more emphasis will be placed on the investigation of clonal ch2 events present in vivo during early post-irradiation haemopoietic repopulation. Consideration will be given to inter-animal variation in CBA responses and their possible dependence on telomere-like repeat (TLR) sequence polymorphism.

Molecular studies

Chromosome 2 libraries : PCR techniques will be used to establish a ch2 FISH "painting" library from microdissected chromosomes. As well as facilitating all cytogenetic studies, these libraries will be used as a source of probes for ch2 encoded TLR loci.

Characterisation of specific TLR sequences : TLR sequences will be cloned and used as probes for in situ chromosome hybridization and electrophoretic characterisation of ch2 breakpoints in AMLs. Of particular importance will be the molecular cloning and chromosomal mapping of the TLR loucs associated with AML predisposition in CBA mice. Different cloning and mapping strategies will be explored and TLR probes willl be used in establishing the origin/inheritance patterns for this polymorphic locus in CBA mice. In collaboration with Mol, the status of this locus in other mouse strains will be investigated. In addition, low dose rate X-ray and neutron induced AMLs from the Petten study will be subject to TLR locus analysis.

AMLs induced in cross-bred mice, : Studies on the induction of AML in the F1 progeny of cross bred mice at Petten will cast light on the genetics of AML susceptibility and provide a resource for further molecular studies. In collaboration with Mol, critical ch2 sequence losses in F1 AMLs will be determined using naturally occurring DNA polymorphisms that distinguish ch2 encoded genes in CBA and other mouse strains. The crosses currently being considered are : CBA x Mus spretus. CBA x CB57BL and CBA x C3H; loss of heterozygosity for ch2 loci in F1 AMLs will be assessed using PCR analysis of polymorphism at locus-specific purine-pyrimidine tracts.

VITO (BE)

AMLs induced in cross-bred mice

As outlined above in the contribution of NRPB and in collaboration, critical chromosome 2 sequence losses in F1 AMLs will be determined using naturally occuring DNA polymorphisms that distinguish chromosome 2 encoded genes in CBA and other mouse strains. The crosses currently being considered are : CBA x Mus Spretus, CBA x C57BL and CBA x C3H. Loss of heterozygocity for chromosome 2 loci in F1 AMLs will be assessed using PCR analysis of polymorphism at locus-specific purine-pyrimidine tracts.

Embryonic stem cell lines

ES cell lines will be established from early CBA/H embryos carrying the different TLR sequence polymorphisms that distinguish AML-sensitive and -intensitive animals. The in vitro patterns of myeloid differentiation in these cells will be studied using growth-factor supplemented medium in order to determine whether the AML-sensitive genotype is associated with altered cellular differentiation. Also, attempts will be made to observe conversion from the pre-leukaemic to the leukaemic state in vitro, with the help of exogenous growth factors. The post-irradiation response of these different cell lines will also be investigated using cytogenetic and clonogenic techniques. Provided that these cell lines are shown to be able to reconstitute in vivo the myeloid lineage of marrow-ablated CBA/H mice, studies to investigate in vitro mutagenesis and myeloid leukaemogenesis will be initiated. Cells will be mutagenised in vitro with X-rays, retroviral vectors (with F. Pedersen, IMV, Aarhus) or homologous recombination and transplanted to host animals. Leukaemias arising in these animals will be characterised using histopathological, cytogenetic and molecular techniques. In the case of retroviral insertion mutagenesis and homologous recombination the location of the integrated vector in leukeamia cell DNA provides a means to identifying target genes for leukaemogenic initiation; such a study would extend beyond the contract period.

ECN (NL)

Studies at ECN will focus on the induction of acute myeloid leukemia (AML) in male CBA/H mice or hybrid mice after exposure to X-rays or 1 MeV fission neutrons.

Induction of AML by low dose-rate X-rays and fission neutrons

In order to investigate the influence of dose rate and radiation quality on the cellular and genetic characteristics of induced AML's, inbred CBA/H mice will be exposed to 2.0 Gy X-rays at 3 mGy/min and 600 mGy/min, or 0.4 Gy fission neutrons at 0.5 mGy/min and 300 mGy/min. The induced AML's will be histopathologically characterized and their malignant characteristics determined using in vivo passage procedures. In addition, DNA samples from AML's and normal host tissues will be collected for TLR sequence polymorphism analysis at NRPB and VITO.

To investigate the effect of dose protraction with fission neutrons and X-rays, all animals without AML will be kept during their life span and be analysed for the induction of other types of tumors.

Induction of AML by X-rays and fission neutrons in CBA/H TLRp homozygotes

Male and female CBA/H mice will be identified on the basis of the TLR sequence polymorphism will be mated to obtain a true TLRp substrain. These animals will be irradiated with X-rays and fission neutrons to determine their leukaemogenic radiosensitivty.

Induction of AML by X-rays and fission neutrons in cross-bred mice

Genetically divergent mice such as CBA/H and Mus spretus will be mated and the F1 progeny will be used to investigate AML susceptibility by means of analysis of critical ch2 sequence losses performed by NRPB and VITO. The crosses currently considered are CBA/H x Mus spretus, CBA/H x C57BL and CBA/H x C3H. F1 hybrids will be irradiated with 3 Gy X-rays.

F1 hybrids will also be back-crossed with CBA/H TLRp mice. AML susceptibility will be determined in the resulting F2 hybrids. DNA from AML and normal tissue will sent to NRPB and VITO for screening with ch2 specific probes.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920017 Studies on radiation induced chromosomal aberrations.

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Description of research work

Introduction:

Chromosomal aberration is considered to be one of the important biological effects arising as the consequence of exposure to ionizing radiation in man. In the case of radiation accidents chromosome aberration frequencies in peripheral blood lymphocytes are used to estimate the absorbed radiation dose. Chromosomal aberrations in germ cells and somatic cells are associated with congenital malformations in newborns and neoplasms. Thus, the understanding of the mechanisms of induction of DNA lesions and chromosomal aberrations caused by ionizing radiation is very important not only at high doses but also at low doses and dose rates. This project (comprising of 4 European laboratories from 4 countries) along with that coordinated by Prof. Olivieri, Rome aims at elucidating some of these important issues.

Radiation induced DNA lesions and their repair relevant to chromosome aberrations:

Among the lesions induced by ionizing radiation, DNA double-strand breaks (DSB) appears to be the most important in the production of aberrations. From the earlier studies (supported by the CEC Radiation Protection Programme) several lines of evidence supporting this concept have emerged. Secondly, dicentric aberrations in human lymphocytes have been shown to be formed immediately after X-irradiation, within the time period in which most of the DSBs are repaired. To what extent this is valid for high LET radiation has to be evaluated. The technique of premature chromosome condensation, by which interphase chromosomes can be easily visualized, has proved to be very useful in these studies.

Radiosensitive mutants have been helpful in identifying the lesions leading to chromosome aberrations. In xrs mutant (CHO) cell lines with differing capacities to repair DNA DSBs, a correlation could be found between the extent of defect and the frequencies of aberrations. With the possibilities of introducing large molecules (such as repair proteins) into cells, the defect in these mutant cells can be identified and corrected. X-ray sensitive mutant cells which are competent in repairing DSBs but respond with increased frequency of radiation induced chromosome aberrations, can be employed to identify DNA lesions other than DSBs which can give rise to aberrations.

Chromosomal aberration is one of the predominant biological indicators of exposure to ionizing radiation and has been used in both basic studies (developing biological models for radiation action - target theory), as well as applied studies (eg., biological dosimetry in case of radiation accidents). Radiation induced DNA lesions responsible for the induction of aberrations have been identified as DSBs and the formation kinetics of aberrations following irradiation have been determined. More information is needed to clarify some of the problems outlined above, especially pertaining to chromosomal repair, low doses and dose rates, high LET radiation and irradiation under in vivo conditions. Most of the techniques to be employed are classical and well established ones. However, there are several new techniques which will be employed in the proposed studies as oulined below.

The project will employ conventional chromosome techniques, namely airdried preparations followed by Giemsa staining for analysis of metaphase chromosomes. However, a newly developed "chromosome painting" technique using chromosome specific DNA libraries and in situ hybridization will be used to detect chromosomal translocations. This technique has already proved to be very sensitive and useful in basic and applied (accidents) studies. In experiments involving kinetics of chromosome aberration formation as well as repair at the chromosomal level, the technique of "premature chromosome condensation" will be employed. For studying DNA repair in conjunction with chromosomal aberration formation, the technique of permeabilizing cells by electroporation or Streptolicin O-poration and introducing cellular extract or specific repair enzymes will be used. In addition, conventional molecular biological techniques to estimate induced strand breaks and their repair, such as alkaline-unwinding, alkaline elution, neutral elution, etc., will be employed.

In this project several aspects of basic mechanisms leading to radiation induced chromosomal aberrations will be investigated. It has been shown earlier that exchange aberrations following X-irradiation of human lymphocytes are formed immediately, whereas the events leading to acentric fragments are repaired slowly (Leiden). Chromosomal repair kinetics will be investigated following irradiation with high LET radiation (neutrons) using the technique of PCC as well as metaphase analysis (Leiden). UV irradiation or hyperthermia can change the chromatin structure and can influence the number and interaction of X-ray induced lesions giving rise to increased frequencies of chromosome exchanges. This problem will be studied using PCC techniques as well as metaphase analysis (Athens, Leiden).

The frequencies of radiation induced chromosome aberrations are mediated through cellular repair processes. Cells defective in DNA DSB repair (xrs mutants of CHO) respond, in comparison to wild type cells, with higher frequencies of aberrations for a given dose. Correction for the increased chromosomal radiation sensitivity of xrs cells will be attempted by introducing wild type nuclear extracts and their fractions into mutant cells by Streptolicin O-poration, in order to identify the proteins involved in chromosomal repair (St. Andrews). A similar approach will be made in radiosensitive mutants which are competent in DSB repair, but still respond with higher frequencies of radiation induced aberrations, by introducing specific enzymes, such as glycosylases following x-irradiation (Leiden).

Stable aberrations such as translocations are very important from genetic and cancer risk point of view as they can perpetuate in the body. Using chromosome painting technique, it has been shown that X-irradiation induces far more chromosomal translocatios than dicentrics (Leiden). The frequencies of x-ray induced translocations will be assessed using probes from different human chromosomes to get an accurate estimate of induction. In addition, the influence of dose fractionation and dose rate, the presence of repair inhibitors on induction of translocations will be studied. The relative induction of dicentrics and translocations will be established for high LET radiation (Leiden). Telomeric probes will be used to discern the nature of acentric fragments (St. Andrews/Leiden).

Inter-individual variability for radiation response is a very important issue especially in the area of radiotherapy. This problem will be investigated by G_2 irradiation of lymphocytes followed by PCC analysis (Athens).

The phenomenon of adaptive response, in which an initial low adaptive dose renders the lymphocytes resistant to challenging higher radiation dose will be further investigated. The question whether in vivo low dose irradiation leads to adaptation for a higher dose in vitro will be addressed using blood samples from uranium miners (Sacavem).

A: Studies on radiation-induced chromosomal aberrations in mammalian cells: Basic mechanisms (Leiden).

1. **Kinetics of formation of exchange type of aberrations:**
   Human lymphocytes will be irradiated with 0.5 and 1.0 Gy of 1 meV neutrons and fused to mitotic CHO cells at different times of recovery to check the time of formation of exchanges and the kinetics of decay of fragments. These results will be compared with our earlier results with X-rays (Vyas et al., 1991).
2. **Frequencies of radiation induced chromosomal translocations:**
   We have shown that translocations (reciprocal, interstitial, terminal) are formed with much higher frequencies than dicentrics following X-irradiation of human lymphocytes, using the chromosome painting technique (Natarajan et al., 1992). This will be extended to low doses (0.1 to 0.5 Gy) to check the feasibility of using radiation induced translocation frequencies as biological dosimeter. The dose response curve for induction of translocations will be established for high LET radiation as well as low LET radiation given as protracted or fractionated doses. The occurrence of symmetrical and asymmetrical exchanges between chromosomes following irradiation of S and G2 lymphocytes will be studied to compare the pattern with that obtained after G0 irradiation.
3. **Studies using X-ray sensitive mutants:**
   X-ray sensitive CHO mutants, such as VC-4 and VG 8 which are competent in DNA DSB repair but respond with high frequencies of chromosomal aberrations will be used to identify critical lesions other than DSBs which can lead to aberrations. These mutant cells will be electroporated or Streptolicin O-porated after X-irradiation to introduce specific glycosylases such as FPG^+ or endonuclease 3 which specifically recognize 8 hydroxy guanine residues or thymine glycols respectively.

B. Utilization of repair deficient cells (St. Andrews).

The project is aimed at studying the chromosomal responses of normal and radiosensitive mutant mammalian cells to radiation, to gain a deeper understanding of the clastogenic effects of low doses of radiation. Previous work shows that chromosomal aberrations are induced by DNA double-strand breaks (dsb) especially those with blunt-ended termini, and the induction of aberrations depends on genetic factors determining both DNA repair and the 'conversion' of dsb into visible aberrations. The proposed work will be a continuation of work and will focus on investigation of possible reasons for the chromosomal radiosensitivity of mutant cell types such as Chinese hamster xrs 5 and human ataxia telangiectasia (AT). This work has already shown: a) a difference in the electrophoretic (SDS-PAGE) profile of proteins in nuclear extracts between xrs 5 and CHO cells; b) altered DNA binding properties of mutant versus WT extracts; and c) a reduction in bleomycin induced chromosome damage in xrs 5 cells treated with WT extract.

In the proposed work, nuclear extracts prepared from wild-type CHO and normal human (lymphoblastoid) cells, prepared by homogenization in non-ionic detergent under specific salt conditions, will be used to treat gamma-irradiated/streptolycin-O porated radiosensitive xrs 5 and AT cells, in order to complement the gene defects in these lines which, in the case of xrs 5 leads to a reduced capacity for dsb repair and in both xrs 5 and AT to high radiation-induced frequencies of chromosomal aberrations. Dsb repair in xrs 5 will be measured by neutral elution and chromosomal aberrations and micronuclei scored in metaphase preparations and cytokinesis blocked (rodent and human) cells respectively. The electrophoretic (SDS-PAGE) patterns of proteins in nuclear extracts, the DNA binding, and other biochemical properties of proteins will be studied using various techniques including South-Western blotting.

The molecular mechanisms of chromosome break formation will be furhter investigated in hamster and human cells using in situ hybridisation of telomeric probes to chromosomes to determine whether dsb termini are irreversibly closed by telomeric sequencies during post-irradiation incubation, leading to visible breaks.

C. Studies of the adaptive response for radiation-induced chromosomal aberrations on high and low exposed individuals working in uranium mines (Sacavem).

An initial small radiation dose (adaptive dose) given to human lymphocytes in G1 reduces the frequency of aberrations induced by a subsequent high dose (challenging dose) in G2, accordingly with in vitro studies of Olivieri and Wolff (1984). Our purpose is to find out if the in vivo dose received by miners acts as an adaptive dose when we score chromatid aberrations after an in vitro exposure to a high dose (1 Gy) of their lymphocytes.

For these studies we selected three groups, one control group with 10 individuals and two studying groups with 20 individuals each. One of these groups will consist of highly exposed individuals, and the other will consist of lowly exposed individuals. Lymphocytes from those individuals will be cultured for 50 hours before receiving a challenge dose of 0.5 Gy of gamma rays. Chromatid aberrations induced in G2 will be scored and the aberration frequencies compared for each individual, with or without the challenge dose, and between groups of high and low level exposed individuals.

D. Utilization of Premature Chromosome Condensation technique (Athens).

1. Elucidation of the molecular mechanisms underlying heat radiosensitization:

Ionizing-radiation-induced cell death is thought to be the result of a sequence of causally related events in the various levels of cellular organization involving energy absorption in the nucleus, damage induction into DNA and, finally, conversion of DNA damage, most likely of DNA double strand breaks (dsb), to lethal chromosome aberrations. Heat-induced radiosensitization can be understood, therefore, as a modulation of one or more of the steps involved in this sequence of events. This modulation may lead to increased production of lethal lesions per cell per Gy, manifested at the cytogenetic level as an increase in the production of chromosome aberrations. The working hypothesis is that heat sensitizes cells to radiation by increasing the conversion probability of DNA dsb into

lethal chromosome aberrations. The enhancement of the conversion probability of DNA dsb into chromsome aberrations is assumed to be caused either by alterations in chromatin structure and function and/or by a reduction in the efficacy of repair. Alterations in chromatin conformation induced by pre-irradiation exposure to heat at 43-45.5°C may lead to structural instability of irradiated DNA molecules and to the enhancement of misrepair processes resulting into exchange type aberrations. Reduction in the efficacy of the repair may explain the radiosensitization by post-irradiation exposure to heat at 41-43°C.

Using conventional cytogenetics and the premature chromosome condensation (PCC) technique, the conversions of dsb in chromosome damage will be analyzed in repair proficient and deficient cells under various experimental combinations of heat and radiation. The results obtained will provide useful information in the development of effective combined treatment protocols in radiation therapy. Such protocols will require lower radiation doses, resulting in an improved radiation protection of the involved patients and radiation personnel.

2. **Elucidation of the mechanisms underlying the enhanced G2 chromatid radiosensitivity of human tumor cells and cells from cancer prone individuals:**

Experiments complementary to those carried out to measure directly induction and repair of chromatid damage in normal and tumor cells in an attempt to elucidate the mechanisms underlying the phenomenon of G2 radiosensitivity fluctuations. In particular, induction and repair processes of chromosomal damage will be studied directly in G2 phase by means of the PCC technique, without the requirement of cells to proceed to mitosis. It will be investigated whether the enhanced G2 radiosensitivity is only expressed by cells that had passed the G2 block transition point at the time of irradiation, and whether chromatid conformation changes are involved in this phenomenon. This research is of significance to the understanding of the mechanisms that predispose cells to become cancerous, and could contribute to the development of an assay to detect genetic predisposition to cancer.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920028 Radiation-induced processes in mammalian cells: principles of response modification and involvement in carcinogenesis.

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Description of research work

Ionizing radiation and UV-light induce a variety of reactions collectively called "stress responses". These can be divided into the early responses which occur shortly after induction of damage, and the late responses which occur many hours or even days after induction of damage and culminate in mutagenesis and carcinogenesis.

The objective of this research proposal is to gain an understanding of the molecular events that control these radiation-induced processes. To this end 8 European laboratories collaborate, which together cover the major research areas in the field. The significance of this joint project is emphasized by the fact that the radiation-induced stress responses appear to represent the molecular basis of both initiation and promotion of oncogenic transformation and possibly also of tumour progression.

A brief outline of the projects is presented below.

The research contributions of the 8 laboratories can be grouped into 3 themes:

A. Initial responses to radiation.
B. Biological consequences of the initial responses.
C. Genetic consequences of radiation.

A. INITIAL RESPONSES TO RADIATION

Early genetic responses to radiation in mammalian cells

Ionizing and ultraviolet radiation induce a wide variety of responses, not only in E.coli but also in mammalian cells. The early responses in mammalian cells include activation of expression of genes coding for transcription factors, growth factors, proteases and other proteins. Evidence has been presented by the lab of **Herrlich** that the inducing signal for UV-light is the non-repaired DNA damage and that the signal chain ends with the activation of pre-existing transcription factors. The signalling chain makes use of cytoplasmic protein kinases (raf-1, MAP-2 kinase) as well as presumably the secretion of growth factors. These experiments will now be extended to X-ray-induced gene expression and studies on the long-term consequences of X-ray-induced synthesis of specific proteins. An important question is how the energy of X-irradiation is converted into a biochemical signal, how X-ray-inducible genes differ from non-inducible or UV-inducible genes and how the signal reaches the responding gene. Another question is whether X-ray-induced gene products play a role in mutagenesis and carcinogenesis.

Role of RecA-like genes in mammalian cells

In E.coli, DNA damage induces stress responses, called SOS, which are controlled by the RecA gene. **Devoret's** group has started a search for RecA-like genes in mammals based on the idea that DNA repair is a universal function that involves cognate proteins. The group has just isolated a mouse gene, Kin17, located on chromosome 2, band A. The encoded kin17 protein, (1) shares a domain with RecA protein, (2) has

an active zinc-finger which results in binding of the protein to DNA, and (3) has a bipartite nuclear localization signal that drives the protein to the mouse cell nucleus. The Kin17 gene has been located on chromosome 10 of the human genome and is being characterized. Kin-17 mRNA seems to be a new molecular marker of neural stress: it is induced by X-rays in the mouse cerebellum and by neuronal treatments that provoke epilepsy. The aim of Devoret's group is to characterize the response of human cells to radiation damage by monitoring the induction of kin17 protein.

Poly(ADP-ribose)polymerase and genetic instability

Another aspect of the UV-induced stress-response concerns the role of poly(ADP-ribose) polymerase (ADPRP), an enzyme involved in the repair of DNA strand breaks and which is studied by **Bertazzoni's** group. The expression of the ADPRP gene and activity of the enzyme will be investigated in response to treatment with DNA-damaging agents in normal and excision-deficient cells. Furthermore, the effect of ADP-ribosylation of nuclear proteins on expression of amplified oncogenes will be determined, by treatment with ADPRP-inhibitors. The group also observed that fibroblasts from XP-patients exhibit spontaneous chromosomal instability. The chromosomal regions in XP cells exhibiting rearrangements will be localized and these regions will be compared with known sites of proto-oncogenes or tumour-suppressor genes, and sites of frequent chromosome breakage, and rearrangements of skin tumours. Similar studies will be performed with cells from UV-sensitive Trichothiodystrophy (TTD) patients who are non-cancer-prone, in contrast to XP patients. Part of the experiments (exchange of patient cell lines, characterization of patients) will be carried out in collaboration with Sarasin's laboratory.

B. BIOLOGICAL CONSEQUENCES OF THE INITIAL RESPONSE

Enhanced reactivation and mutagenesis

XP patients as a rule show a marked predisposition for the development of cancer in sun-exposed skin areas. However, some XP patients have been described who, surprisingly, do not show this property. **Van der Eb's** laboratory has recently found that fibroblasts from these non-cancer-prone XP patients show another abnormality in their response to UV: the cells fail to express the stress-response Enhanced Reactivation (ER), although Enhanced Mutagenesis (EM) is unchanged. Apparently, ER is co-regulated with a process that contributes to the activation of (particular) oncogenes. Further work showed that fibroblasts from patients with other cancer-prone genetic diseases show superinduction of ER, but again normal EM. Future work of the group will concentrate on the identification of UV-inducible processes that are co-induced with ER and could explain the resistance to cancer induction or cancer-proneness.

Oncolytic parvoviruses as a probe to study radiation-modulated gene expression

Parvoviruses can selectively replicate in X-ray-induced tumour cells but not in normal cells. The viral genome will be used by **Rommelaere's** group as a probe for selecting proteins that bind specifically to viral DNA (at a viral promoter region) and which

occur at increased levels in tumour cells. The group will investigate whether the tumour cell-associated changes are due to increased protein levels or to post-translational modification of the proteins, and whether the proteins have transcription-factor activity. The interesting question whether the increase in binding activity occurs already as a consequence of irradiation, i.e. prior to cell transformation, will also be studied. Since parvoviruses are oncolytic in vivo, the group will also make transgenic mice expressing the viral oncolytic protein(s). It will be tested whether these mice are resistant against tumour induction by X-rays.

CD2 expression as a probe for radiation damage

The CD2 protein is a T-lymphocyte-specific membrane antigen that plays a role in the immune response. Expression of the protein is increased following low doses of X-irradiation, which indicates that expression of the CD2 gene may be a sensitive indicator for the effects of radiation. The group of **Thomou-Politi** will try to develop an assay based on CD2 expression to measure low doses of X-ray damage. Furthermore, the secondary effect of radiation damage, e.g. induction of point mutations or small deletions and/or rearrangements of the CD2 gene in X-irradiated cells will be investigated. Use will be made of T-lymphocyte cultures, which express the endogenous CD2 gene, and CHO cell lines expressing a transfected CD2 gene. Sofar, the results indicate that even very low doses of X-irradiation strongly enhance the appearance of CD2 antigen, both in PHA-stimulated and in resting T-lymphocytes. The effect can be demonstrated by a rosette assay or by immunofluorescence. A surprising observation is that CHO cells transfected with a CD2 expression plasmid lose their capacity to express CD2 following X-irradiation. The results so far suggest that this effect can be attributed mainly to point mutations or very small deletions of the CD2 gene in irradiated transfected CHO cells.

C. GENETIC CONSEQUENCES OF RADIATION

DNA amplification, point mutations and other DNA rearrangements

Of the longterm effects of radiation damage, induction of DNA amplification and point mutations may be the most important ones. **Sarasin's** group will focus on the mechanism of induction of these phenomena. Gene amplification will be measured with the use of plasmid constructs containing both the Epstein-Barr virus (EBV) and the SV40 origins of DNA replication. The plasmids can amplify with either origin in response to UV-irradiation. This work will be extended further with a detailed analysis of the accuracy of the amplification process, by studying individual rescued plasmids with the use of restriction-enzyme analysis and more precisely by screening mutations in the supF tRNA gene used as a target. Further work will focus on oncogene amplifications and mutations in skin tumours from Xeroderma pigmentosum (XP) patients. Since these tumours preferentially arise in sunlight-exposed skin regions, involvement of UV damage as the triggering signal is likely. Sofar, it was found that amplification and mutation of ras genes occur more frequently in XP-tumours than in other skin tumours. A similar study will be carried out with the p53 tumour-suppressor gene. The tumour-suppressor function of p53 can be inactivated by point mutations, and analysis of p53 in skin tumours from XP-patients may provide further insight into the mechanism of tumour induction in these patients.

X-ray-induced tumourigenesis

It is generally accepted that cancer induction requires mutational changes in at least 2 cancer-associated genes. This requirement complicates experimental studies of carcinogenesis by DNA-damaging agents, since very high doses of an agent are required to obtain mutational alteration of 2 or more (onco)genes in the same cell. To circumvent this problem, transgenic mice expressing an activated oncogene (ras or myc) or containing an inactivated allele of a tumour-suppressor gene (p53 or Rb), will be made. The mice will then be irradiated with X-rays (which induces, amongst others, DNA rearrangements) or with agents causing point mutations. These two treatments may activate different sets of oncogenes. The tumours that appear in the mice will be characterized with respect to the oncogenes (or suppressor genes) that are mutated by the treatment.

The group of **Simons** will study mice in which the oncogene is expressed from a keratinocyte-specific promoter, allowing expression in epithelial cells, whereas the group of Van der Eb will use various promoters including the immunoglobulin promoter which restricts expression to B- and T-lymphocytes. The group of Simons also proposes to investigate X-ray-induced transformation of mouse keratinocytes *in vitro:* in this study normal and X-ray-sensitive cells will be compared.

Distribution of tasks and complementarity between groups

The participating groups carry out distinct and complementary tasks in the framework of the collaboration. The various levels of damage-induced "response modification" are covered as follows:

A. Initial responses to radiation

- X-ray-induced early responses in mammalian cells (Herrlich)
- role of RecA-like genes in the induction of stress response (Devoret)
- the role of poly(ADP-ribose) polymerase in the generation of the stress response (Bertazzoni)

B. Biological consequences of the initial response

- enhanced reactivation and enhanced mutagenesis (Van der Eb)
- replication of oncolytic parvoviruses (Rommelaere)
- CD2 expression as a manifestation of the stress response (Thomou-Politi)

C. Genetic consequences of radiation

- DNA amplification, mutation, DNA rearrangements (Sarasin, Simons, Bertazzoni), and transformation (Rommelaere, Simons)
- X-ray-induced tumorigenesis (Simons, van der Eb, Rommelaere).
- cell migration as manifestation of stress response (Simons)

Participant No: 1 Laboratory of Molecular Carcinogenesis, Sylvius Laboratory, Leiden, The Netherlands

MECHANISMS OF RADIATION-INDUCED CARCINOGENESIS

Cancer induction requires mutational activation of 2 or more oncogenes and probably inactivation of 2 alleles of a tumour suppressor gene. The purpose of this project is to gain an understanding of the molecular mechanism of radiation-induced carcinogenesis, of which little information is available.

1. Radiation-induced processes in human cells

Exposure of human cells to UV-light results in induction of the stress responses Enhanced Reactivation (ER) and Enhanced Mutagenesis (EM). We have found that fibroblasts from XP-patients, which for unknown reasons do not develop skin cancer on sun-exposed skin areas, have normal EM, but lack ER. Interestingly, cells from cancer-prone genetic diseases show super-induction of ER, but normal EM. This suggests that ER is co-regulated with events that contribute to cancer. The purpose of this project is to identify the processes that are co-regulated with ER and that could explain the above-mentioned phenomena. The processes that will be tested include: 1) expression of UV-inducible genes (myc, MHC, ODC NFkb). Sofar we found that UV-induced expression of c-jun, c-fos, collagenase and Kin17 mRNAs shows no difference between ER^- and ER^+ cells (collaboration with P. Herrlich and R. Devoret), 2) UV-mediated stabilization of p53 (a tumour suppressor protein), in various cancer-prone syndromes, 3) induction of the oxidative stress response, 4) recombination (between introduced plasmids with non-overlapping deletions in the same marker gene, or sister chromatid exchange), gene amplification (collaboration with A. Sarasin) and production of UV-induced secreted growth factors ("EPIF", collaboration with P. Herrlich). This will hopefully lead to identification of mechanisms other than induction of point mutations, that can lead to oncogene activation.

2. X-ray and UV-induced lymphomagenesis.

To investigate the contribution of ionizing radiation in carcinogenesis, transgenic mice will be used expressing an activated H-ras or c-myc oncogene or an inactivated allele of either the p53, the FAP or the Rb tumour suppressor gene. The oncogenes will be regulated by various promoters, e.g. the immunoglobulin enhancer-promoter ($E\mu$), or the H2-K, K10 or K14 (keratin) promoter. In order to get neoplastic transformation in these mice, at least a second, probably more genetic events in the same cell are required. The mice will be irradiated with increasing doses of (fractionated) X-irradiation, and will be monitored for the development of lymphomas (B- or T-cell lymphomas) or other tumours. The lymphomas will be isolated and their clonality determined (rearrangement of immunoglobulin heavy chain genes or T-cell receptor genes). The nature of the oncogene(s) activated by X-irradiation will be identified if possible, using the extensive knowledge on activated oncogenes in mouse B- and T-lymphomas (collaboration with A. Berns). Concomitantly, bone marrow or peripheral blood will be isolated from the transgenic mice and irradiated with UV-light. The UV-irradiated blood will be transplanted into syngeneic recipients and leukemia induction will be monitored. These experiments may reveal whether there is any preference for cancer induction by either X-rays or UV in ras or myc-transgenic mice.

Participant No: 2 Laboratory of Molecular Genetics IRCS Villejuif (France)

BIOLOGICAL CONSEQUENCES OF IRRADIATION IN NORMAL HUMAN CELLS AND IN CELLS OF CANCER-PRONE PATIENTS.

1. Analysis of mutagenesis during the gene amplification process.

We have construced hybrid plasmids containing both the EBV and the SV40 replication origins. These molecules are able to replicate episomally either like an EBV vector or on the SV40 mode if the SV40 large T antigen is provided at the same time. We have used this system in order to determine if such vectors can be amplified *in vitro* as a response to stress. These vectors carry a target gene for mutagenesis studies (*lacZ'*) which should allow us to determine the fidelity of the amplification process in mammalian cells, this amplification being a characteristic of some tumour cells. UV irradiation of both human adenovirus-transformed 293 or SV40-transformed MRC5 cells leads to vector amplification, irrespective of the type of replication origin used for the episomal maintenace.

Our results clearly show that the EBV latent replication origin (OriP) is sensitive to overreplication in UV-irradiated human cells. Since the UV doses were sufficiently low to induce very little damage, if any, on the plasmid sequences, this amplification should be mediated through a cellular factor acting *in trans*. The interest in using shuttle vectors for this kind of study lies in the easy analysis of the amplified vectors in rescued bacterial colonies. The accuracy of the amplification process is monitored by studying restriction maps of individual plasmid molecules or the integrity of a target gene (such *lacZ'*).

2. Oncogene amplification and anti-oncogene modification

Xeroderma pigmentosum (XP) is one of the best examples illustrating the relationship between unrepaired DNA lesions, mutagenesis and carcinogenesis in man. This rare, autosomal recessive hereditary disorder is deficient in excision repair of DNA lesions induced by UV irradiation. This results in a high incidence of cancers of the skin, particularly in sun-exposed parts of the body. Southern analysis of genomic DNA from XP tumours has shown high levels of Ha-*ras* gene amplification and rearrangements not found in skin tumours from normal individuals. Screening for *ras* mutations in skin tumours from XP patients by PCR followed by differential hybridization, result in detection of *ras* mutations in 55% of XP tumours.

Since some tumour suppressor genes such as the p53 gene could be inactivated by point mutations, we started to determine if the p53 gene is modified in UV-induced skin tumours from XP patients. These experiments are carried out using reverse PCR on mRNA isolated from tumours followed by direct DNA sequencing. Around 50% of skin tumours exhibited at least one mutation in the p53 gene.

Participant No: 3 Laboratory Enzymology, CNRS, F-91198 Gif-sur-Yvette, France

MAMMALIAN PROTEINS INVOLVED IN SOS REPAIR

The RecA protein controls the expression of all SOS proteins in bacteria. SOS proteins govern DNA repair and mutagenesis. We have reported in Nucleic Acids Research that we have identified a RecA-like protein in mouse and human cells.

Discovery of the KIN17 gene. We isolated the full-length cDNA of a mouse gene, denoted KIN17, which expresses a polypeptide reactive with anti-recA antibodies. The location of the KIN17 gene is on mouse chromosome 2, band A. We also located the human KIN17 gene on the short arm of chromosome 10. The gene is well conserved among mammals. We observed a high level of KIN17 mRNA in some neural tumour cells.

Properties of the kin17 protein. KIN17 cDNA codes for a basic protein of 43 kDa made consisting of 391 amino acids. Kin17 protein, produced in E. coli, was purified. Kin17 protein binds to double-stranded DNA. It shares with E. coli RecA protein a module of 39 amino acids that is a putative DNA-binding domain. We have proven that kin17 has a Zn- finger responsible for DNA binding. We also found that kin17 protein possesses a bipartite nuclear localization signal that drives kin17 protein into the cell nucleus.

Further characterization of KIN17 induction by radiation. With Simmons' group (Leiden), we have observed an accumulation of KIN17 mRNA after cell treatment with psoralene and UV-light. X-rays also induce KIN17 in the mouse cerebellum in vivo (with J-F Riou, Rhone-Poulenc). We plan to determine the lowest dose of X-rays that induces kin17 protein. We are also setting up a system of detection of kin17 protein on human lymphocytes (G. Frelat, CEA FAR).

Biochemical determination of kin17 domains. The respective roles of the zinc-finger and the recA-like domain for DNA binding will be determined by generating targeted mutations in the two domains. We want to elucidate at the biochemical level the role of mouse kin17 in repair.

Cloning of the human KIN17 gene. We will clone the mouse and the human KIN17 gene and we will determine their structure: regulatory regions, exons, introns, and their localization. The position of mouse KIN17 relative to RAG1 and RAG2 will be determined.

Role of kin17 protein in tumour cells. Our results have led to a plan to study the role of the KIN17 gene in cell lines derived from neuroblastomas.

Characterization of other KIN genes. We have identified another mouse cDNA, called KIN2, which has an extended homology with the N-terminal region of the recA gene. We plan to identify the protein and its role in repair of DNA damage.

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MODULATION OF GENE EXPRESSION IN IRRADIATED AND RADIATION TRANSFORMED HUMAN CELLS

General Aim. Neoplasic transformation can, at least in part, be traced back to alterations of the pattern of cellular gene expression. Accordingly, the present study aims at determining ionizing radiation-jnduced perturbations of nuclear DNA-binding proteins, that may account for the disregulation of gene expression. This leads us to address the related question of the transforming risk of fractionated X-irradiation, and the prevention of radiation-induced carcinogenesis.

Strategy and state of the Art. Human fibroblast and keratinocytes will be analyzed either shortly after X-or γ-irradiation or upon radioinduced neoplasic transformation. In particular, advantage will be taken of the system of *in vitro* stepwise transformation of normal human keratinocytes by ionizing radiation, using resistance to terminal differentiation as a selection criterion, which was developed in our laboratory. In order to identify radiation-modulated DNA-binding proteins, a parvovirus-based DNA probe will be used whose expression has been proved to be transformation-sensitive. Thus, we have demonstrated that transformation of human fibroblasts by ionizing radiation correlates with an increase in their susceptibility to these parvoviruses. The validity of this approach is supported by our recent work showing that : (i) a number of cellular (nuclear) proteins specifically bind to parvoviral DNA ; (ii) some of the latter proteins appeared to be up-modulated, while at least one was down-regulated, in γ-ray-transformed (versus normal) human fibroblasts.

Experimental Approach. Parvoviral probes so far revealed differences in DNA-binding activities for normal human fibroblasts versus γ-ray-induced full transformants. Nucleolin, a prominent nucleolar protein was shown to bind parvovirus DNA and may be processed differently in normal and transformed cells. We are in the process of comparing changes occurring either shortly after cell irradiation or at intermediate stages of neoplastic transformation. This approach will be extended to other systems of stepwise transformation of mammalian cells by radiation (contr.8).

Among the X-ray induced kerotinocyte clones, a single immortalized but non-malignant transformed strain was obtained. Tumourigenic derivatives of the latter strain will be selected upon serial propagation, with or without further repeated irradiation.

The enhanced susceptibility of γ-ray-transformed cells to parvovirus attack, prompts us to investigate whether mice harboring a parvoviral transgene that encodes the proteins responsible for cell killing, are protected against the induction of tumours by ionizing radiation. This approach complements a project from another participant (nr.1), which intends to test whether mice carrying a transforming transgene are, on the contrary, sensitized to radioinduced oncogenesis. It will be of interest to determine whether the antioncogenic capacity of parvoviruses antagonizes oncogenes with regard to the proneness of transgenic animals to radiation carcinogenesis.

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STUDY OF GENETIC INSTABILITY AND ADP-RIBOSYLATION IN MAMMALIAN CELLS EXPOSED TO MUTAGENS.

The enzyme poly(ADP-ribose)polymerase (ADPRP) is stimulated by a variety of DNA-damaging agents, particularly monofunctional alkylating agents, and is required for efficient DNA strand-break repair.

Our study will be pursued along the following lines:

a) Analysis of the regulation of the expression of the ADPRP gene in resting and proliferating mammalian cells. The level and stability of mRNA for the enzyme in control and mutagenized cells will be measured by a quantitative PCR method recently developed in our laboratory.

b) Study of the interaction between ADP-ribosylation of nuclear proteins and oncogene amplification. Human malignant cells (SW 613-S), characterized by a high copy number of c-myc, will be treated with ADRPR inhibitors and the effect on the expression of the oncogene will be evaluated.

c) Analysis of ADP-ribosylated proteins in response to treatment of human cells with inhibitors of DNA topoisomerase II.

The relationship between DNA damage, immediate cellular responses and long-lasting consequences will be analysed in mutagen-sensitive cells derived from patients affected by Xeroderma pigmentosum (XP) or isolated *in vitro* from the Chinese hamster cell line CHO-K1.

The effect of UV light on chromosomal instability demonstrated by us in fibroblasts from unaffected skin of homozygous and heterozygous carriers of XP mutations will be investigated. The chromosome bands involved in clonal rearrangements will be identified and compared with the localization of oncogenes, fragile sites and the breakpoints involved in chromosome rearrangements in cutaneous tumours. The capability to grow in agar will be tested for fibroblast strains carrying clonal chromosome rearrangements and cytogenetic analysis will be performed with anchorage-independent fibroblasts in order to evaluate the possible selective advantage of chromosomally abnormal cells.

The analysis of chromosomal stability, mutability and cellular transformation will clarify whether alterations in these parameters are related to DNA repair defects and tumor proneness. The effect of mutagens on cellular parameters of genetic stability and on the activity of DNA polymerases, DNA ligase, topoisomerases and ADPRP will be investigated in excision-repair-defective CHO mutants, including two that are defective in the same genes altered in two groups of XP.

Participant No: 6 National Center for Scientific Research "Demokritos", Institute of Biology, 153 10 AG. Aghia Paraskevi Attikis, Athens, Greece

CONSTRUCTION AND USE OF EUKARYOTIC CELL LINES FOR THE ASSESSMENT OF RADIATION-INDUCED ALTERATIONS LEADING TO NEW PHENOTYPES

The objective of this study is to obtain an assay for the function of isolated genes in vitro. Such an assay along with current techniques for gene cloning and somatic cell genetics could uncover additional correlations between the expression and regulation of genes that respond to low levels of radiation.

The CD2 gene codes for a cell surface T-cell specific antigen that appears early in thymic ontogeny and is involved in cell-cell adhesion, signal transduction, T cell activation, differentiation and immune response.

Thioguanine-resistant CHO cells ($HPRT^-$) were stably co-transfected with IIH3-CD2 and pSV2-gpt vectors using the calcium phosphate coprecipitation technique. The resulting co-transfected CHO clones are HAT-resistant and constitutively express the cell surface CD2 antigen, as was verified by using the rosette assay and flow cytometric analysis. Moreover, the results of Southern blotting of DNA from cotransfected clones that was probed with digoxigenin-labelled CD2-cDNA suggest that the IIH3-CD2 plasmid is unable to replicatre as an episome and is integrated in the CHO genome.

The transfected cells were further tested by titrating their response to low doses of radiation affecting the quantitative expression of the CD2 gene. The use of these transfectants as biological markers as response to low doses of X-irradiation and their ability to express the CD2 antigen on their surface in relation to dose level has been studied. It was found that very low doses of X-rays did nog affect the level of CD2 antigen. The radioresistance of $CD2^+$ CHO cells in the low dose range 2-6 cGy) suggested that radiation does not affect the CD2-cDNA and/or the gene(s) regulating its expression. However, the irradiated transfectants show a dramatic decrease by 50% of the CD2 antigen at 10 cGy (rads). Higher doses do not further affect the percentage of rosette forming cells. These results strongly suggest that the CD2 antigen is a very sensitive radiation marker. Support for this conclusion comes also from our preliminary findings that very low doses of X-irradiation strongly enhance the appearance of CD2 Ag in PHA- activated and resting T-lymphoccytes. The influence of radiation on CD2 expression may be used as a sensitive indicator of very low doses in conjunction with problems associated with the immune response of human T lymphocytes. Southern blot analysis of genomic DNA from X-irradiated transfectants in a range from 0-200 cGy that were probed with digoxigenin-labeled CD2-cDNA, have shown no extra or missing bands and no amplification at any of the doses used, which indicates that large deletions or rearrangements have not occurred. All analysed DNAs revealed the same pattern as unirradiated control DNA.

It must be emphasized that we did not examine the RNA level after irradiation and it is possible that in our study, a point mutation or very small deletion could have occurred, altering the gene products but remaining undetectable on Southern blots.

In vitro mutagenesis studies of CD2 cDNA by others have shown that for several mutants of CD2, T-cell rosetting is abolished by a single amino acid substitution. This fact supports the notion that a point mutation or very small deletion may have occurred. In conclusion, the radiosensitivity of the $CD2^+$ clone in a dose range from 8 to 200 cGy suggests that the radiation affects either the CD2-cDNA and/or gene(s) regulating its expression. Our future approach will be focussed on the factors influencing the expression of the CD2 gene after radiation.

Participant No: 7 Kernforschungszentrum Karlsruhe, Institute for Genetics and Toxicology

X-RAY-INDUCED GENETIC RESPONSES IN MAMMALIAN CELLS AND COMPARISON TO UV- AND PHORBOL-ESTER-INDUCED RESPONSES

As in E.coli, in mammalian cells various kinds of radiation at low doses (X-rays, α-radiation, UV) induce the transcription and overreplication of specific genes. Within the last few years the research group in Karlsruhe has obtained several leads into the mechanism of signal transduction between the primary stress event (namely UV irradiation or phorbol ester treatment) and the genetic response. We propose now to extend these analyses to X-ray-induced gene expression. Specifically we plan to investigate the following:

Characterization of the inducing event. From the data on induction of gene expression by UV we suppose that DNA damage is a necessary intermediate. In order to investigate whether this holds also for X-irradiation, we will compare dose response curves in wild type and in X-ray-sensitive cells (Ataxia telangiectasia, X-ray sensitive CHO cells, "scid" cells); induction of gene transcription at lower doses in sensitive cells as compared to resistant cells would suggest, that unrepaired DNA damage is an intermediate in the signalling chain. Moreover, we will try to mimic the effects of X-rays by transfecting cells with genes coding for restriction enzymes cloned behind inducible promoters, or X-ray-damaged DNA's. Should this work, introduction of DNA with defined lesions, e.g. oxidized bases, will permit to define the inducing structure. Knowledge of the effective DNA structure will permit to search for the protein(s) that recognize(s) this altered DNA structure and trigger(s) a signal-transduction pathway.

Mechanisms of induction: The induction of gene transcription by UV operates through preformed transcription factors and involves the release of growth factors from irradiated cells and the activation of cytoplasmic protein kinases (raf-1, MAP-2 kinase). These experiments set the stage for the investigation of X-ray-induced gene activation. We will concentrate on the activation of collagenase gene transcription and ask the following questions: Which elements of the collagenase promoter are involved? Is protein synthesis needed? Are protein kinases, e.g. protein kinase C, involved? Which transcription factors are involved? Are growth factors released from the irradiated cells?

Long-term consequences: One of the most threatening consequences of low doses of ionizing radiation is cellular transformation. Evidence has been presented, that radiation does not only induce mutations directly but that it also establishes a cellular phenotype, which is characterized by an enhanced mutation rate (a mutation-prone state). We have described previously, that the Fos-protein is involved in UV-induced mutagenesis by

showing that (i) overexpressing of Fos induces mutations and (ii) antisense fos oligonucleotides inhibit the process. We have also shown, that the culture medium of UV- irradiated cells contains a factor or factors, which induce mutations in non-irradiated cells. We plan to extend these observations to X-ray-induced mutagenesis and to investigate how the expression of Fos and the secretion of radiation-induced factors lead to the mutator phenotype.

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GENETIC AND TUMOURIGENIC EFFECTS OF RADIATION-INDUCED STRESS RESPONSE.

1. X-ray-induced cell transformation in primary epithelial cells.

After improvement of the assay system for TDR (terminal differentiation resistant)-foci (a parameter for cell transformation) in primary mouse skin keratinocytes via the application of improved conditions for growth, the induction of TDR-foci will be studied in both normal and X-ray sensitive cells. The X-ray-sensitive cells are derived from newborn SCID mice known to be X-ray sensitive and deficient in DSB repair as well as to be deficient in the V(D)J recombination pathway. This will indicate whether the defective gene is involved in malignant transformation. A SCID mouse strain is present in our laboratory.

Transformation studies will be extended using cells from transgenic mice without a functioning p53 gene. As these cells have already undergone one step in the process of malignant transformation they require less additional changes for malignant conversion. A complementing oncogene for inactivated p53 could be an activated H-*ras* gene which is known to be activated by point mutation but not by gene rearrangement. Therefore the cells will be treated with either X-rays, which induce mainly chromosome alterations, or with ENU which induce mainly point mutations.

2. Stress response.

a. Stress response and fidelity of DNA synthesis.

Previously it has been found that DNA damage, next to the direct induction of mutations, leads to untargeted mutations due to an infidelity of DNA replication in the progeny of treated cells. This research has been extended by testing the effect of EPIF (extracellular protein inducing factors) known to transmit a stress response to untreated cells. To this end a cell line of mouse lymphoma cells (GRSL) has been treated with EPIF secreted by UV-irradiated cells. It is under investigation whether the progeny of the treated cells hava an enhanced mutation rate. This will be studied with mass cultures as well as with fluctuation analysis. It is also planned to identify growth factors present in the EPIF and to test these individually for their effect on fidelity of DNA synthesis.

b. Stress response and cell functioning.

As there are indications that the induction of a stress response is among others involved in wound healing, an assay system is being developed to assess the induction of MC (migration competence) in mouse keratinocytes. Such an assay system could distinguish between regulated and disregulated stress responses and relate the alteration in phenotype with genetic instability and susceptibility to malignant cell transformation. Therefore it is under investigation whether MC can be induced by DNA damage, tumor promoters, EPIF and cytokines. Subsequently the two phenotypes and their transition stage will be characterized for expression of genes involved in signal transduction pathways.

To monitor genetic instability in these cells a mutational assay system is also being developed.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920031 Studies on radiation-induced chromosome aberrations in mammalian cells. 2) Applied aspects.

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Description of research work

This project is mostly a continuation of EC RTD Project No Bi7-039.

It should be noted that this work forms one segment of a two-part programme, the other part of which is concerned with "basic" aspects of chromosome aberration formation (coordinator Prof. A.T. Natarajan). Close liaison between the two segments is planned, including joint co-ordination meeting to discuss results.

The main objectives of our project are:

A) Validation of the adaptive response

B) Validation of the G_2 phase chromosomal radiosensitive assay in order to identify radiosensitive and cancer prone individuals

C) Achievement of a meaningful score of chromatid-type aberrations that can be used for radiation-induced cytogenetic damage.

A) Low-dose, low-dose-rate effects and the adaptive response.

The evaluation of the genetic risk by low doses of radiation is a very important aspect of radiobiology. In fact, under normal conditions, and in the vast majority of emergency situations, the single doses received by individuals do not exceed levels of the order of O.1 Sv.

Radiobiological dose effects at higher dose and dose-rate levels are satisfactorily elucidated. The relationship of radiobiological effects to very low radiation doses and dose rates, however, still remain ambiguous.

People concerned with estimation of radiation risks extrapolate effects from those experimentally obtained at high doses (usually with a straight line) to the low- dose range. This linear extrapolation is controversial. Moreover there is now a considerable body of evidence showing that exposure of cells to very low doses of ionizing radiation can, "adapt" them such that they show a reduced response to a subsequent higher dose. The initial dose may protect by inducing or priming a repair mechanism. However there are apparent anomalies and donor variabilities in published data so that the so called "adaptive response" requires validation and clearer understanding.

A possible validation of the adaptive repair phenomenon accompanying low- dose radiation exposure could have significant impact on the interpretation of low- dose radiobiological effects and in the considerations of attendant risk assessments.

B) G_2 radiosensitivity and inter-individual variability.

It is known that inter-individual variability in human subjects exists for response to the induction of chromosomal aberrations by ionizing radiations. This individual

radiosensitivity is becoming more interesting for radiation protection and radiotherapy. Some of these traits are inherited as autosomal recessive disorders which often show cancer proneness and are in many cases results of mutation in genes involved in DNA repair. The G_2 phase of the cell cycle is the most sensitive stage for induction of chromosomal aberrations by ionizing radiation and it has been reported that the yields of x-ray induced aberrations are higher in G_2 lymphocytes and fibroblasts of the cancer-prone syndromes ataxia-telagiectesia (A-T) and Fanconi's anaemia. Differential G_2 radiosensitivity may then be used to identify radiosensitive and radio-resistant individuals.

These findings may have relevance for radiotherapy as well as identification of cancer prone individuals.

C) Cell kinetic perturbation and evaluation of radiation-induced cytogenetic damage.

Scores of chromatid-type aberrations are widely used in studies connected with the effects of and protection against ionizing radiation (and many other environmental mutagens).

Unlike chromosome-type aberrations, there is no known cell system where the target-cell population is homogeneous for the production of chromatid-type aberrations. Consequently, the recovered yield of any particular category fluctuates with the time of sampling after treatment; there is no unique yield to set against a given radiation dose. Moreover, the target heterogeneity means that any observed yield will have been profoundly influenced by the population cell kinetics, in particular, mitotic delay. Thus there is no simple relationship between observed yields and the underlying sensitivity patterns within cycle phases. This fact clearly has a bearing upon the interpretation of results from experiments conducted in A) and B) above.

The achievement of new results in these fields are of considerable interest for public health and safety particularly in view of the finding of recent surveys which appear to be revealing carcinogenetic effects, both direct and transmitted, arising from extremely low radiation exposures. Our studies of: 1) effects of low dose, low dose rate irradiation; 2) the validation of adaptive response; 3) the inter individual radiosensitivity and the identification of cancer prone individuals; 4) the more reliable quantitative use of radiation induced chromatid-type aberrations, are likely to make useful contributions in this area.

Moreover it should be noted that there are several collaborative links in the five projects and a large complementarity in the materials and methods that will be used.

Participation of the University of Rome

Factors affecting the radiation - induced adaptive response: interaction of low dose irradiation with subsequent mutagenic treatment (studies with human lymphocytes).

There is now substantial evidence indicating that in the evaluation of the genetic risk by low doses of ionizing radiation (<10 cGy) we must take into account both 1) the direct induction of genetic damage 2) the modifications induced by irradiation in cellular

metabolism, which in turn can interact with subsequent mutagenic treatment. On this respect several attempts have been made over the past few years to characterize a phenomenon observed in human lymphocytes. When human lymphocytes are cultured in tritiated thymidine or exposed to very low doses of X- rays they become less susceptible to the induction of chromatid aberrations by subsequent high doses of X-rays (Olivieri et al. 1984; Wiencke et al. 1986; Shadley and Wolff 1987).

As a continuation of our previous contract, in order to reach a better validation of the adaptive response, we will trying to understand the mechanisms underlying the described exceptions i.e. a) the experimental conditions required to produce the adaptive response or the synergistic effect (pH, temperature, compound added to the cultures, lymphocytes subpopulations) b) the causes of inter- individuals variability (age, life-style and various stress situations of the donors).

At same time we will undertake studies on c) the genetic damage induced in lymphocytes of individuals that for either occupational or medical reasons were exposed to low doses of ionizing radiations d) the mechanisms of "cross adaptivity".

All these studies should contribute to obtain a better insight into topics such as a) the biological effects of low doses and chronic treatments b) the assessment of genetic risk to man.

Participation of the University of Sevilla

Adaptive response to radiation damage in human lymphocytes conditioned with hydrogen peroxide: cytogenetic and molecular studies.

In this coordinated study, our contribution will cover two main aspects concerning the adaptive response to radiation damage in human lymphocytes: a) validation of the adaptive response in unstimulated G0 lymphocytes by means of the micronucleus test in binecluеate cells, and b) analysis of the relative importance of DNA single-strand breaks (ssb) and DNA double-strand breaks (dsb) for the induction of the adaptive response.

a) Adaptive response in G0 lymphocytes

There are controversial reports on the possible induction of the adaptive response in unstimulated human lymphocytes conditioned with low doses of ionizing radiation (Shadley et al., 1987; Sanderson and Morley, 1986; Cai and Liu, 1990).

The analysis of micronuclei resulting from chromosome aberrations in irradiated cultures of human lymphocytes is a rapid and reliable method, recently improved with the use of the cytokinesis block technique with cytochalasin B introduced by Fenech and Morley (1985).

We will make use of this methodology to analyze the adaptive response in G0 human lymphocytes from different donors conditioned with "adapting" doses of hydrogen peroxide or X-rays(low doses) and challenged with X-rays (higher doses) later on.

The conditioning treatment will be given at different times before stimulation of lymphocytes in order to analyze the possible interaction of the expression of putative repair genes with the general phenomenon of stimulation induced by a treatment with phytohemagglutinin (PHA).

The standard procedure followed by different authors to study the adaptive response in cultured human cells has been the scoring of chromatid and isochromatid deletions at metaphase shortly after irradiation. By so doing, only a subpopulation of damaged cells can be analyzed. Our alternative procedure allows us to harvest cells which were irradiated at different moments of their cell cycle, and the picture provided is more complete.

b) Relative importance of different DNA lesions for the adaptive response

In yeast, it has been recently reported that DNA ssb, as those efficiently induced by hydrogen peroxide, appear as more important than DNA dsb to trigger a molecular mechanism that results in a protection, in terms of cell killing, against a further treatment with high doses of ionizing radiation (Boreham and Mitchell, 1990). Human blood samples will be treated with hydrogen peroxide (to induce ssb) or irradiated with high LET radiation before the challenge with X-rays for the scoring of micronuclei as described above.

For the development of the two parts of the Project, an active cooperation with the other groups in the different european laboratories is our basic goal.

Participation of the University of Tuscia

Identification of radiosensitive individuals by means of ionizing radiation induced aberrations in G2.

It is planned to investigate the nature of human interindividual radiosensitivity which is becoming more interesting for radiation protection and radiotherapy.

As experimental approach G2 inter-individual variations for response to ionizing radiations will be studied in human lymphocytes, lymphoblastoid cells and fibroblasts from normal individuals or with genetic disorders predisposing to high risk of cancer such as AT, Fanconi's anemia, bloom syndromes and whenever possible the relative heterozygotes.

Particularly the influence of inhibitors of DNA synthetis or repair will be studied on chromosomal aberrations indueed by low doses of radiation with low or high LET, in order to avoid that, at higher doses, the large extent of DNA damage may hide some step of the DNA repair machinary deficient in radiosensitive individuals, which could only be detected at a low level of DNA damage.

This research project has several links with the other partecipants particularly with the interpretation of the adaptive repair phenomenon which has been observed in the G2 phase of the cell cycle and with the observed phenomenon on the interindividual variability in the "adaptive response".

Participation of MRC Radiobiology Unit, Chilton, Didcot

Evaluation of the effects of radiation-induced kinetic perturbations on the observed frequencies of chromatid aberrations.

As cells transit interphase of the mitotic cycle, the arrangement and physiology of the chromosomes within the interphase nucleus are subject to considerable change.

Ionizing radiation is able to produce aberrations at all stages of interphase, and many, if not all, the types produced are influenced, both qualitatively and quantitatively, by spatial and metabolic factors. Consequently, we expect, and indeed find, that changes on observed yield occur at different sampling times after radiation treatment. The target cell population is not homogeneous with respect to radiosensitivy, and the mixture of cells in the sample for scoring is continually changing with time.

In addition to the production of aberrations, cell progress towards mitosis is also disrupted be "mitotic delay", which is both a dose- and cycle-stage-dependent phenomenon. This perturbs the cell mixture even further.

The net result is that the observed fluctuations in the quantitative yield of chromatid-type aberrations with time, do not reflect, in any simple manner, changes in the pattern of intrinsic radiosensitivity, and the absence of a unique yield to set against a given dose precludes the construction of an interpretable dose-response curve. This limits the usefulness of chromatid-type aberrations as a meaningful end- point for fundamental studies.

Methods are available for marking the position of cells in certain parts of interphase at the time of radiation, so that cell populations can be "unscrambled", and target cells replaced in correct chronological order.

Large computer capacities allow sophisticated modelling of cell population kinetics. Theoretical yield-time aberration curves can be derived from such populations, with and without various models of mitotic delay, allowing delay effects to be monitored, and hypotheses checked.

A number of drugs (eg. Caffeine) are known, which appear to cancel, or mitigate radiation-induced mitotic delay. Thus some aspects of the influence of perturbation on yield can be determined experimentally, and compared with theoretical predictions.

Radiations of different quality produce different cycle responses both for aberration production and delay. Comparative studies will be informative.

We are beginning to use these approaches in an attempt to unravel the effects of delay on aberration frequency, and so determine how best to interpret and use scores of chromatid-type aberrations in the fundamental radiobiological studies generated by the other contractees in this project.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920042 Carcinogenic effects of low radiation doses and underlying mechanisms.

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Description of research work

I Objectives

In this joint proposal the emphasis is placed on the carcinogenic effects of low radiation doses and the underlying mechanisms. The influence of various exposure or host conditions on the probability of cancer development will be studied. The project involves the analysis of data from recently accomplished experiments, the completion of on-going experimental studies, and the performance of a limited selections of new series. This last activity is particularly important to test well founded hypotheses of radiation action based on the results of cellular and molecular biology studies. An effort will also be made towards a rationalization of the experimental data presentation, which needs to be formally organized by an international group of experts supported by adequate funding. Some aspects of the experimental results, in particular the dependence on radiation quality and time regime of irradiation, will be compared with the results of in vitro assays of cell transformation, which are partially proposed within a different contract.

II State of the Art

The assessment of the risk of exposure of man to ionizing radiation requires the knowledge of the probabilities of fatal cancers, and to a certain degree also of non-fatal cancers, induced by radiation. These probabilities are estimated, whenever possible, relying on epidemiological studies. However, data from human epidemiology alone cannot provide the detailed dose-effect relationships needed to regulate radiation protection, in particular regarding the lower dose-ranges, the influence of dose rate on the carcinogenesis after high-LET irradiation, the role of radiation quality, differences in sensitivity of subpopulations, transmission of carcinogenic effects through successive generations, and the influence of host factors, such as the species, age, sex and hormonal status of the irradiated individual. Also, the important questions arising from combined exposures to radiation and other carcinogens and promoters and inhibitors, cannot be unravelled by human epidemiology because of the complexity of our environment and its rapid changes. Furthermore, the following subjects deserve special research efforts: the inherent radiosensivity of individual organs, and the identification of molecular markers associated with specific neoplastic lesions. This latter information has at this moment mainly predictive value, but with improved biotechnological procedures modification of the radiation effect can also be envisaged. For all these reasons the use of biological model systems suitable to study the various aspects of radiation carcinogenesis is essential, and in particular animal studies represent at this moment the most complete and reliable source of data. In order to assess the validity of the results obtained with a model system, a comparison of experimental data on different strains, with different susceptibility to tumour induction is essential. In addition, to investigate the possibility of extrapolating the risk estimates to man interspecies comparisons are also of importance. Mechanistic studies of radiation induced neoplasia using animal models are presently confined to thymic lymphoma, myeloid leukaemia, osteosarcoma and mammary carcinomas in rodents. New insights into carcinogenesis emerging from molecular biological investigations, may provide, by identification of a variety of steps and pathways, much needed

information on the sensitivity to radiation among different cell types, and on cell cycle dependence. Molecular biological approaches may also prove to be instrumental in understanding specificities of responses now observed to be highly dependent on the strain of mice or rats.

III Degree of Innovation

The research of radiation carcinogenesis is based on well consolidated methods. Nevertheless, new insights emerging from molecular and biological investigation will provide useful information on the sensitivity to radiation of different cell types. An important innovation is also represented by the advanced statistical methods of analysis.

IV Economical, Social and Technical Benefits

An understanding of the factors influencing the tissue response to radiation is of a primary importance to assess the radiological risk in the various irradiation conditions. The results of the proposed project should provide information of relevance to shed light on the basic phenomena underlying tumour induction by radiation and therefore will be benefit to the tasks of the regulatory bodies.

V Relationship with other EC Programmes

It is anticipated that collaboration will develop between the work of the ENEA group and the EC funded project of R. Cox (NRPB) and M. Janowski (MOL) on murine radiation leukaemogenesis. In addition ENEA will be involved in a proposal to the EC for support of interlaboratory collaboration on C3H10T1/2 cell transformation studies.

VI Scientific and Technical Description of the Project

On the basis of the considerations exposed in Section II it is proposed to carry out the coordinated research activity in the following areas:

A. Analyses of recent animal experiments

The proposal includes the analysis of the results of recently completed experimental in vivo studies of tumour induction by fractionated doses of fission neutrons from the RSV TAPIRO reactor on long-living $BC3F^1$ male mice.

B. On-going studies

Experimental work will be continued to investigate the induction of myeloid leukaemia in CBA/Cne male and female mice after acute and fractionated doses of fission neutrons. The aim is to obtain further information on the influence of the strain and the irradiation regime on the induction of a relevant lesion by different qualities of radiation, and to validate the model based on the interaction of

transformation and inactivation of identified cells at risk, i.e. hemopoietic cells. These experiments will also provide material for studies of the genetic sensitivity to AML in rodents to be carried out by other European laboratories under a separate contract.

An on-going program on mammary carcinogenesis in the rats includes the effects of multiple low dose fractions (2.5 Mgy), various intervals between fractions and the effects of age at the beginning of the exposure. Verification is being sought for indications provided by human epidemiological studies that the most sensitive age period is before 20 years and that postmenopausal women are least sensitive. These studies of the age factor are carried out with single dose exposures. These investigations will be complemented with fractionated exposures at an early and at an older age. The role of estrogens on mammary cancer incidence is equivocal. Therefore, studies will be continued using low dose estrogen supplements again in young rats as compared to older rats.

C. New experimental series

It appears that, in spite of the difficulty to perform animal experiments, those planned specifically to solve specific problems would be of great importance. In the planning of experiments careful consideration should be taken in maximizing the power and minimizing the number of animals.

An intercomparison between species is necessary to assess the generality of results obtained from one species and therefore the possibility of extrapolating to humans.

An experimental study is foreseen on ovarian tumour induction using partial body irradiation, in order to investigate the influence of hormonal unbalance. It is also intended to study the possible association of characteristic tumours, such as lymphoma and myeloid leukaemia, with paternal irradiation of mice before conception.

An experiment is presently in preparation to test the possible increase of the effect of a low dose of neutrons by protraction over a year. In the experiment three groups of rats (male Sprague Dawley) would be irradiated with a low dose of neutrons. One group would be irradiated over a year (from three months to 15 months of age), two groups would receive a brief irradiation, one at three months and the other at 15 months of age.

The role of physical and biological factors involved in carcinogenesis, on expression of oncogenes, growth factors and their receptors in lung tumours using immunohistochemistry and EuA in situ hybridization methods will be studied. These tumours are induced by irradiation alone at different dose rates, at different ages by irradiation or chemical carcinogens alone or combined action of radiation and chemical toxicants, promoters or complete chemical carcinogens. Tumour samples will be prepared either unfixed or fixed in formaldehyde, frozen and then stored in liquid nitrogen until their histological study. For each tumour, the amount of DNA per tumour cell and the fraction of normal and proliferative cells will be measured on representative samples and the expression of different oncogenes, growth factors and their receptor will be studied using commercially available antibodies and RNA probes.

Based on large differences in susceptibility for induction of mammary carcinomas by radiation observed between different rat strains, e.g. between WAG (high) and the BN (low) strains, a crossing programme will be carried out, using different isozyme markers, trying to localize the gene(s) associated with radiation induced mammary carcinogenesis. Such a localization may be of help to define the molecular basis of differences in susceptibility.

D. Cell transformation assays

As different radiosensivity of different tissues to radiation is an important factor in determining the radiological risk, the susceptibility to neoplastic transformation by radiation of human derived cell lines from various epithelial tissue will be investigated, in addition to the continuation of on-going work in murine fibroblastic C3H10T1/2 cells, proposed within another EC coordinated project.

E. Development of advanced statistical methods

The carcinogenic effect in an irradiated cohort was frequently expressed as an incidence rate with respects to a control group. The incidence rate however not suffice as the only relevant parameter, since the time to tumour induction has been found to shorten appreciably with an increasing applied dose to the cohort. From the analysis of the time distribution to tumour induction relative hazards in the irradiated cohort with respect to the control cohort can be calculated and dose-effects relationships established. The analysis of the time to tumour induction can be undertaken by parametric and non-parametric methods. Parametric Weibull models are optimized via either the maximum likelihood or chi-square method, which are demonstrated to show similar results. It has already been shown that linear-quadratic dose-effect relation are strongly influenced by the experimental conditions such as fractionation and the combined application of drugs.

There is also a need for universally acceptable presentation and analysis of the data gained from animal experiments. We have in the last period concerned ourselves with the statistical methods to plan experiments and to analyze the results of carcinogenesis experiments. Methods have been developed to analyze different types of data (observable and incidental tumours). A set of programs is being prepared with a accompanying description. They should permit the comparison of results coming from different laboratories.

The objectives for the next project would be the application of these sets of routines to the analysis of low dose experiments. Data treatment will include correction for competing risk, the analysis will be performed in terms of tumour incidences and death rates.

F. Model interpretation

The large data base on late somatic effects in the experimental animals produced during the past research activity at some of the proposing laboratories will allow the investigation of cancer risk models. In addition, further attempts will be made to interpret these data using microdosimetric and/or multistage models of carcinogenesis.

VII. Distributions of tasks

ITRI-TNO: Studies on radiation carcinogenesis in the mammary of the rat with fractionated exposures at different ages. Investigations on the enhancing effect of low dose administration.

Explorations of molecular biology parameters, (proto-)oncogenes such as c-myc, N-myc, c-fos, etc. in mammary glands from different strains.

ENEA: Study of the carcinogenic effect of fractionated doses of fission neutrons on BC3F1 male mice. Investigation of the induction of myeloid leukaemia in CBAA/Cne male and female mice after acute and fractionated doses of fission neutrons. Study of ovarian tumour induction with partialbody irradiation. Transformation assays of human cells.

GSF: Development of statistical methods to plan experiments and to analyze the results of carcinogenesis experiments. Preparation of programs to analyze different types of data in cooperation with AZ Leiden and ENEA Casaccia.

AZL: Analysis of tumour induction data by different methods including Weibull-distributions, proportional hazards models, etc. in cooperation with GSF Neuherberg and ENEA Casaccia.

CEA: Studies on the effects of low doses of fission neutrons in Sprague-Dawley rats by a protraction over a year, in comparison with acute exposures at different ages.

IVVO-TNO: Pathology of carcinogenic effects with emphasis on scoring of benign and malignant tumours, origin of the neoplasms and cause of death. Comparison of histopathological criteria and nomenclature for tumours in the experiments performed at the institutes cooperating within this project.

Contribution ENEA-Rome

The contribution of the ENEA Casaccia Laboratory is based on the study of the influence of dose, dose rate and radiation quality on the induction of malignant transformation in different strains of mice of both sexes and various ages, with particular attention to low dose exposures. This includes the analysis of the results of recent experimental series, the completion of on-going experiments, and the performance of a limited selection of new series. New irradiations will be carried out using acute and/or fractionated doses of X rays and fission spectrum neutrons.

In order to investigate the effectiveness of protracted low neutron dose exposures, the results of recently completed experimental in vivo studies of the carcinogenic effect of fractionated doses of fission neutrons on long-living BC3F1 male mice are being analyzed.

Well selected experimental work is planned to investigate the induction of myeloid leukaemia (ML) in CBA/Cne male mice after acute and fractionated doses of fission neutrons. The aim is to obtain further information on the influence of the strain and

the irradiation regime on the induction of a relevant lesion by different qualities of radiation, and to validate the model based on the interaction of transformation and inactivation of identified cells at risk, i.e. haemopoietic cells. These experiments may also provide material for studies of the genetic sensitivity to ML in rodents.

An experimental study is foreseen on ovarian tumor induction after partial body radiation exposure, in order to investigate the direct and indirect effect of irradiation.

It is also intended to study the possible association of characteristic tumors, such as lymphoma and myeloid leukaemia, with paternal irradiation of mice before conception.

The different radiosensitivity of different tissues to radiation is an important factor in determining the radiological risk. Therefore, the ENEA laboratory has long term plans to study the neoplastic transformation induced by radiation in human derived cell lines from various epithelial tissues.

Relevant references

1) Covelli, V., Di Majo, V., Coppola, M., Rebessi, S. The dose-response relationship for myeloid leukemia and malignant lymphoma in $BC3F_1$ mice. Radiat. Res. 119, 553-561, 1989.
2) Di Majo, V., Coppola, M., Rebessi, S., Covelli, V. Age-related susceptibility of mouse liver to induction of tumors by neutrons. Radiat. Res. 124, 227-234, 1990.
3) Saran, A., Pazzaglia, S., Coppola, M., Rebbessi, S., Di Majo, V., Garavini, M., Covelli, V. Absence of a dose-fractionation effect on neoplastic transformation induced by fission spectrum neutrons in C3H10T1/2 cells. Radiat. Res. 126, 343-348, 1991.
4) Di Majo, V., Coppola, M., Rebessi, S., Saran, A., Pazzaglia, S., Covelli, V. Do multifractionated neutron doses enhance mortality in mice? Proc of 9th International Congress of Radiation Research, Toronto, July 1991.
5) Saran, A., Pazzaglia, S., Coppola, M., Rebessi, S., Di Majo, V., Broerse, J.J., Zoetelief, J., Covelli, V. Neoplastic transformation of C3H10T1/2 cells by fractionated doses of monoenergetic neutrons. Proc. of 9th International Congress of Radiation Research, Toronto, july 1991.
6) Covelli, V., Di Majo, V., Coppola, M., Rebessi, S. Neutron carcinogenesis in mice: a study on the dose-response curves. Radiat. Res. 128, 114-116, 1991.
7) Coppola, M., Di Majo, V., Rebessi, S., Covelli, V. RBE modifying factors. Radiation Protection Dosimetry 44, 1/4, 35-39, 1992.
8) Coppola, M., How to model radiation carcinogenesis? In: Biophysical Modelling of Radiation Effects (K H Chadwick, G. Moschini, M N Varma eds.) pp 339-342. Adam Hilger, Bristol, 1992.
9) Di Majo, V., Rebessi, S., Coppola, M., Saran, A., Pazzaglia, S., Covelli, V. Are somatic effects of low neutron doses detectable in vivo? In Low Dose Irradiation and Biological Defense Mechanisms (T. Sugahara, L.A. Sagan, T. Aoyama, eds.) pp. 199-202. Elsevier Science Publishers B.V. 1992.
10) Pihet, P., Coppola, M., Loncol, t., Di Majo, V., Menzel, H.G. Microdosimetry study of radiobiological facilities at the RSV-TAPIRO reactor. Seventh Symposium on Microdosimetry. Gatlinburb, 1992. Radiation Protection Dosimetry. In press.

11) Coppola, m., Specification of fast neutron radiation quality from cell transformation data. Radiation Protection Dosimetry. In press.
12) Saran, A., Broerse, J.J., Zoetelief, J., Pazzaglia, S., Pariset, L., Coppola, M., Di Majo, V., Rebessi, S., Covelli, V. C3H10T1/2 cell transformation after fractionated doses of neutrons of different energies. Submitted to Physica Medica.
13) Rebessi, S., Di Majo, V., Coppola, M., Saran, A., Pazzaglia, S., Pariset, L., Covelli, V. Somatic effects of low neutron doses. Submitted to Physica Medica.

Contribution AZ-Leiden

In the derivation of results from experimental carcinogenesis data, which have been collected over the past two decades in a number of institutes, different approaches were followed for the analysis. The carcinogenic effect in an irradiated cohort was frequently expressed as an incidence rate with respect to a control group. This incidence rate would however not suffice as the only relevant parameter, since the time to tumour induction has been found to shorten appreciably with an increasing applied dose to the cohort. From the analysis of the time distribution to tumour induction relative hazards in the irradiated cohort with respect to the control cohort could be calculated and dose-effect relationships have be established. A correction needs to be applied for right-censoring, if animals are lost from the experiment from causes unrelated to the endpoint. The analysis of the time to tumour induction can be undertaken by parametric and non-parametric methods. Parametric Weibull models are optimized via either the maximum likelihood or chi-square method, which were demonstrated to show similar results. Linear-quadratic dose-effect relations have been obtained, which are strongly influenced by the experimental conditions such as fractionation and the combined application of drugs.

Under a previous CEC contract nr. BI7-0035-C the development of the computer program LifeStat for the data analysis has been performed at the Department of Clinical Oncology (Leiden) in collaboration with GSF. The program currently provides for the analysis of single dose and fractionated experiments, but will be extended to the analysis of protracted irradiation experiments. The program is produced in such a way that it can be distributed among the various European institutes, that are involved in carcinogenesis studies. In a collaboration with ITRI-TNO the analysis of the results on mammary carcinogenesis is performed at our Department. The computer program LifePrep is also developed under the same contract and provides for a conversion of animal tumour induction data into an appropriate format for the LifeStat program. Currently the conversion is adapted to data from the CEN-FAR institute, but extensions to other laboratories will be performed. With the aid of the developed computer tools the data analysis for different large scale experimental programs will be undertaken in the proposed cooperative project.

Recent publications:

Broerse, J.J., Van Bekkum, D.W. and Zurcher, C.: Radiation carcinogenesis in experimental animals, Experientia 45, 60, 1989.
Davelaar, J., Broerse, J.J., Weeda, J., Chmelevsky, D.: Analysis of dose-effect relations for carcinogenesis in experimental animals, presented on 22nd Annual Meeting of the

European Society for Radiation Biology, Brussels, 1989.
Davelaar, J., Weeda, J., Broerse, J.J.: Analysis of animal carcinogenesis data by various mathematical methods, Rad. Env. Bioph., 30, 249, 1991.

Contribution ITRI-TNO

Continuation of research on modifying factors involved in the induction of mammary cancer by radiation in the rat

An ongoing program on mammary radiation carcinogenesis with emphasis on low dose irradiation is being carried out at Rijswijk. These studies include the effects of multiple low dose fractions (2.5 mGy), various intervals between fractions and the effect of age at the beginning of the exposure. Verification is being sought for indications provided by human epidemiological studies that the most sensitive age period is before 20 years and that post-menopausal women are least sensitive. These studies of the age factor are carried out with single dose exposures.

With regard to the role of oestrogens as contained in contraceptive pills, there remains uncertainty notwithstanding a great many epidemiological studies. With oestrogen modification as a replacement therapy for post-menopausal disorders, the effects on mammary cancer incidence are similarly equivocal.

Accordingly, the studies will be continued using low dose oestrogen supplement again in young rats as compared to older rats. In the older rats two groups will be studied, one receiving the irradiation at 8 weeks of age, the second group being irradiated at 64 weeks, both groups starting on oestrogens at 64 weeks of age.

Molecular biology of radiation-induced mammary cancer in rats

Tumours arisen in previous and in ongoing experiments will be tested for the presence of point mutations in either H-, K- or N-ras oncogenes by polymerase chain reaction (PCR) using intronspecific primers followed by dot blot hybridization with selected oligonucleotides or by direct sequencing.

Genetics of susceptibility to radiation-induced mammary cancer

The past experiments carried out at the ITRI-TNO indicated large differences between inbred rat strains in susceptibility to radiation-induction of mammary tumours. A crossing programme will be carried out between susceptible (WAG) and resistant strains (BN) giving rise to F_1, F_2 and backcrosses in order to find an association between different isozyme markers and susceptibility in an attempt to localize susceptibility gene(s).

The department of experimental dosimetry of the ITRI-TNO has been engaged for a long period in the radiation carcinogenesis of the rat mammary gland. The scientist in charge of this project, Dr Peter Bentvelzen, has been engaged for 30 years in the genetics, virology and molecular biology of mammary carcinogenesis in rodents.

- P. Bentvelzen, M.A. Dubbeld, W.H. Koornstra, J.J. Broerse and M.J. van Zwieten. Sera from irradiated rats contain antibodies to a ubiquitos tumour-associated antigen. Eur. J. Cancer 19 (1983) 1255-1263.
- P. Bentvelzen. Presence of fibroblast-transforming genes in normal DNA of several mouse and rat strains. Eur. J. Cancer (1984) 1493-1494.
- J.G.J. Bauman and P. Bentvelzen. Flow cytometric detection of ribosomal RNA in suspended cells by fluorescent in situ hybridization. Cytometry 9 (1988) 517-524.
- P. van Klaveren and P. Bentvelzen. Transactivating potential of the 3' open reading frame of murine mammary tumour virus. J.Virol. 62 (1988) 4410-4413.
- P. van Klaveren, A.G.M. Haaksma, J. Dijk and P. Bentvelzen. Oncogenic potential of retroviral transactivating genes. Rad. Environm. Biophysics 30 (1991) 199-200.

Contribution CEA Fontenay aux Roses

The aim of this project performed at Fontenay aux Roses is to characterize specific or common phenotype and genotype changed occurring in the target cells during their "progression" to lung tumors induced after irradiation, chemical or mineral compound administrations or combined treatments.

Irradiations will be mainly performed locally using gamma-rays exposure or radon inhalation at different low doses levels.

The compounds used will be mainly chemical inducers of lung cytochrome p-450 1A1 including non or poorly (5-6 benzoflavone, 2-3-7-8 tetrachlorodibenzo paradioxin), and strong (methylchloranthrene) carcinogenic agents following systemic administrations, beryllium as metal particules introduced into the lungs and asbestos fibers injected into the pleural cavity.

Cell changes will be characterized by light and electron microscopy using enzymatic, immunologic and in situ hybridization histological methods. Phenotypic studies will include expressions of enzymes involved in basal cell metabolism, xenobiotic metabolism and expression of other factors such as oncogenes, growth factors, and their receptors. Genetic studies will be first confined to measurement of the amount of DNA per cell. Fraction of proliferative cells will also be determined on tissue sections. Different fixed or unfixed samples of lung tissue will be kept at least -80 °C until either the histological processing related to this project or their use for further studies.

This project will allow a better characterization of:

* Tumors induced at a low frequency especially after exposure to low radiation doses.
* Steps occurring during histogenesis of tumors at high frequency so that the different lesions could be regarded specific or unspecific of the carcinogen agent and scored after treatments that induce low tumor incidence using, methods available nowadays in our laboratory.

Moreover, it will provide a kind of "banque" of neoplastic or preneoplastic samples for further studies by us and other laboratories aiming to limit in vivo carcinogenic experiments.

Contribution GSF-Neuherberg

Our present risk estimates are essentially based on human data gained at high doses with low-LET radiation. The extrapolation to low doses of risk estimates gained at high doses remains a central issue of radiation protection. In the past years several experiments have shown that the RBE of neutrons at low doses can reach high values. It is, however, still not clear whether this finding is of general applicability. The influence of a number of factors is insufficiently quantified. These factors are physical factors, such as LET of the radiation, protraction of irradiation as well as biological: age at irradiation, sex, etc.

Human epidemiology does not permit the quantification of risk in the range of doses and under the influence of factors particularly relevant to radiation protection: high-LET radiation, protraction of irradiation, combined influence of radiation and chemicals. The issue is made especially critical by the findings on the influence of protraction of irradiation. While it is accepted that protraction of μ-irradiation can decrease the effects, the reverse tendency has been indicated by a few epidemiological studies for high LET radiation. Experiments from recent years have led to numerous, partly contradictory results. The question is of major importance since a reverse time factor for high-LET would make the high RBE values observed for neutrons even more critical.

An experiment is presently performed at Fontenay-aux-Roses to test the possible increase of the effect of a low dose of neutrons by protraction over a year. In the experiment three groups of rats (male Sprague Dawley) will be irradiated with a low dose of neutrons. One group will be irradiated over a year (from three months to 15 months of age), two groups will receive a brief irradiation, one at three months and the other at 15 months of age. Moreover a program has been started by the European Community to compile animal data obtained from carcinogenesis experiments performed in the past in European laboratories with the purpose of improved accessibility of these data to researchers.

There is therefore a need for universally accepted presentation and analysis of the data gained from animal experiments. We have in the last period concerned ourselves with the statistical methods to plan experiments and to analyse the results of carcinogenesis experiments. Methods have been developed to analyse different types of data (observable and incidental tumors). A set of programs has been prepared at Leiden with our collaboration. With these programs it is now possible to analyse results from life time experiments in terms of tumor incidences and death rates. The tumor rates can be estimated according to the nonparametric lifetable-method or with a Cox regression. Alternatively analytical expressions such as Weibull or log-normal functions can also be applied. The basis of these methods should now be described, i.e. derivation of the likelihoods, the method of optimisation, confidence intervals of the parameters should be given.

The programs will be illustrated on a set of data resulting from experiments performed in the past at the laboratories in Fontenay-aux-Roses or at TNO Rijswijk. RBE values and reduction of life expectancy due to tumor induction will be derived. The work will also include the introduction of the isotonic regression in the program package.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920043 Measurement of oncogenic transformation of mammalian cells in-vitro by low doses of ionising radiation.

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Description of research work

1. OBJECTIVES AND EXPECTED ACHIEVEMENT

This is a collaborative study of dose-response relationships for cell transformation *in vitro* at as low a dose as can be achieved. A realistic minimum dose at which to assess transformation frequency with adequate precision and achieve meaningful comparisons between participating laboratories during the this contract is 0.1 Gy. In addition, subgroups in varying combinations of the participants will continue more specialised investigations as detailed in sections 3.3.2 - 3.3.6. Close links will be maintained with other laboratories working on cell transformation and with groups working on the development of new systems.

Specific objectives include:

1) the preparation of a standard manual for cell transformation assays; this will include precise criteria for scoring transformed foci and an explanation of the classification in terms of examples of transformed and non-transformed foci;

2) intercomparison experiments at doses down to 0.1 Gy of X-radiation;

3) ultimately, by pooling data from each individual laboratory, to extend the determination of dose-response relationships down to a dose of 0.01 Gy;

4) in addition to these studies, and with the same framework which has been established for the C3H 10T½ cell line, to introduce epithelial cell systems and assays that have an increased relevance to human risk estimation, as, and when they become available; and

5) the formation of individual subgroups to continue more specialised investigations: dose-rate effects with densely ionising radiations (3.3.2), role of neighbouring entities of high ionisation density using ultrasoft X-rays and Auger-emitters (3.3.3), transformation frequency and the cell cycle (3.3.4), alternative indicators of transformation (3.3.5) and alternative transformation assays (3.3.6).

2. ECONOMIC, SOCIAL AND TECHNICAL BENEFITS

Standardisation will allow data from different laboratories to be pooled and will ensure that results are interpreted uniformly. The resultant data for low doses will enable current models of radiation risk to be improved. This can lead to increased efficiency and financial savings in radiological protection as the uncertainties in risk estimation at low doses will be reduced.

3. STATE OF THE ART AND SCIENTIFIC DESCRIPTION

3.1 Introduction

The main risk from exposure to low doses of ionising radiation is the induction of cancer. Its quantification, together with the assessment of hereditary and prenatal risks, determines the setting of limits in occupational exposure and action levels for the protection of the general public. Currently the risks of developing radiation-induced cancer are predicted by various methods but these cannot be validated at the low radiation doses that are relevant in radiological protection. Dose-response relationships for tumour induction can be studied using animal models but at low doses these may pose some problems, mainly related to the size of the experiments. A different approach is to use an *in vitro* model such as cell transformation for which a variety of systems are available. *In vitro* cell transformation studies have become an important alternative for the study of the carcinogenic potential of different types of ionising radiation and other cancer-causing agents. They do not provide direct quantitative risk estimates, but produce important information about dose-effect relationships and their dependence on dose-rate, radiation quality and on the combined effects with other agents. By utilising the new techniques of molecular biology it is also possible to gain an insight into the cellular and molecular mechanisms underlying carcinogenesis. However, only one cell transformation assay, the C3H 10T½ mouse fibroblast system, provides the high precision needed for work at low absorbed doses. This system is routinely used in a number of laboratories in Europe and the USA.

There is a substantial range of data using C3H 10T½ cells from various laboratories, but data at low doses are severely limited and there exist substantial differences in the results from different laboratories. Standardisation is therefore an urgent task. If reliable and credible data are to be obtained at low doses a large number of transformants must be scored to reduce the statistical variation. For one laboratory, this may exceed the resources of a single laboratory. Hence since June 1990, six laboratories (at Harwell, Berkeley, Munich, Göttingen (the research team based in Göttingen was located in Frankfurt prior to 1992), Milan and Rome) have collaborated closely on CEC-sponsored contracts the ultimate aim of which is to establish the dose-response relationship for radiation-induced cell transformation *in vitro* at doses as low as 0.01 Gy. A critical step is the standardisation of the assay (which has now been achieved). The present contract is a continuation of these studies.

3.2 Progress to Date

The transformation assay, like most other biological assays, is susceptible to the effects of added influences. Such influences do not obviate the internal comparison of results within a particular laboratory, but they may preclude the direct comparison of results between laboratories unless suitable steps are taken to identify and minimise the effects of relevant factors. The critical factors for cell transformation are the conventions used for scoring transformed foci, the growth conditions used for the assay and the characteristics of the cells used for experimentation. Collaborative work so far has concentrated on establishing a standardised assay for the cell transformation system and this has involved intercomparison experiments and focus-identification exercises.

TABLE: Ratios and Standard Deviations of Individual Score to Consensus Score for Transformed Foci

	Date of Focus Scoring Intercomparison		
Laboratory	July 1990	May 1991	October 1991
Berkeley	1.8 ± 0.6	0.7 ± 0.2	1.0 ± 0.2
Frankfurt	1.3 ± 0.3	0.8 ± 0.3	0.8 ± 0.2
Harwell	1.4 ± 0.2	1.3 ± 0.7	0.8 ± 0.3
Milan	0.8 ± 0.1	1.1 ± 0.3	1.0 ± 0.1
Munich	1.1 ± 0.3	1.1 ± 0.3	0.9 ± 0.1
Rome	--	1.2 ± 0.6	1.1 ± 0.2
OVERALL	1.31 ± 0.19	1.05 ± 0.49	0.95 ± 0.24

One of the first parameters checked by the six groups was the scoring of transformants. For this purpose each group circulated a number of flasks or petri dishes for scoring by the other groups. This was followed up by a joint scoring exercise during which a consensus score for each flask or dish was determined. Altogether three such exercises have now been completed. Initially, there was a relatively large discrepancy between some laboratories (up to a factor of 2), but the disagreement following the third scoring exercise was reduced to a maximum of about 20% (see table).

In addition direct intercomparison transformation experiments have been carried out in order to determine the influence of medium type on the transformation frequency. Cells from one participating laboratory (Berkeley) were irradiated and immediately placed on melting ice at 0°C along with a sample of unirradiated cells. These cells were then despatched to the other laboratories for subsequent culture. A common protocol was adhered to within the limits of each laboratory. This involved the culture of cells in both local medium and in medium as supplied by Berkeley. It was decided initially that these experiments should be of limited size and hence an absorbed dose of 5 Gy was chosen such that an adequate number of foci would be obtained. Altogether four experiments at 5 Gy have been carried out. The conclusions from these experiments are: (i) the viability of cells up to 72 h on ice is not a problem; (ii) the choice of medium (local or Berkeley) has no significant effect on the results; and (iii) the seeding density interval between 1 and 3 viable cells per square centimetre is a region of small variation in transformation values; transformation frequency decreases rapidly at values higher than 4 to 5 viable cells per square centimetre. As a result of these intercomparisons the six laboratories have now established standard protocols for the culture and scoring of transformation in C3H 10T½ cells. Currently transformation intercomparison experiments at doses of 1, 2 and 3 Gy are being carried out. More recently, preliminary data from Berkeley has indicated that an increase in the expression time for transformation from six to eight (or possibly nine) weeks is desirable. It appears that not all transformed cells have had sufficient time to produce easily recognisable foci at the shorter time period. It is expected that by adopting this protocol it will be possible to extend measurements to below 0.1 Gy of X-rays.

3.3 Current Studies

3.3.1 Measurement of Cell Transformation by Low Doses

This is a continuation of the current studies and involves all laboratories. It is anticipated that it will be possible to collectively measure transformation frequencies down to 0.1 Gy during the period of the contract. In addition a manual describing the recommended procedures for carrying out the C3H 10T½ transformation assay is being produced. This will enable other laboratories, not involved in our collaborative work, to carry out experiments using identical protocols to that used for the work described here.

3.3.2 Dose-Rate Effects with Densely Ionising Radiations

The possibility of a reversed dose-rate effect for cell transformation (and by implication for carcinogenesis) with densely ionising radiations is a critical issue in radiological protection. Its resolution is a central issue for the quantification of the risks from neutron and α-particle irradiation and for the determination of suitable quality factors for all densely-ionising radiations. It is also relevant to current concerns regarding environmental exposure to radon and its possible association with lung cancer. Measurements on the effects of α-particles (Berkeley, Milan and Munich) and neutrons (Berkeley and Rome) will be used to undertake further studies relating to the inverse dose-rate effect. These studies will include the use of plateau-phase cultures, lower doses and longer irradiation times than have been used in previous studies. Each source being used has a different radiation quality (2.5 MeV neutrons, fission energy neutrons, curium-244 α-particles, plutonium-238 α-particles and americium-241 α-particles) and so the results are complementary and will provide a unique set of data on dose-rate effects from densely ionising sources.

3.3.3 The Effects of Characteristic Ultra-Soft X-Rays and Auger-emitters

Results on the effects of C_K characteristic ultrasoft X-rays (Energy: 0.28 keV) suggest that closely related entities of high ionisation density, rather than isolated clusters of ionisations, are important for cell transformation. Thus, closely related massive DNA lesions such as double-strand breaks or bulky lesions rather than isolated DNA lesions of these types are critical. Göttingen is investigating the effect of the distance between closely related clusters of high ionisation density using a variety of characteristic ultrasoft X-rays simultaneously generating two or more critical DNA lesions.

Auger-electron emitters produce high, locally concentrated energy depositions in DNA when a radionuclide such as ^{125}I is covalently bound to the DNA. Munich is studying the effectiveness of such radionuclides to transform cells when Auger electrons are emitted during radioactive decay.

These studies, using well defined clusters or distributions of clusters will provide information about the effectiveness of low-energy secondary electrons and Auger emitters and lead to a better understanding of the molecular mechanisms underlying radiation-induced cell transformation and carcinogenesis.

3.3.4 Transformation Frequency and the Cell Cycle

There are suggestions that the inverse dose-rate effect may be a result of a period of high sensitivity (to transformation) during the cell cycle. Work at Milan, Munich and Rome includes systematic studies using fractionation schemes and synchronised cell populations to investigate this hypothesis.

3.3.5 Alternative (Early) Indicators of Transformation

There is a need to provide alternative techniques for the detection of transformation. Ideally such techniques should be very sensitive and provide a measure of transformation frequency at an earlier time than is currently used for the focal assay. Work on this is in progress at Harwell where specific cell surface markers for transformed cells are being sought. At Berkeley, work on the possibility of correlating intracellular ion changes (such as Ca^{++} the concentration of which is known to be reduced for transformed cells) is planned. In addition, other intracellular properties of transformed and non-transformed cells will be investigated using image analysis and immunochemical techniques.

3.3.6 Alternative Transformation Assays

Most human cancers are epithelial in origin and it is important to understand the response of human epithelial cells to radiation, as they may have some different radiobiological characteristics than rodent cells. Berkeley, Harwell and Rome are maintaining close links with other groups involved in the development of human transformable cell lines, and in particular are assessing their suitability for inclusion into the framework of low-dose intercomparison experiments already established for the C3H 10T½ cells. Göttingen is investigating the possibility of using radiation-sensitive (oncogene transfected) C3H 10T½ cells for low dose studies.

4. DETAILED DISTRIBUTION OF TASKS

- Prepare and distribute test populations of irradiated C_3H 10T½ cells: Berkeley
- Prepare manual for cell transformation assay: all laboratories
- Collaborate to measure transformation frequencies for X-radiation at doses down to 0.1 Gy using standard cells and protocols: all laboratories
- Investigate the potential of biochemical techniques and image analysis for the early detection of transformants: Berkeley and Harwell
- Investigate dose-rate effects for X-radiation at low doses and dose-rates: Berkeley
- Investigate dose-rate and fractionation effects for α-radiation at low doses and dose-rates: Berkeley, Milan and Munich
- Investigate dose-rate and fractionation effects for neutrons at low doses and dose-rates: Rome
- Investigate the effects of ultra-soft X-rays and Auger electrons: Göttingen and Munich
- Establish the suitability of using oncogene transfected 10T½ cells for low dose studies: Göttingen
- Use molecular biological techniques to investigate the cellular and molecular mechanisms of cell transformation: Munich
- Investigate the influence of the cell cycle on cell transformation: Milan, Munich and Rome

- Maintain links with other groups developing human cell lines for radiation-induced cell transformation studies: Berkeley, Harwell and Rome

ROLE AND CONTRIBUTION: BERKELEY

The known health effects from exposure to ionising radiation are mainly derived from information obtained at high exposures and exposure rates. In radiation protection information is required on the effects of low doses and dose-rates. For sparsely ionising radiations most radiobiological studies show a reduced effectiveness at lower dose-rates. For radiological protection purposes it is assumed that the dose-response relationship for radiation-induced cancer is linear - at least at doses where cell killing is insignificant. Hence the derived risk is directly proportional to the dose. To account for the reduced effectiveness at low doses and dose-rates a reduction factor - DDREF (dose and dose-rate effectiveness factor) - is applied to the risk estimates derived from high doses and dose-rates to give more realistic estimates for protection level exposures. One current problem is the considerable uncertainty associated with estimates of DDREF which have been considered by several international committees to be in the range 2 to 10. Combined with the uncertainties on the risk estimates from acute exposures uncertainties at low doses and dose-rates span a factor of about twenty. More precise values for DDREF are needed.

The vast majority of studies with cultured cells have utilised log phase cells. Such populations of cells have high growth fractions, high proportions of cells in the S phase of the cell cycle and low proportions of slowly proliferating, G_0 or quiescent cells. However, most tissues *in vivo* are characterised by cell populations in which a relatively large proportion of the cells are noncycling or are cycling very slowly. Thus many studies with log phase cells are not only often complicated by cell cycle kinetic effects but they may not be truly representative of the situation *in vivo*. The only report for cells irradiated as plateau-phase cultures suggests that low doses may be relatively more effective in producing transformants than high doses. More data need to be generated at low doses and dose-rates using plateau phase cultures.

The contribution of the Environmental Technology Branch at Berkeley Technology Centre towards this project will include (a) participation in the *in vitro* cell transformation intercomparison experiments; (b) the measurement of dose-rate effects with ^{238}Pu α-particles, 2.5 MeV neutrons and low doses of X-radiation; and (c) the development of techniques for the early detection of transformants.

(a) We will continue to irradiate and distribute cell samples for intercomparison experiments, with the intention of reducing doses used in these experiments down to 0.1 Gy.

(b) We will use ^{238}Pu α-particles to measure dose-rate effects at dose-rates down to 10^{-4} Gy min^{-1} at doses as low as 0.06 Gy. These investigations will utilise both log-phase and plateau phase cultures and will provide crucial information relevant to the inverse dose-rate effect. We will use X-rays for investigations with plateau phase cells at doses down to 0.1 Gy and for irradiation times lasting up to 3 weeks. The X-ray experiments are specifically aimed at providing information relevant to DDREF.

(c) We will use our image analysis system to investigate a possible correlation between intracellular ion concentrations and cell transformation.

Contribution of the Department of Clinical Radiobiology and Radiation Physics of the University of Göttingen.

(1) Participation in the collaboration on in vitro cell transformation.

The aims of the collaboration are:

- to continue the intercomparison experiments down to doses of 0.1 Gy;

- to improve further the standard protocol so that cell transformation frequencies can be obtained for doses down to 0.01 Gy by pooling data from each participating laboratory.

- to prepare a manual for cell transformation assay including about hundred examples of transformed foci.

(2) Use of radiation sensitive cell lines for cell transformation studies in a pilot study.

In a pilot study it will be elucidated whether oncogene transfected cell lines of CH310T1½ exhibit a higher radiosensitivity so that they become suitable to determine more easily cell transformation frequencies in the low dose region. Three different oncogene transfected cell lines of CH310T1½ (gag-myc, H-ras and gag-myc/H-ras) will be studied. Although their spontaneous transformation frequencies per dose may be higher. For these studies ^{60}Co-gamma-rays at high dose rate are used.

(3) Use of characteristic ultrasoft X-rays to study for neoplastic cell transformation the role of the interaction of primary DNA lesions (DNA double-strand breaks, bulky lesions) induced clusters of high ionization density.

The RBE-values of C_K photons smaller than one at low doses and greater than one for doses higher than 2 Gy suggest that the distance between clusters of high ionization density and thus the distance between specific DNA lesions (DNA double-strand breaks, bulky lesions) is an important parameter for neoplastic cell transformation. Therefore, transformation frequencies in CH310T1½ cells will be determined after exposure with characteristic ultrasoft x-rays which generate two or more clusters at high ionization density along definite (combined) ranges r of the Auger- and photoelectrons arising from the photoabsorptions. Besudes F_K (E=0.68 keV, r=20 nm) and Al_K (E=1.5 keV, r=70 nm) the following characteristic ultrasoft x-rays will be used: Cu_L (E=0.93 keV, r=30 nm), Zr_L (E=2.1 keV, r=120 nm) and Cd_L (E=3.1 keV, r=300 nm). These studies will provide information about the effectiveness of closely related clusters of high ionization densities to transform cells and will contribute data to improve our understanding of the molecular mechanisms of radiation carcinogenesis by α-particles from radioactive decay.

ROLE AND CONTRIBUTION: HARWELL

The work proposed by Harwell Laboratory will contribute to the development of alternative transformation assays. The main programme of work will be the development of a rodent epithelial assay based on rat tracheal cells, which can be used as a bridging system between animal and human models.

Development of the Rat Tracheal Epithelial Cell Bridging System

'Bridging' systems in which cells can be exposed to carcinogens both in vivo and in vitro are of great importance for studies of carcinogenesis as they allow in vivo validation of in vitro measurements. One such system being studied at Harwell is the rat tracheal epithelial cell system (RTE) of Thomassen *et al.* (Carcinogenesis, 7 (12), 2033, 1986) in which cells of the tracheal epithelium can be exposed to carcinogens either *in vivo* or *in vitro.* Pre-neoplastic cells are selected by their resistance to serum-mediated differentiation, forming large colonies of altered cells, known as 'enhanced growth variants' (EGVs), which can progress to produce fully neoplastic cells.

This system has been established in our laboratory and other participants also have appropriate expertise, both directly and indirectly related to the development of the assay. Thus, this seems a useful technique to develop, building upon exsisting strenghts within the group. It is now proposed to undertake a programme comparing the transformation frequency of these RTE cells exposed to high and low doses of radiation both *in vivo* and *in vitro.* Initial work will concentrate on *in vitro* effects, firstly to obtain dose-response relationships for the survival of RTE cells irradiated *in vitro* with both γ-rays and α-particles. The frequency of EGV's will be determined and dose response curves defined; EGV's will be expanded in culture and assayed for tumourigenicity in *nu/nu* mice. Subsequently, the carcinogenicity of radon will be determined *in vivo* using the exposure suite at Harwell. Tracheal cells will be isolated after exposure and the EGV frequency determined, permitting comparison with the *in vitro* data.

In the latter half of this contract, providing sufficient progress has been made, pilot experiments with RTE cells may be conducted in other laboratories within this working group.

ROLE AND CONTRIBUTION: MILAN

(a) Standardization of procedures with 10T½ cell transformation assay.

Results from the existing European collaboration have clearly shown that there is a need to identify common criteria. We will continue the joint efforts to complete the standardization. A standard manual will be prepared and a reference dose-response relationship for transformation with hard X-rays will be determined.

(b) Systematic study of "inverse dose-rate effects" with α-particles.

Enhancement in transformation when the dose is protracted, that is the inverse dose-rate effect, was reported for fission spectrum neutrons from the Janus reactor for the first time in the early eighties. Since then, various studies have been performed but this effect

is far from being clarified. There is evidence that the extent of the enhancement may depend on dose, irradiation scheme and radiation quality. An enhancement after fractionated exposure was found with charged particles of LET between 40 and 120 keV/μm. An enhancement of about 1.4 was found in our laboratory with 4.3 MeV α-particles, LET=101 keV/μm when a total dose of 0.21 Gy was fractionated. Transformation frequency will be determined in 10T½ cells exposed to α-particles of different qualities using various fractionation schemes.

(c) Study of transformation sensitivity in the cell cycle.

One of the most promising of the several interpretations formulated to explain the inverse dose-rate effect is the notion that postulates the existence of a period of high sensitivity to transformation during the cell cycle. Transformation frequency will be determined in synchronized 10T½ populations exposed to 4.3 MeV α-particles at various stages. The influences of irradiation on the progression of the cell in the cycle will be studied.

(d) Development of models for interpreting cell transformation data.

Published data will be analyzed to develop and test models and to study the possibility of inferring dose-effect curves at very low doses.

The irradiation device, an appropriate irradiation chamber with a 42 MBq ^{244}Cm α-source, is already available, as well as dosimetry, beam monitoring and appropriate cell supports for irradiation. The Milan group has many years of experience in the radiobiology of mammalian cells, particularly in cell transformation *in vitro* with both low and high LET radiation.

ROLE AND CONTRIBUTION: MUNICH

General Considerations

In vitro transformation studies have become an important alternative to epidemiology and animal models to study the carcinogenic potential of different kinds of ionizing radiation. They do not provide direct quantitative risk estimates, but produce important information of dose effect relations and their dependence on the temporal distribution of dose, on radiation quality, and on the synergism with chemical and metabolic factors. A variety of these studies can ultimately lead to insight into cellular and molecular mechanisms of radiation carcinogenesis. There is a substantial range of data with 10T½ cells from various laboratories with different kinds of ionizing radiations, but data derived from low dose experiments are limited and there are still substantial differences in the results for dose-effect relations and RBE-values from different laboratories; standardization is therefore an urgent task. A standardization project in six European laboratories has been started in 1990 - supported by the CEC -, and a number parameters have been identified that had earlier led to inconsistent results in different laboratories.

Proposed Work

The first aim of this project is to continue the efforts to obtain precise dose-effect relations for radiation-induced cell transformation at low doses; this can only be achieved, if one is able to pool data from the cooperating laboratories. This study will be performed with sparsely ionizing radiations, i.e. x- and γ-rays, and it will be further extended with regard to densely ionizing particles at low and high dose rates.

After the contradictory findings on the inverse dose-rate effect by Hill *et al.* (1984) with fission spectrum neutrons and by Hieber *et al.* (1987) with α-particles various studies have been performed in different laboratories to assess the influence of the temporal distribution of dose. The contradictory data have, however, not been resolved, and it is still a highly contraversial question whether the inverse dose-rate effect exists and what magnitude it may attain. There is some evidence of an LET dependency of the inverse dose-rate effect; an increased efficiency at low dose rate or after fractionated exposure was seen only with radiations of LET between 40 keV/μm and 120 keV/μm. There are also suggestions that the dose-rate effect may be a result of sensitivity changes within the cell cycle. The problem of a reversed dose-rate factor for cell transformation, and, by implications for radiation carcinogenesis, is critical to radiation protection and its resolution is therefore a central issue for the quantification of risks and for the revision of the quality factor for densely ionizing radiation. To clarify the open problems, transformation experiments will be performed with synchronized cell cultures or with agents, such as caffeine, that modify in other way the cell cycle distribution and radiation-induced division delay.

In view on previous data by Le Motte *et al.* (1982), a further objective of this project is the study of the effectiveness of radionuclides that emit Auger electrons during radioactive decay. These Auger electron cascades lead to high, locally concentrated energy deposition in the DNA when the radionuclide is covalently bound to the DNA, as it can be achieved by ^{125}Iododeoxy-uridine. This study is to provide information about the effectiveness of Auger emitters and lead to a better understanding of the molecular mechanisms underlying cell transformation and radiation carcinogenesis. It will be specifically aimed at an examination for the possibility - indicated in the work of Le Motte *et al.* - that a damaging event anywhere in the DNA can lead to 'pretransformation' and then over several steps to neoplastic transformation of the cell.

These experiments will be closely linked with the study of cellular and molecular mechanisms of cell transformation that will be performed with radiation-transformed Syrian Hamster embryo (SHE) cells. cDNA libraries of primary SHE cells, radiation-transformed SHE cells, and tumour-cell lines derived after injection of neoplastically transformed cells into athymic nude mice have been constructed. By differential hybridization a series of cDNA clones - representing specific mRNAs - have been isolated that are either repressed, deleted or overexpressed in radiation-induced transformants; by this technique oncogenes as well as tumour suppressor genes are to be tracked down. These DNA sequences can be identified after amplification by the use of polymerise chain reaction (PCR) by direct sequencing. In parallel, specific translocations, deletion of genes or non-random chromosome loss will be identified at the chromosomal level by the use of classical cytogenetic techniques, *in situ* hybridization and flow karyotyping. For this purpose chromosome libraries will be constructed and used for the identification of translocations and deletions.

ROLE AND CONTRIBUTION: ENEA - CASACCIA

The contribution of the Laboratory of Carcinogenesis at ENEA Casaccia Centre will include:

a) continuing joint effort in establishing an European standard of procedures for transformation in the C3H10T½ cell transformation system;

(b) use of C3H10T½ cells for dose-protraction studies with neutrons of different qualities;

(c) investigation of age-related sensitivity to neoplastic transformation after fission neutron irradiation;

(d) use of human cells systems for transformation studies.

In the past decade several in vitro studies of neoplastic transformation have suggested that when neutrons are delivered at low dose rate, or in multiple fractions, their effectiveness is increased. Theoretical considerations seem to support this finding, although no conclusive explanation has been given. No enhancement of transformation frequency for the C3H10T½ cell system has been reported by other studies after low dose rate fission neutrons and alpha particle irradiation compared to acute irradiation, and a fractionation study performed in our Laboratory with fission spectrum neutrons in the low-dose range has also shown no inverse fractionation effect. As at the present time there appears to be accumulating evidence that the extent of the enhancement effect may be LET dependent and related to the particular fractionation or protraction scheme, our next experiments will be aimed at investigating different neutron energies, as well as different fractionation schemes. Furthermore, as several suggestions have been made that mitotic cells are the sensitive population to which the enhancement is due, the cell cycle dependence of cellular susceptibility to transformation will be analyzed after fission spectrum neutrons irradiation of the C3H10T½ system.

In order to assess radiation risk, it is important to understand the rsponses of human cells, especially of epithelial origin. Human cells have somewhat different radiobiological characteristics than rodent cells. We are presently in contact with Dr. J.S. Rhim to acquire the human epithelial cell lines RHEK-1 and RHEK-271, consisting of human keratinocytes. Neoplastic transformation by radiation has been shown to be induced in a quantitative manner on the line RHEK-271. Experiments with this line might provide additional information about the dose-protraction effects of high LET radiation. We also intend to use other human cell systems, such as mammary, bronchial and uroepithelial derived cells, from preliminary studies of radiation induced neoplastic transformation.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT920053 Molecular and cellular effectiveness of charged particles (light and heavy ions) and neutrons.

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Description of research work

General summary of the project

1. Comparison of the effectiveness of different charged particles

In the last years a considerable amount of experimental work has been devoted to elucidate mechanisms of radiation action on biological matter. Studies on the biological effects, such as cell inactivation, mutation induction, ssbd, dsb and cromosome aberrations, caused by radiations of different types and energy are considered useful tools for identifying the physical characteristics of the radiation that are relevant to the action on biological targets.

These are the essential steps for developing realistic theories of radiation action on biological matter, in particular:

i) to predict the biological effectiveness of any radiation;
ii) to give a better knowledge about the molecular lesions leading to cellular effects;
iii) to develop deterministic models of radiation action, i.e. biophysical models.

It can be easily recognized that only in this framework it is possible to deal with, and hopefully solve, a number of specific problems, such as:

a) the relationships involved in the chain: energy deposition pattern (spatial and time)-molecular damage-repairability-cellular effects;

b) the interplay between the energy deposition pattern and the structure of the relevant target, essential to obtain a possible explanation of the different sensitivity of different cell lines (e.g. differentiated vs. non-differentiated cells) in terms of the different chromatin organization;

c) the effect of the dose rate, in particular to ascertain whether or not a linear response means independence of the dose.

Recently, it has been shown and confirmed by independent experiments in different european laboratories that different light ions with the same LET and at the same dose level produce different amount of biological damage, that is, they show a different RBE. In particular, low energy protons resulted more effective in inducing cell inactivation and mutation than other heavier charged particles (namely, alphas) in the 10-35 keV/μm LET region. More recently, deuteron beams have been used to extend the earlier proton LET range to higher LET values, studying the induction of cell inactivation.

As aim of the present project it is therefore useful to complete this work and to extend it using heavier charged particles and fast neutrons to perform direct comparison of the biological effectiveness of the different radiations and to gather a set of radiobiological data which could be used as a consolidated data-base to test models of radiation action used in radiation protection for a practical assessment of human risk.

2. Microbeam: single-particle irradiation of cells

At the low dose levels generally encountered in environmental and occupational exposure to high-LET radiations, only a very small fraction of the cells at risk are traversed by a particle and a much smaller fraction by two or more particles. In experiments "in vitro" using conventional broad-field irradiation techniques, it is often not practicable to simulate this situation because at the protection-level doses only a small proportion of the cells will actually receive any dose and respond.

This means that to achieve adequate sensitivity such experiments generally have to be conducted at unrealistically high levels of dose, with cells receiving randomly distributed numbers of traversals, and therefore a corresponding range of individual doses. Where the effect being studied "in vitro" is non linear with dose, this heterogeneity may detract from the accuracy and relevance with respect to low-dose effect "in vivo". The microbeam approach is intended to overcome this problem by irradiating cells individually with exact numbers of particles delivered to precise locations within each nucleus.

By exploiting the experimental opportunity provided by the microbeam facilities, under installation at LNL-INFN and CRC Gray Laboratory, irradiation facilities dedicated to radiobiological studies are planned, in the present project, to start such a kind of investigation.

3. Use of human cellular system

As far as cellular endpoints such as survival, mutation and transformation is concerned, it is generally acknowledged that these are not only the consequences of physical factors, such as energy deposition events, but also the expression of several biological factors. From this point of view the nuclear chromatin organization plays an important role in determining radiation sensitivity, affecting both the amount of initial damage and the accessibility of the repair enzymes to the damaged sites. The majority of the data available in the literature has been obtained using actively proliferating cells. However, in a tissue, the largest part of the cells are in different stages of differentiation, unable to replicate and owning differential structural organization of the genome, different metabolism and enzyme content (including those involved in the repair processes). For this reason, the use of human cells capable to undergo "in vitro" differentiation can represent an important tool for radiobiological studies. This kind of approach allows to better clarify the basic mechanisms of radiation action and have practical implication in radioprotection and radiotherapy. In the present project it is planned to use both the actively proliferating cells and the human (differentiated and non-differentiated) cells and to perform, where appropriate, direct comparison.

4. Development of new assays for DNA damage and biophysical modelling

Biophysical modelling has shown that the distributions of deposited energy in DNA and of complexity of lesions induced vary with LET in a manner that can be related to biological effectiveness. At present, the modelling predictions cannot be validated

directly against experimental data because conventional assays measure only the gross damage to DNA and not its underlying complexity. Development and establishment of different assays for DNA damage will be made by the Demokritos, ISS, Giessen and CRC group. Moreover, the groups of MRC and Giessen will perform theoretical developments related to the experimental results by Monte-Carlo calculations and deterministic approaches, respectively.

Summarizing briefly, the main objectives of the project are to:

1) measure the effectiveness of deuterons in inducing HPRT mutations, as a function of LET;
2) measure the effectiveness of $^3He^{++}$ and α-particles as well as of heavy ions like Li, B, C, N, O and P in inducing cell inactivation and mutation;
3) make direct comparison of the effects of protons/deuterons with $\alpha/^3He^{++}$ for similar LETs as well as of the investigated heavy ions, where appropriate;
4) start comparative experiments of the effectiveness of high and low dose rate irradiation with α-particles;
5) start measurements of the effectiveness of fast neutrons in inducing cell inactivation and mutation;
6) measure the initial yield of DNA dsb in cells irradiated with protons, deuterons and helium ions;
7) start experiments on the effectiveness of a single and few charged particle (proton or alpha) to inactivate a single cell or to induce malignancy;
8) set up a beam line for irradiation of cell monolayers with heavy ions;
9) set up appropriate irradiation facilities based on the use of microbeam systems;
10) make comparative experiments using differentiated and actively proliferating cells to study the influence of DNA conformation on the radiation induced damage and repair;
11) develop several new assays to detect the distribution of lesions within clustered damage sites induced by radiation of various LET;
12) analyse the microscopic track structures of radiations to seek features which correlate with their observed biological effectiveness.

Contribution of the LNL group

Biological Effectiveness of different charged particles and neutrons

In order to extend our previous studies on the biological effectiveness of protons to higher LET values overcoming the limitation due to the proton range in biological matter, we have started systematic experiments at the CN facility with deuteron beams, which should have the same track structure as protons of the same velocity. We propose, in collaboration with the ISS group, to complete these experiments and to perform:

1 -Direct comparison of protons and deuterons with a-particles of similar LETs, furnished by the LNL 16.5 MV XTU-Tandem, to establish well the differences in biological effectiveness of protons and alphas as already reported, using the same cell line, the same physical and biological experimental conditions; we will study cell inactivation and mutation induction.

2 -Analysis of the biological effectiveness of a representative set of heavy ions, like Li, B, C, N, O and P, (obtained from the LNL XTU-Tandem), to gather radiobiological data in the few MeV/nucleon region useful in both the nowadays debated therapy with charged particles, the neutron bio-medical applications and radioprotection. Cell inactivation and mutation induction will be measured. The setting-up of a heavy ion radiobiological facility at the LNL XTU-Tandem is under way.

3 -Analysis of effectiveness of neutrons of energy up to 8 MeV, measuring V79 cell inactivation and mutation induction. Monoenergetic neutrons in the range of few keV-8 MeV are available at the neutron facilities set-up at the LNL CN accelerator for nuclear physics experiments.

Comparison of the effectiveness of high and low dose rate exposures with α-particles

In order to investigate a possible effect of dose rate on the effectiveness of high LET radiation we propose, in collaboration with the ISS group, to measure and compare the lethal and mutagenic effects in V79 cells irradiated with α-particles of 100-300 keV/μm LET with α-source irradiators (^{241}Am or ^{244}Cm), which are especially designed, constructed and tested at the LNL, and which permit continuous irradiation during cell growth at the dose rate of 0.005-0.01 Gy/min. The high dose rate irradiations will be performed at the existing LNL CN radiobiological facility.

Single particle irradiation of cells

In order to investigate the lethal and mutagenic effects of one or few charged particles in a cell, we propose to set-up a dedicated irradiation facility at the "microbeam facility" of the LNL 2MV AN2000 Van de Graaff accelerator to perform V79 cell inactivation and mutation induction experiments using protons and alphas.

Dosimetric measurements

For all the planned experiments, we will perform careful dosimetric measurements with the usual methods based on the use of silicon and CR39 nuclear track detectors. Moreover, regarding the neutron dosimetry, the multisphere Bonner technique will be used too.

Contribution of the CRC group

Biological effectiveness of protons, deuterons and α-particles

We have already studied the protons effectiveness (RBE) in the 17-32 keV/μm LET range. We intend to make a direct critical comparison of protons/deuterons with α-particles/^{3}He^{++} of similar LETs. In preliminary studies, we have compared deuterons with protons where possible matching the LET's. Our earlier work with α-particles, at ~3MeV from a ^{238}Pu source, will be extended to both higher and lower energies. The Gray Laboratory's 4MV Van de Graaff accelerator will be used to accelerate a-particles and ^{3}He^{++} over a wide range of energies up to 8.5 MeV using our already established method of preparing cell monolayers on polyvinylidine difluoride membrane filters. The end

points measured will be cell inactivation, DNA dsb (using neutral filter elution and pulsed-field gel electrophoresis) and the $HPRT^-$ mutation. Repair studies will be included where appropriate. Other work with protons and a-particles will require the new microbeam facility.

Microbeam single-particle irradiation of cells

Development of our microbeam facility is well under way, with a purpose-built irradiation room and Van de Graaff beamline completed and much of the microbeam hardware designed. A new solid-state microscope (BC Cancer Centre, Vancouver) has been evaluated and selected by us as providing the required resolution and sensitivity in the epifluorescence mode to locate cell nuclei precisely with respect to the microbeam. The system is being developed in conjunction with our existing computer-controlled Dynamic Microscope Imager (also from BCCC) which is currently used for single-cell recognition assays of inactivation at doses down to 0.01 Gy.

We propose to exploit the unique opportunities provided by the single-particle microbeam system to investigate cell responses to individual particle tracks using cellular and molecular endpoints, with protons, deuterons and a-particles.

Molecular, physico-chemical and modelling studies of clustered DNA damage

We propose to develop several new assays to detect the distributions of lesions within clustered damage sites induced by radiation of various LET in plasmid and genomic DNA. These include the application of our fast response techniques to probe, via their reactions with oxygen and thiols, the physico-chemical nature of the initial lesions formed by radiations of various LET. This development will contribute to our modelling of the modifying effect of the chemical stage on lesion size. Other methods include the use of gel electrophoresis or HPLC to quantify distributions of DNA fragments and, in conjunction with end-labelling techniques, to score the numbers of sub-lesions induced, with endonuclease treatments being used to recognise specific types of base damage. These studies will be directed at linking biophysical modelling and physico-chemical and molecular endpoints with biological effectiveness and repair.

Contribution of the MRC group

The MRC group propose to provide intercomparisons of selected dosimetric and biological conditions and to investigate the microscopic track structures of the particles used at Legnaro and the other participating laboratories.

In previous collaborative experiments carried out on the Harwell cyclotron we have shown, by direct comparison, that protons of 20 and 23 keV/μm are more effective than α-particles of the same LETs in causing inactivation and mutation of V79 Chinese hamster cells. We propose to compare the dosimetric methods that we used on the Harwell cyclotron (ionisation chamber and CR39 track detectors) with that in use at Legnaro (silicon detector) and to investigate also the profiles of the etched tracks in the CR39 plastic. These comparisons should assist in evaluating any biological or physical differences, or similaries, that may be found between protons and deuterons of the same LET, especially since it is commonly assumed that the differential microscopic track structures of these particles is virtually identical because of identical charged and velocity.

We propose also to provide some basis for biological intercomparison, for mutation induction particularly, at the higher LETs planned for the new radionuclide α-particle sources at LNL and ISS. Our plutonium-238-α-particle irradiator provides α-particles at selectable LETs of about 100-150 keV/μm and selectable doses rates from 10^{-5} Gy/min to 20 Gy/min. It thus allows access to beam parameters beyond those available elsewhere. We shall also provide our own results on cell inactivation and mutation ($HPRT^-$) for specific comparisons with the other laboratories.

We have been using Monte-Carlo track structure simulations with the code MOCA 14 of Wilson and Paretzke to generate tracks similar to those used in the biological experiments of MRC and LNL. These include protons, deuterons and a-particles of selected LETs in the region 20-30 keV/μm. From these we are evaluating, by extensive computer scoring, absolute frequencies of energy deposition in small cylindrical targets, similar in dimensions to DNA, nucleosomes and chromatin fibre, in order to seek microscopic features which do, and those which do not, correlate with observed differences in biological effectiveness of the particles.

We propose:

-to extend these analysis to additional LETs in use in this overall co-ordinated contract;
-to seek other microscopic features that may be of direct relevance to the biological effectiveness of the particles for initial DNA damage and for final cellular effects;
-to estimate the frequencies and spatial magnitudes of the clustered damage (in DNA and associated molecules) that can be produced by these particles.

Contribution of ISS group

Comparison of the effectiveness of different charged particles

In recent years, in the framework of a collaboration between LNL and ISS, we have shown that low energy protons are more effective for cell inactivation and mutation induction than other (notably alpha) particles in the LET range 10-35 keV/μm. In order to extend the range of LET studied, we have carried out experiments with deuteron beams that should have the same track structure as protons because of the same charge and velocity. At present, survival determinations are completed and the results under analysis, while mutation induction and DNA damage assays are still in progress. These experiments will be completed and extended to the analysis of the effectiveness of other particles such as alphas, heavy ions and neutrons in collaboration with the LNL group.

Repair of molecular lesions

Experiments performed on DNA damage induced by protons and alpha particles have shown that the initial yield of double strand breaks (dsb) in quite insensitive to radiation type and LET. This finding seems to support the current opinion that the increased biological effectiveness for radiations of increasing LET can be due to the clustering of the initial damage affecting the repair processes. The repair of DNA damage induced by different charged particles in the same LET range will be investigate and an intercomparison will be performed with the results from experiments carried out by the other coordinated laboratories, where different techniques will be used.

Comparison of the effectiveness of high and low dose rate irradiation with charged particles

In order to analyze a possible effect of the dose rate on the effectiveness of high LET radiations (that present linear responses) we will investigate, in collaboration with the LNL group, the lethal and mutagenic effects in V79 cells irradiated with alpha particles of 100-300 keV/μm. Irradiation at dose rates as low as 0.005-0.01 Gy/min will be performed using the ^{239}Pu or ^{244}Cm source especially designed at the LNL, while the high dose rate irradiation will be carried out using the CN-7MV accelerator.

Use of human cellular system

The use of "in vitro" human cell culture can allow more reliable indication for developing biophysical models of radiation effects useful for the evaluation of risk assessment. In the framework of the collaboration between the ISS and the LNL groups, we will extend the studies with high LET radiation to TK6 human lymphoblasts and K562 human proerythroblasts, possessing different X-ray sensitivities. Actively proliferating cells will be used to establish the correlation between cell lethality and DNA damage after exposure to protons, deuterons and alpha particles in the same LET range. K562 cells at different stages of differentiation, owing a different genome structural organization, will be used for studying the influence of DNA conformation on the radiation induced damage and repair.

Contribution of the Demokritos group

The proposed work aims to a comparative study, mainly at the biophysical level, of the actions of high LET ionizing radiation (protons and α-particles) vs low LET ionizing radiation (gamma rays), on mammalian macromolecular DNA. Monoenergetic accelerated protons up to 8 MeV are available at the proton beam outlet for biological work at the Tandem Accelerator Laboratory of the N.C.S.R. "Demokritos". A Co^{60} gamma rays source (7 Gy/min) and an Am^{241} α-particles source, designed for biological work, (75 MBq) are located at the Radiation Genetics Unit (RGU) where most of proposed work will be carried out. Part of the biophysical experimentation (Dielectric and Conductivity measurements) will be conducted at the Department of Physics of the National Technical University of Athens while Monte-Carlo modelling and analysis work will be carried out at the Radiobiological Laboratory of the Medical School of the University of Athens.

During the last three years the work at the RGU was focused on the adaptation of physical research methods for the study of the effects of ionizing radiation on macromolecular mammalian DNA. These methods include:

- Inverse Gas Cromatography for the estimation of changes in Gibb's Free Energy, Enthalpy and Entropy, arising from the severance of hydrophobic and hydrogen bonds present at the DNA double helix, following exposure to ionizing radiation;

- Perturbed γ-γ Angular Correlation Studies for probing on the energy conduction and the flexibility of macromolecular mammalian DNA exposed to ionizing radiation;

- Thermal Transition Spectrophotometry in conjunction with the development of thermal denaturation model for the study of energy propagation in macromolecular mammalian DNA carrying SSB and/or DSB which facilitate the severance of the hydrophobic and hydrogen bonds along the DNA molecule;

- Conductivity Measurements for the study of alterations evolving on the DNA molecule prior to processes inducing severance of the DNA hydrogen bonding;

- Dielectric Studies for the investigation of the dielectric behaviour of DNA and on the bound water sheath surrounding the DNA molecules.

During the last three years these techniques were used for the study of the effects of gamma irradiation on the genetic material of mammalian cells and preliminary work has been expanded to cover the effect of α-particles.

We propose the expansion of this work on studying the effects of the exposure of mammalian macromolecular DNA to protons of different energies up to 8 MeV and α-particles for radiation quality factors estimation at the molecular level. The work will be backed by comparative studies on survival and chromosome aberrations frequency on V79 cells.

Contribution of the Giessen group

The group works at Giessen with a tandem accelerator for protons and helium ions (max. energy 2.5 MeV) presently adapted for radiobiological studies with mammalian cells; it also regularly works at GSI (Darmstadt) with heavy ions, in the past with energies up to 20 MeV/u and now with energies that will be be extended to several hundred of MeV/u. It will thus be possible to link the work on protons and very light ions with that of heavy particles using the same assay methods. Recently the action of individual ions on single cells has been investigated in some detail in yeast; these experiments are currently extended to mammalian cells. On the theoretical side, models of ion energy deposition and cellular radiation action were developed and tested versus our own experimental results as well as data from the literature. Our group has been recognized (together with other) as a NASA Center of Research and Training in the field of Radiation Health. The studies performed in this context - which are not part of the present proposal - will constitute a link between radiation health problems on Earth and in Space.

Specific contribution:

1- The Giessen group will measure survival, mutation induction, formation of DNA double strand breaks and cell cycle kinetics (using cytofluorometry, available in the laboratory) in mammalian cells (V79 Chinese hamster, P3 human) after exposure to low energy protons and helium ions at the accelerator on site. Cytofluorometric measurements will complement the current understandig of cell cycle kinetics and repair with high-LET radiations. Particular emphasis will be laid on human cells in order to assess differences and similarities between mammalian species and contribute more directly to the problems of human radiation protections. Parallel experiments will be performed at Legnaro.

2 - The method to study single ion effects will be further developed for mammalian cells and tested in Giessen and Darmstadt. It is expected that these studies will contribute to understanding of the microstructure of energy deposition and its biological relevance. The "microbeam facility" at Legnaro will be used for comparative investigations.

3 - Work on theoretical descriptions of energy deposition using deterministic approaches will be continued and compared to the Monte-Carlo calculations performed by the Harwell group. It is hoped that this cooperation will yield useful and practical results applicable in radiation protection.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

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60 KECU

Description of research work

When accidental exposure of persons to ionising radiation occurs, a fast accurate and reliable method of biological dosimetry is needed to estimate whether measures are required. Automated analysis of radiation induced chromosomal changes, once having met these requirements, could be used for a rapid screening of humans exposed to low doses of ionizing radiation.

Commercially available systems for automated chromosome analysis have been trained to recognize standard patterns corresponding to the 23 pairs of normal metaphase chromosomes. Chromosome changes characteristic for specific types of genetic disorders or tumours, often correspond to frequently recurring marker chromosomes. These types of aberrations may also be picked up by high quality automated systems. Radiation, however, produces stochastic chromosome changes that cannot be analyzed in this way, because these random chromosome aberrations are highly variable in shape and DNA content. Other methods of automated analysis should be introduced for these types of aberrations.

The general aim of this project is to develop methods for simplified and automated assessment of radiation induced damage in chromosomes and nuclei, and to make these available for radiation protection purposes. This involves:

- preparation and staining of chromosomes in metaphase and interphase and of micronuclei, suitable for quantitative analysis,
- automated analysis of radiation induced changes in chromosomes, interphase nuclei and micronuclei by fluorescence microscopy, by confocal scanning microscopy and by flow cytometry.
- assessment of dose-effect relationships for radiation protection purposes.

The investigations will be directed at achieving the following objectives:

- To establish reliable, reproducible in situ hybridisation protocols with fluorescent probes for centromeres, telomere marking and for whole chromosome painting
- To establish methods for the isolation of chromosomes and micronuclei that yield suspensions of particles with optimally preserved morphology, and to evaluate a variety of methods for attaching fluorescent probes by DNA in situ hybridisation or immuno labelling to these chromosomes and micronuclei in suspension.
- To establish by microscopic scoring representative dose-effect curves for translocations highlighted by fluorescence in situ hybridisation (FISH). Radiation qualities and dose rates relevant for radiation protection purposes will be used. Methods will be developed for a precise discrimination between stable and unstable aberrations.
- To establish methods for monitoring the morphology of the interphase chromosomes during the cell cycle. The induction of chromosome and chromatid exchanges by radiation should be strongly influenced by the arrangement of the chromosomes in the interphase nucleus with respect to each other.
- To relate these results to other endpoints such as chromosome aberrations observed at metaphase and micronuclei observed in the cell cycle following irradiation.

- To further increase the sensitivity of the micronucleus assay using visual and flow cytometry methods for an improved biological dosimetry in humans.
- To create a fluorescence microscope image processing system for finding metaphase spreads, detecting labelled chromosomes at high resolution and producing an analysis in terms of radiation dose.
- To establish an automated technique for detecting karyotype abnormalities in large numbers of cells using slit scanning of fluorescent chromosomes in suspensions prepared from irradiated cells.

In the course of the project, highly innovative methods will be developed. These involve, in particular, the combination of two-and three-colour fluorescence in situ hybridisation and other fluorescence labelling techniques with automated fluorescence microscopy, (slitscannning) flow cytometry, and 3-D imaging by confocal microscopy. This should improve the sensitivity of the chromosome aberration and micro-nucleus assays. Moreover, joining advanced in situ molecular labelling methods to quantitative detection and imaging techniques is expected to help fill the gap between conventional molecular biology and radiation cytopathology. The results will be relevant for our understanding of the role of the nuclear organisation in the induction of chromosome aberrations. Provided scoring of translocations by FISH can be established for routine use in biological dosimetry, the assay could improve the dose reconstruction particularly of old or chronic exposures. Since we have current co-operations with several scientific institutions in the Community of Independent Republics we can contribute to an extension of dose estimations and risk evaluation of persons, e.g. liquidators exposed to radiation at the Chernobyl accident.

Thus the project will benefit government and medical authorities by providing fast methods for monitoring cytogenetic effects in workers exposed to ionising radiation through their professional activities, and for responding to radiation accidents. Moreover, contacts of the partners with bio-medically oriented industrial organisations in the EC may result in the production and marketing of probes and instruments that are developed within the scope of the project.

Bauchinger, M., Schmid, E., Braselmann, H., Willich, N. and Clemm, Ch. (1989) Time-effect relationship of chromosome aberrations in peripheral lymphocytes after radiation therapy for seminoma. Mutation Res. 211, 265-272.

Bayley, R., Carothers, A., Chen, X., Farrow, S., Gordon, J., Ji, L., Piper, J., Rutovitz, D., Stark, M and Wald, N. (1991) Radiation dosimetry by automatic image analysis of dicentric chromosomes. Mutation Research, 235, 223-235.

Boschman, GA., Rens, W., Manders, EMM, VanOven, CH., Barendsen, GW and Aten, JA. (1991) On-line sorting of human chromossomes by centromeric index, and identification of sorted populations by GTG-banding and fluorescent in situ hybridization. Hum.Genet. 85, 41-48.

Farr, C., Fantes, J., Goddfellow, P and Cooke, H. (1991) Reintroduction and function of human telomeres in mammalian cells. Proc.Natl. Acad.Sci USA 88, 7006-7010.

Green, DK., Fantes JA. and Evans HJE. (1989) Detection of randomly occuring aberrant chromosomes as a measure of genetic change. In: Flow Cytogenetics. (Ed. Gray, JW). Academic Press, new York, 161-171.

Manders, EMM., Stap, J., Brakenhoff, GJ., VanDriel, R. and Aten, JA. (1992) Dynamics of three-dimensional replication patterns during the S-phase, analysed by double labelling of DNA and confocal microscopy. J.Cell Science, 103, 857-962.

Miller, BM., Werner, T., Weier, HU. and Nüsse, M. (1992) Analysis of radiation-induced micronuclei by fluorescence in situ hybridization (FISH) simultaneously using telomeric and centromeric DNA probes. Radiation Research, 131, 177-185.

Nüsse, M., Kramer, J. and Miller, BM. (1992) Factors influencing the DNA distribution of radiation-induced micronuclei. Int.J.Radiat.Biol., 62, 587-602.

Rimpl, GR., Schmid, E., Braselamnn, H. and Bauchinger, M. (1990) Chromosome aberrations induced in human lymphocytes by 16.5 MeV protons. Int.J.Radiat.Biol. 58, 999-1007.

Salassidis, K., Huber, R., Zitzelsberger H. and Bauchinger, M. (1992) Centromere detection in vinblastine- and radiation-induced micronuclei of cytokinesis-blocker mouse cells using in situ hybridization with a mouse gamma (major) satillite DNA probe. Environm. Molec. Mutagen. 19., 1-16.

Schreiber, GA., Beisker, W., Bauchinger M. and Nüsse, M. (1992) Multiparametric flow cytometric analysis of radiation-induced micronuclei in mammalian cell cultures. Cytometry 13, 90-102.

VanOven, CH and Aten, JA.,. (1990) Instrument for real-time pulse-shape analysis of slit-scan flow cytometry signals. Cytometry 11, 630-635.

Contribution of the University of Amsterdam

Slit-scanning flow cytometry analysis of dicentric chromosomes

Flow cytometry is a rapid and accurate method for chromosome analysis. Radiation-induced chromosome aberrations, however, are variable in DNA content and can be analysed by slit-scanning flow cytometry only. The scanning of the fluorescently labelled dicentric chromosomes yields profiles in which the two centromeres are recognised as dips.

Aggregates of chromosomes, however, can also yield profiles with more than one dip. To determine profile characteristics that distinguish real dicentric chromosomes from artefacts, we are developing a high-speed pulse-shape analysis module for sorting chromosomes that correspond to trimodal profiles. These chromosomes are then deposited at well documented positions on slides, to assess the frequency of false-positives. The slit-scanning method described here is based on standard cell sorter instruments and it could be made available to those clinical and research centres that have access to these instruments.

To increase the sensitivity of the slit-scanning analysis, the optical system of the cell sorter instrument will be adapted. In addition, we will develop methods for highlighting the centromeres of isolated chromosomes with fluorescent anti-kinetochore antibodies, in collaboration with Green et al. at Edinburgh.

Chromosome suspensions prepared from cells, irradiated in the G_0-phase of the cell cycle, will be analysed for the frequency of dicentric chromosomes by slit-scanning flow karyotyping and by microscopy analysis. Cells from the same cultures will be plated for assessment of cloning capacity as a test for cell survival. Dose-effect relationships will then be analysed in cell types with different radiation sensitivities and in lymphocytes. In addition, dicentric chromosomes can be sorted for subsequent analysis with FISH, in collaboration with Bauchinger and Green et al., to assess the relative frequencies of exchanges between different chromosome types.

Morphology of chromosome domains in interphase nuclei in relation to the induction of chromosome exchanges

Little is known about the way in which the induction of chromosome exchanges by ionising radiation is influenced by the arrangement of the chromosomes in the cell nucleus. Presently, however, tools for quantitative investigation of the 3-dimensional organisation of the cell nucleus are becoming available.

It is expected that chromosomes that occupy extended or elongated domains in the interphase nucleus will be more sensitive to the induction of inter-chromosomal exchanges than chromosomes contained in more spherical domains. The latter type of chromosome could be relatively more sensitive to the induction of intra-chromosomal exchanges, i.e. deletions and inversions.

To investigate this problem, we will apply fluorescent chromosome paints and replication markers to stain (parts of) chromosomes in the nucleus (collaboration: Bauchinger, Green et al.). 3-dimensional images of cell nuclei will be obtained by two-colour confocal scanning laser microscopy. In addition we will sort cells according to their DNA content, to study the interphase chromosomes as a function of the cell cycle. 3-D image analysis techniques will be developed and applied for the automatic analysis of changes in the size, shape and position of (parts of) chromosomes in the interphase nucleus. The corresponding analysis in meta-phase cells of chromosome exchanges will be performed using paints for whole chromosomes and for parts of chromosomes.

Contribution of GSF (Dr. M. Nüsse)

Analysis of micronuclei using flow cytometry and FISH

A promising alternative to the analysis of chromosome aberrations seems to be the scoring of micronuclei (MN) which is meanwhile well known to provide a quantitative measure of the degree of cytogenetic damage in mammalian cells exposed to ionising radiation and chemicals. The dosimetric potential of this micronucleus assay is currently under investigation by microscopic analysis including image analysis and flow cytometry.

During our earlier EG-project (B17-0038-C, from 1.8.1991 -31.7.1992) we have developed a new flow cytometric technique to measure the frequency of radiation-induced MN in cell cultures and human lymphocytes (Schreiber et al., 1992). With this new technique a suspension of MN and main nuclei is obtained. The DNA of micronuclei and nuclei is stained with two different DNA-specific fluorescent dyes for flow cytometric analysis of DNA content and frequency of MN. MN can be discriminated from unspecific debris by simultaneous measurement of several flow cytometric parameters. Irradiation of cell cultures and lymphocytes in vitro showed that doses as low as about 0.1 Gy can be detected with this new automated technique. Unfortunately, as already known from conventional microscopy, the first results from a number of healthy persons showed that the frequency of MN in unexposed humans is rather high and can vary by a factor of at least 4. This variability reduces the dose sensitivity of the MN-assay since the background level of MN in any individual examined will be unknown and only an average population control value can probably be used. The aims of our new project are to further increase the dose sensitivity of the MN-assay for an improved biological dosimetry in humans. Several possibilities to obtain a lower dose sensitivity will be studied in detail:

1. The age dependence of the frequency of MN will be included in the analysis of MN. It is well known that with increasing age the frequency of MN increases both in males and females. This effect should therefore be included in the dose estimations of humans of different ages exposed to ionising radiation.

2.An even more promising new endpoint will be studied in detail: The chromosomal composition of MN. We have recently shown that it is possible to detect acentric fragments and whole chromosomes in radiation-induced MN using simultaneously telomeric and centromeric DNA probes and in situ hybridisation in mouse cells (Miller et al., 1992, Salassidis et al., 1992). With this technique the frequency of MN containing whole chromosomes and one or more acentric fragments can be measured in the microscope or by image analysis. Differences in the distribution of the chromosomal composition between radiation-induced and spontaneous MN were found. Because ionising radiation mainly induces acentric chromosome fragments, this technique could be used to analyse the chromosomal composition of micronuclei in human lymphocytes with a combination of human telomeric and centromeric DNA probes and, additionally, with chromosome specific and chromosome painting probes.

3. For a fast and automated detection of MN containing certain chromosomes or chromosome fragments identified by FISH with chromosome specific DNA-probes, flow cytometric techniques will be developed to analyse MN stained with DNA probes in suspension. It has already been shown that it is possible to perform FISH with nuclei in suspension. We will try to adapt this technique for flow cytometric analysis of MN in suspension stained with DNA probes. This will possibly enable us to rapidly identify radiation-induced MN and to discriminate them from spontaneous MN found in unexposed humans.

4. With our flow cytometric technique the DNA distribution of MN is measured simultaneously. We have recently shown that several factors can influence the DNA distribution of radiation-induced MN (Nüsse et al., 1992). These factors are: The chromosome size distribution of the cells, the presence of whole chromosomes and of one or more acentric fragments in MN and DNA synthesis in MN. The relative amount of these factors is different in radiation-induced and in spontaneous MN. By analysing the

DNA distributions of radiation-induced MN taking additionally into account the results from the FISH experiments it could be possible to better understand differences between DNA distributions or radiation-induced and spontaneous MN.

Contribution of GSF (Dr. M. Bauchinger)

Scoring of translocations by fluorescence in situ hybridisation (FISH) a new method for biological dosimetry

Scoring of dicentrics in human lymphocytes is still the most sensitive technique to quantify acute individual exposures to radiation. However, the precision of dose estimations of old exposures is limited with this approach since with increasing time after exposure a gradual decrease in the frequency of cells containing such unstable aberrations has been observed in several studies. Scoring of symmetrical translocations which have no selective disadvantage would be more efficient to estimate doses of previous radiation exposures. With the recently developed assay of fluorescence in situ hybridisation (FISH) with composite whole chromosome-specific DNA libraries this aberration type can be efficiently scored in human lymphocytes.

In the current programme, comprehensive studies on the dose-response relationship of radiation-induced stable translocations in vitro are planned. FISH will be performed with a triple combination of painting probes for human chromosomes 1,4 and 12.

Representative calibration curves for translocations induced by radiation qualities and dose rates relevant for radiation protection purposes will be generated. The triple combination of chromosomes 1, 4 and 12 comprises only about 20% of the total human genome. In comparison to conventionally established dicentric data from complete cells it has therefore to be evaluated whether this assay provides dose estimations with a similar precision.

Other chromosomes combinations must be also studied to test for a possible variability of the sensitivity of particular chromosomes or even specific chromosomal regions.

A further aim is to achieve a precise discrimination between stable and unstable aberrations. This is a major prerequisite to avoid that scoring is influenced by cell selection due to cell proliferation. For this reason the application of a two-colour FISH with composite whole chromosome-specific DNA probes and degenerate alpha-satellite pan-centromeric DNA probe will be used that allows a precise discrimination between symmetrical translocations and dicentrics (asymmetrical translocations).

Whereas this project deals exclusively with the microscopic analysis of radiation-induced chromosomes aberrations, the other participating institutions are dealing with the development of automated image analysis or flow cytometric techniques for a rapid detection and assessment of these changes. Similar as with the application of the conventional dicentric assay or the micronucleus assay, microscopic scoring of translocations of FISH can contribute to an evaluation of these automated techniques.

Contribution of MRC

Automatic detection of radiation damage to human chromosomes using in situ hybridisation

Outline: It is proposed that random damage to human chromosomes following exposure to ionising radiation shall be related to dose by the detection and measurement of fluorescence hybridisation signals from DNA probes. Each of the strategies described is targeted at detecting a proportion of the chromosome damage through recognition of either dicentric chromosomes or exchanges involving specific groups of the largest chromosomes. Both automated fluorescence microscopy and fluorescence activated flow cytometry will be developed using probes which detect the alphoid sequences at chromosome centromeres, probes which detect the telomeric sequences on the ends of all chromosomes, and unique libraries of probes for chromosome painting.

Counting dicentric chromosomes: The relationship between radiation dose and the resulting proportion of dicentric chromosomes observed at the first metaphase of cell division is an internationally accepted dosimetry standard. We propose here to use fluorescence microscopy in a manner making it possible to combine images of the DNA outline and highlighted images of centromeres and telomeres of metaphase chromosomes in order to discriminate dicentric chromosomes and acentric fragments from clusters of one or more normal chromosomes. The ability to search for a confirming fragment in the same metaphase as a potential dicentric should make the analysis yet more specific. Work on this approach will be spent on developing labelling and detection regimes which clearly separate centromeres and telomeres of metaphase chromosomes in the presence of a DNA fluorochrome. This will involve experiments with fluorochrome excitation and emission systems and software methods for registration and analysis of three colour images. It will also be necessary to adapt existing automatic metaphase finding technology in order to detect fluorescent metaphase.

Sorted dicentric chromosomes: Here, the performance of dicentric chromosome sorting at Amsterdam will be tested with the fluorescence hybridisation and imaging system at Edinburgh following the deposition of suspected dicentric chromosomes on to a microscope slide.

Chromosome painting: A chromosome painting regime, involving the largest human chromosomes, will first be tested by manual fluorescence microscopy for viability as an indicator of chromosome damage resulting from radiation exposure. This approach will involve scoring chromosome painted regions smaller in size than the expected large painted chromosomes and will not require recognition of chromosome shape, centromere or aggregation. The simplicity of this approach, if it is successfully verified, will easily transfer to the automatic microscope, where in the routine laboratory, metaphase searching and painted chromosome analysis should proceed with little operator intervention. Later on, the addition of centromere and telomere probes, which would add two further fluorescence colours to the analysis, would enable the separation of unstable dicentric chromosomes from stable (and longer lasting) balanced translocation.

Commercial chromosome painting libraries will be used for this project when it is reliable and economic to do so. Experiments will begin using a learning set of irradiated human metaphase cells which have been carefully scored for damage by trained cytogeneticists. The automatic approach will be developed using this data before being applied to a new set of data for which manual scoring results are available only for the measurement of performance.

Flow cytometry: Here it is proposed to test several methods to obtain in situ hybridisation signals on isolated metaphase chromosomes for flow cytometry. Previous published methods result in aggregation and loss. In situ hybridisation and primed in situ hybridisation (PRINS) will be investigated either using metaphase inside the cell membrane or using chromosomes separated in agarose beads. Long standing proposals to attain the painting of chromosomes in suspension have made slow progress, yet this remains an attractive target since success could lead to a very rapid method for dose measurement.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

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Description of research work

Introduction

The induction of chromosomal aberrations is used as one of the basic data bases for judgements made in radiological protection. It contributes to the judgements of radiation quality and to the extrapolation of radiation risk to low doses and to low dose rates. In many instances chromosome damage is believed to be an early step in the long sequence of events which subsequently lead to neoplastic transformation of cells. This project aims to provide more detailed information on the radiation quality dependence of chromosome aberration production and a better understanding of the production of such aberrations, both in the first cell cycle and after many cell divisions. All projects described here have developed from results obtained from work previously funded by CEC.

State of the art

Chromosomal aberrations in human lymphocytes following irradiation in the G_0 phase of the cell cycle have been measured for many years. With regard to the quality dependence of the yield, results exist for x rays, γ-rays, several neutron energies, natural α-particles, track segment protons, helium-3 and neon-20 ions. The crucial track segment data covering the important LET range from 10 to 200 keV μm^{-1} are lacking. The PCC technique of fusing lymphocytes in G_0 with mitotic chinese hamster cells to produce prematurely condensed chromosomes is relatively new and used routinely in few laboratories. It enables the timescale of repair of chromosomal breaks to be studied. This gives more information concerning the mechanism by which visible chromosome aberrations at the first metaphase are formed. Data exist for low LET radiation but are lacking at high LET. Recent observations, also in human lymphocytes in G_0 have shown a difference in the ratios of aberration types induced by high and low LET radiation. As LET increases the complexity of the exchange increases and aberrations involving 3 or more chromosomes become more common. After high LET radiation chromosome breaks are found more commonly than exchange type aberrations. After cell division the frequency of cells carrying such lesions decreases by about 30% per division. Similar differences in the ratio of exchanges to breaks have also been shown using mammalian cell lines.

Besides these significant differences at the first metaphase following irradiation there is some evidence of effects occurring after many cell cycles which appears to be specific to high LET radiation. Khadim et al (1992) and Sabatier et al (1992) have observed that cells which survive high LET radiation transmit to their daughter cells instabilities which lead to an occurrence of new chromosomal aberrations many cell cycles later. There is some evidence that these instabilities do not occur at random but are concentrated on a few chromosomes at specific sites.

The observation of chromosomal instabilities following heavy ion irradiation of human fibroblasts has lead to the following scheme.

1. Irradiation by heavy ions induces multiple forms of chromosomal aberrations proportional to particle fluence.

2. The damaged cells, containing unstable aberrations, do not give rise to viable descendents which results in the observation of mostly normal karyotype metaphases after 5-10 passages.
3. A few passages later some cells exhibit damage which had previously been undetected and thought to be transmitted by an instability.
4. This instability appears preferentially on some chromosomes and in human fibroblasts the telomeric regions of chromosomes 13, 16 and 1p are implicated.
5. Some clones containing chromosomal rearrangements and imbalances progressively develop and invade all the cultures after approximately 25 passages.

These observations seem to mirror the clinical advance of solid tumours and leukaemias in humans. In human solid tumours, chromosomal instability and exchange aberrations are generally observed in premalignant or low grade malignancies (Kovacs et al, 1988; Mandahl et al, 1985; Patak et al, 1988; Aledo et al, 1988). Clonal rearrangements leading to typical imbalances are detected in more advanced malignant tumours. This is true, not only for human solid tumours but also for acute non lymphocytic leukaemia secondary to chemo or radiotherapy (S-ANLL). In these S-ANLL deletions occur during leukaemogenesis, several months or years after exposure to the genotoxic agent and thus are obviously not directly induced. These anomalies are not random, affecting chr 5,7,11,17 recurrently (Mamuris et al, 1989, Dutrillaux et al, 1989). Thus in this pathology, the chromosome imbalances appear to be a long term consequence of the exposure to a mutagen. Nothing is known about genetic or chromosomal lesions occurring in radiation induced human solid tumours.

The overall aims of the project are as follows. The first is to understand more fully the process by which double strand breaks are converted to first metaphase chromosomal aberrations and also to provide basic data for testing models of biological effect. The second is to investigate and understand the cellular consequences of the induced chromosomal instabilities.

Description of the project

The project is in two parts. In the first part human lymphocytes will be irradiated at GANIL to score chromosomal aberrations at the first metaphase and PCC fragments and dicentrics at times soon after the irradiation when repair is only partially complete. In the second part, human fibroblasts and CHO cells will be used to follow the disappearance of aberrations after cell division and the subsequent appearance of aberrations caused by transmitted instabilities.

Thin samples of blood (for conventional aberration analysis) and separated lymphocytes (for PCC analysis) will be prepared and exposed to known doses of carbon-12 ions. About 8 doses will be selected to give reasonable dose effect relationships. Experience has shown that no more than one ion energy can be accommodated in any one visit to an accelerator and that no more than two visits can be made in a period of 17 months which is the period of the contract. Consequently the project is limited to two particle energies. The range of LET to give measurable yields has been determined to be 10-200 keV/μm. This limits the ions that can be used to ions lighter than neon. GANIL are able to generate carbon ions but the energy range is restricted to 20-100 MeV per nucleon covering the LET range from about 30 to 100 keV/μm.

After irradiation lymphocytes will be cultured, metaphase spreads prepared and scored according to standard techniques. Standard PCC preparations will be made from the isolated lymphocytes immediately after irradiation and, following fusion with mitotic CHO cells, scored for excess fragments. The chromosome analysis will be done at NRPB and the PCC analysis at Leiden. Analysis of the results will be done at NRPB.

At present very little information is available on the repair of DNA damage induced by charged particles. Staff at Leiden will study the kinetics of repair of induced chromosome breaks by fusing irradiated lymphocytes with mitotic CHO cells to create PCCs, at different time points from zero time and determine the decay of the chromosomal fragments as an indication of repair. They also plan to interfere with repair processes by using specific DNA repair inhibitors (such as ara C, and aphidicolin) to estimate the influence of these inhibitors on the yield of fragments in PCCs. If enough mitotic preparations become available, they will employ chromosome painting to determine the frequencies of induced translocations.

NRPB will co-ordinate the study and will arrange with staff from the French National Institute for Nuclear Physics (IN2P3) for the irradiations at the GANIL accelerator at Caen in France. Responsibility for dosimetry will lie partly with IN2P3 - GANIL and partly with NRPB. NRPB will score metaphase spreads for chromosomal aberrations and LEIDEN will score for PCC fragments. Analysis of results will be done at NRPB.

CIR from Paris will study seven clones of human fibroblasts, each known to contain a transmissible radiation induced chromosomal instability using cytogenetic and molecular techniques. The cell lines will be cultured until senescence or immortalisation is reached. By this means the qualitative objectives of the project will be achieved. However, it is difficult to use this cell line for quantitative studies because of the slow proliferation of human fibroblasts in primary cultures. To test the effect of energy, atomic number and fluence of the particles a more rapidly dividing cell line will be used by G.S.I. The CHO cell system will be used for this purpose with the intention of culturing to the 40th generation. In order to test the specific nature of sites of instability, which has so far been observed in human cells only, a hybrid cell system will be used. This is a CHO cell with a single human chromosome. Using the fluorescence in situ hybridisation technique (FISH) it is possible to identify the number and positions of breaks on the human chromosome.

References

Kadhim, MA, MacDonald, DA, Goodhead, DT, Lorimore, SA, Marsden, SJ and Wright, EG. Nature 355, 738-740 (1992).

Sabatier, L, Dutrillaux, B and Martins, B. Nature 357, 548 (1992).

Kovacs, G, Muller-Brechlin, R and Szucs, S. Cancer Genet. Cytogenet. 32, 93-101 (1987).

Mandahl, N, Heim, S, Kristofferson, V, Mitelman, F, Rooser, B, Rydholm, A and Willen, H. Hum. Genet. 71, 321-324 (1985).

Patak, S, Wang, Z, Dhaliwal, MK and Saks, PC. Cytogenet. Cell Genet. 47, 227-229 (1988).

Aledo, R, Avril, MF, Dutrillaux, B and Aurias, A. Cancer Genet. Cytogenet. 33, 29-33 (1988).

Dutrillaux, B, Mamuris, Z and Aurias, A. in Workshop on cell transformation system relevant to radiation induced cancer in man. Dublin 5, 169-175 (1989).

Mamuris, Z, Prieur, M, Dutrillaux, B and Aurias, A. Cancer Genet. Cytogenet. 35, 65-77 (1989).

Contribution of the NRPB (Chilton, UK)

The role of NRPB is to co-ordinate the human lymphocyte study and liase with staff at IPN and GANIL to arrange beam time and specify particle fluences in order to determine doses. At NRPB, suitable sample holders to suit the beam characteristics will be made. At the GANIL accelerator, samples of blood will be prepared and irradiated. Blood lymphocytes will be cultured and metaphase spreads will be prepared for about 8 doses. Microscope slides will then be taken back to NRPB for staining and microscope analysis, scoring for dicentrics, centric rings and acentric fragments. An analysis of the dose effect relationship in terms of a linear or linear-quadratic equation principally to determine the linear coefficient α. This will then be compared with measurements of α using other radiations.

Contribution of the University of Leiden

The role of Leiden is to attend GANIL at the time of irradiation and prepare samples of separated lymphocytes. The lymphocytes will be fused with mitotic CHO cells to create PCCs, at various times following irradiation. It is planned to use specific DNA repair inhibitors to interfere with repair processes to estimate their influence on the yield of fragments in PCCs. Microscope slides containing PCCs will be prepared at GANIL and taken to Leiden for staining and microscope analysis. If sufficient mitotic preparations become available chromosome painting techniques will be used to determine frequencies of induced translocations. In the seventeen month period of the contract, two separate visits to GANIL will be made to use two different ions.

Contribution of IPN (Paris) and GANIL (Caen)

i) The technical staff from the GANIL accelerator will provide the heavy ion beams and the standard equipment of the beam line (vacuum pump, beam diagnostics...)

ii) A team composed of about ten physicists, engineers and technicians from the French National Institute for Nuclear Physics and Particle Physics (IN2P3) and from the GANIL will be in charge of the physical aspects of the experiments (prepare a homogenous beam over the required area, determine the particle energy and fluence with the required accuracy, and participate, with NRPB, in the conversion of these data to dose).

Contribution of CIR (Paris)

This study is to follow the induction and the transmission of radiation induced chromosomal rearrangements in human fibroblasts. Seven irradiated cultures have been performed: Neon ions (E=10.74 MeV/u, LET=386 keV/μm), (fluences of 10^6, 2.10^6 and 4.10^6 particles/cm^2), Argon ions (E=10.52 MeV/u, LET=1207 keV/μm), (fluences of 10^6, 2.10^6 and 4.10^6 particles/cm^2) and Lead ions (E=9.5 MeV/u, LET=13600 keV/μm), (2.10^6 particles/cm^2). Karyotypes were studied each 5 passages up to 25 passages (about 50 cell divisions). This showed increased life span since the control culture could be prolonged to 20 passages only. Radiation induced lesions were observed at the first passage studied, decreased and totally disappear around passage 10. However, after the 15th passage, a chromosome instability appeared involving chromosomes 1, 13 and 16 in multiple telomeric associations or dicentrics. When our cultures were prolonged clonal chromosome imbalances were noticed between the 20 and 25th passages, in particular a monosomy 13, this chromosome being the most frequently involved in telomeric associations.

We shall continue this study by 1) prolonging the cultures, 2) look for a possible relationship between the losses of chromosome 13 and of the expression of RBI gene, which is a major tumour suppressor gene from chromosome 13, 3) performing new irradiations to confirm our first findings and to investigate a relationship between the LET and the observed effects.

Technical approach

Cytogenetics

- cell culturing of human fibroblasts until senescence (or immortalization)
- cytogenetic study, using R-banding, every 5 passages, to follow the appearance of chromosome instability and the formation of clonal rearrangements
- in situ hybridization to search for eventual translocation of chromosome 13, and in particular of the RBI (retinoblastoma) gene, when one chromosome 13 is missing
- in situ hybridization of telomere specific probes to the type of instability (telomeric associations or dicentrics = TAS or DIC)
- in situ immunodetection of 5-methylcytosine antibodies
- NOR staining, to study the activity of the nucleolar organisers where the short arm of chromosome 13 is involved in TAS or DIC.

Molecular biology

- preparation of probes for in situ hybridization
- northern blotting to look for a possible defect of the expression of the RBI gene
- southern blotting to look for possible loss of heterozygosity, and gross structural rearrangement of the RBI gene.

Contribution of G.S.I. (Darmstadt)

Based on recent experiments we shall focus on the processes which occur between the induction of radiation damage seen at the first postirradiation mitosis up to the 40th cell generation. We shall use an established hamster cell line (CHO cells) characterised by a much shorter cell cycle time (12 hours) than the slowly proliferating human cells (cell cycle time: 48 hours). After exposure the cells will be subcultured every 2 to 3 days corresponding to 4 to 6 cell generations. The exact number of cell divisions will be recorded and chromosome analysis will be performed. The studies using CHO cells will last only for about 4 to 6 weeks instead of 6 to 7 months for the human cell system. Only by the use of a fast proliferating cell system like the hamster cell line will it be possible to study the dependence of late radiation effects from particle parameters like atomic number, energy and particle fluence within a reasonable time.

Secondly, we shall test the hypothesis of a nonrandom pattern in chromosomal instability - at least in human cells. Specific chromosomes were mainly involved in the late appearance of aberrations in humans but not in murine cells. For this study, a hamster-human hybrid cell line (CHO-cells containing one human chromosome) will be used. This cell line has the same generation time as the parent CHO cell line. With a novel technique, the fluorescence in situ hybridization, it is possible to identify the human chromosome and count the number of breaks or illegitimate fusions between hamster and the human chromosome. The number of breaks induced in this specific human chromosome will be compared to the amount of damage induced in hamster chromosomes of the same size.

B13 Cellular, molecular and animal studies to determine the risk of stochastic somatic effects of radiation with respect to low dose, low dose rate and radiation quality.

Contract FI3P-CT930067 Development and investigation of systems for the quantification of radiation induced carcinogenesis in humans.

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Total Contribution by the Commission: 190 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Introduction

Quantitative estimation of cancer risk is a major problem in radiation protection, especially at low doses. There is very little data available on the consequences of low dose exposure to human populations. Extrapolation must be made from existing high dose data, from animal experiments and from in vitro transformation data using rodent C3H 10T1/2 cells. The development of new systems using human epithelial cells is essential if the risks of cancer induction are to be quantified in a human population.

The partners in this proposal will thus focus on developing suitable human epithelial culture systems which could provide quantifiable end-points for evaluation of radiation-induced transformation. The human epithelial culture systems to be investigated will include primary cultures of gastrointestinal tract, skin, breast and ureter; established human cell lines of bronchial epithelial cells, keratinocytes, urothelial cells and thyroid cells; and organotypic cultures of skin. There is also an increasing recognition of the need to fully assess age-related changes in radiosensitivity and the risk of radiation exposure in utero and in early childhood from all sources of radiation. Concern centres in particular on the induction of childhood leukaemia and the radio-sensitivity of the brain during in utero development. With regard to effects on the brain, epidemiological studies have shown an increased incidence of brain tumours as well as mental retardation in irradiated populations after in utero exposure. Because of the severity of these tumours, this risk requires further consideration. Studies with rats have shown that in utero and early neonatal irradiation results in central nervous system tumours in adults which are not observed after irradiation at later stages. Most of these tumours are of glial origin.

To predict adequately the likelihood of particular effects on human populations exposed to radiation at low doses and dose rates, the requirement for an understanding of the cellular and molecular mechanisms involved is increasingly clear.

Evidence of progression to the tumour phenotype will be established using xenografts in athymic nude mice. Another approach will be to identify or develop in vitro markers indicating advancement along the carcinogenesis pathway. These will involve molecular biological approaches, including amplificaton of potential marker gene sequences using the polymerase chain reaction, DNA sequence analysis and in situ hybridisation. Studies will be made of p53 and RB mutation and allelic deletion. In addition, changes in the expression of growth factors will be studied and a search for novel transformation related proteins and their corresponding genes made. The growth patterns of cells in soft agar and varying serum concentrations will be defined to indicate progression towards carcinogenesis.

This contract, therefore, aims to examine the development of quantitative epithelial cell culture systems for studying radiation induced transformation in humans and to compare radiation induced transformation with that induced by chemicals in adult human epithelial and fetal glial cells with a view to addressing the problem of radiation induced transformation in adult and fetal tissues.

The emphasis in the overall project is to develop systems using primary cultures of cells from adult and fetal organs which retain differentiated properties of the tissue of origin. These systems can then be used to answer specific questions concerning the carcinogenicity of radiations of various qualities or administered at various dose rates. They can also address the comparative risks of radiation and carcinogenic chemicals. The problem of producing results which allow transformation frequency to be quantified is central to the project, as is the focus, where possible, on fresh material of human origin.

Objectives and Expected Achievements

The main objective of the project is to define suitable epithelial and glial culture systems which will allow the investigation of the cancer risk to humans from exposure to low doses and dose rates of ionising radiations. The objective will be pursued by:

(1) identifying and establishing suitable human cell lines for radiation-induced transformation studies;

(2) continuing animal experiments using pregnant rats, where high levels of central nervous system tumours in adult offspring can be obtained;

(3) identifying suitable end-points for quantitation of transformation and for elucidating the steps in the process, using molecular biological approaches to define transformation markers and growth factor expression;

(4) linking end-points defined using in vitro clonogenic assays, in vivo xenograft growth and markers.

It is expected that at the end of the 17 month contract period, the proposers will have defined suitable systems to investigate transformation of human epithelial and rat glial cells and provided an approach to quantification of the responses of radiation using a range of defined end-points. This will provide the basis for future studies relevant to radiation protection and radiation carcinogenesis in man.

Complementarity between participating organisations, expertise to be contributed and benefits to be gained by collaboration

The five partners in this proposal each bring unique skills to the overall problem of the development of a quantitative human radiation transformation system. All are already collaborating with at least one other partner and exchanging material for assay. Each partner has a unique culture system to examine, a unique assay service, or material to provide to the other partners. The benefits of this collaboration are that it provides a rapid means of assessing the usefulness of a number of primary cell systems and transformation markers without the need for the develop-ment of each potential assay system in one laboratory.

The specific contributions of each laboratory are as follows.

DIT Kevin Street will culture primary human urothelium and produce abnormal foci. They can handle the HPV cell line and supply irradiated cultures to other partners. The group can also do immunocytochemical analysis, in situ hybridisation, and routine histological examinations, for markers of interest to the group.

St. Andrews University will culture SV40 transfected HUC-1 and HTori lines and has facilities to test cells for tumour growth in nude mice. They can recover cell lines from tumours and perform marker analysis and histology.

Gray Laboratory will PCR, amplify and sequence the p53 gene in supplied material and can grow bronchial epithelial cells. This group has extensive facilities for irradiation, including chronic low dose ^{60}Co and α irradiation.

CEN-Saclay will carry out p53 protein expression analysis, and will determine if protein levels correlate with radiation dose. They will culture HaCaT cells and skin keratinocytes from human biopsy material on collagen in a skin equivalent system.
CEN-FAR will culture fetal glial cells obtained from irradiated pregnant rats and can supply cultures and tissues at various stages of brain development for analysis of relevant markers.

Contribution of the Dublin Laboratory

The work to be carried out by the Dublin Laboratory is a continuation of the previous contract to develop a quantifiable end-point for radiation carcinogenesis using human cells. The proposed work programme for the next 17 months is as follows:

1. HPV transfected cells will be irradiated using chronic tritium exposure or X-rays, and passaged (according to protocols established in the last contract) prior to inoculation into nude mice to test for tumourgenicity. The nude mouse work will be undertaken in collaboration with St. Andrew's University.

2. Cultures at each passage during the above experiments will be tested for cloning efficiency and for mutations in the p53 gene in collaboration with the Gray Laboratory.

3. Primary epithelial cultures will be established and exposed to tritium or to X-rays. The frequency of immortalised cmyc positive foci of primitive carcinoma-like cells will be quantified and related both to the number of cells at risk in the original explant and to the number of cells which survive the radiation exposure. Techniques enabling this information to be obtained were developed during the last contract.

4. We have recently detected mutant p53 protein in a small number of discrete foci from urothelial cultures exposed to two doses of irradiation 5 days apart, and maintained for several weeks. A programme of experiments to determine the nature of the mutation and its frequency under different conditions of radiation exposure will be undertaken in collaboration with the molecular biology groups at the Gray Laboratory, Saclay and CEN-FAR to define and further explore this result.

Contribution of the St. Andrews Laboratory

The proposed work to be carried out at the University of St. Andrews will be performed by Dr. Andrew Riches and Dr. Peter Bryant and is a continuation of work in progress on radiation-induced carcinogenesis of human epithelial cells. The approach to studying neoplastic transformation in human epithelial cells will be to utilise the human urothelial cell lines isolated after immortalisation with SV40 by Dr. Catherine Reznikoff and the human thyroid epithelial cell line isolated after immortalisation with SV40 origin minus by Dr. David Wynford Thomas. The tumorigenic potential of these cells will be monitored in nude mice and cell lines rederived from these for further testing. Results of studies so far indicate that nodules containing human cells can be detected in nude mice following transplantation of irradiated cells.

The proposed work will utilise the human urothelial cell line (SV-HUC-1) and the human thyroid epithelial cell line (HTori-3). The dose-effect relationships for survival and micronucleus production in cells irradiated with different types of radiation will be compared. Changes in cell morphology, clonogenicity, p53 expression and in vitro growth will be defined after exposure to irradiation. The transformation responses of single and fractionated doses of gamma and alpha irradiation will be compared.

Evidence for transformation of irradiated cells will be monitored by passaging into nude mice and observing tumour formation. Human keratinocytes from the Dublin group will also be tested in our laboratory. Both normal and irradiated, transformed human urothelial and thyroid cells will be analysed in the partner laboratories. The tumour suppressor gene p53 status and the analysis of mutations will be undertaken at the Gray Laboratories by Dr. Janet Arrand and Dr. Mike Joiner. The expression of selected growth factors and oncogenes in irradiated human cells will be evaluated by Dr. Michèle Martin at Saclay. A collaborative chronic low dose rate experiment will be undertaken jointly at the Gray Laboratory using human cell lines from the partners.

Contribution of the CEA-Saclay Laboratory

The contribution of the LRA will be performed by Dr. Michèle Martin and Dr. Denis Biard, as part of a new programme developed in the laboratory on skin cell transformation.

We propose to develop for the group the use of p53 gene expression as a marker of mutations in p53 gene, and consequently of radiation-induced cell transformation in epithelial human cells.

Recent publications demonstrate that normal cells, which are wild type for the p53 gene, have a low endogenous expression of p53. DNA damaging agents induce in these cells an increase in the level of the protein, which is concomitant to a G1 arrest of the cell cycle. These processes probably prevent the replication of a damaged DNA template. On the contrary, cells with mutant p53 genes exhibit high endogenous levels of p53 proteins, no increase in protein amount after exposure to DNA damaging agents, and no arrest in G1.

We will assess the use of variations in the level of p53 protein as a marker of mutations induced by ionizing radiation in the p53 gene. As gene sequencing will be performed concomitantly on each cell lines, we will test whether this assay could be predictive, and could thus be used to screen for mutant cells.

Alterations of the protein levels will be quantified by immunoprecipitation and Western-blotting. Alterations of the RNA levels will be studied by Northern-blotting. The G1 arrest and variations of p53 protein level as a function of the cell cycle will be studied using a flow cytometric assay and specific antibodies.

The assays will be performed on epithelial cells before irradiation, immediately after high dose irradiation (2GY, ^{60}Co), and after low dose irradiation and selection for abnormal clones. In the laboratory, normal primary keratinocytes and HaCaT cells will be used. The latter are immortal but non-tumorigenic keratinocytes. Cell culture, high dose irradiation and p53 gene expression studies will be performed in the laboratory. Low dose irradiation and DNA sequencing will be performed in collaboration with the Gray Laboratory. St. Andrews Laboratory will study the tumorigenic potential of abnormal clones selected after low dose irradiation in nude mice.

As p53 is a DNA-binding protein, we will further address the role of p53 in the G1 arrest and the type of genes that this transcription factor can regulate, such as repair genes or growth factors.

Contribution of CEA-FAR Laboratory

Comparison of ex vivo transformation of rat fetal glial cells induced by radiation or chemicals

Experimental data and epidemiological studies show that there is a period of radiosensitivity during *in utero* development for Central Nervous System (CNS), in particular glial cells. Irradiations of pregnant rats induce an increase in incidence of CNS tumours in progeny, this is not observed when adult rats are irradiated.

The objective of this study is to provide information on the sequence of events occuring during *ex vivo* transformation of fetal glial cells. Comparison of radiation and chemically induced transformation may help elucidate factors affecting this process.

Rat fetuses will be treated *in utero* with ethylnitrosurea or irradiated with ^{60}Co. Cultures will be initiated with glial cells isolated from rat brains. These cells will be treated with TPA to try to accelerate the transformation process. Appropriate controls will be studied in parallel.

The following paramters will be studied to characterise these cells: morphology, proliferation, clonogenicity, tumorigenicity, expression of oncogenes and differentiation markers.

Besides providing informations about cell transformation, this project represents a new approach to the study of radiosensitivity of developing CNS and brain tumours induction.

This proposal is complementary with proposals from other participants which are focused on human epithelial cells. As work progreses, comparison of human and rodent cells could be interesting and convergence between the projects should become more evident.

Contribution of the Gray Laboratory

The Gray Laboratory possesses the only Dynamic Microscope Image Processing Scanner (DMIPS) in Europe; the use of this sophisticated computer controlled microscope overcomes the statistical uncertainty in the number of cells plated, which limits the resolution of conventional colony forming assays, since individual cells can be located and revisited and their life history followed to determine viability following exposure to very low doses of ionising radiation. Using this machine, we have shown that two of the epithelial cell systems used as radiation transformation targets by partners in this contract (human lung epithelial cells, L132, and human urothelial cells (SV-HUC-1) exhibit hypersensitivity to killing by low X-ray doses (<0.5 Gy) compared to higher doses, 2-D gel analysis of proteins from unirradiated cells when compared to those from cells irradiated with 0.5 Gy, showed differential expression or post-translational modification of a number of proteins suggesting that the effects observed on cell survival at low doses may be due to inducible repair. We will extend these studies to the epithelial cells being used as radiation targets by our other partners as required.

We are now attempting to correlate the cell survival end-point with mutation frequency at low doses and dose rates; a low dose rate facility has recently been commissioned for this project at the Gray Laboratory. This facility will also be available for low dose rate transformation studies on all the epithelial cell targets proposed for use in this contract and the results of ongoing low dose rate survival and mutation assays will enable us to define the parameters for the transformation experiments. In addition, we have immediate in-house access to an unparalleled range of radiation sources for use by all partners in this contract group. This will ultimately facilitate studies of the dependence of transformation and the appearance of molecular markers on LET.

Our continuing analysis of low ionising radiation dose effects on human epithelial cells concentrates on the identification of differentially expressed proteins and the isolation of the coding gene sequences from in-house cloned cDNA libraries made from irradiated and unirradiated cells. Some of these differentially expressed proteins may prove to be useful as early transformation markers.

We propose to test the correlation of transformation with alterations in such marker genes and will initially concentrate our efforts on identifying the status of the tumour suppressor p53 gene and its product in the target cells used by each contract partners using PCR/DNA sequencing and immunofluorescence/FACS. As radiation-transformed derivative cell lines are generated, these will be included in the analysis to identify genetic alterations. Such studies can be extended to the RB gene and others identified as potential tumour progression markers during the course of this and future contracts.

B14 Assessment of genetic risks in man.

Contract FI3P-CT920005 Radiation-induced genetic effects in mammals and the estimation of genetic risks in man: a concerted approach using theoretical, epidemiological, cytogenetic, biochemical and molecular methods.

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Total Contribution by the Commission: 175 KECU
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Description of research work

Introduction.

Genetic effects of ionizing radiation are those associated with mutations and chromosomal aberrations induced in the parental germ cells and transmitted to the progeny. Some of these induced changes can lead to one or another kind of genetic disease (Mendelian, chromosomal or multifactorial) in the progeny of those exposed. Owing to the paucity of direct human data on radiation-induced germ cell mutations and chromosomal aberrations, animal data from radiation genetic studies are used together with human epidemiological data on naturally-occurring genetic diseases to estimate genetic risks. While the methods and risk estimates themselves have evolved over the years with additions to knowledge in this multidisciplinary area, data are still insufficient for an assessment of the risk of chromosomal and multifactorial diseases and uncertainties remain with respect to the estimation of genetic risks to females.

Objective.

The main objective of this project is to strengthen and expand the conceptual framework, data-base and methods relevant for genetic risk estimation through concerted efforts. The items on our agenda are: assessment of chromosomal damage in mouse germ cells using molecular cytogenetic techniques, studies on the induction of congenital abnormalities (scored in the F_1) following low dose-rate gamma irradiation of mice, further studies to assess the suitability of the guinea pig female as a model for cytogenetic radiosensitivity of human resting oocytes, exploration of potentially useful model systems to study induced germ cell mutation effects on multifactorial traits in mouse and man and finally, development of theory, models and methods to make risk estimation for multifactorial diseases in man.

A. Radiation-induced chromosomal aberrations in mouse germ cells.

In these studies which will be carried out at the MRC Radiobiology Unit, Chilton, England, the *fluorescent in situ hybridization* (FISH) technique will be employed. In the initial phase of the mouse work, repeat sequence DNA probes for the X and Y chromosomes will be used to screen mature spermatozoa and paternally-derived pronuclei of one-cell embryos for aneuploidy. The sensitivity of the assay will be examined through a compa-rison of the rates of anomalous spermatozoa and embryos in three groups of males: chromosomally normal controls; males hemizygous for an X-autosome Robertsonian translocation that increases the rate of sex-chromosome non-disjunction and, males that had received an X-ray dose known from published studies to increase the frequency of sex-chromosome loss. These studies, which will be carried out at the MRC Radiobiology Unit in Chilton, England, should provide an initial assessment of the sensitivity of the FISH technique as an assay for radiation-induced chromosomal damage in germ cells. Subsequent experiments will concentrate upon analysing dose and dose-rate effects, strain and sex differences in response and are expected to provide data which can be compared with those generated in conventional cytogenetic experiments.

B. Induction of congenital malformations by low dose-rate gamma irradiation of mouse germ cells.

In this part of the project, to be carried out in Essen, the question addressed is whether and to what extent, low dose-rate gamma irradiation of mouse germ cells will produce malformations scorable in F_1 fetuses. The germ cells to be studied will include: immature oocytes in female mice (the predominant cell population in the mammalian ovary), developing oocytes (in fetuses of pregnant mice) and of different stages of spermatogenesis in male mice. Immature oocytes will be irradiated at a dose-rate of 0.016 Gy/h, which is 25-fold lower than the one used in earlier work (0.4 Gy/h); in the earlier work, these oocytes had been found to respond mostly with lethal events. For developing oocytes and spermatogenic stages in male, again, low dose-rate irradiation will be used. In addition to the scoring of malformations, tissue material from both malformed and normal fetuses will be used in biochemical assays to screen for alterations in protein pattern.

C. Development of model systems for genetic risk assessment.

This part of the project will be carried out in Mol and Leiden. In the work at Mol, the focus is on the development and vali-dation of the female guinea pig as a model for asceraining the chromosomal radiosensitivity of human immature oocytes. From earlier studies, on reproductive performance (among others), it is clear that the guinea pig oocytes are indeed more resistant to cell-killing than mouse oocytes; it is very probable that the guinea pig oocytes may be more comparable to the human oocytes in this regard. In favor of this assumption is the finding that the "large" resting oocytes of the guinea pig are morphologically similar the human resting and maturing oocytes. Oocytes from irradiated guinea pigs will be cultured to the first meiotic metaphase and then processed for cytogenetic analysis and scoring of chromosomal aberrations (fragments, translocations etc). The emphasis will be on the large resting oocytes. Additionally, the possible influence of pregnancy on chromosomal radiosensitivity of the adult "contracted" resting oocytes will also be determined, complemented by cytogenetic analysis of somatic cells.

In Leiden, we propose to initiate work to ascertain the genetic basis of specific multifactorial conditions in humans to explore their potential utility as models for multifactorial conditions and risk assessment. These include cleft lip/palate and intrauterine growth retardation [resulting in "low birth weight" babies] in humans and dwarfism or "runting" in mice. The stimulus for the work on intrauterine growth retardation comes from the studies of Czeizel and colleagues in Budapest which show that babies born to parents several months or years after self-poisoning with high doses of chemicals (in suicide attempts) have a significantly low birth weight, possibly due to the induction of germinal mutations which have adverse effects on intrauterine growth. We will use known human cloned growth factor genes in RFLP analysis of DNA from low birth weight babies. In the mouse, there are sufficient data to suggest that a higher proportion of offspring from mutagen-treated parents were in fact detectably small at weaning.

The material (blood or other tissue samples) for the mouse work will be provided by Dr. J. Favor (Neuherberg) and we envisage using, initially, the pit-1 gene in mouse as the molecular probe in the RFLP analysis. The general rationale is that, when once the genes involved are identified, this information can be used both for developing experimental models and for theoretical investigations (see below, item D) on the risk of multifactorial conditions.

D. Theoretical studies and mathematical modeling for risk estimation for multifactorial diseases.

This part of the work will be carried out in Leiden. The rationale is that little progress has been made with respect to the effects of induced mutations on multifactorial diseases in man (common congenital abnormalities and common diseases of adults; these contribute very substantially to morbidity and mortality and far more than Mendelian and chromosmal diseases together) which in turn is due to the lack of an adequate theoretical framework, lack of precise knowledge on the numbers of genes involved and the paucity of usable experimental results. An extensive analysis of the existing information on human multifactorial diseases will therefore be carried out with particular attention being paid to the interaction effects and mechanisms of maintenance in the population. This will provide the basis for developing mathematical models to inquire into the impact of induced mutations on multifactorial conditions.

Contribution of the collaborating laboratories.

LABORATORY 1. Department of Radiation Genetics and Chemical Mutagenesis, University of Leiden, Leiden, The Netherlands

Theoretical analysis of human multifactorial conditions.

This work will consist of (i) a systematic and comprehensive analysis of our current knowledge on common congenital abnormalities and of other multifactorial conditions (prevalence, aetiology and mechanisms of maintenance in populations); (ii) examination of the suitability of the models (e.g., the multifactorial threshold model) which are currently used to explain their prevalence, calculation of recurrence risks in families etc., and (iii) formulation of models which will permit risk estimation. Using specific and realistic input values to define mutation, selection and threshold, mathematical modeling and computer simulations will be carried out to inquire: effects of changes in mutation rate on disease prevalence, the effects of changes in prevalence on the variance and heritability of the trait and the relationships between changes in mean, variance or both of the distribution on disease prevalence in the population.

Experimental approaches.

Molecular studies using RFLP markers will be carried out to ascertain the genetic basis of selected common congenital abnormalities for which patient and family material (blood samples) can be obtained. We intend to start with cleft lip/palate and depending on progress made, extend these studies to other suitable congenital abnormalities.

Additionally exploratory studies will be performed to examine the involvement of major genes in some specific multifactorial indicator traits --intrauterine growth retardation (resulting in "low birth weight babies") in humans and "dwarfism" in mice. For this part of the work, blood samples will be made available by Dr. Czeizel (Budapest) and we will use known human cloned growth factor genes in RFLP analysis of low birth weight babies. The material for the mouse work will be provided by Dr. J. Favor (Neuherberg) and we envisage using, initially, the pit-1 gene in mouse as the molecular probe in the RFLP analysis.

LABORATORY 2. MRC Radiobiology Unit, Chilton, England

Studies on radiation-induced chromosomal anomalies in mouse germ cells using molecular techniques.

The aims of this part of the work are to study radiation-induced chromosomal aberrations in mouse germ cells using the technique of *f*lourescent *i*n *s*itu *hy*bridzaition (FISH), to explore the strengths and limitations of the technique and to compare the data generated with those accumulated in conventional cytogenetic and genetic assays. The currently available chromosome-specific DNA repeat sequences will be used in these studies.

Initial work will focus on the use of repeat sequence DNA probes for the X and Y chromosome to screen mature spermatozoa and paternally derived pronuclei of one-cell embryos for sex-chromosomal aneuploidy. Sensitivity of the assay will be examined through a comparison of the rates of anomalous spermatozoa and embryos in 3 groups of males: (i) chromosomally normal controls; (ii) males hemizygous for an X-autosome Robertsonian trans-location that increases the rate of sex-chromosome non-disjunction and (iii) males that have received an X-ray dose known from published studies to increase the frequency of chromosome loss. The data generated should provide an initial assessment of the sensitivity of FISH as an assay for radiation-induced chromosomal damage in germ cells. Subsequent experiments will concentrate upon analysing dose- and dose-rate effects, strain differences and inter-sex comparisons to provide data which can be compared with those from conventional cytogenetic studies.

LABORATORY 3. CEN/SCK, Mol, Belgium

Development and validation of the female guinea pig as a model for assessing the chromosomal radiosensitivity of human immature oocytes.

In mammals, the immature or resting oocytes are present from birth throughout reproductive life; they constitute about 90% of the total oocyte population in the mature ovary and are thus the most relevant stage for the study of genetic effects of radiation in females. Mouse immature oocytes which have thus far been studied in radiation work, unlike human resting oocytes, are extremely sensitive to killing effects of ionizing radiation; further, the nuclear morphology of the mouse resting oocyte (dictyate) is also quite different from that of the typical diplotene human resting oocyte.

The extension of radiation studies to an other oocyte system which is more comparable to the human one both in terms of sensitivity to killing and nuclear morphology is therefore strongly indicated. The guinea pig resting oocytes represent such a system, particularly the "large" resting oocytes which are present in the newborn.

The work consists of three parts: (i) improvements of the techniques (developed earlier in our laboratory) to obtain and culture guinea pig oocytes *in vitro* and to make cytogenetic preparations; (ii) investigating the cytogenetic radiosensitivity of the "large" resting oocytes by irradiating newborns; our preliminary results show that chromosomal aberrations can indeed be induced in them, in contrast to the situation known for mouse oocytes and (iii) examining whether the level of gestation hormones in the female (progesterone, oestradiol), as has been recently shown in the mouse (in liver cells from fetuses and bone-marrow of mothers; M. Ricoul and B. Dutrillaux, Mutation Res 250, 331-335, 1991), influences the chromosomal radiosensitivity of the adult, "contracted" resting oocytes.

LABORATORY 4. Inst. Medizinisch Strhlenbiologie, Univ. Essen, Germany

Induction of congenital malformations by low dose-rate gamma irradiation of mouse germ cells.

The impact of radiation exposure of human immature oocytes, especially at low dose-rates, is of concern from the standpoint of genetic risks. The results of earlier mouse experiments in our Institute, as those from similar studies conducted in other laboratories, have confirmed that high dose irradiation (1 Gy/min) of female mice causes almost exclusively killing of immature oocytes. At a lower dose-rate (0.4 Gy/h) however, the cell-killing effect was markedly reduced with a concomitant increase in the frequency of malformed fetuses; this was particularly true of irradiation of very immature oocytes in fetuses.

The work will focus on (i) extending these studies on the radiation response of immature oocytes to an even lower dose-rate, namely, 0.016 Gy/h and to spermatogenic stages in the male and (ii) gaining insights into the mechanisms of induction of malformations by radiation through a comparison of the protein patterns in normal (control) and malformed fetuses. In all these studies, a mouse strain which has been found to respond to radiation exposure (during oogenesis, spermatogenesis and pre-implantation stages) with increased frequencies of malformed fetuses will be used.

B14 Assessment of genetic risks in man.

Contract FI3P-CT920055 Genetic risks associated with exposure to ionizing radiation.

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50 KECU

Description of research work

An adequate understanding of the genetic risk associated with radiation exposure as well as the mechanisms by which mutations may be induced are essential for an informed action to protect mankind from the harmful effects of radiation. To this end our project will scientifically address the strategy of risk estimation based on experimental data in animals as well as to undertake studies to determine factors important in the mechanisms of mutation induction by radiation exposure to germ cells. Researchers specializing in germ cell genetics from six leading European laboratories active in radiation mutagenesis (GSF-Neuherberg, Leiden University, MRC-Radiobiology Unit, Utrecht University, Universitat Autónoma de Barcelona and West Midlands Regional Genetic Services) will participate in a coordinated research effort to improve risk extrapolation procedures as well as to determine the influences of DNA damage repair and cell killing on the levels of mutation induction by radiation. Further, molecular techniques will be employed to assess DNA damage to the genome as well as to molecularly characterize the DNA changes associated with radiation induced mutations. Details of scientific hypotheses to be tested and experimental methods are outlined by the individual participants.

Strategies of genetic risk estimation

Estimates of genetic risk to man resulting from an increased mutation rate due to radiation exposure of necessity must be based on experimental results in animals. Two sets of data are required for a valid estimation of genetic risk to man from results in other species; (1) those based on a number of genetic endpoints, several of which can be applied in several species studied, and (2) those derived from a number of different species, strains or genotypes, based on one or more genetic endpoints. Two methods are available to estimate the human radiation genetic risk: The direct approach estimates the frequency of induced dominant deleterious mutations in man based on experimental results in the mouse for induced dominant mutations. An estimation of the frequency of induced chromosomal aberrations is extrapolated from a combination of results in the mouse and primates. The second approach, the indirect method, estimates the frequency of dominant deleterious mutations in man based on the estimate of the radiation mutational doubling dose for recessive specific locus mutations. Both the direct and indirect methods of risk estimation require the assumption of no species differences in the sensitivity to the induction of mutation by radiation to extrapolate genetic risk estimations in man based on experimental animal data. The indirect approach requires the additional assumption that the doubling dose estimated in the mouse for recessive specific locus mutations is representative for dominant deleterious mutations. Experiments to determine the validity of the above two assumptions will be carried out.

Mechanisms of radiation-induced mutagenesis

The induction and transmission of mutations in germ cells of animals is dependent upon the interaction of the rate at which primary DNA lesions are induced as well as with the rate at which such lesions are repaired, misrepaired or lead to cell death. Differences in the capacity of the various germ cell stages to the repair of induced DNA damage have been demonstrated and correlate with the sensitivity to radiation

induced mutation rates. Further, the shape of the dose response curve in the mouse suggests differences in the sensitivities of the stem cells to mutation induction by radiation, and this may be dependent upon the stage of cell cycle of stem cell spermatogonia. In order to investigate the effects of DNA repair and cell death on the induction by radiation of mutation in germ cells, the sensitivities of the germ cell populations will be manipulated either genetically or via chemical or radiation pretreatment. Mouse strain 101/H has been shown to be radio-sensitive to induced-cell killing as was the mouse dominant mutation steel although the yield of translocations was reduced. Further, chemical pretreatments may alter the sensitivities of the stem cell spermatogonial population to radiation: Triethylenemelamine pretreatment results in a non-specific cell killing of stem cell spermatogonia and leads to an altered dynamics of cell proliferation. Hydroxyurea pretreatment kills any cells in S-phase, so that a partially synchronized spermatogonial cell population exists at the subsequent radiation exposure. Using the vitamin A deficiency/ vitamin A replacement protocol it is possible to synchronize the testis in such a way that high numbers of radiosesistant or exclusively radiosensitive stem cells are present. Finally, 3-aminobenzamide pretreatment sensitizes cells to radiation. These sensitization methods will be applied to determine correlated effects on DNA damage, cell death, as well as mutation and translocation yields.

Radiation-induced DNA alterations

To bridge the gap between DNA lesion induction and mutation fixation, studies will be initiated to screen for radiation-induced deletions and micro-deletions as well as to molecularly characterize radiation-induced mutations recovered in germ cells of mammals. Cattanach and co-workers have recently observed a class of dominant mutations recovered in radiation studies associated with extremely large deletions. An analysis of a larger group of mutations from radiation experiments will be carried out to characterize the spectrum of radiation-induced DNA lesions associated with mutations in germ cells. Complementary to these studies will be a project to screen coding and non-coding regions of the mouse genome via pulsed field gel electrophoresis to determine if such methods are feasible in detecting induced genetic damage directly at the DNA level. Together these studies should provide data required to characterize the nature of radiation-induced DNA lesions ultimately resulting in mutation and thus to elucidate the mechanisms of mutation induction by radiation in mammalian germ cells.

Finally, new methods have been recently developed to determine genotoxic effects in mammals based on transgenic mice. These methods combine the elegance and sensitivities of microbial genetics with the in vivo mammalian situation. The methodologies have not yet been adequately tested. If they prove to accurately reflect the in vivo situation, mutagenicity experiments of much greater precision and reduced costs could be anticipated. Leiden University is intimately involved in the development of these methodologies and will carry out initial studies to determine the biological relevance of such transgenic systems in determining the mutagenic effects of radiation in germ cells of the mouse.

All aspects of the project have been carefully discussed to coordinate experiments including choice of species and genetic endpoint examined as well as genetic and chemical manipulations of germ cell populations to alter radio-sensitivities. Results

derivied from these studies will lead to a better understanding of the problem of cross species extrapolations as well as the interactive roles of DNA damage induction, cell death and DNA damage repair in radiation mutagenesis.

Contribution of GSF-Neuherberg

The experiments to be undertaken by the GSF-Institut für Sägetiergenetik will be orientated towards testing two critical assumptions required in extrapolating from results in animals to an estimate of the radiation genetic risk in man, i.e. the question of extrapolation across species as well as possible genetic endpoint differences in mutagenic response to radiation. Mutagenicity testing procedures have been developed in Neuherberg to systematically screen for genetic endpoints which are not dependent upon special tester strains of animal. Further, the genetic endpoints studied (dominant cataract and enzyme activity) are relevant in any mammalian species considered and allow a direct species comparison of the sensitivity to mutation induction by radiation at identical genetic endpoints. Finally, the genetic endpoints chosen for study are directly comparable to known genetic diseases in man. Dominant cataract mutations exist in man and, more broadly, the dominant cataract test is a superb animal model for the entire category of dominant deleterious mutations in humans. The enzyme activity mutations are direct animal models of inborn errors of metabolism in man.

Studies to evaluate the extrapolation of genetic risk across species will be carried out utilizing the laboratory mouse Mus musculus and the golden hamster Mesocricetus auratus. Specifically the sensitivity to mutation induction by radiation in oocytes of the two species will be compared.

To determine if both recessive and dominant mutations are subject to similar mechanisms of radiation mutagenesis, experiments will be undertaken to determine the effect of sensitizing pre-treatments such as 3-aminobenzamide, hydroxyurea or triethylenemelamine on the radiation-induced mutation rate to recessive specific locus and dominant cataract mutations in the mouse.

Finally, recessive and dominant mutations recovered in mutation experiments will be phenotypically, genetically and cytogenetically characterized in cooperation with parallel studies at the MRC Radiobiology Unit (Cattanach et al.).

Contribution of the University of Utrecht

The studies conducted are designed to evaluate the Leenhouts/Chadwick model for the induction by radiation of reciprocal translocations and point mutations in stem cell spermatogonia. The model explains the humped shaped dose-response curve, and the values of a number of parameters (e.g. the ratio between the numbers of stem cells resistant or sensitive to cell killing and induction of translocations) are assumed. This project will be carried out in close cooperation with Drs. Cattanach and van Buul, and will determine many of these parameters in the 3H1 hybrid mouse. With the help of image analysis the total number of spermatogonial stem cells per testis will be determined. Further, an estimation will be made of the numbers of stem cells per

testis, resistant or sensitive to the cell killing effect of radiation. To this end a fission neutron irradiation experiment will be carried out. As the dose-response for fission neutrons has no shoulder, extrapolation to dose zero is possible and the numbers of resistant and sensitive stem cells in the unirradiated situation can be estimated. Furthermore, the sensitivity of spermatogonial stem cells for cell killing by a second dose of X-rays 24 hr after the first dose will be determined. This will render the D0 value of stem cells that were resistant and of those that were sensitive during the first irradiation. When time permits fractionation experiments with longer intervals (e.g. 4-5 days) will also be carried out.

The radiosensitivity for the induction of reciprocal translocations in stem cells resistant to cell killing by irradiation and in stem cells sensitive to cell killing also will be determined. This will be done in mice with a synchronized seminiferous epithelium via vitamin A deficiency /vitamin A replacement method.

The data obtained during the project will be used to evaluate the Leenhouts/Chadwick model. Estimates of the model parameters will facilitate species and strain comparisons, carried out by the groups of Cattanach, Ehling, and van Buul, as they will provide a better understanding of the differences in the dose response relationships found.

Contribution of the University of Leiden

In the estimation of genetic risks ensuing from exposure to ionizing radiation, the emphasis is on the risk due to mutations having dominant effects, especially those that arise in pre-meiotic germ cell stages. This subproject proposes to investigate the induction frequencies and nature of dominant mutations by irradiation in Drosophila spermatogonia. Recessive visibles at a number of selected loci will be studied simultaneously and this wi11 enable comparison of induction frequencies and molecular nature between dominant and recessive mutations. Although it is well known that the frequency of recessive lethal mutations is much higher than of dominant ones, for risk assessment they are not considered a matter of primary concern. However, over 50% of these mutations are multilocus deletions. The dominant effect of such multilocus deletions in heterozygotes will be investigated. In Drosophila 149 loci where dominant visible mutations (wings, eyes, bristles and body) can arise are well known and allow detection in forward mutation experiments. In addition many well defined multilocus deletions are available.

Offspring resulting from irradiated spermatozoa and spermatogonia will be scored. Isolated recessive visible mutations of the vermilion locus will be genetically and molecularly analyzed. Isolated dominant mutations will be characterized genetically. A subset of the dominant mutations may be analysed at the molecular level to establish their exact nature. Crossing schemes are devised to estimate the dominant effects of multilocus deletions on fitness traits.

The data obtained with Drosophila may provide important information, complementary
to that recorded in the mouse (see projects Dr. Cattanach, MRC, and Dr. Ehling, GSF).

Contribution of the University of Leiden

1) In order to elucidate the recorded differences in the recovery of radiation induced chromosomal translocations from stem cell spermatogonia of rhesus monkeys and mice two approaches will be taken. First, dose-response studies for the induction of translocations and cell killing analysis in steel (Sl) and dominant spotting (Wv) mice with "primate type" spermatogenesis will be completed. Feasibility studies on translocation induction in DNA repair deficient severe combined immunodeficient (scid) mice will be conducted. Second, collection of data on cell killing and translocations in specific stages of the spermatogonial stem cell cycle via combined hydroxyurea (HU), 3-aminobenzamide (3-AB) and X-rays are planned. In recent years it has become clear that both the radiosensitivity for cell killing and for the induction of translocations varies during the spermatogonial cell cycle but simple correlations seem not to exist. Thus, more detailed information is essential for our understanding of the mouse-rhesus monkey differences. All translocation work is closely related to the work of Cattanach (MRC-Radiobiology Unit) on the induction of genetic changes in strain 101 mice and other mouse mutants and that of de Rooij (Utrecht) on cell killing in C3H/101 mice with vitamin-A deficiency.

2) For further characterization of mouse germ cells the induction of DNA strand breakage and its repair will be studied in oocytes of different mouse mutants employing a microgel electrophoresis technique. For comparisons with man somatic cells can be used. This work is closely linked to the cytogenetic studies of radiation exposed human populations by Hulten (West Midlands Regional Genetic Services) and the in vitro radiation studies of human spermatozoa by Miro Ametller (Universitat Autónoma de Barcelona).

3) Preliminary experiments to determine the suitability of the available transgenic mouse strains harboring bacteriophage lambda shuttle vectors with selectable LacZ gene in detecting X-ray induced spermatogonial mutations will be carried out. These studies are strongly interactive with the genetic endpoint comparisons of Cattanach (MRC-Radiobiology Unit), Eeken (Leiden), and Ehling (GSF-Neuherberg).

Contribution of MRC - Radiobiology Unit

The core work continues mouse mutation studies to elucidate factors influencing the mutagenic response of spermatogonial stem cells to radiation. Two strategies will be pursued. First, investigation of mouse strains which show unusual responses to irradiation will be conducted. The inbred 101/H strain has a uniquely high sensitivity to stem cell killing but has given lower translocation yields than typically obtained with the C3H/HeH x 101/H F_1 hybrid standardly used. Recent investigation has, however, suggested that the strain shows a normal specific locus mutation response. A reduced translocation response might be due to technical factors associated with grossly damaged testes, affecting scoring. Alternatively, the responses of the inbred and the hybrid to the different endpoints may genuinely differ. Translocation induction will be reinvestigated using a more refined chromosome preparation procedure. Second, the mutagenic response of 'altered' stem cell populations will be asssessed using chemical pretreatments which either, a) kill cells non-specifically to deplete the population (triethylenemelamine), b) kill cells in S phase (hydroxyurea) or, c) sensitise cells to

radiation damage (3-aminobenzamide). Dependent upon the interval between chemical and radiation treatments, the results should identify differences in response between repopulating and normal stem cell populations and between cells in different stages of their cell cycle.

A further major component of the work will involve analyses of mutants induced. This will specifically include a newly identified class representing large chromosome deletions but will also include molecular analyses of mutations at the mottled locus, which is homologous to the locus for Mencke's kinky hair disease in man. Feasibility studies will additionally be conducted to determine if radiation induced genome rearrangements can be detected by pulsed field gel electrophoresis.

The translocation studies are relevant to and coordinated with the studies of van Buul (Leiden) and de Rooij (Utrecht). The mutations studies are directly relevant to the studies of Ehling and Favor (Neuherberg).

Contribution of the West Midlands Regional Genetic Services

There is little information available on the relation between amounts and types of radiation-induced chromosome abnormalities in the soma and germ-line of man. We propose to study both somatic cells and germ cells of exposed humans to fill an important gap in our knowledge of radiation effects of particular relevance for the evaluation of induction by radiation of genetic disease in future generations of man.

The fluorescence in situ hybridisation (FISH) technique will be employed for the identification of radiation induced chromosome aberrations in men following (1) radiotherapy, (2) occupational, and (3) accidental exposure. The study populations include Sellafield nuclear power plant workers and subjects exposed to radiation at the Chernobyl nuclear accident as well as adequate controls. Somatic chromosome preparations and semen samples have already been banked for analysis. We will utilise the whole chromosome specific probes, repeat-sequence DNA centomeric probes and chromosome segment specific probes, which allow a more sensitive assay for chromosome aberrations. We are especially interested in the dose-response as regards stable chromosome abnormalities such as reciprocol translocations and insertions as indicators of heritable genetic lesions which may compromise future generations. For an assessment of radiation effects in soma cells, in vitro cultured lymphocytes at metaphase and at interphase will be examined. For the germ-line studies, chromsome gains and losses will be determined in individual spermatozoa from semen samples of the same control and exposed populations of men as examined for effects in soma cells.

The proposed studies represent a direct assessment of genetic damage in man and thus serve as the most critical data set with which to compare laboratory mammal experimental results. The human cytogenetic data are most comparable with the cytogenetic studies proposed by van Buul (Leiden University) and Cattanach (MRC). In addition, the human results will be most important to the studies of germ cell and species differences (Neuherberg, MRC) as well as the dose response studies of de Rooij (Utrecht University).

Contribution of Universitat Autónoma de Barcelona

Cytogenetic studies of human germ cells employing the human-hamster embryo technique offers the advantages of providing a large number of cells for analysis by non-invasive techniques. Standard cytogenetc studies scoring for chromosomal aberrations have been successful in establishing the interspecific fertilization techniques and have shown an increased aberration rate in mutagen exposed cancer patients treated with radiation adnd/or cytostatic drugs. An alteration of the above techniques to assay for micronuclei would have as an advantage a simplification of the cytogenetic procedures as well as to provide data for the human in vivo germ cell situation comparable to the vast data base for laboratory mammals as well as human somatic cell studies. To this end the proposed project will involve a dosimetric study on human spermatozoa for radiation-induced micronuclei via the human-hamster embryo technique.

2-cell human-hamster embryos will be scored for micronuclei in a feasibility study extending the capabilities of our laboratory, which already is competent in the interspecies fertilization and cytogenetic techniques. To differentiate between the human or hamster origins of the micronuclei, fluorescent in situ hybridization procedures utilizing chromsome specific probes for human chromsomes will be carried out. Finally, control and in vitro irradiated human spermatozoa will be analysed by these techniques to establish a radiation dose response for micronuclei as assayed in human-hamster embryos.

As a direct measure of genetic damage in human germ cells, the proposed research is highly relevant to the coordinated research effort "Genetic risks associated with exposure to ionizing radiation". The results anticipated will be most directly relevant to the projects on cytogenetic damage assessed in laboratory mammals (Leiden University, MRC) as well as the cytogenetic studies on exposed human populations (West Midlands Regional Genetic Services).

B15 Action of radionuclides on target cells in relation to radionuclide metabolism and studies on biological models for radionuclide-induced cancer.

Contract FI3P-CT920021 Dose assessment early cellular and late carcinogenic effects of exposure to radon and its progeny.

Coordinator CEA - FAR
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Total Contribution by the Commission: 140 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The link between occupational exposure to high levels of radon and increased lung cancer incidences has long been established. Epidemiological studies of lung tumour incidences in relation to environmental and domestic exposure have shown no or even negative correlations. The risk factors for radon exposure are derived from the risk at high levels of exposure and the effect of low doses and low dose rates are not known. Another considerations are the effect of environmental factors modifying physical forms of radon daughter aerosols.

Animal studies on the effects of radon exposure have been very limited and in those that have been conducted only a limited characterisation of the exposure conditions has been conducted making the comparison of the results from different studies impossible.

There are currently three animal radon exposure facilities in Europe. In France, COGEMA and the CEA operate a large facility which accomodate up to 700 rats for whole body exposure. A new facility has been constructed at AEA Harwell which will allow exposure of up to 120 rats or 200 mice by whole body exposure. The advantage of the latter system is that exposure can be continuous for periods of up to six months, whereas the other facility is limited to exposures of a few hours per day.

Comparison of the effects of radon and radon daughter exposure in the exposure facilities requires a common index of exposure. The Working Level Month is the unit currently used to describe exposure levels. However differences between the facilities in the characteristics of the exposure conditions in terms of the equilibrium of radon with its daughters and the degree of attachment to adventitious or carrier aerosols is known to cause differences in deposition throughout the regions of the respiratory tract and hence differences in the local dosimetry.

This project is mainly confined to dosimetric studies using physical and biological methods. It also includes studies of some early and late effects induced by radon exposure such as induction of cell proliferation.

Specific Objectives

1) To establish standard radon metrology between participant laboratories.

This will ensure that the methods and equipment currently used in the metrology of radon and radon daughters by each of the participating laboratories yield comparable results.

2) To study the effect of exposure conditions on the deposition of radon daughters in the respiratory tract.

This will enable more accurate estimates of radiation dose to the different regions of the lung to be made and provided that the methods can be standardised, the results from the participating laboratories can be compared.

3) To develop new assays for biological dosimetry in the different parts of the respiratory tract using cytological methods.

This will provide a more accurate measurement of delivered doses that can be expressed as standard units after a comparison with measurements performed in animals, following homogeneous irradiation of the respiratory tract by external local irradiation.

4) To continue with the studies on the early subacute of radon exposure on the epithelial cells of the rat respiratory tract, using cell proliferation indices and the incidence of nuclear aberrations in alveolar macrophages as indicators of radiation damage.

5) To initiate life-time carcinogenicity studies to continue investigations on the effect of dose and dose rate on lung tumour induction.

The CEA Fontenay aux Roses study:

The main contribution of the laboratory in this contract is development of a new assay aiming to perform local biological dosimetry in the different parts of the respiratory tract after inhalation of radon and its daughters. Exposures are performed under controlled conditions measuring different physical parameters. The assay is based on measurement of genetic damages visualized by cytological methods.

Induction of cell proliferation in the respiratory tract.

Cells of the respiratory tract are poorly proliferating so that very rare mitosis are observed. We have developped an experimental model, exposure of rats to 2 ppm ozone during 5 hours, to induce cell proliferation which was observed mostly in alveolar macrophages and bronchiolar and bronchial epithelial cells between 24 and 48 hours after the treatment. As many as 8 % of macrophages and more than 20 % of the epithelial cells were labelled 2 hours after a single administration of DNA precursor. Thus, this experimental model is appropriated to study metaphasis or post replicative cells in different parts of the respiratory tract.

Cytological measurement of micronuclei after induction of cell proliferation.

Preliminary results have been obtained in alveolar macrophages that allows to estimate doses delivered to alveolar tissue. After irradiation, ozone treatment increases about 2 fold frequencies of micronuclei compared to irradiated controls; measurements limited to post replicative cells increase this value by a factor 4 to 5. Determination of a dose effect relationship is in progress in animals exposed to radon under "static" (high attached fraction) or "dynamic" (low attached fraction) conditions during 24 hours. First results show a similar induction of micronuclei under "dynamic" compared to "static" conditions and will be compared to those obtained in rats irradiated locally by gamma rays.

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Future developments.

A similar cytological study will be performed on isolated tracheal epithelial cells. Chromosome painting would be used to score chromosome rearrangements on mitosis. Assuming the use of a confocal microscope, these measurements will be performed in situ, on thick sections, that allows a study all along the upper airways.

Others contributions mostly correspond to maintain Razès facility, metrology and experimental studies of 214 lead deposition in the respiratory tract.

The Razès facility is used for interlaboratory comparative metrology and exposed animals can be provided. As far as possible, our biological dosimetric studies will be carried out in animals exposed in other facilities and tissue or cell samples are prepared in order to allow complementary or comparative experiments between the laboratories.

The AEA Harwell study

Under this contract AEA are involved in four main aspects of the studies:

Metrology standardisation.

AEA is taking the lead on the metrology standardisation. Methods for monitoring radon and radon daughter concentrations are being compared. Comparison and standardisation are necessary because of the different methods and equipments used by the two groups for their measurements. A first intercomparison of measured radon and radon daughter levels (attached and unattached) at the CEA/COGEMA facility during animal exposures showed generally good agreement between the groups. The maximum spread in results was around 40%. It did however highlight some differences in calibration methods. These were reviewed prior to a further intercomparison.

Measurements of flow-rate, and radon daughter concentration, over a range of PAEC (potential alpha energy concentration) levels, "unattached" fractions (fp) and equilibrium factors were made. The importance of sample pump flow rate measurements was highlighted. Generally good agreement was seen between the participants in their measurements (within 17%).

A final intercomparison of metrology is planned at AEA Harwell. These studies provide full characterisation of the exposure conditions in the two facilities and forms the basis for the comparison of the metrology between the two groups.

Deposition.

AEA have developed a method for the measurement of deposition of radon daughters in different regions in the lung, trachea and head. Deposition measurements under well defined conditions in both facilities will indicate whether there are underlying differences in the exposure conditions which result in marked differences in deposition and hence in doses to the lung.

Early subacute studies.

AEA will measure the cell proliferation indices in differents regions of the respiratory tract and the incidence of nuclear aberrations in alveolar macrophages.The effect of dose and dose rate will be studied. The AEA results from animals exposed in the CEA/COGEMA facility will be compared with the results on the same animals obtained by the CEA group. Results from these animals will be compared with those from animals exposed under similar conditions at AEA.

Lifetime carcinigenicity studies.

AEA will initiate life-time carcinogenicity studies in both rats and mice using different cumulative doses and dose rates. The results from these studies will be compared with those obtained from animals exposed in the CEA/COGEMA facility.

B15 Action of radionuclides on target cells in relation to radionuclide metabolism and studies on biological models for radionuclide-induced cancer.

Contract FI3P-CT920051 Induction of osteosarcoma and leukaemia by bone-seeking alpha-emitting radionuclides.

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Total Contribution by the Commission: 520 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The objective of this project is to facilitate the more precise quantification and understanding of the risk of radiation-induced carcinogenesis. Exposure to low doses of alpha-emitting bone seeking radionuclides has been selected as an experimental model.

Although therapeutic use of alpha-emitting radionuclides has been largely discontinued, human exposure may still occur in the environment from the use of nuclear energy, disposal of radioactive waste and exposure to radon and its daughters. This pattern of exposure can result in a low-dose irradiation of sensitive tissues, particularly the physiologically interrelated bone and bone marrow cells. Thus, protection of the general population through definition of carcinogenic dose levels, and identification of at risk individuals remain a necessary goal of radiation protection. Identification of the radiation-induced molecular lesions that directly lead to carcinogenesis will allow a more accurate assessment of the dose-effect relationship, and will permit a monitoring of at risk individuals. Our experimental goal is to use a range of model systems to identify those molecular mechanisms involved in radiation-induced tumorigenesis. We have adopted an integrated approach to this problem, combining animal studies with *in vitro* cell culture systems and molecular biological analysis.

The project is sub-divided into three inter-related tasks:

1) Comparison will be made of the in vivo behavior of different alpha-emitters, with the objective of relating incidence and tumour type to the distribution of dose at the cellular level and to the target cell populations in sensitive tissues. This approach is designed to allow more precise identification of those cell populations specifically at risk from alpha-radiation effects. The effect of age, including *in utero* exposure, will be examined, and the importance of incorporation of short-lived ^{224}Ra and other radionuclides into target cells will be investigated.

2) Molecular analysis of the genetic events of carcinogenesis will focus upon initial mutational events and the activation of mobile elements in the genome after exposure to alpha radiation. The consequences of these phenomena for gene expression will be studied. The molecular basis for retrotransposon activation and their role in the early stages of radiation-induced tumour development will be determined. Alterations to the tumour suppressor gene p53 will be followed, and new loci mutated by radiation will be identified. Knowledge of these molecular changes will be of potential use as sensitive indicators of early damage during the latency period.

3) Using new model systems of haematopoietic and osteoprogenitor stem cell populations we will identify and localize those cells that are at risk from alpha-radiation. This is an essential complement to Task 1, where the dose received by these cells will be estimated more accurately. The influence of prior somatic cell mutation will be determined in an *in vivo* model of paternal oncogenesis. The use of all of these model systems in conjunction with molecular studies will facilitate quantification of molecular changes after exposure to radiation, and will provide additional information for risk analysis.

Contribution of the Institut of Pathology, GSF-München

Radiation-sensitive loci in ostesarcoma and transgenerational carcinogenesis.

To assess the risk of carcinogenesis at both the individual and population levels it is necessary to identify radiation-induced molecular events that lead to, or predispose for, a malignant phenotype. However, very little is yet known about the precise number of sensitive loci involved in radiation-induced carcinogenesis, or the frequency of these different forms of genetic alterations. In this project we will analyse radiation-induced osteosarcomas to identify DNA loci deleted by irradiation. To identify these potential targets for alpha-irradiation inactivation we have developed a molecular technique to identify loci inactivated during radiation-induced carcinogenesis. The technique relies upon the identification of those genes whose normal transcript has been deleted in a radiation-transformed cell. The basis for the method lies in the subtraction from a normal osteoblast cell cDNA library of all sequences still expressed in a radiation-induced osteosarcoma. The sequences that are unaffected by the subtraction procedure are *a priori* absent in the tumour cell. By cloning these sequences we will be able to establish if there is a pattern to inactivation events in a range of osteosarcomas, and thereby isolate and identify loci that are frequently inactivated. Sequences thus identified will be made available to partners using model systems designed to quantify early events in tumorigenesis.

Loss of gene loci which control the normal proliferative activity of dividing cells is known to result in an increased risk of carcinogenesis. Thus, inheritance of a mutated allele via the germ line or due to somatic radiation damage will influences tumour susceptibility. One model we are using to study this phenomenon is the effect of prior parental exposure to mutagenic substances upon the susceptibility of an F1 generation to radiation-induced carcinogenesis. Male parents of the murine 102 x C3H stock were treated with a mutagenic dose of ENU and crossed with T-stock females. A proportion of the F1 offspring were exposed to ^{227}Th to determine the influence of paternal effects upon radiation risk. This project is a continuation of B17-0068. In addition to assessing risk we will determine if inheritance of mutated alleles of the tumour suppressor locus p53 influences carcinogenesis in this system. The assay of p53 mutation will also be used to screen osteogenic colonies grown in vitro after alpha contamination of mice (in conjunction with VITO).

Contribution of the Biomedical Effects Department, NRPB.

Distribution of alpha-emitters within the bone marrow-skeletal system: Role of local concentrations in determining tumour frequency and type.

In the main study, the toxicity of the three bone-seeking radionuclides ^{239}Pu, ^{241}Am and ^{233}U is being compared with the objective of relating differences in the distribution of dose within the skeleton, and the extent of irradiation of the different cell types, with the observed incidence and distribution of osteosarcoma and myeloid leukaemia. In addition, studies are in progress on the distribution and retention of ^{210}Po in the skeleton of rats and marmosets after administration of either ^{210}Po or ^{210}Pb with the objective of assessing alpha-doses and leukaemic risk from these naturally-occurring nuclides. Measurements have been made of the distribution of

^{239}Pu, ^{241}Am and ^{233}U in the mouse skeleton at times up to 448 days after administration. Further analyses are being undertaken on bones from each mouse in the toxicity study to provide direct information on the retention of activity at death. Autoradiographic studies to quantify the distribution of alpha-activity within the bone are in progress. Fission track autoradiographs of femur sections have been produced and analyses of the distribution of activity and dose with time after injection are planned. It is intended to model the burial of bone-surface deposits of the nuclides and their transfer to bone marrow. Photographic emulsion autoradiographs of mandibular condyle and rib have been prepared for use in comparisons of doses to different cell types (coperation with GSF-P).

In the comparison of the toxicity of ^{239}Pu, ^{241}Am or ^{233}U, groups of 50-100 mice were given systemic injections of the nuclides at three dose levels. For ^{239}Pu, the injected activities were 5, 15 or 25 kBq kg^{-1}. The corresponding amounts for ^{241}Am and ^{233}U, to give equivalent average bone doses 6, 17 and 29 kBq for ^{241}Am and 40, 118, and 197 kBq kg^{-1} for ^{233}U. To date, approximately 80 % of the animals have died. Eighteen bone tumours have been observed, 10 in animals given plutonium. Identification of other suspected bone tumours is in progress, looking at gross changes shown by X-ray analysis and tissue histopathology. Eight myeloid leukaemias have been identified and suspected myeloid leukaemias are being histopathologically and cytogenetically evaluated. Under B17-0037 a number of the leukaemias have been karyotyped and have shown chromosome 2 rearrangements characteristic of acute myeloid leukaemia. DNA obtained from the brain and spleen of leukaemic animals will be analysed using molecular techniques in order to extend knowledge on the involvement of telomere-like repeat sequences in myeloid leukaemogenesis.

Contribution of the Radiobiology Unit, MRC

i) **Effect of age and dose rate on the induction of tumours by alpha-particle emitters in CBA/H mice.**

ii) **Irradiation damage of the murine haematopoietic stem cell system.**

In order to identify early events following exposure to alpha-particle irradiation we are using a clonogenic culture system to stimulate the clonal growth of cells derived from stem cells. Preliminary studies have been concerned with the identification of cytogenetic aberrations in individual colonies of haemopoietic cells derived from irradiated stem cells. Exposure to alpha-particles (but not X-rays) produced a high frequency of non-clonal aberrations in the clonal descendants, compatible with alpha-emitters inducing lesions in stem cells that result in the transmission of chromosomal instability producing visible abnormalities many cell cycles later. It is possible to postulate that such instability could, on occasion, disrupt a region of the genome involved in leukaemic transformation. As stem cells have the property of self-renewal, such a change could arise in a daughter stem cell and thus represent an apparent initiating lesion although the actual initiating lesion would be the radiation-induced instability. Future studies will investigate further the observed instability by cellular, cytogenetic and molecular investigations of irradiated cells *in vitro* and in mice transplanted with alpha-particle-damaged stem cells.

The main overall aim of the long-term mouse studies has been to investigate the leukaemogenic and osteosarcomagenic effects of alpha-particle emitters. Integrated with this aim have been the modifying effects of age (including in utero) and of the period over which the alpha-particle emitter was administered. In those twelve week old mice given ^{224}Ra in a single injection, the incidence of myeloid leukaemia is more than twice that of osteosarcoma. Other results suggest that the incidence of myeloid leukaemia is greater in mice given ^{224}Ra in multiple injections than in those given the activity in a single injection. Three experiments are continuing in which the effects of age of the mice at contamination are being investigated. In one, male mice were given ^{224}Ra when four weeks old either as a single injection or as sixteen injections spaced over eight weeks. In another, pregnant mice were injected with ^{239}Pu on either day 4 or day 13 of gestation. In the third, male mice were injected paratibially with ^{228}Th when twelve weeks old, to investigate the effects of continuous contamination of the skeleton with ^{224}Ra.

Contribution of IMV-GSF

Cooperation of low dose alpha-irradiation, retrotransposons and celleular genes in induced neoplasia

Radiation-induction of osteosarcomas in mice -a model system for radiation carcinogenesis in man- is associated with a dose-dependent activation of endogenous retroviruses. These viruses induce skeletal tumours in the mouse, indicating that provirus activation, the expression of infectious retroviruses and new integration into the DNA of affected host cells represent critical steps in radiation osteosarcomagenesis. Further, the data suggest that activation of endogenous retroviral genes may serve as an early molecular marker for osteogenic cells which have been affected by irradiation. Additional genetic or epigenetic events are also involved in radiation osteosarcomagenesis. These include alterations in p53, c-fos/v-fos and c-myc expression.

The objectives of our group are to study the initiation, fixation, enhancement and/or accelaration of radiation-induced genomic alterations by radiation-activated retroviral genes. Cooperating effects of alpha-irradiation and infection with radiation-activated retroviruses on differentiation and neoplastic transformation of skeletal cells will be analysed. Early effects of radiation-induced provirus activation, retrovirus infection and new integration in osteogenic target cells will also be determind. Affected cells and tissues will be analysed for the expression of protooncogenes, structural genes typifying osteogenic differentiation, and genes associated with and regulating cell proliferation.

As in vitro systems we will use cell and tissue cultures from bone-forming cells established from c-fos, c-myc, and p53 transgenic mice, and hybrid mice carrying combinations of these transgenic traits. These strains have been successfully bred in our laboratory. Cells will be irradiated with doses of ^{223}Ra which are known to activate endogenous proviruses and to induce cell transformation. In addition, irradiation will be preceeded or followed by infection with molecularly cloned retroviruses which in previous experiments have shown to induce 100% benign bone tumours in newborn mice. The molecular analysis will be done in collaboration with GSF-P and DMB.

Contribution of the Department of Molecular Biology, University of Aarhus

Molecular mechanisms of radiation induced retroviral activation and determination of pathogenic sequences.

Radiation carcinogenesis is presumed to involve a number of events in the host cell leading to quantitative and qualitative changes in the expression of certain cellular proteins. Proviral genes, proto-oncogenes and tumour suppressor genes are amongst specific target genes associated with carcinogenesis. Endogenous proviral genes form large gene families in the chromosomal DNA of mammalian species. We focus on the family of murine leukaemia viruses because: 1) the complete nucleotide sequence has been determined for a number of proviruses, 2) proviruses of this group are found expressed during radiation-induced osteosarcomagenesis and leukaemogenesis, 3) murine leukaemia viruses are able to induce a variety of leukaemias and bone tumours when injected into newborn mice, 4) proviruses of this group have, in many cases, been found to activate oncogenes either by provirus integration or by recombination, and inactivation of a tumour suppressor gene by provirus insertion has also been observed, 5) related families of proviral genes have been found in man.

Molecular mechanisms of radiation carcinogenesis will be studied with respect to early effects of radiation upon gene expression using endogenous retroviral elements as markers. In a cell, radiation and other forms of stress may activate transcription factors as a result of an overall biochemical response without direct physical hits to their genes. One such transcription factor is the protooncogene product c-Fos that, in complex with other proteins, may bind to specific regulatory regions of DNA and thereby transactivate other genes. Transcriptionally silent endogenous proviruses in some strains of mice have been found to be activated by alpha radiation. One possibility is that c-Fos and related proteins, produced in response to radiation, stimulate provirus transcription. Our ongoing studies indicate that c-fos expression has some stimulatory effect upon transcription from a murine leukaemia virus LTR after gene transfer to a monolayer culture of osteogenic cells. The possible activation of a provirus by alpha radiation in cultured cells rather than in animals will facilitate studies of molecular mechanisms (with GSF-IMV). Monolayer cultures derived from a mouse strain with a transcriptionally silent endogenous ecotropic provirus will be irradiated with doses of ^{223}Ra which are known to activate endogenous proviruses and induce osteosarcomas in mice. If provirus expression can be detected by sensitive PCR-techniques as an early result of irradiation, our studies will focus on the mechanism behind this activation, and on the role of other potential target genes.

Contribution of Institut für Allgemeine Pathologie, Technische Universität München

i) **Development of methodology for analysis of gene mutations in archival human materials**

ii) **Gene inactivation: Identification of mutational events at the p53 locus**

The central role of the tumour suppressor gene p53 in tumorigenesis has been established by many different lines of evidence. We have been able to detect p53 mutations consistently in murine osteosarcomas induced by internal contamination with ^{223}Ra. Mutations identified are predominantly small to moderate deletions of exonic

DNA, consistent with alpha-particle mediated damage. The high frequency of mutation indicates that further assessment of the nature of the mutations, their distribution and their frequency will be a useful procedure for recognising and quantifying early events in tumorigenesis. The localisation of frequent mutation sites is essential for future large scale analytical studies using archival material or biopsies taken from pre-malignant states or for screening purposes. The facilities of the TUM are geared to the routine analysis of gene mutation events by the direct sequencing of polymerase chain reaction (PCR) amplificates of the p53 locus. We are routinely involved in the analysis of p53 in a range of human tumours. Whilst a small number of fresh human osteosarcomas will become available during the study period, and we have an established bank of some 10 frozen tumours, we recognise the urgency for the analysis of archival material. As part of our previous proposal we have established the technology for the amplification and sequencing of specific DNA sequences in tumour material microdissected from formalin-fixed paraffin embedded tissue, and for the analysis of p53 mutations in fresh frozen osteosarcoma tissues. We will continue to develop this PCR-based methodology for use on archived murine and human osteosarcoma samples.The pending EULEP proposal for the establishment of a european collection of tumour material (EULEP-ARCHIVE), and our own experience with the human radiation-induced osteosarcomas in ankylosing spondylitis and bone tuberculosis now makes it feasible to begin to analyse archival materials of human osteosarcoma from irradiated subjects.

In addition we intend to expand our knowledge of the location and frequency of p53 mutations in the murine osteosarcoma model, allowing us to identify mutational hot spots. These will be compared with the hot spots already described from a wide range of spontaneous human tumours, and to the human osteosarcoma material available to us.

Contribution of Departement Leefmilieu, Vlaamse Instelling voor Technologisch

i) **Irradiation transformation of the murine osteoprogenitor system.**

ii) **Identification of osteogenic target cells in the bone marrow.**

Our contribution will be directed to the identification and quantification of cellular, molecular and genetic alterations which occur in bone marrow target cells at early stages in radiation-induced osteosarcomogenesis. The availability of early and sensitive markers of radiation damage will improve risk assessment. In previous years we developed a mouse model for osteosarcomogenesis and *in vitro* techniques to study osteogenic precursor cells among adult murine bone marrow. *In vitro* culture of bone marrow from irradiated female Balb/c mice (40 Bq ^{241}Am/g mouse, skeletal a-dose rate: 7 mGy/day) which show a high risk (> 50 %) for osteosarcoma development, has shown to be sensitive to a-particle irradiation. Therefore this technique allows detection of long lasting changes in the osteogenic differentiation capacity (mineralization *in vitro*) of bone marrow stromal cells.

The next two years should allow

1. Further phenotyping of the precursors of bone cells in the marrow and purification of the cell culture system. Subpopulations of bone marrow stroma

cells with osteogenic potential will be obtained and Fluorescence Activated Cell Sorting (FACS) analysis will be used for immunologically phenotyped.

2. Phenotypic tracing and quantification of radiation induced changes in osteogenic marrow cultures. In relation to a-radiation dose, the changes in cell proliferation and expression of differentiation markers such as collagen Type I, alkaline phosphatase, osteonectin, osteocalcin will be further quantified using incorporation of radio-labeled precursors (^{3}H-Tdr, ^{3}H-proline), immunocytochemistry, *in situ* hybridization and subsequent image-analysis and ELISA-technique.

3. In marrow cultures from a-irradiated mice, or marrow cultures irradiated *in vitro*, tumour suppressor genes (p53, Rb), and oncogenes assumed to be involved in tumour development and/or bone differentiation (fos, myc, jun, ras) will be studied by *in situ* hybridization and sensitive gel electrophoresis following PCR (with GSF-P).

4. Development of an *in vitro* transformation assay by a-irradiation of marrow *in vitro* (^{223}Ra added to osteogenic marrow cultures) or *in vivo* (marrow from ^{241}Am injected mice), with and without addition of a tumour promotor (With GSF-IMV).

B2 Non-stochastics effects of radiation

B21 Radiation syndromes and their treatment after exposure of large parts of the body.

Contract FI3P-CT920008 Research on the management of accidentally radiation exposed persons.

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Description of research work

Objectives and expected achievement:

The Collaborative Programme on the Management of Accidentally Radiation Exposed Persons is executed by 4 scientific working groups in The Netherlands, France, Italy and Germany. The objectives of this research work are to contribute by preclinical and clinical research to the area of "non-stochastic effects of radiation" as outlined in the Radiation Protection Research 1992-1993 of the Nuclear Fission Safety Programme of the European Communities.

Pathophysiology of radiation effects: (table 1, part 1.1, 1.2 and 1.3)

It is essential to broaden the basic knowledge of the effects of ionizing radiation on the mammalian organism in 3 main areas: the immuno-hematopoietic system is of particular importance for the survival of the acute phase of the radiation syndrome observed after doses larger than 0.5-1 Gy. The potential failure of the specific and unspecific resistance of the organism against infection and foreign proteins and the potential failure of the blood cell forming tissues require new approaches utilizing the recent advances in molecular biology and cell cloning. Even if the failure of the immuno-hemopoietic system can be treated,, one will be faced with the radiation injuries of the respiratory and gastrointestinal system including the slowly turning over stromal cells in which radiation damage may well be stored for long periods of time and show up only in terms of "late effects". The central nervous system became an organ of interest especially in the Hiroshima studies. Therein, a potential risk of mental retardation after radiation exposure was observed.

Thus, the research groups in Rijswijk and in Ulm will address the problem of the radiation sensitivity of hemopoietic stem cell populations at various dose rates and radiation qualities as well as mechanisms of replication and differentiation as the essential basis for prophylactic and therapeutic measures using human, nonhuman primates and canine cell models. Of equal importance to these groups is the question whether new avenues for intensive care will influence the LD50 of bone marrow damage. This approach employs pathophysiological models in order to improve the understanding of the mechanisms of the kinetics and regulation of hematopoietic regeneration with and without bone marrow and/or blood stem cell transfusion using new systems simulation approaches. The group in Rome will widen the scope of studies to the field of the lymphocyte population as a key element for understanding the immune system after accidental irradiation. The group in Paris but also in Ulm is particularly interested in the study of the interaction between stem cells and cells of the bone marrow stroma. This is of particular relevance: the key question for the restauration of hemopoiesis after TBI is, whether the microenvironment (sinusuidal system, stroma- and endothelial cells, myelinated and unmyelinated nerves) is capable of supporting stem cell replication and differentiation quantitatively and qualitatively (e.g. production of growth factors by the cells of the stem cell microenvironment). The group in Paris, however, will expand the research agenda to the cells of the respiratory and the gastrointestinal system. It is well known, that vascular changes and fibrotic lesions are consequences of local and whole body radiation exposure and present - as late effects - life threatening symptomatologies,

especially when complicated by viral infection. It is envisaged, that advances will be made resulting in new directions for measures of "secondary prevention". The study on the CNS is of particular importance to improve the understanding of pathophysiological mechanisms of radiation response relevant for both diagnostic and therapeutic possibilities. It is envisaged to study the CNS by applying new aproaches of EEG changes in order to establish the role of such approaches for the early diagnosis of the acute radiation syndrome and for the development of predictors for the reversibility of CNS impairments.

Diagnosis of the radiation syndromes: preclinical and clinical approaches
(see table 1, part 2).

As pointed out in the research task concerning the non-stochastic effects of radiation it is of great importance to assess both early and late biological effects on the human organisms after accidental exposure to different radiation doses and different radiation qualities. While some information is already available with respect to the diagnosis of the effects of high doses of radiation in excess of 1 through 10 Gy a new dimension must be recognized. Due to the previous work of the research group in Rijswijk there is evidence that the pluripotent hemopoietic stem cell responsible for the hemopoietic recovery is less sensitive to radiation than previously thought. The research of the group in Ulm has clearly demonstrated the importance of the migration potential of hemopoietic stem cell in the mammalian organism. Thus, any inhomogeneity of radiation exposure to the human organism works in favour of the spontaneous potential of the hemopoietic system to recover.

Therefore, it is of vital importance to improve existing and develop new tools in order to recognize the response of the human organism towards external and internal radiation exposure. The key question is to assess as early as possible (within hours after exposure) whether an organism has the potential for spontaneous recovery or whether it is necessary to apply therapeutic measures such as bone marrow-, blood- or cord blood-stem cell transplantation. In addition, the previous experience has shown that very often in accidental situations rescue personnel and clean-up workers have to enter in a planned way radiation fields. It has also become evident that in situations of large nuclear catastrophy situations the public will be involved to a smaller or larger extent. Therefore, it is essential to develop better diagnostic tools to recognize not only acute radiation effects, but also biological effects of repeated or chronic low level radiation as a basis to plan and administer therapeutic measures.

With this in mind, the 4 participating groups are planning to contribute to this particular field of research. The group in Rijswijk proposes to study the use of hemopoietic growth factors to stimulate the stem cell system for autochtonous regeneration. The group is planning to apply an optimal combination of growth factors to study the prognostic value of the early response to such factors with the question in mind whether the early response is useful to distinguish between patients which show a rapid recovery using converentional supportive care or whether the number of residual stem cells might be insufficient for an adequate growth factor response. In addition, the Rijswijk group will address transient engraftment of positively selected hemopoietic stem cells as a means to overcome periods of profound pancytopenias, electively supported by growth factor treatment.

The group in Ulm proposes to study the complex signs, symptoms, and blood cell deviations after accidental overexposures and to establish a system of early indicators relevant for answering the question whether the damage inflicted to the immunohemopoietic, the respiratory and the gastrointestinal systems is reversible. This group, in addition, is suggesting to further develop indicators for DNA damage in individual cells in order to relate such DNA damage after acute or protracted irradiation to other prognostic indicators. It is of great importance for this group to answer the question whether hemopoietic systems have responded to ionizing radiation or not and whether residual damage is likely to persist, resulting in the development of non-neoplastic or neoplastic lesions, such as hemopoietic failure due to leukemia or aplastic anemia.

The group in Italy proposes to use certain indicators of the immune system to assure whether this system has responded to high or low level radiation or not which is of great importance with respect to the specific resistence of the organism towards infections.

The group in Paris is suggesting to investigate the efficiency of growth factors and to develop models to relate biological indicators to physical dosimetry.

All in all it is expected that the research activities of the participating laboratories will greatly improve possibilities of the medical profession to recognize the biological consequences of accidental whole body radiation exposure and to establish prognostic criteria enabling the medical profession to better plan and execute therapeutic measures.

Prevention of the radiation syndromes:
(see table 1, part 3)

The reports of the Chernobyl catastrophe and of the Goiania accident have indicated that one has to consider not only the persons involved directly as a consequence of the accidental radiation exposure. But in addition there will be "clean-up" operations that might persist for a long time as shown in the Chernobyl reactor facility. In Chernobyl, even 5 years later, the clean-up operations have by no means been finished yet. They require again and again that persons take the risk of being exposed to substantial amounts of radiation. In addition, the population surrounding the radiation facility has been exposed to elevated levels of radiation ever since. The same was true in the Goiania accident also where a large number of people received overexposures even after the radiation source had been discovered.

Under these circumstances a new field of research becomes evident. Can one, and if so with what possibilities, decrease the effect of radiation and increase the potential of survival and of a "restitutio ad integrum" for the persons involved?

The 4 participating groups propose to study this question in detail. All groups are suggesting to use growth factors prophylactically to prevent the development of radiation lesions. However, each group will use its particular biological models and test systems to complement and not duplicate the efforts.

The group in Paris is proposing to study certain enzymes in order to prevent the development of fibrotic changes in lung and of vascular late effects. The group in Rijswijk and in Ulm are studying the hypothesis that the prophylactic administration of hemopoietic growth factors may alter the sensitivity to total body irradiation and is prepared to study this question in the preclinical rhesus-monkey and the dog models. In addition, the group in Ulm wants to study this question in patients in cooperation with the autologous blood stem cell transplantation group at the MD Anderson Hospital in Houston directed by Dr. Martin Körbling, whereas the Rijswijk group has a strong collaborative tie with the Hematology Department of the Rotterdam Cancer Center and Academic Hospital, headed by Dr. B. Löwenberg. Again, it is the question whether the prophylactic administration of recombinant growth factors will alter the radiation sensitivity and improve the chance for a spontaneous stem cell recovery.

Therapy of the radiation syndromes:
(see table 1, part 4)

Of particular importance is, of course, the question whether one can improve the presently available therapeutic possibilities for the treatment of patients with acute radiation injury or develop new approaches.

All 4 groups will contribute to this specific area with their own approaches that are complementary to each other. The group in Rome is proposing to study the improvement of the rapidity for HLA typing for preparing for bone marrow transplantation. Indeed, it is essential to know as early as possible whether there is a potential donor for a given radiation accident victim. This is of great importance for the entire strategy of therapy planning.

The group in Rijswijk is proposing to study the combination of recombinant hemopoietic growth factors to accelerate the autochtonous regeneration of hemopoiesis even if only very few stem cells are left.

The group in Ulm will study the question whether the use of recombinant growth factors is influencing the migration of stem cells between different hemopoietic sites and thereby will enhance hemopoietic recovery. Along the same line of thinking, the group in Ulm is suggesting to use hemopoietic growth factors to mobilize stem cells into the peripheral blood for easy collection and storage.

This leads to the approach of the Rijswijk group to improve the possibilities to replace conventional intensive supportive care by the use of specifically selected hemopoietic growth factors. The group in Paris wants to concentrate on new approaches for the allogeneic repopulation of radiation damaged bone marrow. This group is suggesting to use cord blood cells in order to study a new source of hemopoietic stem cells. The group in Ulm is proposing to study new approaches to use blood derived stem cells for bone marrow reconstitution. This work is planned to be carried out in collaboration with the autologous blood stem cell transplantation unit at the MD Anderson Hospital in Houston. The group in Rijswijk and in Paris will give particular attention to use purified stem cells for even a temporary engraftment considering in particular, the chances to prevent graft versus host disease.

Table 1: Survey of the Scientific Programme

	Prof. Jammet FRANCE	Dr. Wagemaker NETHERLANDS	Prof. Covelli ITALY	Prof. Fliedner GERMANY
1. Pathophysiology of radiation induced alterations of 1.1 Immuno-hematopoietic system 1.2 Respiratory and gastrointestinal systems 1.3 Central Nervous System	- Radiation induced depression of different cell lines of blood cell formation with particular emphasis on interaction of stem cells and stroma cells. Kinetics of various categories of lymphocytes. - possibilities and limitations to tolerate single, repeated radiation exposures in relation to dose and dose rate - development of radiation induced changes in the CNS: neurological and psychic consequences. EEG changes	- Reassessment of the radiation sensitivity of hemopoietic stem cells - Influence of intensive care on LD50 of bone marrow damage	- The influence of ionizing radiation on functions of lymphocyte populations	- Replication and differentiation of hemopoietic stem cells after total and partial body irradiation - kinetics of hemopoietic reconstitution after total and partial body irradiation: development of biomathematical models to simulate pathophysiological courses of events to assess the remaining and initiating stem cell pool - assessment of radiation damage to the microenvironment of the bone marrow, the lung and the GI system
2. Diagnosis of the radiation syndromes: preclinical and clinical approaches	- Indicators of TBI and PBI. Indicators of efficiency of growth factors, models relating biological indicators and physical dosimetry.	- Stimulation by hemopoietic growth factors a test for the capacity of autochthonous regeneration	- Biological indicators to evaluate the radiation induced damage of the immune system and to predict the pathological consequences	- Blood cell deviation (quantity, quality) as prognostic indicators for or against stem cells transplantation (granulocytes, blood stem cells, platelets) - Chromosomal and single cell DNA injury changes as indicators for hematopioetic cell system damage

More →

3. Prevention of the radiation syndromes	- Use of enzymes to prevent the development of fibrotic changes (lung, skin) and of vascular late effects	- Hemopoietic growth factors as "radioprotectors"	- Recombinant cytokines as radioprotectors	- Prophylactic administration of growth factors to prevent radia-tion hemopoietic faailure - Research on the stablishment of "stem cell banks" for persons "at risk"
4. Theraphy of the radiation syndromes	- Methods for inhibition of graft versus host disease (GvhD) in-cluding selective elimination of lymphocytes - Improvement of stem cells transplantation (incompati-ble, puri-fied stem cells, cord blood stem cells) - Use of haematopoietic growth factors - Improvement of radioactive decorporation in order to reduce the radiation effects on haematopoietic and in-mune systems	- Acceleration of autochto-nous regeneration by hemo-poietic growth factors - Possibilities to replace con-ventional intensive supporti-ve care by a specific combi-nation of hemopioetic growth factors - Transient engraftment of pu-rified stem cells	- Modulation of the immune response by recombinant cytokines - Improvement of the rapidity in HLA typing for preparing for bone marrow transplan-tation - Selection of bone marrow donors with appropriate immune responsiveness to prolong survival and reduce morbidity of radiation chi-meras	- Evaluation of drugs that en-hance the endogenous pro-duction of hemopoietic growth factors - Stem cell mobilization into the peripheral blood by drugs or by growth factors - Improvement of approaches to use blood derived stem cells for bone marrow re-constitution

Contribution of Department of Clinical Physiology, Occupational and Social Medicine, University of Ulm, Germany

The Ulm group is prepared to contribute to the Collaborative Research Programme on the Management of Accidentally Exposed Persons in 4 problem areas. (1) The Ulm group will study new approaches to establish a procedure which allows gene expression analysis at the level of a single or a few cells as a prerequisite to study gene expression of hemopoietic stem cell subpopulations under physiological and pathophysiological conditions. In addition, using new computer based cell system simulation models (develped in cooperation with the University of Ulm Department of Electrical Engineering) the Ulm group will study the reconstitution kinetics of hemopoietic cell systems as observed after accidental and therapeutic whole body irradiation with and without stem cell transplantation. (2) As a contribution to area 2 (diagnosis of the radation syndromes: preclinical and clinical approaches), the Ulm group will study the most promising approaches to analyse early after accidental radiation exposure (within 72 hours), whether one is dealing with a reversible or irreversible damage at the hemopoietic stem cell level requiring either a sophisticated "supportive therapy" or a "stem cell transplantation therapy". On this basis, it is suggested to investigate new approaches to support clinical decision making in radiation accident situations which will be based on pathophysiological models of cell renewal systems. (3) The Ulm group will also contribute to the field of prevention of the development of lethal forms of the acute radiation syndrome by investigating the possibilities to establish "stem cell banks" for persons "at risk". (4) As far as the therapy of the radiation syndromes is concerned, the Ulm group will perform preclinical and clinical studies on drugs that may enhance the endogenous production of hemopoietic growth factors, on the migration and mobilization of stem cells in the peripheral blood and on the appropriate use of blood derived stem cells (purified by new technologies).

Contribution of ENEA, Cassacia, Italy

The work will be carried out at ENEA Casaccia as a part of the collaborative programme for Research on the Management of Accidentally Radiation Exposed Persons. The research is focussed on the problems related to damage and recovery of the immune system after radiation exposure. This aspect is of particular importance for the treatment of radiation accidents because they result in a decreased resistance to pathogens and in development of auto-immune diseases and cancer. The adopted experimental approach, using genetically homogeneous animals and standardized irradiation and treatment conditions, is likely to provide reproducible data to evaluate the consequences of radiation accidents and to design appropriate strategies for effective medical intervention.

Mouse populations will be studied after whole-body exposure to acute X-ray doses ranging from 2 to 10 Gy. Irradiated mice will be left untreated or, starting immediately after irradiation, will be treated by one or more injections of immunoregulatory molecules such as recombinant cytokines. Untreated and treated survivors will be studied to assess the number and activity of blood leukocytes and to evaluate the immune functions of spleen lymphocytes (mitotic responses to Con A and LPS, antibody production, T helper cell activity, cytokine production and expression of their receptors). Comparison of the effects of various treatments after different radiation doses should indicate the conditions for optimal intervention to accelerate recovery of the immune system and to prevent radiation death.

Cell typing for bone marrow transplantation in humans requires rapid and precise techniques. Currently available alloantisera are not always specific enough to identify single antigenic specificities and are difficult to obtain from multiparous women or from volunteers subjected to planned immunization. Also mouse monoclonal antibodies (mAb) do not always distinguish between HLA class I or class II alloantigens. Conversely, human transformation and subculturing, are more powerful than mouse mAb to identify HLA alloantigens and will be investigated. Another approach, which will be pursued, consists of the amplification of small amounts of DNA by polymerase chain reaction and subsequent hybridization by allele or sequence specific oligonucleotide probes. These ongoing techniques will be improvedby increasing the rapidity of their application and by use of non-radioactive reagents such as biotin and avidin. This research on cell typing will be carried out in cooperation with Prof. G.B. Ferrara from the Cancer Institute of Genoa.

Contribution of Erasmus University Rotterdam and Department of Radiobiology, Rijswijk, The Netherlands

The program of the Rotterdam/Rijswijk group is directed at radiation induced damage and recovery of the hematopoietic system. A major problem in the management and treatment of heavily irradiated victims of a radiation accident is the identification of patients that will recover spontaneously following conventional treatment and those that will need a bone marrow transplantation for recovery. The program is directed at answering the following questions by prospective experimental studies in rhesus monkeys:

1 What is the maximum achievable LD50/30d for whole body irradiation by giving maximal supportive care?
2 Which combination of hemopoietic growth factors will shorten the duration of pancytopenia at TBI doses > 6 Gy?
3 Will the optimal combination of growth factors significantly shift the LD50/30d without supportive care, which is of practical importance in case of large scale accidents?
4 Will the early response to an optimal combination of hemopoietic growth factors serve as an indicator for the dose of TBI received, i.e. as a biological indicator for bone marrow damage of sufficient accuracy for the development of an individual management strategy?
5 Will the use of CD34 positive bone marrow grafts at higher doses of TBI (> 7 Gy), aiming for transient engraftment and/or partial hemopoietic chimerism, elective supplemented with growth factor treatment, facilitate the management of high dose radiation victims?

The data obtained with the rhesus monkey model will be compared with the hematological data of human radiation accident victims, that are being collected and analyzed at Ulm to design recommendations for the treatment of radiation victims. In the past contract period, studies relevant to objectives 2-4 were completed with respect to the radioprotective properties of GM-CSF and IL-3, either alone or in combination, and studies were initiated on the radioprotective properties of IL-6 and combinations of several doses of SCF and IL-3. Results obtained with IL-6 were extremely encouraging in that IL-6 appeared to promote the recovery of immature bone marrow cells after TBI, in addition to promoting the recovery of thrombocytes. In the recovery phase after TBI,

up- and/or down-regulation of growth factor receptors (to help predicting which growth factors will be important to accelerate recovery) can now routinely be applied for IL-3, IL-6, SCF, GM-CSF, G-CSF and erythropoietin. Relevant to objective 5 is the improvement of the quality of CD34 positive cell preparations by the availability of a panel of CD34 positive cells directed against different epitopes of the CD34 antigen and the further subfractionation of CD34 positive cells to identify the long-term repopulating stem cell.

Contribution of Centre International de Radiopathologie, Paris, France

Until the end of 1991 clinical researches have been carried out from patterns of total body irradiation. In 1992 we will have the use of a source of Cobalt 60 specifically allocated to clinical research in radiopathology. It will allow to use new technics of exposure on the spatiotemporal level.

1. Physiopathology

Concerning the study of the interaction between stem cells and stroma medullary cells, it is known that "in vitro" stem cells cultures in the presence of stroma cells do not give satisfying results. We propose the in vivo primary introduction of stem cells in the scid mouse and the in vitro secondary amplification in the presence of macrophages, endothelial cells, fibroblasts and human lipocytes.

The clinical studies on the kinetics of different categories of lymphocytes after total body irradiation will be continued while using new spatiotemporal modes of exposure. The expected results should allow to quantify the role of this two parameters in man. Researches will also be made on thephysiopathological consequences, specially on the induction and evolution of lung radioinduced fibrosis according to the irradiation modes.

During these irradiations the brain dammage expressed by the EEG will serve as a basis for an experimental study of the lesions induced in the encephale and of the distribution of the neuromediators.

2. Diagnosis on the radiation syndrome

According to the conditions of irradiation the dose-effect curves for the cortisol, the ACTH and the serious analysis will be established. IL1, IL2, IL6, TNFÓ and GMCSF will also be analysed. It has been observed that the IL6 can be significantly increased in certain patients submitted to total body irradiations.

The retrospective dosimetry by cytogenetic analysis will be systematically carried out in order to study the role of the irradiation models.

In any case, a physical dosimetry will be carried out in order to get the most of the post-accidental dosimetric reconstruction.

3. Prevention

It has been shown that the administration of super-oxyde-dismutase modifies the evolution of the seated radioinduced fibrosis. If the lung investigation showed the development of a radioactive lung fibrosis, we envisage to give the SOD precociously in order to study if this enzyme is able to stop the process of evolution.
Concerning the experimentation on the mouse irradiated with a lethal dose, we propose to use metallic complexes such as copper disopropyl salicylate, to stimulate the hematopoiesis and increase the number of graft samples.

4. Therapy

In order to destroy the allo-reactive T lymphocytes at the origin of the GvH reaction, it is envisaged to use an anti-body, anti IL2R, coupled with ricine. This study will be carried out on the mouse irradiated at 9 Gy.

The researches on the use of the funiculus cells will be continued in man as well as in animals to define the immunological properties of the neonatal cells, to purify the hematopoietic stem cells and to study the placenta's growth factors.

We will do research work on the time interval during which it is still possible to carry out an exact HLA typing, according to the spatiotemporal conditions of the exposure.

There are radio-elements such as rare earths and actinides which have a tropism for the hematopoietic tissues or their environment. We will look for the possibilities of using new complexing molecules in the series of the Licam, in animals.

B21 Radiation syndromes and their treatment after exposure of large parts of the body.

Contract FI3P-CT930069 Radiation effects and their treatment on the connective and vascular tissues in various organs.

Coordinator CIR
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Total Contribution by the Commission: 65 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

1 - Objectives and expected achievements: the objective of the projects is the development of new methods of diagnosis, prevention and treatment of fibrosis, sclerosis, thrombosis, necrosis and associated lesions in various organs following radiation overexposure, based on an increased knowledge of physiopathological alterations. The organs or tissues considered will be the lung, lens, osseous and tendinous tissues, muscles and brain. Haematopoietic tissue and skin will not be considered. The expected achievements of the project are i) a better assessment of the evolutive potential of lesions to the vascular and supporting tissues and its implication on the organic function of irradiated organs, ii) a better knowledge of the mechanisms of action of compounds capable to prevent or reverse such lesions, and iii) new modalities of diagnosis, prevention and treatment of radiation induced lesions to these organs.

2 - State of the art : whereas much research effort on radiation induced fibrosis concentrates on skin, little is known on the contribution of radiation induced damage to vascular and connective (or supporting) components of other organs to their atrophy or loss of function after overexposure. There is however increasing evidence, supported by recent biochemical or physiological investigations, that alterations of mesenchymal tissues contributes to alterations of epithelial tissue homeostasis. New tools are now available to study at the molecular level these interactions in organs other than skin after irradiation. Some compounds have proven efficacy in prevention or treatment of fibrotic process in irradiated skin, but the demonstration of their activity in other organs is still lacking and suffers from delivery problems.

3 - Degree of innovation : still little knowledge is available in the physiology of vascular and connective tissues of internal organs after irradiation. The expression of growth factors, oncogenes, etc..., is a new approach to investigate alterations of these tissues. New antidegenerative compounds have to be tested experimentally (SOD on glial cells,...), and new modalities of delivery have to be evaluated in controlled clinical protocols for prevention and the treatment of radiation damage. A specific protocol of SOD delivery to the lung by aerosol will be evaluated in the treatment of radiation induced lung fibrosis.

4 - Economic, social and technical benefits : early diagnosis and efficient treatment of radiation induced lesions of vascular and connective tissues will spare or limit accompanying or subsequent alterations of vital function of irradiated organs.

5 - Scientific and technical description: the scientific and clinical project comprises:

- Pathophysiology of radiation effects on tissue biopsies, cells in culture and animal models
- Clinical and paraclinical (physical, biochemical methods of diagnosis of fibrosis, thrombosis and necrosis of internal organs), including IRM
- Prevention of radiation syndroms related to fibrotic or thrombotic process in internal organs

- Treatment of irradiation syndromes in internal organs.

6 - Detailed distribution of tasks among participating organizations

Experimental Research :

CIR (Paris): - mesenchymal-epithelial cells interaction in cell culture after irradiation. Role of paracrine action of growth factors.

University Hospital (Nijmegen) : role of oxidative injury and oxygen radical scavenging mechanisms in radiation injury of the CNS.

Clinical Research :

CIR (Paris): - histopathological parameters of fibrotic evolution in tissue biopsies

- physical methods of diagnosis (IRM) in lung, muscle and bone

- treatment of radiation induced fibrosis of the lung by SOD

University Hospital (Nijmegen) : non invasive (MRI and MRS) evaluation of brain injury after radiotherapy.

7 - Complementarity and benefits from the collaboration : both Institutions have complementary experimental and clinical expertise concerning radiation damage to various tissues and organs. The group from Paris will share its experience in biochemical diagnosis and treatment. The group from Nijmegen will share its experimental data for the development of new treatments.

8 - Project management : CIR is the coordinator of the project. Experimental and clinical data will be exchanged regularly.

Contribution of the CIR (PARIS)

Radiation effects on the connective and vascular tissues in various organs.

Due to their relative radiosensitivity, radiation-induced damage of the vascular and connective tissues largely determines the subsequent atrophy and loss of physiological function of various organs. Clinical and accidental situations of overexposures encountered at the Institut Curie (Paris) allows the study of the human physiopathology and the evaluation of preventive or curative treatments of radiation overexposures to the lung, lens, osseous and tendinous tissues, muscles and vascular and glial tissues of the central nervous system.

1 - Pathophysiology of connective and vascular tissues in irradiated organs

Histochemical methods, including immunohistochemistry and in situ hybridization, and biochemical methods, including ligand binding or bio- or immuno-assays will be used on tissue biopsies. The analyses will focus on the characterization of altered paracrine interactions between mesenchymatous and epithelial cells : growth factors (EGF, TGFα, PDGF, FGF, TGFß, NGF, ...) and their specific receptors. The relation between these alterations and those of organic function, differentiation or induced cell death will be studied.

2 - New paraclinical investigations

The comparative worth of modern techniques of deep tissue investigation (SPECT, PETscan, CTscan, NMRI, NMRspectroscopy, ultrasonography) will be evaluated in the diagnosis and follow up of treatment of fibrosis, sclerosis, thrombosis or necrosis in irradiated lung, lens, skeleton, muscles and brain. The vascular component to radiation induced lesions of the CNS will be analyzed.

3 - Prevention and Treatment

There is now convincing evidence that radiation induced damage to mesenchymal tissues can be prevented, limited or even reversed by superoxide dismutase (SOD). Various modalities of delivery will be developed according to the targeted organ : aerosols for lung, systemic or intramuscular infusion for deep lesions. Delivery to the CNS will be envisaged following the results of the companion project from University Hospital-Nijmege. The efficacy of treatment of radiation damage to the lens with recombinant growth factors will be evaluated.

4 - Experimental research

The effect of experimental cranial irradiation on brain regional capillary permeability and ageing in the rat (Dr JY Delattre).

Contribution of the University Hospital, Institute of Radiotherapy (NIJMEGEN)

Radiation injury in the CNS : clinical and experimental studies on the modification of oxidative injury combined with non-invasive monitoring by NMR techniques.

This part of the collaborative project will focus on the central nervous systems as a model of early and late delayed radiation injury, in particular on the role of connective (glial cells and their precursors) and vascular (endothelial cells) tissue elements and their interactions.

The components of the study are aimed at a maximal interaction and parallelism with the project carried out at the Institut Curie/CIR. In addition to some common approaches (MRI and MRS as noninvasive diagnostic tools to study pathophysiology of radiation injury, use of superoxide dismutase for *in vivo* prevention and treatment), for several parts of the study the work in Paris will concentrate on the *in vivo* aspects, while the *in*

vitro counterpart in Nijmegen will employ primary cultures or co-cultures of glial and endothelial cells.

In the two-year project, two key areas will be pursued :

1. Pathophysiology of glial and endothelial cells : the role of oxidative injury and oxygen-radical scavenging mechanisms in radiation injury in astrocytes, oligodendrocytes and glial progenitor cells.

We will combine this project with elucidating the role of different modes of cell death (mitotic and apoptotic) in specific glial cell populations as preliminary work has shown large differences in the relative contributions of these types of cell death after irradiation. The role of SOD and other antioxidant enzymatic systems (catalase, GSH-reductase) will be elucidated in the context of the various modes of cell death. It is envisaged that in a later phase of the study primary cerebral endothelial cell cultures, as well as astrocyte/endothelial co-cultures will be added to these studies. The development of these latter *in vitro* models are currently performed in the laboratory in Nijmegen. In particular, the role of TNFα (produced by astrocytes) in cellular necrosis and changes in endothelial permeability are studied in a parallel project not included in the present proposal.

2. Clinical studies : non-invasive (MRI and MRS) evaluation of brain injury after radiotherapy in combination with administration of nicotinamide and carbogen breathing.

This new rationale of brain tumour treatment has recently been started in Nijmegen University Hospital as an attempt to overcome tumour hypoxia by relatively non-toxic additives of radiotherapy. However, in addition to an enhanced tumour effect also an enhanced toxicity of normal brain tissue has been observed in animal studies. This indicates that under normal conditions the brain may be (slightly) protected by radiobiological hypoxia. In the clinical study ^{1}H & ^{31}P-MR spectroscopy will be used to measure biochemical changes pertinent to radiation-induced injury. In addition measurements will be made in the rat brain using a 6.3 Tesla system for animal experiments. These studies will be combined with the superoxide dismutase investigations in animals, and possibly in humans in a later phase conjunction with the Paris project.

Histochemical methods, including immunohistochemistry and in situ hybridization, and biochemical methods, including ligand binding or bio- or immuno-assays will be used on tissue biopsies. The analyses will focus on the characterization of altered paracrine interactions between mesenchymatous and epithelial cells : growth factors (EGF, TGFα, PDGF, FGF, TGFß, NGF, ...) and their specific receptors. The relation between these alterations and those of organic function, differentiation or induced cell death will be studied.

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vitro counterpart in Nijmegen will employ primary cultures or co-cultures of glial and endothelial cells.

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B22 Irradiation and committed exposure from incorporated radionuclides.

Contract FI3P-CT920064b Reduction of risk of late effects from incorporated radionuclides.

Coordinator NRPB
GB-OX11 0RQ CHILTON, DIDCOT
Tel. 44-235831600

Total Contribution by the Commission: 170 KECU
23 months 1/07/92 to 31/05/94

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Description of research work

1. OBJECTIVES

The aims of the studies with laboratory animals described in this project are to assess the reduction of risk resulting from treatment of incorporated radionuclides and to provide guidance to those involved in the treatment of accidental overexposure. The work will involve experiments designed to

- optimise the efficacy of 3,4,3-LIHOPO for enhancing the excretion of plutonium, americium and thorium after uptakes of various chemical forms by intravenous injection, inhalation and wound contamination. The toxicity of the substance will also be investigated
- optimise the efficacy of orally administered DTPA for enhancing the excretion of inhaled transportable forms of plutonium and americium
- synthesise and test new substances for removing uranium from the body after the injection and inhalation of transportable forms.

In addition, studies carried out by others in this field will be kept under review and appropriate work will be initiated to substantiate reports of significant developments.

2. GENERAL DESCRIPTION OF THE PROJECT

The current agents of choice for enhancing the excretion of plutonium (Pu) and americium (Am) from the body after overexposure to their transportable forms are trisodium calcium or zinc salts of diethylenetriaminepentaacetic acid (DTPA). They are usually administered by slow intravenous injection. However, the ligands are not completely effective and some of the most severe cases of internal contamination have required their repeated administration over months or years. The development of superior substances is considered an important aspect of radiological protection. During the last decade increasing interest has been shown in the potential therapeutic uses of synthetic analogues of siderophores. Several of them have been tested in experimental animals eg. LICAM(C), DTPA-DX, DFO-HOPO. However, the most promising ligand identified so far is 3,4,3-LIHOPO.

Studies carried out with 3,4,3-LIHOPO under a previous CEC-NRPB Association Agreement have shown that it is substantially more effective than DTPA for enhancing the excretion of plutonium after inhalation as nitrate and tributylphosphate, and after its intravenous injection as citrate. The ligand is also effective for americium inhaled as nitrate. Significantly, a pilot experiment has shown that 3,4,3-LIHOPO also mobilises thorium deposited in the lungs. This is important since no effective treatment for inhaled thorium is presently available.

Together the above observations could represent a most important development in the reduction of risk of late effects from intakes of plutonium, americium and thorium. One of the aims of the proposal for 1992-94 is to optimise the efficacy of 3,4,3-LIHOPO for these actinides. Due account will be taken of the different chemical forms of the actinides, their route of incorporation particularly by inhalation and wound contamination, the variations in mass concentrations of the actinides at the site

of entry which reflect different accident scenarios, different routes of administration of the ligand eg by injection, infusion, orally or as an aerosol, and the dose effectiveness of the ligand.

The toxicity of 3,4,3-LIHOPO with particular emphasis on pathological damage to the lungs, liver, kidneys and gastrointestinal tract and its effect on the excretion of essential trace metals will also be evaluated.

The oral administration of DTPA for decorporating plutonium and americium has the advantage that it can be self administered, it is more likely to be accepted by the patient than other treatment regimes and it is particularly relevant for accidents involving large numbers of people.

Experiments performed under the 1990-92 contract showed for the first time, that the administration of DTPA in drinking water was an effective method for removing inhaled transportable forms of plutonium and americium from the body. However, further studies are needed to ascertain the optimum treatment regimen and the toxicity of the ligand after this mode of intake.

Accidental overexposure to industrial uranium compounds is becoming of increasing concern yet no effective treatment regimes are currently available. Studies performed under the 1990-92 contract show that treatment using tiron and certain phosphonate derivatives were only partially effective.

More effective compounds are considered a priority and in 1992-94 the work will include the screening of a selected number of polyaminopolyalkylpolyphosphonic acids, bisphosphonates, phosphonoalkylphosphinates, and the 'clathrate type' calixarenes with a known or expected affinity for uranium.

For convenience, the substances will be tested initially after the injection of uranyl nitrate. The most promising substances will subsequently be investigated after the alveolar deposition of other transportable industrial compounds such as UO_3, UF_4.

3. INTERACTION BETWEEN PARTNERS

The interaction between the partners is shown diagrammatically in Figure 1. It demonstrates the close cooperation that is required to fulfil the objectives of the project. Interaction in this way maximises the expertise and facilities at each establishment.

The synthesis of 3,4,3-LIHOPO and some of the substances to be tested for uranium decorporation will be undertaken at the Pierre and Marie Curie Univ; other substances for uranium will be prepared by SHI Pierrelatte or obtained from elsewhere.

The toxicity of 3,4,3-LIHOPO will be examined by NRPB and CEA Bruyères-le-Châtel.

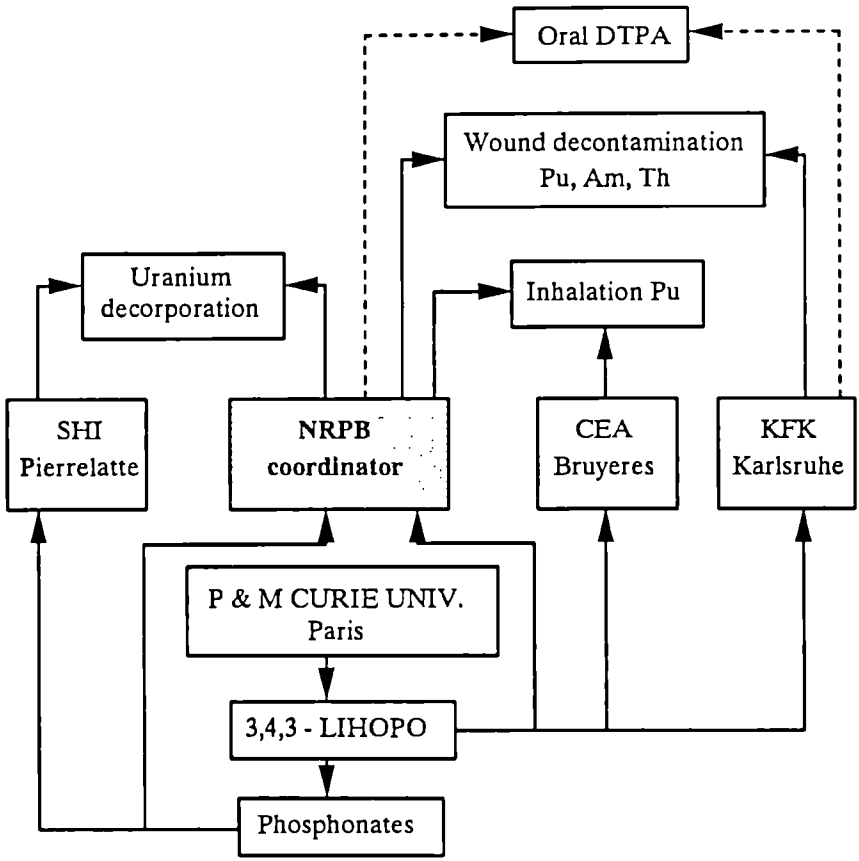
The NRPB and CEA will collaborate on optimising treatment regimes with 3,4,3-LIHOPO after inhalation of plutonium in different chemical forms; inhalation studies with americium and thorium will be undertaken at NRPB.

Treatment with 3,4,3-LIHOPO after wound contamination by plutonium, americium and thorium will be investigated at KfK and NRPB.

NRPB and KfK will collaborate on studies designed to investigate efficacy and toxicity of orally administered DTPA.

The SHI Pierrelatte in collaboration with NRPB will examine substances for enhancing the excretion of uranium from the body.

Figure 1. Interaction between Partners



Other collaborators:

Univ. Paris Nord: Collaborate with P & M Curie Univ. on synthesis of LIHOPO and phosphonates
Dept. of Chemistry, Univ. of California, Berkeley: Synthesis of siderophore analogues for actinide decorporation
Lawrence Berkeley Laboratory: Screening of siderophore analogues
Shanghai Medical University: Synthesis and screening of substances for thorium
Norwich Eaton Pharmaceuticals Inc., New York: Synthesis of phosphonic acid derivatives for uranium
Albright & Wilson plc, UK: ibid
Univ. Louis Pasteur, Strasbourg: ibid
Churchill Hospital Oxford: Toxicity of chelating agents
Institute of Hygiene and Epidemiology Prague: Decorporation of thorium

CONTRIBUTION OF THE NATIONAL RADIOLOGICAL PROTECTION BOARD

The NRPB will act as the coordinator for the contract. The NRPB will be involved in experiments designed to optimise the efficacy of 3,4,3-LIHOPO for removing plutonium, americium and thorium from rodents. The studies will involve incorporation of the actinides by inhalation and wound contamination and the administration of the ligand by injection, infusion or as an aerosol. Limited studies on the toxicity of the ligand will also be undertaken. The mass concentrations of the actinides in the lungs will simulate realistic accident scenarios and those for thorium in particular will differ by several orders of magnitude cf ^{228}Th and ^{232}Th.

The NRPB will also undertake experiments designed to optimise treatment with orally administered DTPA for removing inhaled transportable forms of plutonium and americium from the body. The toxicity of the substance after this route of administration will also be investigated.

The NRPB will examine the efficacy of the most promising substances for enhancing the excretion of uranium. The substances must be provided by the P & M Curie Univ and others. Initial testing will involve the administration of uranyl nitrate but the work will be extended to include inhaled transportable forms such as UO_3, UF_4 when significant progress has been made.

CONTRIBUTION OF KERNFORCHUNGSZENTRUM KARLSRUHE

The work at KfK is intended to consolidate and to complete the present studies on the efficacy of 3,4,3,-LIHOPO, the most effective substance yet tested, for the reduction of the body content of plutonium and americium following accidental intake. Studies will be carried out in rats exposed under the conditions simulating either a single intake (intravenous injection) or continuous absorption into the bloodstream from a contaminated wound (intramuscular injection). The work will concentrate on the optimisation of treatment schedules by administration of the chelating agent either by continuous infusion, via "minipumps", by local treatment of a wound site, or by oral administration in drinking water.

The work will also be extended to include investigations of the ability of new chelators, such as 3,4,3,-LIHOPO and various new substances synthesised recently by colleagues in the USA and China, to reduce the body content, and hence the risk of late radiation effects, following accidental intake of thorium. Satisfactory treatment schemes for thorium contamination have not yet been developed. The studies will be carried out in rats injected intramuscularly with thorium in order to simulate a contaminated wound.

CONTRIBUTION OF COMMISSARIAT A L'ENERGE ATOMIQUE

Studies at CEA will concentrate on the efficacy of the siderophore analogue 3,4,3-LIHOPO for removing plutonium (^{238}Pu) after inhalation in rats as Pu-tributylphosphate complex. This form is described as the most difficult compound to decorporate, especially after an internal contamination by a large amount of plutonium.

Experiments will be undertaken to define the most effective treatment by the chelating agent in terms of the route of administration (intravenous, intramuscular or inhalation), concentration of the ligand (3 to 30 μmole kg^{-1}) and schedule for the treatment.

The 3,4,3-LIHOPO, synthesized by the chemistry staff (P&M Curie University), will be compared to the DTPA, as the reference molecule.

The toxicology of the substance will be investigated in rodents and non-human primates after different routes of administration. Pathological damage in organs (lung, kidneys, liver, gastrointestinal tract) and cellular lesions will be analysed by classical histology and electron microscopy.

CONTRIBUTION OF SERVICE D'HYGIÈNE INDUSTRIELLE - PIERRELATTE

The aim of the work of SHI is to synthesize and test new molecules to chelate the uranyl ion.

The SHI Pierrelatte will synthesize new substances for enhancing the excretion of uranium from the body. The stereochemistry of the molecules synthesized will be taken into account.

For the molecules synthesized at SHI Pierrelatte and at the University Pierre et Marie Curie, the complexation constant between the ligand and uranium, and the effect of pH on the stability of the complex will be determined, in order to test *in-vivo* only the most promising substances.

Initially, the SHI will examine the efficacy of these substances after injection of uranyl nitrate. The efficacy of different routes of administration such as injection and infusion will be investigated for the most promising materials. Subsequently, these will be tested after inhalation of different chemical forms of uranium dusts including UO_3 and UF_4.

CONTRIBUTION OF ADFAC, LABORATOIRE DES ORGANOELEMENTS PARIS

One important part of the work will involve the synthesis of 3,4,3-LIHOPO. The substance will be used at NRPB Chilton, CEA Bruyères-le-Châtel and KfK Karlsruhe for studies of its efficacy in rodents and, in association with CEA Fontenay-aux-Roses, for the evaluation of its toxicity in baboons.

New, sterically rigid, chelating agents will be synthesised for the decorporation of uranium. Essentially these substances consist of three molecular fragments, a diphosphonate group acting as the complexing centre, an organic group acting as an orientation carrier and a linker or spacer group connecting the other two. The substances will be tested at IPSN Pierrelatte and NRPB Chilton.

B23 Radiation syndromes and their treatment after local exposure to skin and subcutaneous tissues.

Contract FI3P-CT920059 Radiation effects and their treatment after local exposure of skin and sub-cutaneous tissues.

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Total Contribution by the Commission: 275 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Introduction

The aim of the collaborative group is to bring together experimental and clinical researches, performed by the different participants, on diagnosis, prognosis, pathogeny and treatment of local radiation exposure of the skin and subcutaneous tissues.

Acute localized irradiation and external contamination are the most frequent accidental events in radiation protection hazards. Cases of over-exposure of skin and underlying tissues, including skeletal muscles, can occur in both medicine and industry. In a major accident situation, as indicated by experience from Chernobyl and Goiania, significant numbers of persons would be affected. The severity of a radiological damage depends on the energy deposited in the tissues, the dose-rate, the surface irradiated and the radiosensibility of the subjects. Early diagnosis and late effects prognosis are fundamental to evaluate the extent of the injuries and to manage the treatment.

Dose-effect curves have been established for the various endpoints (erythema, dry or moist desquamation, permanent ulcer or healing) from experiments in animal models and from observations in radiotherapy patients. These studies have provided a suitable basis for improving radiological protection criteria for the skin. The pathogenesis of different late effects, including atrophy, fibrosis and sclerosis, observed after local exposure of skin and underlying tissues, has been assessed in the pig, the rabbit and the mouse in the first 18 months after exposure. Skin has been widely used in radiation carcinogenesis studies because of the accessibility and visibility of the tumors in this tissue. Both rat and mouse models have proved to be sensitive, reproducible systems to study the dose and time response of cancer induction following different mode and qualities of radiation exposure.

All these responses have been documented in human skin after accidental overexposure; the clinical studies on the Chernobyl and Goiania accidents victims made obvious combined injury, adding the effects of local and regional or total exposures. The problem of hot particles has been posed in these cases more acutely (than as a hazard in the nuclear power plants).

At least some remarks would sum up the general objective of these joined contracts. Localized overexposure accidents are rather rare and generally due to industrial sources manipulation; their clinical evolutions are dramatic and surgical interventions are accepted rather bilatedly by the patients because of the extremely painful and hopeless nature of the pathological process in these injuries. Experience and knowledge supplied by both clinical following up of such lesions and experimental observations should be useful to give scientific basis for setting a common and well accepted management protocole of these radiological injuries. Studies on the pathogeny, the prevention and the treament of fibrosis, the pre-transformation of certain sort of skin cells, will be directly applicable to the management of the late effects of radiotherapy. Combining clinical and experimental researches worked out by the different contractants seem to be more efficient way to obtain rapidly practical methods for localized radiation injuries management.

Experimental researches

In the pig, after irradiation of a 2 cm diameter area with a single dose of about 120 Gy of either ^{192}Ir γ-rays, ^{90}Sr / ^{90}Y or ^{170}Tm B-rays, acute ulcerations of the skin occur. This effect was more marked after irradiation with ^{192}Ir γ-rays where a 100% incidence of acute ulceration was seen after exposure of a 2 cm diameter area of skin. This 120 Gy dose represents the ED10 for this effect after "hot particle" type exposures from beta-emitting sources.

It is proposed to examine the effects of high dose irradiations to the skin designed to represent "hot spots" in a more generally exposed area. These studies will be carried out in the pig with beta-emitting sources (Oxford). Studies will be developped to evaluate the effects of beta-rays of different energies, and/or small radioactive particulates when combined with partial body ^{60}Co gamma-rays (Saclay) and X-rays irradiation (Oxford). The different responses of the skin and sub-cutaneous tissues will be compared on the basis of a standardized pig skin response to ^{90}Sr / ^{90}Y beta-irradiation between the two groups. The early and late effects of low dose-rate and non-uniform exposure to sources of ^{90}Sr / ^{90}Y will be investigated in pigs to establish the dose effect relationship, and hence to estimate the threshold doses, for acute ulceration, moist desquamation and late dermal atrophy.

On the other hand it is proposed to pursue in the mouse model the synergistic effects of whole body radiation with localized beta irradiation of small areas of skin. An initial protocol involving 20 mice per group has yet begun, with 2x4 mm field plus 0, 1, 2, 4, 6, 8, 9 and 11 Gy of whole body ^{60}Co radiation. The whole body doses were chosen to cover the whole ranges of sublethal to supralethal doses (London).

Diagnosis

Needs for earlier and more accurate diagnosis methods do exist either for the early inflammatory reaction (hyperemia, hyperthermia, oedema), for the secondary reaction (late erythema, ischemia, necrosis) or for the following fibrosis and sclerosis processes. NMR imaging and NMR spectroscopy, blood and lymph flow measurement and gamma scintigraphy seem the best non invasive methods to be developed in animals models before to be validated in man, both for diagnosis purposes and for quantifying the therapeutic assays (Saclay & Oxford).

Cellular and Molecular Mechanisms

Concerning the pathogenesis of early and late radiation-induced damage to the skin, the role of any increased expression of cell adhesion molecules, ICAM-1, ELAM-1 and VCAM on the surface of endothelial cells will be studied. The adhesion of leucocytes to up regulated endothelial cells would appear to be an important initial step in the management of radiation damage to skin (Oxford).

Post-irradiation fibrosis is a severe complication of acute localized irradiation. In pig skeletal muscles the limit of fibrosis expansion was reached at a depth dose of 14 Gy for skin surface doses exceeding 48 Gy. Extensive studies on pig were performed on

fibroblasts isolated from fibrotic tissues. These cells exhibited abnormal proliferation and extensive chromosomes anomalies. Studies on the mechanisms of collagen deposition and growth factor action allowed to understand important control steps of fibrosis development. It is proposed to carry out studies to determine the responsability of the other target cells in the different types of effects observed. Further cellular and molecular studies related to the pathogenesis of radiation damage will be continued. The modifications of gene expression will be assessed for two types of genes: oncogenes and growth factor genes in vivo, in irradiated pig skin and in vitro, in cultured fibroblasts and endothelial cells (Saclay).

In the mouse model, the fibrotic process has only been studied by light microscopy and the progressive nature of dermal fibrosis is well understood. The project involves a cellular and molecular biology programme to link the two effects (early epidermal reaction and late neoplastic change) by studying the chronic promotional hyperplasia that begins with an inflammatory reaction and progresses to dermal fibrosis (London). These intermediate reactions will be monitored, as carried out in the previous pig model, using histology, immunohistochemistry, cell culture and molecular biology techniques, especially TGF-b expression (Saclay & London).

Clinical Researches in Man

Paraclinical investigations will be developed in man both for initial diagnosis and monitoring the efficiency of new treatment modalities: static and dynamic telethermometry, as well as capillaroscopy will benefit from the development of computerized image analysis. Periodic thermographic recording with thermostimulation will be continued in radio-exposed workers to evaluate a possible dose-effect relationship (Roma & Paris). First attempts for using NMR imaging and spectrometry will be carried out (Paris). There is still a need for sensitive and specific indicators of acute local irradiation in easily accessible biological fluids: interleukins (IL1, TNF, IL6) and markers of lipoperoxidation (malonat-dialdehyde, lactoferrine) will be assayed in patients submittted to acute local irradiation therapy (Paris).

Treatment assays

In experimental animals, the treatment of the acute phase symptoms on the one hand and the prophylactic treatment of the late radiation-induced damages will be studied using systemic or topical application of pharmacological agents. Treatments will be directed either towards the reduction of inflammation and oedema, or towards the improvement of the vascular supply, trying to avoid the reperfusion injury: haemorrheological agent, anti-inflammatory and platelet anti-agregant drugs, anti-adhesion molecules and inhibitors of iron-dependent lipid peroxydation (Oxford & Saclay) will be assayed.

Protocols for treatment will be developed by a joint effort of participants with emphasis on optimisation of treatment during the acute and late phases in a way to reduce free radical production, and improvement of different methods to cover the combined lesions such as skin grafts, skin flaps and artificial skin used first as transitory dressing. Encouraging results have been obtained in pigs treated with

liposomal superoxyde dismutase given by intramuscular injections 6 months after irradiation: the reduction of the fibrotic scar was highly significant (Saclay). The wound healing problem in irradiated skin is a further aspect of accidental overexposure. Trauma may be associated with accidental irradiation, but surgery may have to be carried out in irradiated areas to repair necrotic lesions. This will be evaluated in pig skin after irradiation with sources of various energy (to observe lesions at different depth) using a standard surgical wound, with or without different decontamination processes (Saclay).

Protocols for surgical management of radiological burns will be tested taking into account 1) the extent of the lesions according to the depth in the irradiated tissues, 2) the delay for the intervention and 3) the technics used: skin graft, skin flaps, and combined surgical treatment (Saclay & Oxford).

CONTRIBUTION OF CIR-CEN-FAR, FONTENAY AUX ROSES

A) Laboratoire de Radiobiologie Appliquée GIF SUR YVETTE;

Most of the available information in human deals with skin response after high dose irradiations from a collimated photon or electron source, or from uniform beta-emitting plaques. In accidental irradiation such uniform and well delimited exposures are rarely seen, and an experimental approach is essential. Consequently, the early and late non-stochastic effects of radiation will be studied in pigs, which is unanimously considered as the best experimental model.

Pig skin will be exposed to beta-particles (local irradiation) with $^{90}Sr/^{90}Y$, combined with graduated doses of ^{192}Ir or ^{60}Co gamma-rays given on a larger area (regional irradiation). The clinical evolution of the lesions will be followed up and quoted. Quantification of the lesions will be performed by:

- NMR spectroscopy of the skin,
- measuring the drainage ability of the irradiated skin with colloid isotopes clearance,
- evaluating the healing capacity of the irradiated skin after surgical wound,
- measuring early after exposure the skin content of TNFa and TGFB mRNA, and the c-jun, c-fos and p53 gene expression, which were previously seen as dependent of the dose; these early gene expressions will be investigated both in vivo on pig skin and in vitro on cultured human and pig cells.

All these parameters will be assessed with and without pharmacological treatments (topical or systemic) and/or decontamination processes.

On the other hand a dermal equivalent model was developed with human and porcine keratinocytes, fibroblasts and endothelial cells, in order to study changes in the interactions between the different cells, gene expression and protein synthesis (matrix, proteases...) after irradiation in vivo and in vitro.

Fibrosis prevention and treatment will be assayed comparing the effects of different pharmaceutics (corticoids, anti-oxydants, SOD) given by systemic ways.

Irradiated muscle functionnal and metabolic data will be studied combining gamma scintigraphy, electromyography and NMR spectrometry and imaging.

B) Laboratoire de Radiopathologie - Physiopathologie, Institut Curie - PARIS

The ultimate objective of the project is the prevention and treatment of the deleterious effects of acute local irradiation affecting skin and subcutaneous tissues. New and specific treatments can be expected from a better understanding of the pathophysiology of irradiated tissues, in the light of recent acquisitions in modern biology. In this respect, it is also essential to develop or improve the potential usefulness of certain methods of paraclinical investigation such as thermometry, capillaroscopy and nuclear magnetic resonance imaging and spectroscopy. The clinical activity at the Institut Curie finally allows the evaluation of new modalities of treatment (and prevention) of early and late forms of radiation induced damage to the skin.

Pathophysiology of the irradiated skin

We propose to extend our current studies of the physiological alterations of human skin after irradiation (therapeutical and accidental). It is envisaged to extend our studies on the phenotypic and genetic alterations of dermal fibroblast and epidermal cells, with special focus on growth factors and their receptors (epidermal growth factor, EGF, and its receptor, EGFR; fibroblast growth factor, FGF and FGFR; platelet derived growth factor, PDGF and PDGFR), on cytokines involved in inflammation and fibrosis (IL6, TGFb,...), on proteases (plasminogen activators, tPA and uPA). Methodology comprises immunohistochemistry, in situ hybridization, biochemical analysis of altered gene expression (proteins and mRNA) and biological behaviour of fibroblasts and epithelial cells in co-culture.

Functionally related alterations of cell membrane-associated growth factors are alterations in signal transduction (tyrosine kinase, protein kinase C,...) and gene transcription (transcription factors). We propose to investigate the expression and/or the activity of such signal transducers and transcription factors in irradiated skin biopsies and in irradiated skin-derived cultures of dermal fibroblasts.

There is still a need for sensitive and specific indicators of acute local irradiation in easily accessible biological fluids. Interleukins (IL1, TNFa, IL6) and markers of lipoperoxidation (malondialdehyde, lactoferrine,...) will be assayed in patients submitted to acute local irradiation treatment.

Paraclinical investigation of local irradiation

As means of both initial diagnosis and monitoring the efficacy of new treatment modalities, non invasive paraclinical investigations need further developments:

Telethermometry (static and dynamic) as well as capillaroscopy will benefit from the development of computorized image analysis.

Semiology of Magnetic Resonance Imaging (and spectroscopy for certain limbs) of irradiated human tissues need to be developped.

Prevention and treatment of radiation induced fibrosis and radiodermitis

Fibrosis

Superoxide dismutase (SOD) is efficient in reducing radiation induced skin fibrosis in patients treated by radiotherapy (results of a clinical evaluation on over 100 patients). We propose to evaluate the relative efficacy of alternative sources of SOD and of new pharmacological agents, potentially effective in the treatment of skin fibrosis such as antisens oligonucleotides against PDGF receptors.

Radiodermitis

Recombinant EGF, which is available to us in pharmacological quantities, may help heal radiation wounds and will be tested in clinical situation of radiation burns.

Soluble synthetic polymers will be studied as treatment for radiation induced ulcerative wounds.

Contribution of CRC - Normal Tissue Radiobiology Research Group, Oxford

Radiation effects, their diagnosis and treatment after local exposure to skin and subcutaneous tissues.

The contribution of the Oxford group involved in this collaborative programme will be in three main areas:

1. Establishment of dose response relationships for non-uniform exposures to different energy beta-emitters;
2. Pathogenesis of early and late radiation damage to the skin and
3. Prevention of early and late radiation-induced sequelae in the skin.

The majority of these studies will be carried out on the skin of the pig, a species whose skin is most similar to that of man, and from which results will have the greatest clinical applicability.

Dose-effect relationships

Accidental over exposure of the skin may result from low dose-rate contamination, highly non-uniform exposure to a significant area of skin and irradiation with discrete radioactive "hot" particles. The early and late effects of low dose-rate and non-uniform exposure to sources of $^{90}Sr/^{90}Y$ will be investigated to establish the dose-effect relationships for acute ulceration, moist desquamation and late dermal atrophy. From those dose-effect curves it will be possible to estimate threshold doses for each effect.

Studies involving the assessment of acute ulceration will be for irradiation from discrete ^{60}Co particles. This will mimic operational problems frequently encountered in pressurised water reactors.

Pathogenesis of early and late radiation-induced damage to the skin

Cellular and molecular aspects of radiation damage to the skin will be investigated. The role of any increased expression of cell adhesion molecules, ICAM-1, ELAM-1 and VCAM, on the surface of endothelial cells will be studied. The adhesion of leucocytes to up regulated endothelial cells would appear to be an important initial step in the pathogenesis of late radiation damage to skin. It is hoped that these studies will give further guide to the use of potential prophylactic measures in the management of radiation late effects in the skin.

Prophylactic treatments

Treatments aimed at reducing late radiation effects will be directed at mechanisms known or believed to be involved in the pathogenesis of such reactions; these to include the use of inhibitors of iron-dependent lipid peroxidation (re-perfusion injury), the correction of imbalances in eicosanoid metabolism, the use of haemorrheological agent and finally the use of anti-adhesion molecules may be considered.

Contribution of thr Radiation Biology Department, St. Bartholomew's Hospital Medical College.

This laboratory has for somme years concerned itself with radiation protection aspects of both the early and the late radiation effects in mouse skin. Our acute studies using a range of alpha and beta sources continue to be done in collaboration with the pigskin work of Dr J. Hopewell of Oxford University.

In paralllel with these early effects we have developed a highly reproducible skin cancer induction system in four different strains of mice - SAS/4, CD1, CBA and C57B1. The significant difference in radiogenic skin cancer proneness among these mouse strains is now being investigated at the molecular level under the current 1992-1994 CEC contract.

Since the majority of our murine skin tumours are of fibroblast origin we are concentrating research on the response of the cytokine that is central to the fibrosis that seems to precede neoplasia, namely, TGF-beta. For this we are using in situ hybridisation and the biochemical analysis of the protein and mRNA as indicators of altered gene expression.

Recent reports in the literature (including the work of our collaborators in Saclay) point to the important role played by the so called "early response" genes such as c-jun and c-fos in the initiation of the pathophysiological effects of radiation.

We are therefore putting an effort into clarifying the part played by such proto-oncogenes in the skin reactions of the four strains of mice.

Contribution of Instituto Dermatologico S. Gallicano - Rome

Telethermography with thermostimulation technique in the medical surveillance of radiation workers.

Infrared thermography is a very useful paraclinic tool to examin vascular damage in chronic radiodermitis, being the temperature of the skin expression of the skin blood flow. This method give us the immediate visualisation of all thermal points of the skin surface, presented as a video imaging, without the necessity of plotting or mapping, as happens for other thermometric procedures.

In particular telethermography examines the superficial, nutritional vascular network, formally the capillaries of dermal papillae and subpapillary microvessels, which supply is mandatory for the avascular epidermis. Moreover it is to be emphasized that at this papillary-subpapillary layer of the skin there is, physiologically, a critical condition of perfusion (25% of total skin blood flow), so that minimal damages of dermal vasculature will be responsable of important alterations of blood flow at this level.

The particular sensitivity of thermography can be enhanced employing a thermal stress on the examined skin area and following in real time the thermal recovery. Thermal recovery times after thermostimulation are the exact expression of thermogenicity of the area, that means its supply. This stress is performed by using a special thermal probe. This technique among other things permits to eliminate the problem of a prolonged immobility of the patient and its adaptation to the room temperature and humidity.

Starting from these data we have studied thermographically in the last three years 150 radiation workers, dividing them in two groups: those affected by severe cutaneous lesion radiodermitis and those without or affected with minimal lesions. 200 healthy subjects age and sex paired were examined.

Materials and methods: we employed a Teletermograph AGEMA 870, a thermostimulator Surgicon, a PC TIC 8000. The subjects were subdivided as follows: I group (n=200). Normal subjects, never professionally exposed to radiations, without hypertension, diabetes or sistemic disease, not smokers; II group (n=15) chronic professional radioexposed workers with clinical evident radiodamaged fingers: scleroatrophy, severe onicodistrophy); III group (n=135). Chronic (more than 20 years) professional radioexposed workers without clinical apparent lesions, or having minimal lesions (eg onicodistrophy).

Results (preliminary data): in the first group (normal, not exposed subject) the values of thermal recovery time (TRT) were: 2-6 min ±40 sec in 180 subjects (90%), less than 1 min in 2 subjects (1%), more than 7 minutes in 18 subjects (9%). As that concerns the second group (subjects with evident severe cutaneous radiolesions) the fingers affected showed a TRT of less than 30 sec in 12 subjects (80%), values between 2 min and 6 min in 2 subjects (13%), and in 1 case a TRT over 10 min. In the III group (subjects professionally radioexposed, with no evident or minimal clinical lesions), we had the following results: TRT 2-6 min: 90 subjects (67%); more than 7 min: 35 subjects (26%); less than 1 min: 10 subjects (7%).

Project 1992-1994

The significant different percentage of subject with prolonged thermal recovery times (TRT) between normal (9%) and radioexposed subjects (26%) let us to study the possibility to evaluate a specific difference between these two groups.

The proposal of the project is to examin and analyse the curve of thermal recovery time in the aim to better differenciate physiological from pathological curves.

In our opinion is the first phase of TRT the true phase indicating the blood supply of the skin, being the second phase hardly dependent from other conditions in particular room temperature and humidity.

The aim of this study is to evaluate the possibility to differenciate through the study of thermal recovery time curve normal subjects from subjects affected by radiodermitis. 150 radioexposed workers will be enrolled.

Will be employed: telethermography with thermostimulation technique. Also will be performed histological, histochimical and ultrastructural studies.

B24 Radiation damage to lens, thyroid and other tissues of relevance in radiation protection.

Contract FI3P-CT930076 Thyroid and its proximate tissues radiation dosimetry; stochastic and deterministic biological effects in humans and model systems.

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17 months 1/01/93 to 31/05/94

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Description of research work

This thyroid project as a whole has the objective of providing a more reliable scientific background against which public health officials, radiation protection organisations and governments may evaluate risks from thyroid irradiation, establish dose limits, intervention levels and monitoring techniques appropriate to normal time and/or in the event of an accident. Within this general framework it is expected to :

a) advance foetal and neonatal thyroid dosimetry, and the dosimetry for sensitive target organs/tissues (e.g. brain), liable to be irradiated by radionuclides contained in the foetal thyroid;

b) render more reliable the iodine kinetics models which underlay foetal and neonatal dosimetry;

c) provide studies of thyroid mass for different ages for use in dosimetry studies;

d) use newly available data on Thyroid Mass and iodine Kinetics, to provide a country by country (and where necessary region by region) map for the probable distribution of Thyroid Radiation Dose throughout Europe after an incident in which radioiodine is inadvertently distributed;

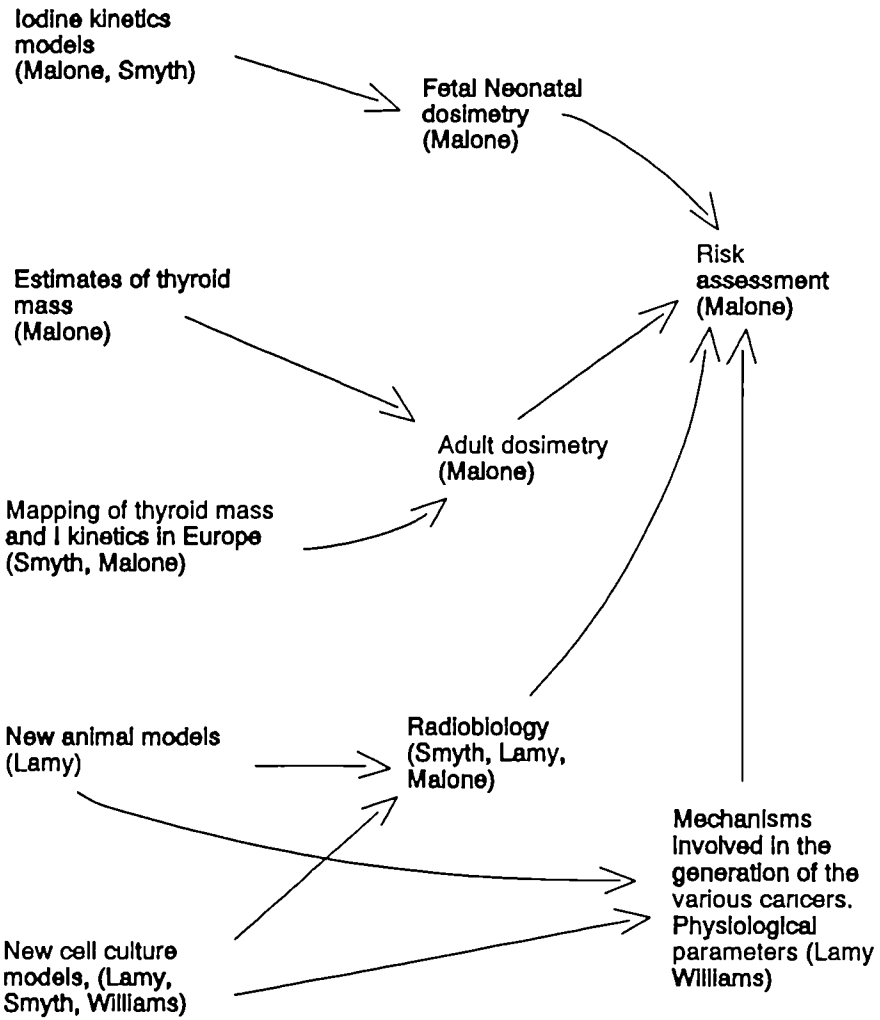
e) derive a more scientifically reliable, and possibly easier to measure, unit to incorporate this unit as part of the process of risk assessment and risk statement;

f) use newly created animal models and develop new animal models of transgenic mice for the study of the carcinogenetic process in thyroid in vivo and the role of environmental factors (eg. iodine supply, level of stimulation, etc);

g) establish issue culture models for radiation induced thyroid diseases (including neoplasia);

h) use the human cell lines models for studying radiation and other mutagenic induced neoplasia. Definition of the steps of immortalization.

DISTRIBUTION OF TASKS



B3 Radiation effects on the developing organism

B31 Damage to the central nervous system and hematopoiesis.

Contract FI3P-CT920015 Effects of protracted exposures to low doses of radiations during the prenatal development of the central nervous system.

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Description of research work

1. General Project Description:

a. Project aims to further evaluate the effects of low doses and particularly, of low dose rates of ionizing radiations on the developing central nervous system of the foetal rat.

b. State of the Art: In previous studies sponsored by the CEC, the repair capabilities of the foetal rat brain were found to be extremely limited, specially during the period of the organogenesis of the cerebral cortex. At that time indeed, an acute exposure to a dose of neutrons as small as 30 mSv (10 mGy) induced a 2% brain atrophy which was noted in the 3, 15 and 24 month old offspring. More important even, a protracted exposure to a dose of 160 mSv (given over as many as 4 days, using, in this case, a very low dose-rate gamma ray exposure) produced the same level of damage.

Such findings raise a series of problems among others, it would be useful to better characterize the brain atrophy: is it caused by a reduction in the size or in the number of the brain components (including growth factors, neuroreceptors and neurotransmitters)? Is there a relationship between the atrophy and the naturally occuring cell death of the neurons ? Is there a disorganization of the radial glial cells which represent the early guidance system of the neurons? Is there a difference between the atrophy caused by a protracted or an acute exposure to radiations? In the future, it will be necessary to investigate the possible correlations between the presence of small brain lesions and the development of severe mental retardation.

c. Importance of the projected studies on prenatal exposures to low doses of irradiation: The preceding observations (severe effects after protracted prenatal irradiations and also, the measurable atrophy of the foetal brain after an exposure to as little as 1 cGy of neutrons) are of importance for the sake of the radioprotection of the human population. They raise a series of questions which form the basis of the present project:

1) Can brain injury be observed after a prenatal exposure lower than 30 mSv ? During our preceding contract, the minimal dose able to produce a significant brain atrophy was found to be much lower than previously quoted, actually as low as 1 cGy (30 mSv) after an acute 600 KeV neutron irradiation. This level is very close to the maximal annual limit of 20 mSv now proposed by ICRP for atom workers; however, it can be speculated that the constant improvement of the methodologies will allow to reveal in the near future, a number of small though real abnormalities, down to dose levels still presently considered as safe.

2) Protracted versus Acute exposures. No data from protracted exposures of the foetal CNS to low doses of neutrons have yet been published but it can be suspected from our recent gamma irradiation data that a continued neutron irradiation even to doses as low as **1 cGy/day** (during 4 days) could elicit a detectable brain injury. Since similar prolonged exposures are susceptible to occur (in general after a negligence) more frequently than the acute ones (which only happen in cases of war, radiotherapy

or in atomic accidents), these situations are worth of serious consideration. More generally, it is clear that even basic knowledge over the **mechanisms** at work in protracted foetal irradiations is still urgently needed.

3) Severe mental retardation risk. The main reason to deal carefully with the consequences of a prenatal brain irradiation comes from the risk of radioinduced severe mental retardations. After their recent re-estimation of the doses administered at Hiroshima and Nagasaki, Otake and Schull estimated that a dose of 1 Gy of irradiation could induce as much as 48% of severe mental retardation in the offspring of women bearing foetuses of 8 to 15 weeks at the time of the exposure; the severity of this injury appears even more clearly when it is put in contrast with the comparatively "modest" 4% rise in tumour incidence which will occur in humans exposed to the same dose. Unfortunately and contrarily to the case of cancer, no treatment is to be expected for mental injury.

H. Coffigny adds "that the effects of the atomic bombs on the head size and/or mental retardation were found to be dose dependent. The small head size could be due to the death of nerve cells (see: I. Ferrer's proposal below) or to a mis- or un-development of brain structures as observed in morphological studies of the irradiated brains. Mental retardation was mainly characterized by low IQ and bad results at school; it is still very poorly understood at the anatomical level: some associative brain areas in the neocortex and in the older phylogenetic structures such as the hippocampus could be involved in mental retardation."

2. Project objectives:

The F13PCT920015 project has 2 main objectives:

1) Objective one, the Cf-252 experiment, is the study by the 3 participants of a common experimental model, namely the effects of a protracted neutron exposure to be carried out in France by the CEA. So, the 3 contractors will apply their own methodologies to specimens which have been irradiated in strictly identical conditions of exposure.

2) Objective two: Satellite experiments: each laboratory will also analyze pilot experiments more specific to each group and which will be developed hereafter (in the part of this summary redacted by each participant). This second objective aims at the solution of a range of issues in foetal radioprotection, but also at the development of new assays for future investigations using low dose and dose-rate levels after a variety of irradiation procedures.

These 2 objectives of the program are detailed hereafter:

Objective 1: Cf-252, the common research program.

FOETAL BRAIN INJURY AFTER A PROTRACTED EXPOSURE TO VERY LOW NEUTRON DOSES.

In this part of the project, the exposure conditions are similar for the 3 contractors.

The aim of Cf-252 is to evaluate the effects of a protracted exposure to low doses (from 1 to 7 cGy/day) of neutrons (using a Californium 252 source). Exposure will be given during 2 different periods of the pregnancy of the rat: from D12PC (day 12 post-conception) to D16PC (i.e.: the most radiosensitive period of the brain organogenesis) and from D16PC to D20PC (this period was shown by Sienkiwickz - 1991, personal communication- to be critical for hippocampus mediated tasks).

This experimental protocol has been established as a logical follow-up of the protracted gamma irradiations studied by the contractors during the recent B17-0003 project. They had found that prolonged exposures, during selected periods of the organogenesis of the brain, were nearly as deleterious as acute ones. The main question will now be to know whether a protracted exposure to high RBE radiations could produce brain lesions down to the very low 1 cGy/day dose-rate.

Exposure protocol:

The neutron irradiation will take place in France; the neutron source is located in the E.T.C.A., a military facility in Arcueil (near Paris), connected to the CEA. The irradiated brains will be collected by the 3 contractants and analyzed in their respective laboratories. The principal advantage of this common protocol is to eliminate the sources of experimental and regional variation in the irradiation and to provide the 3 investigation groups with biological samples as homogeneous and comparable as possible (with respect to: dose, type of radiation, rat strain and origin, maintenance of the animals, etc). This common strategy is derived from a "protracted" experience inside the EULEP organization which has very much contributed to the development of international scientific collaborations.

Assays:

A number of parameters of the collected material will be assayed according to the techniques currently in use by the respective contractors. They will be detailed below under each contributor's special part but are already given in short as follows:

The CEN-SCK at Mol will mainly deal with anatomical and morphometric measurements in the white matter but also with immunocytochemical assessments of the synaptic contacts (labelled by synaptophysin) and of the glial progenitors (GFAP, S-100 labellings) in the hippocampus and the cerebral cortex.

The CEA at Fontenay aux Roses will study the behaviour of a range of neurotransmitters in irradiated glioblast and neuroblast cultures (from both the hippocampus and the cerebral cortex).

The Hospital "Princeps d'Espanya" in Barcelona will evaluate the differences between the radioinduced and the physiological neuronal cell death phenomena tracing the mechanisms down to the gene level. They will also assess the effects of radiation on selected neuroblast populations (parvalbumin and calbindin positive) in the cerebral cortex (and in particular, the cingular cortex).

Objective 2: Satellite activities. (including more data over the methodological aspects of each contractant laboratory to the Cf-252 experiment).

1. CEN-SCK, Mol, Belgium,

"Focus is set hereafter on the technologies involved in the analysis of the biological material from the Cf-252 experiment and on the satellite exposure protocols to be carried out at Mol.

A. Description (with respective objectives) of the different biological assays to be performed at the CEN-SCK laboratory:

1. **Brain weight** measurements: a sensitive test for the presence of **microcephaly.** 2. **Cerebral cortex thickness, corpus callosum and cingulum volume** assessments: the cingulum was previously found to be the most sensitive anatomical indicator of a radioinduced prenatal damage. 3. **Subsurface cistern development** in cerebral cortex layer-I neurons: this electron microscopic assay could recognize metabolic alterations down to the neuronal level after 56 cGy gamma rays given in 4 days. 4. **Synaptophysin** immunocytochemistry: to reveal possible synaptic losses to be correlated with the decreased dendritic arborization which was detected by other methods. 5. **S-100 and GFAP** immunocytochemistry: to test a possible astroglial involution in cortex and hippocampus.

B. Other irradiation protocols:

Limited scale experiments with protracted X-ray, neutron, proton or gamma ray irradiations will be used to cast more lights on low dose-rate prenatal exposures. The influence of the **rat strain** will continue to be comparatively assessed. Part of the brain material will be sent to Barcelona for comparative assessments."

2. CEA, Fontenay-aux-Roses, France,

"The basic steps of brain development are similar in all mammals. The rat brain model was chosen in order to study the effects of irradiation specially on radial glial cells which act as guides in the migration of neurons during brain development, but also on the cortical and the hippocampal nerve cells involved in learning.

Radial glial cells from 2-day neocortex cell cultures taken on days 13, 14, 16, 18 and 20 of gestation will be studied after 0.25, 0.50, 0.75 and 1.00 Gy of acute 60-Co gamma irradiation. Radial glial cells will be identified on morphological basis but also after immunohistochemical labelling of their cytoskeleton (vimentin). The survival of these cells and the changes in shape and length of their processes will be measured. The radiosensitivity of the **cortex nerve cells** of 16 to 20-day-old fetuses will be studied in 2-day cell cultures either after acute irradiation (as above) on freshly isolated cells or after protracted gamma or 252-Cf neutron irradiation (0.01 and 0.025 Gy/day) between day 12-16 of gestation prior cell isolation and culture. General mortality, length and branching of neurites will be measured. Wherever identification of specific neurons by immunocytochemistry of their neurotransmitters (DA, GABA, ACh, serotonin) is possible, cell populations and neurite characteristics will be studied after

irradiation. **Hippocampal cells** taken on day 17 or 18 of gestation will be irradiated in the same conditions as for the cortex cells, cultured 2 days and then analyzed. Cell survival, the length and the branching of the neurites and the number of GABA positive cells will be measured. On the other hand, in infant rats, cell death and neurotransmitter distribution in hippocampus will be studied on brain sections after specific staining.

This proposal is fully integrated with those of the other participants. In particular, brain cell death will be studied at short (cell culture; Fontenay), mid (7 day-old rat; Barcelona) or long term (90 day-old rat; Mol) and surviving cells will be identified by immunohistochemistry. For all participants, rat foetuses will be exposed either to acute or protracted gamma or 252-Cf neutron irradiation at specific ages. This later irradiation will be carried out in France and the material analyzed by the three research groups."

3. Universidad de Barcelona, Hospital "Princeps d'Espanya", Hospitalet de Llobregat, Spain,

"**Naturally occurring cell death** is a widely distributed phenomenon during development of the nervous system which probably serves to match the number of neurons in different neuronal subsystems with the number of their respective targets. Recent observations in our laboratory, most of them carried out under EEC sponsorship, have shown that irradiation during development dramatically transforms the programme of naturally occurring cell death in the cerebral cortex. For example, early induced cell death produced by irradiation at appropriate stages of gestation may cause microcephaly and cortical malformations depending on the dose and the timing of exposure. Naturally occurring post-natal cell death is however reduced in these animals. In contrast, naturally occurring cell death is markedly increased after irradiation exposure during the postnatal period. Possible **mechanisms** commanding naturally occurring and irradiation induced cell death in the nervous system are barely understood; our purpose is to study the effects of **protein synthesis inhibitors** (i.e. cycloheximide) and **inhibitors of the transcription** (i.e. actinomycin D) on natural and irradiation induced cell death. It is also our aim to know the modifications of **nerve growth factor receptors** after acute irradiation, and the expression of several immediate-early genes (i.e. the **proto-oncogene c-fos**) after irradiation. Finally, we will continue our study of the particular vulnerability of different **types of cortical neurons** after acute and protracted irradiation.

Experiments will be carried out in different **strains of rats** exposed to different sources and doses of irradiation. Exposure to neutrons will be carried out in France. Gamma irradiation will be carried out in Belgium. Animals will be exposed to X-rays in Spain. Acute and protracted effects will be examined. Although high dose effects will also be examined, emphasis will be made on the effects of low doses."

B33 Transfer of radionuclides in utero.

Contract FI3P-CT920064c Dosimetry and effects of parental, fetal and neonatal exposure to incorporated radionuclides and external radiation.

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Description of research work

OBJECTIVES

There is an increasing recognition of the need to assess fully age-related changes in radiosensitivity and the risks of radiation exposure in utero and in early childhood from intakes of radionuclides and external radiation. Concern centres on the induction of childhood leukaemia and the radiosensitivity of the brain during in utero development. With regard to childhood leukaemia, the suggested association with paternal radiation exposure has shown the need to consider the genetic transmission of damage. To assess risks, it is necessary to estimate radiation exposure of the sensitive tissues in humans and, using animal models, establish the relationship between dose and risk, taking account of changes in sensitivity during development. To predict adequately the likelihood of particular effects on human populations exposed to low doses of radiation, the requirement for an understanding of the cellular and molecular mechanisms involved is increasingly clear. This proposal brings together work on radiation effects, biokinetics and dosimetry and considers natural alpha-emitters, fuel-cycle radionuclides and external irradiation. The objectives can be summarised as follows:

1) Human data: To measure concentrations of natural ^{210}Po and fallout $^{239/240}$Pu in human fetal tissues; use the data to assess exposures and in the development of biokinetic/dosimetric models.

2) Animal studies - biokinetics/dosimetry: To study the transfer of radionuclides including ^{85}Sr, ^{141}Ce, ^{238}Pu, ^{239}Pu, ^{241}Am, ^{237}Np and ^{210}Po to the embryo, fetus and associated tissues of mice, rats and guinea pigs for intakes during pregnancy and transfer from existing maternal deposits; for selected radionuclides, measure transfer to the neonate during suckling, to paternal reproductive tissues prior to conception and long-term retention in tissues of the offspring; estimate doses to the sensitive tissues and cells including the yolk sac membrane; use the animal data to estimate doses to the human fetus and child.

3) Animal studies - effects on haemopoiesis: To determine the relative long-term risk to haemopoietic tissue from low or high LET radiation in terms of functional disturbance and chromosomal damage following irradiation at different stages of development; include studies of the risk from pre-mating parental (in particular paternal) damage expressed in offspring; use the animal data to determine whether estimates of risk of late radiation effects should be modified for damage arising at different developmental stages or from inherited genetic damage; study the modification or amelioration of long-term haemopoietic damage using haemopoietic growth factors.

4) In vitro cellular and molecular studies: To study the cellular and molecular nature of long-term deficiencies in haemopoietic development and of promotion of recovery;

locate and characterize the radiosensitive subpopulation among the heterogeneous stromal cells; determine the role of haemopoietic growth factors in the regulation and recovery of radiation induced damaged tissue; evaluate the combined effect of embryonic exposure to radiation and murine parvoviruses with marked tropism for haemopoietic progenitors; study the induction of neoplastic transformation by in vitro alpha-particle irradiation.

5) Animal studies - effects on the developing brain; To study changes in the brain of rats after in utero exposure to bone-seeking radionuclides including ^{85}Sr and ^{141}Ce; determine the radiation dose delivered in bone and to the brain from these nuclides using techniques of biological dosimetry and compare effects with results obtained previously for external low and high LET irradiation.

The proposal represents an extension of work funded previously under two Contracts: B17-001 and B16-347d. Close collaboration with Cox (Contract PL920067-B13) will focus on chromosomal changes in haemopoietic stem cells. There will also be close collaboration with Contract PL920041-B15 which includes a long-term mouse study on leukaemia induction after in utero exposure to ^{239}Pu.

GENERAL DESCRIPTION

1) Human data

For the natural alpha-emitter, ^{210}Po, and for $^{239/240}Pu$ from fallout, it is possible to measure exposure of the human fetus directly. Measurements have been made of ^{210}Po concentrations in fetal tissue samples from Oxford, Bristol and Cumbria in the UK showing that levels are greatest in the developing skeleton in late gestation. Values obtained for $^{239/240}Pu$ concentrations in selected samples were on the limit of detection by mass spectrometry but provide valuable information on the relatively low concentrations of Pu in the fetus compared with fetal Po concentrations and with maternal Pu concentrations.

Human fetal tissue samples will be obtained from either second trimester terminations or stillbirths and areas of collection will be extended to include Cornwall because of high ^{222}Rn levels in the area. For $^{239/240}Pu$, measurements by mass spectrometry on pooled samples are planned to improve the reliability of results obtained. Measurements of ^{238}U and ^{232}Th are also planned. For ^{210}Po and ^{226}Ra, measurements using track detector techniques will concentrate on levels in fetal spine. Possible relationships between radionuclide accumulation and geographical location, and maternal and fetal age will be investigated.

2) Animal studies - biokinetics/dosimetry

Animal studies to determine radiation doses to the fetus and associated tissues and to neonates from incorporated radionuclides are required both in the interpretation of the significance of different effects observed at different stages of development and to enable estimates to be made of possible doses to the human fetus. Studies under existing Contracts B17-001 and B16-347d have provided information on the transfer of

^{210}Po, ^{238}Pu, ^{239}Pu and ^{241}Am to the embryo and fetus and associated tissues in mice, rats and guinea pigs. In addition to radiochemical measurements of nuclide concentrations in fetal and maternal tissues, autoradiographic techniques are being used to study the distribution of activity within tissues and determine the local concentrations of activity, particularly in haemopoietic tissues during development. The measurements have included transfer of ^{239}Pu and ^{241}Am to neonatal mice during suckling. These studies will be completed during 1992/3 and will be extended to include transfer from existing maternal deposits, including ^{210}Po transfer after contamination with ^{210}Pb, and irradiation of paternal reproductive tissues prior to conception. Studies with other radionuclides are planned; preliminary measurements of chronic transfer of ^{224}Ra in mice will be extended and limited studies of the transfer of ^{237}Np, ^{95}Nb, ^{106}Ru and ^{144}Ce in rats and guinea pigs will be undertaken. Measurements of the placental transfer of ^{239}Pu, ^{241}Am, ^{237}Np and ^{210}Po in the baboon in late gestation are in progress; measurements of transfer during organogenesis are planned. Studies of the transfer of ^{85}Sr and ^{141}Ce to the fetus in rats will be undertaken to determine accumulation in and doses to the skeleton in relation to doses to the brain

The results of these studies will provide data for the calculation of doses to the embryo and fetus; ICRP are currently considering doses to the fetus as part of the work of the Task Group on age-dependent doses to members of the public from intakes of radionuclides.

3) Animal studies - effects on haemopoiesis

The sensitivity of haemopoietic tissue to irradiation during development has been demonstrated in experiments using mice. Long-term bone marrow cultures derived from mice contaminated in utero with ^{241}Am during organogenesis and the fetal period showed damage to the stromal microenvironment with a reduced capacity to maintain granulocyte-macrophage progenitor proliferation. Experiments with ^{239}Pu indicated that the sensitivity of haemopoiesis to alpha-irradiation, as shown by the development and long-term deficit of stem cells in the marrow of offspring, was highest in the embryonic stage. The results indicated that damage in early gestation was due to a direct effect on haemopoietic stem cells while irradiation during fetal development caused long-term damage to the stromal microenvironment. Differences in sensitivity to bone marrow irradiation between neonates and adult animals have been shown using external X- and gamma- sources. In general, the greatest effect on stem cell survival was seen in neonates. Paternal contamination with ^{239}Pu or ^{241}Am before mating has also been shown to have significant effects on haemopoiesis in offspring. Further studies are needed to resolve the observed effects.

Mice will be contaminated with radionuclides (specifically ^{239}Pu, ^{241}Am, ^{224}Ra, ^{210}Po) at specific developmental time points from pre-conception through to weaning (using acute or chronic administration). Evaluation of haemopoietic stem cell changes in the developing haemopoietic tissue of offspring will be made from birth to old age, measuring multipotent CFU-s, their proliferative activity and self-renewal capacity; lineage committed progenitor cells - including granulocyte-macrophage progenitors, assessment of the platelet-producing megakaryocyte CFC and the marrow repopulating cells. A parallel assessment of the regulatory microenvironment will be made by assay of the stromal cell number (CFU-f), of the capacity to generate a new

microenvironment under the renal capsule and of its haemopoietic supporting capacity in long-term bone marrow culture. Associated chromosomal damage will be studied using standard cytogenetic techniques. Where appropriate, spatial distribution of cells in the marrow spaces will be determined. Effects will be compared to those similarly obtained using homogeneous low LET external irradiation at different dose rates and different radiation quality. The competitive expression of haemopoietic stem cells obtained from co-isogenic or transgenic mice, irradiated at different stages, will be analysed.

These data will be used to determine the relative sensitivities of haemopoietic tissue and its regulatory microenvironment to damage at different stages of development, the capacity of the tissue to compensate for damage, and to correlate the nature of the damage to any subsequent development of myeloid leukaemia. A long-term mouse study to determine leukaemia incidence after in utero exposure to ^{239}Pu is in progress at MRC (Chilton).

4) In vitro cellular/molecular studies

In studies of the cellular nature of the changes in haemopoietic development caused by alpha-irradiation, it has been shown that stromal cells from ^{241}Am contaminated animals perform better than controls in supporting the production of in vitro colony forming cells (CFC) when they are reseeded with total bone marrow cells. However, when stromal cells are recharged with purified haemopoietic stem cells, the stroma derived from contaminated mice showed a decreased capacity to maintain haemopoiesis. Studies of long-term effects on haemopoiesis have shown metabolic changes in neutrophilic granulocytes from mice after external irradiation or ^{239}Pu contamination. Preliminary observations have shown activation of the functional capacity of granulocytes with enhancement of superoxide anion production and protein production in cells taken from either the peripheral blood of mice or in long-term marrow cultures.

Studies will be undertaken on stromal bone marrow cells from ^{241}Am contaminated mice to determine the cellular and molecular changes relating to residual radiation damage. Studies will include haemopoietic growth factor induction, production of extra-cellular matrix molecules, expression of cell adhesion molecules and production of heat shock proteins. The contribution of these components to the interaction between stromal and haemopoietic cells will be examined. The heterogeneous stromal cell population will be fractionated to determine the contribution of its subpopulations to haemopoiesis. Cellular and molecular characterisation of the subpopulation will be undertaken and their ability to maintain haemopoiesis in long- term cultures will be investigated.

Mechanisms of repair of residual damage will be studied, determining the role of haemopoietic growth factors in the regulation and recovery of tissue damage after low dose alpha-irradiation and external X- or gamma- irradiation. In addition, changes in haemopoietic growth factor expression and growth factor receptor expression will be studied after external X-irradiation. Neutrophilic granulocyte function will be assessed after external irradiation or alpha-contamination using cells from peripheral blood or long-term marrow cultures.

An attempt to induce neoplastic transformation by direct alpha-irradiation haemopoietic stem cell lines, in vitro will be made.

5) Animal studies - effects on the developing brain

The effects of in utero irradiation of the developing brain have been studied using rats and mice, comparing exposure to gamma (^{60}Co) and neutron (^{252}Cf) sources. For exposure throughout gestation, brain weight in the fetus and offspring was reduced with increasing dose-rate and cumulative dose. In the offspring, the effect was confined to the cerebrum (forebrain + midbrain) with no significant change in the hindbrain. The RBE for neutron irradiation was about 4 for this effect on the cerebrum. Experiments in which gamma-irradiation was continued for only part of pregnancy showed that exposure during the last third of gestation was as effective as exposure throughout gestation. Studies of associated histological changes in the brain have shown agenesis of the corpus collosum and the presence of ectopic grey matter as well as perturbations of the alignment of pyrimidal cells in the hippocampus.

In rats exposed to ^{85}Sr, ^{141}Ce and ^{239}Pu in late gestation, changes to the brain will be studied. Biological dosimetry, measuring apoptosis in the cerebrum, will be used to assess doses and effects will be compared with previous results from work under existing Contract B16-347d in which dose-effect relationships were established using whole-body gamma and neutron irradiation.

CONTRIBUTION OF THE NRPB

The NRPB will continue with measurements of $^{239/240}Pu$, ^{210}Po and other alpha-emitters in human fetal tissues and animal experiments on the placental transfer of radionuclides. Studies will be extended to include the effects of in utero exposure to ^{239}Pu on haemopoietic development in the mouse.

Measurements have been made of concentrations of ^{210}Po and other alpha-emitters in fetal tissues obtained from second trimester terminations carried out in the Oxford area and in Cumbria. No correlation was observed between ^{210}Po concentrations and either geographical location or maternal age. Ethical Committee approval has been obtained to extend this study to include fetal tissue samples from high ^{222}Rn areas in Cornwall. Measurements have also been undertaken of $^{239/240}Pu$ concentrations in selected samples. The values obtained are on the limit of detection by mass spectrometry but provide valuable information on the relatively low concentrations of Pu in the fetus compared with fetal Po concentrations and maternal Pu concentrations. Measurements on pooled samples are planned to improve the reliability of results obtained. It is also intended to collaborate with UB in the measurement of $^{239/240}Pu$ in tissues from still-births.

Animal studies using rats and guinea pigs have concentrated on the placental transfer of ^{238}Pu. Studies on the transfer of ^{210}Po to the embryo/fetus and comparisons of the behaviour of ^{238}Pu and ^{241}Am are in progress and will be completed during 1992/93. Limited experiments with other radionuclides, including ^{237}Np, ^{95}Nb, ^{106}Ru and ^{144}Ce are planned. Studies will consider transfer from existing maternal deposits as well as exposure during pregnancy. Measurements of transfer and distribution of ^{238}Pu and

^{241}Am in mice will be undertaken in support of effects studies (NRPB, VITO and PICR). In addition to radiochemical measurements of nuclide concentrations in fetal and maternal tissues, autoradiographic techniques are being used to measure concentrations in the embryo and associated tissues at the blastocyst/egg cylinder and during organogenesis. These measurements will include the concentration of nuclides in the yolk sac, the first site of haemopoiesis in mammals and the origin of the germ cells. A collaborative experiment with CEA is in progress to measure the placental transfer of ^{239}Pu, ^{241}Am, ^{237}Np and ^{210}Po during late gestation in the baboon. Measurements of transfer during organogenesis are planned. The results from animal experiments will be used to calculate doses during in utero development; particular attention will be given to doses to haemopoietic tissues, taking account of yolk sac development, and subsequent stem cell irradiation in the fetal liver and bone marrow.

Studies of the effects of in utero ^{239}Pu contamination on haemopoiesis in CBA mice are planned. Damage expressed in the offspring will be determined, initially measuring changes in haemopoietic stem cell populations by clonogenic assays and studies of chromosomal changes (the latter in collaboration with Contract PL920067-B13).

CONTRIBUTION OF THE UNIVERSITY OF BRISTOL (UB)

The proposed project builds on current work in which levels of natural long-lived alpha-emitters have been determined in autopsy fetal tissues obtained from a number of geographical areas. To date 53 samples have been obtained from the Bristol area and 5 from West Cumbria. Analysis has been completed for 17 samples from around the Bristol area. In soft fetal tissues, the levels of activity present are below the detection limit, < 50 mBq kg^{-1}, for the samples that are obtained. In fetal spine the activity from ^{210}Po ranges from the detection limit to 450 mBq kg^{-1}. These values have been compared with model predictions, developed in the project, for fetal transfer of ^{210}Pb by which the ^{210}Po is assumed to be supported. The level of alpha-activity in fetal spine appears to show an association with distance of mother's residence from the Severn estuary. Further data are required to determine whether this association is real.

The programme of collection of autopsy material from Bristol and West Cumbria in the UK will continue. Future analysis will concentrate on levels in fetal spine where in general the activity present is sufficient to be resolved in the small samples obtained. As before, TASTRAK (CR-39) plastic track detectors will be employed to measure concentrations of ^{210}Po and ^{226}Ra activity and their distribution in tissue samples. The TASTRAK plastic autoradiographs will be stored for up to one year in order to record the low activity present at natural exposure levels. The analysis of tissues under existing storage will provide improved statistics on which to determine the activity concentration in the fetus as a function of fetal age and to compare with model predictions for fetal transfer of natural alpha-emitters. Similarly, added data will provide a more rigorous test of the apparent correlation of activity with proximity of mother's residence to the Severn Estuary. The analysis of samples from West Cumbria will reveal whether levels in these tissues are different to those taken from the Bristol area.

CONTRIBUTION OF VITO (SCK), MOL

After various regimes of ^{241}Am administration in mice (paternal contamination pre-conception, protracted contamination during gestation, contamination at 14 days of gestation and/or during lactation), effects on the bone marrow will be studied in the offspring (14 kBq ^{241}Am administered to the dams, 6 kBq to the fathers).

Haemopoietic and stromal stem cell studies will be continued in order to identify the most radiosensitive periods during development. Particular attention will be given to the effects on haemopoiesis in offspring after paternal contamination before conception. External irradiation (X-irradiation or gamma-irradiation) will be used for comparison with internal contamination and to study the effect of dose rate and radiation quality (collaboration with CIEMAT). To simulate the situation of continuous chronic intake of radionuclides from the environment, osmotic pumps containing ^{241}Am will be implanted into pregnant mice for continuous exposure during organogenesis and the fetal period. Studies of the transfer and distribution of ^{241}Am after acute (single injection) and chronic (osmotic pump implantation) contamination will be used to estimate average radiation dose in femur and liver of the offspring.

Autoradiographic studies of bone samples will be undertaken at NRPB to determine the distribution of dose within the tissue. The relationship between doses and observed effects will be studied.

Studies on the stromal bone marrow cells will investigate the cellular and molecular changes related to residual radiation damage to stem cell proliferation, initially comparing the effect of in utero contamination on day 14 of gestation, continuing during lactation, with contamination of adult mice. Stromal cell cultures will be characterised with respect to: 1) growth factor production (IL-1, IL-3, IL-6, GM-CSF, SCF, M-CSF, G-CSF) in the conditioned medium and bound to the extracellular matrix (collaboration with CIEMAT); 2) extra- cellular matrix production (collaboration with UCM); 3) expression of cell adhesion molecules (integrin); 4) heat shock protein production. The contribution of these components to the interaction between stromal and haemopoietic cells will be examined. Techniques will include the use of growth factor dependent cell lines, in situ hybridisation, Northern blot analysis and immunocytochemistry. Depending on the results obtained, effects in other experimental groups will be studied at other developmental ages. Studies will also be undertaken on fractionated stromal cell populations in collaboration with TNO in order to evaluate the contribution of subpopulations to haemopoiesis (see detailed description of TNO contribution). Purified stromal layers will be characterised at the cellular and molecular level (expression of membrane markers, extracellular matrix growth factor products). They will also be reseeded with separated haemopoietic stem cells to evaluate their supportive capacity for haemopoietic stem cell proliferation.

Mechanisms for the repair of residual radiation damage after contamination with ^{241}Am will be studied with respect to the cellular mechanisms which control the growth and differentiation of haemopoietic cells. In this context, there is a lack of information on the role of haemopoietic growth factors in the regulation and recovery of damaged tissue after low dose internal alpha-contamination.

CONTRIBUTION OF THE PATERSON INSTITUTE FOR CANCER RESEARCH

The proposed studies will investigate a) whether estimates of risk of late radiation effects should be modified for damage arising in utero and during early development, b) the extent of transmitted genetic damage is expressed in haemopoietic tissue, c) the cellular and molecular nature of long-term deficiencies and d) the mechanisms and promotion of recovery.

The placental transfer of radionuclides in mice (specifically ^{239}Pu, ^{224}Ra (via ^{228}Th implant) and ^{210}Po) and their distribution in fetal and neonatal tissues will be studied following acute or chronic administration at various phases of pregnancy and infancy. Since considerable data have already been accumulated following acute administration of ^{239}Pu, more attention will now be given to other radionuclides.

Assessments will be made of long-term damage to the haemopoietic progenitor cells (including marrow repopulating cells, pluripotent spleen colony forming cells (CFC), committed granulocyte/macrophage and megakaryocyte CFC and the stromal fibroblastoid CFC) in terms of total number, proliferative activity and self-renewal capacity after radionuclide contamination or after homogeneous low LET irradiation given at different dose-rates. These cell populations will also be assessed in offspring of mice (particularly paternal) contaminated with ^{239}Pu before mating and with potential follow-up using external irradiation. These studies will include determination of long-term disturbances in the spatial distributions of haemopoietic progenitor and stromal cells in the marrow spaces.

Long-term damage to the haemopoietic stromal microenvironment will be assessed using marrow cultures and the manipulation of cell production using growth factors, as well as determining the capacity of the stroma to generate a bone capsule supporting haemopoiesis under the renal capsule.

Induction of factor independence in haemopoietic stem cell lines by transformation in vitro using alpha- particle irradiation will be investigated.

The proposed work will complement studies at MRC (Chilton) on long-term induction of neoplasia in mice following similar contamination protocols. It will also complement information obtained for other radionuclides studied at VITO, for external irradiation at CIEMAT and provide material for an assessment of any functional damage arising in the mature cell populations at UCM.

CONTRIBUTION OF THE ITRI-TNO, RIJSWIJK

Stromal and haemopoietic stem cells of normal and irradiated bone marrow and fetal liver will be analysed and sorted by a combination of cell separation techniques and culture methods in collaboration with VITO.

The analysis and sorting of the haemopoietic stem cells will be performed by combining density gradient centrifugation and FACS sorting for wheat-germ agglutinin-positive, monoclonal antibody 15-1.1 negative cells with medium forward and low perpendicular light scatter intensity. Subpopulations of this cell fraction will

be sorted using Rhodamine 123 and a new antibody directed against the product of c-kit. The different subpopulations will be compared with respect to their short-term and their long-term repopulating ability in vivo by transplantation studies at TNO, whereas these characteristics will be analysed in vitro by culture on stromal layers at VITO. For this purpose the cells are transported to VITO after the sorting procedure. Pilot experiments indicated that this does not impair the culture of the cells on stromal layers. Pilot experiments also indicate that long-term growth of purified haemopoietic stem cells on stromal layers may require the presence of accessory cells. This will be further investigated and cell sorting will be used to identify such cells.

The stromal stem cells will be characterized by separating bone marrow cells using a variety of antibodies against haemopoietic differentiation markers (at TNO) and by subsequent culturing of the sorted fractions under conditions facilitating the growth of CFU-F (at VITO). Attempts will be made to find a combination of antibodies which yields a high enrichment of CFU-F. This cell fraction then will be studied under conditions that facilitate the formation of stromal layers maintaining haemopoiesis (at VITO).

Subsequently, TNO will regularly deliver sorted stromal stem cells and some weeks thereafter sorted haemopoietic stem cells to VITO where the interaction between the strictly stromal layers and individual pluripotent haemopoietic stem cells will be analysed at the molecular level. As sources for stem cells, the fetal liver and adult bone marrow from normal and ^{241}Am contaminated mice will be used.

Fluorescent in situ hybridisation methods will be developed at TNO to detect which cells in the stromal layers are producing biological response modifiers, haemopoietic growth factors and inhibitors, and to what extent that production can be damaged by radiation. In addition, adhesion molecules will be analysed with respect to their role in the interaction between stem cells and stromal layers by employing PCR on cDNA from sorted haemopoietic stem cells and by using neutralizing/blocking antibodies in long-term cultures. Upon identification of relevant adhesion molecules, their function in long-term cultures of cells from ^{241}Am contaminated mice will be analysed and compared with normal bone marrow and fetal liver cells.

CONTRIBUTION OF UNIVERSIDAD COMPLUTENSE DE MADRID (UCM)

We have observed that treatment of mice with either external irradiation or internal contamination with alpha- emitters leading to persistent stromal damage, produces compensatory mechanisms probably in stromal cells, that stimulate the release of excess CSF-S. These factors prime granulocytes in such a way that their functional capacity is activated.

Our contribution to this project will be focussed on the impaired oxidative metabolism of granulocytes obtained from peripheral blood and long-term bone marrow cultures, to analyse the mechanisms involved in the priming effect. The studies will be carried out in mice of different ages, newborn, young and adults, irradiated with single and repeated doses of external X-irradiation or contaminated with the alpha-emitters, ^{239}Pu or ^{241}Am (in collaboration with PICR and CIEMAT).

The role of radioinduced factors in increasing glucose uptake in granulocytes, thereby controlling glycolytic flux and intracellular ATP levels will be analysed. With the aim of correlating increases in superoxide anion and levels of NADPH, oxidation of glucose via hexose monophosphate shunt will be quantified.
Because of the close correlation between activation of protein kinase C and neutrophil function, particularly the activation of the respiratory burst, studies will be undertaken of the phosphorylation reactions mediated by protein kinase C. Possible modifications in the characteristics of binding between individual factors such as mr-GM-CSF and their specific receptors in mature granulocytes and purified populations of CFU-S will be investigated (in collaboration with CIEMAT).

In addition, stromal cells from irradiated or contaminated animals will be analysed for changes in intracellular levels of the principal metabolites and the activities of their main glycolytic enzymes will be quantified.

Proteins from whole cell lysates will be analysed by two dimensional isoelectric focussing/gel electrophoresis. Special attention will be paid to the polypeptide composition of the matrix molecules synthesised by stromal cells. These experiments will be done in close collaboration with VITO.

Different growth factors will be used in order to study the qualitative and quantitative specificity of their activation on neutrophil function.

CONTRIBUTION OF CIEMAT

After the X-irradiation of mice in newborn and embryonic stages of growth, the number of haemopoietic progenitors (CFU-S, CFU-GM, BFU-E, CFU-MK and CFU-Mix) in the marrow and spleen will be determined up to one year post-irradiation. After identifying the most radiosensitive periods in the development of the embryo, the influence of the dose rate and quality of radiation on both stromal and haemopoietic precursors will be investigated. These studies will be performed in coordination with PICR and VITO where particular attention will be paid to the precursors of the haemopoietic microenvironment.

To obtain information on the nature of haemopoietic failures induced as a consequence of the external irradiation of the mice, the expression of haemopoietic growth factors will be determined by Northern blotting and RT-PCR. These experiments will be coordinated with studies at VITO, where similar determinations will be performed in ^{241}Am contaminated mice.

To investigate whether the long-term haemopoietic injury produced in irradiated mice reflects changes in the function and/or expression of growth factor receptors in granulocytes and haemopoietic progenitors, purified populations will be prepared by elutriation and biochemical studies will be undertaken by UCM.

With the aim of analysing the most radiosensitive embryonic stage for the totipotent haemopoietic stem cells, the competitive expression of haemopoietic stem cells obtained from co-isogenic or transgenic mice irradiated at different stages of growth, will be analysed.

Using bone marrow genetically marked by retroviral vectors, it will be further determined whether radiation restricts the longevity, clonal succession and differentiation potential of haemopoietic stem cells. The consequences of the direct interaction of radiation with the haemopoietic cells and other indirect mechanisms mediated through the irradiated stromal cells, will be considered. These experiments will be coordinated with the other collaborators to establish the most appropriate irradiation protocol.

Finally, in order to ameliorate the haemopoietic syndrome and/or the haemopoietic residual damage induced by the irradiation, different growth factors and other indirect stimulating molecules will be used, in experiments coordinated with PICR.

CONTRIBUTION OF CEA

The proposed project aims to estimate the doses delivered to the different parts of the skeleton during its development from the beginning of calcification, after the administration of alpha and beta emitting radionuclides of various energies. This will involve measurement of the skeletal distribution of these nuclides and analysis of the biological effects induced. The studies will initially be particularly focussed on the uptake and distribution in the skull as a source for irradiation of the brain during development. This represents a continuation of work undertaken during the last two years, funded under CEC Contract B16-347d in which dose-response relationships were studied for protracted external irradiation (gamma and neutrons) with low dose rates (15 to 375 mGy/day) and biological effects, particularly changes in the development of the brain.

Experiments will be performed mostly on rats but also in non-human primates (baboons) if possible. For the development of dosimetric models for the human fetus, information on radionuclide transfer and distribution in a primate species is clearly very valuable. A collaborative experiment is currently in progress with NRPB to determine the placental transfer of ^{210}Po, ^{237}Np, ^{238}Pu and ^{241}Am in the baboon in late gestation. Ethical Committee approval is required for further studies and is being pursued. The use of techniques that do not require sacrifice of animals will be considered, including the use of medical imagery such as scintigraphy and nuclear magnetic resonance spectroscopy.

Initially ^{141}Ce and ^{85}Sr will be the radionuclides used for the study and later actinides such as ^{239}Pu will be used. Animals will be given radionuclides by intravenous or intraperitoneal administration at various stages of pregnancy during the fetal period.

The initial distribution of radionuclides will be determined in rats from day 19 post conception to weaning. Histological and autoradiographic studies will be performed to localise the activity in the different bones. Remodelling of activity in bone will be estimated as a function of time after administration. Histological studies will be performed to localise the activity in the different bones. To establish the biological alterations in the brain, malformations including corpus callosum agenesis, the formation of ectopic grey matter and perturbations of hippocampal pyramidal cells alignment will be studied.

Histological and cell culture methods will be developed to estimate the doses delivered to bone and adjacent tissues. Biological dosimetry will be done on the cerebrum by counting apoptosis at different depths from the tissue surface. A dose effect relationship is already available for 14 day old rats, after whole body irradiation by ^{60}Co gamma rays at levels between 0.1 and 0.2 Gy.

C) RISKS AND MANAGEMENT OF RADIATION PROTECTION

C1 Assessment of human exposure and risks

C12 Exposure to natural radioactivity and evaluation of parameters influencing these risks.

Contract FI3P-CT920025 Retrospective assessment of radon exposure from long-lived decay products.

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Total Contribution by the Commission: 70 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

OBJECTIVES

The main purpose of the project is to establish the prerequisites for using long-lived decay products of radon (Rn-222) in the indoor environment for the assessment of lung cancer risks from radon exposure. Radon chamber investigations of plate-out and implantation daughter processes will be carried in parallel with *in-situ* studies of long-lived daughters (LRnD) in dwellings, aiming at a better understanding of the long-term rates of plate-out in the realistic indoor environment. With the knowledge gained it will be possible to reach a consensus concerning the choice of objects in a dwelling suitable for retrospective monitoring.

BACKGROUND

The long-lived decay products of radon (Rn-222), starting with Pb-210 ($T_{1/2}$= 22 y), remaining in dwellings, can be used to integrate past radon concentrations over several decades, but only the part of the activity well hidden from human influences (e.g. cleaning) or other destructive actions is useful in practice. Decay products implanted into indoor surfaces by alpha recoils fulfil this latter requirement, and daughter nuclides which have built up inside large-volume, porous materials can also be useful.

In the first phase of the RARE project (EC Contract No. B17-CT90-0013) it was shown that an analysis of glass surfaces for the alpha emitting granddaughter of Pb-210, Po-210, gives the necessary sensitivity and specificity. With specially developed open-flow pulse ionization chambers the alpha-spectrometric measurement of Po-210 can be performed without dismantling or destroying the sample.

Very few, if any, dwellings exist in which the radon concentration has been followed continuously over several years. This means that straightforward *in-situ* "calibration" of the retrospective glass-polonium method is not at hand. In order to demonstrate the merits of the method, radon chamber studies under well controlled conditions must be performed to support and complement results from dwellings.

Competitive ways to estimate past radon exposures involve extrapolation over long periods of time and the precision of such estimates is normally low. It is obvious that large uncertainties are also associated with the glass-polonium method if the exposure and plate-out history of the exposed surface is poorly known and that the method will work only if openly exposed glass surfaces of well-known age are available. If this is the case, however, the conclusion of the ongoing CEC RARE project is that implanted Po-210 in household glass sheets, already at the present stage of knowledge, can be a useful tool in radon epidemiological studies.

STRATEGY AND COLLABORATION

The complexity of the chain of processes leading from the dose-giving airborne daughters to implanted Po-210 activity in surfaces under realistic indoor conditions presents a real challenge to the project. A multi-lab approach is necessary and a

certain degree of overlap is justified in order to guarantee general validity of the results. All the involved laboratories are well equipped for the purpose and have long experience of radon and radon daughter research. Compared with the previous phase of the project, field measurements of Po-210 will be more frequent, and SCK will explore the new approach of Po-210 in large-volume porous materials. RUG (with CR-39 devices from UCD) will test autoradiographic track-etch methods for measuring Po-210 in dwellings. As before, pulse ionization chambers (PIC) will be used as reference detectors for implanted Po-210. RUG with technical support from Denmark Technical University (Dr N.Jonassen), DTU, will test and further develop the open-flow PIC for non-destructive measurements of large-area objects.

DESCRIPTION OF THE CONTRIBUTION OF S.C.K (STUDIECENTRUM VOOR KERNENERGIE, MOL)

In the living environment a lot of parameters influence the conversion factor between the implanted ^{210}Po activity of glass surfaces and the long term radon exposure. Important parameters are: the deposition constant of the unattached decay products, the attachment rate, the ventilation rate, the removable fraction of the activity deposited on theglass surface, the extend to which a layer of dust or grease can prohibit implantation, the escape probability of implanted ^{210}Pb recoil nuclei, the influence of convection and turbulence.

A sensitivity analysis of the conversion factor will be performed in collaboration with the university of Gent and will result in a better understanding of the relative importance of the different parameters. The results will be compared to the experimental values reported by the and other partners. The analysis can help to indicate the objects and locations which are suited to conduct the ^{210}Po measurements (window with or without convector, frame of a picture, etc.).

Next to the modelling effort, experiments will be set up to investigate the long term radon exposure from the ^{210}Po activity inside spongy materials like mattress, the padding of sofas or cushions, etc. The materials must have a long diffusion length for radon and must be inaccessible for the decay products. Some years ago Falk investigated this technique for wooden furniture. The usefulness was limited due to the variable ^{226}Ra and ^{210}Po background of the wood. In recent years more and more spongy synthetic materials are used in homes which have no ^{226}Ra or ^{210}Po background. ^{222}Rn can enter the material so that the ^{210}Po activity inside the material depends only on the duration and on the average radon concentration in the room. The interpretation of the measurement result is thus much easier then in the case of the measurement of the ^{210}Po activity implanted in glass surfaces.

Different spongy materials will be exposed in the laboratory with known radon concentrations. The ^{210}Po activity accumulated in the material will be measured. The material will be solved with an acid solution and the ^{210}Po will be separated. Then the activity will be counted with a low background α-counter. The detection limit is of the order of 1.5 mBq per sample. For a sample with an internal air volume of 1 l and for a integration period of 20 years this corresponds with an average radon concentration of only 3 Bq/m^3. This is less than the average outdoor concentration in Belgium. The feasibility of the technique will be investigated and the experimentally determined

calibration factor will be compared to the theoretical value. Several types of spongy materials will be examined. In a few houses where the actual radon concentration is known, samples will be collected and analysed. The results will be compared to the theoretical predictions.

DESCRIPTION OF THE CONTRIBUTION OF UNIVERSITY COLLEGE DUBLIN

The proposed work will consist of three principal parts as follows which will be carried out concurrently during the contract period:

(1) To assist the University of Gent group in their retrospective exposure assessment of the cases and the controls in the radon epidemiological study being carried out in the Ardennes. The assistance will involve the supply, processing, image analysis interpretation of CR-39 alpha track autoradiographics of long lived radon decay products from in-situ exposures to glass in the chosen dwellings. Improvements in alpha track energy resolution should make it possible to quantify the area specific activity of recoil implanted Po-210 in the glass surface examined.

(2) Laboratory based work to improve the energy resolution under automatic image analysis of CR-29 alpha tracks from radon and its decay products. This will make the form of using specially prepared sources of radon decay products and also by intercalibration with a pulsed ionization chamber.

(3) Investigations into the effect of aerosol loading, airflow and other variables on the plate-out rates of the individual short lived radon decay product. This work will be carried out in a thirteen cubic metre steel radon room using the UCD CR-39 plateometer as the principal measuring instrument. This phase of the work should directly benefit the investigations of the coordinator into the build up of long lived activity on surfaces due to alpha recoil implantation.

DESCRIPTION OF THE CONTRIBUTION OF THE UNIVERSITY OF GENT

Exposures of the past play a crucial rule in the determination of the indoor radon risk. Monitoring the Po-210 implanted in glass and other surfaces is up to now the only way to obtain some information about past radon situations and about possible changes to the indoor climate.

In the previous phase the feasibility of auto-radiographic track-etch methods for measuring Po-210 has been tested in a small-scale study. In the coming two years period these techniques will be further developed in order to obtain a practical measuring device for past radon and instant plate-out. Therefore, an open pulse-ionization chamber will be constructed as a reference instrument for the analysis of large samples. Glass samples with known embedded Po-210 surface activity (of the order of 50 Bq/m^2) will be prepared for calibration purposes and performance tests. This chamber will also be intercompared with other chambers of similar construction.

According to a protocol set up in collaboration with the University College of Dublin (UCD) a number of energy selected CR-39 devices will be installed in selected houses of the Ardennes epidemiological study for long-lived radon decay product and instant plate-out measurements. The analysis of the measuring devices will be taken charge of by UCD.

The experimental obtained Po-210 values will then be compared with the values calculated by means of the room model and the present radon determination.
The instant plate-out measurement will improve the estimation of the unattached fraction and in this way also of the lung dose.

In a limited number of dwellings the non-destructive CR-39 based technique will be compared with the results of the destructive technique applied by the Nuclear Research Centre (SCK).

C12 Exposure to natural radioactivity and evaluation of parameters influencing these risks.

Contract FI3P-CT920034 Characteristics of airborne radon and thoron decay products.

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Description of research work

Objectives

The inhalation of the short-lived decay products of radon and thoron determines the natural radiation exposure. Besides the airborne activity concentrations, the size characteristics of the unattached and attached decay products and the quantity of unattached activity are important parameters in all models for dose calculations. The size characteristics are also essential for understanding and describing the behaviour of the airborne activity in indoor and outdoor environments.

After the decay of ^{222}Rn (radon) and ^{220}Rn (thoron), most of the decay products (^{218}Po, ^{216}Po) are positively charged ions. These ions react with vapours of NO, NO_2, H_2O and other trace gases in the air, grow by cluster formation, and become neutral by recombination processes. During and after these processes, the clusters attach to aerosol particles in the air and form a radioactive aerosol. The air activity concentrations of the radon progeny depend on the decay constants, the exhalation rate of the radon gas, ventilation, and recoil processes. The size dependence of the attachment rate and of the deposition velocities or deposition rates determines the size distribution of the airborne radon decay products, the amount of the unattached activity, f_p, and the equilibrium factor F.

State of the art and progress to be achieved

1) The equilibrium factor F, the ratio of the equilibrium-equivalent concentration of the radon daughters to the radon gas concentration, ranged in indoor atmospheres from 0.1-0.9 with median values between 0.3-0.6. However, most results in the published literature were obtained under unknown and variable atmospheric conditions. Only some detailed studies show the dependence of F on the ventilation rate and the aerosol particle concentration in the air. The large variation of these results can mainly be explained by the uncertainties combining different experimental methods, by the non-steady-state conditions during the measurements, and by statistical variability.

Improved measurements of F require detailed information about aerosol particle concentrations and sizes, because these parameters basically determine the degree of disequilibrium in airborne activity.

2) Measurements of the complete activity size distribution of ^{218}Po show for an aged aerosol two well separated size fractions with median diameters between 0.5-3 nm and about 200 nm. These results suggest that the activity in the ultrafine size range less than 5 nm can be defined as the unattached fraction. However, in some cases unattached fractions of potential alpha energy, f_p, up to 0.34 were obtained from measured activity size distributions in the presence of different aerosol sources, because aerosol-attached activities with diameters up to 20 nm were included. Other measurements in closed rooms without additional aerosol sources yield an average unattached fraction, f_p, of 0.1 at average aerosol particle concentration of 6100 cm^{-3}. With additional aerosol sources, such as cigarette smoke, the aerosol particle

concentrations ranged up to 10^5 cm^{-3} and the values of f_p decreased below 0.005. More recently published results of measurements in poor ventilated rooms without aerosol sources yield at low aerosol particle concentrations of 2500 to 3000 cm^{-3} unattached fractions in the range 0.2 to 0.4. Surprisingly high f_p values between 0.6-0.8 were found with a vacuum cleaner operating and with aerosol particle concentrations of 1500 to 2000 cm^{-3}.

Some authors calculated the unattached fraction with a room model using different sets of deposition, attachment and ventilation rates and obtained values of f_p between 0.05 to 0.15 for poorly ventilated rooms with an aged aerosol.

Improved measurements of the unattached fraction f_p require a sophisticated measurement technique and data evaluation method. The determination of the aerosol composition, that is, aerosol particle concentration and size characteristics, are necessary to explain the results of f_p measurements and to avoid the possibility of misinterpretation.

3) The representation of the unattached fraction f_p with a single diffusion coefficient in dose models is a crude approximation. In reality, the unattached activity is a cluster mode in the diameter size range between 0.3-10 nm. Different kinds of chamber studies with high concentrations of activity demonstrate the effect of humidity, the reaction with oxygen or trace gases such as SO_2 and NO, and deal with radiolysis, the initiation of chemical reactions, and aerosol particle production from the radioactive decay of radon daughters.

However, most published results of chamber studies cannot be transferred to realistic living conditions, because the atmospheric chamber conditions (high concentrations of activity and trace gases) are quite different.

Due to experimental difficulties only few measurements were performed in ambient air under realistic indoor living conditions. Measurements in poor ventilated rooms with an aged aerosol yield median diameters 0.5-3 nm for ^{218}Po clusters. Other publications did not specify values of aerosol or other room-specific parameters and found median diameters of the potential alpha energy up to 10 nm.

Diffusion batteries with different screen and mesh numbers are commonly used for size distribution measurements in the nanometer range because other methods must be excluded owing to insufficient sensitivity. Unfortunately, the potential misinterpretation of aerosol-attached activities, because of the ambiguity of screen penetration and the poor size resolution have to be taken into account. Further uncertainties arise from non steady-state conditions during measurements and from data evaluation methods, which are mostly based on non-linear optimisation procedures.

The limited information available in the literature shows that it is necessary to determine aerosol and room conditions (such as aerosol particle concentrations, ventilation, type of aerosol and vapour sources, humidity) during measurements. In most studies, only the size distribution of the potential alpha energy concentration, rather than the individual radionuclides, was measured. Such results my be useful for older dose models, but improved techniques for spectrometric activity measurements

allow one to measure the size distribution of ^{218}Po, ^{214}Pb and ^{214}Bi. To discover differences for these radionuclides is a major goal in the understanding of the dynamics of cluster growth in the nanometer size range.

4) Various methods are used to measure the size distribution of aerosol-attached activity in the ambient air. For an aged aerosol, the size distribution can be described as a single mode, the activity being associated with particles of median diameter in the range 100 to 300 nm (accumulation mode). The geometric standard deviation of the lognormally distributed activity is in the range of 2 to 3. Aerosol particles produced by different types of aerosol sources may yield complex distributions with nucleation and coarse modes.

The poor size resolution of the screen diffusion battery technique, especially in the diameter size range of interest (accumulation mode), the ambiguity of the penetration of the screen, and the uncertainties associated with data evaluation, often yield inadequate results.

Experimental methods dependent on the analysis of the electric mobility of aerosol particles have good size resolution. However, uncertainties in the charging probability and in the attachment theory have to be considered determining activity weighted size distributions.

The best devices for measurements of size distributions of aerosol-attached activities are low-pressure cascade impactors. They have lower cut-off diameters down to 50 nm and excellent size resolution. These impactors must however be calibrated with monodisperse aerosol particles, because theoretical calculations of the response functions of low-pressure impactor stages are not accurate enough.

Despite these problems, the results cited for indoor measurements and considering an aged aerosol show that different experimental techniques yield similar size distributions. Unstable aerosol conditions and studies in the outdoor atmosphere, however, yield quite different values. These results raise some questions about the accuracy of different measurement techniques - especially diffusion methods - and also about data evaluation procedures. Side-by-side sampling is necessary to ensure such differences are real and to resolve disagreements between different techniques.

Expected achievement in this project

Values for the size distribution of the unattached and aerosol-attached activity in the domestic environment and the magnitude of the unattached activity are quite limited and consequently uncertain. For the correct determination of the real variation of these parameters under normal living conditions more efforts will be done to reduce uncertainties concerning activity measurements, to calibrate and improve size fractionating instruments, to compare different data evaluation methods, and to simulate the behaviour of airborne activities by model calculations. Most of these questions can only be solved performing experiments in the laboratory or special radon chambers under controlled and stable atmospheric conditions.

Other topics of the projects deal with the measurement of the size characteristics in different domestic environments (including working places), the determination of activity deposition in the lung (direct and indirect methods), intercomparison measurements, and the determination of the properties of thoron and their airborne decay products:

1) Chamber studies: In a radon chamber of 0.05 m^3 the IL, Göttingen, will study the influence of the trace gas SO_2 (0.01-100 ppm), humidity, and different kind of aerosol sources on cluster formation processes.

2) Metrology: The IL, Göttingen, will calibrate and test a rotating screen diffusion disk and an electrostatic classifier - in connection with a CNC and surface barrier detectors (alpha spectroscopy) - for measurements of clusters in the size range between 0.3-10 nm. Lund unversity will continue their build up, test routine and calibration of a five-stage multi-orifice impactor. University of Brest will improve the activity measurements derived from the SDI 2001 sampling device (impactor with granular bed diffusion batteries) by using alpha spectrometric methods. University of Barcelona will check their set-ups for measurement of radon gas (electrostatic method), the radon decay products (by alpha spectroscopy), and for determination of the unattached activities (by a single-screen method).

Side by side measurements of the different groups are planned to improve the accuracy of measurement techniques and to avoid systematic uncertainties. The collaboration with other groups of USA, Australia , Canada, and South Africa will be continued in this field.

3) Field measurements: Measurements will be performed by all groups with high priority in the domestic environment to determine the real variation of F, f_p, and the complete activity weighted size distribution and to estimate the influence of different living conditions of the habitants in various geographical and climatical regions of Europe. The influence of different kinds of aerosol sources and sinks (cigarette smoke, air cleaning) on these parameters are included in this studies.

Detailed measurements concerning the differences of the size distributions of the different nuclides RaA, RaB and RaC and of indoor/outdoor distributions will be performed by the IL, Göttingen, with a new developed online alpha impactor. The Ghent/Mol groups will perform measurements with combinations of different screens which simulate the deposition of activity in the human respiratory tract. All groups will also evaluate their experiments with regard to the thoron decay products ThB and ThC.

4) Theoretical studies: All experimental results of the chamber studies will be completed by model calculations on the basis of attachment and plateout processes. Room model calculations will be improved concerning the size dependence of non-steady state conditions of the concentrations of the airborne activities and aerosol particles.

Considerable effort will be spend in simulation studies and intercomparison measurements to find out the influence of different optimisation procedures and of experimental uncertainties on the final results of the size distribution measurements.

The analysis of the dynamic behaviour of the airborne radon decay products requires extended data evaluation routines considering the time dependence of the activity concentrations.

Contribution of the Isotopenlabor (IL), University of Göttingen, Germany.

Main priorities of the current radiation protection programmes of the IL are the measurement of the size distributions of the "unattached" and aerosol-attached radon decay products and the determination of the properties of thoron gas and the thoron decay products in the ambient air.

Special parts of this programmes shall be involved in the CEC project and deal with controlled chamber studies of the cluster formation processes including modelling, the development and calibration of size fractionating instruments, and the improvement of data evaluation methods:

1) A radon chamber of 0.05 m^3 was built in the IL to study the influence of the trace gas SO_2, humidity and different kinds of aerosol particles (monodisperse, cigarette smoke) on the dynamics of particle growth in the sub-nm region, the attachment to aerosol particles and radiolysis processes. The major parts of this chamber are an electrostatic classifier and a rotating screen diffusion disk for measuring particles in the diameter size range between 0.5-10 nm. After check of the performance of this complex chamber the size distributions of the radioactive particles as well as the inactive particles will be measured. The size fractionated activities will be measured by alpha spectroscopy (with surface barrier detectors) directly on the screens after air sampling, and during and after air sampling using the electrostatic classifier. The number size distribution will be determined with a calibrated condensation nuclei counter (TSI, Model 3025) with a 50% registration efficiency of 3 nm. The trace gas concentration of SO_2 shall be varied and monitored between 0.01-100 ppm to transfer the chamber results to real environmental conditions.

2) Parallel to these chamber experiments theoretical work is necessary. The size distributions will be evaluated from the measured size fractionated aerosol particles using different optimisation procedures as Twomey-, Simplex-, EM- algorithms. It is planned to find out the influence of these different fitting routines, the implication of experimental uncertainties, and the poor size resolution of diffusional methods on the final results. Especially in this field of the data evaluation procedure the close cooperation with the groups of the US-DOE (E.A. Knutson, A.C. George, USA), Clarkson University (P.K. Hopke, USA), ARL (S. Solomon, Australia), Elliot Lake Laboratory (J. Bigu, Canada), and Chamber of Mines (R. Rolle, South Africa) will be continued and further intercomparison measurements are intended.

Another topic deals with the modelling of the experimental results of the chamber studies (radioactive and nonactive particles) on the basis of the attachment theory and the size dependence of the deposition processes in the chamber.

3) Partly connected to the CEC project are size distribution measurements in the real domestic environment concerning the aerosol-attached part. To investigate differences of the size distributions of the different decay products RaA, RaB and RaC, to find

out differences of indoor and outdoor size distributions, and the investigation of the dynamic behaviour during non-steady state conditions are important topics of this project. For this purpose an online alpha impactor for low-level measurements was built and calibrated in the IL during the last two years. The size fractionated activities are deposited on surface barrier detectors and the emitted alpha particles are measured during air sampling.

Contribution of the Laboratoire de Physique des Aerosols et de Radioactivite Atmospheric (LPARA), University of Brest, France

One of the main objective of this work is to improve the a-activity counting procedure of radon daughters collected by the SDI 2000 sampling device in indoor environments. Finally, the choice was given to the carrying-out of a nine-channel counting system including nine PIPS detectors, five of 450 mm^2 area and four of 2000 mm^2, assigned to count a-activity on the filters and the last four plates of the Andersen Impactor constituting the SDI 2000, respectively.

The global counting protocol is planned to be conducted from a PC computer on the basis of a deconvolution procedure perfected at the Commissariat à l'Energie Atomique. Otherwise, it will be necessary to modify the geometry of filterholders downstream the granular beds of SDI, in order to get out samples as fast as possible.

That would result in a better evaluation of individual Radon-daughter concentration on the SDI collection samples. The prototype of this set-up would be ready by the end of April 1993 before to be experimented in field.

In parallel, a specific instrument to measure unattached fraction of Rn-daughters, time integrated over a large sampling time, is in progress of experimentation. The sampler consists in two coaxial cylinders forming an angular diffusion channel of 30 cm length, 4 mm width.

Penetration performances were numerically calculated for unattached fraction through the classical theory of diffusion but applied to an angular space of internal and external radius R_1 and R_2.

During sampling, ultrafine Radon daughters were deposited on the central cylinder covered firstly with LR 115 nuclear a track film and then with a Mylar film of thickness fitted to allow the recording of ultrafine Po-218. First experiments carried out in a test chamber, showed a non negligible number of tracks due to radon gas. Attempts of optimisation on dimensions of the channel and on sampling conditions are still in progress in order to quantify and to minimise this effect.

This research is also made in collaboration with the "Laboratoire de Physique de Métrologie des Aérosols" (LPMA) of the Commissariat à l'Energie Atomique (Dr. Boulaud).

Contribution of the SCK/CEN, Mol, Belgium

The deposition of the radon decay products in the respiratory tract is very dependent of the activity size distribution. In the recent NAS report on comparative dosimetry of radon in mines and homes the unattached fraction and the AMD of the attached fraction that are assumed to represent exposure conditions in homes vary significantly. For the unattached fraction the range is between 1% in living rooms during smoking and 16% in a well insulated bedroom. The resulting exposure dose conversion coefficient during smoking is only 4% of the normal value and in a bedroom with a low exchange rate with outdoor air the conversion coefficient is 51% higher than the values applicable to the normal bedroom atmosphere. The NAS has reviewed the research needs and strongly recommends to collect data on the size distributions of radon progeny in the indoor environment and to examine the effects of various indoor aerosol sources on these distributions. The main objective of the current research project is to collect these data in a representative number of Belgian dwellings.

In the last CEC contract (BI7*-0047.C) a measurement system has been built, according to the specifications of Hopke et al. (Health Phys. 58, 291ff (1990)), to simulate the deposition of the decay products in the nasal cavity and in the tracheobronchial region. The system is based on wire screen penetration theory and consists of three sampling heads. The simultaneous measurements are comparable within 1% so that accurate determinations can be made of the deposition in the nasal cavity and in the tracheobronchial region. The system is calibrated in laboratory conditions. The calibration was checked during the intercomparison exercise of the IAEA at Badgastein, 1991.

In the collaboration with the University of Gent, studies will be set up in more than 20 Belgian dwellings with average radon concentrations above 100 Bq/m^3. In each dwelling data will be collected during one or more days. The SCK will measure the nasal and bronchial deposition while the university of Gent monitors the aerosol concentration and inactive size distribution every 20 to 30 min. Both groups will measure the radon concentration, one group by sampling continuously and the other by grab-sampling. Aerosol with different size distributions will be produced by smoking and cooking. The experimental data will be compared with the results of the university of Göttingen and with the data available in the literature. In particular it will be investigated whether the unattached fraction and the AMD of the attached fraction for the selected exposure scenarios in the NAS report are valid. Finally the data will be fitted by the room model to test the hypothesis whether radon is a good measure of the dose in the indoor environment.

Contribution of University Ghent, Belgium:

The size distribution of the attached and unattached radon daughters are crucial parameters for dose calculations. The deposition of these decay products in the nasal cavity and the tracheo-bronchial region can be simulated by means of a measurement system based on the wire screen penetration theory (Hopke et al., Health Physics 58, 1990).

As part of the previous contract (BI7*0047.C), such a device has been built by the team of the Nuclear Research Centre in Mol and tested in the Badgastein intercomparison (October 1991).

In the current project this device will be thoroughly calibrated. In some 20 selected dwellings, with radon levels of more than 100 Bq/m^3, this instrumentation will be used in combination with the condensation nucleus counter apparatus and electrostatic classifier.

The radon concentration will be monitored both in the continuous and discrete mode. By smoking and cooking aerosol distributions of different size will be generated. In these selected houses the practicability of the passive plate-out measuring device - under development by the RARE group - for the determination of the unattached fraction, will be investigated.

The data will be fitted by the room model to control if for indoor situations radon is reliable replicate for dose. Finally the gathered information will be of basic interest for clarifying the discrepancies encountered recently in radon risk estimate.

Contribution of Polytechnical University of Barcelona, Spain

In the last two years the spanish group at the Polytechnical University of Barcelona has developed a training and equipment task linked to the Göttingen University group in the framework of the last CEC Radiation Protection Programme.

The main aims accomplished in that period are gathered in the following items:

1) Training programme of two researchers in Göttingen.

2) Development of some experiments using grab sampling measurements and charcoal adsorption techniques.

3) Setting up of a facility devoted to the continuous radon and daughters concentration measurements, using surface barrier detectors and an electrostatic deposition chamber.

4) Experimentation of the thoron daughters concentration determination by grab sampling and gamma measurements.

In the next period the following programme is planned:

1) Selection of a set of 40 to 50 dwellings representative of the habitat in the catalan region. In spite of being this spanish community a typical mediterranean area, has a wide variety of climates and geographical features.

2) Implementation, in these dwellings, of a campaign of radon concentration measurements in order to select some of them of particular significance and interest.

3) Determination of the equilibrium factor and attached fraction at real conditions in the dwellings selected in point 2. This task could be complemented with aerosol size distribution and concentration measurments. However, the latter would be dependent on the final approval by the spanish research authorities of the necessary funds to purchase the appropriate equipment. These funds have already been requested and the final decision is expected within the next three months.

4) It would be also possible to conduct the same measurements in a coal mine sited in the Barcelona province.

5) Regarding to the thoron studies we plan to carry on a set of experiments directed to optimise the method to determine the ThB/ThC fraction using alpha and gamma spectroscopy.

Contributions of the Swedish Radiation Protection Institute and the University of Lund.

Sweden is an EFTA country. The contributions towards the Swedish Radiation Protection Institute (Stockholm) and University of Lund (Lund) will be published in an addendum after signature of the Association Agreement.

C12 Exposure to natural radioactivity and evaluation of parameters influencing these risks.

Contract FI3P-CT920061 Study of the different techniques to mitigate high radon concentrations level disclosed in dwelling.

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Description of research work

It has been known for long that high levels of radon involve health hazard for underground miners of uranium-rich ores. But radon has also become a domestic word since the middle of the eighties, with the growing concern that this radioactive noble gas – and its progeny – may induce lung cancer at inhaled doses encountered in some houses. The uncertainties in evaluating excess risk attributable to high radon concentrations indoor find expression in the discrepancies between recommended maximum radon levels and, up to now, in the lack of national or international regulations. Despite these uncertainties, it is clear that remedial actions should be taken in houses with the highest radon levels (*e.g.*, over 1,000 $Bq.m^{-3}$ for existing buildings, as recommended in Sweden, or 400 $Bq.m^{-3}$, as recommended by CEC). These houses are more likely to be found in areas with the highest potential of radon emanation from rock basement and soil (accessorily, from well water). Radon surveys, that make use of geology, soil science and hydrology, but also of direct measurements of radon concentration in soil gas, are at a different stage of achievement from one country to another.

In several European countries, however, measurement campaigns – some of them within the framework of a CEC Research Action – have been carried out to assess radiation exposure of man in dwellings. The approach, that could prepare national screening programs, is to carry out measurements of indoor radon in houses located in areas exhibiting the highest radon potential. Beside geological setting indeed, radon levels in the domestic atmosphere depend on house design, living habits and, to a lesser extent, building materials.

Once it has been demonstrated that there is a radon problem in a house, a wide range of radon mitigation techniques are at the homeowner's disposal :

* ventilation (either natural or forced-air);
* house pressurization control;
* closure of soil-gas radon entry pathways;
* wall surface sealing;
* ventilation of drain-tile, subslab, block-wall, etc.

The general objective of the studies carried out under the CEC contract is an evaluation of these mitigation techniques, conceivably leading up to improvements of the existing techniques, or to thoroughly new approaches. The relative efficiency of ventilation and of radon barriers will be measured. Experiments will be carried out either in the laboratory, on ventilation loops and on material samples, or in actual houses, before and after completion of step by step remedial actions.

The number of contractors, from five European countries (namely, France, Italy, Portugal, Spain and Greece), guarantees that a wide range of specific situations will be investigated, that will be representative of most houses "at risk" in the region under concern. Emphasis will be put on differences in house design and living habits from one country to another but, simultaneously, the possibility to establish a common "house laboratory" will be investigated. Some contractors already include in their project the settling of a life-size laboratory in their respective countries.

Contribution of CEA-IPSN - Département de Protection de l'Environnement et des Installations - Centre d'Etudes de Saclay.

Since 1983, within the framework of a previous CEC contract, measurement campaigns have been carried out in France in order to assess the public exposure to indoor radon. As of 1992, about half out of the hundred French departments have been investigated, and the survey should meet with completion by 1995. With 3,500 measurements so far, in different geological settings, it is already possible to emphasize a strong correlation between radon indoor and emanating power of parent soils.

In western France, an area of higher radon potential, and with the agreement of the homeowners, it has been possible to screen the various parameters affecting radon concentrations in houses:

* architectural design;
* radon entry pathways;
* way of life of occupants.

Radioactive and environmental measurements had been carried out in these selected houses in order to design a cost effective method for determining the causes of a radon problem in a house. It has been concluded that the remedial actions should apply primarily to the source term and the ventilation.

Under the CEC contract, the efficiency of various concrete slabs, gas-tight membranes, paints, floor-coverings, sealants and drains as a radon barrier will be tested in the laboratory. Most of the samples will be provided by companies that market the corresponding material for the building trade. If possible, the testing procedure will eventually be standardized.

In parallel, the experimentally tested materials will be implemented in actual houses, with the homeowner's agreement or explicit request. The radon levels will be simultaneously monitored in order to determine the efficiency of each remedial action undertaken.

The ventilation, either natural or forced-air, will be also considered as a radon mitigation technique. The results of mathematical models describing the ventilation regime and its effects on radon and radon daughters concentration will be validated by experimental studies on actual houses. These studies will conceivably yield recommendations to be implemented on an individual or a nation-wide basis, through a building trade becoming sensitive to the radon problem.

Contribution of ENEA - Direzione Centrale Sicurezza Nucleare e Protezione Sanitaria

Italy is characterized by a large variety of dwelling types, because of the different climate conditions and the change in building practices throughout the centuries.

In spite of the fact that the Italian national radon survey is still under way, different areas with high indoor radon concentrations have already been identified with, in several instances, a strong correlation of radon levels with the building materials and design. In the ENEA project, two of these areas have been chosen for research on remedial actions.

The first area is located in the north-eastern part of Italy, and has been chosen for:

* its high levels of indoor radon in dwellings, with relatively little parent radionuclides in the soil;
* its high seismic activity, which results in a very stringent building code, and may also be responsible for the higher level of indoor radon.

The second area has been located in the centre of Italy, with the following characteristics:

* relatively high levels of radionuclides in the soil;
* building practices based on the use of local materials (typically, volcanic tuff), materials that may be similar to the soil beneath the houses.

It has been anticipated that the necessary remedial actions may be different from one area to the other. The major scope of the project is to study and identify the appropriate remedial actions for these two different settings. In an aside, remedial actions in the area from north-eastern Italy will also be studied in relation to the stringent building code of seismic areas.

Contribution of LNETI - Departamento de Protecção e Segurança Radiológica

During the survey which has been carried out in Portugal for the last years, a significant percentage of dwellings with indoor radon concentrations higher than 400 $Bq.m^{-3}$ was found. An undoubted influence of the geological make up of soils on the resulting indoor radon concentration levels was also observed.

Considering the high values that have been measured in some dwelling houses, it is important to study several remedial actions adapted to the characteristics of the country, taking into account a cost/benefit analysis.

So, the research program will consist in the following:

* as a first step, it is proposed to analyze and describe in detail the special characteristics of the dwellings from the regions where high indoor radon concentrations were found. Statistical data on climatic aspects, predominant living habits of the concerned population, typical occupancy factors, etc., will also be compiled;
* in order to identify the main radon source term in such houses, measurements of radon emanation from ground and walls will be carried out, using classical methods;
* laboratory radon mitigation experiments will be performed, considering the soil beneath the houses as well as some building materials with high radium contents, like phosphogypsum. Methods to be used in these experiments will be harmonized with the other participants. The cost/mitigation efficiency will be analyzed before trying to apply the countermeasures in real houses;
* according to the results obtained in the laboratory tests, and the willingness of the householders, remedial actions will be tested, in one or two actual rooms, and their efficiency monitored and compared.

Contribution of Universidad de Cantabria - Departamento de Ciencias Médicas y Quirúrgicas

Masonry materials such as stone, concrete and brick are often the primary source of radon from building materials. Typical building materials contain less than 70 $Bq.kg^{-1}$ of ^{226}Ra. However, there are situations where building materials that incorporate residues from industrial processes such as fly-ash, phosphorus and blastfurnace slags, etc., may contain ^{226}Ra in excess of 400 $Bq.kg^{-1}$, and hence be an important source of radon indoor.

The extent to which these materials are a major source of indoor radon, however, will depend on the parameters that determine radon transport : e.g., diffusion coefficient, permeability, porosity, emanation factor and pore water content. In order to assess the potential of building materials as a source of radon, an additional difficulty is the measurement technique and the analysis of data. To benefit from existing experience, and to allow intercomparison of data, UNIVERSIDAD de CANTABRIA (UC) will co-operate with LNETI and NCSR"D" on measurement protocols and evaluation of radon emanating from building materials.

The purpose of UC project is to evaluate the contribution of granite and concrete with high radium content on indoor radon, not only in the laboratory but also through *in situ* measurements.

Laboratory measurements of the parameters referred to above will be performed on these particular building materials, analyzing the advantages and problems of the different measuring techniques presently in use. These data are basic for the study of remedial action in the high-radon houses. Samples from areas where high indoor radon levels were found during the national radon survey carried out by UC team, within the framework of another CEC contract, will be selected. Specific intercomparison exercises will be organized between the participants.

A test structure will be built in the relevant UC laboratory, where to compare measurements on different kinds of granites and thicknesses of samples with the values derived from mathematical models. Techniques to mitigate high radon concentration levels in this test structure will also be investigated at last. Some experiences will be implemented to find out the conditions under which the permeability of the building materials may be responsible of the presence of high radon levels in houses. UC shall also evaluate the efficiency of some caulking compounds that could be used for radon mitigation, not only in ancient dwellings, but also in new ones.

Contribution of NCSR "DEMOKRITOS" - Environmental Radioactivity Laboratory

For the 1992-1993 period, the Environmental Radioactivity Laboratory of NCSR"D" intends to concentrate on the topic of indoor radon sources, with emphasis upon the contribution from building materials. This is of special interest for Greece, as long as:

- the modern building practices lead often to pylon-based constructions characterized by the absence of basement spaces and the enhanced isolation from the soil radon sources. In such cases the contribution of the building materials is expected to dominate;

* materials of high ^{226}Ra content are commonly used, either directly or as components, for building purposes. The characterization of these materials as indoor radon sources is far from being satisfactory.

In the field of methodology, NCSR"D" shall closely collaborate with LNETI and UNIVERSIDAD de CANTABRIA on the measurement protocols and evaluation of radon coming out from building materials. At the same time, NCSR"D" shall work on the development of simple, but reliable, procedures (non- or nearly non-destructive) for *in situ* evaluation of the exhalation rate of radon from building materials.

NCSR"D" shall pay special attention to the use of fly-ash and other components of enhanced ^{226}Ra content in cement, as well as to the application of some decorative materials (floor and wall slabs) of high radium content. Suitable models will be developed in order to evaluate the contribution of these sources to the total indoor concentration of radon under various conditions (material usage, surface and volume parameters of the building, ventilation rates, etc.).

In a further phase, attention will be paid to those building materials, which appear (experimentally) or are supposed (theoretically) to be sources of enhanced indoor radon concentrations. In these cases the project will be gradually shifted towards the test of alleviative counter-measures directed against the specific building material revealed as radon sources.

NCSR"D" shall collaborate with the Department of Medical Physics of the ATHENS UNIVERSITY (AU), on the interpretation of the results of the AU national survey, in terms of the building material contribution as a source of indoor radon.

Contribution of the Athens University - Department of Medical Physics

The Department of Medical Physics (MPL) has been setting Terradex radon detectors since 1987, in dwellings of several cities and other selected locations in Greece. In 1991, however, MPL started implementing track-etch detectors of its own. The radon measurement method by means of MPL-designed detectors has been subjected to testing and intercomparison with commercially available Terradex detectors. In parallel with radon measurements, spectroscopic analyses of soil samples from areas close to the same dwellings have been carried out, in order to measure ^{226}Ra in surface soil and possibly to correlate its concentrations with indoor radon levels.

The selection of the above mentioned locations are based on unpublished results from a research program implemented by the Nuclear Engineering Section of the National Technical University of Athens, at 588 sampling locations throughout the country, and aiming to map the natural radioactivity of soils in Greece.

The detector measurements have been carried out with the use of a microscope, a time consuming method that introduces subjective errors, since it is based on optical observations. Therefore, the use of a dedicated image analysis system is being considered in order to derive the measurements automatically. At this stage, MPL is in the process of obtaining such a system, that could be made operational before mid 1993. In this prospect, and given the appropriate manpower, MPL plans to carry on the national

survey of indoor radon concentration. The statistical sample will be determined on the basis of the population and size of each region of Greece.

Other objectives of MPL future research include intercomparison of optical and automatic measurements, and expansion of efforts on the study of correlations between indoor radon concentration in dwellings and uranium concentration in the adjacent soil. It is understood that an important consequence of the study will be the transfer of technology for radon measurements and the training of young scientists in radiation protection techniques.

In parallel, a procedure of studying the dependence of the indoor radon concentrations on the type of building materials found in the Greek dwellings will be developed. The relevant measurements will be performed on samples of various building materials used in Greece and/or on specially designed models, in close cooperation with NCSR"D".

C12 Exposure to natural radioactivity and evaluation of parameters influencing these risks.

Contract FI3P-CT920064d Radon sources models and countermeasures.

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Description of research work

Objectives:

The objective of the project is to enable people to avoid excessive exposure to radon decay products by improving the understanding of the sources of radon, how it moves through the ground and materials and into buildings, and the measures to counteract ingress. This will be achieved by developing techniques to map the probability of high radon levels occurring indoors, by the use of models and experiments on the movement of radon in the ground, building materials, sub-floor spaces and buildings, and by developing and testing preventive and remedial measures.

State of the art:

Radon mapping techniques are available for certain well-defined geologies, but a better understanding of radon and geology and more generally applicable techniques are required. The movement of radon in soil as a function of the permeability, porosity, pressure differentials and other factors can be modelled, but present models do not always agree with measurements. Further development of models and comparison with results of laboratory and field experiments are required. Similarly the understanding of movement of soil air into and within buildings is imperfect. Effective intervention techniques have been developed for certain situations, but their reliability needs testing. The development of countermeasures will be advanced by the improvement of understanding of radon transport.

Degree of innovation:

The techniques to be used in this project are established, but being applied to new areas of study, or are developments of techniques that have been used by the collaborating laboratories in the first stage of the project.

Economic, social and technical benefits:

Radon has been clearly established as a cause of lung cancer, as recognised by the CEC in its recommendation 90/143 Euratom, Protection of the Public against Indoor Exposure to Radon. The purpose of this project is to provide the means for governments and individuals to avoid excessive exposures to radon.

Relation to other RTD programmes:

The CEC is supporting various other projects on radon. These concern studies to refine the risk estimates for radon exposure, development of a retrospective measurement technique, and examination of the behaviour of radon decay products and implications for lung doses. These projects are complementary to the project described here, but are sufficiently self-contained not to require day-to-day links. Nevertheless, the scientists working on this project have extensive and regular contacts with those working on the other projects and in similar fields both within and outside the EC.

Scientific and technical description:

This project covers a large area of work, and for the purposes of coordination and collaboration it has been organised in two sub-projects: radon mapping and radon models and experiments.

Radon mapping

Two laboratories, NRPB and BGS are working in this area.

BGS will study the geological aspects of radon potential. The proposed study will extend the examination of granite and limestone areas to areas adjoining these, and also to other areas with limestones, calcareous shales, clays and sands. In particular the implications of high radon concentrations in limestone areas will be examined in relation to the large areas of Europe which are underlain by this rock. The primary measurement technique used by BGS will be measurement of radon and thoron in soil gas, with measurement of uranium, thorium, radium and radon concentrations in water and rocks where appropriate. The results will be studied in relation to the measurements of radon in homes made by NRPB, and the radon potential of different rock types mapped.

NRPB will provide assistance to BGS in calibrations and measurements, supplying BGS with results of measurements in homes and testing the radon potential maps produced by BGS. NRPB will also use its database of more than 100,000 radon measurements in homes to develop and test techniques for mapping radon affected areas accurately and economically. Different modelling and smoothing techniques will be compared to determine the most accurate means of mapping radon based on limited data. The use of population density maps with radon potential maps to identify areas where large numbers of homes need to be remedied will also be examined.

Models and experiments

Five laboratories are involved in this area of work: CSTC-WTCB, RISOE, RD-TNO, KVI and NRPB. Collaboration between the laboratories will take place in various areas: a summary is given at the end of this section.

KVI will study transport of radon in soil and from soil into crawl spaces. In the previous funding period a large vessel containing sand was built, with multifunctional probes inserted to measure various parameters. The base of the vessel can be pressurised or depressurised or filled with water. Radon profiles will be measured as a function of water level, pressure difference, height of the lid and ventilation rate under the lid. The data collected will be used to validate radon transport codes. The effect of countermeasures will also be studied in the vessel.

RISOE will continue its studies of steady-state radon transport and entry with detailed modelling based on measured soil parameters and comparison with experimental results obtained at their test structure. This structure is a 40 litre stainless steel cylinder in an excavation on the RISOE site. An investigation will be made of how detailed both soil characterisation and modelling need to be to account for soil

inhomogeneities. The model will be tested by comparison with data from houses monitored by KVI; it will also be used to investigate the influence of building and soil related factors on radon entry rates.

CSTC-WTCB will use a finite difference code to simulate radon mitigation strategies, particularly the use of subslab depressurisation in cases where there is no highly-permeable gravel layer beneath the slab. The model will also be used to evaluate the flow of soil gas into the test house. CSTC-WTCB will also use a ventilation model to study the effects of weather, building characteristics and ventilation systems on radon transport both with and without the deployment of radon countermeasures. Detailed investigation will be made of one or more buildings with high radon levels in order to determine the factors influencing radon entry and to identify appropriate remedial measures.

RD-TNO has developed a model to predict radon infiltration into buildings, and have linked this to a ventilation and transport model. The model uses a large number of input parameters, some of the values of which are not well known. RD-TNO will examine the literature on the normal ranges of values of parameters, and evaluate their interrelations. It will also examine the influence of weather conditions, soil parameters and construction-related parameters. The basic aim is to identify those parameters with the largest influence on radon concentrations.

The studies of the effectiveness and durability of countermeasures against radon in new and existing homes being conducted by NRPB will continue. It will also study the correlation of permeability under slabs with building characteristics and ground conditions. Further work will be carried out to obtain a better understanding of ventilation under suspended timber floors, and to develop more reliable remedial techniques for this type of construction.

Collaboration on models and experiments:

KVI and RD-TNO will verify their respective indoor radon flow models using data from the KVI research house. CSTC-WTCB will collaborate on the evaluation of the flow of soil gas into the research house. KVI, RISOE and CSTC-WTCB will cooperate on the evaluation of the pressure-dependence of the radon entry rate from soil into crawl spaces for the KVI test house. KVI and RISOE will evaluate the time-dependent radon profile in the KVI research vessel. TNO, KVI, and RISOE will collaborate in the verification of TNO's model concerning the radon entry rate.

Complementarity, expertise contributed and benefits to be gained

Each laboratory has its own expertise in radon, in fields such as geology, measurements or modelling. The laboratories have been grouped together under two headings: radon mapping, and models and experiments. There is some overlap between these groups. The benefits to be gained from the collaboration vary. For instance, in the first group NRPB will supply radon house data to BGS, to assist in the production of radon potential maps. In the models and experiments group, various data sets from measurements in the field and the laboratory will be shared to allow the testing of models.

CONTRIBUTION OF THE NRPB

The National Radiological Protection Board will act as coordinator of the project.

It will contribute to the development of radon potential mapping in two ways: by collaborating with the British Geological Survey (BGS) in its study of the geological correlates of high radon in homes, and by developing mapping techniques based on the measurement of radon in homes. NRPB will calibrate BGS radon measurement equipment, will provide data on radon levels in homes and other buildings, and will test the radon potential maps that BGS produces.

NRPB will also develop mapping techniques based on measurements of radon in homes. It has a database of about 100,000 radon measurements in homes made under standard conditions in the UK, the great majority in southwest England. The high density of measurements in this area allows the use of the data to develop and test techniques for mapping radon affected areas accurately and economically. A subset of the data will be used, with modelling and smoothing techniques, to create a map of the probability of radon levels in homes exceeding a given threshold.

The accuracy of the map will then be tested by comparing it with the full data set. Different modelling and smoothing techniques will be compared to determine the most accurate means of mapping radon based on limited data. The size of the subset used as initial input data will be varied to determine the minimum data requirements for accurate mapping. The effects of including data from a questionnaire for householders, such as storey of measurement and presence of double glazing, will be tested. The use of population density maps with radon potential maps to identify areas where large numbers of homes need to be remedied will also be examined. The outcome of the exercise will be a set of recommendations for survey, analysis and mapping techniques to allow the rapid and economical production of accurate radon potential maps and the identification of areas that need attention.

The studies of the effectiveness and durability of countermeasures against radon in homes being conducted by NRPB will continue. The studies so far have shown a great variation in effectiveness depending on the countermeasures adopted. New homes with preventive measures and existing homes with remedial measures will be studied with repeated measurements over the contract period to determine the durability of the measures and to expand the database on the effectiveness of different techniques. One factor that strongly affects some radon remedial measures is the permeability of the ground immediately below solid floors.

Measurements have been made in the USA and the UK of the horizontal extension of low pressure fields induced under floor slabs. NRPB will study the correlation of permeability under slabs with building characteristics and ground conditions.

Remedial measures have proved ineffective in some homes with suspended timber floors. Further work will be carried out to obtain a better understanding of ventilation under suspended floors, and to develop more reliable remedial techniques for this type of construction.

CONTRIBUTION OF THE UNIVERSITY OF GRONINGEN KVI

Transport of radon through porous media, like soil and building materials has proven to be a complex problem, both in the theoretical description as in the measurements techniques and analysis. The objective of this project is to obtain better understanding of transport of radon in soil and in particular transport from soil to crawl space. This objective will be met by studying under controlled conditions both transport of radon in soil and radon exhalation by soil as a function of a number of parameters.

This project is a continuation of the project started under the 1990-1991 Radiation Protection Programme of the CEC. In the initial stage a large vessel was built and installed. Multi-functional probes, inserted radially, allow the measurement of pore water content, air permeability of the soil, and radon concentration in soil gas in the centre of the vessel. The vessel is covered with a lid that can be adjusted in height. The vessel is filled with sand; in the sand near the bottom a box with a diameter somewhat smaller than the diameter of the vessel is installed to (de)pressurise the soil with respect to ambient. A flange at the bottom of the tank contains in addition to the feed-through for the (de)pressurisation valved in- and outlets for water. In this way a water level can be set.

The transport mechanisms of radon in soil are studied by measuring and analysing radon profiles in soil as well as radon concentrations under the lid as function of water level, pressure difference, height of the lid and ventilation rate of the space under the lid. The first investigations will be carried out with a homogeneous, well defined sand of which grain size distribution, water retention capability (pF), radium and thorium content, and emanation factor for radon and thoron were determined separately. The level of understanding will be improved by validating recently developed radon transport codes.

In this project we work in collaboration with A. Damkjaer (Tech. Univ. Denmark) (permeability measurements), Drs. P. Cohilis and P. Wouters [CSTC/WTCB] (calculation of pressure fields) and with Drs. B. Majborn and C. Andersen [RISO] (calculation of radon profiles).

CONTRIBUTION OF RISOE

Motivation and background: The objective of this project is to obtain a better understanding of radon transport in soil and entry into houses using numerical modelling and related experimental field-studies at a special radon test structure. Our two-dimensional model solves the equations for steady-state soil-gas and radon transport including emanation, decay, diffusion, advection, and partitioning of radon between gas and liquid phases. The model is flexible, and problems related to our test structure or to real houses can be analyzed in detail. The test structure provides a relatively well-defined object for model studies. The (de)-pressurization of the structure can be set by a mass-flow controlled pump, and pressure couplings, radon concentrations, and gas permeabilities can be measured in more than 20 probes located in various depths of the subsoil.

Contributions: (1) We will continue our studies of steady-state radon transport and

entry with detailed modelling based on measured soil parameters and comparison with experimental results obtained at our test structure. We will attempt to improve the soil characterization, and to minimize the uncertainties related to the experimental and modelling procedures aiming at a reduction of the observed discrepancies between initial model calculations and measurements. In addition, we plan to investigate dynamic effects, i.e. entry rates of radon in response to time-dependent depressurizations of the test structure. (2) The steady-state model will be tested by comparisons with data from real houses. This work will be carried by KVI (the Netherlands) using experimental data obtained at their respective research houses. Subsequently, the model will be applied to investigate the influence of building and soil related factors on radon entry rates with an emphasis on slab-on-grade houses.

Interactions: (1) Risoe will collaborate on the study of radon entry using the Risoe model and data from the research house. (2) Risoe, KVI, and CSTC (Belgium) will collaborate on the evaluation of the pressure dependence of the radon entry rate from soil into crawl space for the KVI research house. (3) Risoe and KVI will evaluate time dependent radon profiles in the KVI research vessel. (4) Risoe, KVI, and TNO (the Netherlands) will collaborate in the verification of TNO's model concerning the radon entry rate.

CONTRIBUTION OF CSTC/WTCB

The studies begun under the 1990-92 contract are being continued under the new contract, with special attention to air movement into and inside buildings, and also in the ground. The general objective is to study, from theoretical and experimental point of view, the role of ventilation and air flow patterns in the framework of radon, with a particular attention to radon reduction techniques related to these parameters.

a) Prediction of pressure fields and air flows from the ground into buildings:

The TRISCO model will be used as a tool to simulate radon mitigation strategies (sub-floor ventilation strategies). For example, an important point concerns the application of the subslab depressurization (SSD) technique in difficult situations. The influence of some important parameters on the characteristics (number/location of the suction pipes, pressure inside the pipes,...) needed to obtain enough depressurization below the slab will be studied. This study can help to gain some feeling on how to define and implement SSD systems with a high probability of success. The model will also be used in a collaboration with SSI (Dr. L.Hubbard) for the evaluation of the flow of soil gas into the SSI research house.

b) Study of the influence of climatic and building parameters on the air flows and pollutant transport in buildings, and on the performances of radon reduction techniques based on building ventilation:

Air flow patterns within a building influence the propagation of airborne pollutants. They are influenced by climatic and operational factors which are, together with other information, included in the VENCON model. We plan to use this model in a way that takes into account the peculiarities of the radon problem. In particular, we will consider the influence of the climatic conditions, the building characteristics and the

use of ventilation systems on the radon transport inside buildings, and also the influence of climatic conditions, building airtightness, and occupants behaviour on the performances of ventilation strategies aimed to reduce radon levels in buildings. This will show the importance of some parameters for the definition, with a good probability of success, of remedial actions based on natural/mechanical ventilation. For the evaluation of the pressure dependence of the radon entry rate from soil into crawl spaces the model will be used in cooperation with KVI (Dr. R.J.de Meijer) and RISOE (Dr. B.Majborn), using the KVI test house data.

c) Investigations in buildings:

Investigations will be performed in one or more buildings with high radon levels. The factors influencing the radon entry and determining the concentrations measured inside the buildings will be investigated. A number of countermeasures will be proposed, evaluated and compared. Radon concentration measurements, pressurization techniques, pressure distribution measurements and (if necessary) tracer gas techniques will be used. Besides of developing, testing and gaining experience concerning remedial actions, the aim of these studies is also to suggest answers to important questions concerning, for example, the kind of investigations that have to be performed in order to allow the proposal of appropriate remedial actions.

CONTRIBUTION OF THE NERC

The role of the British Geological Survey in the project is to provide the geological and geochemical expertise and information. This has been in the production of radon in soil gas distribution maps, and provision of data sets for existing and new measurements for the naturally occurring radioactive elements in rocks, soils and groundwaters. It has investigated the use that existing geological and geochemical data sets; new measurements of soil and bed-rock radioelement concentrations; and in particular the determination of soil gas radon levels, might have to the efficient recognition of areas with high radon values in houses.

The contribution of the BGS has included field and laboratory measurements of radon and thoron in soil gas, mine and cave air. Where relevant, groundwaters have been analysed for uranium, radon, radium and other related decay products. The uranium, thorium and radium contents of soils and rocks have been determined. It was proposed to extend the areas investigated to include regions adjoining those covered in detail by the current investigations. The housing stock for the whole of the county of Derbyshire is being investigated by the NRPB and it is proposed to study the logistics of extending out from the well surveyed Chapel en le Frith area, mapped in the first round, to include similar lithologies in neighbouring areas and hence to develop procedures for covering larger areas more efficiently.

Another area of concern is that of Somerset, which adjoins the radon affected counties of Cornwall and Devon, and where a large NRPB housing data set is available. Soil gas surveys carried out last summer will be compared with the NRPB house data set. Rock types ranging from various limestones through calcareous shales to clays and relatively unconsolidated sands are available for testing.

Limestones have been shown to generate large quantities of radon. Most of the related work to date has been in areas of cavernous limestone (heavily karstified). Preliminary data indicates that other limestones which have not been so extensively subjected to karst forming processes also produce high levels of radon in houses, and current and future investigations extend to areas underlain by these rock type variants. Data obtained from Somerset and Derbyshire will be used in this more thematically based investigation. These rocks are known to underlie large areas of Europe and several major towns are located upon limestones.

Most collaborative work has been with the NRPB but good contacts have been maintained with colleagues from Athens University and the Technical University of Denmark, with particular attention paid to establishing uniformity of calibration and interpretational procedures.

C12 Exposure to natural radioactivity and evaluation of parameters influencing these risks.

Contract FI3P-CT930074 Evaluation of the combined helium/radon in soil gas mapping methodology as an indicator of areas in which elevated indoor radon concentrations may be found.

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Total Contribution by the Commission: 95 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Radon mapping methodologies, based on established soil gas sampling techniques, are being developed in several EC and non EC countries. The ability of these mapping procedures to successfully identify areas with elevated indoor radon concentrations has not so far been critically assessed. This project aims, for the first time, to assess the effectiveness of the combined Helium/Radon in soil gas mapping methodology in identifying such areas. The technique of ground probing radar will also be assessed in the context of its usefulness in site investigations and foundation investigations. In addition the meteorological influences on radon exhalation from the ground will be investigated, as will the temporal and spatial patterns of indoor radon concentrations in houses.

The objectives and expected achievements of the project are

(a) To assess the effectiveness of the Helium/Radon in soil gas mapping methodology as an indicator of areas in which elevated indoor radon concentrations may be found.
(b) To investigate the temporal and spatial patterns of indoor radon concentrations in houses.
(c) To investigate the variations in radon exhalation from the ground due to meteorological conditions, and
(d) To improve our understanding of the geological factors which control the generation, migration and ingress of radon into houses.

Scientific and Technical Description

Radon monitoring in Ireland to date has indicated that the majority of the elevated indoor radon concentrations are spatially associated with limestone regions, which comprise up to 40% of the land area of the country.

Previous investigations in a limestone region in western Ireland (contract Bi7-0059) have shown that this rock type is prone to dissolution by percolating groundwater (karstification), and that the consequent increase in permeability is probably significant in facilitating the migration of free radon to the surface. Consequently built up areas underlain by such limestone which have not been extensively surveyed for indoor radon concentrations may also have a significant proportion of houses with elevated indoor radon levels.

A multidisciplinary team approach by institutes in Ireland (RPII, GSI) and Germany (Enmotec, GAU) will take place in two stages. The first stage will involve the execution of an integrated field campaign by the project team in a preselected carboniferous limestone region. The elements of the field campaign will comprise

(a) Application of the combined Helium/Radon in soil gas mapping methodology at reconnaissance and detailed scales to delineate soil gas radon concentrations and radon migratory routes to buildings (Enmotec).

(b) Simultaneous and continuous measurement of meteorological parameters (wind speed and direction, soil and air temperatures., atmospheric pressure, pressure differentials between soil and air and precipitation) at selected sites. Continuous measurement of the soil radon profile will be made using the electroprecipitation method and the result compared with measured soil radon exhalation rates into a test volume at surface (GAU).
(c) Computer integration of available geodata for the study region (RPII).
(d) Detailed indoor radon investigations of houses in the study region incorporating (i) Active and passive radon measurements and (ii) an evaluation of occupancy patterns for residents in the study region (RPII).

The second stage of the work programme will involve evaluating the effectiveness of the mapping methodology as an indicator of areas within the study region in which elevated indoor radon concentrations may be found. The elements of this part of the work programme will comprise.

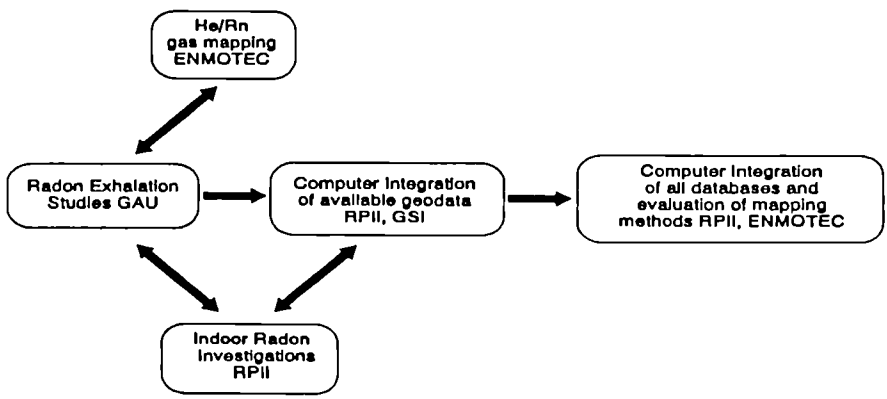
(a) Computer integration of all the databases generated in the field campaign (stage 1) (RPII, Enmotec), and
(b) Possible further indoor radon monitoring if coverage proves to be low in relevant areas of the study region (RPII).

It is envisaged that the results of the research will have wide applicability in other EC member states where many population centres are situated on common bedrock lithologies.

In terms of the schedule of work, it is proposed that the project should commence on 1st December 1992 and end on the 31st May 1994 (18 months)

Distribution of Tasks and Collaborative Links

The distribution of tasks between the participants are summarised under the technical description given above and the collaborative links are shown below. The USGS (Dr. G.M. Reimer) will participate in the project on a self-funded basis to provide expertise in He soil gas mapping methodologies.



Contribution of the Radiological Protection Institute of Ireland

The Radiological Protection Institute of Ireland (RPII) is the principal Government Agency with responsibility for radiation matters in Ireland. Since 1989 the RPII has been actively engaged in indoor radon gas surveys and has recently extended its radon monitoring programme to incorporate contract work for the private sector and other Government Departments, mainly in schools and workplaces.

The contribution of the RPII to the project will incorporate.

(a) Indoor radon gas measurements using closed, alpha track (Cr-39) radon detectors. The exposure period will be for three months with measurements replicated throughout the duration of the project.

(b) At selected houses within the designated survey area indoor radon measurements at several locations within each home using (Cr-39) alpha track detectors. The exposure period will be for three months with measurements replicated throughout the duration of the project.

(c) At selected houses continuous radon gas measurements over periods of three months will be carried out using Pylon AB-5 Radiation System in parallel with passive radon gas measurements.

(d) If possible one or two houses will be selected for detailed analysis incorporating

i) continuous ground floor radon gas monitoring
(ii) continuous sub-slab radon gas monitoring
(iii) continuous indoor/sub-slab pressure differential monitoring
(iv) continuous indoor/outdoor temperature difference monitoring

Measurement periods will be of the order of several months.

(e) Evaluate occupancy patterns for residents in the survey area

(f) Compilation of available geodata with the assistance of GSI

(g) Preliminary digitization of all databases prior to final computer integration.

Contribution of the Geological Survey of Ireland

The Geological Survey of Ireland (GSI) is the national geoscience agency responsible for the acquisition and dissemination of geodata in Ireland. The agency acted as co-ordinator of a previous project funded under the Radiation Protection Programme of the C.E.C. (Contract Bi7-0059) aimed at assessing the geological factors which influence the occurrence of high indoor radon in karstic limestone regions. The findings of that research suggest that the enhanced permeability of such karstified limestone rock sequences strongly influences surface radon availability. Zones of enhanced permeability

(e.g. fractures, faults, shear zones) in the vicinity of dwellings may account for high indoor radon concentrations. Such zones are best delineated by active soil-gas sampling of both radon and helium.

The current proposal is aimed at further evaluation of active soil-gas mapping techniques in a preselected region underlain by granite and/or limestone sequences. The precise source and migratory routes of radon to dwellings in this area is not well understood but the development and application of effective soil-gas mapping methodologies should constrain possible models.

The GSI will support the detailed and systematic geological mapping of the selected area and its immediate environs by providing geological maps and data for the designated areas. Together with other partners (RPII, Enmotec) GSI will assist in the assessment of the predictive effectiveness of the mapping methodology and contribute to recommendations on its application to mapping of other EC radoniferous environments.

Contribution of ENMOTEC G.m.b.H., Tübingen, Germany

ENMOTEC is an independent consulting company which has for years been active in environmental protection projects financed by the EC, the German Government, the Government of Burundi (Africa), international government institutions and private companies.

Dr. Günter van den Boom, senior scientist, was project leader of the German group in the EC Radiation Protection Programme project: "Assessment of the geological factors influencing the occurrence of radon hazard in areas of karstic terrain" (1990-1992) co-ordinated by the Geological Survey of Ireland. The guarantees a continuation of the application of new combined radon-helium soil gas method used during that programme.

Enmotec's contribution to the current project will incorporate the following fields of applied research:

1. Application of the combined helium-radon method to detect and quantify hazardous areas with radon contaminated soil-gas in population zones in Western Ireland. New sampling techniques and fast reliable radon analysis will supply the field party with immediate results to enable maximum flexibility in sampling campaigns.

2. Analysis of helium concentration in soil gas will allow detection of radon migration paths in underlying rock units and soil cover. Location and regional distribution of migration paths for terrestrial gases are important for modelling the behaviour of radon distribution in the soil and selection of areas with high radon potential.

Delineation of high radon potential areas will allow indoor radon measurement programmes to be properly targeted, thus reducing the time required for these measurements. The applied research work will furthermore include testing a new permeability method for radon availability studies.

The work will begin with intercomparison and calibration measurements among project participants and will incorporate systematic repeat measurements at permanent stations to determine the parameters that might influence the acquired soil gas data. A time-series study to evaluate these measurements will follow.

During the field programme (summer 1993) a radon and helium survey will be carried out in the designated survey region. A radon potential map will be produced showing endangered populated areas. Potential radon migration zones will be shown on a map based on the helium survey and geological data (fault and joint systems).

In summer 1993, in close co-operation with other partners, it should be possible to establish a correlation between radon potential in soil gas, proximity to migration paths and radon contaminated houses. This will allow definition of areas of radon risk on sites reserved for house construction with the possibility of preventative measures being taken prior to habitation.

Contribution of the Isotopenlabor (IL), University of Göttingen, Germany

The aerosol/radon research group of the isotopenlabor has long experience in the field of radon gas measurements in indoor and outdoor environments and the laboratory is well equipped with high sensitivity monitors for continuous measurements.

In most field studies the transport of radon in the soil is determined by taking grab samples of radon soil gas or by measuring the radon entry into a test volume. From these data radon exhalation rates are calculated. Since field measurements are performed over a period of days, it would be of interest to measure the changes of radon exhalation due to meteorological conditions.

The daily and seasonal variation of radon concentration in the open atmosphere is mainly determined by the change of the turbulent exchange rate. However, we have no information about variation of the exhalation rate, its convective fraction and the influencing parameters. The results of some preliminary measurements in a dwelling show clearly the great influence of the meteorological parameters on the pressure difference (indoor-outdoor) and its strong correlation with the radon concentration indoors. From these measurements we can conclude that the magnitude and variation of the radon concentration can only be explained by assuming a convective radon transport from soil into the indoor air, which changes with weather conditions.

It is proposed to monitor radon transport in the soil (near the surface) over a time period of several months while the main meteorological parameters like windspeed, temperature in soil and between soil and atmosphere and precipitation are measured simultaneously and continuously. In addition to these measurements, grab sampling types of exhalation measurements will also be carried out and the radon concentration in the air above the ground will be measured continuously.

The radon transport in the soil can be registered by measuring the vertical profile of the radon concentration in the soil air. This method must be modified to allow continuous measurement without disturbing the concentration profile. Therefore, the sampled air from the soil depth must be pumped back after the measurement. This can be achieved

by using a continuous radon measurement technique, like the electroprecipitation method. The results of this method will be compared to another method which measures the radon entry rate by molecular diffusion into a test structure which is connected tightly to the soil surface. The latter will be applied simultanuously with the profile technique. The electroprecipitation method with on-line alphaspectrometry will be used as a radon sensor. Temperatures will be measured with ventilated Pt-100 resistors, wind speeds with cup anemometers and soil humidity with a tensiometer in ten-minute values.

C13 Comparative assessment of exposure and risks.

Contract FI3P-CT920019 Comparative assessment and management of radiological and non-radiological risks assiciated with energy systems.

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Total Contribution by the Commission: 160 KECU
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Description of research work

INTRODUCTION

With the rising demand for energy and the increased awareness of the importance of environmental protection, it becomes increasingly necessary to focus not only on the health impacts of energy systems but also on the link between energy production and the environment. The initial aim of energy comparison studies was to rank the various electricity production systems. It is now recognised that this was merely an academic exercise, with a limited impact in decision-making. If comparisons, and choices, are to be made, it is essential that a consistent approach is taken to assess each energy system of interest. During the first phase of this project a general framework was developed to incorporate health and environmental risks, in a consistent manner, into the decision-making process. This framework is currently being utilised for a comparative risk assessment between the coal and nuclear fuel cycles for CEC-DGXII- Radiation Protection, a decision-aiding system for EdF-Mission Environment, and a DGXII-Environment programme to assess the External Costs of Fuel Cycles. During development and the initial implementation, issues that still need to be resolved have been identified.

During the next phase further developments of the risk indicators for the health and environmental risks will be carried out, and the results of the analysis will be incorporated into a framework for multi-criteria utility analysis. A preliminary assessment of the uncertainties associated with the risk assessment and the comparative analysis will also be conducted.

OBJECTIVES

The objectives of this work will be to complete the final development of the methodology for the comparisons of the risks involved in the generation of energy and to conduct a test case for the comparison between the nuclear and coal fuel cycles in France and Germany. More specifically this will involve:

(1) the further development of health and environmental indicators of risk within a time/space matrix;
(2) integrating the results of the risk assessment into a framework for multi-criteria utility analysis using the data for coal and nuclear fuel cycles ; and
(3) estimating the range of uncertainty associated with the risk estimates used in the multi-criteria utility analysis.

CURRENT STATUS

During the 1990-1991 Radiation Protection Programme, the project "Expression of Radiological and Non-radiological Detriments in the Comparative Assessment and Management of Risks Associated with Energy Systems" was initiated to develop a methodological framework for the assessment of environmental and health impacts of energy sources.

The aim of the work was to collect the information available and develop a coherent methodological framework to assess and manage the health and environmental impacts of the nuclear PWR cycle compared with that of those of the coal cycle. The framework has been developed to allow for direct comparisons between the two fuel cycles and allow for incorporation into decision making processes related to risk control.

The key issue for developing a comparative work has been to attain consistency between the fuel cycles. Each stage of the two fuel cycles are treated separately, as well as the analysis of construction, decommissioning and transportation of materials between stages. To assess the health and environmental risks from environmental releases the impact pathway approach has been utilised.

The classical dimensions used in comparative studies are the health impacts on the public and workers under normal and accidental operations have been maintained. The dimension of time is addressed in short, medium and long term impacts, and space is divided into local, regional and global impacts. For each step of a fuel cycle a matrix of time and space was constructed and the assessment of the risks applicable for each cell of the matrix was completed. The transfer of the releases through the environment and ultimately to the receptor is accomplished by pathway analysis specific to the reference environment and the composition and type of release.

The health impacts for the public and workers have been reported using indicators of: mortality, morbidity, genetic effects, years of life lost, working days lost and occupational injuries. At this time, direct comparisons between the two fuel cycles can be made using the indicators of deaths or worker-days lost for the occupational population. New indicators have also been proposed to describe permanent disabilities.

DETAILED DESCRIPTION OF TASKS

(1) the further development of health and environmental indicators of risk

Environmental indicators, are needed to report the resulting environmental damage from the air and water pollution (including agricultural and recreational use). The different pathways between nuclear and coal will be harmonised in terms of the start point and endpoint of the pathway, common indicators describing the damage/risks will be reported in consistent time/space scales . Direct impacts on the environment will also be considered for normal and accidental operations.

(2) integrating the results of the risk assessment into a framework for multi-criteria utility analysis

The risks that are reported for the coal and nuclear fuel cycles will be used to conduct a comparative assessment using multi-criteria utility analysis methodology. Once the hierarchy of criteria are set and the risks are estimated, it will be necessary to find a way to rank the criteria in terms of importance and societal preferences. The challenge for this part of the work will be to find a way to quantify the importance of the risks. It is planned that a workshop be organised to bring together experts for the purpose of

estimating weighting factors needed to conduct the comparative analysis of many different factors. Using interactive software it will be possible to determine the sensitivity of many of the factors.

(3) estimating the range of uncertainty associated with the risk estimates used in the multi-criteria utility analysis.

Whether the impact data are used for academic risk analysis or for energy and policy making, it is necessary to have an idea of the uncertainty associated with the estimates. This is a complex and difficult task. In many cases the information needed is not available. Within the context of this project, work will be conducted to try to ascertain an idea of the level of uncertainty that is associated with the many different methods used for estimating the health and environmental risks.

NUCLEAR FUEL CYCLE - CEPN

An overall assessment of the risks of the nuclear fuel cycle has been completed during the past phase of the project. In some cases these risk estimates will be revised and more detailed assessments will be included. As the health and environmental indicators are finalized the estimation of these impacts for the nuclear fuel cycle will be completed. If more work is required to clearly report these data in terms of the time and space matrices, this work will be performed.

The compilation of all the risk data available for use in the multi-criteria utility analysis will be incorporated into a small database to be utilized by available software A model will be defined including the relative criteria to be evaluated, evaluation of the alternatives with respect to the criteria (estimates of the risk indicators), and evaluation of the relative importance of the criteria (weighting). It is recognized that the weighting of the criteria is the most subjective part of this work. A sensitivity analysis will be carried out to investigate the robustness of the decision to changes in scores and weights.

In order to determine a set of appropriate weights for the criteria, a workshop will be organized. It is expected that the weights that could be assigned to different criteria may vary between different populations. The results of this workshop will be compared to published data, and will help to guide the future work required for the development a decision-aiding system.

COAL FUEL CYCLE - IER

The air pollution-human impact pathways have to be completed taking into account the most important pollutants of the coal fuel cycle, which are sulphur dioxide, nitrogen oxides, particulate matter and secondary pollutant ozone. Main effects are expected from exposure to ozone. Modelling of ozone ambient air concentration requires the use of rather complex air chemistry models. An analysis of health effects from heavy metals will led to the identification of additional pathways that should be analyzed in the future.

To compare health effects of the coal and nuclear fuel cycles, appropriate health indicators must be developed describing occupational and public mortality and morbidity.

These indicators will be integrated into a comparative framework based on a multi-criteria analysis methodology. The results based on the application of a mult-criteria analysis-based methodology will be compared with those derived from the estimation of the external costs of the different fuel cycles. The specific advantages and differences of each concept will be discussed.

RIVM

The environmental effects of both the nuclear fuel cycle and the coal fuel cycle are very diverse, ranging from local effects on a short time scale (e.g. atmospheric pollution in power plant operation) to global effects on a long time scale (e.g. the greenhouse effect or an increase in background radiation levels). Following the identification of the important environmental impacts, useful indicators will be determined for use in the methodological framework for a comparative assessment. In this way the importance of the environmental impacts can be taken into account in the decision-making process.

During the development of the methodological framework for the comparison of risks associated with different energy systems, it was ascertained that large uncertainties can be associated with the assessment of both the human health and the environmental impacts. These uncertainties depend on the different steps in the impact assessment (e.g. source term, dispersion of the discharged substances, dose-effect relationships) and on the discharged substances considered (e.g. radionuclides, toxic and non-toxic substances). Especially in the area of dose-effect relationships, large uncertainties, as well as large differences in the uncertainties, exist depending on the extrapolation models used to determine the effects a low levels of exposure.

Therefore, it is recognised that some measure of uncertainty must be included in a well-balanced methodological framework for a comparative assessment of the human health and environmental impacts of energy systems. A rigorous uncertainty analysis will not be possible at this time, but at least a global assessment of the uncertainties associated with the framework will be performed with a crude estimation of the range of uncertainty in the associated steps. In this evaluation, a special effort will be made to compare the assessments of human health and environmental impacts to exposures from toxic substances to the impacts from radionuclides.

C13 Comparative assessment of exposure and risks.

Contract FI3P-CT920064c Studies related to the expression of the detriment associated with radiation exposure.

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Total Contribution by the Commission: 100 KECU
23 months 1/07/92 to 31/05/94

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Description of research work

INTRODUCTION

Recent years have seen a significant increase in the quantity and quality of predictive information available on the health effects associated with exposure to radiation. The latest reevaluation of cancer risk coefficients from the Japanese atomic bomb survivors, together with new and updated information from other human exposure and animal experiments (detailed in the most recent UNSCEAR[1], BEIR[2] and ICRP[3,4] reviews), has coincided with increased application of computing facilities within the health physics environment. This has meant that these data are more readily analysed and modelled than previously. In addition, there has been a gradual increase in the epidemiological data from medical exposures.

One major consequence of this has been that various national and international organisations have produced data on the distribution of health effects in populations following exposure to low levels of radiation. These analyses are often not directly comparable, since typically they are based on individual combinations of models, populations and assumptions on the use of the data.

Another area that has seen significant development is the definition of 'detriment'. Although ICRP, in Publication 60, have applied aspects of detriment, most notably in the revised definition of effective dose equivalent - 'effective dose' - it is clear that the quantification of radiation detriment is exceedingly complex. Whilst in their revised recommendations ICRP discuss a number of attributes of detriment that may be relevant to risk assessments, as yet there is neither a consensus on the units and quantities that should be assessed in terms of detriment, nor are there methodologies for aggregating the attributes. This is likely to become a focal point for radiological protection research as there will be a need to consider the adequacy of various measures of detriment for the definition of limits and other radiological protection standards, and for ALARA purposes.

Given the increasing range of health effects and interpretative data on radiation risk, as a first step, a tool that allows a rationalisation of these developments would plainly be beneficial. This would provide a common framework for developing the application of radiation detriment, and would act as a structure for analysing those aspects of detriment that require social judgements. ASQRAD (Assessment System for the Quantification of Radiation Detriment), a personal computer code being developed jointly by CEPN and NRPB, will be such a system. Both organisations have previously written software to analyse the significance of radiation exposure, and this project draws on this experience.

ASQRAD acts as a focus for CEPN and NRPBs' work on radiation risk and detriment, for the system necessitates both a full review of the present situation, and development of approaches for the quantification and application of detriment measures. What follows is a brief review of the objectives of the project and the progress to date.

OBJECTIVES

It is envisaged that ASQRAD (see Figure 1) will be applied in the development of policies where the radiation risk incurred or averted will be an important factor in decision-making. It will also be of use for ongoing research on radiation detriment; should find application in risk communication and training; and may also have a role in compensation issues. To this end, there are several features, that are deemed essential.

(i) *completeness:* ASQRAD will accommodate all the principal health effects models, in particular those that interpret A-bomb survivor and the main medical exposure data, and a comprehensive range of national population parameters;
(ii) *a capacity for simple up-dating and adaptation:* the user will be able to input other demographic statistics: moreover, the code is being constructed in such a way as to ensure that revised models can be readily installed;
(iii) *simplicity of use:* the system will be menu driven and contain default pathways for common applications; and will be supported by help and library facilities;
(iv) *sensitivity analysis* will be fully integrated; and
(v) *display and graphic facilities:* a range of these will be available.

OUTPUT

Fatal cancer risk and detriment estimates are obtained by combining 'fitted risk coefficients' with demographic details for a given exposure scenario using assumptions about risk projection. These health effects models have been developed either for specific organ sites or for groupings of sites, for example 'digestive'. The code will, however, provide the user with a choice of all the relevant risk projection models and coefficients for each tissue. The default data for hereditary effects will those recommended by ICRP, although the user will be able to test other estimates.

The demographic details required are a breakdown of the populations by age and sex cohorts in terms of numbers, death rates, and the cancer death rates for each site. In addition, two other sets of population statistics are used: the lethality fractions for each cancer type are needed for calculating the probability of non-fatal cancers; and the fertility rates by age and sex (the probability of an individual of given age and sex having a child during the forthcoming year) are used to calculate the probability of hereditary effects in progeny.

Using these data, it will be possible to calculate many measures of radiation detriment, and to perform extensive sensitivity analyses on the factors. The library functions will provide background information on the data, and guidance on the legitimacy and consistency of these combinations.

Measures of detriment will be calculated according to the following scenarios:
(i) individual whole-body exposure: acute or extended (at variable annual dose), to an individual of given age or sex, or assuming an average for both sexes;
(ii) population whole-body exposure: a single low dose and dose-rate exposure to a population (either default or user input) of mixed age and sex; and
(iii) non-uniform exposure: assuming doses to a single organ or a combination of organs

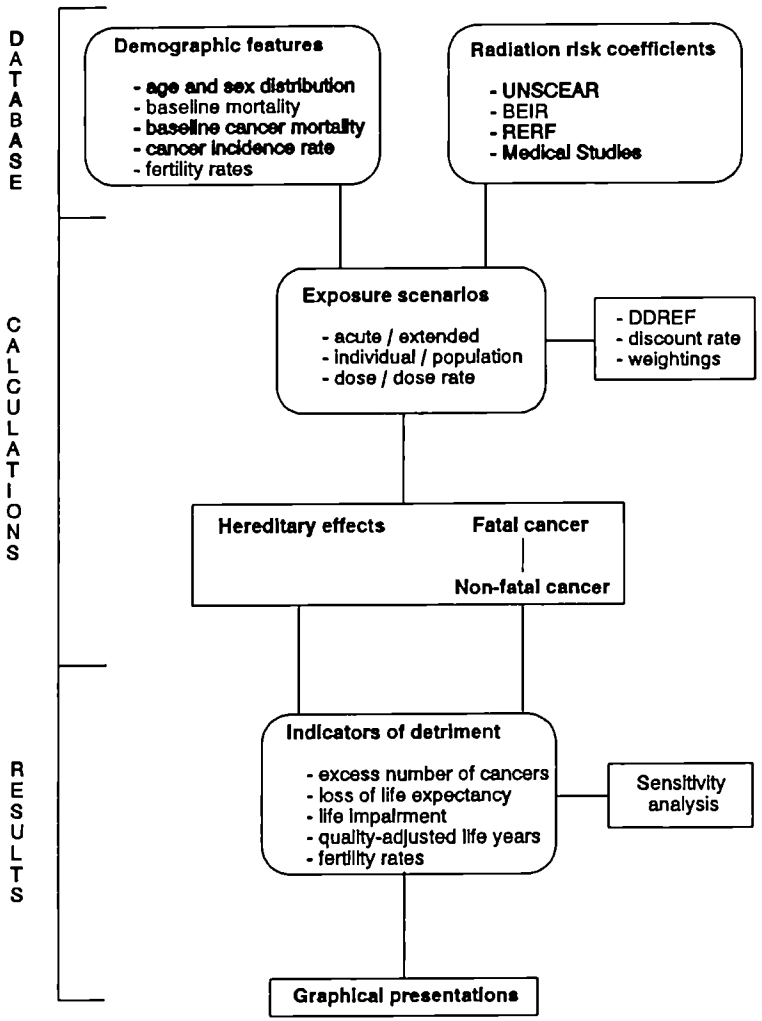

Figure 1. Outline of ASQRAD

The excess risk will also be assessed in terms of the excess lifetime risk (this takes into account the probability that individuals dying from radiation-induced cancer deaths may in any case have died of cancer from other causes, by subtracting this probability from the attributable cancer rate) as used by BEIR V[4], or the lifetime risk of exposure-induced death, as used by UNSCEAR[1], and in most other analyses.

For each of these situations, probability of cancer and hereditary effects, and associated loss of life expectancy (years of life lost, or YOLL) can be calculated directly. Moreover, additional measures of detriment are proposed to take into account the severity of these effects in terms of morbidity and lost quality of life. Thus, some of the aggregated measures of detriment discussed in ICRP publication 60 will be provided; and two other expressions are introduced - years of life impaired (YOLI), and the quality-adjusted life year (QALY). The library facility will provide information on any other measures of detriment that may be of interest in addition to those calculated, and also information on the variability and uncertainty of the data.

PROGRESS TO DATE

Experience has shown that unless systems such as ASQRAD are comprehensively planned prior to the commencement of coding, the program may be generated in a piecemeal fashion, becoming inefficient and incomplete. For this reason, the development of ASQRAD has followed three distinct phases:

(i) Planning paper.

This was prepared to give an overview of the range of models, calculations, end-points, and output required for the code; as well as the features that the various users would expect in the system. In particular, this noted all the data requirements, and the connections between these, highlighting those areas where work would be necessary in order to establish full data sets. This planning paper was completed in early 1991, and forms the basis of all the subsequent work.

(ii) Development and application of the database.

The development of the database is part of an ongoing programme of work, and has contributed towards the detailed planning of the software, particularly in relation to the health effects models. There are two components to the development of a rationalised set of health effects models. The first of these requires a clear elucidation of the individual sets of data and assumptions used: for example; in each of these studies, for a given organ how are the risk coefficients derived? what form does the projection model take? and what demographic details have been used? The present database contains several models and populations.

The second relates to the calculations themselves - the application of the risk projection models. A comparison study was performed using the present NRPB and CEPN codes to identify any differences in the assumptions and methods employed. For instance, the way in which survival probability of the over-85 age group is modelled differed, but can be very important in estimates of radiation detriment.

(iii) Detailed planning and preliminary coding.

The system is being coded using the APL language. The basic calculations mentioned above under point (ii), were linked by an initial set of menus, thus providing a framework around which the rest of the code, the graphics, and the library and help facilities can be built. This was designated ASQRAD 0.1, and was subsequently replaced by ASQRAD 0.2 towards the end of 1991. The rest of the system will be constructed around this preliminary code.

CURRENT WORK

In the context of ongoing research into the application of radiological detriment, the developments necessary for ASQRAD can be summarised under five areas:

(1) data requirements;
(2) sensitivity and uncertainty analysis;
(3) coding;
(4) library/help facilities; and
(5) output.

(1) Data requirements

Data sets for the elements of ASQRAD are being compiled. Most of these are straightforward, if time-consuming, but one substantial topic has to be finalised: the rationalisation of measures of radiation detriment.

(2) Sensitivity and uncertainty analysis

It is only the simplest approach to uncertainty analysis that is feasible, and this is to carry out calculations on ranges of data rather than single points. The appropriate data for these ranges are being formulated, as is the most efficient yet user-friendly way of including it in the code. On-line sensitivity analysis will be provided with the user being advised on the options open to him or her.

(3) Coding

ASQRAD is being coded using APL code, a vectorial programming language that is particularly suitable to scientific calculations and large data-set manipulations.

(4) Library/help facilities

Throughout the system, the user will have the option of help facilities: if the user is being asked to make a choice or feed in data, the help facility will provide relevant details. The library will operate at a deeper level, giving background information on the components and the calculations, and advising on the applicability of whatever choices are available. Setting up these facilities within the code is relatively simple. However, preparing the detailed text is time-consuming, requiring considerable literature review.

(5) Output

In addition to tabular output of both data and associated results, basic graphics will be available as part of the system. A primary set is being defined using the inbuilt graphic capabilities of the APL language. These should be adequate for most uses, but a further option available is being introduced that will allow export of the data/results to standard worksheet/plotting packages.

SUMMARY

ASQRAD is now developed such that its structure is fully planned, and a preliminary version including all the major calculations, linked by a basic set of menus, has been coded. The second stage of the contract is building on this foundation.

References

1. UNSCEAR. Sources and Effects of Ionising Radiation. 1988 report to the General Assembly, with annexes. New York, UN (1988)

2. BEIR V. Health Effects of Exposure to Low Levels of Ionizing Radiation. National Academy of Sciences, National Research Council, Washington DC (1990)

3. ICRP. 1990 Recommendations of the International Commission on Radiological Protection. ICRP Publication 60, Ann ICRP 21, Nos 1-3 (1991)

4. ICRP. Risks Associated with Ionizing Radiation. ICRP Publication 61. Ann ICRP 22, No 1 (1991)

CONTRIBUTION OF THE NRPB

NRPB contribution to the present phase of this project is best detailed with reference to four main areas of commitment. These are:

(a) collation of data sets;
(b) development and incorporation of measures of detriment;
(c) provision of sensitivity and uncertainty analyses; and
(d) preparation of the on-line help and library facility.

(a) Collation of data sets

Complete data sets for each of the components of the code have to be finalised. This work will be undertaken jointly by NRPB and CEPN, although there will be separate individual tasks.

(b) Measures of detriment

NRPB will devote considerable effort to the interpretation, quantification, and application of indicators of detriment. This work will measures of detriment that are calculated as outputs in ASQRAD.

(c) Sensitivity and Uncertainty analyses

NRPB will be jointly responsible with CEPN for planning the detailed incorporation of sensitivity and limited uncertainty analysis into the code. The two organisations will construct the necessary database of value ranges required from the work under (a).

(d) Help and library facility

Ultimate responsibility for the library and help facilities resides with NRPB. Parallel to the coding at CEPN, NRPB will develop a shallow help facility that will provide basic guidance to the user, and a deeper library that explains all the options provided by the database together with detailed background information.

There will be close collaboration with CEPN in all areas of the work with each organisation reviewing the other's contribution.

CONTRIBUTION OF CEPN

The contribution of CEPN to the present phase of this project will focus on three areas of commitment. These are:

(a) review of health effects models and collation of demographic data;
(b) development of the system coding; and
(c) development of new software facilities.

(a) Review of health effects models and collation of demographic data

ASQRAD currently includes information from the main health effects models published by international and national organisations. It is planned that the various risk coefficients derived from epidemiological studies related to medical exposure be reviewed in order to determine if further data can be incorporated as default options. A first analysis has been performed on studies of radiation-induced cancer incidence. The next phase will include other cancer sites, for example lung and red bone marrow.

(b) Development of the system coding

The initial coding is well advanced with all the fundamental calculations introduced for the risk of fatal cancer, non-fatal cancer, and hereditary effects. The principal improvements to be introduced are those to the user interface, and the integration of help and library facilities. Furthermore, the graphics have to be defined, and an export facility coded.

(c) Development of new software facilities

Sensitivity and uncertainty analysis options will be developed with NRPB. The former will consist of options on the parameters and assumptions chosen, or introduced, by the user. The uncertainty analysis will be limited to ranges of values.

There will be close collaboration with NRPB in all areas of the work with each organisation reviewing the other's contribution.

C14 Epidemiological studies in human populations.

Contract FI3P-CT920047 Investigation of late effects in humans after artificial irradiation (Thorotrast-patients) - Follow-up study.

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Total Contribution by the Commission: 170 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Background

Thorotrast was the trade-name of an medical x-ray contrast medium used from about 1930 to 1950. The predominant form of application was an intraarterial injection, especially for cerebral angiography. It consists of a 25% colloidal suspension of thoriumdioxide (^{232}Th). After intravascular injection, ThO_2 aggregates are stored lifelong in the reticoloendothelial system (RES). According to KAUL and NOFFZ (1978) the distribution of Thorium-232 in a standard patient can be estimated as follows: liver 59%, spleen 29%, red bone marrow 9%, calcified bone 2%, lungs 0.7% and kidneys 0.1%.

^{232}Th has a natural radioactivity; the half life is more than 10^{10} years and 95% of the radiation is alpha-particels. KAUL and NOFFZ (1978) calculated the mean values of the annual radiation dose to the organs of the RES. A mean intravascular injection of 25 ml Thorotrast in a 70 kg person causes the following absorbed dose rates: liver 25 cGy/year, spleen 70 cGy/year, bone marrow 9 cGy/year, endothelial layer and bone 16 cGy/year, kidneys 0.4 cGy/year. The radiation dose to the lung tissue is mainly caused by the daughter product ^{220}Rn which is exhaled by the breath. The dose rate to the endothelial layer mainly results from 1) the alpha-radiation originating from ThO_2 particles in the adjacent bone marrow and 2) the radium-224 translocated from the deposits in the RES in the bone surface.

Aims:

- to uncover the late effects of chronic alpha-irradiation caused by incorporated colloidal thoriumdioxide with epidemiological observation and clinical and biophysical examination of the patients
- to compare the results with those of the general population as well as of different clinical control groups
- to reevaluate the dose rate to the depositing organs with special regard to bone marrow and lung as well as to the non-depositing organs
- to assess a relationship between late effects and radiation dose
- to study the unspecific life shortening effect especially in the still living patients injected with low Thorotrast volume at young age
- to follow-up the off-spring of Thorotrast patients to calculate the risk caused by irradiation of the germ cells of the male Thorotrast patients and the fetus of female Thorotrast patients
- to examine genetic alterations in Thorotrast-induced liver tumors

Expected results

From the results of these studies we expect contributions to high LET risk estimates for liver cancer, non-lymphocytic leukaemia, plasmacytoma, bone sarcoma, and lung cancer caused by ^{220}Radon, a daughter product of thorium.

Furthermore, we expect information on the late effects in the so-called non-depositing organs, which are exposed however to much higher doses compared to the natural external and internal irradiation dose. These organs contain a minute amount of Thorotrast and additionally are chronically exposed by the daughter products circulating in the blood.

We expect additional information on the life shortening effect by chronic internal alpha-irradiation by analyzing the diseases leading to death.

The Danish und the German Thorotrast studies with more than 3,000 well documented and biophysically examined patients present the largest number of Thorotrast patients in the world. A combined analysis of these patients who are very similar in life style and socioeconomical conditions contribute to the statistical power of the risk calculations.

These results will contribute fundamentally to the knowledge of human radiation biology and radiation protection.

Program of work

Clinical study:

- Clinical, biochemical and radiological examinations of the Thorotrast patients and the control group
- Biophysical examinations to calculate the tissue dose due to the thoriumdioxide deposits and their radioactive daughter products
- Identification of the causes of death of Thorotrast patients and members of the control group
- Ophthalmological examinations to evaluate the cataract response in Thorotrast patients caused by the daughter product ^{224}Ra

Reevaluation of the dosimetric calculations for selected organs:

- Lung dosimetry based on the recent lung model. The dose to the target cells will be calculated from three sources of irradiation: Thorotrast particles stored in the interstitial space, ^{220}Rn and its alph-emitting daughters in the capillaries and in the airways (exhalation).
- Dosimetry of the non-depositing organs:
  autopsy material of patients of the study will be examined by different methods (see U.K. study).
  Combining these results, the dose to the non-depositing organs will be calculated.
- Dosimetry of the bone marrow:
  assessment of the distribution of Thorotrast in the yellow and red bone marrow with regard to individual bones.

- Animal experiments to assess spatial distribution and the time-dependent distribution of Thorotrast in the bone marrow in comparison with autopsy results of human beings.

Molecular biological investigations:

- Detection of mutations and deletions of the tumor suppressor gene p53 in liver tumors of Thorotrast patients
- Identification of hepatitis B virus in liver tumors of Thorotrast patients

Late effects in the off-spring of Thorotrast patients:

- Epidemiological observation of the children of male and female Thorotrast patients and comparison to the control group or the normal population
- Estimation of the dose to the germ cells of male and female Thorotrast patients and to the fetus during pregnancy

Statistical evaluation of the study:

- Separate analysis of the epidemiological data of the Danish and German Throtrast study
- The German Thorotrast study until now compares its results to a special control group. The patients of the Thorotrast and the control group now will be compared to the results of the cancer registry of the state of Saarland.
- Combined evaluation of special diseases in the Danish and the German Thorotrast study

The German Thorotrast Study

The German Thorotrast study includes 2326 patients who had a cerebral arteriography (70%) or arteriography of the upper and lower limbs (30%) with Thorotrast and 1890 patients of the control group. The control group was selected from patients who were treated at the same time in the same hospitals. Up to now 2222 Thorotrast patients and 1520 patients of the control group have died.

The living patients (474 in both groups together) are followed up every two years by out-patient examination including biophysical measurements, radiological and clinical examinations.

Results

The recent status of the study is summarized in Table I. The results demonstrate a clear excess rate of liver tumors, non-lymphocytic leukaemias (NLL) and liver cirrhosis.

An excess rate is very probable for carcinomas of the extrahepatic bile ducts, pancreas and esophagus, Non Hodgkin's lymphoma, plasmacytoma, mesothelioma and bone sarcomas; but one has to consider that the number of the still living control patients is twice the number of the still living Thorotrast patients (van Kaick et al., 1991).

Thorotrast patients die earlier than patients of the control group: Even after excluding the influence of life-reducing diseases due from radiation like liver cancer, cirrhosis or leukaemia the life expectancy of these patients is reduced compared to the controls. This phenomenon depends on the amount of Thorotrast injected. Therefore we can state that there is a Thorotrast dependent influence on age at death.

Table I

German Thorotrast Study - Diseases with Excess Rates

Status '92	Thorotrast	Control
Cause of Death	n=2326	n=1890
Liver cancer	436*[+5](18.74%)	2 (0.11%)
Liver cirrhosis	190 [+173](15.60%)	47 [+2] (2.10%)
Non-lymphatic leukaemia	36 [+3] (1.68%)	5 (0.26%)
Bone marrow failure	30 (1.29%)	5 [1] (0.32%)
Ca ext. bil. ducts and gall-bladder	28 [+3] (1.16%)	6 (0.32%)
Ca. pancreas	18 (0.77%)	5 (0.26%)
Ca. esophagus	7 [+1] (0.34%)	2 (0.11%)
Ca. larynx	6 [+1] (0.30%)	1 [+1] (0.11%)
Non Hodgkin lymphoma	13 [+2] (0.64%)	3 (0.16%)
Bone sarcoma	4 [+1] (0.21%)	1 (0.05%)
Plasmacytoma	7 [+2] (0.38%)	1 (0.05%)
Mal. Mesothelioma		
Pleural	5 [+1] (0.25%)	0
Peritoneal	4 [+1] (0.21%)	0

[] Additional cases with another disease leading to death
* 5 patients with combined carcinoma and sarcoma
() Inclusive additional cases related to n

Objectives for 1993

The working program will be continued according to the recommendations of the coordinating committee.

- Regular correspondence with about 470 patients of the Thorotrast and control group as well with the respective family physicians
- Out-patient reexaminations of Thorotrast carriers and patients of the control group at two year intervals
- Calculation of the dose to the germs cells of the male Thorotrast patients and to the fetus of female Thorotrast patients during pregnancy and to follow-up the off-springs of Thorotrast patients
- Combined analysis of the leukaemia cases in the Danish and German Thorotrast patients

- Cooperation between the British and the German group in analysing bone samples of Thorotrast patients
- Molecular investigations on liver tumors and comparison of the German and Danish results.

References

Kaick G van, Wesch H, Lührs H, Liebermann D, Kaul A (199") Neoplastic diseases induced by chronic alpha-irradiation - Epidemiological, biophysical and clinical results of the German Thorotrast Study. J Rad Res, Suppl 2, 32:20-33

Spiethoff A, Wesch H, Wegener K, Klimisch HJ (1992) The combined and separate action of neutron radiation and zirconium dioxyde on the liver of rats. Health Physics 63:111-118

Spiethoff A, Wesch H, Wegener K, Klimisch HJ (1992) The effects of Thorotrast and quartz on the induction of lung tumors in rats.
Health Physics 63:101-110

Research Program On Thorotrast in the United Kingdom

Intravascular injection of the radiographic contrast medium "Thorotrast" produced a life-time burden of alpha irradiation to those organs into which it deposited. The alpha exposure to bone marrow leads to an increase of non-lymphocytic-leukaemia and lethal myelodysplastic syndrome. The high LET risk estimates of ICRP cannot be confirmed by these results.

The U.K. group will make a re-evaluation of the effective dose to bone marrow and skeleton in Thorotrast patients. The studies undertaken will look at the microdistribution of Thorotrast in both yellow and red bone marrow in both human and primate skeletal tissues. The primate study will enable the determination of whether the Thorotrast changes its distribution with time thus producing temporal changes in the distribution of dose.

The emphasis on the studies undertaken to date have been on the Thorotrast distribution in a human case USUR-Th1001, and in monkeys that were injected with Thorotrast. Examination of these skeletal tissues has been made using the following methods.

- Secondary ionisation mass spectrometry (SIMS)
- Scanning electron microscopy with microprobe analysis
- Autoradiography using solid state nuclear track detectors
- X-ray fluorescence analysis (XRF)
- X-ray fluorescence using synchrotron radiation (SXRF)
- Particle induced X-ray emission spectrometry (PIXE)
- Histological and morphological demonstration technique

All of the above have been used to determine the distribution pattern of Thorotrast in the tissue. These results are being used to assess the fraction of tissue potentially exposed

to alpha-radiation and the degree of heterogeneity in the distribution of dose across the tissue. These factors may be important when assessing any dose/response date provided by the epidemiological studies.

We have now also been provided with skeletal samples from Germany and skeletal and soft tissues from Japan.

At the present time comparison is being made between the results obtained from the USUR Th1001 case and monkey results for post mortem tissues obtained 7 days post injection.

Samples are being processed from a monkey sacrificed 4 years post injection and some data from these tissues will be ready towards the end of February.

The Danish Thorotrast Study

p53-GENE Mutations in Alpha-Particles induced hepatic neoplasms diagnosed in Danish patients injected with Thorotrast

Approximately 1000 Danish residents were injected with Thorotrast, a 20-25% colloidal solution of thoriumdioxide, during 1935-47. Sixty percent of the injected substance will accumulate and be retained in the liver, where the decay of the principal isotope, ^{232}Th, exposes the individual to chronic alpha-radiation. The incidence of primary liver tumors in these patients is increased more than 100-fold (Andersson M, Storm HH. J Natl Cancer Inst 84: 1318-1325, 1992) and experimental studies strongly imply radiation to be causative. It has been suggested that a specific point mutation of the p53-gene is essential in the pathogenesis of primary liver tumors induced by aflatoxins. In order to examine whether such alterations are also implicated in the development of radiation-induced liver tumors, archival formaline fixed and paraffine embedded tissues from 38 histologically verified liver tumors are currently screened for p53 mutations by amplification by PCR of exons 5-8, denaturing gradient gel electrophoresis and sequencing of mutated regions.

Related Research on Thorotrast

Offspring of both male and female Thorotrast injected patients have been identified and followed. The fertility of female patients and the incidence of cancer and the mortality will be related to that of the general population. For the offspring of male patients, the cancer incidence will also be related to the estimated paternal testicular radiation dose (in cooperation with the Harwell Laboratory, UK).

The incidence of leukaemia and other related haematologic disorders among the Thorotrast patients has been assessed and related to estimated radiation dose to the bone marrow (Andersson M, Carstensen B, Visfeldt J. Radiation Research 1993, in press). In cooperation with DKFZ a joint analysis of the leukaemia incidence is being planned.

C14 Epidemiological studies in human populations.

Contract FI3P-CT920056 The risk assessment of indoor radon exposure.

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Description of research work

The relation between radon exposure and lung cancer incidence is well documented in numerous epidemiological studies of uranium miners and other exposed underground workers. Projection of these results to the house environment suggests that indoor radon may be an appreciable cause of lung cancer. These extrapolations however suffer of many uncertainties, due to differences between the mining and indoor environment, the circumstances and temporal patterns of exposure, and the biological characteristics of miners and members of the public. Therefore epidemiological studies conducted in the population are necessary to clarify the lung risk of indoor radon.

At the moment, correlation studies are available from several countries, relating the incidence of lung cancer to radon exposure based on geographical areas. Such descriptive studies are of limited value due to the wide range of radon exposure within a region and because of the difficulty of taking smoking habits into account adequately.

Case-control studies are a better approach. Persons with lung cancer are included and also comparable persons without lung cancer characterised with respect to (past) radon exposure and other causal factors such as smoking and occupational exposure. Several preliminary studies of this type have been reported with a variety of outcomes. However in the early studies radon exposure and cigarette smoking were not always accurately assessed and additional high quality studies are needed.

The project consists of three major studies about the health risk of radon :

- Radon and lung cancer in the Ardennes-Eifel region;
- Radon dans les habitations de Bretagne et du Massif Central et risque de cancer du poumon;
- Investigation on the relationship between lung cancer and radon in houses.

The ultimate objective of interest is the quantification of the risk of indoor radon exposure with sufficient precision to be informative.

The Ardennes-Eifel study is a hospital based case-control study, situated in a geological distinct area covering parts of Belgium, France, Germany and Luxemburg. Out of national and regional surveys it became clear that in this region the range of indoor radon exposures is quite large and that an important fraction of the population is living in high radon houses.

It is a multi-centre study with participants in France (Centre d'Energie Ato-mique), Germany (University of Wuppertal), Luxemburg (Division de la Radioprotection) and Belgium (University of Gent), and with scientific support from Britain (Imperial Cancer Research Fund). In this study a common protocol is used, dealing with general items such as the selection of cases and controls, the residential criteria for inclusion in the study and the specifications of the radon measurements. An appropriate hospital control

group was derived as well as a list of ineligible diseases strongly related to tobacco. A common core questionnaire is adopted, including items such as residential history, occupational history, exposure to passive smoke (for non-smokers and occasional smokers only) and educational attainment of the partner.

The study is aimed to arrive at complete data of some 1200 cases and 3600 controls over a period of 5 years. Based upon local population distribution patterns and geological extent of the region a total amount of 1800 subjects is planned to be recruited in Belgian and German hospitals, while some 600 will be delivered by French and Luxemburg clinics. Only subjects who are currently living within the defined study area and who lived there for at least 25 out of the last 35 years are included. The radon history is reconstructed through 6 months measurements in the living and bedroom of the different dwellings. The comparability of the detectors and associated procedures used in the different centres is investigated through yearly quality control exercises.

The cases are selected in the departments of pneumology, bronchoscopy and thoraxsurgery. Only histologically confirmed cases are included in the analysis. Patients aged 75 years or over on their first visit to hospital are excluded. In each country a local reference pathologist will review the pathological material. The radon inter comparisons are organised by the Belgian participant (University Gent), who acts as co-ordinator for this project.

The common statistical analysis will be performed at Wuppertal. To achieve this a common coding schedule is compiled. According to this scheme every centre is submitting data periodically to the co-ordinator in Gent, as a standard ascii-file. Here a data-filter is applied in order to transfer all data in an appropriate form to Wuppertal.

The treatment of the data will be done in close collaboration with the epidemiological research team from Oxford. This unit is also taking part in the setting-up of the study-protocol.

The estimation of missing radon values is one off the secondary challengers of this study. Geographical location, geological data, soil and building characteristics are often the only available information to work with. The predictive ability of this information is rather limited. Therefore the suitability of Po-210 as tracer for past radon exposures (project "Retrospective assessment of Radon Exposure from Long-Lived Decay Products"), will be thoroughly investigated. The fact that after all only radon exposures of the past are relevant for lung cancer risk experienced today, opens perspectives for such a retro-spective technique.

Whenever unacceptable high radon levels are encountered in this study, control measurements will be performed and the responsibles for mitigation actions (project "Radon sources, models and countermeasures") will be contacted for advice and actions to take.

The set-up of the case-control study organised in Brittany by the C.E.A., in close collaboration with the university of Brest and INSERM, is in complete conformity with that of the Ardennes-Eifel study. It is aiming at collecting complete data for some 600 cases and 1200 controls. The data from these studies will also be included in the final statistical analysis.

In view of carrying out a pooled analysis on a large European scale, a feasibility study will be carried out from the British Centre, participating also in the Ardennes-Eifel study. It will consist partly of discussions with the principal investigators of ongoing and completed studies in Europe and partly of a meeting of the possible collaborators in the pooling exercise, to be held in Oxford at the end of 1993. At this meeting some of difficulties of achieving a common data set suitable for a pooled analysis will be discussed. Conclusions will be drawn as to the feasibility of carrying out a scientific valid pooled analysis of European studies.

The British Centre involved in the Ardennes-Eifel project is also, together with NRPB, the principal responsible in the epidemiological study about radon in Cornwall-Devon. In this project, with a protocol very similar to that of the Ardennes-Eifel, a total of 600 confirmed cases of lung cancer, for whom a full radon history can be obtained, are aimed to be included. This necessitates interviewing some 1350 cases of suspected lung cancer. The study is based in 5 hospitals. First interviews were carried out in mid-1988. Case enrolment should last in each centre for about three and a half years and should be complete by approximately the end of '92. Control interviews and radon measurements should last up to the end of '93.

Contribution of the Laboratory of Nuclear Physics - University Gent - Belgium

RADON AND LUNG CANCER IN THE BELGIAN ARDENNES

The Laboratory of Nuclear Physics of the University of Gent is the Belgian participant and co-ordinator of the European multi-centre study, called Ardennes-Eifel project. The study area consists of the 4 provinces Hainaut, Liège, Luxembourg and Namur, where high radon values have been registered and where the range in radon exposures is much greater than in the remaining part of Belgium.

Cases as well as controls are recruited in six hospitals. In each centre interviewing should last for about 4 years.

The protocol used is the common protocol worked-out and agreed-upon by all partners during the preparative phase of the European epidemiological study on radon and lung cancer.

All radon related work is accounted for by the radon research group of the Lab of Nuclear Physics.

Current inhabitants of houses previously occupied by subjects of the study are contacted with the help of the local authorities and through an application to collaborate of the Ministry of Public Health.

During the period 1992-1994, 1200 patients (cases + controls) are planned to be interviewed and most of the related radon measurements in present as well as previous houses will be performed.

As co-ordinator of the European study, the Nuclear Physics Laboratory in Gent is also responsible for collecting the data of all participating countries and for transferring them in appropriate form to the team of Wuppertal for statistical analysis .

Therefore a draft version of a common coding-schedule has been prepared in close collaboration with the teams of Oxford and Wuppertal and a data-filter is being constructed to facilitate the data-entry for common analysis.

Special attention is also be paid to the problem of missing radon data. As experts themselves have pointed the limited predictive ability of geological and soil information, the suitability of techniques (the use of Po-210) for estimating past radon exposures is tested during this study. As co-ordinator the Laboratory of Nuclear Physics is also responsible for organising quality control exercises between the radon devices used by all participants.

Contribution of Institut de Protection et de Sûreté Nucléaire - CEA - France

EPIDEMIOLOGICAL CASE-CONTROL STUDY ON THE RISK OF LUNG CANCER FROM DOMESTIC EXPOSURE TO RADON IN FRANCE

This study, coordinated by the ISPN, is in progress in the following areas of France:

- The Ardennes;

- The 6 provinces of the Brittany-Vendée area; namely Finistère, Morbihan, Loire-Atlantique, Vendée, Côte d'Armor and Ille et Vilaine.

It is part of the international Ardennes-Eifel study through the intermediary of the Société Ardenaise de Cancérologie at Charleville Mézières.

In Britanny-Vendée the Pneumology Services of the following hospitals have agreed to participate : Brest, Nantes, Rennes, St Brieuc, Vannes, Quimper, St Nazaire and Cholet. Their geographical distribution ensures a representative recruitment of lung cancers in the area under consideration.

The protocol for this study conforms in every respect to the protocol defined at the European level during the feasibility study. The study began in the Ardennes at the end of 1990 and in the Britanny in June 1991.

The same protocol will also be adapted to the Massif Central area where contacts with hospital doctors are being made.

As the risk of lung cancer linked to domestic exposure is probably low, a minimum number of 750 cases (150 in the Ardennes and 600 in Britanny-Vendée) and double this number of controls will be necessary in order to evaluate that risk. The results of this study will form part of an analysis at European level, bringing together for the Ardennes-Eifel, more than 1200 cases and controls. In France, this study is scheduled to last 5 years. It is hoped that in the period 1992-1994, about $^2/_3$ of the planned questionnaires, as well as most of the radon measurements in the corresponding homes will be realised.

In order to carry out this study, it is necessary to recruit about 15 interviewers. In Britanny-Vendée the logistics of this study are being achieved thanks to the collaboration of the Laboratory of Aerosols and Natural Radioactivity of the University of Sciences of Brest, led by Professor G. Tymen.

Contribution of the Department of Labor Safety and Environmental Medicine - Bergische Universität Wuppertal - Germany

LUNG CANCER AND RADON IN THE EIFEL REGION

The Department of Labour Safety and Environmental Medicine at the University of Wuppertal is acting as the German participant in this European Study. The responsibility of the study group includes the selection of cases and controls in the 5 participating hospitals, the distribution and collection of the radon detectors and the preparation of the regional data base of the German study area. The study region was defined as the three administrative districts of Trier, Koblenz and Saarbrücken.

In close collaboration with the co-ordinating team of Gent and the consulting team of Oxford, data checks, co-ordination of the common data base, statistical analysis namely in-between-analysis, common proposals of separate analysis and overall-analysis will be done in Wuppertal. Therefore first programs to do descriptive statistical analysis on the common data bases are evaluated with the software SAS.

The practical experience of co-ordinating the common data base should support international activities of a feasibility study into the possibility of carrying out a pooled analysis of European environmental radon studies.

For this reason pooling and data-inter comparison exercises will be supported.

All radon measurements are performed by the group of Prof. G. Keller of the university of Homburg

Contribution of the Radiation Protection Service - Ministry of Pubic Health - Luxemburg

During the period 1992 - 1993, the epidemiological survey of lung cancer cases and controls of the same age and sex, living in the Grand Duché du Luxembourg for a minimum of at least 30 years, is being continued. The age limit for those included in the survey is 75 years old.

The study will, from now on, cover the whole of the Grand Duché du Luxembourg which forms a clear geographical unit. In this way the numbers of cases to be included in the study and which meet the established criteria will be increased. Only cases which have been confirmed pathologically will be included.

As agreed by the institutes of the four countries taking part in the study, the choice of controls will be made partly at hospital level and partly from the general public.

Controls included at the hospital level are identified by the relevant department of general radiology.

The controls drawn from the general public will be chosen at random, using census lists available from local authorities. Contact with the controls will be initiated by officers of local fire-brigades in order to limit refusals as much as possible.

The basis of the survey will be the questionnaire elaborated by the institutes of the four participating countries.

Measurements of radon in the homes of the cases and the controls will be carried out according to the protocol agreed by the four institutes.

The radon level in all houses occupied for the last 30 years by cases and controls will be measured. Each measurement will cover 6 months and 2 dosimeters will be placed in each house. The method of nuclear track detectors is being used in the present study.

Contribution of the Cancer Epidemiology Unit - Imperial Cancer Research Fund - England

INVESTIGATION OF THE RELATIONSHIP BETWEEN LUNG CANCER AND RADON IN HOUSES

The present study is being conducted in Devon and Cornwall, where surveys have shown that there is a wide variation in radon concentrations, including a substantial proportion of houses with relatively high concentrations of radon and also areas with low concentrations. A standard questionnaire is used to obtain detailed residential, smoking and occupational histories, and people in the following categories are interviewed :

i) patients under 75 years of age with a presumptive diagnosis of lung cancer,

ii) an age/sex matched sample of patients admitted to the same hospitals with presumptive diagnoses of a wide variety of conditions other than those strongly related tot smoking,

iii) an age/sex matched sample of community controls chosen at random from Family Practitioner Committee lists for Devon and Cornwall.

Patients with a presumptive diagnosis of lung cancer rather than only those with a confirmed diagnosis are interviewed for two reasons. Firstly, as survival among lung cancer patients remains poor, it maximises the change that the interview can be completed and all the information necessary for the study obtained before the patient becomes too ill or dies. Secondly, when diagnoses are checked after discharge, the group of patients whose final diagnosis is not, after all, lung cancer provides an exceptionally valuable addition to the control group free of any possible interviewing bias that might be associated with knowledge of the nature of the disease.

The residential histories obtained from the interviews enable a list of houses to be prepared in which measurements of radon concentrations are subsequently made, in order to build up a picture of the radiation exposure of each person in the study. Two measurements are made in each house for a period of six months, and annual exposure estimated from these. Important alterations to houses are noted whenever possible, and appropriate corrections to the measured concentrations will be made.

About half the people interviewed have migrated recently into the counties in which the survey is being conducted, and the inclusion of such people would weaken the power of

the study, as the variation in radon levels would on average be less in other areas. Individuals are therefore included in the study if they have lived in Devon and Cornwall for at least 20 years during the period starting 35 years and ending 5 years before the interview.

From the data obtained it will be possible to estimate relative risks for different degrees of exposure, standardised for sex, age, smoking habits, and occupation.

During the period 92-93 the scientific support to the Ardennes-Eifel project will continue as in the past and a feasibility study into the possibility of carrying out a pooled analysis of European environmental radon studies will begin.

Contribution of the National Radiological Protection Board - England

INVESTIGATION OF THE RELATIONSHIP BETWEEN LUNG CANCER AND RADON IN HOUSES

The National Radiological Protection Board (NRPB) is collaborating with The Imperial Cancer Research Fund (ICRF) on a study of radon in homes in southwest England and the incidence of lung cancer.

NRPB has several roles in this study. Its principal role is to provide measurements of radon in homes. This is done by issuing passive radon detectors for use in the present and past homes of cases and controls. In order to estimate the long-term average radon concentration in the homes, two detectors are issued to each home for a six-month period, one for the living room and one for a bedroom. The concentration of radon in each home is calculated by NRPB on the basis of the results and a knowledge of the dates of placement and removal of detectors. This result is passed to ICRF, and a seasonal correction is applied on the basis of NRPB data on average seasonal variations of radon in homes.

For measurements in the present homes of subjects in the study, NRPB supplies detectors to ICRF interviewers for placement. For measurements in past homes of subjects, ICRF passes a list of addresses to NRPB, which makes direct contact with householders by post and carries out radon measurements with their agreement. In case of difficulty, the details are passed to ICRF interviewers to make personal contact with householders.

NRPB supplies facilities for a steering group for this study, which meets twice yearly.

C14 Epidemiological studies in human populations.

Contract FI3P-CT920062 European childhood leukaemia/lymphoma incidence study.

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Description of research work

BACKGROUND

Following the accident at the Chernobyl nuclear power plant on 26 April 1986, radioactive materials were deposited over large areas of Europe. There were three successive 'plumes' of material affecting (1) the eastern USSR, Poland and Sweden, (2) central Europe, especially Austria, Bavaria, north Italy and part of Switzerland, and finally (3) Romania and Bulgaria. Most exposure was to ^{151}I, ^{134}Cs and ^{137}Cs, and was generally of rather low magnitude. Exposure to humans was both external (mainly from ground deposition) and internal (from ingestion of contaminated food). Estimated average exposures require rather complex models and differ according to the methods used. UNSCEAR (1988) has produced estimates for all national populations in Europe (and by sub-region within some countries). Nationally, the highest average exposures outside the USSR were in Bulgaria (760 μSv) and the lowest in Portugal (2 μSv). These figures can be compared with the average (worldwide) exposure from natural sources of 2400 μSv per person, although there is considerable geographical variation.

Outside the immediate vicinity of the accident, the predicted health effects due to radiation are rather small. Nevertheless, these health effects should be monitored for several reasons. Firstly, it is a matter of great public concern, and already there are reports of clusters of leukaemia, excess infant mortality and excess premature births among malformed children in sub-national areas with higher than average exposures. Investigation of apparent clusters is greatly facilitated by a large scale systematic study. Secondly, it is possible that either dose estimates or the models used in predicting cancer risk are in error, and the excess will be higher than expected.

Childhood leukaemia is the most logical choice of adverse health effect for monitoring. Radiation-induced leukaemias appear early (2-10 years) after exposure, and provide the largest excess incidence of any cancer. Background incidence of leukaemia is relatively constant in Europe. Finally, a fairly comprehensive monitoring scheme is already in place in the form of registers of cancer and childhood cancer, so that a study of geographical and temporal trends in incidence requires no special data collection systems in most countries. It should be noted that the relatively good prognosis of childhood leukaemia in many countries and likely improvements in therapy and survival in others mean that mortality data are virtually useless for monitoring risk.

METHODS

The study began in 1987. It involves cancer registries in some 21 European countries (Table 1). Each collaborating centre agrees to follow a common protocol. This requires that they provide, each year, data on all recorded cases of childhood leukaemia and lymphoma occurring in the populations covered. Collation of data from the different centres and their analysis are coordinated by IARC. Registries send an updated file of every case registered, with details of age, date of birth, date of diagnosis, place of residence, and histological diagnosis, at annual intervals.

Table 1 shows the status of the data received on 11 January 1993. In addition to the case-listings, the collaborators are requested to provide annual estimates of the childhood population at risk, for the same sub-national areas as those for which estimated radiation exposure information is available. The estimated excess dose of radiation due to the accident (as dose equivalents in μsSv) for the first and subsequent four years post-accident were supplied by UNSCEAR (see Table 2).

RESULTS

The first report of the study, incorporating cancer data to the end of 1988, has now been published (Parkin et al., 1992). In addition, a revised analysis has been undertaken, using data from most centres to the end of 1989. In this revised analysis, the observed numbers of leukaemia registrations were assumed to have a Poisson distribution, with the mean proportional to the equivalent population at risk, and depending on region, age, sex, and year of diagnosis. Models with these terms were fitted using GLIM, and relative risks estimated for each of the variables. Age was fitted in three categories: 0-4, 5-9 and 10-14.

In addition, an extra variable, dose, was calculated for the cases and person-years at risk, and fitted in the model to determine the effect on risk of leukaemia.

The assumptions made in estimating the excess dose received by a particular child were as follows:

1. Exposure to the (excess) radiation can affect only children born before the year of exposure.
2. The effect appears one year after exposure, and lasts seven years, as a corollary:
3. Children aged under one year would not be affected (by radiation exposure received after birth).
4. The dose received by the foetus is assumed to be 0.25 of that of a child aged 1.
5. The effect of radiation received as a foetus would persist, and the dose equivalent at every age is equal to the estimated foetal dose in the year of birth.

Thus, the effective dose can be estimated by region, age-group, and year. In this population-level analysis, dose cannot vary within region and year, so that it is not possible to estimate its effect from a model including all of these parameters.

Dose was considered at five levels (within five-year age-groups):

Level	Average annual dose equivalent
1	0 (no excess radiation dose)
2	0.1 < 60 μSv
3	60 < 130
4	130 < 300
5	300+

In each model, the significance of the various factors was assessed by the change in residual deviance which resulted from its omission, and comparing this with the X^2 distribution for the appropriate degrees of freedom.

RESULTS

First, to look at the effect of Region, the model 'Age + Sex + Year + Region' was fitted for the period 1980-86 (i.e., when no excess exposure effects are possible (dose level = 1)). The effect of region was highly significant (Figure 1). Region was therefore regrouped into three categories of pre-existing risk: (1) low risk (RR < 0.92); high risk (RR > 1.15); and (3) medium risk (other). This categorization (area) is independent of estimated dose. A model containing 'Area + Sex + Age + Year' was then fitted. The effects of age and sex were, as expected, highly significant - the relative risk for girls was 0.86; and, by age, expected differences were observed (Figure 2). The risk by year was studied relative to 1980. Although none of the risks for the individual years was significantly different from 1.0 (the reference year, 1980), there is a distinct impression of a rising incidence over time (Figure 3). The trend test is positive ($X^2 = 6.81$, df = 1, p = < 0.01).

Substituting Dose in this model, instead of Year, results in no significant trend of risk according to dose (Table 3) (Trend test: $X^2 = 1.65$, df = 1, p ~0.2).

CONCLUSIONS

As in the earlier correlation analysis (Parkin et al., 1992), there is no systematic trend of risk with increasing dose. The present analysis uses rather more data than the earlier one, including, for example, results from 1989 for several centres. However, one cannot really expect to see any effect until the 1990 data are entered. The regression model used allows a rather more sophisticated analysis than previously, particularly the control for the underlying time trend in incidence. Dose has also been estimated rather more accurately, with an attempt to produce cumulated dose for each child. However, defects in the denominator (population-at-risk data) mean that coarse groupings of children (five-year groups) must be used, with inevitable misclassification of dose within the category (the validity of individual doses based on estimates for wide geographical areas produced by somewhat arcane simulation models is another issue, but outside the scope of the ECLIS study). Population-at-risk data by single year of age have been requested from all collaborators, and have now been received from almost all of them. It is clearly imperative also to include the data from Belarus and the Russian Federation which have been promised but not yet received.

REFERENCES

Parkin, D.M., Cardis, E., Masuyer, E., Friedl, H.P., Hansluwka, H., Bobev, D., Ivanov, E., Sinnaeve, J., Augustin, J., Plesko, I., Storm, H.H., Rahu, M., Karjalainen, S., Bernard, J.L., Carli, P.M., L'Huillier, M.C., Lutz, J.M., Schaffer, P., Schraub, S., Michaelis, J., Möhner, M., Staneczek, W., Vargha, M., Crosignani, P, Magnani, C., Terracini, B., Kriauciunas, R., Coebergh, J.W., Langmark, F., Zatonski, W., Merabishvili, V., Pompe-Kirn, V., Barlow, L., Raymond, L., Black, R., Stiller, C.A. and Bennett, B.G. (1993) Childhood leukaemia following the Chernobyl accident: the European Childhood Leukaemia-Lymphoma Incidence Study (ECLIS). Eur. J. Cancer, 29A, 87-95.

United Nations Scientific Committee on the Effects of Atomic Radiation (1988) Sources, Effects and Risks of Ionizing Radiation (1988 Report to the General Assembly, with annexes). United Nations, New York.

Table 1

(Data received on 11 January 1993)

Austria	1980-87	Leukaemia/Lymphoma
Bulgaria	-	-
ex-Czechoslovakia: Bohemia	1980-90	Leukaemia/Lymphoma
Moravia	1980-90	Leukaemia/Lymphoma
Slovakia	1980-89	Leukaemia/Lymphoma
Denmark	1980-88	Leukaemia/Non-Hodgkin lymphoma
Finland	1980-89	Leukaemia/Lymphoma
France: Bas-Rhin	1980-89	Leukaemia/Lymphoma
Dijon	1980-90	Leukaemia/Lymphoma
Doubs	1980-87	Leukaemia
Isère	1980-87	Leukaemia/Lymphoma
Lorraine	1983-89	Leukaemia/Lymphoma
PACA & Corsica	1984-88	Leukaemia/Lymphoma
ex-German Democratic Rep.	1980-89	Leukaemia/Non-Hodgkin lymphoma
Germany, west	1980-89	Leukaemia/Lymphoma
Hungary	1980-88	Leukaemia
Italy: Piedmont	1980-89	Leukaemia
Varese	1980-87	Leukaemia/Lymphoma
Netherlands	1980-89	Leukaemia
Norway	1980-87	Leukaemia/Lymphoma
Poland	1980-89	Leukaemia/Lymphoma
Slovenia	1980-89	Leukaemia/Lymphoma
Sweden	1980-89	Leukaemia
Switzerland: Basel	1980-88	Leukaemia/Lymphoma
Geneva	1980-88	Leukaemia/Lymphoma
Neuchâtel	1980-88	Leukaemia/Lymphoma
St-Gall	1980-88	Leukaemia/Lymphoma
Vaud	1980-88	Leukaemia/Lymphoma
Zürich	1980-87	Leukaemia/Lymphoma
UK: England & Wales	1980-88	Leukaemia/Lymphoma
Scotland	1980-89	Leukaemia/Lymphoma
ex-USSR: Belarus	-	-
Estonia	1980-89	Leukaemia/Lymphoma
Lithuania	1980-90	Leukaemia/Lymphoma
Russian Federation	-	-

Table 2

Radiation doses in European countries from the Chernobyl accident (estimates prepared by UNSCEAR): effective dose (μSv)

	First year	0-4 years
Austria	670	1101
Bulgaria	760	900
region 1	720	790
region 2	800	1020
ex-Czechoslovakia	350	440
region 1	275	320
region 2	355	450
region 3	340	390
Denmark	30	55
Finland	460	730
France, region 3	150	210
ex-German Democratic Republic	210	340
region 1	260	370
region 2	340	540
region 3	175	290
Federal Republic of Germany	130	200
region 1	67	100
region 2	130	200
region 3	490	780
Hungary	230	285
region 1	280	370
region 2	175	203
Italy, region 1	374	485
Netherlands	57	90
Norway	230	330
Poland	270	370
Slovenia	620	1045
Sweden	150	270
region 1	390	960
region 2	85	100
region 3	105	150
Switzerland	270	320
region 2	315	380
region 3	205	240
region 4	120	145
United Kingdom	27	32
region 1	12	14
region 2	105	140
region 3	190	250
ex-USSR*	260	350
region 1	1960	2680
region 3	445	630
region 4	140	190

*Belarus, Estonia, Lithuania, Russian Federation

Table 3

Fitting Area, Sex and Age + Dose

Dose level	1	1.0
	2	1.02 (0.96-1.07)
	3	1.20 (1.13-1.28)
	4	0.96 (0.90-1.01)
	5	1.03 (0.96-1.10)

No significant trend

(X^2 = 1.65, df = 1, p = 0.2)

Relative Risk by Region (Period to 1986)
Adjusted for Age+Sex

Figure 1

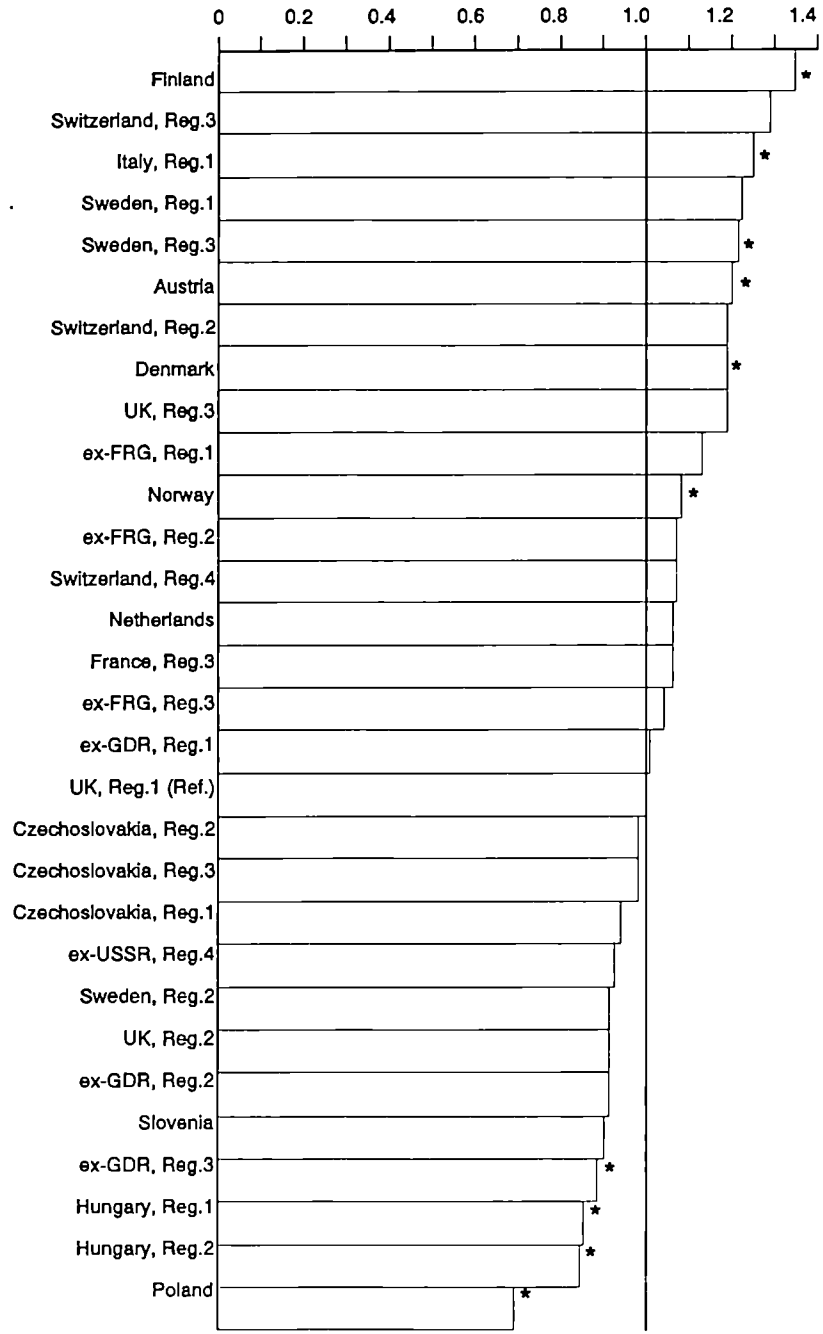


Figure 2

Relative Risk, by age (adj. for sex, region, year)

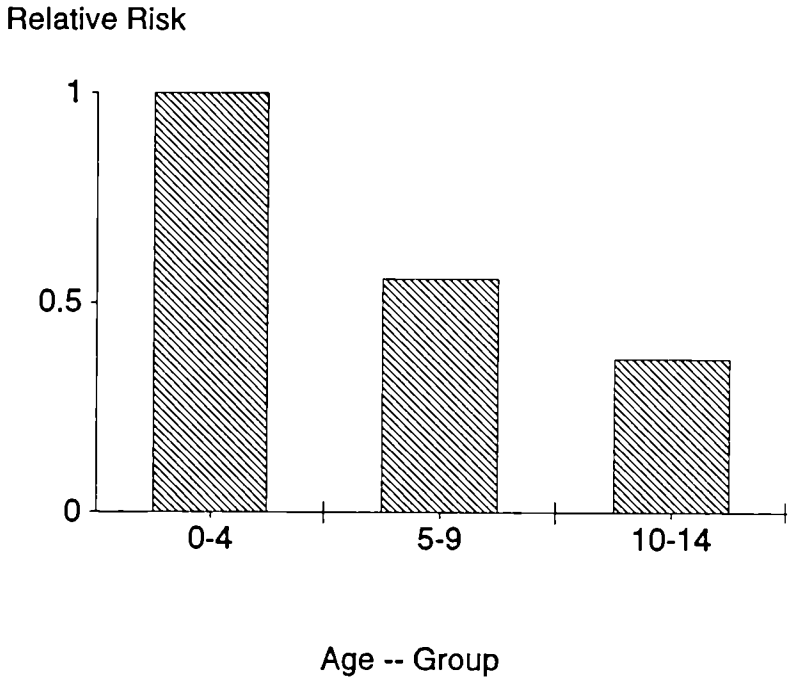
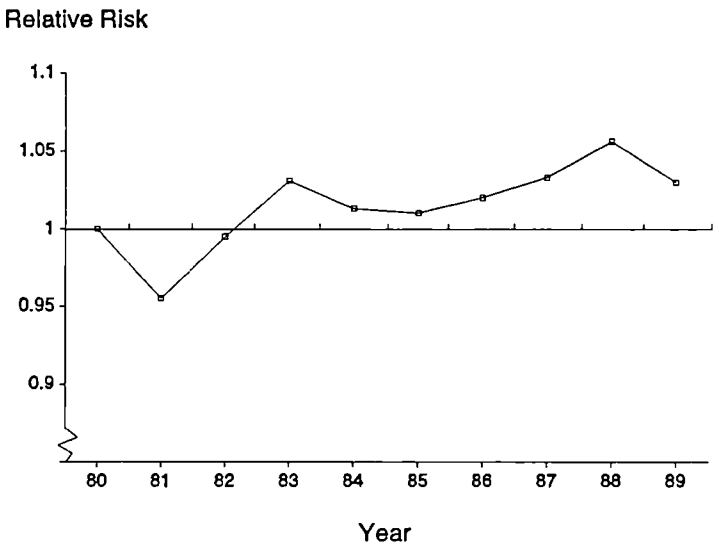


Figure 3

Relative Risk, by year (adj. for age, sex, region)



C14 Epidemiological studies in human populations.

Contract FI3P-CT920064f Epidemiological studies and tables.

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Description of research work

Objectives of project:

To conduct epidemiological studies and obtain information on radiation-induced cancer risks based on the following medically-irradiated cohorts:

(a) patients with exposures to I-131 or I-123;
(b) persons irradiated in childhood for skin haemangioma;
(c) persons given radiotherapy for a first cancer, either in childhood or in adulthood, and followed to determine second cancer incidence;
(d) patients injected with Ra-224.

To analyse and summarise data on populations exposed to high doses, such as the Japanese atomic bomb survivors. To examine models, both empirical and mechanistic, for radiation-induced cancer risks. To continue the construction of radioepidemiological ('probability of causation') tables for Europe.

To study cancer mortality around French nuclear installations and in French new towns.

To examine and apply statistical methods for analysing the geographical distribution of disease.

To review information on childhood cancer risk in relation to in utero and preconception irradiation.

To continue and extend a study of occupational radiation exposure and mortality, namely the UK National Registry for Radiation Workers.

Description of project

1. Background

(a) Risk Models

The International Commission on Radiological Protection (ICRP) has recently published its new recommendations (Ann. ICRP, no. 1-3, (1991)), which contained revised estimates of the risks of radiation-induced cancer. These values were derived largely on the basis of the follow-up of the Japanese atomic bomb survivors, based on the DS86 dosimetry system. However, several factors give rise to some uncertainty in these estimates, as follows.

(i) The latest published follow-up of the Japanese survivors only covers the period up to 40 years following exposure. To estimate risks of radiation-induced cancer over a lifetime therefore requires a model for projecting risks over time. Depending upon which model is used, the estimate of lifetime risk can vary by a factor of about 2 to 3, and the uncertainty is greater for those irradiated early in life (Muirhead, Radiat. Prot. Dosim., 36, 321 (1991)).

(ii) The Japanese survivor data do not allow the dose-response relationship to be estimated precisely, although analyses of these data do give some indication of which types of relationship are consistent with the data.
(iii) There is uncertainty in how to transfer radiation-induced cancer risks in a Japanese population to a western population with different baseline cancer rates.
(iv) The data from the Japanese survivors for some individual types of cancer are not strong enough to allow risk coefficients to be derived accurately.
(v) The Japanese survivor data provide little information on the effects of exposure to high-LET radiation.

Two ways in which it is intended to address these topics are to obtain data on other groups - in particular, those irradiated for medical reasons - and to make optimal use of the available data by developing and implementing appropriate statistical techniques.

One of the benefits of better quantifying the risks of radiation-induced cancers is the opportunity to estimate the probability that a cancer which has occurred was induced by prior radiation exposure.

Radioepidemiological tables for this 'probability of causation' were developed in 1985 by the US National Institutes of Health for an American population, based largely on the Japanese data for the earlier T65DR dosimetry. It is intended to use risk models developed under the proposed programme of work in the construction of radioepidemiological tables for European countries.

(b) Low Dose Studies

In recent years there has been considerable interest in reports of raised incidences of childhood cancer - in particular, leukaemia - near several UK nuclear installations. However, these observations have not been explained on the basis of assessments of the radiation exposure of the relevant populations (Stather et al, NRPB-R215 (1988)). To examine this issue further, studies have been carried out around nuclear installations in other countries in order to determine whether these observations can be replicated, and it is intended to continue a study of this type in France.

Information on the underlying geographical distribution of childhood cancer would be of help in allowing the results around nuclear installations to be viewed in context, and it is proposed to analyse data from Britain on topic. Two specific hypotheses put forward for childhood leukaemia clusters, namely 'population mixing' (suggested by Kinlen and supported by studies in British new towns) and pre-conception radiation exposure (suggested by Gardner et al (Brit. Med. J., 300, 423 (1990)) will be addressed by a study of French new towns and by examining epidemiological data relating to pre-conception exposure.

There have also been suggestions that the geographical distribution of leukaemia may be influenced significantly by exposure to natural radiation - in particular, radon (Henshaw et al, Lancet, 335, 1008 (1990)). Whilst an analysis of British data did not support this (Muirhead et al, Lancet, 337, 503 (1991)), it is intended to perform further analyses that take account of socio-demographic variables and also to develop other statistical approaches for analysing such data.

In view of uncertainties in the use of data from studies of high dose and high dose rate exposure to predict risks associated with occupational exposure to low doses at low dose rates, an alternative is to conduct a study of groups with occupational exposures. However, since the associated risks are likely to be low, it is important to base such a study on as large a cohort as possible, in order to maximise its statistical power. The UK National Registry for Radiation Workers currently includes about 100,000 workers and results from its first analysis have recently been published (Kendall et al, Br. Med. J., 304, 220 (1992)). It is intended to continue and extend this study.

2. Work Proposed

(Further details are given under the description of each participant's contribution).

2.1 Studies of Medically Irradiated Populations

It is proposed to undertake work on the following epidemiological studies of populations irradiated for medical reasons.

i) University of Saarland

A study of thyroid cancer incidence among just under 12,000 patients in the Saarland (Germany) who received I-131 exposures will continue. The first follow-up of this cohort is currently being completed, and it is intended to continue the follow-up into the future. It is beneficial to obtain epidemiological data on thyroid cancer risks in relation to I-131 exposures, because of the importance of this radionuclide if a reactor accident were to occur. It is also intended to study another group of patients whose exposures were to I-123. Comparison of thyroid cancer risks in the two cohorts will be informative because the dose per unit activity is much lower for I-123 than for I-131.

ii) Karolinska Institute

Sweden is an EFTA country. The contribution towards the Karolinska Institute will be published in an addendum after signature of the Association Agreement.

iii) University of Florence

It is intended to continue a study of second tumours among persons given radiotherapy for a first tumour. The study has so far been concentrated at the University of Florence, where about 1,800 new patients receive radiotherapy each year. Follow-up of about 1,400 patients treated for breast cancer provides some evidence of an excess of leukaemia as a second cancer (6 observed versus 1.3 expected).

It is proposed to look at patients treated for other types of first tumour and to involve other Italian centres, such as those at the Universities of Modena and Pisa, so as to increase the statistical power of the study.

iv) INSERM-U.351

A cohort study of second cancer incidence among children who have survived at least two years following a first cancer is commencing, based on various centres throughout Europe.

To date about 1,500 children have been included from France, UK and Spain, and it is aimed eventually to include about 5,000 children from about twenty centres. Doses to individual organs will be estimated, and the dose-response relationship for second cancers will be investigated, along with the joint effect of radiotherapy and chemotherapy. This project and that by participant number 5 will be important in obtaining further information on the risks of exposure early in life.

v) GSF
University of Munich

Two studies of late effects among patients in Germany treated with Ra-224 for ankylosing spondylitis or bone tuberculosis will continue (participant numbers 10 and 11).

Statistical analyses of these data will be conducted, and the results compared with those from other studies, particularly as regards the excesses of breast and kidney cancers observed in the older cohort. Cataracts will also be evaluated using a Scheimpflug camera system (participant no. 12).

2.2 Risk Models and Radioepidemiological Tables

(i) NRPB

Data on populations exposed to high radiation doses, such as from the continued follow-up of the Japanese atomic bomb survivors, will be studied with respect to the effect of factors such as dose, age and time on the risk of radiation-induced cancer. As well as empirically-based models, models that attempt to describe mechanisms of carcinogenesis will be examined. Non-cancer mortality among the Japanese survivors will also be examined, in view of indications of an excess at doses above 2 Gy.

(ii) University of Munich

Models for projecting radiation-induced risks over time, such as a model based on attained age, will be examined and the implications studied. Non-parametric data analyses will be performed, and illustrative diagrams will be produced to help in understanding important features of data on the Japanese atomic bomb survivors and on uranium miners.

It is intended that the models derived will be used in constructing radioepidemiological tables for the 'probability of causation' (in collaboration with participant no. 3).

(iii) GSF Neuherberg

Work on the construction of radioepidemiological ('probability of causation') tables for European countries will continue. Particular attention will be given to the construction of Tables for leukaemia and, in the instance of radon exposure, for lung cancer. Information on the risks of other types of cancer will be reviewed.

2.3 Geographical and Low Dose Studies

(i) NRPB

Further study will be made of a statistical technique, developed at NRPB under the previous stage of the Association Agreement, for detecting over-dispersion in geographical disease rates. This technique can be used in examining whether a disease has a 'natural' tendency to cluster. It is intended, in collaboration with the Childhood Cancer Research Group (CCRG), University of Oxford, to apply this technique to the CCRG's data on the geographical distribution of childhood cancers throughout Britain.

An analysis performed under the previous stage of the Association Agreement did not show any statistically significant correlations between NRPB's data on natural radiation levels and CCRG's data on childhood leukaemia incidence in small areas (districts) throughout Britain (Muirhead et al, Lancet, 1991). A further analysis in which adjustment is made for socio-demographic variables is proposed. It is also intended, in collaboration with INSERM-U.170 (participant no. 7), to examine the properties of recently developed statistical methods for analysing geographical correlations, and it is hoped to apply these methods to the above NRPB and CCRG data, in conjunction with CCRG.

Epidemiological information on childhood cancer risk in relation to irradiation in utero and preconception irradiation will be examined.

(ii) INSERM-U.170

Statistical methods developed under the previous CEC contract BI6-126-F for testing for associations between the geographical distribution of disease and possible risk factors will be examined in the context of small area statistics. In collaboration with NRPB (participant no. 1) the properties of other methods will be studied. It is hoped to apply these methods, in collaboration with NRPB and the Childhood Cancer Research Group (CCRG), to small area data throughout Britain on childhood cancer, natural radiation and socio-demographic variables. Problems relating to the interpretation of geographical correlation studies, such as the possibility of bias, will be investigated using simulations.

(iii) Institut Gustave Roussy

A study of mortality at ages less than 25 years around French nuclear installations will continue. In view of earlier observations, the literature on clustering of Hodgkin's disease will be examined. In another study, leukaemia mortality among young persons in French new towns during 1968-89 will be examined, in order to test the hypothesis of L.J. Kinlen that childhood leukaemia may have a viral aetiology.

(iv) NRPB

Following the publication in 1992 of the first analysis of the UK National Registry for Radiation Workers, covering approximately 95,000 workers, the follow-up of this cohort will be extended and additional groups of workers added. This will increase the statistical power of future analyses of radiation exposure and cancer in this study.

3. Distribution of Tasks Amongst Participants

As indicated above, the work proposed falls into three areas. Within these areas there will be particular links, for example between GSF and the University of Munich on the construction of radioepidemiological tables and on studies of patients treated with Ra-224, and between NRPB and INSERM-U.170 on statistical methods for analysing geographical disease correlations. However, each of the participants will have the opportunity to provide an input to the work of the other participants, both within and outside the three main areas, through the project's Scientific Co-ordinating Committee which will meet at regular intervals. The participants are drawn from a wide range of disciplines, including epidemiology, statistics, medicine, radiation dosimetry and radiation biology, and it is intended that the participants will share their expertise on these topics. Furthermore, it will be possible, through the Scientific Co-ordinating Committee, for participants to keep each other informed of, for example, developments in methodology and new epidemiological results obtained in their work.

Contribution of NRPB

Statistical Studies of Radiation Risk

(a) Modelling Radiation Risk in Populations Exposed to High Doses

The International Commission on Radiological Protection (ICRP) has recently published its revised recommendations, which included estimates of radiation-induced cancer risks derived from the follow-up of the Japanese atomic bomb survivors based on the DS86 dosimetry system. However, several factors give rise to some uncertainty in these estimates.

First, the latest published follow-up only covers the period up to 40 years following exposure, and the future pattern of risk will have a substantial effect on the risk incurred over a lifetime, particularly for those exposed when young. Second, while analyses of the dose-response relationship among the atomic bomb survivors provide an indication as to which kinds of dose-responses are consistent with the data to date, the data lack sufficient statistical power to allow the dose-response to be estimated precisely; however, the power may be increased by longer follow-up of the survivors. Third, there is uncertainty in how to transfer radiation-induced cancer risks in a Japanese population to a western population with different baseline cancer rates. Fourth, the data for some individual types of cancer are not strong enough to allow risk coefficients to be derived accurately, and for cancers that are generally non-fatal data from other populations have to be utilised.

It is intended, in collaboration with the University of Munich (participant no. 2), to study these topics using data on the Japanese atomic bomb survivors made available by the Radiation Effects Research Foundation (RERF) in Japan and on other irradiated populations. RERF has already released mortality data based on the follow-up of the survivors to 1985 and more data will become available with further follow-ups.

In particular, RERF will shortly be publishing a follow-up to 1987 of cancer incidence, which will allow an examination of non-fatal cancers. Data of this type will be analysed to examine how the cancer risk varies according to dose, time since exposure, age at exposure and sex. Data on western populations will also be studied to obtain a better idea as to how cancer risks vary across populations.

As well as considering empirically-based risk projection models (ie. relative and absolute models, plus variants thereof), models that attempt to describe mechanisms of carcinogenesis will be examined. Some analyses have already been performed using the Armitage-Doll multi-stage model and it is intended to look at other types of mechanistic model, such as that of Moolgavkar and Knudson. Study will be made of the weight of evidence in favour of the various mechanistic models, including how well they fit the epidemiological data, as well as the implications for the calculation of lifetime risk estimates. A new report from RERF suggests that there may be an excess of non-cancer mortality among the Japanese atomic bomb survivors at doses in excess of 1-2 Gy. It is intended to analyse the detailed data set from RERF in order to examine the implications for exposures at low doses.

b) Geographical and Low Dose Studies of Childhood Cancer

In recent years there have been several reports of raised incidences of childhood cancer - in particular, leukaemia - near several UK nuclear installations. One of the problems in interpreting these reports has been the lack of information on the underlying geographical distribution of childhood cancer in small areas which would allow the results around nuclear installations to be viewed in context. However, a database on the distribution of childhood leukaemia throughout Great Britain has now been constructed by the Childhood Cancer Research Group (University of Oxford), which will soon be extended to cover all cancers.

Various statistical methods have been suggested for the detection of localised clustering of disease, and the International Agency for Research on Cancer recently performed a comparative study of some methodologies. One of the methods included in this study was a technique, developed at NRPB under the previous phase of the Association Agreement from a method described by Potthoff and Whittinghill (Biometrika 1966) to identify clustering over different sizes of area.

It is intended during 1992-94 to make further study of the statistical properties of this technique and to examine whether it could be extended to look for over-dispersion in disease rates after adjusting for other geographical variables. It is also intended, in conjunction with the Childhood Cancer Research Group (CCRG), to apply this technique to the CCRG's data on the geographical distribution of childhood leukaemia and other cancers in Great Britain, both on the basis of the address at diagnosis and the place at birth.

There was considerable interest in recent reports of a statistically significant correlation between the incidence of leukaemia and mean radon levels in different countries.

However, the interpretation of this result was affected by differences in data quality between countries. An analysis of data on natural radiation levels held by NRPB and on childhood leukaemia in small areas (districts) throughout Great Britain held by CCRG did not show any statistically significant correlations (Muirhead et al, Lancet, 1991). This analysis also suggested that results based on larger areas (counties) may have been affected by geographical confounding factors.

It is therefore intended, in conjunction with CCRG, to analyse the CCRG data on childhood leukaemia and other cancers, in relation to socio-demographic variables as well as the NRPB data on indoor radon concentrations and indoor and outdoor gamma dose rates. It is also proposed that INSERM-U.170 (participant no. 7) will study related methods and apply them to the CCRG and NRPB data in collaborative analyses.

There is continuing interest in the risk of cancer following irradiation in utero, based on studies of prenatal x-ray exposure and of exposure from the atomic bombings of Hiroshima and Nagasaki. Also, the observations of Gardner et al (BMJ, 1990) have raised the question of whether preconception irradiation affects the risk of childhood leukaemia. Relevant epidemiological information on irradiation of the embryo and fetus and on preconception irradiation will therefore be studied.

Contribution of the Radiobiological Institute of the University of Munich

Modelling of Radiation Risks and Construction of Radioepidemiological tables

Quantitative risk estimates have become increasingly important for the practise of radiation protection. The dose revision and the new evaluation of the study of atomic bomb survivors have led to increased risk estimates for radiation carcinogenesis and to a reduction of the dose limits in radiation protection regulations. The postulate of a dose reduction factor in the risk estimates by ICRP has by some been criticised as being arbitrary, and, this has led to requests for further restrictions in the dose limits. The proposed work is aimed at a critical assessment of the hypotheses underlying the derivation of risk estimates. This includes:

- A critical assessment of the choice of different models in the analysis of BEIR V and an effort to reduce the number of models.

- A comparison of the age-attained models that are usually employed with the radon data, for example in BEIR IV, with the models applied to the Japanese data. This will include a detailed analysis of the various models to project relative risk in time after exposure and in age attained.

In part this will be a continuation of present work that indicates that an age-attained model is in better agreement with the trend of the Japanese data for solid cancers, specifically of those exposed at young ages, and that the changed projection in time after exposure leads to lifetime attributable risks that are, averaged of all ages, smaller than the ICRP estimates by about a factor of two.

- A non-parametric analysis of the dependencies of relative risk on age attained and age at exposure.

- The extension of illustrative diagrams to provide better understanding and visualisation of the Japanese data and the data of the major cohorts of uranium miners.

- Use of the results to radioepidemiological tables of the probability of causation and their adaption to the use in European countries.

Contribution of GSF Neuherberg

To establish probabilities of causation (PC-tables) for lung cancer due to radon inhalation and for leukaemia. These are essential components within a wider effort by GSF to prepare PC-tables for use in the European countries.

PC-Tables for Lung Cancer Induced by Exposure to Radon and Radon Daughters

In the preceding period we have performed, jointly with the group in Prague, an analysis of the S-cohort of uranium miners with methods and models similar to those used by the BEIR IV-Committee. In agreement with the conclusions of the BEIR IV-Committee, it was shown that the added risk of lung cancer due to radon inhalation is strongly dependent on age attained. Although there are indications of non-linear dependencies on exposure, ie. an increased excess per unit exposure at low exposures, the analysis confirmed essential aspects of the model chosen by the BEIR IV-Committee. The step functions employed by the BEIR IV-Committee are not suitable for the construction of PC-tables; to avoid the resulting discontinuities we have used analytical functions. On the basis of this analysis it is now possible to develop a model which agrees with the essential findings in the major cohorts of uranium miners. Probabilities of causation will be computed for various circumstances of exposures, ie. for different ages at start of exposure and for different patterns of exposure. The recently initiated studies on the uranium miners of the Wismut AG in the former GDR will duly be accounted for in the work.

PC-Tables for Leukaemia

The NIH-Committee, in establishing radioepidemiological tables for the USA, has used an absolute risk model with dependence of the total risk on age at exposure. The BEIR V-Committee, on the other hand, has advocated a relative risk model in which the characteristic wave-like dependencies on time after exposure were quantified in terms of simple step functions. The objective of work in the past project period was to update the PC-tables established by the NIH-Committee after revision of the study on the atomic bomb survivors. We have compared the relative risk and the absolute risk model under the assumption of a gamma-distribution in time after exposure. The reason for this choice instead of a log-normal distribution as used by the NIH-Committee has the purely practical reason that it allows the use of the computer package EPICURE used in the Japanese studies. According to our analysis a relative risk model fits the Japanese data

better than an absolute risk model. It is sufficient to use two different gamma-distributions, one for ages at exposure <30 years and one for ages >30 years. Within the project we will now establish the radioepidemiological tables for leukaemia according to our best model, ie. a relative risk model with two different gamma-distributions according to age at exposure. The relative risk model leads to a considerable simplification in the calculations and in the use of the PC-tables, since the PC-values are then independent of the age-specific rates of leukaemia.

The continuation of the work will be to review the present risk estimates and the evidence on which they were based for other sensitive organs.

Contribution of the University of Saarland

Epidemiological studies of thyroid cancer following medical exposures to radioactive iodine

1. Completing the study "Incidence and mortality from thyroid cancer in a cohort with medical exposures to I-131".
   This study involves 11776 patients who had been given I-131, for reasons either of diagnosis or of treatment, at the Department of Nuclear Medicine at the University of Saarland (Homburg) between 1962 and 1977. Funding for this study was provided during 1990-91 under CEC contract BI6-347h, to which these proposals form a continuation. Since we are dependent on the Cancer Registry of the Saarland for determining the incidence of thyroid cancer in the cohort, it will be necessary to continue the study into the new contract period.

2. Continuing the follow-up of the cohort with medical exposures to I-131.
   In the case of thyroid carcinoma latent periods of 30 years and more are observed. Most of our former patients have not yet been followed for this time. Therefore the cohort of patients treated with I-131 at our department should be followed up beyond the end of the present study.

3. A study to determine incidence and mortality rates for thyroid carcinoma in a cohort with medical exposures to I-123.
   Not only the patients treated with I-131 but also these patients who were treated with I-123 since the year 1977 should be followed-up. The incidence and mortality rates of thyroid carcinoma for the cohort of patients with medical exposures to I-123 can then be compared with those for the cohort with medical exposures to I-131. It should be noted that the organ dose from I-123 exposure is a factor of 100 lower than that from I-131 if the applicated activity is the same.
   It will be necessary to create a second database containing all the patients who were or are treated with I-123 at our department. Just as in the current study it will be necessary to distribute a questionnaire, on the one hand to find out the state of health of our former patients and on the other hand to prove data like name, address and date of birth.

   The second database will then be compared with those persons who are registered in the Cancer Registry of the Saarland.

Contribution of the Karolinska Institute

Sweden is an EFTA country. The contribution towards the Karolinska Institute will be published in an addendum after signature of the Association Agreement.

Contribution of the University of Florence

Epidemiological study of second tumours in radiotherapy patients

Studies of radiation carcinogenesis present remarkable difficulties mainly related to the lack of a large enough number of cases during the follow up. Radiotherapy represents a good source of subjects exposed to ionising radiations. In fact the dose to the irradiated volume is accurately measured and the dose absorbed by other parts of the body can be calculated with sufficient accuracy since the conditions related to the radiation source and to the patients are well known.

Recently, many thousands of patients with tumours in different organs and tissues underwent radiotherapy at the Radiotherapy Centre (University and Hospital) of Florence. Clinical records of these patients document well the dose, irradiated volume and treatment schedules.

Patients (about 2500 per year) were followed up for many years (some of these for over 30 years) for periodic evaluation of the disease and for the appearance of late radiation damage. About 1800 new patients undergo radiotherapy every year. Different radiation sources (telecobalt unit, 3 linear accelerators, after loading sources) were used for external and endocavity radiotherapy. Moreover radiometabolic treatments with 131-I for thyroid carcinoma, hyperthyroidism and toxic adenoma have been performed. Conventional fractionation (2 Gy/die, 5 days/week) is used for external radiotherapy with X rays, 60 Co, X rays, X and accelerated electron beams from linear accelerators. More recently different fractionation schedules (1 Gy x 3/die, 2 Gy x 3/die and 2 Gy x 2/die) have been introduced.

The use of these different radiation sources and treatment schedules makes possible the evaluation of the incidence of a second primary tumour in the irradiated area or outside of it. Most cases resulted from primary breast cancers, gynaecologic tumours, head and neck tumours, and thyroid carcinomas.

During the treatment and the follow up, besides clinical and radiological examinations, the concentrations of some important biochemical indicators were assayed in order to evaluate the correlation of these parameters with the condition of the patient, prognosis, recurrence, and the appearance of a second tumour.

At the end of this study the epidemiological analysis will be carried out to find the correlations between irradiation and tumour incidence.

The Radiotherapy Sections of the University of Modena, Pisa and Siena will participate in the study with their own cases. Agreements are ongoing with the Radiotherapy Centre of the University of Brescia.

Contribution of INSERM-U.170

Statistical methods, biases and interpretation of geographical correlation studies, with application in analysing the geographical association of cancer and radiation exposure. There has been growing interest in the use of geographical analyses to pursue some epidemiological aspects of the difficult question pertaining to the potential effects of low dose radiation on human health.

In particular, childhood cancer incidence around nuclear installations has been assessed in the U.K. and geographical studies of associations between natural background radiation or radon levels and cancer incidence or mortality (for some sites) have been carried out. In our previous contract (BI6 126 F), an extensive review on the use and pitfalls of correlation and regression methods for testing associations between the geographical distribution of risk factors and disease was completed, leading to the development of new tests of geographical correlations and implementation guidelines for spatial regressions.

These methods were formulated and discussed in a framework where the variability within each geographical unit is small in comparison to the between units variability, as is the case for medium size geographical units (for instance French départements) and not too rare diseases. Currently, within the availability of good geographical resolution of cancer incidence, small area studies have attracted considerable epidemiological interest. The work proposed under the Association Agreement extends and complements our previous work in several respects.

a) The methods developed under our previous contract will be examined in the context of small area statistics. Simulations will be implemented to assess their performance in this context.

b) Interest will be focussed on other statistical methods which allow for the influence of covariates on disease incidence at a small area level and on a comparison of these different approaches. These other methods include an extension of the technique developed at NRPB (cf. participant no. 1) for identifying clustering over different sizes of area, extension which allows for consideration of explanatory variables. They also comprise a Bayesian hierarchical model approach. This latter method was originally proposed by several authors as a way of reconstructing the underlying geographical distribution of rare diseases. The inclusion of covariates within this model has recently been suggested.

c) In collaboration with NRPB (Dr. C. Muirhead) and the Childhood Cancer Research Group (CCRG), it is hoped to be able to use these methods in analyses of the CCRG data on childhood leukaemia and other cancers in Britain in relation to socio-demographic variables and to the NRPB data on indoor radon concentrations and indoor and outdoor gamma dose rates.

d) Problems relating to the epidemiological interpretation of geographical correlation studies will be reviewed and some specific sources of biases of geographical studies will be investigated. These biases are related to some extent to the difficulty of correctly allowing for the joint effect of risk factors at a group level (eg. geographical units). They will be quantified using a flexible simulation model of joint effects.

Parts (b) and (c) of this proposal will be carried out in close collaboration with Dr. C. Muirhead of NRPB (cf. participant no. 1).

Contribution of the Institute Gustave Roussy

(a) Mortality around French nuclear sites.

France derived 75% of its electricity from nuclear energy in 1989 and the first nuclear unit producing electricity started operating industrially in 1962.

An excess mortality from leukaemia has been observed in the vicinity of nuclear sites in Great Britain in the population under age 25.

We are studying mortality patterns under age 25 around French nuclear sites. The first results have been published. A detailed publication is in preparation. A cluster of deaths from Hodgkin's disease has been observed in the vicinity of a reprocessing plant. We think that the cluster is not likely to be related to radiation and plan to review the literature on the clustering of Hodgkin's disease.

(b) Leukaemia mortality between 1968 and 1989 among children living in French towns with a recent and large increase in population.

Kinlen suggested that the excess risk of leukaemia around nuclear sites might be related to the pattern of infection associated with the large influx of population caused by the building of the installation.

The exposure to viruses at a young age associated with the mixing of population of different origins might lead to an increased risk of leukaemia. This hypothesis has been tested first in Scotland on the new town of Glenrothes (Kinlen 1988), and subsequently on fourteen new towns throughout Britain (Kinlen 1990). The results are consistent with an infectious aetiology for leukaemia, the mixing of population of diverse origins increasing the risk of exposure to infections particularly in utero or during early childhood.

The aim of our study is to confirm or refute the hypothesis of a viral aetiology in childhood leukaemia. We plan to compare the mortality from leukaemia of children aged 0 to 24, living in French towns having had a recent increase of population, to national mortality rates.

This work will be performed on the 1968-1989 period, for the 90 towns of over 10,000 inhabitants whose population has increased by more than 50% between the 3 last censuses. This corresponds to 720,000 subjects in 1968 and 1,800,000 in 1990, leading to 33,500,000 person-years of observation, among which 2,400,000 are for those under 5 years of age. These data will allow us to show an increased risk of leukaemia death of 50%, with a power of 90%.

Contribution of INSERM-U.351 (Institut Gustave Roussy)

Second cancers incidence after first cancer in children: A cohort study.

Aims of the study

A cohort study of 5,000 children having survived at least 2 years after their first cancer recruited from about twenty European centres.

The objective of this study is threefold:

- Establish the relation between the radiotherapy dose received at a particular anatomical site during treatment for the first cancer, and the probability of observing a second cancer at the same site.
- Estimate with precision the carcinogenic effect of antimitotic treatments prescribed during the treatment for the first cancer and quantify the dose-effect relationship.
- Investigate the existing relationship between the first and second cancer.

We will study in priority:

- the shape of the second cancer incidence curve after radiotherapy alone, so as to test the additive and multiplicative risk models
- the relation between the radiotherapy dose received by the thyroid and the risk of developing a thyroid tumour, which is still not well known
- the effect of the chemotherapy-radiotherapy association
- the successive preferential appearance between two given cancer types, which may provide hypotheses about the existence of certain genetic mechanisms common to the origin of the two cancers.

Methods

The data for general and clinical characteristics as well as the doses and lengths of use of each chemotherapeutic agent will be collected by the paediatricians from each participating centre. For each drug, the dose will be expressed in milligrams.

The estimation of the radiation doses received by the children on the 160 different anatomical sites will be evaluated with the help of a program developed by the Department of Radiotherapy Physics at IGR and tested on a preliminary study.

These estimates will be given to the paediatricians, radiotherapists and physicists of the participating centres who may use them as they deem necessary. The program takes into account the characteristics of the machines, the individual conditions of irradiation, the bones and the lungs, and, particularly, the control films.

State of the study

The inclusion has begun at Institut Gustave Roussy (France), Institut Curie (France), Provincial Hospital of Barcelona (Spain), Childhood Cancer Registry (Oxford) and Thames Cancer Registry (Sutton). The inclusion of 5 new French centres is beginning.

At this time 1535 children have been included, of whom 63 have developed a second primary cancer.

Contribution of GSF

Late effects in Ra-224 treated ankylosing spondylitis patients.

This project comprises more than 1500 ankylosing spondylitis patients treated with repeated intravenous injections of radium-224. The usual injection scheme, which was applied until very recently, consisted of 10 to 12 injections of about 1 MBq each, given at weekly intervals. The resulting α-dose to the skeleton is 0.56 to 0.67 Gy for a 70 kg man. This is much lower than the doses applied to patients in the project of participant no. 11, corresponding to the objective of evaluating the late effects risk to humans for bone tumours and other lesions potentially related to α-emitters below that lowest dose of 0.9 Gy found in participant no. 11's study, where patients having received lower doses were not sufficiently represented. In addition, in this project, there exists a control group of about 1500 ankylosing spondylitis patients not treated with radioactive drugs or X-rays which was formed in order to provide comparative information on causes of death and lesions possibly related to the basic disease itself or to chemotherapy.

Up to now three cases of malignant primary bone tumours (according to the Histological Typing of Bone Tumours of the WHO) have been observed in the exposure group: one fibrosarcoma of bone, one reticulum cell sarcoma (malignant lymphoma) of bone, and one medullary plasmocytoma (multiple myeloma) originally observed in the bone marrow of sternum and pelvis. The expected number of spontaneous bone tumours for the present average follow-up time of 19 years is estimated to be 0.7-2.4 cases, determined from the age-dependent spontaneous rates for bone tumours from the three cancer registries of the Federal Republic of Germany. Observed diseases of the haematopoietic tissue among the study population included: bone marrow failure (12 cases in the exposure group vs. 6 in the control group and leukaemias (9 vs. 6). The increase of total leukaemias is, compared to a standard population, highly significant for the exposure group (9 cases vs. 2.7-2.8 expected, $p < 0.003$) and striking also for the controls (6 cases observed vs. 3.3-3.5 expected, $p = 0.14$).

Possibly the increase of leukaemias in the control group may indicate an effect of the mostly considerable intake of pain-killing or other drugs for the treatment of the basis disease. Subclassification of the leukaemias shows a clear preference for chronic myeloid leukaemia in the exposure group (3 cases observed vs. 0.8 expected, $p = 0.047$), whereas in the control group (1 case observed vs. 1.1 expected) the observed cases are within the range of expectancy.

For the coming years continued attention will be given to the evaluation of the diseases of the haematopoietic tissue, especially the myeloid leukaemias, in order to investigate a possible correlation with lower dose rates which has already been demonstrated for animals. Further attention will be given to bone tumours, kidney and liver diseases, and other lesions which have been known or supposed from the study of participant no. 11 to be elevated.

Contribution of the University of Munich (Dr. H. Spiess)

Late effects in Ra-224 treated juvenile and adult patients.

This project continues the study of patients that were treated in a German hospital shortly after World War II as juveniles for bone tuberculosis and as adults for ankylosing spondylitis.

The data, particularly for bone sarcomas, are largely complete, but additional recent observations on various other radiation effects are still being collected and special attention is given to the determination of these stochastic and non-stochastic radiation effects and their dose, time, and age distribution. In particular:

1. Continued inquiries from the registered cohort of radium-224 treated patients, from their physicians, and from medical institutions.

2. Regular individual medical reexaminations of the patients, investigation of the diseases that resulted after the radium-224 treatment. These investigations will be performed, as far as possible, by visits to the patients.

3. Evaluation of causes of death and diseases observed within the group of patients. Special attention will be given to the occurrence of benign and malignant bone tumours and also to soft tissue carcinomas. The increased prevalence of liver and kidney diseases, which has been observed so far demands added attention, as does the prevalence and evolution of cataracts (see project of participant no. 12).

4. Cytogenetic investigations will be extended. For 20 patients chromosome aberrations have been analysed, and controls will be examined.

5. A control group of patients not treated with radium-224 has been set up and they are now being examined according to the same procedure as the exposure group. The control tuberculosis patients were drawn from a file of a specialised lung clinic.

6. Statistical analyses of the relationship between the occurrence of late effects and the dose from radium-224 treatment as well as their timing are being continued in cooperation with participant no. 12 and with colleagues from NCI, Bethesda, USA.

Contribution of the University of Munich (Prof. Dr. A.M. Kellerer)

Epidemiology of radiation carcinogenesis and cataract studies.

The project will continue the epidemiological and statistical analyses of the results obtained in the studies by participant numbers 10 and 11. The modelling of the dose, age, and time dependence of the bone tumours has been completed in earlier work, and it is unlikely that additional cases will change the basic conclusions.

For the soft tissue tumours, especially for the breast carcinomas, the situation is different; substantial excess incidences are being seen now, especially for the breast carcinomas, and this necessitates a two-fold effort. One needs to re-examine the doses due to radium-224 to individual organs, and to relate these doses to the observed cases of soft tissue cancers. Secondly, it will be necessary to obtain information from control patients, especially from tuberculosis patients who were not treated by radium-224, but who are otherwise comparable, for example in the frequency of past X-ray diagnostic exposures. An important new component in the project is the technical preparation and continuing support of the evaluation of radium-224 induced cataracts among the ankylosing spondylitis and tuberculosis patients. A Scheimpflug-camera-system has been acquired and it is now being modified for electronic image storing and image processing to be utilised in this project.

The system will be fitted into a mini-van to permit the programme of repeated ophthalmological investigations with the Scheimpflug-camera without calling the patients to the ophthalmological clinic in Munich. In this way a broad cost-efficient system of inhouse-examinations or of examinations in nearby clinics will be possible. While all the ophthalmological work is done by Prof. Stefani and his colleagues from the ophthalmological clinic of the University of Munich, the present project will provide the technical support in terms of hardware development as well as imaging processing and evaluation. A suitable RISC workstation is available for the purpose, and essential parts of the software for the image processing have been prepared.

Contribution of the NRPB

Second analysis of the National Registry for Radiation Workers (NRRW).

After extensive consultations with interested parties, the National Radiological Protection Board (NRPB) set up the National Registry for Radiation Workers (NRRW) in 1976 to provide the largest possible epidemiological study of UK radiation workers. The essential prerequisite for a study of this type is a detailed and unbiased database describing the population to be studied; their eventual dates and causes of death; general factors affecting mortality (eg. age and sex); and the factor under investigation, namely ionising radiation. The assembly of such a database from records which were originally kept for other purposes is an exceedingly labour-intensive task. This has been completed for the 95,000 individuals in the first analysis of the NRRW - at least so far as their dose and employment histories up to 1986 and their mortality experience up to 1988 is concerned.

The mean lifetime dose is about 34 mSv, corresponding to a total collective dose of about 3200 man Sv. Many of these in the study population were born in the period 1945-59 so

these individuals were mostly in their thirties or forties at the end of the follow-up period (1988). This accounts for the fact that only about 7% of the study population had died by the follow-up date. It is clear that subsequent analyses, when more mortality experience has accrued, will have considerably more statistical power for making quantitative statements about the risks of exposure to ionising radiation. Furthermore, mean annual doses are falling with calendar period. This suggests that it is important not only to follow the existing cohort into the future, but also to add other groups who may have been employed in the earlier years when doses were higher. Both these extensions to the study group are planned.

The main findings of the first analysis of the NRRW (Kendall et al, Br. Med. J., 304, 220 (1992)) were as follows.

a) There was the expected Healthy Worker Effect. The Standardised Mortality Ratio (SMR) for all causes was 85 and the all cancer SMR was 86. Most cause-specific rates were also below 100.

b) There was a positive association between mortality from cancer and radiation dose, though the trend did not reach statistical significance ($p=0.10$).

c) There was a positive association between mortality from leukaemia (except chronic lymphatic leukaemia (CLL)) and radiation dose and this trend was statistically significant ($p=0.03$).

Estimates of the lifetime risks for cancers and for leukaemia (except CLL) can be made from the NRRW using projection models. In both cases the resulting risk coefficients are about twice the values estimated by the International Commission on Radiological Protection (ICRP). However, the 90% confidence intervals are very wide and include the ICRP values. Furthermore, a study of US radiation workers, albeit smaller than the NRRW, found no association between malignant disease and dose.

For the second analysis of the NRRW, NRPB will continue to liase with the nuclear plants and installations from whom data are collected and also with the organisations providing flagging information. The organisations include:

1) The National Health Central Register for England and Wales.

2) The General Register Office for Scotland.

3) Offices in Northern Ireland, the Isle of Man, Guernsey and Jersey.

4) The Department of Social Security Central Office.

5) The MOD Medical Statistics Unit.

6) The Royal Army Pay Corps Computer Centre.

7) Many Dose Record Keeping and Personnel Departments in the Nuclear Industry. The major organisations involved are:

Amersham International
Atomic Weapons Research Establishment
British Nuclear Fuels Limited
Defence Radiological Protection Service
Nuclear Electric
Rolls-Royce and Associates
Science and Engineering Research Council
Scottish Nuclear
United Kingdom Atomic Energy Authority

By extending the follow-up and adding other groups of workers, the second analysis of the enlarged NRRW database will have increased statistical power; it is estimated that 90% confidence intervals will be 20%-30% smaller. This analysis will thus provide more information on the magnitude of risks associated with occupational radiation exposure.

C14 Epidemiological studies in human populations.

Contract FI3P-CT930065 Risk estimates of lung cancer from the follow-up of uranium miners.

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Total Contribution by the Commission: 180 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Radon and its daughter products constitute the most important source of natural radiation exposure of the population. There are various animal data on the induction of lung cancer by radon and its daughter products, but neither these data nor epidemiological studies of domestic radon exposure can as yet provide quantitative risk estimates. Such estimates need to be based on studies of uranium miners exposed to higher levels of radon.

Data for major cohorts were recently reviewed in a document of ICRP (ICRP 50), and reevaluated by a special Committee of the US-National Academy of Sciences (BEIR IV). When the analyses of BEIR IV were performed, it was not yet possible to include in the computations the extensive data from the follow-up of the Czech uranium miners; these data could only be accounted for in summary form from publications based on simple computations. The evaluation of the original data with the same mathematical methods as used by the BEIR IV Committee remained therefore an important task.

This has been started by the group involved in project 1 in collaboration with the Institute of Epidemiology in Prague. The results of a preliminary analysis, although in essential agreement with those of the BEIR IV Committee, show important differences. It appears therefore necessary to complete the analysis by a comparison between models currently used for the Japanese data to those used for data from uranium miners. As a further important step the analysis would have to be extended to the very large study that has been initiated on the several hundred thousand miners of the Wismuth AG in Thüringen, Germany.

Accordingly our project has a two-fold aim. One objective is the comparison of models to be used for the analysis of such studies, their application to the cohorts of Czech uranium miners (project 1 and 4), to the cohort of French miners (project 2), and to the miners of the Colorado Plateau (project 3). This includes the examination of confounding factors such as time after exposure or duration of mining. A special effort will be done to quantify the influence of smoking on the basis of the data from cohorts L and N of Czech miners (project 4). Second aim of the project will be the joint analysis of two cohorts of the French and the S-cohort of Czech uranium miners as a preliminary step to the impending analysis of the cohort of the Wismuth AG.

Contribution of GSF - Neuherberg

Models applicable to the risk of radon exposures and analysis of cohorts of uranium miners.

Our group has closely collaborated in recent years with the Institute of Hygiene and Epidemiology in charge of the follow-up of the Czech uranium miners. A detailed description in terms of illustrative diagrams and an analysis based on the relative risk model have been concluded. This analysis is largely comparable to the earlier studies of other major cohorts of uranium miners by the BEIR IV Committee, but it leads to a number of specific and partly different conclusions.

We propose for the impending research period to extend this analysis by a comparison of the age attained model with models currently used for the analysis of the Japanese data, i.e. relative risk models with an age at exposure dependency. A notable result of our study has been the apparent non-linearity of the excess lung cancer mortality in its dependence on exposure, and there has also been an indication of a substantial influence of the duration of mining. The analysis will therefore include a detailed examination of the influence of duration of exposure and of non-linearity in total cumulated exposure. In particular the influence of further confounding factors will be examined.

A subsequent step will be the joint analysis of the data from the Czech S-cohort together with the French cohort of uranium miners. This will be the preliminary step for a later analysis of data from the important new cohort of uranium miners from the Wismut AG. The work in this project should contribute tools required for the evaluation of data from the Wismut AG.

With these new data, as soon as they will become available, it is planned to perform a case-control study with the models developed for the Czech miners. The computations would be done with the software package EPICURE which has already been used with the data from the S-cohort.

Contribution of IPSN Centre d'Etudes Nucleaires of Fontenay-aux-Roses

Contribution of the French uranium miners cohort to the Modellisation of Lung Cancer Risk due to exposure to Radon and its decay products.

A cohort of French uranium miners has been followed since the date of first exposure to radon to the 31st of December 1985. The Poisson regression modelling, applied to the mortality by lung cancer, expressed as a relative risk, in function of the cumulated exposure to radon, indicates a positive linear trend, with a risk coefficient of 0.6 % per WLM (working level month). The results of this analysis confirm the linear trend observed in other uranium miners cohort (Colorade, Ontario..), but may differ in several points from the CSSR cohort.

In the two following years the objective is to define and apply a common approach in the analysis of the European cohorts of uranium miners studies, mainly in order to estimate more precisely the risk linked to low chronic exposures spread over a long working period. The French uranium miners are characterized by low annual exposures in comparison to the CSSR miners (project 1) or to Colorado miners (project 3). The results presently available may show differences that are not necessary incompatible. These differences may be linked to diffferent modelling or to different exposure or background conditions; age attained or time since last exposure may also interfere.

We intend to use this common approach with epidemiologists from France (CEA, INSERM.), from England (NRPB), and Germany (GSF), directly involved in studies of chronic exposure to ionizing radiation with time dependent risk factors.

The statistical analysis should include features specific to the various cohorts. Once the conditions for a correct comparison of all the data are established, a joint analysis can be performed.

Contribution of the NRPB

Methods and Analyses for Uranium Miner Data

There are a number of uncertainties concerning the risk of lung cancer among miners exposed occupationally to radon. These include the temporal pattern of risk, which affects estimates of the risk over a lifetime, and the joint effect of radon and smoking, which affects estimates of how the radon-associated risk is distributed amont smokers and non-smokers. Furthermore, there has been recent interest in the possibility of an "inverse exposure rate effect", ie. whether the risk per unit exposure increases with decreasing exposure rate (Darby and Doll, Nature 344, 824 (1990)). There are also some methodological issues associated with the analysis of data on miners, related to the fact that exposures are received over a period of time rather than instantaneously. It is intended that NRPB will provide an input concerning the methodology for analysing data on uranium miners and the modelling of radon-induced lung cancer risks. In relation to this, data on Colorado Plateau (US) uranium miners will be analysed.

Contribution of the Imperial Cancer Research Fund - Cancer Epidemiology Unit

Project to be carried out in conjunction with Dr. Emil Kunz and Dr. Ladislav Tomasek of the Institute of Public Health, Prague.

Over the last six months, we have started working collaboratively with Dr. Ladislav Tomasek of the Institute of Hygiene in Prague on new analyses of mortality in the major Czech uranium miner cohort started by the late Dr. Josef Sevc. This work forms an extension of the work recently carried out within project 1 jointly with the Prague Institute. Six major aims have been identified:

i) Extension of the follow-up of the first cohort of miners first employed between 1948 and 1957 S cohort) from 1985 until the beginning of 1990, and improvement in the quality of the follow-up. Investigation has revealed that the existing method of follow-up, which relied on Ministry of Interior records, missed some deaths, especially in the period since 1980. A new exercise, using more reliable follow-up sources, is now under way to provide rigorous checks on the vital status of each individual.

(ii) Improvement of the quality of the exposure data. A review of the estimated radon doses is being carried out, using all available sources of information, and information on exposure to arsenic is also being compiled. Initial results show that radon doses may be changed considerably for the early years of the study, and that substantial exposure to arsenic has occurred.

(iii) Extension of the cause of death coding to enable mortality from a wide range of cancers and other diseases to be examined. This will allow investigation of recent suggestions that radon may be the cause of cancers other than lung cancer and also to

evaluate mortality from a range of non-neoplastic diseases.

(iv) Use of standard modern analytical methods for the analysis of the risk of lung cancer and other diseases, to give rigorous analysis directly comparable with those from other cohort studies, and directly interpretable in relation to possible residential risks. This aspect of the project is a continuation and extension of the work already carried out in conjunction with GSF.

(v) Analyses of lung cancer risk in relation to histology in the S-cohort using data already collected, and investigation of the possibility of obtaining further histological data.

(vi) Investigation of the status and potential of two other radon exposed cohorts of miners which have not been formally analyzed before: recent uranium miners first employed before 1968 (N cohort) in conjunction with Dr. Vaclav Placek, and the cohort of burnt clay miners (L cohort) in conjunction with Dr. Alena Heribanova. Both of these cohorts include information on cigarette smoking.

The objective of the work to be carried out under this proposal is the completion of this extensive programme of work. The proposal is complementary to the co-operation of GSF with the Institute of Public Health in Prague, and adequate interfacing between the three institutes will be established.

C14 Epidemiological studies in human populations.

Contract FI3P-CT930066 International collaborative study of cancer risk among nuclear industry workers.

Coordinator IARC
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Total Contribution by the Commission: 150 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Risk assessment is mainly based at present on epidemiologic studies of subjects receiving moderate or high doses of ionizing radiation over a short time period and on large, long-term animal carcinogenicity experiments. The use of these results to set protection standards for low dose chronic exposure requires application of controversial extrapolation models which cannot be validated on the basis of these data alone. Epidemiological studies of populations exposed chronically to low doses of ionizing radiation are necessary to assess the adequacy of extrapolation models and to guide radiation protection practice. Populations of workers occupationally exposed to ionizing radiation in the nuclear industry are ideal for this purpose because of the detailed dosimetric and employment records which are kept in this industry.

The current contract concerns the setting up and coordination of an international collaborative study of cancer risk among nuclear industry workers from at least 11 countries (8 from Europe, including 6 EEC member states) expected to last five years, until March 1988. The specific objective of the study is to provide data on the effects of low dose chronic exposures to low LET ionizing radiation on risk of all forms of cancer, and particular types of cancer defined a priori, for comparison with the risk estimates derived from the high dose/high dose-rate studies. This will provide a direct test of the adequacy of the current extrapolation models used for risk assessment and for the setting of radiation protection standards, and may assist in the construction of improved risk assessment models.

The first three years of the project will mainly be focused on collection and validation of data. In the first six months, detailed procedures documents and detailed questionnaires will be prepared at IARC. They will be reviewed at meetings of the Epidemiology and Dosimetry Sub-committees. Following this, construction and collection of entry data on cohorts and follow-up for death and cancer morbidity will be carried out at the national level.

Contribution of IARC

The coordinator will provide expertise in the management of large international collaborative projects, especially in relation to cancer risk in occupational settings and quantitative risk assessment. The coordinator will also provide expertise in the statistical analyses of cancer data and on the biological effects of low level radiation. The coordinator's functions will be:

1) to take the responsibility for the coordination of the international study;

2) in collaboration with national study group members, to provide guidelines on the abstraction of data from personnel records in the various countries;

3) to assist participating countries in the supervision of data collection in individual facilities for employment and exposure history;

4) in consultation with participating countries, to provide guidelines for checking the accuracy and the completeness of the data collected from facilities and of the follow-up procedures;

5) to take responsibility for carrying out statistical analyses of the data of the international cohort in consultation with the study group;

6) whenever necessary, to provide statistical and programming assistance at the national level;

7) to organize meetings of the study group, to discuss on-going problems of the study, results, preparation of reports and publications;

8) to select further countries to participate in the study, and provide a link among the participating countries between the meetings of the study group;

9) to maintain the international database;

10) to coordinate the publication of results of the international study;

11) to keep national investigators informed of study progress via national study group members.

The management of the collaborative study is by means of regular contact with participants. This implies written or telephone correspondence; meeting of the study group as necessary (at least every two years during the conduct of the study) to discuss its developments, approaches to the analyses and the interpretation of results as well as to prepare the publications; site visits when appropriate during the data collection and data analyses phases. The list of participants (members of the Study Group) responsible for the basic data collection in the participating countries is attached.

C2 Optimization and management of radiation protection

C2 Optimization and management of radiation protection

Contract FI3P-CT920004 Development of fundamental data for radiological protection.

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Total Contribution by the Commission: 100 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The primary aim of radiological protection is to provide an appropriate standard of protection of man without unduly limiting the beneficial practices giving rise to radiation exposure. To achieve this end, it is necessary to quantify the detriment associated with exposure and to develop a general policy of protection of exposed populations and for individuals.

Much of the work of the International Commission on Radiological Protection (ICRP) in the next few years will be a consequence of the new basic recommendations in radiological protection, published in ICRP Publication 60 (1990 Recommendations of the ICRP). One immediate effect was the publication of new interim Annual Limits on Intake of Radionuclides by Workers (ICRP Publication 61) based upon the new recommended radiation and tissue weighting factors.

Task Groups within the four standing committees of the ICRP will provide updated information on radiation-associated risks of cancer and severe hereditary effects; on the application of the basic recommendations in terms of secondary limits; and on the optimisation and management of radiological protection.

Work in progress and expected achievement under the period of the contract

It is envisaged that progress will be made in several important areas in relation to radiological protection.

A Task Group on Risk Estimates from the Committee on Radiation Effects is now considering the uncertainties in the present ICRP risk estimates which are based primarily upon the Life Span Study of the Japanese survivors of the atom bombings in 1945. They also intend to consider other sources of epidemiological data to supplement that from the Life Span Study.

Working Parties have been formed to consider the appropriateness of risk projection models and the method of transfer of estimated risks from one population to another; and to examine the information available from Soviet epidemiological studies.

A Task Group has been formed to study genetic susceptibility to cancer. Its terms of reference include a study of the mechanisms of radiation oncogenesis with respect to mutations; frequencies of gene mutations in human populations; interaction between genetic and environmental factors; and the role of genetic factors in cancer risk.

Other topics under consideration by the Committee on Radiation Effects are to review the effects of radiation on the developing central nervous system and the influence of radiation on the incidence of multifactorial hereditary diseases. A report of the Task Group on Age Dependent Dosimetry from the Committee on Secondary Limits will be published as Part 2 of ICRP Publication 56 (Age Dependent Doses to Members of the Public from Intakes of Radionuclides).

Doses per unit intake following ingestion will be given for isotopes of sulphur, cobalt, nickel, zinc, molybdenum, technetium, silver, tellurium and polonium using the new tissue weighting factors. New age-specific biokinetic models for the alkaline earth elements and lead, and for plutonium, americium and neptunium will be recommended for use in calculating dose coefficients. Dose coefficients following inhalation of these radionuclides are not included, but they will appear in a future publication, together with updated values for those radionuclides published in Part 1. Future publications will provide dose coefficients on the elements antimony, uranium, thorium and iron; as well as doses to the embryo and fetus following chronic intake of radionuclides by the mother before conception and during pregnancy.

A report on a Human Respiratory Tract Model for Radiological Protection will be published. This model updates that used in ICRP Publication 30 (Limits for Intakes of Radionuclides by Workers), based upon increased knowledge of the anatomy and physiology of the respiratory tract and of the deposition, clearance, and biological effects of inhaled radioactive particles.

The new model will:

- provide calculations of doses for members of the public, in addition to workers;
- be useful for predictive and assessment purposes as well as for deriving limits on intakes;
- account for the influence of smoking, air pollutants, and respiratory tract diseases;
- provide for estimates of respiratory tract tissue doses from bioassay data; and
- be equally applicable to radioactive gases as well as to particles.

The workload of the Task Group on Dose Calculations has been increased and will in future be shared between the US Oak Ridge National Laboratory, UK National Radiological Protection Board and the German Bundesamt für Strahlenschutz. The workload of the Task Group on Reference Man has been reviewed and effort will be directed in the foreseeable future to identifying anatomical and physiological parameters in Caucasian man (with an appendix on non-Caucasian man), with special reference to the skeleton, gastrointestinal and respiratory tracts and the haematopoietic system.

A joint Task Group has been established with the International Commission on Radiation Units and Measurements (ICRU) to clarify the conceptual differences between the protection quantities defined in ICRP Publication 60 and field quantities defined by ICRU. The Task Group will also provide fluence to effective dose calculations for a variety of radiations and energies for different ages and fluence to field quantity conversion factors.

The objectives of other Task Groups and Working Parties of the Committee on Secondary Limits will be modified or extended in order to accommodate the ultimate revision of ICRP Publication 30.

A report on Radiological Protection in Biomedical Research will be published which defines the criteria for the implementation of radiological protection principles for patients and volunteers exposed to ionising radiations in the course of the research programme.

An addendum to ICRP Publication 53 (Radiation Dose to Patients from Radiopharmaceuticals) will also be published, providing information on the biokinetics and calculated tissue doses following the intake of six new radiopharmaceuticals now in common use.

The Committee on Protection in Medicine will consider the advice to be given on optimisation of protection of patients in diagnostic radiology and on the hazards of potential exposures of patients due to mechanical or human failures.

Three reports from Task Groups of the Committee on Application of the Commission's Recommendation will be published.

The first report on Protection from Potential Exposure will provide a conceptual framework upon which to apply radiological protection principles. There will be recommendations for annual probabilities from which constraints on potential exposures might be chosen. This important document forms an initial bridge between the radiological protection and nuclear safety philosophies, while giving practical guidance that can be used for much simpler sources.

The second report on the Principles for Intervention for Protection of the Public in a Radiological Emergency will update and extend the advice given in ICRP Publication 40 (Principles for Protection of the Public in the Event of Major Radiation Accidents), particularly in the application of Publication 60 principles to control of contaminated foodstuffs and relocation of population groups.

A Task Group has been formed to produce recommendations on application of the principles for the restriction of radon exposure in homes, above-ground workplaces and in mines. The report from this task group will be an amalgam of work done previously which considered the restriction of exposures to radon in homes and above-ground workplaces and on limits for exposure to radon in mines.

C21 Optimisation of radiological protection.

Contract FI3P-CT920033 ALARA in installations.

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Total Contribution by the Commission: 200 KECU
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Description of research work

INTRODUCTION

One of the fundamental principles underlying radiation protection, since ICRP 26, is that all exposures shall be kept As Low As Reasonably Achievable. This principle is an explicit requirement of the CEC Directives laying down the basic safety standards for radiological protection (EURATOM Directive). The new ICRP 60 recommendations have emphasised the role of this principle not only for collective dose but also for the number of people exposed and the distribution of individual doses; it has also introduced the concept of constraint within the optimisation framework.

The first joint project in ALARA (1986-1989), involving NRPB (UK) and CEPN (France) focused mainly on fundamental concepts and ALARA methodology. The end product was the book "ALARA from theory towards practice". The second CEC / NRPB-CEPN contract (1990-1991) began with the development of ALARA programmes and ALARA tools. As far as "ALARA in Installations" is concerned, the main objectives of this phase were the development of : - a software for performing simple cost effectiveness analysis (CEPN), - a feasibility study for setting up an accident/incident data base in the non nuclear industry (NRPB), - a generic procedural guide for implementing ALARA during normal operation of installations (CEPN). The content of this guide has been tested and improved during the four ALARA CEC Training Courses organised at Saclay in 1990, 1991 and 1992, jointly by NRPB and CEPN under the auspices of CEC.

OBJECTIVES

The next period (1992-1994) will be devoted to a larger and more detailed programme. Four teams (CEN MOL, CEPN, GRS, NRPB) from four CEC member states (Belgium, France, Germany, United Kingdom) have been selected for this joint project. The area of research will cover : - the setting up of Programmes of optimisation of radiological protection in nuclear and industrial installations from the design stage up to the decommissioning of the facility, - further development of tools and software, - as well as transverse themes such as the interface between design and operation, and the interface between radiation protection and safety requirements. This joint project will also take into consideration the impact of ICRP 60 recommendations.

The next CEPN/NRPB CEC Training Courses on ALARA will include Belgian and German lecturers as a means of presenting ideas and obtaining feedback on the new CEC Joint Projectdevelopments. Twice a year, the group will meet to discuss specific topics of common interest (such as dose constraints), and each participant will act as national contact to provide data from his own country on dismantling, inspections, work management, etc.

DETAILED DESCRIPTION OF TASKS

Integration of ALARA at the design and operation stages

The initial study developed the role of ALARA in design, beginning with the optimisation of radiation protection for the workers within a new French fuel fabrication facility (the MELOX plant). This study will be developed in order to propose a generic guide to help in defining ALARA design objectives, criteria and procedures. As far as work management is concerned, a model will be developed, assessed upon some practical experiments and surveys performed within the French nuclear industry.

Integration of ALARA at the decommissioning stage

During the previous period, decommissioning was not in the ALARA research programme. Even though dismantling is comparable with some big maintenance operations, it presents many specific concerns such as the amount of wastes to be managed, and the role of the "time schedule", as there is no critical path. Therefore, the objective will be to develop a generic ALARA Programme for the decommissioning of installations as well as a European feedback data base structure. This will be mainly performed using the experience of the BR3 PWR decommissioning programme.

Development of tools, methodologies and databases

* The aversion, equity and prudence concepts analysed previously (1990-1991) in the valuation of the man-Sievert for exposure of public and workers associated with normal operation will be integrated at the implementation level. Within this framework, coherent with the new ICRP 60 recommendations, the use of discount rate, "return rate", as well as "time in general" will be considered.

* The software package for performing simple and extended cost-benefit analysis will be completed; in order particularly to take into account the new framework recommended in ICRP 60 recommendations.

* Methodologies will be developed for the derivation of dose and risk constraints, as defined by the ICRP 60 recommendations, and how they can be integrated into radiation protection programmes.

* A database for recording information on failures and accidents will continue to be developed.

Interface between Radiation Protection and Safety

A pilot study, mainly focused on some German plants, will evaluate the dose contribution of testing and inspection tasks in Nuclear Power Plants, in order to optimise doses with respect to the necessity and the frequency of these tasks.

CEPN PARTICIPATION

In the course of 1990 and 1991, CEPN has been actively involved in a project aiming at optimising the radiological protection for the workers within a new fuel fabrication facility under construction in France (the MELOX plant). This project delineates the key issues about the implementation of the ALARA principle at the design stage. This study is being developed in collaboration with the French Nuclear Industry (EDF, COGEMA) in order to propose a generic guide to help in defining ALARA design objectives, criteria and procedures. CEPN is also implementing, in cooperation with EDF and FRAMATOME, an overview on the impact of work management factors on ALARA achievement. A model based on surveys and practical experiments will be developed.

CEPN will integrate, at the implementation level, the aversion equity and prudence concepts analysed previously (1990-1991) in the valuation of the man-Sievert for exposure of public and workers associated to normal operations. Within this framework, the use of discount rate, "return rate", as well as "time in general" will be considered. Feedback from the nuclear industry, as far as radiological protection actions management is concerned, will be of considerable interest as input for this study.

In conjunction with NRPB staff, CEPN will complete the software package for performing simple and extended cost-benefit analysis, in order particularly to take into account the new framework recommended in the ICRP 60 publication and the developments in valuing the man-Sievert.

SCK / CEN MOL PARTICIPATION

The question of dismantling of nuclear installations is of current interest, as many of the first generation nuclear power plants are coming near to the end of their operational life. Dismantling operations are very specific, differing in various aspects from the normal maintenance operations in nuclear reactors. As a consequence, the radiological optimisation of dismantling operations also demands a specific, adapted approach, which requires much R&D concerning the factors typical to dismantling.

Important radiological protection factors (in the meaning of ICRP 55 recommendations) for the optimisation of dismantling operations are situated in the domains of - the exposure of workers, - the waste generation and disposal, - long-term risks for the population, - high monetary costs of protection.

As much as possible is to be learnt from specific operations in pilot projects. One of such pilot project is the BR3 pressurised water reactor pilot dismantling project at the

SCK / CEN. Apart from this, the SCK / CEN intends to discuss specific aspects of dismantling operations in a generic way. Typically, these aspects will include the long time scales and trade-offs related to different timings of certain operations or interventions such as trade-offs between radioactive decay of contaminations or wastes and degradation of equipment, operation of installations and institutional memory.

The programme of work will be mainly focused on four topics :- an inventory of typical decisions for decommissioning relevant to Radiological Optimisation, - a time scale model for describing decision factors evolution such as costs, doses..., - a description of an ALARA data base structure for decommissioning, - proposals for specific ALARA decision-aiding tools.

GRS PARTICIPATION

In the Federal Republic of Germany, on the average of all PWR's, 40 to 50 % of the annual collective dose of the personnel is caused by recurrent tasks such as tests, inspections, etc.

Considering the development of minimisation and ALARA concepts, it is worthwhile to evaluate the dose contributions of these activities and to optimise the doses with respect to the necessity and the frequency of the tasks to be carried out. In this case, the influence of testing and inspection of the plant safety during operation must be considered..

As a base for these evaluations, the doses of the special tasks in the above mentioned fields, the dose rate at the relevant places, the frequency of the tasks and information about the failure rate of the systems considered as a function of test frequency, will be collected in a pilot study carried out at two old plants: one PWR and one BWR. The German data will be, as far as possible, put into perspective with British, Belgian and French ones.

NRPB PARTICIPATION

The NRPB would further develop the use of tools such as the ALARA Procedure, ALARA reviews and Predictive ALARA Plans within the broader and important contexts of Work Management, Organisational Structures and Commitment. In conjunction with CEPN staff, NRPB staff will also assist in the completion of the software package for performing simple and extended cost-benefit analysis in the optimisation of radiological protection.

ICRP's definition of optimisation of radiological protection in Publication 60 has explicitly introduced the concepts of dose and risk constraints. Work would be carried out to develop the methodologies for the derivation of constraints and how they can be practically integrated into radiation protection programmes. Particular attention will be given to the use of generic optimisation as a means of deriving constraints and the interactions between constraints and design targets.

The explicit extension of the ICRP Recommendations to cover potential exposures necessitates further consideration of how the ALARA concept can be fully implemented into the design of safety system in industrial facilities. It is recognised that there is inadequate information on failures of various safety systems such as interlocks and in the first instance NRPB staff will continue the development of an appropriate database to record information on failures and accidents. This could eventually provide details on where problems exist for the European Community as a whole leading to improvements in design.

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920014 Digital Medical Imaging: Optimization of the dose for the examination.

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Total Contribution by the Commission: 130 KECU
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Description of research work

1. Summary of Background Information:

During the last 2 decades major innovations in Fluoroscopic and Digital Medical Imaging Equipment occurred. The associated clinical applications include Digital Cardiac, Vascular Imaging and new approaches to traditional Barium Studies. In Europe the population dose from the latter alone is comparable with that from CT. Taken together the population doses from Fluoroscopic and Digital Techniques provide the largest single contribution from diagnostic medical sources. This project aims to bridge a major gap that has opened up between the rapid and comprehensive development of the equipment and it's clinical applications on the one hand, and the underdeveloped condition of the necessary support studies in Dosimetry, Quality Assurance and Optimization on the other.

The need for dosimetry and optimization studies is further exacerbated by the recommendations of ICRP-60, which have altered the basis for risk estimates in situations involving irradiation of the abdomen and thorax.

The project addresses the Dosimetry, Quality Assurance and Optimization issues raised above. The available Patient and Staff Dosimetry literature has serious gaps for the newer clinical techniques, and many of the available assessments are for older equipment which are unlikely to be valid for the present generation of equipment. These questions, and those arising from ICRP-60 will be refined and addressed. With regard to Quality Assurance, a set of techniques/protocols suitable to the technology and it's application will be pioneered.

The approach involved will be dominated by objective measurements such as Signal to Noise Ratio, Modulation Transfer Function etc., but semi-subjective methods will also be employed as required. Tolerances of the measurement method, Limiting Values of the technical parameters and Recommended Performance Levels will be investigated for four situations: Acceptance Tests, Quality Assurance, Routine Constancy Checks and Equipment Write-Off. While the available relevant literature on Dosimetry and Quality Assurance is limited, that on dose reduction and optimization is almost non existent. Hence a series of Optimization Studies is proposed both at the level of the Physics/Technology and it's Clinical Application. The later will include a number of studies of the optimum dose for specific clinical applications using specific techniques.

2. General Framework for Objectives:

This project is an extension and development of a former contract. It has the object of correcting a serious imbalance that has occurred in the application of new digital and fluoroscopic radiological technologies. On the one hand technical and clinical development has been rapid while, on the other, the associated development in dosimetry, quality assurance and optimization have been at best patchy.

The general objectives and longterm aspirations are summarized in the following points:

* Gain a clear insight into the underlying philosophy of the automated/software controlled mechanisms of exposure selection used in all modern systems.
* Identify and recommend Optimized Exposure Levels for Specific Technologies and Imaging Tasks. These might, in turn, be adopted in the automated systems.

* Develop the medical and the technical aspects of a set of proposed protocols for digital and fluoroscopic imaging equipment that will operate at 4 levels. They are: Acceptance Testing; Periodic Regular Quality Assurance; Frequent Constancy Checks; and Write-Off of the Equipment. The latter is particularly important and is essential to compliance with the Patient Directive of the European Community. The protocols will concentrate on whole systems, though in many cases subsystems will have to be studied also.

* Identify and define the parameters to be assessed and the appropriate norms, tolerances/limiting values for each protocol. The parameters/methods of measurement will concentrate on objective approaches, although semi-subjective approaches will also be used where necessary.

* Determine, in optimization studies, how much dose can be usefully employed by specific imaging devices. The effect of increasing or decreasing dose will be determined using objective image quality endpoints such as Signal to Noise Ratio. These studies will be extended to look at specific features of phantoms.

* Perform Optimization Studies of Clinical Problems such as those in Digital Vascular, Cardiac, Gastrointestinal, Orthopaedic and Chest Imaging.

* Address the specific Patient and Staff Dosimetry issues raised by new technology, ICRP-60, the new types of examination being undertaken, and the requirement for guideline/dose-constraint values.

* Address the question of the ICRP-60 dose limit to the abdomen of Pregnant Workers in selected Medical Imaging Procedures to see if it provides adequate protection for the foetus.

* Study of Multiple Modalities of Chest Imaging to determine the most effective in the Optimization context.

* Develop approaches to facilitate transfer of results from one location/equipment supplier to another, so as to allow the optimization results produced to be propagated in practice.

* Identify approaches to software QA appropriate to Digital Imaging Technology, including development and use of On-Line KBS based systems.

* Organise expert meetings to develop consensus on the solutions reached and other meetings/educational activities to propagate the results produced.

The list is not exhaustive, and in many cases the results produced will have to be brought forward eventually in documents or protocols expected to guide practice, and provision is made for this in the planning of the project.

3. Specific Objectives:

The work in this contract will be conducted within the above framework. The main tasks to be performed in the area can be identified under four headings:

PROJECT 1: Optimization of Dose/Factor selection in Digital and Fluoroscopic imaging Systems.

PROJECT 2: Quality Assurance, Acceptance and Write-Off of Digital and Fluoroscopic Systems.

PROJECT 3: Patient and Staff Dosimetry with Digital and Fluoroscopic Systems.

PROJECT 4: Optimization of Patient Examinations with Digital and Fluoroscopic Systems.

In addition, progress in the area as a whole will require significant progress on specific items under each heading, and is expected within the contract. The project also expects to contribute to addressing the need for a continuing series of expert meetings and training.

Within the contract, the work already initiated under each of the above project headings will continue. However, the work it will be possible to perform is necessarily limited and in deciding which areas to give priority to a number of criteria have been adopted including:

- No work involving Interventional Radiological Procedures will be performed within the contract.

- Dosimetry and Optimization Studies will be directed towards examinations with a high frequency.

- Dosimetry and Optimization Studies will be directed towards examinations with a high individual dose.

A further criterion for the selection of the studies to be performed is that they should have a high probability of success in their own right, or that the intermediate knowledge they generate should be likely to be able to be used as technical models for further studies within this or other projects. Finally, in areas where knowledge is sparse, particularly in respect of Optimization of the newer technologies, it will continue to be necessary to undertake some more fundamental work within the areas of Physics, Technology, and Dosimetry, with a view to preparing the ground for Clinical Optimization Studies. Such studies are likely to be particularly enlightening and to contribute to the knowledge base necessary for the above applied studies. Within this framework studies will be performed with a view to:

- Gaining knowledge of the best way to describe the image information content/quality in respect of the examination selected.
- Conduct of patient and staff dosimetry including the necessary developments with respect to quantitative units and methodology.
- Definition of the criteria for optimization of the above.
- Developing a Quality Assurance Programme for a limited range of equipment within the above categories.
- Segmentation of the Quality Assurance Programme into levels that correspond to Scientific, Technical, Clinical and Managerial Reality.
- Development of the scientific information from which technical criteria for the write-off of equipment may be brought forward.

It is expected to carry out all of the above work within the project management arrangements which have already established their worth.

Dublin

The work within the Dublin Group will be carried out in the framework of the preceding overview. However, the work within the Group will concentrate mainly on:

Project 1: Optimization of Dose/Factor selection in Digital and Fluoroscopic Imaging Systems.

Project 2: Quality Assurance, Acceptance and Write-Off of Digital and Systems.

From the Mannheim meeting it appears useful to define and segment Quality Assurance(QA)/Quality Control (QC) into a series of levels corresponding to, for example: (a) acceptance/write-off/rigorous quality control measurements; (b) routine constancy checking; (c) external audit, and so on. Much work in this area is confused, and a function of this project will be to clearly define the work and methodology appropriate to each level.

Within the project, work will be continued on (a) the characterization of the range of automated control systems (ACS); (b) the mechanism of operation of ACS (c) the performance of these devices; and (d) the development of an optimization protocol for these devices. Where possible these protocols will be brought forward in a form that allows them be applied to other studies.

Criteria for Image Quality appropriate to the different segmented levels in QA and QC will be defined, and where possible performance limits with respect to the types of examination, or a limited range of equipment for these devices will be identified with a view to applying them practically as set out below.

Optimization Studies permitting the optimization of entire systems and of each component within the system will be undertaken on a highly specific research basis, with a view to modelling this process for more general applications where standards, performance limits, tolerances, and limiting values may be applied. This approach will include some theoretical modelling.

Work in support of the Dosimetry Studies to be undertaken within Project 3 and 4 will be performed. This will include an approach to gastrointestinal dosimetry based on the analysis of video tapes of examinations conducted by different Clinicians.

Work to develop Image Quality Criteria for Clinical Studies that come within the ambit of those defined in the overview will be initiated. The most probable candidate for such studies will be Invasive Diagnostic Cardiology, not involving interventional procedures.

Work will be continued on establishing write-off criteria. This will include the development of performance limits, desirable and achievable performance levels, and the segmented approach to QA identified above.

Newcastle

The Group will co-ordinate the various patient and staff dosimetry studies performed by the consortium. These studies will be undertaken to establish baseline data for a range of common fluoroscopy or digital imaging procedures on both adults and paediatric patients.

Particular emphasis will be placed on gastro-intestinal studies in which digital imaging techniques will be compared with other modalities. The optimization of personal protection during fluoroscopy will be established. In another study, in collaboration with the Mannheim Group, the correlation of personal doses with dose-area product will be investigated.

A theoretical model to predict the contrast-detail dose characteristics of any imaging system will be refined. This model will be adapted to take into account quantitative image quality parameters, imaging technique and observer viewing conditions. A trial will be performed to establish the accuracy of the model in a series of experiments on fluoroscopy and digital imaging systems using a set of quantitative optimisation test objects. Comprehensive optimisation investigations will then be pursued on equipment located at the three participating centres.

Measurement protocols for the assessment of dose levels at the entrance surface of the patient and the input receptor of fluoroscopy and digital imaging systems operating under automatic exposure control and automatic brightness control will be developed in collaboration with the Dublin Group. A large scale international survey will be performed to establish the operating levels on both new and existing equipment.

Further investigations in collaboration with the Dublin Group will be performed to study the important practical problem of writing-off old imaging equipment. A multi-centre survey of the imaging performance of fluoroscopy systems will be undertaken to establish guidelines values. The results of previous attempts at writing-off old fluoroscopy equipment will be reviewed with the intention of formulating minimum performance standards.

Mannheim

The main topics of the Mannheim group will be the optimization of exposure parameters for new digital imaging methods (storage phosphor radiography, image intensifier

radiography). In addition to the selection of suitable and unsuitable clinical indicators for these new imaging methods (in comparison to conventional film/screen radiography) the preselectable dose values have to be chosen as low as possible to fulfil the requirements of necessary image quality. This optimization has to be done for imaging of the skeleton, lung, gastrointestinal tract and angiography (DSA). Comparative dose measurements (patients and staff) have to be done for patient examinations and phantom studies. The aim of this procedure is to give common recommendations for the selection of the optimal imaging method and exposure parameters depending on the clinical situation.

There are now five analog and digital methods available for chest imaging (conventional film/screen, storage phosphor, AMBER-system with film/screen, AMBER system with storage phosphor, digital image intensifier radiography). Further clinical and phantom studies have to be performed to determine the clinical value of these methods.

Image quality criteria, which already exist for conventional film/screen radiography have to be applied to digital radiography. These criteria have to be adapted to the new imaging capabilities of digital radiography (e.g. postprocessing). The aim of this project is to define new modified criteria for these new imaging techniques.

We gained experience with constancy tests for digital image intensifier radiography in the 1990-1992 period. The development of constancy tests for storage phosphor systems should be a subject of this new programme.

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920020 Quality assurance parameters and image quality criteria in computed thomography.

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Total Contribution by the Commission: 180 KECU
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Description of the research work.

The steady increasing use of computed tomography as a general tool in diagnostic radiology seems not to weaken, and recent developments in technology makes this high dose modality even more attractive. Computed tomography allows for better diagnosis than conventional x-ray, and, in a considerable number of indications, has completely replaced plain and contrast radiographic studies. In many clinical situations, it can be necessary to use high values of mAs and thin slices, thus increasing the dose to the patient. Special concern is related to the use of computed tomography in paediatric radiology where recent information emphasize that in children there is an additional hazard of detrimental radiation effects. CT-examinations on infants and children have yearly increased in frequency in spite of the availability of ultrasound and magnetic resonance imaging. Information of the use of CT in infancy and childhood will be produced from appropriate survey questionnaires especially adapted for paediatric patients on an Europe-wide level.

Image quality in computed tomography is affected by many parameters, some of which are called scanning parameters and can be controlled by the operator and should be selected using clinical and anatomical considerations about the examinations and patient in question. Important parameters are KV and mAs settings, slice thickness, number of slices and table increments, field of view and matrix and the type of reconstructive algorithms. The effect of these parameters on image quality and patient dose will be assessed quantitatively by phantom measurements and is essential information for the quality criteria be formulated on the clinical information wanted in the images.

The complexity of the technology and the variability of scan parameters results in higher numbers of possible quality reducing factors in computed tomography than in conventional radiology. For investigations of the unnecessary high doses to the patients general acceptable image quality criteria for the most common examinations are needed both for adult and paediatric patients. Such criteria will be tested in clinical practice and their influence on patient doses demonstrated by calculations of organ doses and the effective doses compared with the same calculations for previous standard protocols. Studies of quality assurance parameters and image quality criteria in computed tomography for infants, children and adults will be performed.

A further evaluation of the methods for dosimetric measurements of CT-scanners will be performed. The correlation between the two different methods used for CTDI determinations (computed tomography dose index), the measurements of dose profiles free-in-air at the centre of rotation and the measurements on phantom surfaces, has to be generated in order to avoid misinterpretation of scanner performance and dose assessments.

As a background of this research project studies will be performed to extend image quality criteria for paediatric conventional radiography to the age of 5 years. It has been demonstrated in conventional radiology both for adults and for infants that wide variations in radiographic techniques and entrance surface doses exists and significant dose reduction can be achieved without any loss in image quality.

These studies in conventional radiography serve as a reference to the extension to computed tomography and the rating will be made by the Lake Starnberg Group based on the criteria defined in the "Working Document: Quality criteria for diagnostic radiographic images in paediatrics" which has been available recently.

Project 1 (Århus University Hospital).

Image quality criteria recently formulated for computed tomography for adults in high contrast regions (chest) and for low contrast regions (abdomen) will be tested and extended to other examinations. Standard protocols used for the latest installed CT units in Denmark (91/92) will be evaluated and compared with the image quality criteria formulated. The influence on expected dose reduction will be assessed by dose measurements and phantom measurements of image parameters.

Information on paediatric computed tomography performed at the departments having paediatric patients regularly will be collected and compared with the information gathered by the other contractors and image quality criteria tested. Further studies of the correlation between dose descriptions for computed tomography will be performed using TLD and ionisation chambers, free-in-air and in phantoms.

Project 2 (LNETI).

Image quality criteria for paediatric CT will be developed based on optimization and field studies. Optimization will be carried out for each examination type with objective to get diagnostically acceptable CT images with the lowest patient doses. This study will be done in Lisabo University Hospitals through the definition of: minimum level of image quality that keeps diagnostic information; scan parameters to produce those images; physical data associated with scanners and scan parameters; patient doses.

Field studies will be carried out to collect information on technical CT parameters used to perform paediatric examination of head and thorax and test image quality criteria formulated in this project and by other partners.

Additional studies will be done about the influence of scanners technical specifications and parameters on patient dose and image quality. Work within the contract period will be focused in the most frequent typed of paediatric examinations: head and thorax for babies, infants and children.

Project 3 (Universität München).

This project can be divided into two parts:

I. A continuation and extension of the surveys of radiographic technique, dose measurements and evaluation of image quality in conventional diagnostic X-ray examinations in paediatrics, and II. development of quality criteria for paediatric CT examinations and test of their implementation along with dose measurements in a pilot study in the south of Germany.

I. In the previous contract period a thermoluminescent dosimetry study (TLD) was confined to the chest of a premature baby and the 10 month old infant. The Lake Starnberg Group (LSG) recommended to extend the project to X-ray examinations of the 5 year old child, including contributions from non EC-countries, i.e. Scandinavia, Austria, Switzerland, and Eastern Europe, as well as the extension of the survey on the intravenous pyelography (IVP) to the 10 months old infant, thus profiting from the considerably higher frequency of this examination in this age group. The X-ray examinations using the standard IVP, chest and skull in 2 projections necessitate changes in the questionnaire, the data collection and the data analysis.

II. A pilot study on paediatric CT examinations will survey the respective CT technique and will measure X-ray dose free-in-air using appropriate TLDs. These measurements will be conducted together with Dr. Horwitz (Universität Würzburg) and will be analyzed by the GSF (Neuherberg).

Quality criteria for paediatric cranial CT examinations (skull, brain, face, orbits and hypophysis) have been drafted and were sent to the professionals interested in the project for evaluation and possible use in their studies. The LSG revise the paediatric quality criteria for CT examination for the skull and discuss respective criteria for other organs (chest, abdomen, spine and pelvis).

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920024 Diagnosis related dose: an investigation on patient risk and image quality in european hospitals.

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Description of research work

The main sources of low dose radiation to the population of Europe are the medical diagnostical examinations in radiology and nuclear medicine. Large effort has been put on lowering these doses by investigation of (entrance) dose to patients, intake of radionuclides, concertation on guidelines for good radiological practice, quality assurance programs,... mainly examination or equipment related.

In this project we will investigate factors that influence the amount of radiation given before a certain diagnosis is made. This amount depends on factors of medical, technical and/or organising character:

- in the medical sector we identify the justification of an examination and the protocol leading to a diagnosis. Little work is done on the influence on the dose by these items so far.
- in the technical sector many variables are identified and great effort has been noticed to evaluate the effects of quality assurance and to establish guide-lines for the improvement of image quality and the reduction of patient dose.
- in the organising sector many extrinsic and logistic factors -e.g. factors that are linked to the differences in the organisation of the health services- can compete with an optimum cost/benefit relation.

Present status.

No data are available that relate the (estimation of) doses and therefore the risk to establish a correct diagnosis in different European countries, to those three factors. A method that can contribute to a better understanding of this relationship is the concept of diagnostic groups (DG's): a DG is the set of examinations that has led to one and the same diagnosis.

A pilot study (CEC Radiation Protection Contract Bi7-054) was devoted to the inventory of existing useful methods of dose assessment in diagnostic radiology. A methodology for patient data and equipment data collection is available, and the goal of this project is to include more hospitals in the study, to develop the use of both the Image Quality Figure (see BIR Report 20) and the dose, to rank different strategies mathematically, and inventoriate and discuss the reasons that can lead to the choice of examinations and number of images and finally evaluate the medical and/or sociological data.

For two frequently occurring diagnoses (Renal Cell Carcinoma and Lumbar Hernia Discalis) a protocol for inclusion in the study is available and was tested: around 30 patients per diagnosis and per centre were analysed in the two participating Institutes, Nijmegen St. Radboud, and Brussels, University Hospital; V.U.B.. as well as all data relevant to patient exposure (type of examination, number and size of films,...). In parallel, the centres agreed upon a protocol describing the determination of equipment parameters, needed for the dose calculations. Data were collected on the equipment used in the radiological rooms used in Brussels and Nijmegen.

A first rough analysis of the patient data showed that within an institution the practice is rather consistent but some significant difference in diagnostic practice occur between the two institutes (e.g. digital angiography is a common practice in Nijmegen for diagnosis in Renal Carcinoma, while in Brussels Computed Tomography (CT) is preferred).

For conventional X-ray images, entrance surface dose for the used examinations is presently evaluated. For CT, patient dose will be evaluated using the method published by Shrimpton and Jones in NRPB Report 249.

Perspex test objects were used for dose determination, and with the Contrast-Detail (CD) phantom , consideration is also given to image quality in the equipment evaluation. In the past, the Nijmegen team has gathered much experience in the quantification of dose and image quality by the use of a single image of a CD test phantom. The Image Quality Factor (IQF) was developed for this reason.

Objectives.

The pilot study "Diagnosis related doses" showed that a diagnostic group can be established and clearly described -at least for some diagnoses- and that the elements determining the dose can be collected. This proposal will investigate this topic further and collect the data required to give a starting point to a discussion on recommendations for optimisation of diagnostic radiology procedures and a better use of national health resources.

To compare the risks of different pathways leading to a given diagnoses, a common risk factor must be handled for conventional and for CT examinations. Organ doses are available for a set of standard examinations and projections (B. Wall, NRPB Report 186), but these differ often from the angles and field sizes used in the examinations used. So a number of them must be calculated, using the same methodology as used in NRPB Report 186.

The specific objectives can be summarised in following items:

- exploitation of the results obtained in the pilot study, and improve some approaches;
- evaluate and refine the data collection procedures and data sheets for selection of patient files, of examination information
- completion of the data collected by data from partners in Madrid, Paris and Udine and extra data from Nijmegen nand Brussels
- calculation from skin-dose and used imaging techniques into absorbed dose or dose equivalent, so far it can be done with the help of the available literature data;

- use of the Image Quality Figure (IQF) (see BIR Report 20, p.29) and the dose to rank different strategies mathematically;

- inventory of reasons that can lead to the choice of examinations and number of images and evaluate the medical and/or sociological data;

- describing the methodology of the diagnostic groups in a publication so that the dose and risk evaluation that can be done in other institutes.

The project will be executed by the two centres that initiated the Diagnosis Related Dose project, with contributions from three centres with experience in quality assurance and patient dose reduction.

Example of selection criteria for inclusion:

Radiological diagnosis of "Lumbar Hernia Discalis".

Diagnosis:

Detection by imaging procedures such as standard radiography, tomography, computed tomography, MRI, myelography or discography of a hernia discalis resulting in surgical treatment.

(Hernia discalis = expulsion of the nucleus pulposus through a ruptured annulus fibrosus into the vertebral canal or into the inter vertebral foramen).

Golden standard:

Hernia discalis proved by operation. (Operation protocol).

Exit point:

Surgical treatment of the hernia.

Entry point:

First radiological examination following the complaint of LBP or sciatalgy.
In case of chronical LBP we take into account examinations up to one year before surgery.

Criteria of exclusion:

We can only accept patients who can account for all their radiological examinations, even if some of them are taken in other hospitals.

We will also exclude patients who already had surgery for a hernia discalis.

Examinations to be reviewed for dosimetric study:

Standard radiography, conventional tomography, computed tomography, MRI, myelography, discography, bone scintiscan and other pre-op examinations.
Examinations performed in another centre or private practice will be included as if they were perfomed in the participating centre.

Data collection:

Relevant information will be collected on a data sheet.

-patient identification

-chronological summation of all the radiological examinations as stated above

-technical data (physics)

-exposure data including beam direction, FOV, kV and number of films exposed.

University Hospital AZ-VUB, Brussels:

Contribution comes from the University Hospital, with its large Radiology Department (2 MR, 3 CT, 16 radiology rooms,..). The team will co-ordinate the project.

The team of Brussels University and University Hospital will:

1] Be responsible for the co-ordination, meeting organisation and reporting.
2] Cooperate with Nijmegen-St Radboud to further develop the equipment radiation measurement protocols, including CT. This includes further development of a measuring protocol for the skindose of a standard patient at each piece of equipment that is used in the production of those images. The results must be directly comparable with other published data.
3] Cooperate with Nijmegen-St Radboud on the calculation of absorbed dose from the skindose and the X-ray techniques used to absorbed dose, dose equivalent and if possible, to radiation risk per examination and per patient using available literature data;
4] Develop the data base and centralise the results; criteria for the data-base will be set together with Nijmegen, user of the data.
5] Give assistance to the measurement of physical parameters in the minor subcontractors' institutes, to warrant a coherent dose and risk evaluation.
6] Co-ordinate and contribute to the comparison, the interpretation and the final report: inventory of the reasons that led to the examinations and number of images for the patients, evaluation of the medical and./or socio-economic data.

St Radboudziekenhuis, Universiteit Nijmegen

In the department of Radiology a group of 7 physicists is working on Diagnostic Imaging, MRI and MR Spectroscopy, and mammography. They are running the Dutch National Reference center for breastcancer screening, with daily control by telephone line of over

30 screening units in the Netherlands and abroad. Reduction of patient dose in one of the main research lines in the department, and equipment and experience is available.

The items of St Radboud University Hospital Nijmegen are the following:

1] be responsible for the collection of data on the number and size of X-ray images and the X-ray techniques used in physical measurements;

2] Together with the team in Brussels, further development of a measuring protocol for the skindose of a standard patient at each piece of equipment that is used in the production of those images. The results must be directly comparable with other published data.

3] Be responsible for the calculation of absorbed dose from the skindose and the X-ray techniques used to absorbed dose, dose equivalent and if possible, to radiation risk per examination and per patient using available literature data.

4] The quantification of image quality in terms of the Image Quality Figure (IQF) of the images of which the dose is measured. We expect that the Nijmegen group is able to trace the differences in dose per image down to differences in image quality, type of image receptor or processing. The main object is to use both the IQF and the dose to rank different systems mathematically. In this project it will be used to possibly explain the differences in dose found in the participating hospitals.

5] Contribute to the comparison, the interpretation and the final report: inventory of the reasons that led to the examinations and number of images for the patients, evaluation of the medical and./or socio-economic data.

For specific data collection subcontractors have been concluded with:

Universidad Complutense de Madrid - Cátedra de Física Médica

Unità Sanitaria Locale No7 , Servizio di Fisica Sanitaria, Udine:

Centre d'Assurance de qualité des Applications Technologiques dans le domaine de la Santé (CAATS) - Cachan Paris

In order to:

1] Collect patient and examination data for the diagnoses Renal Carcinoma and Hernia Discalis for 30-40 patients, retrospectively, following the protocol developed within contract Bi7-054 and amended by St-Radboud and Brussels;

2] Determine, in co-operation with the physics team of Brussels, the relevant equipment data;

3] Contribute to the comparison, the interpretation and the final report.

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920037 Optimisation of image quality and reduction of patient exposure in medical diagnostic radiology.

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Total Contribution by the Commission: 270 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

This coordinated project has the general objective "optimisation of the radiation protection" and aims at implementing all relevant initiatives which can reduce radiation risk and improve image quality in diagnostic radiology. The project is concerned with two basic areas of radiation protection in medicine:

1) The practical implementation of Quality Assurance in diagnostic radiology;
2) Development of an Expert System for Quality Assurance.

This project is a development of the previous coordinated contract Bi7-0019.

1) The practical implementation of Quality Assurance in diagnostic radiology

In the practical implementation of Quality Assurance programmes in Diagnostic Radiology it is important to obtain the required image quality at the least possible dose. It is essential therefore that the correct choice of equipment and operating parameters is made. In addition the concerted choice of parameters to be monitored and the existence of Quality Control protocols are fundamental to the quality system.

The objective of this part of the project is therefore to identify the parameters which control image quality and patient dose and to develop methods for their optimisation and monitoring. This will be achieved by the use of computer modelling and experiments, and by assessment of data gathered from existing Quality Control protocols.

2) Expert System

Diffusion of scarce expertise concerning the management and interpretation of Quality Control tests should be encouraged and made available to the personnel involved in Quality Control practices.

The key for solving this problem is the design of a computerised tool capable of simulating and trasferring such an expertise.

Such a tool (Expert System) concerning mammography is in development since the previous contract period.

The work programme of this part of the project will deal with:

- Improvement of the theoretical knowledge base;

- Validation of the Expert System for mammography.

- Initiation of the extention of the Expert System to other radiological techniques.

The programme work of the project is based upon the active collaboration of all coordinated partners.

DETAILED DESCRIPTION - OBJECTIVES AND EXPECTED ACHIEVEMENTS

Over the past 10 years, the Radiation Protection Research Programme of the CEC has facilitated the development of an interactive collaborative research programme in the medical radiological field.

This has led to the creation of a framework for patient dose reduction strategies in which image quality is a key element, and which adequately underpins the Euratom Directives 84/466 and 80/836.

There are two main objectives in this coordinated project :

1) To reduce patient radiation risk while improving image quality in diagnostic radiology;
2) To continue to contribute to the effective implementation of these Directives in all member states.

Concerning the first objective, the selection of imaging conditions to provide the required image quality at the least possible dose will be explored theoretically and experimentally.

From the theoretical point of view, the project deals with the extention of existing computational models to take account of patient inhomogeneities, contrast and noise transfer by the image receptor, and image unsharpness, while allowing for variations of these effects over the image plane. Many different types of radiological imaging can be explored using such powerful models.

Within this project, factors which influence image quality and radiation risk will be investigated (choice of x-ray spectrum, scatter rejection method and image receptor). Those technical parameters which minimise radiation dose while achieving the desired image quality throughout the image plane will be identified and disseminated. Attention will be paid to imaging techniques which use conventional screen-film receptors, receptors for digital radiology and even equalisation radiography systems. Information gathered through this approach will be used interactively as inputs during the extention of the expert system to other imaging equipment.

In tandem with this theoretical research, experimental work will be undertaken to test the parameters for their influence upon the dose and the image quality. This work will be concentrated upon mammography and CT-scanning. The results of these experiments will also be used for the extension of the expert system.

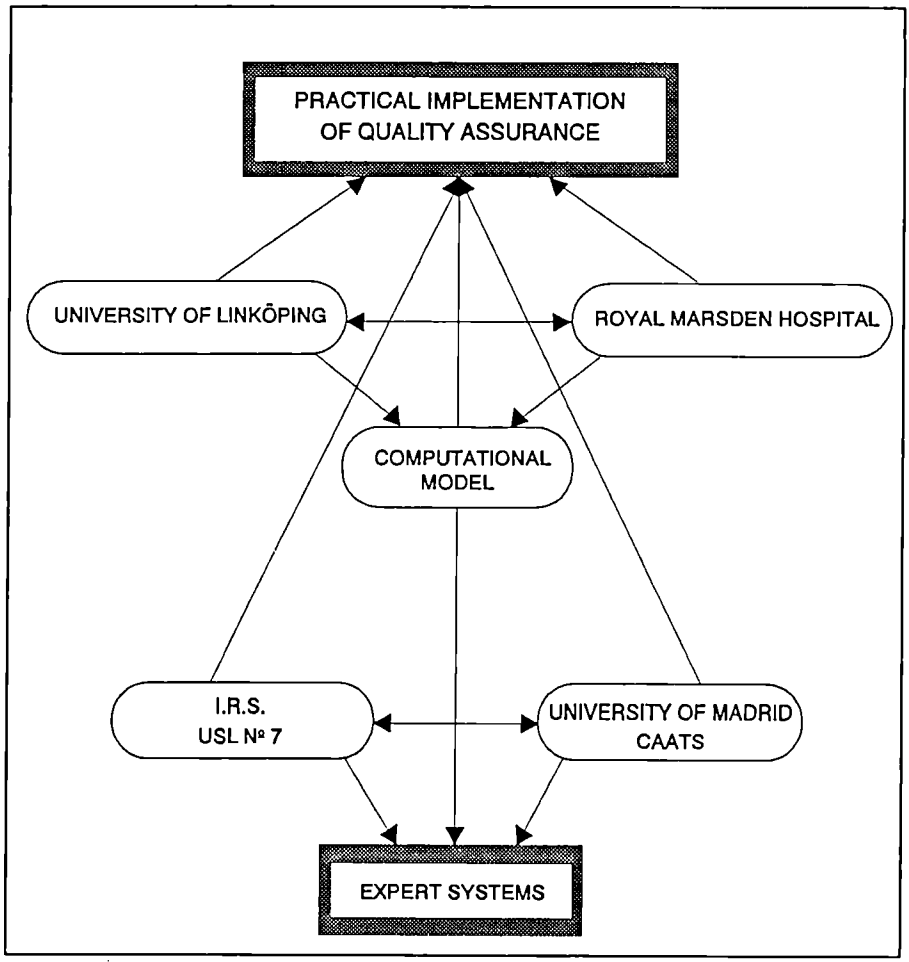
Concerning the second objective, recent efforts have focused upon the definition of European Quality Control protocols in mammography.

These efforts have revealed the need, for access to expert advice concerning the evaluation of Qualtiy Control programme results, and the implementation of findings in order to improve local practice.

This objective will be attained by the establishment of an Expert System for quality control which will permit:

- the diffusion of scarce expertise concerning the management and interpretation of Quality Control tests;
- the automation of certain routine Quality Control tasks;
- the education of untrained staff in Quality Control procedures.

The programme work of the project is based upon the active collaboration of all coordinated partners together with additional laboratories outside of this project. The interactions of all participants in this coordinated project are described in the following scheme



IRS, USL in Udine, University of Madrid and CAATS altogether will be actively involved in the expert system approach for mammography, in its further development and validation.

University of Linköping together with the Royal Marsden Hospital will share the development of a computational model which will include patient inhomogeneities and incorporate transfer of noise, contrast and image unsharpness by the receptor.

Interactive collaboration will be established between the two groups in order to atteint the global objective of the coordinated project. The benefits gained from such a collaboration include the sharing of expertise of the two Group Leaders, and the merging of different skills and approaches with experiences gained in diagnostic radiology physics and radiation protection in different countries.

CAATS contribution.

a) PARTICIPATION IN THE ELABORATION OF AN EXPERT SYSTEM FOR QUALITY ASSURANCE IN DIAGNOSTIC RADIOLOGY (improvement of the existing prototype)

The management and the scientific interpretation of the results of a Quality Control programme in diagnostic radiology constitutes a complex and difficult task requiring access to an expertise which, at the present time, is not always available.

In the case of mammography screening, access to expert advice is of vital importance. The prototype of the Expert System for Quality Control, developed under the previous CEC radioprotection programme, is designed for problems generated by this radiographic technique. Although somewhat limited in its use, this prototype has demonstrated the feasibility of such an approach (creation of an adequate knowledge base for the automatic resolution of problems posed by the exploitation of results of quality control measurements).

Within this context CAATS will improve this knowledge base according to a protocol which will be established jointly by the principal laboratories concerned (Udine, IRS and Madrid). Specifically, three initiatives will be taken within the project once the protocol has been established :

1) Collection and analysis of information from the image production system (x-ray generator), from the image reception system (cassette, screen, films), and from the image processing system (processor), from at least 50 mammography installations involved in the screening campaign in Bas-Rhin (France) presently under surveillance by CAATS.

2) Experimental study of modifications of image quality (resolution, contrast) and dose caused by malfunctions and sub-standard functionning of one or more links in the imagery chain of production (critical analysis of responses from different types of image phantoms, experimentation and simulation of failures).

3) Contribution to the methodological reflection on the extention of the prototype to other types of equipment.

b) CONTRIBUTION TO THE DEVELOPMENT OF IMAGE QUALITY CRITERIA

The optimising process of the radiation protection of patients undergoing CT examinations is in constant evolution:

- Significant progress towards the harmonisation of this radiological practice has been accomplished through the research conducted during the previous CEC radiation protection programme. This work has identified the radiological techniques used, the frequency of examinations by country, the doses per procedure and the collective doses (BI6-317-DK, BI6-136-I, BI6-135-UK, BI6-132-F).

- From the technical point of view of image quality (resolution, noise, signal to noise ratio), the importance of QC procedures has also been assessed (BI6-317-DK).

- From the radiological point of view of image quality, a first tentative towards the determination of quality criteria is in progress in one of the CEC member countries. This effort follows the same methodological approach used for conventional radiological examinations in adults and children (CEC trials).

Once these criteria for image quality will have obtained a wider European consensus, we propose to accomplish a synthesis of the relevent results concerning the technical aspects of the image quality.

The objective is to establish a coherence between the radiological and the technical dimensions of image quality by determining a minimal protocol of Quality Control measurements for the CT-scanner.

This objective will be attained by the simulation on some types of equipment with known dosimetric performances, using the technical conditions observed in real practice. Different types of CT phantoms will be used.

The collaboration of other laboratories is essential for the selection of radiological techniques to be assessed.

IRS contribution.

The project is concerned with the development, implementation and evaluation of an expert system approach to quality control programmes in diagnostic radiology. It is being developed initially for mammography. The IRS will link the fundamental research and development aspects of the proposed project to its implementation and evaluation in routine use, as well as contribute to its further development. The development of an expert system for use in quality control programmes has now reached a stage where the above can take place. With experience in the implementation and evaluation of quality control programmes in diagnostic radiology as well as close collaboration with a large number of hospitals throughout the Mersey Region, the IRS is ideally placed to evaluate the expert system approach in routine practice. Also based upon its involvement in the quality control programme of the Mersey Region Breast Screening Programme, the IRS will help to further develop the expert system.

Expert Systems for Quality Control

One of the prime requirements for the expert system approach to quality control in diagnostic radiology is the transportability of the system itself and its compatibility with standard computer hardware. The first action which will be undertaken will be to assess this aspect by transferring a system, developed in Udine, to Liverpool.

When the system is fully operational, a logic tree will be installed within the system which relates findings from a quality control programme to the most probable causes. The logic true and probability profiles will be developed from a data base analysis of quality control data established over a long time period for a number of breast screening units. This analysis relates the findings of quality control tests, such as phantom image results, to the radiographic factors employed. The data base provides the normal range of measured parameters to expect so that variations outside "tolerance" can be highlighted and probable causes indicated.

A prime requirement for any viable expert system is that it be user friendly so that scientifically untrained staff can operate it. This project will evaluate the software requirements for development of user friendly systems which can be operated on a European wide basis. Also, the data base analysis of quality control results which actually provides the logic and probabalistic elements of an expert system must reflect local conditions. Quality control problems and their magnitude are not universal but very much relate to local circumstances. Therefore, the facility to modify existing data bases to reflect local circumstances must be developed and implemented in an easy to use approach.

It is intended that expert system will be evaluated under routine conditions so that effective advice and guidance can be provided to the Commission regarding their potential in this area of radiation protection. It is also intended to evaluate their potential as teaching aids since the framework for quality control will form an implicit component of the system as well as eventually full diagnostic profiles relating cause and effect of variations in performance. This approach could help to standardise throughout Europe education and training in this field.

University of Madrid contribution.

1) Development of Image Quality Criteria

The image quality criteria proposed at present by a group of European experts will be applied in several X-ray Departments. The obtained technical results together wiht the opinion of the field radiologists about the selectiveness of the criteria will be used to score the images. At the same time image quality will be scored in a number of X-ray rooms, by using the image parameters obtained from suitable test objects. Such a score will be compared with that gathered from the application of the EC criteria and its suitability to image optimization will be analysed, even in those cases where the quality criteria proposed by the EC expert group are thoroughly fulfilled.

2) Risk evaluation in medical X-ray exposures

The group from Madrid will continue updating and enlarging its radiodiagnostic patient dose data bank, likewise the population statistical data, in the scope of updating the evolution of the radiological risk in the Madrid area, with a possible extrapolation to the rest of Spain. For the period 1992-1993, the contribution to the knowledge of patient doses will be increased, particularly in mammography and paediatric radiology, so that sectorized optimization programmes can be promoted in cooperation with other european groups. Evaluations wil be carried out to estimate the dose reduction and image quality improvement, through the QC and QA programmes already started in some radiological centres as an optimization pilot experiment. The benefit derived from the application of Radiation Protection and Quality Assurance training programmes in X-ray services will be evaluated in terms of dose reduction (occupational and patients), as well as of improvement of the image quality.

3) Elaboration of an Expert System for Quality Assurance in Diagnostic Radiology (improvement of the existing prototype).

The Madrid research group will continue collaborating to the development of an Expert-System for Quality Assurance in Radiodiagnostic which is being designed by the italian group (Dr. R. Padovani). The main objective will be to complete the available database and to bring the know-how of the Madrid research group to the achievement of the existing prototype. Some simulations of real actions already implemented in the practice to verify the prototype behaviour will be carried out.

USL nº7 contribution

Quality Control (QC) in medical diagnostic radiology can benefit from the development of Knowledge Based Systems (KBSs) which aim at emulating the way followed by human experts for solving problems. The main objectives of a KBS for QC are:

- to distribute scarce expertise concerning the management and interpretation of QC tests; it will help to introduce more effective QC procedures in every radiological department;

- to automate routine aspects of QC: frequent tests require human resources hardly available; automation of measurements, recording and interpretation of data will result in most favourable conditions to introduce a QC programme;

- to formalise practices of QC not yet embodied in a formal set of rules and procedures; building a KBS will have a role to play in organising QC knowledge, introducing consistent measurement regimes and suggesting hints for new research.

To demonstrate the feasibility of this approach we have built a prototype of KBS whose main task is to find out causes of insufficient performance of a mammographic system by analysing the images of TOR(MAX), a mammographic phantom of widespread use for daily checks of performance. During the routine use of this phantom in QC

programmes, test images are compared with a baseline reference image to find out deterioration in quality. The results of this surveillance procedure are used by the KBS as a first input for hypotheses on malfunctions which may account for the observed variations in the test image. Although the prototype is limited in use, its development was able to successfully test the adequacy of the knowledge formalisation and of the basic underlying ideas.

Objectives

We propose to revise first and then extend the prototype of the KBS along the following lines:

• modelisation of the problem solving process according to the KBS development methodology proposed by Esprit Project P1098, with special concern for the intricate interplay between equipment malfunctions and phantom test results ;

• knowledge acquisition. A critical step in the development of a KBS is the acquisition of expert knowledge. Even though many human experts are willing to make explicit their expertise, the process of knowledge elicitation is not an easy task since this expertise is hardly formalised in a way directly suitable for a KBS. A protocol that reflects the present formalisation and representation of knowledge will be designed for the collection of relevant knowledge and circulated among the most prominent experts in QC. The knowledge thus gathered including criticism for the protocol will allow the improvement of the knowledge base and the refinement of the representation language ;

• introduction of log-book data in the inferential process to exploit redundancy for supporting conclusions and to increase the system ability to make differential diagnoses. In particular, analysis of the results of QC tests recorded in the log-book allows the detection of early symptoms or trends in the behaviour of the equipment which may confirm or exclude some types of malfunctions ;

• generalisation to other phantoms. Presently, the KBS can infer diagnoses by observing images of a specified phantom (TOR[MAX]). The system should be modified so as to allow the (simultaneous) use of other phantom. Furthermore, the architecture of the system will be able to accept new phantoms without any major revision ;

• experimental simulations of malfunctions and their effects on phantom test images will help to identify advantages and drawbacks of a phantom for discriminating between different malfunctions by showing different variations of their measurable details ;

• extension to other equipments will be taken into consideration during the developmental and refining stages of the KBS.

The Royal Marsden Hospital and the University of Linköping joint project.

The principal aim of this project is the reduction of radiation dose and the improvement of image quality by the optimisation of the various parameters which control the performance of the radiological imaging system.

This will be achieved by the development and use of realistic computer based models of the radiological imaging system. Present computational models of the radiological imaging system have proved very useful in studying individual features, but the results must always be treated cautiously because they do not take the whole system into account. They are based on simple geometries which treat the patient as a uniform tissue slab, and by their very nature, cannot make a proper assessment of the variations over the image plane which occur in the real situation.

In this real situation, image quality changes with position in the image because (1) the patterns of primary and secondary photons vary due to patient inhomogeneities and (2) the transfer of noise and contrast by the image receptor depends on the energy imparted per unit area. The receptor also influences image quality because of its limited spatial resolution. Patient dose decreases with increasing thickness of the receptor but image quality is degraded due to increased blurring. The influence of dose and blurring will depend upon both the atomic composition of the receptor and the energy spectrum of the incident photons.

We wish to develop a computer model which approaches this realistic situation, and to use this model as a basis for the optimisation of the radiological imaging system in various situations including both adult and paediatric examinations, film-screen and digital imaging.

The first stage of this process is the development of the computational model. This model must be able to estimate the various physical quantities of interest within small areas of the image plane. This is not possible using straight forward Monte Carlo strategies because of large statistical uncertainties. We will therefore use the collision density technique which is able to estimate quantities at a point with high precision. Our present Monte Carlo code uses this method and provides a very good starting point for the development of the realistic model, although extensive additions will be necessary because of the much increased sophistication of this model.

The development of the realistic model will involve (1) the introduction of an anthropomorphic phantom to facilitate the evaluation of image quality as a function of position in the image plane and (2) the inclusion of the transfer characteristics of the receptor as regards contrast, resolution and noise. These two initial steps will be performed separately at the Royal Marsden Hospital and the University of Linköping, but good contact will be maintained throughout.

The second stage of the project is the use of the realistic model by both institutions to study the performance and optimisation of the radiographic process. Various test situations for adult and paediatric examinations will be investigated as follows:

1. Study of the ratio of scattered to primary radiation at various positions within the image. This ratio will differ substantially from that for a homogeneous phantom because the scattered and primary photons can traverse regions of very different compositions and densities. The study of this quantity is important because of the substantial degradation of image contrast which scatter produces, and because of the variation of the image contrast with position on the film characteristic for film screen imaging systems.

2. Optimisation of tube potential, grid design and film latitude for film-screen radiology taking into account the variation of contrast across the image plane, choice of screen and tube potential. Present optimisations may only be valid for parts of the image plane and take no account of the limited latitude of film and variations of contrast with optical density. An overall optimisation is necessary. The optimisation should be done for selected radiological procedures (adult PA chest and paediatric examinations).

3. Optimisation of tube potential and grid parameters for digital radiology, and for the same examinations as for (2) above.

4. Study of the effect of receptor thickness and atomic composition on image quality.

5. It is noted that the results available from both the realistic model proposed here and our existing model of the mammographic system may be of value to other contractors, including those developing expert systems. We should be pleased to produce such results where appropriate, but this way may be at the expense of some of the items listed above.

The Royal Marsden Hospital contribution

The project will be jointly managed by the project leaders in the two institutions. During the first stage, the Royal Marsden Hospital will develop the computer code to include the effects of tissue inhomogeneities.

The starting point will be the code already developed which uses the collision density estimator to achieve adequate statistical precision in the calculations. It accepts input from the various data files we have developed, which specify the parameters for X-ray spectra, patient and test object composition, grid geometry and construction and receptor composition. Major extension of the program is required for the present work.

The initial stage is the development of an appropriate phantom model. We shall use a geometric phantom similar to that employed by the group at NRPB, which is itself based on that developed by Cristy.

This phantom will be coded in such a way that alterations to patient size, and the positions and sizes of the internal structures can be readily made. This will facilitate the use of the phantom for both adult and paediatric examinations. The geometric phantom and the methodology for photon transport through the inhomogeneities will be discussed with the NRPB and the final code will be based on both our own previous experience in dealing with inhomogeneities and this discussion. At this stage, the programm will be able to calculate patient dose, and image contrast and quantum noise at any point in the image plane, but will take no account of the effect of the receptor transfer functions or noise in the image. The patient dose estimation can be made both in terms of the energy imparted to the patient and effective dose, with the latter quantity either being obtained using conversion factors published or supplied by NRPB, or estimated directly from the program itself.

The University of Linköping will develop models for the receptor transfer functions and noise whilst we are coding the patient inhomogeneities.

Close cooperation will be maintained between the two centres in the development of all computer models. The style and general approach to coding is already decided by the two institutions, and the program structure, algorithms and outline of the code will be agreed at each stage. All programms will be carefully tested.

In the second stage of the project, the code will be used by both institutions in the investigation and optimisation tasks identified earlier. We shall allocate these between the two institutions once the codes have been developed. Both institutions will collaborate closely in the detailed planning of these tasks, the analysis of the results and the production of recommendations on the choice of operating parameters.

The University of Linköping contribution

The project will be jointly managed by the project leaders in the two institutions. During the first stage, the University of Linköping will develop a computer code to take into account receptor resolution and noise characteristics.

In the present model, receptor response is expressed in terms of energy imparted to the receptor. Values of contrast (film-screen radiography) and signal-to-noise ratio (digital radiography) are derived assuming the energy to be imparted at the site of the primary photon interaction. We shall use Monte Carlo simulation to calculate the spatial distribution of this energy caused bu the diffusion of the secondary radiations. In particular, when incident photons have energies above a K-absorption edge of the atoms in the receptor, the K-radiation liberated may travel a considerable distance before absorption, thereby degrading resolution and adding spatially correlated noise to the signal. This is an important feature of many modern receptor materials containing Cs, Ba, I, La, Gd with their absorption edges within the energy range of commonly used X-ray spectra. The emission and diffusion of light photons in fluorescent screens will be considered to allow derivation of the receptor point spread function and, finally, the modulation transfer function (MRF) and quantum noise characteristics (Wiener spectrum). These functions should be derived as a function of photon energy as well as of energy spectrum (tube potential and filtration), receptor thickness and atomic composition.

This part of the code will give the basic information about the relationship between the physical image quality parameters and the fundamental interaction processes in the receptor. Image quality is also influenced by the particular equipment used in managing the imaging system. We will in addition have to consider the film characteristics in film-screen systems and electronic and digilisation (system) noise in digital systems. These components, depending on the actual equipment, must be added from empirical data.

The Royal Marsden Hospital will develop models for the patient inhomogeneities whilst we are coding the receptor transfer functions and noise.

Close cooperation will be maintained between the two centres in the development of all computer models. The style and general approach to coding is already decided by the two institutions, and the program structure, algorithms and outline of the code will be agreed at each stage. All programms will be carefully tested.

In the second stage of the project, the codes will be used by both institutions in the investigation and optimisation tasks identified earlier. We shall allocate these between the two institutions once the codes have been developed. Both institutions will collaborate closely in the detailed planning of these tasks, the analysis of the results and the production of recommendations on the choice of operating parameters.

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920052 Patient dose from radiopharmaceuticals.

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Total Contribution by the Commission: 75 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Scientific background

Absorbed dose estimates are needed in clinical diagnostic work for judging the risk associated with the use of specific radiopharmaceuticals, both for weighing against the possible benefit of the investigation and giving adequate information to the patient. These estimates also provide guidance to ethics committees having to decide upon research projects involving the use of radioactive substances in volunteers, who receive no individual benefit from the study.

The main source of information concerning patient doses from radiopharmaceuticals is Publication 53 of the International Commission on Radiological Protection (ICRP) entitled "Radiation Dose to Patients from Radiopharmaceuticals" (Annals of the ICRP, **18**, No. 1-4, 1987). Since its publication, some new radiopharmaceuticals with potential for wide application in the field of nuclear medicine have come into use. For example, a new generation of mTc-labelled substances (HMPAO, hexamethylpropyleneamine oxime; MIBI, methoxy-isobutyl-isonitrile; Mab, monoclonal antibodies, etc) has been introduced for diagnostic purposes. Investigations with these radiopharmaceuticals can lead to relatively high effective doses in the order of 10 mSv. This is largely due to the fact that these investigations, especially when they incorporate tomographic imaging, can yield valuable diagnostic information provided the administered activity is high enough to ensure the required image quality. Another reason stems from differences in excretion patterns of different labelled substances. Several of the newer substances are excreted via the hepatobiliary system and GI tract.

Consequently, for the latter there is particular emphasis on the need to estimate doses to various parts of the GI tract, including the stomach wall. This is important, because, according to the 1990 Recommendations of the ICRP (Annals of the ICRP, **21**, No. 1-3, 1991), the risk weighting factors for parts of the GI tract are comparatively high and absorbed doses received by them will influence the effective dose to a greater extent than the previously used effective dose equivalent (Annals of the ICRP, **1**, No. 3, 1977).

There is an increased interest in therapeutic applications of radiopharmaceuticals, such as labelled MIBG and various types of monoclonal antibodies. In therapy, good dosimetry is essential, as optimal administered activities lead to doses close to tolerance levels for vital tissues such as bone marrow and liver. Estimates of dose variation within tumours is also important for effective management of treatment regimes.

For more than twenty years internal radiation dose estimates have been based upon the mathematical description of MIRD-type anthropomorphic phantoms (Snyder *et al.*, MIRD Pamphlet 5. J. Nucl. Med. Suppl. 3, **10**, 1969). In comparison, the recent innovation of 'Voxel' phantoms presents certain advantages, particularly with respect to more realistic and detailed representation of organ shape, size and location, which are important parameters for internal dose estimation. Voxel size and overall dimensions of phantoms are variable factors, providing a powerful tool for assessing

organ doses in individuals differing in stature from standard phantoms. In the first stage of this programme, the suitability of Voxel phantoms was investigated for this purpose. The phantoms were developed at GSF and are extensively used for the estimation of doses resulting from external radiation fields. The Voxel phantoms are derived from computed tomographic (CT) data of individual humans and are used with Monte Carlo simulation techniques to calculate absorbed fractions for organ pairs. So far, three Voxel models have been constructed, two paediatric (an 8 week old baby and a 7 year old child) and one of an Alderson Rando phantom. An adult male phantom is currently undergoing these procedures and these are the first members of a growing family of Voxel phantoms representing various ages and both sexes.

Work programme

The current project is a direct continuation of an ongoing CEC Radiation Protection Programme project with the same title. It is carried out in close cooperation with the Department of Radiation Physics, Malmö, Lund University, Sweden, which was one of the participating organizations in the previous contract. The objectives of the new project are to produce dose data for new radiopharmaceuticals and to further improve the accuracy of the current data on absorbed dose to patients. This will mainly be achieved by measurements on patients and volunteers. There is a special need to improve the collection of biokinetic and dosimetric data for babies and children of various ages,because little information is presently available in these younger age groups compared to adults. This information is required to establish a base to optimize the activity administered and the measuring procedure for various investigations, so that the administered activity can be kept as low as possible, whilst fulfilling the diagnostic requirement. The work programme will incorporate the following main objectives:

1. **Kinetics.** Collection of biokinetic data will continue for selected radiopharmaceuticals by serial uptake and retention measurements on patients and healthy volunteers. Special emphasis will be given to:

   a) new radiopharmaceuticals recently taken into clinical use, and

   b) biokinetics and dosimetry for children and the new-born.

   Biodistribution and organ retention measurements will be performed using gamma cameras and whole-body counters, together with measurements on samples of blood and urine.

   Studies of patients will be supported, when appropriate, by experiments with phantoms.

   Special attention will be paid to the application of recently developed excretion models especially the dynamic urinary bladder model (MIRD Pamphlet No. 14, J. Nucl. Med. 33, 783-802, 1992) and a new GI tract model incorporating age and gender dependent factors (Stubbs and Barker, Proc. 5th Int. Symp. Radiopharmaceutical Dosimetry 229, 1992).

In view of the relative importance of the new risk weighting factors for the GI tract, it will be important to investigate methods to distinguish between radioactivity in the lumen and in the walls of this organ.

2. **Physics.** Attention will be paid to improving the physical basis for the dose calculations by using detailed voxel phantoms based on CT data for calculations of new Specific Absorbed Fractions and Specific Effective Energies for the determination of doses due to incorporated radionuclides, by using existing and new Voxel phantoms together with Monte Carlo codes. The Voxel phantoms, based on CT-data of real patients have been extensively used for external dosimetry and partly for internal dosimetry. The construction of phantoms is continuing with paediatric and adult phantoms.

   Where necessary, studies will involve the assessment of techniques, such as transmission attenuation correction and emission computed tomography to improve quantitation of organ uptake determinations.

3. **Dose calculations.** Biokinetic information will be used to quantify the mean absorbed dose (D) to various organs, as well as the effective dose (E). Numerical comparisons with calculated effective dose equivalent (HE) values will also be made. The results will enable comparisons of different radionuclide labels for various pharmaceuticals with the aim of balancing diagnostic benefit and radiation dose and to determine the appropriate activity to be administered in order to avoid unnecessary radiation exposure to the patient without losing pertinent information.

   The proposed studies will also offer opportunities for a comparative evaluation of the ICRP 30 GI-tract model used in ICRP 53 and the aforementioned model due to Stubbs and Barker (1991).

4. **Image quality.** Diagnostic image quality for children of different ages will be studied as a function of amount of administered activity. Phantom studies, as well as information from clinical investigations will be used. The first aim is a choice of activity leading to equal image quality for different ages. Various mTc-labelled diagnostic radiopharmaceuticals will be investigated.

   Except for our previous contract, there has been no other similar programme within the CEC Radiation Protection Programme. The United States Radiopharmaceutical Dose Information Centre at Oak Ridge carries out calculations based on biokinetic data collected at various hospitals. We have good contacts with the Oak Ridge group through various channels. The project coordinator is a member of an ICRP Task group on the updating of ICRP Publication 53.

   In the field of dose to patients from X-ray investigations, there are several projects within the CEC Radiation Protection Programme. There is a project supporting the construction of further Voxel phantoms and their use in diagnostic radiology.

Distribution of tasks amongst participating organisations.

MRC Clinical Research Centre

Project coordination (temporary). Proposed work will include the collection of biokinetic data for new diagnostic radiopharmaceuticals and dosimetry studies on children over a wide age range undergoing routine diagnostic imaging investigations at Northwick Park Hospital, Harrow, U.K. In collaboration with the Medical Research Council's Cyclotron unit at Hammersmith Hospital, collection of biokinetic data and modelling for short-lived positron emitters will start and be progressively increased. Dose calculations will be carried out.

GSF

The physical basis for absorbed dose calculations will be improved by using detailed voxel phantoms based on CT-data for Monte Carlo derivation of absorbed fractions incorporated in new S-values for combinations of organ pairs. At a later stage, calculations of absorbed doses for beta-gamma emitters and pure beta emitters in the skeleton will be carried out.

University of London, The Institute of Child Health

The proposed work involves the collection of biokinetic data for children with special reference to Tc-99m DMSA, and the assessment of diagnostic quality of imaging as a function of administered activity.

Especially in the field of paediatric dosimetry, it is of importance to have a joint project as the number of patients in each age group and each centre is limited.

The work programme covers both experimental and theoretical aspects of the problem. It focuses studies on children and together we have contacts with practically all types of modern nuclear medicine investigations, now also including PET studies.

Contribution of MRC Clinical Research Centre

a) **Temporary project coordination.** b) **Myocardial perfusion imaging agent,** Tc-99m P53. Comprehensive biodistribution and dosimetry investigations have been carried out on 12 normal healthy male volunteers and the results reported. In addition, data obtained in this study have been used to compare two methods of quantitation in human studies of biodistribution and radiation dosimetry. Radiation dose estimates will be compared with those for other myocardial perfusion imaging agents.

b) **Labelled monoclonal antibodies.** Biodistribution studies of a Tc-99m labelled monoclonal antibody against atherosclerotic plaque, have been performed in two patients with peripheral vascular disease. Analysis of data for dose

calculation is proceeding. There is a temporary halt on further studies with this substance but these two studies will be progressed as part of the investigation of the labelled antibodies in general. Biodistribution studies have also been performed using In-111 antimyosin in twelve patients with inflammatory muscle disease. Two myocardial infarct patients without evidence of inflammatory muscle disease also will be studied also with this substance for comparison. Image analysis and dose calculations for these subjects are proceeding.

d) **Paediatric dosimetry.** Biodistribution studies have been attempted with variable success in six children (4-8 years) following administration of Tc-99m DMSA to investigate renal disease. With one exception, time scales of follow-up have been limited to the first 3 to 4 hours after injection because of difficulty in getting return visits by these out-patients. However, these studies will continue with a view to establishing reliable biokinetic data and dosimetry for this substance.

e) **Dosimetry of positron emitters.** In conjunction with the MRC Cyclotron Unit of Hammersmith Hospital, the opportunity now arises to investigate the internal radiation dosimetry of a variety of radiopharmaceuticals labelled with positron-emitters (principally ^{14}C, ^{15}O, ^{18}F) for use in a clinical research programme which covers neurology, psychiatry, cardiology and oncology. Labelled substances could include blood flow agents, metabolic substrates, opiates, pyrimidines, and other drugs. Currently, ^{18}F - 5-fluorouracil is under investigation in cancer patients. In addition, in view of the short lifetime of many positron emitters, alternative methods of dose calculation based on kinetic models, used with recorded arterial blood concentration and tissue tracer retention will be investigated.

Contribution of GSF-Forschungszentrum für Umwelt und Gesundheit.

a) Calculations of Specific Absorbed Fractions and Specific Effective Energies will be continued by using existing and new voxel phantoms, together with Monte Carlo codes. The suitability of voxel phantoms to quantify doses due to incorporated radionuclides has already been demonstrated from the calculations for the 8 week old baby. For the present project, the voxel phantom of the 7 year old child and the new adult male voxel phantom, which is at the moment under construction, will be used.

b) Organ specific scaling factors will be assessed for each radionuclide and each age class to enable the determination of organ doses for individuals whose physical dimensions deviate from those of the phantoms used. This will be achieved by changing the voxel size of the phantom and consequently the body dimensions.

Contribution of the University of London, The Institute of Child Health

a) Biokinetic data of radiopharmaceuticals in children is not as well documented in children as it is in adults and it is also known that children have different biokinetics, particularly neonates. As DMSA scans represent one of the most frequently used radioisotope examinations at this hospital, it is proposed to collect biokinetic data on 15 children in the age group 3 months to 2 years and 15 children in the age group 2 to 5 years.

The geometric mean method will be used to obtain the time integrated activity in the kidneys, liver, spleen, bladder, knees and whole body after the injection of Tc-99m DMSA. Blood and urine activity will also be measured. From this biokinetic data and using the MIRDOSE 2 programme as supplied by Oak Ridge Associated Universities, estimations of the absorbed radiation doses to these organs will be calculated.

b) Diagnostic quality of imaging as a function of the amount of administered activity will be assessed. The definition of quality will be defined, then two sets of children who received proportionately different amounts of activity will be assessed. This will begin with 60 children aged 3 - 10 years undergoing Tc-99m MAG3 scans for indirect cystography, divided into 2 equal groups. This study will be completed within the time scale of this application.

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT920064g Medical dose assessment and evaluation of risk.

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Total Contribution by the Commission: 170 KECU
23 months 1/07/92 to 31/05/94

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Description of research work

Objectives

The contract combines a number of projects concerned with radiation dosimetry and quality control in diagnostic radiology aimed at assessing radiation doses and risks to patients and reducing them without compromising the diagnostic value of the examinations. The objectives are to improve the ease and accuracy with which doses to patients from x-ray examinations can be estimated and to develop a common basis for patient dosimetry and risk evaluation within the European Community. Particular attention will be paid to meeting these objectives in the fields of paediatric radiology and mammography.

Scientific Description

Computer modelling of medical x-ray fields using Monte Carlo techniques and geometrical or tomographic (voxel) phantoms has been performed at a number of centres worldwide, including 3 of the laboratories taking part in this contract. A range of geometric phantoms representing patients from infancy to adulthood has been developed as well as 3 voxel phantoms representing an 8 week old baby, a 7 year old child and a commercially available adult physical phantom (Alderson Rando). These phantoms have been used in Monte Carlo simulation of conventional and CT x-ray examinations to calculate normalised organ doses but mainly for adult patients. There is increasing interest in dosimetry for paediatric radiology because of the likely higher sensitivity of children to the effects of radiation. Thorough evaluation of paediatric radiology practice in both specialised and general hospitals is needed to provide realistic input data for these dose calculations. NRPB and GSF will continue with the development of Monte Carlo radiation transport programs in patient-like phantoms, to calculate normalised organ doses for an extended range of x-ray examinations and to include paediatric patients. Information on typical paediatric radiology procedures and doses, and a comparison between practice in a specialised children's hospital and a general hospital, will be provided by St George's Hospital: it will also implement and assess the impact of a complete Quality System following EN29000 on the performance of a paediatric radiology department. Close co-operation with the children's hospital of the University of Munich (Dr Schneider) will also be maintained and, in particular, GSF will help it to estimate organ doses to children from CT examinations.

National breast cancer screening programmes using mammography have been implemented in the UK and the Netherlands. However, the existing national protocols differ in the methods and data used to derive average glandular tissue dose in the breast. Breast phantoms of different thicknesses are used to derive air kerma free-in-air at the entrance position of the beam and the factors used to convert the air kerma into average glandular tissue dose are not derived in the same way. It is essential to assess the impact of these differences on the specification and measurement of breast dose and to arrive at a common European protocol. A unique calibration facility for mammography dosemeters at St George's Hospital will be used for an on-site intercomparison of the dosimetry systems of TNO and St George's Hospital. This will be valuable in the improvement of absolute dosimetry in mammography.

Average glandular tissue doses normalised to unit incident air kerma will be calculated by TNO using Monte Carlo techniques and used to evaluate the impact of the different modelling procedures and exposure data used in the various national protocols. The results of the calculations will be compared with experimental depth-dose distributions in phantoms and entrance dose measurements on patients to arrive at a standard method for specifying and measuring breast dose in mammography.

The International Commission on Radiological Protection (ICRP) has recently published new recommendations incorporating the latest radiation health effects models and including a definition of effective dose with an extended list of radiosensitive organs. Medical dose assessment and risk evaluation will, in future, have to embrace these new concepts. NRPB will investigate the possible application of the latest radiation risk models and the recent recommendations of the ICRP to medical exposures. The important influence of age at exposure and gender on risk coefficients, effective dose and radiation detriment will be assessed.

NRPB and GSF will continue to supply dosimetric support to the CEC initiatives for developing quality criteria for radiographic images in adult and paediatric radiology.

CONTRIBUTION OF THE NRPB

Organ dose calculations and risk evaluation

1. Monte Carlo organ dose calculations will be extended to include all the organs required to calculate the newly defined quantity effective dose and will include more x-ray fields enabling a wider range of examinations to be simulated. These fields will include those used during complex examinations involving fluoroscopy, such as barium meals and enemas. Organ doses will be normalised to both entrance surface dose and dose-area product.

In collaboration with St George's Hospital and GSF, x-ray fields and exposure conditions representative of routine paediatric radiography will be defined and used in the Monte Carlo program to simulate x-ray examinations on a range of geometric phantoms of different sizes representing patients from newborn to 15 years old. The corresponding normalised organ doses will be calculated to provide improved data for estimating radiation risks to children.

2. The 1990 recommendations of the ICRP, together with the latest epidemiological data and radiation risk models, will be studied to evaluate the risks to patients from x-ray examinations. The new ICRP concept of aggregated detriment and the corresponding definition of effective dose will be considered, with regard to their possible application to medical exposures.

In particular the influence of age at exposure and gender on risk coefficients and on the definition of effective dose will be assessed, to see if a system can be devised to account for these factors when estimating radiation detriment to patients and particularly to children. A link with other measures of health detriment (or benefit) such as Years of Life Lost (YOLLs) or Quality Adjusted Life Years (QALYs) will be sought.

3. NRPB will continue to provide advice and support on the dosimetric aspects of two initiatives that are being developed by Study Groups set up from within the group of contractors working under Topic 2.2 of the CEC Radiation Protection Research Action. These two initiatives involve the preparation and European-wide trials of quality criteria for diagnostic radiographic images in adult and paediatric radiology respectively.

CONTRIBUTION OF GSF

Phantom development and organ dose calculations

1. Continuation of construction of voxel phantoms, using images of contiguous transverse slices of real patients obtained by computed tomography. This will be achieved by a segmentation technique being developed, which is based on standard image analysis software running on a Mipron/Contron computer. This technique offers a high degree of automation. An adult male phantom is under development; data for constructing further phantoms are available.

2. Extension of the calculations of organ doses from medical exposures, using existing and new phantoms together with Monte Carlo codes for:

- CT examinations of children (in cooperation with the Children's Hospital of the University of Munich).
- Assessment of the impact on paediatric patient doses of the recommendations for "good radiographic techniques" in the CEC Quality Criteria for Diagnostic Radiographic Images in Paediatrics.

3. Dose measurements and field studies in routine x-ray diagnostic procedures to support coordinated CEC projects and initiatives. Special emphasis will be given to the determination of patient exposure in paediatric CT examinations (in cooperation with the Children's Hospital of the University of Munich).

Contribution of the St. George's Hospital

Patient dosimetry and quality control in paediatric radiology

There is a shortage of data on patient doses in paediatric radiology and it is believed that x-ray practice in a specialist children's hospital is often superior to that of a general hospital because of the benefits of staff specialisation and training. This project will assess the difference in radiographic, radiological and referral practice between two independent children's hospitals, a number of general hospitals and a teaching hospital. The impact of installing a Quality System meeting the requirements of EN29000 on patient dose and image quality in a children's hospital will be studied, and the potential benefits of applying the good radiographic practice in a children's hospital to general x-ray departments will be assessed. The study will include all paediatric age groups within the target hospitals and will comprise an evaluation of the following:

a) the frequency of all types of paediatric x-ray examination according to age group;
b) radiological referral criteria for undertaking examinations, and radiological and

radiographic practice and technique;

c) dose measurements using TLD and dose area product meters as appropriate for simple and complex examinations respectively;

d) the influence of radiographic and radiological technique on patient dose and image quality, image quality being assessed using objective test tools with which considerable experience has already been gained;

e) the improvement in patient dose and image quality when a quality system meeting EN29000 is applied in a specialist hospital;

f) the improvement in patient dose and image quality in a general hospital when agreed paediatric radiologic and radiographic criteria are applied;

g) dose reduction techniques in a specialist hospital including an evaluation of film screen sensitivity using realistic diagnostic x-ray spectra.

This project will be in collaboration with the Children's Hospital of the University of Munich and NRPB.

Calibration of a reference standard ionisation chamber used for dosimetry in mammography will be carried out for TNO in St George's calibration facility using realistic mammographic x-ray spectra.

CONTRIBUTION OF TNO

Dose measurements in mammography

Average absorbed dose in glandular tissue, which is recognised as the most appropriate quantity for dose specification in mammography, cannot be measured directly and has to be derived from measurements of air kerma free-in-air and appropriate conversion factors. Measurements are confounded by the fact that most national standards laboratories do not provide calibrations at true mammographic x-ray qualities. St George's Hospital (participant no. 3) has a unique calibration facility with real mammography x-ray sets which will be used in a valuable intercomparison of mammography dosemeters with TNO.

Air kerma to glandular dose conversion factors have been published by various authors as a function of half value layer (HVL) for simple breast phantoms. Recent studies at TNO have shown that depth-dose distributions provide a better measure of radiation quality than HVL in mammography. Moreover, different radiation transport codes, photon interaction data, photon spectra and composition and geometry of the breast phantom have been used by the various authors in calculating the conversion factors. Protocols for dosimetry in mammography in different European countries (UK, Netherlands, Sweden) use results from different authors. TNO will study the impact of these differences by calculating, with the MCNP radiation transport code, conversion factors for photon spectra fitting previously measured depth-dose distributions as well as the spectra used by other investigators (possible variations 10-20%). Different breast geometries and compositions will be assessed (possible variations 10-20%) including tissue compositions recently recommended by ICRU. Data will be presented for the range of reference breast thicknesses used in existing national protocols (4.5 -5.5 cm) which correspond to about a factor of two difference in glandular dose.

Average glandular tissue doses will also be obtained by integration over appropriate depths of the available experimental depth-dose distributions inside homogeneous phantoms. The results will be compared to calculated conversion factors. Available data on entrance doses for phantoms of various thicknesses and for actual mammography patients will be analyzed to obtain insight into dose variations for individual women, covering the whole range of thinner to thicker breast (2 to 8 cm. compressed).

C22 Reduction of patient exposure in medical diagnostic radiology.

Contract FI3P-CT930070 Evaluation of dose and risk due to interventional radiology techniques.

Coordinator Klinikum Nürnberg
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Total Contribution by the Commission: 65 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Interventional radiological procedures are increasing all over the world. The diagnostic and therapeutic interventions usually cause high occupational exposures and patient doses. Both arguments force the determination of the contribution of interventional radiology to the mean collective exposure due to diagnostic radiology.

There are two main objectives in this coordinated project:

1) Occupational and patient dose assessment;
2) Quality Assurance in interventional radiology.

Concerning the first objective, important efforts will be made in each country participating in the coordinated project to evaluate the frequency of examinations, the technical parameters used and to measure dose of patient and staff. Concerning the second objective, the selection of imaging conditions and parameters to be monitored to provide the required image quality at the least possible dose will be explored experimentally.

The determination of radiation dose and its interpretation will be used as scientific fundamentals for the estimation of the individual and collective risk of the somatic and hereditary effects due to the exposure of diagnostic radiology. For the development of uniform radiation protection concepts the evaluation of radiation exposure is required. Hence the possibility is given to optimize the radiation protection.

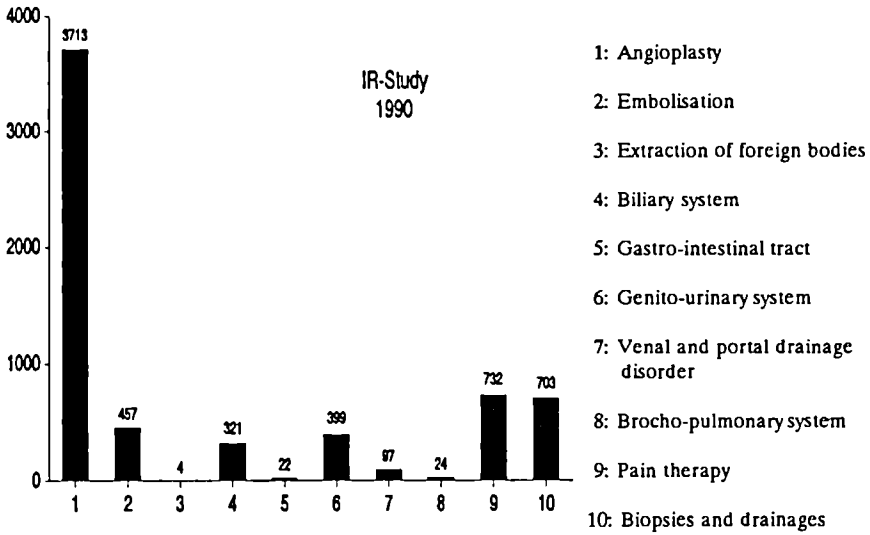
The programme work of the project is based on the collaboration of three European working groups, Germany (Klinikum Nürnberg), France (CAATS) and Italy (USL n°7 Udine).

For dose measurements a protocol will be established, which prescribes both the calibration of TLDs and dose area product meters and the methodology of the measurements. Using harmonized risk relevant dose quantities, it will be possible to compare the results of the participants to some extent.

Contribution of Klinikum Nürnberg

The most extensive documentation of the number of interventional radiological procedures in Germany is conducted by Prof. Dr. med. E. Zeitler, Klinikum Nürnberg. More than 100 institutes will participate in the central documentation. In the first year (1990) of the study 6477 interventions of 35 institutions were submitted. The figure shows, that the most frequent interventions are Angioplasties with nearly 60%. For this kind of intervention the radiation exposure of the patient will be determined by measuring the dose area product at the Klinikum Nürnberg. Additionally TLD - measurements will be done.

Interventions



The exposure of the radiologist and the assistency will be measured with TLDs. Tissue and organ doses, like fingers, head (eyes, thyroid gland) and body, will be determined separately. It will be investigated what kind of correlations exist between the exposure of staff and the exposure of patients, the kind of interventions, the X-ray unit, the qualification of radiologists a.o..

This documentation, adding extended inquiries, is the unique chance to evaluate the radiation exposure of staff and patients depending on the interventional methods and localizations.

Contribution of CAATS

The Interventional Radiology (IR) technique is applied to all anatomic regions and, from the technical point of view, includes sophisticated and precise radiological tests.

Radiation exposure of patients as well as of medical and paramedical staff involved should receive particular attention, whether generated by puncture-biopsies, angioplasties, embolisations or nucleolysis. All these exams generally require multiple radiographic series of images as well as prolonged fluoroscopy screening time.

So far no study has been conducted in France which might permit the quantification of the impact of this imaging technique according to the frequency of examinations, the technical parameters used, characteristics of radiological equipment, the number of paramedical and medical personnel involved and medical protocol used during different procedures.

As a first step, the evaluation of the number and the frequency of IR examinations practiced in France will be undertaken. As far as the assessment of the risk attributable to these techniques is concerned, a selection of the most relevant categories of examinations will be made in collaboration with the other two European laboratories involved in the study.

Three different objectives will be pursued:

a) The quality control of the equipment.

The radiation quality of different types of radiological equipment used when performing a specific examination may often be responsible for large variations of both patient and occupational dose.

In order to reduce and interpret such variations, a quality control protocol will be commonly established together with the other two partners of the project and applied to a restricted number of x-ray units used within the context of the study.

b) The occupational exposure.

Recent international publications have clearly addressed the problem of well training and adequately monitoring all personnel taking part in interventional radiology procedures.

For the selected categories of examinations, individual occupational dose measurements will be performed following a common methodology established together with the other European laboratories.

A reasonable number of examinations of each category will be considered in order to get statistically significant dose figures. Particular attention will be paid to estimate individual Equivalent Dose values from monitoring devices worn outside or under the protective led apron.

c) The patient exposure.

An IR procedure may be of capital importance to the patient's life.

Practice of a procedure may generate highly variable exposure in relation to the duration of the examination and the patient's condition. Therefore, the individual dimension of the problem should be kept in mind when trying to evaluate the dose received by the patient.

A specific group of patients presenting comparable symptoms and treated with a similar protocol, may be of interest to approximate the range of patients' received dose and will be taken into consideration for the aim of the study.

Contribution of USL n°7 - Udine

1. Justification and purpose

Interventional Radiology (IR) represents one of the most recent applications of modern imaging techniques in radiology and in many cases it is a substitute for surgical intervention. It is applied to all anatomical regions of the body in order to make puncture-biopsies, angioplasties, embolisations and nucleolysis.

The procedures associated to interventional radiology require prolonged use of flouroscopy and multiple series of radiographs. In reason of the increasing diffusion of these procedures more concern is due to radiation doses delivered to both patients and medical and paramedical staff.

The main objectives of the study, made in collaboration with the Italian Association of Neuroradiology and the Institute of Neuroradiology of Udine hospital, is the evaluation in Italy of:

- frequency and type of IR procedures performed in neuroradiological practice,
- patient exposure in the different types of procedures,
- exposure of the staff involved in the practice.

2. Description and objectives

We propose to conduct the study along the following lines:

- in collaboration with the Italian Association of Neuroradiology, data on **type and frequency of IR examinations in neuroradiological practice** will be collected in a sample of neuroradiological institutes in Italy;

- for every examination performed **data on patient, staff, type of examination and technical parameters** used will be collected with a questionnaire;

- on a sample of Italian installation and for each type of neuroradiological procedure, an **evaluation of patient and staff exposure** will be performed.

Patient exposure will be evaluated measuring the total dose-area product quantity with a flat transparent ionisation chamber mounted on the tube housing and, for selected organs, measuring dose by means of TLDs attached to patient's skin. Organ dose and effective dose will be evaluated by means of Monte Carlo conversion factors.

Ranges of effective doses to the staff will be evaluated for each type of examination measuring doses with TL dosemeters attached on different parts of the body of medical and paramedical staff involved.

C23 Management of radiological protection in normal and accident situations.

Contract FI3P-CT920013b Evaluation and management of post-accident situations. Project 1: Database and decision-aiding techniques.

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Total Contribution by the Commission: 180 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Following an accident with radioactive releases affecting people and the environment, consequences must be assessed and countermeasures must be proposed. These two points rely heavily on availability and quality of the data, imply efficient computer codes, and need establishing managerial procedures. The project deals with data acquisition and decision aiding techniques for the management of countermeasures. The project is divided in two sections.

Section 1 : The EUROGRID data base

Objectives and expected achievement

To collect and organize data allowing the assessment of individual and collective doses, and of economical impacts of an accidental release of radionuclides at the european scale. This is a continuing task, new data being introduced in the base and old ones being updated.

Current state of the data base

Data files currently available concern populations, land uses, agricultural productions, livestock and number of people in the various socio-economic groups. These data are given for all the meshes of a grid covering the twelve european members of the European Community. The surface of each mesh is 10 000 km^2. For populations, data are also available for a one hundred time finer grid, obtained by dividing each side of the large meshes in ten equal parts. For an easy consultation of the data base, numerical and graphical softwares have been developed (QUERY, MICROSTATION). All data and softwares are usable on a PC micro-computer.

Degree of innovation

Such european data are annually published by EUROSTAT, on a regional basis. With such a presentation, data cannot be associated to dispersion and deposition models. For good use of these models, it is necessary to have the data at a finer level so that contamination can be assumed as homogeneous. Spatial units, like meshes regularly disposed in a grid, make the comparisons of risk and of impacts easier.

Economic, social and technical benefits

EUROGRID is needed to assess the collective and individual doses resulting from an accidental release. It also allows the evaluation of economical impacts of the accident and of the available countermeasures, so that action can be optimised. The interest of general data structures trough a grid is not restricted to radioactive contamination. EUROGRID can also be used in other circumstances than nuclear accidents, and is also relevant in case of chemical pollutions.

Relation to other EC RTD programmes

The EUROGRID data base is part of the data used in the COSYMA Accident Consequence Assessment code that is developed by KFK and NRPB under the auspices of the CEC. Parameters to be included in are selected with cooperation of these organizations.

Technical description

- Updating the data

Updating is necessary because most of the data are more than ten years old. It began in 1991 for populations at the both levels, 10 000km^2 and 100km^2 . It will pursue for agricultural and land use files on the basis of recent national agricultural surveys. These later statistical data allow to document both the large meshes and the small ones.

- Adding data : new parameters

More details are needed concerning the milk path, and particularly the milk products. Collected milk is the current parameter in EUROGRID; it does not permit accurate individual dose calculations because transformations and the delay between the collect and the consumption are not taken into account. To deal with this problem, milk products must be introduced.

An other demand concerns housing data in order to estimate doses due to inhalation and external exposure. To this end, parameters describing houses and living habits are necessary (i.e., types of housing, ventilation rates, time spent indoor and outdoor).
A feasibility study is in progress on the interest of including water data in EUROGRID. Depending on the conclusions of the study, data on water resources and uses could be introduced, but only for selected meshes characterized by high potential risk.

- Adding data : new countries

The recent political changes in Europe, and the assessment of risk for eastern nuclear power plants introduce the need for extending EUROGRID to new european countries. Firstly are concerned the east part of Germany and some east countries. Secondly, and for completion, Switzerland is to be introduced. Data from Nordic countries could be welcome too.

Section 2 : Decision Aiding Systems

Objectives and expected achievement

During the last ten years, interest in assessing the impacts of accidental situations and in the management of post accidental countermeasures has increased dramatically. There are now sophisticated computer codes for the different situations occurring. In

parallel, the search for expertise has been developed and experts have been involved in the many crisis exercices. Decision making revealed to be a crucial point, so decision aiding methods have been proposed. In practice these three aspects are used independently and not on an integrated mode. The goal of the proposed research is to study how the necessary relations between results from ACA codes, expert judgement, and decision aiding tools may be improved. To avoid unduly general considerations, a fruitful approach is to choose a particular context, as for instance the management of contaminated foodstuffs.

Degree of innovation

The proposed research emphasises the study of the links between ACA codes, decision aiding techniques, and expertise. This makes the originality of the project.

Economic, social and technical benefits

The development of integrated systems for decision aiding is of major importance to improve emergency planning and the management of post accidental situations. The methodological approach and the research proposed is intended to solve radiological problems; nevertheless it is of more general value because large tecnological accidents imply the same type of decisions.

Scientific description

Management of post accidental situations relies on the acquisition of exposure data (actual or calculated), the use of extended data bases, computation of the expected consequences, need for expert's judgements, and the use of procedures and/or methods to help and achieve decisions. It must be also emphasized that countermeasure implementation depends on the time scale: some countermeasures are adequate for a particular period of time, and not justified or possible for other periods. For instance, sheltering and evacuation are short term decisions when contaminated foodstuffs management is a medium or long term problem. In any case and at any time, there are several choices which involve many risk managers. Up to now there is no computer system integrating all the aspects of the crisis from data to decision; computer codes are existing only for some aspects.

Evaluation of countermeasures to be taken after a nuclear accident takes into account several categories of parameters. The first category includes dosimetric and economic data or results, and is necessary to perform cost-effectiveness analysis of the different countermeasures that can be envisaged. This analysis constitutes a first approach in optimization procedures.

A more sophisticated approach takes into account further parameters, that may be uneasy to quantify, as expert judgements.

The objectives of this proposal are: to achieve the DACFOOD system which consists in a cost-benefit analysis; to investigate methods allowing to account for expert judgements.

One main point will be to collect the expertise. This will be done using interviews : experts will be requested to give their opinions during simulations obtained by running the DACFOOD system, or others. Before introduction of the expert judgements in an operationnal system, it is necessary to investigate extensively the existing methods, and to understand why some decision methods are in use and others not. This point will be a major task of the project.

TASKS OF THE CONTRIBUTORS

a)Institut de Protection et de Sûreté Nucléaire

The Institut de Protection et de Sûreté Nucleaire (IPSN) is coordinator of this project and will contribute in the two sections of the project.

Section 1 : The EUROGRID data base

Updating the data

IPSN has achieved in 1991 the updating of the population data at the both levels, 10 000 km² and 100 km². It will poursue for agricultural and land uses files on the basis of recent national agricultural surveys. The part of IPSN in updating these data will cover all the EC countries, excepted the Iberic Peninsula.

Extension of the data base to other parameters and countries.

IPSN will collect other parameters (milk products, data concerning housing and living habits) for the EC countries, Spain and Portugal excepted. It is in charge of the on-going feasibility study concerning the interest of including water data (resources and uses) in EUROGRID. IPSN will investigate the availability of these data in other countries : the east part of Germany and some east countries, Switzerland and Austria, Nordic countries. All the collected data, both by IPSN and the Polytechnical university of Madrid will be organized and introduced in the data base by IPSN.

Section 2 : Decision Aiding System

The task developed by IPSN will mainly be oriented to achieve the DACFOOD system, which consists in a cost-benefit analysis, by improving the presentation and the interactivity of the system, using experience gained during the french nuclear crisis exercices.

It will also be responsible of the collect of the expertise. This will be done using interviews : experts will be requested to give their opinions during simulations obtained by running the DACFOOD system, or others. IPSN will participate in the comparison and evaluation of existing decision aiding methods, with special attention to the application of countermeasures following a nuclear accident.

b) Universidad Politécnica de Madrid

The University of Madrid (UPM) is involved in the section 1 of the project: With regard to the update and development of the EUROGRID data base, the UPM will collect data of the Iberic Peninsula and Balearic Islands to give them to the IPSN, based on the most recent surveys available in national statistics offices. Data referred to population, land uses, cultivated areas, vegetal productions, livestock and livestock productions including specifically milk and milk products, will be collected and distributed, when possible at both levels of the grid.

The availability of new parameters concerning housing data and living habits to be used for the assessment of doses due to inhalation and external exposure, will be investigated as well as the data referred to water resources and uses. Every effort will be made to colect useful informations, at least at the large meshes level.

c) Leeds University

The contribution to the decision aiding project from the Leeds university will be focused on the following points:

- Input on the use of the multi-attribute value/utility methods for comparison with the outranking/Electre method. The research would focus particularly on exploring the difference in perspective offered by the methods. It is suggested that decision makers should be offered as wide a range of informative perspectives as possible on their problems so that they can appreciate their choice more clearly.

- The development of the decision conferencing approach to use in these circumstances. Leeds University would attempt to trial the approach with groups of decision makers or at least, groups from the radiation protection community who understand the issues well anough to act as proxies for the decision makers

- The apply the sensitivity analysis developped by this laboratory in the context of decisions in post accident situations, to test the robustness of decisions.

d) Paris IX university

Paris IX university will mainly be concerned by the modelling stage using and adapting methodologies developped and tested in the laboratory. Some of the major points of interest will be :

- the precise definition of the set of alternatives : The set of alternatives consists of possible countermeasures in a specific context. Some difficulties occur at this level since a countermeasure is actually composed with a serie of elementary countermeasures which can be taken at different periods of time. Several interesting options are available when defining such rather complex alternatives. The choice of a specific option will largely influence the way of conducting the whole research.

- The construction of criteria : The evaluation and comparison of countermeasures should be based both on quantitative aspects (resulting e.g. from specialized codes which calculate the effects of some countermeasures) and qualitative ones (resulting e.g. from expert judgements). Considering these aspects, the purpose, at this level, is to construct a rather limited set of criteria which are sufficiently exhaustive but also easily understandable by decision makers faced with a situation of crisis.

- the imprecision of available data : The data to be used to evaluate the alternatives on each criterion are partly imprecise. Some techniques (based e.g. on fuzzy set concepts) must be developped to handle such imprecisions.

- the existence of multiple decision makers : The methodology must take into account different decision makers with specific interests. The decision to be made can be seen as resulting from a negotiation among these decision makers. Decision aiding tools or methods must be designed so as to favour such negociation.

In order to validate such a methodology, Paris IX university will test it on the specific context of the management of contaminated foodstuffs.

C23 Management of radiological protection in normal and accident situations.

Contract FI3P-CT930068 Assessment and management of post accidental situations. Radiation detriment, risk perception and risk communication.

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Total Contribution by the Commission: 160 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

Objectives and expected achievement

Risk perception and communication studies are essential for responsible risk management in that they provide : a) a basis for assessing the likely response of workers and the public to advice and information on radiation protection which is important for both normal (operating) conditions and abnormal situations; b) a route to appraising the effectiveness/efficiency of radiation protection; c) criteria and guidelines for coping with problems of risk attenuation and risk amplification.

The project proposed has two objectives :

(1) to undertake research in order to improve the understanding of risk perception in relation to radiation detriment ; and
(2) to evaluate current approaches in risk communication, and to develop appropriate strategies for radiation risk communication in the European Community (EC).

State of the art

For a long time, only scientists and managers dealt with the risks from radiation exposures. But the large scale expansion of nuclear activities, and particularly of nuclear power programmes, during the last two decades led to an upsurge in people's beliefs about risk, that has been extensively mediatized. This fact introduced the necessity to know more about reactions of the public. Attention given to the perceptions of radiation risk by people then increased.

Risk perception research has been initiated in the 1970's mainly by Slovic and co-workers, and it is still continuing. Dimensions of perceived risk have been set up, and tentatively used to understand acceptability and tolerability of risks by people. These dimensions point out what is of importance for individuals, and they give some guidance for risk communication. Risk communication studies have become a focus of concern more recently.

Risk perception and risk communication research activities are disparately spread throughout the Community. Much of the work has been concerned with the need of action following the Seveso directive. The amount of research effort directed at radiation detriment was lower. Nevertheless, there is a good source of related expertise within the Member States which may be drawn upon for further activities. So there is now a need to synthetize the results obtained and the experience gained.

Degree of innovation

The project emphasizes the synthesis and critical analysis of risk perception studies and risk communication practices in the domain of nuclear activities.

Economic, social and technical benefits

Recognizing that an efficient communication is a key point in the management of radiation risk, outputs of the project will include : a) recommendations for harmonization of risk communication programmes and policies; b) proposals for the improvement of risk communication in cross-cultural settings; and c) advice for the development of training programmes in risk communication.

Scientific description

The project includes five tasks.

Task 1. Review of risk perception studies.

Objectives : to evaluate the concepts of risk perception ; to assess their quantification in view of improving radiation risk communication; and to generate an appropriate reference framework for developing communication strategies.

Task : Risk perception associated with any hazard (situation, technology, or product) depends on the social representation of the hazard, especially when the hazard is not experienced bodily by individuals during their everyday life, e.g., as with nuclear power. Therefore the social representations of hazards play a significant role in public debates on radiation risks. However, each hazard has not only one social representation but many. This plurality of representations is due to many factors. Some factors are related to the person ; demographics, experience and skills, personality, social status, cultural level, as well as political positions. Other factors are related to the risk source : economics, institutional and legal constraints. So the first requirement is to clarify the social representations of radiation risk and the factors which may contribute to the amplification or attenuation of radiation risk perception.

The life cycle of social representations is also important. The changeability of a representation after particular events (such as catastrophes, political elections, or communication campaigns) must be taken into account. Case studies, e.g., on Chernobyl or on Sellafield, could be used to enlight the point. Furthermore, the debate about hazard issues can be better understood when embedding it in the context of other social debates.

At the individual level beside risk perception, coping strategies should also be taken into account; possible coping strategies range from apathy, no worry, avoidance, information seeking, changes in life style, inter alia. All these may be accompanied by manifestations of stress or somatic disordres. A clear understanding of these coping strategies is necessary for the development of better risk communication programmes.

Task 2. Inventory of risk communication approaches

Objectives : Identification of current approaches at Commission, national and regional levels within the European Community. Paying attention to information policy ; information networks, actors, strategies, and accomplishments.

Task: Collection of information about existing programmes. Two of the most important areas of concern reflect the potential for attenuation and amplification of risk. There are the problems of radon in the home and radioactive waste disposal respectively. Experience in communicating the risks of radiation exposure for these two areas should be collated and analysed.

There are well documented risk communication studies of the radon problem in the United States - both from the efforts of the US Evironmental Protection Agency and individual State Departments of Nuclear Safety. Similarly, the Swedish Institute for Radiation Protection (SSSI) has promoted radon protection which deserves careful assessment, and an inventory of European Community approaches should be pursued.

Experience of radioactive waste disposal programmes in the Member States of the EC is well documented. However, very little of this information relates specifically to risk communication practices. There is some good associated experience of risk communication from toxic waste disposal and incineration programmes which mirror the amplification problem and attention should be paid to these also.

Finally, further critical appraisal of the effectiveness of Commission activities in the field of risk communication, training, provision of advice and information is needed.

Task 3. Review of evaluation techniques for risk communication programmes.

Objectives : (1) identification of key risk communication evaluation techniques implemented to date inside and outside the Community (for example, in Sweden, by the Environmental Protection Agency (US), inter alia); (2) review of evaluation studies on risk communication in the field of radon and radioactive waste.

Task : a key problem is to identify criteria and techniques for the effectiveness of risk communication.

Three general points are relevant for the evaluation of risk communication programmes: (1) **Content-orientation** : the focus is on the substantive characteristics of the information communicated to the target audience ; (2) **Outcome-orientation** : final results and consequences of the employed activity/programme are to be measured; (3) **Process-orientation** : all stages of the intervention and the development of effects are surveyed.

A comprehensive collation of available evaluation studies on risk communication should be undertaken. The aim is to identify the factors influencing risk communication effects. Beside the analysis of existing evaluation studies, a survey on national and regional risk communication programmes in the EC should be conducted to identify informal evaluation efforts. The survey would assemble information necessary for comparing which kinds of communication strategies are judged to be effective by different organisations within the EC.

Output of this task is data sources and information for Tasks 4 and 5.

Task 4. **Assessing organisational factors influencing social learning in radiation protection**

Objective : Assessment and appraisal of current learning capabilities for anticipating and responding to public and workers' concerns in relation to radiation protection.

Task : The question "Under what conditions is learning about radiation protection progressive?" is a matter for empirical research. An inclusive view of what can be learned, certainly includes facts and skills, but also extends to theories, expectations, values and norms. The focus of the study is on how the facts, expectations and values to the management of risks evolve through processes of innovation, selection, and diffusion. Accepting the possibility of social/organisational (as opposed to merely individual) learning, we can set out to discover which organisations and social groups are involved in which aspects of the risk communication process. The study therefore accepts the important role of domestic and Community institutions in shaping risk management via processes of communication.

An essential part of preparedness consists of the ability of responsible institutions properly to represent needs and expectations in the public. It is therefore instructive to assess the fit between the values, norms and expectations of those institutions and of those existing or likely to emerge in the general public. Two empirical approaches offer themselves.

The first would be to employ psychometric techniques (e.g. SYMLOG ratings) on interview data to map the values carried out through into institutional action on radiation protection/risk communication in relation to those identified in sections of the general public. This provides evidence of social learning process within the responsible institutions and provides information for training purposes.

The second would be to employ interview techniques, policy analysis and content analysis of communication materials to establish the predominant organisational form from a cultural theory perspective. Again such an approach audits the responsible organisation to determine the driving social and organisational factors which influence the form and effectiveness of risk communication activities.

Task 5. **Identification of strategies for coping with risk amplification and attenuation problems**

Objective : To propose a communication policy for the responsible organizations in the public debate on radiation risks.

Tasks : This topic will seek to integrate the research findings (task 1 to 4) and draw conclusions. The focus is on the development of guidelines for risk communication concerning the performance of specific risk management functions. These are :

(1) Risk characterisation, that is, understanding the nature, cause, and consequences of radiation risks;

(2) Risk evaluation, that is, the position of the radiation risks being studied relative to each other and to other risks on the current agenda;
(3) Safety measures, that is, what are thought to be possible solutions to the radiation risk management problem;
(4) Code of conduct, that is, standard setting, rule following, enforcement, and optimisation of risk management.

Outputs are guidelines for policy and training.

Research Network for Risk Perception and Communication

Given the need to develop, exchange, apply and evaluate an array of existing information and expertise on risk perception and communication throughout the Community, the research project should reinforce its working links within the European research community. This will involve:

- a research-information network;
- a series of workshops to disseminate the work and findings from each project task within the wider network of European researchers and organisations responsible for radiation protection.

Distribution of tasks

Task 1.	Review of risk perception studies
Task 2.	Inventory of risk communication approaches
Task 3.	Review of evaluation techniques for risk communication programmes
Task 4.	Assessing organisational factors influencing social learning in radiation protection
Task 5.	Identification of strategies for coping with risk amplification and attenuation problems

Contribution of Institut de Protection et de Sûreté Nucléaire (IPSN- FRANCE) Laboratoire de Statistique et d'Etudes Economiques et Sociales (LSEES).

The Laboratory will coordinate the research project and will participate in Tasks 1, 4 and 5. In Task 4 will be analyzed how responsible institutions meet public demands and expectations in radiation information; data on how institutions are perceived, i.e. on their social representations, will be used.

Experience of the Laboratory

The Laboratory of statistics and socio-economic studies has large experience in the analysis and systematic follow-up of public opinions relative to nuclear energy, and more generally to practices involving radiation. Since 1977, national representative surveys have been performed in France dealing with perception of radiations, of their uses, and with acceptability of nuclear energy. Hazardous activities are studied by opinion polls within a comparative approach, using people beliefs about risks, benefits, and credibility of the safety systems (both the measures and the participating organisations). Different people have been interviewed: lay people in most cases, but also farmers, safety experts, and nuclear workers. Attention has been given to nuclear events, as the Chernobyl accident, and their impacts in public opinion characterized by major changes in perceptions and attitudes. Conflictual issues raised by the siting of nuclear installations have been studied too, by interviewing individuals in the communities concerned and by press-content analysis.

During the last ten years, risk research in the Laboratory focused on the conflictual nature of major hazards, both in their assessment and management. This point of view introduces social and cultural aspects, some of them being risk specific, others not. Present research in the LSEES is how to use in risk communication the previous aspects and others related to people knowledge and information on risks.

Contribution of Institut für Soziologie (IFS) der RWTH-Aachen GERMANY

Role and contribution of RWTH-Aachen

The main will be to do a review of evaluation techniques for risk communication programmes, i.e.

- identification of key risk communication techniques implemented in countries inside and outside the Community (Germany, France, Sweden, USA);
- review the evaluation studies on risk communication in the field of radon and radioactive wastes in countries inside and outside the Community.

Another task of IFS will be to:

- identify criteria for the effectiveness of risk communication processes in the selected fields; and based on this
- to develop techniques for the improvement of risk communication.

The Institut IFS will also conduct a survey on national and regional risk communication programmes in the selected fields and also in related fields by case studies. Aim of this part of the project done by IFS is to assemble informations about what kinds of communication strategies are judged to be effective by different organisations in the Community in different fields of risk communication.

Based on the experiences of the conducted studies and surveys mentioned above, the Institut will furthermore take part in the identification of strategies for coping with risk amplification and attenuation problems together with the other teams.

Contribution of the Stockholm School of Economics-Center for Risk Research, SWEDEN

Sweden is an EFTA country. The contribution towards the Stockholm School of Economics-Center will be published in an addendum after signature of the Association Agreement.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT920023 CEC/USNRC joint project on uncertainty analysis of probabilistic accident consequence codes.

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Total Contribution by the Commission: 160 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

The objectives of the proposed joint project are to further develop and apply expert judgement elicitation techniques in estimating the uncertainties associated with the predictions of probabilistic accident consequence assessment codes, and to investigate the use of the results of these studies as input to uncertainty analyses of such codes.

Within the project the following contributions will be made:

(i) To further develop methodological backgrounds and implementational aspects of expert judgement elicitation techniques to be applied in the joint project (TUD contribution).

(ii) To further develop mathematical techniques for handling modelling uncertainty (TUD contribution).

(iii) To perform modifications and extensions of the System for Uncertainty and Sensitivity Analysis (SUSA), that emerge from methodological improvements, for instance from (ii), as well as from the practical applications of the system to results from the COSYMA and possibly MACCS codes (GRS contribution).

(iv) To provide the necessary detailed information on the relevant parameters and models, in order to:

(a) assist in the selection of appropriate experts in specific subject areas of consequence assessment (NRPB-KfK-SRD-ECN contributions),

(b) provide experts both in the general area of accident consequence assessments, and in particular modelling aspects, who will interact with and advise the chosen experts in other fields (NRPB-KfK-SRD-ECN contributions),

(c) consider the implications of the results of the expert elicitation with regard to an uncertainty analysis of the COSYMA code (NRPB-KfK contributions).

The following subject areas will be investigated:

- Atmospheric dispersion and deposition (KfK-NRPB)
- Behaviour of deposited material and related doses (SRD-NRPB-ECN)
- Plume rise (KfK-SRD)
- Internal dosimetry (NRPB-SRD)
- Early health effects (ECN-KfK)
- Late health effects (NRPB-ECN)
- Foodchain modelling (NRPB-KfK).

It has been decided to commence the project with the area of atmospheric dispersion and deposition (November 1992 - November 1993).

The project is being undertaken jointly by the CEC the USNRC (United States Nuclear Regulatory Commission). CEC project manager is Louis Goossens, USNRC project manager is Frederick Harper (Sandia National Laboratories, Albuquerque, New Mexico, USA).

Contribution of EC participants.

Delft University of Technology (TUD), Delft, the Netherlands

(1) The further development of the expert judgement elicitation techniques will be performed by both the Safety Science Group and the Dept of Mathematics and Informatics of TUD. Such techniques have been developed in the recent years by TUD. The major aspects for the implementation in this joint project are the problem identification phase, the expert identification phase, the question formulation phase and the seed variables selection phase. The methodological aspects and the implementational features will be delivered and applied by TUD supported by detailed computer code information on the parameters from the participant institutes NRPB, KfK, SRD and ECN, and the USNRC efforts in this field.

(2) The further development of mathematical techniques for handling modelling and parameter uncertainty will be performed by the Dept. of Mathematics and Informatics of TUD.
The key to this approach is that distributions over modeling parameters should be fitted to subjective distributions over hypothetical experiments. The method can be sketched as follows:

1). Modelers specify which experiments they would perform, supposing these to be possible, to determine the values of parameters in their codes. The experiments must be physically possible in every intended application of the code. This step requires the modelers to think carefully about the meaning of terms in his/her model.

2). Subjective probability distributions over possible outcomes of the indicated experiments are obtained via existing expert judgement techniques.

3). Distributions over the parameters in the model are fit to these subjective distributions.

The resulting distributions over the modeling parameters reflect the uncertainty in predicting results of experiments with the given models. Hence they reflect both the uncertainty in the models and the uncertainty in the values of the parameters of the models. The fitting problem in step three above is similar to yet distinct from traditional problems in which parameter values (not distributions) are fit to empirical distributions and the quality of the fit is assessed.

The basic mathematical techniques have been worked out and a research grade code has been written to implement these ideas. Results are encouraging and demonstrate the

feasibility of the above approach. However, many problems of a purely mathematical nature remain. In particular, optimal fitting (step three) can only be done when the subjective probability distributions and the model's parameter space are discretized. The manner in which this is done can influence the results significantly, and a method for optimal discretization under reasonable constraints must be found.

GRS, Garching, Germany

GRS will contribute to the study in the following ways:

i) To provide the system for uncertainty and sensitivity analysis (SUSA) and to assist in establishing the link between SUSA and the accident consequence assessment code (or module) subject to analysis;

ii) To perform modifications and extensions of SUSA that emerge from methodological improvements to be achieved by this project as well as from the actual application of the system to results from the COSYMA and possibly MACCS codes.

National Radiological Protection Board (NRPB), U.K.

NRPB will contribute to the study in the following aspects:

i) To assist in the selection of appropriate experts in specific subject areas of consequence assessment (see below)

ii) To provide experts in both the general area of accident consequence assessment, and in particular modelling aspects (see below), who will interact with and advise the chosen experts in other fields.

iii) To assist KfK in considering the implications of the results of the expert elicitation with regard to preparation for an uncertainty analysis of the COSYMA code.

NRPB will contribute, as identified in (i) and (ii) above, to the following three subject areas:

- Internal dosimetry (joint responsibility with SRD)
- Late health effects (joint responsibility with ECN)
- Foodchain modelling (joint responsibility with KfK)
- Atmospheric dispersion and deposition (joint responsibility with KfK)
- Behaviour of deposited material and related doses (joint responsibility with SRD and ECN)

Kernforschungszentrum Karlsruhe GmbH (KfK), Germany

KfK will contribute to the study in the following aspects:

i) To assist in the selection of appropriate experts in specific subject areas for consequence assessment (see below)
ii) To provide experts in both the general area of accident consequence assessment, and in particular modelling aspects (see below), who will interact with and advise the chosen experts in other fields.
iii) To consider, with NRPB, the implications of the results of the expert elicitation with regard to an uncertainty analysis of the COSYMA code.

KFK will contribute, as identified in (i) and (ii) above, to the following three subject areas:

- Atmospheric dispersion and deposition (joint responsibility with NRPB)
- Plume rise (joint responsibility with SRD)
- Early health effects (joint responsibility with ECN)
- Foodchain modelling (joint responsiblity with NRPB)

The methodological basis for uncertainty assessments will be established and preparations will be made for uncertainty and sensitivity analyses with COSYMA based on the results of the expert judgement elicitation process.

Uncertainty and sensitivity analyses of the program system UFOMOD have been performed at KfK for several years. This experience may build an important input to the planned expert judgement elicitation project and the subsequent analyses with the program package COSYMA, commonly developed by NRPB and KfK.

Safety and Reliability Directorate (SRD), AEA Technology, U.K.

SRD will contribute to the study in the following ways:

i) To assist in the selection of appropriate experts in specific subject areas for consequence assessment (see below)
ii) To provide experts in both the general area of accident consequence assessment, and in particular modelling aspects (see below), who will interact with and advise the chosen experts in other fields.

SRD will contribute, as identified in (i) and (ii) above, to the following two subject areas:

- Behaviour of deposited material and related doses (joint responsibility with NRPB and ECN)

- Plume rise (joint responsibility with KfK)

- Internal dosimetry (joint responsibility with NRPB)

Netherlands Energy Research Foundation (ECN), Petten, the Netherlands.

ECN will contribute to the study in the following ways:

i) To assist in the selection of appropriate experts in specific subject areas for consequence assessment (see below)
ii) To provide experts in both the general area of accident consequence assessment, and in particular modelling aspects (see below), who will interact with and advise the chosen experts in other fields.

ECN will contribute, as identified in (i) and (ii) above to the following two subject areas:

- Early health effects (joint responsibility with KfK)
- Late health effects (joint responsibility with NRPB)
- Behaviour of deposited material and related doses (joint responsibility with SRD and NRPB).

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT920036 Development of a comprehensive decision support system for nuclear emergencies in Europe following an accidental release to atmosphere.

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Total Contribution by the Commission: 570 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

Within its 1990/1991 Radiation Protection Research Programme, the Commission of the European Communities (CEC) has embarked on a major project aiming at the development of an integrated and comprehensive real-time on-line decision support system (RODOS) for nuclear emergencies in Europe applicable for the vicinity of the release to far distant areas and able to make consistent predictions unperturbed by national boundaries. At present some 18 European institutes are involved in the joint development of RODOS, which is being carried out within the framework of 3 sub-projects, dealing with

(1) analysis and forecast of meteorology and atmospheric dispersion in real-time systems from local to European scale (contract No. FI3P-CT92-0044).

(2) development of hardware and software configuration, acquatic and terrestrial transfer, modelling of countermeasures and consequences (this contract).

(3) decision aiding techniques for evaluating countermeasure options (contract No. FI3P-CT92-0012).

All three projects will be fully integrated.

A collaborative programme has also been initiated with institutes in the Community of Independent States (CIS) within the framework of the agreement between the CEC and Belarus, Russia and Ukraine on investigations of the health and environmental effects of the Chernobyl accident (contract No. COSU-CT92-0020).

One of the main benefits of these coordinated R&D activities within the RODOS project is that they will facilitate the allocation of limited resources to those areas where they are most needed, for example methodological problems and modelling weakpoints where data and information are missing. In addition, they will also help to minimise the development of many different systems, which may provide information of different kinds and quality, and to harmonise methods and models used in already existing or planned decision support systems.

For these benefits to be attained, the overall structure and objectives of the RODOS system have been specified and accepted by all contractors involved in the first phase of the project. Its basic features can be summarized as follows:

As decisions are required from the very early stages of an accident up to many years after the release of radioactive material, RODOS should operate in real-time and on-line coupled to meteorological and radiological data networks. For aiding decisions on early emergency actions in the near field, such as sheltering, evacuation and distribution of stable iodine tablets,cycle times of about 10 minutes are considered to be acceptable. Decisions on countermeasures in the later phases of an accident (i.e. after the passage of plume and/or in the far field), such as relocation, decontamination or food-bans, are less urgent, so that longer computing times are unproblematic.

The dialogue between RODOS and the user should be possible in both an automatic and an interactive mode. In the early emergency situation, a pure dialogue system would probably be inadequate; because of stress and psychological pressure, it could create confusion, nervousness and mistakes. Therefore, the system should offer the possibility to automatically present all information relevant for decision-making. Early accident phase interaction with the system has to be limited to a minimum of user input, i.e. just that is necessary to characterize the actual situation and to adapt models and data to reality. At later stages of the accident, when longer-term countermeasures have to be considered, and quick decisions are no longer necessary, the interactive communication with the systems becomes more important.

Additionally, RODOS should be a comprehensive system, which integrates models and data for assessing, presenting and evaluating the accident consequences in the near, intermediate and far distance ranges under due consideration of the mitigating effect of countermeasure action. It should be operationally flexible to cope with differing site and source term characteristics, differing amounts and quality of monitoring data, and differing national regulations and emergency plans. Its potential application all over Europe requires the software to be developed as a transportable package; in particular it has to support the integration of external programs developed by many of the contractors. Therefore, the structure of RODOS has to be modular in form, allowing an easy exchange of models and data, and thus facilitating the adaptation of the system to the accident situation. Finally, the system has to provide a variety of access tools to cope with the different capabilities, knowledge and aims of the potential users.

This flexibility, together with the various modes of operation, would allow the system to be used not only in actual accidents, but also as a powerful tool for education and training of personnel involved in the decision-making process and in preparing and exercising emergency plans.

Based on the key features and the potential applications of RODOS set out above, the following conceptual structure has been developed. RODOS comprises three main subsystems, which are controlled by an operating subsystem OSY:

(1) analysing subsystem ASY:
continuously updated estimation of the present and future environmental distributions of activity concentrations and derived doses/dose-rates together with monitoring/measurement data and the uncertainties in the absence of countermeasures;

(2) countermeasure subsystem CSY:
quantification of the benefits and drawbacks of alternative courses of actions, such as individual/collective doses, health effects and economic costs, areas and number of people affected, together with the technical and personnel aid required and the associated uncertainties;

(3) evaluating subsystem ESY:
judgement of alternative courses of actions under the aspects of practicability in the actual situation, acceptance by and behaviour of the population, and socio-political implications.

Each of the subsystems has a modular structure to enable an easy exchange of models, algorithms and data sets. The detailed parameterization of calculational procedures implemented in the various models will allow the tuning of the system to consider differing

- amounts and quality of measured meteorological and radiological data,
- environmental, demographic and economic information of the region considered,
- national regulations, emergency plans, decision-making structures, and needs of the user.

The division of RODOS into the three subsystems ASY, CSY and ESY should be considered as a conceptual design. To ensure an effective execution of the calculations, the actual software structure will consist of a variety of submodules, each of them developed for a specific type of calculations. The development of each module can be accomplished for the most part independently, thus facilitating the specification of project structures and task descriptions.

An important task of the first phase of the project was the development of the structures and the hardware and software framework of RODOS. The first prototype version has been realised in autumn 1992 as a transportable software package on UNIX workstations, which integrates programs developed by contractors, in particular the complete software of the German real-time system RESY. They comprise models for near range atmospheric dispersion, early emergency actions, dose calculations, food-bans, health effects and economic costs. The RODOS prototype was presented during the 3rd CEC workshop on Decision Making Support for Off-site Emergency Management, Schloss Elmau, Bavaria, October 25 - 30, 1992.

Much R&D work has still to be done before the goal of a comprehensive decision support system for application in Europe will be achieved. Two main areas can be distinguished in the research programme of the sub-project (2) described in this summary:

A. Further development of the hardware and software framework, and improvement and completion of the model and data ingredients.

B. Application of the system for training and exercises.

Topic A comprises connecting the system to on-line networks of meteorological information and monitoring data, developing data assimilation methods to approach reality by making best use of both model predictions and measurements, providing simulation models for all kinds of countermeasures and their possible combinations together with appropriate models for assessing doses, health effects and costs, quantifying the uncertainties of the various endpoints presented by the system, and further investigating the applicability of decision-aiding techniques to facilitate quantitative comparisons between options of countermeasures strategies by evaluating all kinds of consequences.

To build a system, which is relevant for practical decision-making, and which is capable of being broadly accepted, it is essential to establish an early involvement in the development of RODOS of people engaged in the decision-making process. To that purpose the use of the system in emergency exercises as well as in the organisation of training courses is a key issue for the next development phase (Topic B). This includes the development of illustrative scenarios, the preparation of material for exercises and training courses, and their evaluation.

The detailed descriptions from the partner organizations contributing to this project are attached. The principle work areas of each organisation and the collaborative links between these areas are as follows:

KfK

Further development of the hardware and software framework of RODOS, supported by a subgroup of contractors with expertise in this area, including the University of Leeds, NRPB, EdF and SCK/CEN. Incorporation of software products provided by the contractors, including simulation models for early emergency actions (sheltering, evacuation, issue of stable iodine tablets) under improvement and completion at KfK. Continuation of work on the expert system for ranking alternative countermeasure strategies developed at KfK. On-line link of RODOS and preparation of exercises and training courses together with EdF, Nuclear Electric, Mol.

EdF

Support in the on-line operation of RODOS using real-time meteorological data as well as data from simulation exercises. The scenarios cover a scale of several kilometres up to the regional level. Based on its experience in the management of accident situations, EdF will help to further develop the system with respect to the needs and requirements of the decision-makers in a nuclear emergency.

GSF

Further development of the dose modules for external and internal exposure, in particular foodchain and ingestion pathways, in order to meet the requirements of the countermeasure models under improvement at NRPB and KfK. Parallel investigations are aimed at a methodology for providing uncertainty estimates of the results obtained with the foodchain and dose models under real-time conditions.

University of Leeds

Use of Bayesian modelling techniques to forecast and handle uncertainties in RODOS. Work already started on assimilating monitoring data in plume forecasts will be continued and expanded to other modules. Development of appropriate forms of presenting uncertainties to non-numerate decision-makers. Support with extensive knowledge and expertise on decision analysis and software engineering.

SCK/CEN Mol

Development of data assimilation methods for atmospheric dispersion and deposition predictions based on both mathematical procedures of fitting model parameters and expert judgement of graphically presented measurements and model predictions. The usefulness of fuzzy logic to cope with the variability of observatons will be investigated. Assistance will also be offered to KfK and the other contractors in defining the system requirements, based on the experience with monitoring networks and training courses.

NRPB

Development of models for assessing the consequences of long-term countermeasures, such as relocation, food interdictions, and decontamination. This includes the consideration of different intervention criteria, the feasibility of measures, and the interaction with other modules developed by GSF and KfK or other contractors.

Nuclear Electric

Advice and support will be provided in the development of illustrative scenarios and course material to be used for training and exercising those involved in the decision-making process. From the expertise gained a systematic approach to training will be followed in the preparation of the relevant training material associated with RODOS project.

Participation of Kernforschungszentrum Karlsruhe GmbH (KfK)

Objectives and Programme of work

Within the first phase of the project, KfK has outlined the basic concept of RODOS, laid down the modular structure of a detailed hardware and software framework, and finally realised its first prototype version in accordance and continuing cooperation with the contract partners.

Main objective of the next Research Programme will be to take benefit of an extended effective cooperation of competent organisations in aiming at the further development of the existing system to an operational flexible and comprehensive decision support system applicable within the EC member states and other European countries in the case of a real accident as well as for education, training and exercises.

To achieve these goals, KfK work will concentrate on the following topics:

(1) Further development of the hardware and software framework including the graphics subsystem of RODOS.

2) Incorporation of software products and data provided by the contractors, test of their functions, and generation of the appropriate user interfaces, in close cooperation with the developers.

(3) Further development of simulation models for emergency actions in the early phase (sheltering, evacuation, issue of stable iodine tablets), adaptation of models for estimating economic costs and health effects to the needs of the sytem, and completion of data bases with geographical and economic information.

(4) Continuation of work on the already existing rule-based system, which also contains elements of multi-attribute decision analysis.

(5) Connection of the system with on-line networks of meteorological information and radiological monitoring data to achieve its full function.

(6) Generation of illustrative scenarios for training and exercises, preparation of systematic course material, and evaluation of the experience and response with respect to improving the endpoints of RODOS and their presentation.

As coordinator of this RODOS project and the Joint Study Project 1 of the agreement between CEC and Belarus, Russia and Ukraine, KfK will organise a close co-operation between all contractors and the other two RODOS sub-projects to achieve an equal-balanced development of all parts of the system.

Participation of Electricité de France, EdF

Background and Programme of work

As part of its operation of an installed base of nuclear power plants comprising 56 units, Electricité de France has implemented a structure and a method designed to manage accident situations in compliance with the recommendations of the International Commission on Radiological Protection (ICRP).

EdF's structure performs the following functions: diagnosing the accident, weather analysis, staff protection, first-aid for wounded and contaminated persons, analysis of radioactive releases, and providing information to the local and national authorities.

EdF carries out simulation exercises to maintain and improve its operational crisis organization. These exercises are based on accident scenarios which call on nuclear facility simulators, and use preset data for extreme situations.

Within the scope of the European decision support system project RODOS, EdF is offering its experience in operating models using real-time meteorological data, as well as supplying data from its simulation exercises to enable comparison of the different systems.

Based on its experience in implementing the crisis structure and improvements due to the simulation exercises, EdF is also offering its expertise in this field to help define and apply a decision support system in case of a nuclear accident in Europe.

Participation of GSF-Institut fuer Strahlenschutz

Objectives and Programme of work

During the first phase of the RODOS project, GSF has developed the modules for estimating the transport of radionuclides within foodchains as well as for the assessment of internal and external doses within the analysing subsystem ASY. These modules are also used for the dose assessment considering alternative courses of actions within the countermeasure subsystem CSY.

In the present phase of the project, the following tasks will be performed:

(1) Further development of the foodchain transport and the dose modules in order to meet the requirements of the RODOS system. Since these modules, and the countermeasure modules as well as the operating system OSY are under parallel development by different institutes, a close collaboration with those groups is necessary who are developing OSY and the countermeasure modules. The main tasks will be:

- Further adaptation of the modular structure of the programs in order to apply them in the automatic and interactive mode of the analysing and the countermeasure subsystems.
- Model extensions in order to allow dose assessments considering various countermeasures not considered in the system up to now.

(2) Development of a methodology of providing uncertainty estimations together with the results of the foodchain transport module and the dose module. Since "classical" uncertainty analysis as e.g. Monte-Carlo calculations can not be performed in the real-time application of the system, other techniqes of providing information on uncertainty have to be developed. This is done in performing Monte-Carlo calculations considering the uncertainty ranges of the model parameters for a number of scenarios. The resulting uncertainties will then be parameterized with respect to the type of result, radionuclide, foodstuff, time of deposition, etc. and fitted by empirical functions. These functions can be used in the real-time application of the system for a fast estimation of the uncertainty of the model results.

The main task will be to develop the basic methods to be applied, to develop a prototype version of the respective modules and to identify remaining problems.

Participation of The School of Computer Studies, University of Leeds.

Objectives and Programme of Work

In collaboration with the Statistics Department at the University of Warwick, the School of Computer Studies at the University of Leeds will be contributing to the methodology for modelling uncertainty used in RODOS. Specifically, the two prime aims of the contribution are:

* The development of Bayesian modelling techniques to forecast and handle uncertainties in RODOS. Initially, this will focus on the inclusion of uncertainty measures within the plume forecasting module (using the RIMPUFF model) of the ASY subsystem. In addition, methods for the assimilation of monitoring data will also be developed and tested. Specifically the following will be investigated and implemented:

- assimilation of monitoring data;
- estimation of release height;
- reporting of means and variances of predictions at given sites;
- estimate the source term with allowance for autoregression;
- make allowances for modelling error;
- consider the computational advantages of working on a variable grid.

* Subsequently, attention will widen to consider uncertainty measures in the forecasting of the efficacy of countermeasures. We will consider passing appropriate measures of uncertainty between modules in RODOS and providing an audit trail to justify the predictions made by the system.

* The development of methods to communicate uncertainty to non-numerate decision makers.

In addition to the above, Leeds and Warwick will also contribute general expertise on decision analysis, statistics and uncertainty modelling, numerical methods and software engineering.

Participation of Studiecentrum voor Kernenergie (SCK/CEN)

Objectives and Progamme of Work

A. Given the division of RODOS in three subsystems ASY, CSY and ESY and considering the requirements of the respective subsystems, the main programme of work of the SCK/CEN within this project releates to the development of a module in ASY to obtain a reliable picture of the radiological situation in the near surrounding of a nuclear facility following an accidental release to atmosphere. To achieve this goal the initial uncertainty of the main variables governing the radiological assessment (source term, wind direction, plume height,...) has to be reduced as quick as possible to an acceptable value.

During previous activities related to this project, the SCK/CEN has performed a study, mainly based on document tracer experiments, to compare the feasibility of implementing several methodologies. One cornerstone of the system will be the realization of a modules to assimilate early environmental survey data and to process them by means of an atmospheric dispersion and dose assessment model.

First a protocol concerning environmental data assimilation (spatial and temporal acquisition of nuclide dependent air concentrations and surface contamination and of gamma dose rate measurements) will have to be worked out, given the boundary conditions of the general operating system (OSY). This has to be considered together with an adapted environmental sampling scheme.

Secondly, a mathematic procedure will be implemented to reduce the radiological uncertainties. Given the complexity of the task, a quite simple general dispersion model (e.g. segmented plume Gaussian model) will be at the base of it, in order to concentrate efforts on the most important parameters.

In an on-going process, a combination of straightforward physical principles to extract fundamental information and of a comparison of model predictions versus observations to adjust the main model parameters to their optimal value will be applied. The introduction of fuzzy set theory to infer information fom fuzzy quantities is promising to be a valuable tool in the process.

B. In collaboration with KfK, EdF and Nuclear Electric, the SCK/CEN will help to develop criteria to extend RODOS for use in existing or future training courses and to define specific scenarios in these training courses for which RODOS can be applied.

Participation of The National Radiological Protection Board, (NRPB)

Objectives and Programme of Work

The work of NRPB within the current phase of the project is concerned with the treatment of countermeasures in the RODOS real-time system. The principal objective is the development of a module of RODOS for assessing the consequences of long-term countermeasure options.

As a part of the RODOS project, a module - ECOAMOR - for the assessment of doses from accidental releases of activity is being developed at GSF in Germany. At NRPB, a module for predicting the effects of longer term countermeasures is being developed, using input from other modules of the system, and primarily from ECOAMOR. This countermeasures module is called FRODO.

The countermeasures to be considered in FRODO will be: relocation, food countermeasures, and decontamination. The effects of these countermeasures considered in FRODO include the dose saved by introducing the measure, the time over which the countermeasure would be in force, the areas of land and numbers of people affected, the amount of food affected by restrictions.

During the work, the interaction with the dose assessment module ECOAMOR, the decision-aiding aspects of the system and the economics module of RODOS will be considered. Hence, the primary interactions with other contractors will be with GSF in connection with ECOAMOR, and with KfK in connection with other aspects of the system.

Participation of Nuclear Electric

Background and Objectives

Nuclear Electric, as the operator of 24 power reactors at 10 sites in England and Wales has, for the past 30 years had in place effective arrangements for response to a major accidental release of radioactivity from one of these sites. As part of the Company's policy of continually reviewing and refining its emergency arrangements, considerable effort has been put into improving the way in which post accident monitoring and other information is gathered, collated and presented so as to aid the decision making process. Use is currently made of various predictive codes and computerised information handling systems which are currently being further developed. Within this context Nuclear Electric intend to contribute to the development of the CEC decision support system RODOS by bringing to it the practical needs of a potential user of the system and our considerable experience in emergency scheme training and exercising.

Our objectives will be to ensure that:

1. advice and support is provided in matters relating to training and exercising;

2. the many practical lessons arising from Nuclear Electric's significant exercise programme and experience in scenario development are fully taken into account in the development of training scenarios;

3. that a systematic approach to training is utilised in the production of the relevant training material associated with the project.

Programme of Work

Provide advice and support to the project in the development of course materials and scenarios to be used for training and exercising staff in the use of the system.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT920038 Deposition of artificial radionuclides, their subsequent relocation in the environment and implications for radiation exposure.

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Description of research work

BACKGROUND

Typically, deposited activity plays a major role in the radiological consequences of atmospheric nuclide releases. For example, external exposure from deposited ^{137}Cs would contribute significantly in many cases to the lifetime doses to members of the public from anthropogenic radionuclides. A contribution to dose, that has not yet been considered adequately, arise from ß-emitting aerosols deposited on human skin. Also the intensity, spatial distribution and subsequent fate of the deposit in urban environments must be adequately addressed. In the past there has been a lack of information to enable the particular characteristics of weathering in the urban situation to be explicitly taken into account. For example, external exposure in many urban scenarios is strongly influenced by migration into the soil of caesium deposited on grassed areas; this has not been investigated sufficiently well hitherto.

A wide range of potential weather conditions has to be considered in consequence assessment, and it is necessary to ensure that there is adequate treatment of those particular situations that lead to high deposits, such as heavy rain, snow or fog. Although causing relative high deposition rates, the last two have not usually been treated explicitely in consequence assessments to date since adequate models and parameterisations are not available.

Experiments performed with monodisperse artificial aerosol to examine indoor air concentrations and depositions have so far used particles with sizes from two to four micron. As the particles from the Chernobyl accident deposited in Western Europe had a size from 0.1 to 5 μm it is important to extend the size range to the sub-micron region. In the sub-micron size range, additional phenomena determine deposition behaviour: diffusion, electrostatic influences, thermophoreses and other processes. New techniques for labelling artificial submircron aerosols, as surrogates for natural aerosols, should permit this.

The technology of producing, sizing and releasing such particles differs from that for the larger silica particles used so far, but is completely feasible. Deposition experiments using these sub-micron particles should fill the gap in deposition data concerning sub-micron particles.

So far, deposition experiments have been done in three different houses, two in Denmark and one at the Building Research Establishment, BRE, in UK. Data obtained from measurements in other houses and under a wider range of conditions are needed to improve the reliability of the parameters. To allow a description of the contribution to the external gamma dose from indoor deposited material experiments demonstrating the distribution of indoor deposition as a function of size, shape and furnishings need to be performed.

Not all material deposited in the environment remains in its initial deposit location. Resuspension of deposited activity may recontaminate cleaned areas and there are still many unanswered questions regarding its degree of localization and its dependence on the prevailing climate. Radionuclides may also be transferred via

water movements in sewage systems. The fate of such material is presently not followed in detail in consequence-assessment codes, and there is a need to assess whether or not significant contributions to health or economic consequences are being omitted.

OBJECTIVES

It is intended to improve where necessary the models and parameterizations used in estimating the intensity and spatial distribution of deposited activity and the total health/economic impact of such deposits in assessments of the consequences of accidental releases of radioactivity. To this end a better understanding should be attained of:

- the influence of various weather conditions on deposition, particularly weather conditions which can lead to high deposition fluxes such as fog, snow or intense rain;

- the skin deposition velocity, for a range of aerosol sizes, in controlled environments and under normal conditions;

- the indoor air concentration and deposition for micron and sub-micron-particles. The latter will be achieved by developing new methods for labelling, dispersing and measuring sub-micron particles, labelled with neutron activatable tracers;

- the weathering of deposits in urban and rural environments and its impact on long-term external exposure;

- resuspension of deposited ^{137}Cs activity;

- the ultimate fate and dosimetric impact of radionuclides carried by run-off water.

A new model will be developed calculating the inhalation dose, skin dose and the external dose originating from indoor deposits and based on data concerning the cloud of pollution and the dwelling in question. An additional objective is to review and consider the results from the experimental programmes with a view to the future incorporation into accident consequence assessment codes and computer support systems for use in emergency planning and response.

WORK PROGRAMME

A) Wet deposition of atmospheric aerosol

In addition to the parameters describing the aerosol, the characterisitcs of hydrometeors such as the droplet size distribution or the morphology of snow flakes are determined during field measurements. The droplet distribution will characterize the different rainfall types and will allow a more detailed description of the deposition process.

The deposition of trace substances during fog events can be much higher than during events with rain or snow because the element concentration of fog droplets is higher than that of rain drops or snow crystals. Using an 8 stage Berner impactor aerosol samples with different sizes are collected. The elemental and ionic aerosol composition is used as tracer for the deposition process. The liquid water content of the foggy air is an important parameter for the deposition and is also measured. Airborne fogwater is collected using active type fog water collectors with cut offs for different droplet sizes. Deposited fogwater is collected on polyethylene deposition surfaces. The fogwater is analysed for the same components as it is done for the aerosol.

The deposition of fogwater to natural surfaces is measured using a balance lysimeter. Deposited masses of the substances under consideration can then be derived by using the results on trace substance concentrations in the fog water samples.

B) Deposition on skin

The deposition velocity to human skin is measured for resting people and under working conditions in normal surroundings. It is intended to measure the deposition of tracer marked material in two ways: on cellophane taped to the skin and by doing a very thorough shaving.

In addition experiments are carried out in a new 2 meter cube aerosol test chamber to confirm the range of aerosol concentrations and air exchange rates over which consistent deposition samples may be taken from skin, clothes etc. These experiments will encompass the range of aerosol types that have been developed for experimental study. While aimed principally at sampling techniques, using surrogate surfaces and models, e.g. shop window dressing models, the experiment will also serve to define the safety design of future measurements using human subjects.

Given a satisfactory safety case and experimental design in this test chamber, measurements of deposition on human subjects will be undertaken. Further, the test chamber offers the possibility of studying deposition on particular indoor surfaces in air concentrations higher than could be conventionally achieved in a house, thus making measurements easier.

C) Labelling and dispersion of sub-micron monodisperse aerosols

Tests are carried out to confirm the feasibility of labelling sub-micron particles, for use as surrogates for natural submicron particles, with suitable neutron activatable tracers such as dysprosium, indium, europium or caesium.

Two alternative types of particle are investigated. Firstly, polystyrene particles, which are available in a wide range of monodispers size are used. Two approaches to labelling these particles will be investigated, namely service attachment and the complexing of the tracer with a suitable bonding structure. Secondly, it is tried, to generate solid submicron aerosols with an aerosol condensation generator from the actual tracer compound. This clearly has the potential to yield higher labelling levels.

D) Indoor air concentration and deposition

Air exchange rates and deposition constants will be measured in new houses to improve the knowledge on the distribution of these parameters. On basis of the improved numerical material a relationship between the surface to volume ratio of the rooms and the deposition constant will be established. By taking samples of different surface types placed in the ceiling, on the wall and on the floor and determining the amount of tracer on them, it should be possible to get a picture of the deposition velocities to the different surfaces in a room.

In addition the influence of variations of the internal air circulation, the particular furnishing types and the human activity will be investigated in the experimental terrace house of the british Building Research Establishement.

E) Weathering of ^{137}Cs in urban and rural environments

In-situ γ-ray spectrometry will be carried out at about 50 locations in urban, suburban and rural environments in Bavaria which have been continuously investigated since the reactor accident at Chernobyl. The spectra will be evaluated with respect to the reduction of the γ-dose due to weathering. The data sets will be used to check recently developed external dose models, and , if necessary, to derive an improved model.

Run-off and weathering for unpervious urban surfaces will be investigated in areas, which have been relatively high contaminated after the reactor accident of Chernobyl, e.g. in the town of Gävle in Sweden. The surfaces considered will include busy roads, pavements and roofs; three test roofs at Risø will be used, supplemented by a test roof from Gävle rebuilt at Risø. Also pervious surfaces such as surfaces covered with grass, trees, and bare soil will be examined.

F) Resuspension of deposited ^{137}Cs activity

The measurements of resuspension, translocation and weathering of ^{137}Cs in the urban environment of Goiania (in co-operation with the Instituto de Radioproteção e Dosimetria, Rio de Janeiro) will be continued. The experimental work will include measurements of size differential aerosol concentrations during different weather episodes and measurement of street dust throughout the city. The data will be used in the urban exposure model PARAT I to assess the resulting exposure to critical groups in this city from various pathways and the effectiveness of various decontamination measures.

Resuspension is also investigated at sites which have been contaminated after the Chernobyl accident. Rain-collectors with different elevations above ground are used for these measurements.

G) Ultimate fate of deposited radionuclides

The sewage sludge in the town of Gävle will be investigated in order to identify the dominant mechanisms of retention and redistribution.

H) Consideration of experimental results for external exposure and other irradiation pathways in the context of accident consequence assessments and real-time response systems

Using the experimental results a model of particulate pollution ingression into and distribution within dwellings will be developed. Input variables will be the air exchange rate of the house-rooms, the concentration of the pollutant in the air, the duration of the cloud passage and the content of different isotopes in the cloud. The aim is to calculate the inhalation dose based on the mentioned input parameters. Based on the measured deposition velocity to skin the model will also give the ß- and γ-dose to the skin.

The amount of the material deposited in the dwelling and its distribution will be calculated, resulting external exposures will be estimated.

Ways of incorporating the results of this project and of other CEC projects(on long term countermeasures and on decontamination in rural and urban environments) into accident consequence assessment codes and computer support systems for use in emergency planning and response, specifically those under development for the CEC will be considered and evaluated.

BENEFITS

This programme of work will result in increased confidence in the models and parameterizations used in assessing the consequences of accidental releases of radioacitivty, thereby enhancing the utility of such assessments in decision-making processes. It will provide valuable data and theoretical underpinning for the computer codes being developed on a European scale for probabilistic consequence assessment and for real-time emergency evaluation.

The possible applications of the investigations are not restricted to the area of radioacitivity. Results on wet deposition of atmospheric aerosol, deposition on skin, ingression into and deposition inside dwellings can be applied in other air-pollution models. The amount of inhaled matter is the same, given identical conditions and particle type.

CONTRIBUTIONS

Participant 1: GSF

GSF is the coordinator of the project. Among its main activities in the project are field measurements of wet and moist depositions of the aerosol. Elemental and ionic trace elements on aerosols and hydrometeors are used to characterice the deposition process. Besides sampling devices with high temporal resolution and meteorological equipment the measurements need analytical capabilities with low detection limits and certain special items of equipment, for example size specific collections of fog droplets or a highly sensitive outdoor balance for the measurement of the amount of fog deposition water.

GSF performs in situ gamma spectrometric measurements at 50 urban and rural sites in Bavaria, which have been examined since the reactor accident of Chernobyl. It also contributes its experiences and data obtained in the close cooperation with the Ukrainian Scientific Centre on Radiaton Medicine, Kiev, on the migration of cesium and cerium into the soil and on the external exposure of the Ukrainian population after the reactor accident of Chernobyl. Available data sets on external exposures will be evaluated. The quality of existing models will be investigated using these data sets, and improved if necessary.

Concerning resuspension, GSF contributes experiences and data obtained in the close cooperation with the Instituto de Radioproteção e Dosimetria, Rio de Janeiro, on the cesium redistribution after the accident in Goiania.

Participant 2: Risø

The deposition velocity to human skin will be measured when people are resting and under working conditions in normal surroundings. The amount of tracer marked material deposited on skin shall be measured in two ways: on cellophane taped to the skin and by doing a very thorough shaving.

Air exchange rates and deposition constants will be determined in a few new houses to improve the knowledge on the distribution of these parameters. If the new method using sub-micron particles under development at Imperial College succeeds these particles will also be used. On the basis of the improved numerical material a relationship between the surface to volume ratio and the deposition constant will be established. These experiments will be carried out in close collaboration with Imperial College. By taking samples of different surface types placed in the ceiling, on the wall and on the floor and determining the amount of tracer on them, we should be able to get a picture of the distribution of particles to the different surfaces in a room.

Risø will continue to investigate weathering processes and the influence of traffic etc. on the decrease of the radioactive contamination in the urban area. This will be done using in situ measurement in the relative highly contaminated city of Gävle. The weathering processes will also be followed on test roofs contaminated with radioactive deposition from the Chernobyl accident.

Resuspension is still not well described, one of the major uncertainty is the contribution of the local resuspension to the re-deposited activity, compared to the contribution of material which has been resuspended faraway. This will be investigated further by use of rain collectors elevated to different heights.

The fate of the radioactive material deposited in the urban area will be investigated by measuring the activity of sewage sludge in the city of Gävle.

Risø will develop a model of particulate pollution ingression into and distribution within dwellings using the experimental results. As input we want to use the air exchange rate of the house, the concentration of the pollutant in the air, the duration of the cloud passage and the percentage of the different isotopes in the aerosols. The first aim is to calculate the inhalation dose based on the mentioned input parameters.

Based on the measured deposition velocity to skin the model will also give a γ-dose to the skin. We want the model to give an estimate of the external gamma dose rate originating from indoor deposits.

Participant 3: NRPB

The external irradiation of people from radioactive material deposited on the ground and the subsequent movement of this material in the environment makes an important contribution to exposure and risks following accidental releases of material to atmosphere. As part of this project continued experimental work is proposed on the aspects of deposition and weathering in urban and rural environments and on resuspension. Indoor deposition and air concentrations and deposition on the skin will be investigated. These experimental studies will provide additional valuable data on irradiation exposure routes and the mechanisms involved in the spatial and temporal distribution of radionuclides and doses in rural and urban environments.

NRPB is to review the data from these experimental projects, and to consider and evaluate ways of incorporating the results of these experimental programmes into accident consequence assessment codes and computer support systems for use in emergency planning and response, specifically those under development for the CEC.

There will also be collaboration with other participants in the CEC CHECIR project in connection with long term countermeasures and, in particular, decontamination in rural and urban environments.

Participant 4: Imperial College of Science, Technology and Medicine

Imperial College will continue the past collaboration with RISÏ where the two organisations have demonstrated their ability to work together in an innovative way. To give an example, in earlier joint work, Risø has contributed its expertise in air exchange and aerosol measurements inside buildings, while Imperial College has contributed original developments in labelling artificial monodisperse aerosols with neutron-activatable stable tracers.

The specific contributions of Imperial College will include:

- continued application of its expertise in labelling and measuring aerosols using stable tracer techniques together with the College reactor;

- the development (in collaboration with the Chemical Engineering Department) of new techniques for labelling, generation and analysis of sub-micron aerosols - of size near the theoretical minimum in deposition velocity;

- deposition measurements on human subjects in a controlled environment and participation with Risø in joint experiments in a natural environment;

participation in joint experiments in houses both involving skin deposition of aerosols and on indoor surfaces, and involving a wider range of parameters influencing deposition than has been studied so far;

the use of the Energy Systems Section aerosol test chamber for a range of initial proving experiments relating to the measurement of aerosol deposition on skin - relating to the modelling of beta dose.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT920044 Coordination of atmospheric dispersion activities for the real-time decision support system under development at KfK.

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Total Contribution by the Commission: 315 KECU
21 months 1/09/92 to 31/05/94

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Description of research work

1 Background

From the beginning in 1990, the project coordination of the comprehensive decision-aiding system (RODOS) has been the responsibility of KfK, both with respect to software developments and implementation, and also with respect to the systems hardware configurations including interfaces and graphics development etc. The system involves multiple disciplines of which atmospheric dispersion is only one.

However, emerged from the discussions during Phase-I of this project, it became clear that the ability to estimate specific atmospheric dispersion scenarios is a high-priority task and is of uttermost importance for the subsequent success or failure of the decision support system to guide off-site emergency situations.

Therefore, in order for KfK to fully concentrate on the development of the decision supports systems itself, including all of its soft- and hardware configurations, graphics packages etc., the atmospheric dispersion related work of this project is now during Phase-II separated out in a supporting sub-group, which is in close collaboration with the system-coordination group at KfK.

1.1 Progress regarding atmospheric dispersion during Phase I:

At present, the Phase-I contractors in the field of dispersion and deposition related matter are in the midst of accomplishing a series of activities, which in combination contributes and provides a series of practically useable components to the decision support system. Presently, the on-going Phase-I activities fall under the following headlines:

A. NEAR SITE - LOCAL SCALE:

1. Developing, ranking and selecting a hierarchy of suitable flow and dispersion models for the near-range dispersion.

2. Conducting and evaluating local scale dispersion experiments for model-evaluation, system training and uncertainty analysis.

3. Establishing and testing schemes for meteorological preprocessing of meteorological data used as inputs to the flow and dispersion module.

4. Feed-back feasibility studies for straight-line gaussian plume models.

5. Feasibility studies for real-time data assimilation procedures for non-gaussian models (the Leeds-Warwich group).

B. MESO-SCALE AND REGIONAL SCALE:

1. Investigation of the influence of orography and cloud structures on rain and deposition patterns.

2. Basic modelling/development for diagnostic mesoscale dispersion including complex terrain and orography.

3. Investigating reliability/uncertainties in regional scale numerical weather forecasts for prognosis dose-calculations (pilot study).

4. Feasibility studies using data assimilation (Back-fitting) with a regional scale Eulerian grid model.

5. Investigation of the usefulness of probabilistic statements of precipitation predictions.

In this context, we will distinguish activities on:

a) Local or Near-site scales: 0-20 (30) km range, and
b) Meso-scale (out to 100 (200) km), and
c) the Regional (or European) scale (out to 5000 km).

For the local scale, both models and experimental data bases for the real-time system are presently under development and test implementation at KfK. At present, no principal problems seems to remain at this range, except of course, the always challenging but also very difficult problem of now- and fore-casting dispersion foot-prints over severe complex terrain, including situations with stagnant flow.

For use on the meso- and regional scales, however, both appropriate real-time models and accompanying experimental test data will have to be pointed out during the work.

Summary of objectives and work programme

High priority is now given to real-time dispersion and deposition assessment modelling on the meso- and regional scale ranges, with special consideration for providing the decision support system with user-friendly codes, user friendly interactive menus, interactive graphical systems, etc.

In addition, new and interesting challenges for the real-time decision support system includes now- and forecasting meteorology of winds and precipitation, and also "on-line" data handling and transfer of large quantities of prognostic met-data from operational and numerical weather forecast centers in Europe.

Furthermore, we propose to continue those activities started during Phase-I with respect to integration of model results with measurements (back fitting/data assimilation), in addition to continuing efforts regarding appropriate model interpretation and clear presentation of results, - taking due account of the uncertainties in the assessment.

A more detailed break-down of these and other activities include:

1. Integration of early radiological measurements with model results.
2. Establishing "ease-of-use" and "hands-on" training and data interpretation facilities, taking into account the uncertainties in the assessments.
3. Graphical presentation in a clear form will be assigned a high-priority area as an important element to compare superimposed maps and time-sequence displays of measured and calculated doses, and to indicate areas where uncertainties are large.
4. Establishment of an on-line data transmission between the decision support system and an operational weather-center producing routinely numerical forecasts for the regional (European) scale thereby providing hour-by hour forecasts up to +36 hours. Regional centers like ECMWF in Reading, UK, and the HIRLAM-collaboration in the Scandinavian countries including Holland and Ireland exists already and several of these centers are already operational. Regional forecasts covering the entire CEC can in this way be provided from National, - or alternatively, European weather centers. The present coordination includes collaboration with the HIRLAM project in this field.
5. Investigating of simplified and fast models or methods for "nesting" the output from these numerical weather forecasts models, presently running on a 50 km by 50 km resolution, as "prognostic drivers" for our local- and meso-scale flow models. We are searching a "gap-filler" method to "nest" the regional scale meteorological fields provided on a 50 km by 50 km grid, down to the meso- and local scales for dispersion calculations in the initial phase, by taking into consideration the local topography and terrain types in question. Such "Gap-filling" models, relating regional scale winds to local scale winds, are already in use for wind energy assessments, and is also to be considered for this development.
6. Like has already been accomplished on the local scale, a hierarchy of suitable and useable (read reliable and fast) dispersion and deposition models should be established for taking over the calculations where the near-field models start to fail (at the 20 - 30 km scale, say), and which then tracks the pollution out on the European scale. The selected models must be able to use the information on stability, rain and cloud distributions provided by the regional scale weather forecasts.
7. New strategies for data-assimilation within the decision support system is also to be investigated regarding meso- and regional scale transport, since the numerical weather centers providing these forecasts, will in general be dislocated from the system and its users. On the local and meso scale, we can most likely proceed the data assimilation activities along the lines now being proposed by the Leeds-Warwick group. However, in praxis, this activity (data assimilation and back fitting) seems to be closely depending on the type of flow and dispersion models in use, which in turn means that the back-fitting method chosen will be model-dependent. Consequently, it is necessary to select the type of model (Gaussian puff, Lagrangian particle, Eulerian grid etc.) to use first.
8. For user training and modelling exercises, we envision further to incorporate a real-time experimental data base on the meso- and regional scales as well: Suitable and well documented meso- and regional scale experiments, accompanied by their meteorological conditions under which they were obtained, must be identifies and incorporated in the decision support systems data-base for users to train with, and for familiarizing with uncertainties and data interpretation procedures.

Contribution of the Coordinator (Risø)

Coordination of activities relating to atmospheric dispersion will require participation of the coordinator in several of the proposed activities, in particular with respect to integrating activities and to guide the participants to produce practically useful software, graphical concepts, and providing the necessary sub-modules for the decision support system.

In particular, it is envisioned that the coordinator contributes with a useful model for the "gap-filler nesting" necessary between the Regional scale prognosis calculations, and the local scale winds (in collaboration with DMI).

Furthermore, the coordinator envision to participate actively in the establishment of a "Reference and Training Experiment Data Base", suitable for local and mesoscale model evaluation and used-training:

1) Existing data sets from suitable and well documented meso-scale diffusion experiments will be identified and made available for inclusion in the systems database for training and evaluation purposes on this scale.

2) Furthermore, as the only experimental counterpart in this project,- continued measurement campaigns, based on high-resolution lidar measurements will continue, but now approaching the larger (meso) scale. Risø National Lab has recently allocated funding for a 2. generation Lidar-system, which is designed for 1000 times the sensitivity of the mini-LIDAR system successfully operated during Phase- I.

We consequently continue with our experimental activities, now on larger scales, by conducting full-scale and detailed smoke and aerosol plume experiments, with the purpose to capture and study individual atmospheric dispersion scenarios relevant for model validation and training with RODOS.

Contribution of ICSTM

Real Time Modelling of Atmospheric Dispersion on trans-European Scale.

Within the current Radiation Protection Programme on Real Time Modelling, the Air pollution Group at Imperial College is developing a computer model, 3DRAW, capable of simulating transport and deposition of radionuclides on a trans-European scale. The model operates in both real-time and predictive modes, using output from a meteorological forecasting model. It is designed to be flexible with respect to the data available, and to interface with other models for short and medium range dispersion developed by other contractors. In addition the model produces output files for direct use as input to the dose response module under development by GSF for assessment of exposure, contamination of food chains and other consequences.

With the aim to integrate the 3DRAW into RODOS, ICSTM next engage in two main tasks:

i) Integration of early radiological measurements with model results to revise and improve assessments.

ii) Analysis and presentation of results - implementation of a "user friendly" code.

Contribution of ENEA-DISP

Atmospheric dispersion modelling in the RODOS real time emergency response system under development.

Suitable modules of the meteorological Preprocessing software PAD will be implemented into the Analyzing System (ASY) of RODOS, to provide the necessary link between primary meteorological and environmental data available, and the short range plume or puff models which is are going to be integrated into ASY.

The variables which can be evaluated with different methods are boundary layer scaling parameters (Monin-Obukhov length, friction velocity, heat flux), mixing height, wind speed vertical profiles, atmospheric stability category, turbulence parameters (the three components of wind direction fluctuation and Lagrangian time scale), plume rise.

For a limited number of variables, the preprocessor will be extended to the evaluation of two-dimensional fields for the application of mesoscale particle models that will be the future, by using simple the estimated values where integrated into the system in interpolation schemes between meteorological observations are available.

The software of past and future development of PAD is written in FORTRAN language in a DEC-UNIX environment (ULTRIX), based on considerations of cost-effectiveness and portability; the graphics are based on OSF-MOTIF and NCAR graphics.

Contribution of Risø (II)

Computer system for real-time modelling of atmospheric dispersion on the local and mesoscale including back-fitting and training.

Objectives for Phase-II RIMPUFF/LINCOM modelling activities, within the framework of real-time facilities under RODOS, include:

i) Integration of early radiological measurements with models to revise and improve assessments

Improve the doses/concentrations predicted of the model in cooperation with the Universities of Leeds and Warwick on forecasting techniques. Further, "back-casting" will be considered to update earlier estimates of dose/contamination levels occurring in the light of later observations.

ii) Presentation of results - implementation of a "user friendly" code:

In accident situations, simple menu-driven information displays is a very important requirement. Substantial effort will consequently be assigned to methods of assessing and interpretation of these results, and to their communication with the user through user friendly graphics and display systems.

iii) Manual and training aids

To facilitate the use of RIMPUFF/LINCOM and its associated software, a number of examples involving hands-on operation of the code will be provided.

iv) Coupling to Regional scale wind prognosis data (HIRLAM)

For long-range dispersion the mesoscale model RIMPUFF and the long range dispersion model, RAM, developed by SMHI, will be combined.

v) Uncertainty estimates using HIRLAM

A number of "hands-on" examples will be provided for RODOS.

vi) Preprocessing of meteorological data

RIMPUFF/LINCOM will be integrated with PAD developed by ENEA.

Contribution of NCSR "Demokritos"

The NCSR "DEMOKRITOS" is concerned basically with the use and development of the modules DELTA, ADREA-wind and ADREA-dispersion.

The DELTA module is basically the ground description module for the ADREA modules.

The tasks for the present phase II of the RODOS project are outlined as follows.

1. The integration of ADREA-wind and dispersion codes into the RODOS Decision Aiding System for Off-Site Emergency Management, including links with regional forecasting. This task reflects also close collaboration with KfK,DMI, SMHI and Risø.

2. Demonstration of module reliability and quantification and uncertainties. A systematic effort will be undertaken to compare predictions with well documented field data. The first such activity will be concentrated on the Rocky Flats

Experiments directly related to the EG & G Nuclear facility radiation protection. Comparisons with the best US computational tools is also intended through this study. Other existing European data will also be investigated and utilized for comparisons. Based on the experimental comparisons, evaluation of uncertainties will be attempted.

3. In the present models certain important aspects of deposition have to be looked on more systematically. The effort will be concentrated on the dry deposition over seas and the first order estimation of the "radioactivity load" in water accumulated areas. The capability of the DELTA module to describe ground in detail, permits a relatively fast identification of the water pathways.

Contribution from DMI:

DMI proposes to contribute to the development and validation of the RODOS real-time decision support system using meteorological forecast data from the Danish HIgh Resolution Limited Area model (DK-HIRLAM) as input for the atmospheric dispersion models for dose calculations on both local and regional scales.

The role of DMI will be to link DK-HIRLAM to already existing dispersion models, and to establish a real-time system enabling dispersion models to run in predictive modes to forecast concentrations, deposition and dosages of radioactive materials.

It shall be emphasized that the dispersion models will not be integrated into the RODOS system, but RODOS will be supplied with outputs from HIRLAM, and one major issue of this work will be to examine which of the many parameters from HIRLAM can be used with advantage in dispersion models. Validation studies will be undertaken both concerning the quality of relevant products from DK-HIRLAM and for the dispersion models in concern. The validation studies will mainly be based on new (actual) data, but studies will also be made for the Chernobyl case.

Investigation of the possibility to transfer large quantities of met-data on-line via the new European network ISDN will also be performed in collaboration with Risø and KfK.

Contribution of GRS

Implementation of a mass consistent flow model to the RODOS real time decision support system:

In order to achieve a realistic approximation of a pollutants transport and diffusion in complex terrain a three dimensional wind field is needed as an input for subsequent dispersion calculations. Here, the inhomogeneous wind field is the most important driving mechanism for a pollutant's transport.

The contribution of GRS to the atmospheric dispersion activities in RODOS is to provide an already existing mass consistent flow model (MCF) to the decision support system, and to assist the implementation.

Contribution of SMHI

Sweden is an EFTA country. The contribution towards SMHI will be published in an addendum after signature of the Association Agreement.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT920057 Methodology for evaluating the radiological consequences of radioactive effluent released in accidents - the MARIA project.

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Description of research work

Summary of Project and Objectives:

The MARIA (Methods for Assessing the Radiological Impact of Accidents) project was initiated by CEC in 1983. There have been three earlier phases of the project; the first from 1983-84, the second from 1985-89 and the third from 1990-91. The current fourth phase of the project runs from 1992 to 1994.

The aim of the project was firstly to review and subsequently to further develop methods and models for accident consequence assessment (ACA) within the EC. A further objective of the later phases of the project was to develop a computer program for carrying out probabilistic accident consequence assessments, and to make this system widely available throughout the EC. This program package - named COSYMA (COde SYstem from MARIA) - has now been issued, together with supporting documentation. It was released and distributed to 28 institutes in February 1991 as Version 90/1 and with updates in December 1991 as Version 91/1.

Throughout the period of the MARIA project, the main contractors have been the National Radiological Protection Board (UK) and the Kernforschungszentrum Karlsruhe (Germany). In addition, a number of other contracts have been placed with organisations in most other EC countries. Contracts have covered work related to most aspects of accident consequence assessment, including atmospheric dispersion and meterological sampling, external exposure from deposited material (particularly in urban areas), decontamination, the modelling of foodchain transfer and early health effects, risk coefficients for the induction of late health effects, and models for estimating the economic impact of accidents. The project has also included studies on the uncertainty inherent in model and ACA code predictions.

Many of the results of these studies have been incorporated into the COSYMA program package, which is a very flexible system consisting of a set of ACA modules together with a number of subsidiary programs and datafiles.

The current state of accident consequence modelling is that sufficient knowledge and experience has now been gained for adequate results to be determined for many applications. However, effort and further research is still required in a number of areas.

These areas are:

i) The maintenance, support and development of the COSYMA system. This includes modelling and data improvements and updates to both the system itself and the supporting documentation, and continual adaptation of the system to the needs of users.

ii) The further development of a PC version of the COSYMA system, to allow users to undertake ACA studies without access to large computer resources and to benefit from an interactive interface. The software for the PC system, like the full COSYMA system, will be available to EC organisations through the CEC.

iii) Participation of the COSYMA system in the CEC/NEA ACA code intercomparison exercise, and documentation of the results.

iv) Further work on the development of techniques for assessing economic impact, including impact beyond the areas affected by countermeasures, the incorporation into the COSYMA system of an alternative economics model, and site-specific features of economics modelling.

The objective of the current fourth phase of the MARIA project is therefore to achieve by mid-1994 consistent and up-to-date probabilistic accident consequence assessment tools which will run on both main frame and PC computers, and which will be generally available through the CEC to EC countries for use in risk assessment studies.

Distribution of tasks

The collaboration between the four participating organisations is as follows.

The maintenance, support and development of the COSYMA system is being undertaken predominantly by KfK and NRPB. The development of a PC version of the COSYMA system is also being undertaken with collaboration between NRPB and KfK. The emphasis at KfK is on the mainframe system and at NRPB is on the PC system.

The participation of the COSYMA system in the CEC/NEA ACA code intercomparison exercise, and documentation of the results, is being lead by KEMA, with assistance and cooperation from the other participants.

The further work on the development of techniques for assessing economic impact is being undertaken primarily at the Polytechnical University of Madrid, with assistance and discussion with KfK and NRPB as necessary. KfK and the Polytechnical University of Madrid will collaborate on the incorporation of the MECA economics model into the COSYMA code.

The principle work areas of each organisation, and the collaborative links between these areas, are described below.

Contribution of the National Radiological Protection Board (NRPB), UK

NRPB objectives:

- **to assist KfK in the maintenance, support and development of the mainframe COSYMA system**, primarily in the area of data improvements and updates to the system and the supporting documentation.

- **to take the lead, with assistance from KfK, in the development of the PC version of the COSYMA system**, with the aim of producing a first version for release in 1993 and a revised version with graphical display features in 1994. Documentation of the PC system will be provided. The bulk of NRPB effort in this phase of the MARIA project will be devoted to this topic.

- **to participate in the CEC/NEA ACA code intercomparison exercise**, together with KfK, KEMA and other EC organisations, in the running of the COSYMA code, and the documentation of the results. Work is also being undertaken in support of other organisations using COSYMA in this exercise, in collaboration with KEMA and KfK, to explain reasons for any differences in the results which may be obtained by the various organisations involved.

- **to undertake a review in 1994, with assistance from the other participants in the project, of the current position of probabilistic ACA modelling and codes**, with a view to identifying the need and priorities for future research.

  This will involve a review of the state of modelling for the many steps involved in accident consequence assessment at that time, leading to recommendations on what further work is required.

Contribution of Kernforschungszentrum Karlsruhe (KfK) (Germany)

KfK objectives:

Four topics are covered, with emphasis on the first and fourth:

- **Support, maintenance and further development of the full COSYMA system, with assistance from NRPB.** The program package COSYMA requires continuous updating, testing and improving of identified weak points in models and data sets. This includes an on-going adaptation of the program descriptions and the user guide, and the preparation of further documents giving more detailed information or guidance. A specific aspect is the refinement of models for assessing the off-site economic costs of accidents, and assisting the University of Madrid in incorporating the MECA economics model into COSYMA.

- **Distribution of the program package COSYMA and interaction with users.** Interaction with these users, in particular support during implementation and testing and help in applying the code for special tasks (see fourth topic), is an ongoing activity. This includes the further distribution of COSYMA and the preparation of training courses for potential users.

- **Further development of models and databases for the PC version of COSYMA** in support of NRPB. An aim of this continuing work is to achieve low computing times and storage requirements while preserving flexibility in the PC system.

- **Participation in the CEC/NEA code intercomparison exercise.**
  A single agreed set of formal COSYMA results has been submitted by KfK and NRPB. Work is also required in support of the other organisations using COSYMA, including in co-operation with KEMA the provision of help in explaining any differences in the results obtained by the various organisations involved.

Contribution of the Polytechnical University of Madrid (Spain)

Polytechnical University of Madrid's objectives:

- **to undertake further work on the development of techniques for assessing the secondary, or indirect, economic impact of nuclear accidents**, including the impact beyond the areas directly affected by countermeasures. Different methods that could be used for this study will be reviewed and evaluated, taking into account the experience from Chernobyl and other past events, with assistance and discussion with KfK and NRPB as necessary. The first step will be the assessment of the feasibility of the various models for evaluation.

  If the problem seems amenable to modelling as part of standard ACA, the second step is the development of a model applicable to ACA studies, including indirect economic losses due for instance to the impact on tourism or to the effect of the accident on the marketing of uncontaminated agricultural produce.

- **to incorporate the MECA2 model (Model for Economic Consequence Assessment) into the COSYMA system** as an additional economics module. A close collaboration with KfK will be established to ensure the correct implementation of the module into the code system. To allow site-specific features of economics modelling to be considered, the socio-economic database used by MECA2 will be adapted to the grids and general data structure used in COSYMA. The performance of the new module will be tested against reference cases.

Contribution of KEMA (Netherlands)

KEMA's objectives:

- **to take the lead in co-ordinating the efforts, and preparing, evaluating and documenting the results of the COSYMA users who are participating in the CEC/NEA ACA code intercomparison exercise.**

  The COSYMA code is being used by several participants from EC countries in the CEC/NEA ACA code intercomparison exercise. In collaboration with KfK and NRPB several extra exercises have been defined for the participating COSYMA organisations, which have been designed to enable an intercomparison of the results of the different users of the COSYMA code.

  KEMA is co-ordinating the efforts of the different organisations. Work is being undertaken, also in co-operation with KfK and NRPB, to evaluate the results and to explain the differences. Several extra test runs will be carried out to verify the explanations.

  A questionnaire has been sent to the participants in the intercomparison exercise and to all other users of the COSYMA code to ask for their experiences and comments in using the code.

The evaluation of the results of the COSYMA users intercomparison exercise, together with the replies from the questionnaire, will be documented in a EUR report which will be published in 1993. This document will be a valuable feedback for the CEC on the use of its code and for improvement and updating the system. It will also be of use to present and future users in running the COSYMA code.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT930073 Analysis and modelling of the migration of radionuclides deposited in catchment basins of fresh water systems.

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Total Contribution by the Commission: 145 KECU
17 months 1/01/93 to 31/05/94

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Description of research work

The migration of radioactive substances from catchment basin to water bodies is a very complex process involving a variety of phenomena depending on the nature of the radionuclides, on the characteristics of the drainage area (such as the basin geomorphology, the composition and amount of vegetation cover, the impact of human activities on the basin etc.) and on seasonal effects (ice and snow melting, floods etc.).

Moreover the transport of radionuclides is related to the complex dynamics of water movement in the drainage area. Surface runoff, infiltration, percolation, ground water flow, transport of eroded particles etc. play different roles in the migration of radioactive substances depending on the characteristics and the state of the catchment basin.

A considerable fraction of the initial deposited activity over a catchment basin is transferred to the sediments. This will give an important potential long-term radioactivity source to the water body and to the biological parts of the ecosystem. The experience gained up to now suggests that various components of aquatic systems may show high levels of contamination in the long run due to the radionuclide remobilisation from sediments and the migration from catchment basin to water.

The main aim of the research project is the analysis of the most important phenomena concerning the migration in fresh water systems (drainage areas, rivers, lakes, reservoirs, etc.) of radioactive substances deposited in catchment basins to develop generic models for assessment of short and long term behaviour of radionuclides in the aquatic compartments of contaminated areas. Such models can be a valuable support in dose evaluations following an accidental release of radionuclides to the environment.

In this study the processes of the migration of radionuclides in drainage areas will be analysed and quantified. The research will deal with the migration and the remobilisation, due to the transport by rivers and by run-off and wash-off waters and to the resuspension from sediments, of deposited radionuclide in the environment. The study will lead to generic models for the assessment of the short- and long-term behaviour of the most important radionuclides such as ^{90}Sr and ^{137}Cs in different types of catchment basins. Such models can be applied extensively in the EC, subject to the appropriate local characteristics of the catchment basins. The developed models will describe the consequences and the behaviour of the radioactive contaminants in the aquatic part of the environment after a nuclear accident.

The evaluation of the model results and the uncertainty and sensitivity analysis, will offer the opportunity of identifying processes and parameters that are of high importance for the out-come of the models in different ecosystems and during varying circumstances. As the models are being developed for radiation protection purposes, the work may be done in close co-ordination with the EC-project on Decision Support Systems (DSS) for off-site emergency management.

The research project will deal with fresh water systems showing a great importance from an economic and social point of view (fishery, drinkable waters, irrigation waters etc.). Catchment basins located in various European areas (Italy, Netherlands, Sweden, United Kingdom) and showing a range of environmental characteristics and situations will be analysed. In one of the studied catchment the migration of radionuclides in a major European river (the Rhine) plays an important role.

The fallout from the nuclear-weapon tests in the atmosphere of past decades and the contamination due to the Chernobyl accident, provide stimulating opportunities to analyse the behaviour of the long living radionuclides ^{90}Sr and ^{137}Cs in catchment basins and to evaluate the quantitative migration of radionuclides to water bodies.

As lakes represent point of accumulation of radionuclides transported by rivers, by wash-off and run-off waters, the processes of migration in catchment basin may be analysed and quantified taking advantage of the possibility of evaluating the radionuclide accumulation in lakes.

Following a preliminary analysis the following typologies of catchment basin systems were selected:

- volcanic (small drainage areas with deep lakes as accumulation points, formed by volcanic action)
- coastal (wetland and their transfer to shallow lakes)
- glacial (large drainage area formed by glacial action with deep lake systems as accumulation points)
- forested (forested areas and their tributaries to lakes)
- continental (i.e. the drainage area of the river Rhine and its tributaries, with lake Ijsselmeer as accumulating point)
- agricultural (predominantly agricultural drainage areas and transfer via tributaries to lakes).

The program will include a detailed analysis of the balance of radionuclides in the examined systems (evaluation of the amount of radionuclide depositions on the catchment basins, content of radionuclides in water and in sediment etc.) and will be based on two phases :

a) collection of experimental data of radionuclide contamination and of information about basic characteristics of the drainage area to support the model development;

b) model development, validation and verification with uncertainty analysis.
Phase a) will allows the evaluation of time dependent "transfer functions" of the radionuclides from the drainage areas to water bodies.

The model development will be based on a detailed analysis of the involved processes : movement and balance of water in the basin, erosion, sorption-desorption mechanism

of radionuclides with soil components and eroded particles, role of acidification, role of stable element concentrations, etc.

The main goal of the study is the **development of models assessing the behaviour of radionuclides in catchment basins** in view of the importance of the long term exposure to man from increased levels of radionuclides in water and, consequently, in fishes.

Causal correlation between the catchment basin characteristics, the transfer functions and the compartment model structure will be examined.

Each participant will devote its attention to problems relevant for the specific environmental situation of its country. The collaboration will offer a great deal of benefits such as:

a) possibility of evaluating the relative importance of some phenomena related to specific characteristics of the drainage areas;

b) exchange of information about the phenomena involved in the migration of radionuclides through drainage basins relevant to different environmental situations;

c) exchange of information about model characteristics and ability to predict some aspects of radionuclide migration;

d) application of models to different environmental situations.

CONTRIBUTION OF ENEA, ITALY

Italian contribution to the study will consist in the analysis of some catchment basins, located in various Italian regions, showing different geomorphologic characteristics:

a) catchment basins formed by glacial action with deep lakes as accumulation points characterised by a large ratio "area of catchment basin/area of lake water" (drainage basins are located in the North Italy);

b) small catchment basin formed by volcanic action with deep lakes as accumulation points characterised by very long mean water retention time;

c) coastal wetland characterised by frequent exchange of water between the water body and the sea;

d) artificial lakes characterised by relevant amount of accumulated sediments.

The programme will consist of following steps:

a) collection of hydrogeological data (rain precipitation, water fluxes, amount of suspended matter in waters, etc.);

b) collection of radiological data (radionuclide depositions, contents of radionuclides in waters, sediments etc.);

c) analysis of the acquired data to evaluate and investigate the migration of radionuclide in catchment basin and the radionuclide resuspension from sediments;

d) model development, validation and sensitivity and uncertainty analysis.

The behaviour of ^{137}Cs and of ^{90}Sr will be investigated. The programme will include also the collection and the analysis of some contamination data collected in Italy for roughly 30 years.

CONTRIBUTION OF KEMA, NETHERLANDS

The Dutch lake Ijsselmeer is a very shallow lake, with a mean depth of approximately 4 m and an area of 1140 km^2. Originally the lake was a sea (the former Zuidersea), but it was transformed into an artificial fresh water lake after closing it at the north end by a dike in 1930. It is fed by the river Ijessel, which is a branch of the river Rhine. Therefore lake Ijsselmeer has a very large drainage area, i.e. the catchment basin of the river Rhine. Apart from these characteristic hydrological properties of lake Ijsselmeer, which are very different from the other selected lacustrine systems, also another factor might play an important role in the behaviour of, especially, radiocesium in the lake ecosystem. Lake Ijsselmeer has a fairly high ionic strength, due to industrial releases of salts in the river Rhine. As these salts contain also a certain amount of stable caesium, the lake water will have a different ratio of stable and radioactive caesium as compared to the other lacustrine systems in the study.

The contribution of KEMA will be first to collect data in the Netherlands, Germany and Switzerland on radionuclide depositions, remobilisation, concentrations in water, suspended materials and sediments in the drainage area of the river Rhine, together with hydrological and geochemical data that characterise the different parts of the Rhine basin. In the second phase the relations between these data sets will be studied in detail, to evaluate the most important factors that dominate the time dependent migration of radionuclides from the drainage area into the lake.

In the third phase the results of the evaluation will be used for developing a model to describe the time dependent migration of the radionuclides in the drainage area. The relations between the different compartments of the model for the Ijsselmeer system will be compared with the results of the other participants studying the other selected lacustrine systems.

CONTRIBUTION OF THE SRD, UK

There is a need to examine the ultimate fate of the activity translocated from the sites of deposition by weathering or other transport processes to ensure that there are no pathways by which it can deliver significant doses to man that have not currently been taken account of in risk assessment methodologies. SRD has already considered this question for the urban-drainage pathway and will now address catchment areas in general.

This work will commence with a review of the mechanisms of transfer of radioactivity deposited in rural catchment areas to freshwater systems taking account of the range of catchment types and characteristics found within the UK. This will be followed by the development of a generic mathematical model representing the freshwater environment and the pathways from deposition to delivery of dose to man. Location-specific details will be kept to a minimum at this stage to allow general scoping calculations to take place for a range of representative freshwater conditions. Using this model a sensitivity study will be undertaken to determine important routes of exposure and identify areas of the freshwater environment requiring more detailed modelling.

Contribution of STUDIVISK AB

Sweden is an EFTA country. The contribution towards Studivisk AB will be published in an addendum after signature of the Association Agreement.

C24 Probabalistic risk assessment and real-time models for assessing the consequences of accidental releases and for evaluating effectiveness and feasibility of countermeasures.

Contract FI3P-CT930077 Multifractal analysis and simulation of Chernobyl radioctive fall-out in Europe.

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17 months 1/01/93 to 31/05/94

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Description of research work

ABSTRACT

Recent developments in theoretical and applied geophysics show that phenomena exhibiting extreme variability over a wide range of scales (e.g. atmospheric turbulence, wind, cloud formation, rain-fall, temperature fluctuations, ...) are best described by *multifractal* methods and techniques; indeed, multifractals provide a coherent theoretical framework and a powerful operative tool to analyse their properties. Actually, the natural phenomena involved in the Chernobyl fall-out feature this high variability, as much as the radioactive pollution released in the environment. Hence multifractals might significantly improve the knowledge of its time-space evolution and provide parameters (of easy empirical estimate) whose meanings and values are directly related to the statistical properties of the available sets of data. It is then our goal to provide a practical multifractal characterisation and modelling of air pollution and/or cumulative deposition in some European Countries.

1. INTRODUCTION

We first outlined the possibility of using *fractals* to analyse chemical and radioactive environmental pollution within the project "Post Chernobyl Action 1: Improvements of Reliable Long Distance Atmospheric Transport Models" [Contract BI6*-0241-I(A)]. In particular, we provided empirical monofractal models of both the Dioxin (TCDD) ground pollution over the Seveso (Italy) territory and the radioactive air pollution (^{137}Cs) in Northern Italy [Contract BI7*-CT90-0062 MNLA]. Below we briefly summarise the results obtained.

1.1 A study case: the monofractal model of the Seveso pollution

The Data Bank of the Seveso accident contains some thousands measurements of the TCDD pollution on the soil collected after the Seveso Accident during sevveral years. The experimental evidence that the TCDD pollution shows fractal features suggested the use of these data to improve an empirical monofractal model of the Dioxin distribution on the soil. The model was based on the Theory of the *Fractal Sum of Pulses* ("FSP" originally created for the study of atmospheric phenomena), which states that a (fractal) phenomenon can be *recovered* (simulated) by means of a properly distributed series of randomly overlapping Primary Pulses. The generation and the displacement of the pulses followed proper probabilistic laws and was "driven" by using the fractal information contained in the given data (measurements network). The output reproduce the original features of the data, while recovering (estimating) the intensity of pollution in areas where data were missing (i.e. in the "gaps" of the network). This study case represent a significant test for the feasibility of the fractal approach in describing and recovering the distribution of pollutants in the environment. Moreover, it allowed us to fix and improve some procedures and algorithms, which turned out to be of primary importance when investigating the radioactive air pollution in Northern Italy.

1.2 A monofractal model of radioactive air pollution in Northern Italy

In order to investigate the possibility of a model able to describe the distribution of the radioactivity in air in Northern Italy and to guess reliable values where no measurements were available, we made the following assumption:

1) the environmental distribution of the radioactive pollution is due mainly to the meteorological conditions (which show fractal properties);

2) the transport of radioactive substances does not change these features.

First step we provided a parametric function able to recover the trend of ^{137}Cs pollution in air and tested it on the data belonging to Italy and France. Then, we recovered the maximum information about ^{137}Cs from other sampled nuclides. Finally, we applied the FSP model on the ^{137}Cs air data. Once again, the results indicated the feasibility of a fractal modelling, providing a new useful tool to investigate (both in space *and* in time) the distribution of air radioactivity in the environment.

2. EXPLOITING MULTIFRACTALS

The recent literature on fractals show the possibility of improving the power of fractal modelling exploiting the new mathematical tools offered by *multifractals* (in view of the fact that multifractals give a better description of several geophysical phenomena). An advantage of using multifractals rather then monofractals is that they can characterise the detailed structure of the pollutant distribution over the entire range of scales, from the strictly local concentration up to the largest spatial average. Moreover, all the non-linear mechanisms involved in dispersing the pollutant are expected to be multifractal processes which, in turn, justify the application of a multifractal analysis to the study of these phenomena. In spite of the good and reasonable results obtained so far, the rigidity of our (monofractal) assumptions became soon clear and the necessity of a more flexible approach became evident.

2.1 A multifractal description of cumulative deposition in Europe

Within the contract BI7*-CT90-0062 MNLA we provided an original procedure (based on "Lévy Flights" and "Potentials") which exploits the opportunities offered by the theory of *geometrical* multifractals and uses of the available information about intensity and sparseness of the cumulative deposition of ^{137}Cs in Europe. In particular, we attempted to recover the features of the measured pollution and to estimate its intensity in regions not sampled by the monitoring networks, getting a deeper insight into the global distribution of the pollution.

Indeed, from our analyses it turned out that the radioactive distribution shows multifractal features. Our preliminary results are fairly interesting and seem to provide realistic simulations of the evolution of radioactive pollution over several ranges of scale, both in space and in time. Moreover, this study allowed us to calculate some characteristic (multifractal) parameters, to be used in further modelling exploiting the *stochastic* framework of Universal Multifractals.

2.2 A Universal Multifractals description of Seveso chemical pollution

Within the contract BI7*-CT90-0062 MNLA we again tested the possibility of exploiting a (stochastic) Universal Multifractals approach to the Seveso pollution. This study definitely point out the feasibility of universal multifractals to characterise the ground distribution of TCDD. Furthermore, we also investigate the role played by the structure of the monitoring network: actually, the (typically sparse) nature of a network may significantly alter the inferred statistical properties of a phenomenon measured on it. Treating the *density of stations* as a *multifractal measure* (rather than the stations themselves as a *fractal set*) we showed that it is possible to statistically correct for such effects. Overall, we gained a deeper insight into the "violent" fluctuating behaviour of the pollutant distribution at all scales: its strong fluctuations are not regarded as anomalous and discarded, on the contrary they are kept as an essential feature of the phenomenon itself.

3. THE PROPOSED TASK

The project involves an *a priori* analysis of the available data, since the selection of reliable measurements/networks and some quality assurance is needed in order to provide certified and consistent samples as a starting point. Once these preliminary analyses are done, the estimate of the multifractal features of the radioactive fall-out is sought, along with the calculation of the influences and the effects of the used networks. This step should allow to identify the (turbulent) processes generating the fall-out in terms of multifractal parameters. Quite recently the *Double Trace Moments (DTM) technique* for estimating the multifractal parameters of stochastic processes has been developed. In principle, this algorithm can easily be applied to any kind of data and it turns out to be a robust estimator (in the statistical sense), overcoming the difficulties arose in the past by the previously used (less efficient) methods. Moreover, given any real network, it is likely that "gaps" are left; in our case this leads to the problem of estimating the radioactivity distribution in locations not covered by the networks.

Besides, even if natural phenomena are independent of the *networks* used to measure their evolution, nonetheless it turns out that the geometry of (fractal) networks affects the analyses of the data, introducing (unavoidable) biases. This fact implies that special techniques are needed in order to properly take into account the networks effects. Now this can be done in terms of multifractals; such a task is not easy at all and most of the available statistical techniques (e.g. those used in rain-fall analysis) give unsatisfactory results. This is due to the fact that such techniques make unrealistic assumptions about the smooth behaviour of the field. On the contrary, multifractal interpolating techniques can be developed to exploit the mathematical structure of multifractal fields to obtain: (a) estimates of the extremely variable field and (b) estimates of the global statistic of a phenomenon starting from a partial knowledge of it.

This step would require, first, an analysis of the local versus global properties of multifractal fields and, second, the simulation of fractal networks and fields with known features in order to test the consistency and the validity of any recovering algorithms. Finally, these techniques might be applied to the available data collected using a given network in order to improve the "information" given by the original measurements (i.e. filling the gaps of the given network providing estimates of the radioactive pollution in non-sampled locations).

All the above opens the possibility of making some relevant progresses in the near future. Based on the already mentioned results, we are confident that within the present contract it should be definitely possible to formulate a practical multifractal model able to provide a reasonable description of air pollution and/or cumulative deposition in several European Countries.

Multifractals could also be the optimum theoretical and practical environment to develop a new class of predicting schemes, exploiting some general (statistical) properties of random variables related to multifractal fields. Being almost all crucial parameters under control and/or within reach, given the competence available within the collaborative effort, it is reasonable to expect that the goal of having a first formulation of such a describing/predicting model might be reached a couple of years in advance with respect to the common present expectations. To this purpose, the collaboration of the group led by Prof. Shaun Lovejoy (McGill University, Montreal, Canada) is foreseen. We will also develop cooperation with the group led by the vice-president of the Ukrainian Academy of Sciences Academician Victor G. Baryakhtar and its scientific correspondents in the Community of Independent States for collaborative efforts on multifractal analysis of their Data Bank concerning the Chernobyl accident and developments of dynamical simulations.

Contribution of the University of Pavia

The University of Pavia will take care of the *a priori* analysis of the available data, performing specific quality assurance operations and selecting reliable measurements and/or networks. This shall provide certified and consistent samples, to be used in further analyses. Then, the estimate of the multifractal features of the radioactive fall-out will be obtained, along with the calculation of the influences and the effects introduced by the presence of the measuring networks. This will enable us to give (statistical) estimates of the radioactivity distribution in locations not covered by the networks. Actually, the University of Pavia will develop multifractal interpolating techniques to investigate the structure of multifractal fields; this, in turn, will enable them to obtain both (statistical) local estimates of extremely variable fields and estimates of the global statistic of a phenomenon starting from a partial knowledge of it. In order to achieve this point, they will use a two-steps procedure: first perform an analysis of the local versus global properties of multifractal fields; second, simulate fractal networks and fields with known features in order to test the consistency and the validity of any recovering algorithms. Finally, they will apply these techniques to the available data collected over all Europe using a well defined network; this will enable them to improve the "information" given by the original measurements, by filling the "gaps" of the given network and providing estimates of the radioactive pollution in non-sampled locations.

Contribution of the Centre National de Recherches Scientifiques

The Centre National de Recherches Scientifiques will take in charge the problems of time evolution and of prediction. These are the most challenging and difficult to deal with given: (a) the high number of variables involved and (b) the poor partial knowledge of the extremely variable/intermittent phenomena playing some significant role in the radioactive fall-out. However, going far beyond Generalized Scale Invariance, a rather

powerful multifractal framework ("Lie cascades") has been recently developed to deal with complex interacting fields. In this framework one may simulate and analyse the scaling relations between the different components of a multicomponents field (e.g. vector or tensor fields) on a wide range of scales. For instance, dynamic multifractal simulations of (liquid water) clouds have already been developed and analysis of the full complexity of strong interactions between the dynamics of clouds and rain fields are in progress.

The corresponding techniques will be readjusted in order to dynamically simulate in space and time the radiation fall-out in the framework of radioactivity environmental studies. We will then exploit the temporal long-range interactions properties of these multifractal processes to derive new *stochastic predicting schemes*. These schemes will be "optimal" in the sense that, contrary to others, they will predict the extreme variability of the data over a wide range of scales and intensities (e.g. contamination levels).

PART II

International collaboration on the evaluation of the consequences of the accident at the Chernobyl Nuclear Power Plant

RADIOECOLOGY

Radioecology

Contract COSU-CT920015 Contamination of surfaces by re-suspended material.

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Total Contribution by the Commission: 560.500 ECU
12 months *1/11/92* to *31/10/93*

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Description of research work

MAJOR OBJECTIVES

For an understanding of the consequences of radionuclide deposition it is necessary to study concentrations and size distributions of airborne radioactive material, properties of hot particles in the air and soil, and the time-dependent behaviour of various radionuclides in surface layers. This surface contamination acts as a source of continuing exposure principally from external radiation and from internal radiation due to transfer through foodchains.

In addition, the resuspension of radioactive material by agricultural activities, fire events and traffic may enhance considerably inhalation doses. Resuspended material has a fraction of large particles (>10 μm) and the adaption of models for inhalation dose calculations to this large particle fraction and to hot particles needs to be investigated. Resuspension has the potential for a net transfer with time of material from more to less contaminated areas; such transfer processes have not been investigated in any detail and could be important in the long-term management and control of contaminated areas and, moreover, could have implications for the level to which it may be worthwhile decontaminating more heavily populated areas. This project is directed to a more detailed investigation of the potential importance of resuspension (caused by both natural and man-made phenomena) in these contexts and has the following main objectives:

- to predict airborne contamination levels and the levels of deposition on to surfaces by the resuspension of material
- to assess the significance of resuspension in terms of recontamination of surfaces previously decontaminated
- to determine whether buffer zones are needed around contaminated areas to ensure that airborne concentrations of resuspended material are reduced to acceptable levels and, if so, to determine their magnitude in general and for specific operations, eg, ploughing, etc.
- to investigate the effects of fire events of traffic and of agricultural activities (field work with tractors) on the amount, size distribution and other characteristics of the resuspended material
- to improve the dose assessment for inhaled large particles and hot particles

The objectives are met, firstly, by compiling and interpreting data already obtained in the former Soviet Union (partly during the 1991/92 phase of this project) and elsewhere and, secondly, by performing supplementary measurement programmes within the 30 km zone around Chernobyl and elsewhere. Meteorological parameters should be measured during the experimental campaigns to enable appropriate correlations to be explored and the results should be integrated with results obtained previously.

Investigations will be focused in the following areas:

a) Characterization of sources of resuspension

This work focuses on the identification and characterisation of sources of resuspension with an emphasis on the potential importance of the resuspension of small and large "hot particles". The experimental programme is directed towards the following:

- to characterize the surface soils in the regions of Chernobyl where resuspension measurements are made
- to evaluate the solubility of various gamma-emitting radionuclides

b) Characterization of resuspended material

Material resuspended under natural conditions and during agricultural activity will be investigated. A field campaign in Zapolie, May 1993 will address the effects of wind resuspension from grassland and of agricultural and vehicular activities. During a second field campaign in Chistogalovka, August 1993 the same effects will be studied for another soil type and another source distribution (at the measurement site in Chistogalovka the contamination is relatively high compared to its environment). A feasibility study about an experiment on the resuspension by a grassland fire will be performed.

The size spectrum of aerosols, in the range below 10 μm, will be measured at different heights above ground. In order to relate the results to the environmental conditions, the size distribution of particle mass in the same range should be determined as well as the airborne radionuclide concentrations in different diameter ranges. Particular emphasis will be given to the contribution of alpha emitters.

The large particle fraction of the resuspended material will be analyzed with a wide range aerosol classifier (WRAC) by an isokinetic sampler and by size fractionating air samplers. In addition, with rotating arm impactors the height dependence of the aerosol size distribution in the large diameter regime will be measured. For analyzing the data, image analysis techniques and instrumentation are being developed so that routine use can be made of rotating arm impactors with known collection efficiency and operating at three different heights simultaneously. Air concentrations of hot particles will be determined.

c) The importance of large particles in the transfer of material

The following experimental studies should be performed to elucidate and quantify the contribution made by large particles to the further spreading of deposited material and to investigate the influence that various environmental and meteorological parameters have on their transfer:

- the measurement of the rate of spread of contamination from more to less contaminated areas

- the determination of the deposition of large particles as a function of height above ground level and

- the evaluation of the results in terms of the transport of contamination by resuspended large particles.

In addition, an analysis should be made of the potential importance of relatively rare yet extreme meteorological phenomena or events as a means for dispersing large amounts of deposited material over considerable distances.

d) Resuspension measurements outside the former Soviet Union

Significant amounts of radioactive material released during the Chernobyl reactor accident were deposited on territory outside the former Soviet Union. Measurements of resuspension in these areas can be compared with those made in the 30 km zone where the nature and radionuclide composition of the deposited material is likely to have been different. Important insights on transport and resuspension processes may be gained by such comparisons, in particular the influence of the nature of the deposited material and of the characteristics of the surface at the time the deposit occurred and subsequently.

In selecting data to be analyzed for this purpose, preference should be given to complete or largely complete time-series of measurements of resuspended concentrations of radioactive material in air at different heights above ground having different surface covers (eg, urban, rural).

e) Inhalation risks from resuspended material

The size distribution of resuspended material is different from the size distribution of the average atmospheric aerosol. The applicability of current dose assessment models to resuspended matter will be investigated.

The biological effects of inhaled hot particles needs to be clarified. A review of the current research in this field will be given. In addition, a computer code for simulating track structures in cellular matter will be adapted to this problem.

CONTRIBUTIONS OF GSF

To assess the inhalation risks from resuspended large particles, measurements will be made of the aerosol size distribution in the regime above 10 μm diameter. The resuspension by wind stress as well as by agricultural activity will be studied. As sampling instruments, rotating arm impactors will be adapted in geometry and rotating frequency to sample size fractionated large particles at three different heights simultaneously. Image analysis technique will be developed to determine the particle size distribution in a routine procedure.

The application of an existing model on the distribution of pollutants in the respiratory tract after inhalation to the problem of dose assessments for large particles will be investigated. A review will be given on the biological effects of hot particles. A radiation transport program for the generation of track structures will be adapted to the problem of hot particles in biological matter.

Data collected by partners in the CIS will be edited for publication in the open literature.

In addition, resuspension data collected in the urban environment of Goiania, Brazil, will be documented and analyzed in comparison with Chernobyl and Neuherberg resuspension data.

CONTRIBUTION OF AEA Technology

To elucidate uncertainties in the role of large particles as vectors for the spread of deposited activity and how the resuspension of such particles will depend on various environmental and meteorological parameters. Experimental work will be in four areas:

1. A measurement of the rate of spread of contamination into a clean area from adjacent contaminated areas,

2. To determine the deposition of large particles as a function of height above ground level and to evaluate the results in terms of the transport of contamination by resuspended large particles,

3. To evaluate the airborne concentrations of radionuclides associated with large particles,

4. To collaborate with GSF in the collection and interpretation of available Soviet data.

AEA Technology will also liaise with CIS scientists in relation to the provision of meteorological and environmental parameters.

Experimental work to be carried out during 1992/93 will be based on complementing the 1992 measurements. These additional measurements are necessary in order to provide an assessment of the variability of resuspension with environmental conditions.

The technique used for the collection of large particles has incorporated several impaction surfaces positioned within a small rotational wind tunnel. Two of these samplers have been specifically built for the task.

CONTRIBUTION OF CEA

Air sample collection without disturbing the size distribution and concentration of large particles.

To achieve this goal, a high volume air sampler has been developed in 1992 which allows sampling in near isokinetic conditions (the air velocity inside the inlet is equal to the outside wind speed).

To determine the importance of the anisokinetic operation of aerosol collectors, the first experiments done during 1992 will be complemented by new sampling periods. The atmospheric concentration of gamma-emitting radionuclides will be determined under isokinetic and anisokinetic conditions.

Characterisation of the size distribution of surface soil particles.

To achieve this goal, a laser equipment will be used. This technique is based on laser beam diffraction according to particle size in 0.1 to 600 micrometers. For each sample analysis of gamma-emitting radionuclides will be conducted.

Caesium desorption in surface soil.

For the resuspension, the characteristics of the activity in the surface soil layer are of importance. Concentration profiles as a function of depth will be determined and the decontamination of the surface layer will be predicted.

CONTRIBUTION OF CIEMAT

To provide experimental basis for assessing the inhalation risks from resuspended particles, especially those smaller than 10 μm and to characterize the resuspended material measurements will be done under natural conditions and for agricultural activities. Emphasis should be given to the contribution of alpha emitters, as well as the contribution of gamma emitters. It will help to elucidate the behaviour of these radionuclides in function of the factors dominating the resuspension processes. Air concentration measurements will be done at different heights and total deposition using planchets will be measured.

Experimental work will be focused on the following areas:

1) Determination of activity distribution in function of size particles in the range size below 10 μm. The size spectrum of aerosols will be measured simultaneously at two heights above ground using high-volume cascade impactors. Airborne radionuclide concentrations will be evaluated.

2) Determination of the total deposition per unit area at different heights above ground.

3) Resuspension and deposition parameters in function of meteorological conditions will be obtained. Data obtained during 1992 about superficial distribution and migration in depth of alpha and gamma emitters will be used in order to characterize the source of resuspension.

CONTRIBUTION OF Fh - ITA

As far as measuring large particles is concerned, a suitable instrument is available at Fh-ITA. This Wide Range Aerosol Classifier (WRAC) is unique in its capability to collect large particles and to determine their size distribution.

Size-resolved resuspension rate of g-emitters will be measured as a function of wind speed for various vegetations (bare soil, crops, grassland etc.) and - weather and time permitting - different humidities and farming activities. For selected samples, autoradiographic localization of hot particles with subsequent electron microprobe

analysis will be performed. With a passive sticky foil sampler (VDI guideline 2119, part 4) weekly averages of the deposition flux can be determined. All measuring equipment necessary is available at Fh-ITA for the project. This includes the WRAC, wind sensor, rain sensor, sticky foil sampler, high-resolution g-spectrometer, microbalance, data acquisition computer and institute's car.

Radioecology

Contract COSU-CT920016 The transfer of radionuclides through the terrestrial environment to agricultural products and livestock, including the evaluation of agro-chemical practices.

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Total Contribution by the Commission: 665.500 ECU
12 months *1/11/92* to *31/10/93*

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Description of research work

Migration and extractability of radionuclides in soil

Migration of radionuclides in different soil types will be completed from last years sampling campaign on the nine selected experimental sites. A common sequential extraction procedure will be applied in order to follow the type of activity and solubility of radionuclides in the soil profile. The analyses will comprise gamma-emitting nuclides and on selected experimental areas, isotopes of strontium, uranium, americium and plutonium. In addition the soils will be characterized according to an international soil classification system. The soil types are classified as soddy podzol, sandy loam, peat and chernozem. The experimental sites are located in Ukraine, Belarus and Russia and have been contaminated by radionuclides in condensed form as well as by fuel particles.

A model will be devised for the description of the migration of radionuclides in the soil profile for different soil types. In order to characterize the migration parameters a column experiment will be set up under laboratory conditions for sandy and peaty soils.

The desorption measurements of caesium for freshly contaminated and aged soils will be completed from last years studies. The fixation dynamics will be related to the ionic status of the soil (Ca, Mg, K).

The extractability of radionuclides from hot particles will be studied on selected sites. In addition the influence of soil type and additives on the destruction of fuel particles will be studied in a model experiment.

Soil-plant transfer

Bulk samples of soil and vegetation from the nine sites selected and sampled last year will be determined on their radionuclide content. The vegetation consists of oat, ray grass, barley and potatoes. The experiments will continue for the forthcoming year with addition of lupine as one of the experimental crops. The analyses will comprise gamma-emitting nuclides and, on selected experimental areas, isotopes of strontium, uranium, americium and plutonium.The extraction procedures for radionuclide in soil samples are comparable with the ones used in the migration experiment above.

The external contamination of plants from resuspension and soil splash will be studied. Straw mulch will be assessed as a potential countermeasure to prevent external contamination of plants. Radionuclides as well as stable Sc and Ti will be measured. Particle size will be determined by scanning electron microscopy.

Countermeasures

Experiments will be carried out under laboratory conditions to determine promising additives (lime, fertilizer, zeolites etc) and to optimize application rates in order to reduce the transfer from soil to plants.

Experiments will be initialized in Ukraine, Belarus and Russia in order to study the effect of additives under field conditions.

Animal related studies

The bio-availability of radiocaesium (and possible radiostrontium) from different sources for absorption in the ruminant gut will be studied by in vitro measurements of availability and in-vivo measurements of absorption. The literature will be reviewed on the relationship between body size/age of domestic animals and transfer coefficients and biological half-life. In addition countermeasures will be will be evaluated with respect to trace elements in forages, pasture grasses and grazing animals.

Publication

Papers will be prepared for publication based on results obtained within the framework of the ECP-2 project in perspective (including) previous results obtained in the CIS and EC.

Contribution of the Swedish Radiation Protection Institute, Sweden

Soil-plant transfer

Completions of extraction procedures in four steps for soil samples from selected experimental plots. Gamma emitting nuclides and Sr-90 are to be analyzed for each extraction procedure in order to follow the type of activity and solubility of nuclides in the complete soil profiles. In collaboration with AEA the residue from the sequential extraction procedure will be analyzed for hot particles.

Completing gamma emitting and Sr-90 analyses of vegetable crops from the experimental plots (inside and outside the 30 km zone). This will provide the basis for soil-plant transfer calculation. Transfer data and evaluation of nuclide migration from soil to crops is made in close collaboration with joint EC and CIS laboratories.

Countermeasures

In order to reduce the transfer of strontium from soil to crops laboratory tests will be performed in order to select promising additives and to optimize application rates. The results obtained in the laboratory tests will be used in designing field experiments in the forthcoming years.

A primary test for a rotary cultivator equipment aimed for homogeneously mixing of soil together with additives on the field will be tested in Sweden within the framework of the Swedish Radiation Protection Program. This part of work is intended to be in collaboration with the contractor and Department of Radioecology at the University of Agricultural Sciences in Uppsala. If the method appears to be promising the rotary cultivator equipment will be introduced in the ECP-2 project in the forthcoming years.

Animal related studies, collaboration with ECP-5

Analyses Sr-90 contents of bones and of internal organs will be performed for native feral pigs originating from inside the 30 km zone. This part will contribute to the understanding of the turnover of strontium in pigs and is done in collaboration with Prof. B. Jones (ECP-5) at the University of Agricultural Sciences in Uppsala.

Contribution of the Catholic University of Leuven, Belgium

Sorption-desorption experiments in soil

These studies are intended to complete radiocaesium desorption measurements for freshly contaminated and aged soils, and to attempt to connect the fixation dynamics of different soils with the ionic status (Ca, Mg,K) of the soil.

They are also meant to study the fixation response of different types of soil to calcium treatment for short aging times (days). If such a response is significant, a treatment with soluble Ca-salts can be a simple countermeasure directly after a nuclear incident.

Modelling

In collaboration with CEN/SCK, a number of model chromatography experiments for connecting migration behaviour of radiocaesium with soil properties will be set up. This should provide a link to modelling input of Ioannina University.

Countermeasures

Laboratory studies on the efficiency of a set of natural zeolites as countermeasures for radiocaesium and strontium in types of soils will be carried out.

Contribution of the University of Barcelona, Spain

Resuspension and rain splash experiments

For the year 1992-93, two different approaches are proposed. The first one will be an extension of the approach started last year. The aim of this experiments is not only to evaluate radionuclide contamination in corps by near field resuspended soil particles, including rainsplash effect, but also to take advantage of this experiment in order to check the use of mulching as a possible countermeasure to prevent resuspension.

Therefore, a field experiment similar to the one carried out last year is proposed, but replacing the net placed on the soil by both contaminated and non-contaminated straw. With this experiment, information about foliar absorption and root uptake will be also obtained.

Sequential extraction in soils

It is proposed to carry out an evaluation of the sequential extraction schemes as a tool for predicting radionuclide mobilization in contaminated soils. With this aim three schemes will be applied: a) the one especially designed by the ECP-2 group for caesium and strontium, b) the one widely used for heavy metals but optimized for radionuclides and c) the one especially designed to study the role of the organic matter in radionuclides retention in soils. These schemes will be applied to the different soils collected last year in the different sites studied including soils containing both fuel and condensed particles.

Moreover, sequential extraction schemes will be applied to soils amended with zeolites and with calcium solution immediately after contamination with soluble or particulate radiocaesium and radiostrontium.

From the results obtained in this study and the values obtained by the Swedish Radiation Protection Institute for crops cultivated in the studied soils an evaluation of the methodology will be carried out.

Contribution of CEN/SCK , Belgium

Soil-plant transfer

The gamma spectrometry analyses of the soil and plant samples collected in 1992 will be carried on. The radiochemical analyses for U, Pu and Am determination will be performed on samples from the sites contaminated by fuel particles. The soil-to-plant transfer factors will be estimated and compared with data from the previous years. The migration processes in undisturbed soils will be modelled (in collaboration with Univ. Ioannina) in order to fit the actual distribution profiles.

Modelling

In order to develop predicting tools, controlled experiments will be conducted under laboratory conditions to characterise the migration parameters of various radionuclides (Cs, Sr, Ce, . . .) in problem soils (sandy and peat soils). The data obtained from leaching experiments on soils columns will be modelled by Univ. Ioannina and related to the soil characteristics determined by K.U. Leuven and Univ. Barcelona.

Similar tests will be carried out after application of different counter-measures (fertilising, liming,..)
to quantify their influence on the behaviour of the radionuclides in soil.

Countermeasures

The CEN/SCK will also contribute to the field experiments to be conducted in Belarus, Russian and Ukraine and will participate to the assessment of the efficiency of the different counter-measures applied.

Contribution of the Institute of terrestrial Ecology, UNITED KINGDOM

Animal related studies

1. Bio-availability

ITE and MLURI have developed an *in-vitro* CsCl extraction technique for predicting the availability of radiocaesium for absorption in the ruminant gut, mostly using sheep as the experimental animal. We hope to adopt a similar approach to radiostrontium and to validate it further with measurements of true absorption in cattle. Initially ***in-vitro* extractions** will be performed on a wide range of contaminated sources which are either routinely fed to, or ingested by grazing ruminants. Subsequently some of these sources (with varying availabilities) will be fed to cattle and the **true absorption** of radiocaesium (and radiostrontium if possible) measured. Preliminary experiments to look at methods of measuring the true absorption of Sr are currently being conducted under the CEC Radiation Protection programme.

2. Preparation of papers for publication

Many of the results of studies in animal radioecology after the Chernobyl accident have been published in Russian-language literature and are therefore have a limited distribution worldwide. CIS participants will summarize studies which they would like to publish in English in journals and translate them into English. ITE will then prepare paper based on these documents and collaborate with the authors in getting them to a suitable state for submission to journals.

3. Literature review on the relationship between body size/age of domestic animals and Ff, Fm and T1/2b

Initially we proposed a study to look at the relationship between age/ body size and F_f and $T_{1/2b}$ of radiocaesium. After group discussion we decided to postpone this experiment pending a detailed review of literature already available and to include all domestic livestock and other radionuclides such as strontium, iodine, ruthenium and curium etc.

Further consideration will then be given to experimental planning for a suitable experiment in 1994.

Contribution of the University of Ioannina, Greece

Modelling

A new model will be devised for the description of the diffusion of trace elements, deposited on surface, in various soils. A laboratory experiment will be carried out (C. Vandecasteele) with undisturbed columns of a variety of soils and depth profile data after 4, 8 and 12 months will be employed for the validation and calibration of the model. The model will be eventually applied to field data.

The animal model (P.A. Assimakopoulos, et al, Health Physics, 61;245-253; 1991) developed at Ioannina will be appropriately modified and applied to the analysis of data from the experiment to be conducted by the animal group (B. Howard, et al.) for the investigation of animal size dependence on transfer rates of radiocontaminants from an animal's diet to it's tissues and milk.

The above animal model will be applied to existing data, e.g. the decontamination data obtain from heavily contaminated bulls, transferred to uncontaminated pasture, to be supplied by our CIS colleagues (R.M. Alexakhin, et al.).

Contribution of the Agricultural University of Norway, Norway

Animal related studies

The effect of countermeasures on trace element levels in forages, pasture grasses and grazing animals will be assessed (AUN).

Contribution of the AEA Environment and Energy , UNITED KINGDOM

"Hot particles"

The behaviour of fuel particles deposited on soil as a result of the Chernobyl accident was, and still is, a cause for concern.

Currently, there is interest in fuel particles both in the field and in the laboratory. In the field, the behaviour of particles is critically dependent on two processes viz, leaching and adsorption. One of these processes will be rate-determining: the latter process will also be soil specific. A knowledge of these rate constants is essential input for computer modelling of the situation. The laboratory requirement is to understand the behaviour of particles in the sequential extraction schemes now favoured by laboratories within the ECP-2 project. The extractability of the gamma emitting radionuclide plus ^{90}Sr will be studied in aged particles using the sequential extraction procedure formulated or agreed by the ECP.2 project. The condition of fuel particles in soils after sequential extractions carried out by Dr. Melin and Dr. Rauret will be studied.

Contribution of the Paul Scherrer Institute, Switzerland

Resuspension and soil splash experiments

The aim will be to study the contribution of rainsplash to foliar absorption of resuspended 137 Cs by preventing root uptake and using another crop. Two experiments, the first using straw mulch and the second with no treatment, will be carried out.

Radionuclides as well as stable Sc and Ti will be measured in the majority of the samples. Particle size of the aerosol will be determined by scanning electron

microscopy.

Contribution of the Belarus Institute of Agricultural Radiology, Belarus

Soil-plant transfer

The experimental plots on which investigations have been carried out are preserved. In the period from October 1, 1992 to September 30, 1993 the following works will be carried out:

Analytical work

a) determination of Cs, Sr and Pu radionuclides concentration in vegetation and soil samples.
b) sequential extraction of soil samples.
c) intercalibration for determination of Sr-90 in vegetation and soil samples.

Field works

a) planting and sowing of new groups in the experiment winter and spring rape, leguminoses.
b) applying of countermeasures.
c) sampling.

Animal related studies

Investigations on cattle-breeding

a) estimation of Cs-137 availability from different sampling
   - measurement of Cs-137 specific activity in fodder and milk
   - diet structure estimation
   - presentation of results to Brenda Howard
   - training of a specialist in the UK

b) Estimation of basic microelements concentration in soil, fodder-crops, blood and milk of agricultural animals kept on the territory contamination by radionuclides
   - selection of typical farms
   - sampling of fodder, soil, blood and milk
   - microelements determination

Contribution of the Russian Institute of Agricultural Radiology, Russia

Countermeasures

Countermeasures to reduce transfer of 137 Cs, 90 Sr into crops

1. Field experiments of study on effects organic fertilizing (peat-manure compost, carbonate sapropel) on transfer radionuclides in crop. Tula Region, Plavsk

2. Study on effects of application sorbents (zeolite and natural sorbents). Tula region, Plavsk

3. Study on effects of application mineral nutrition (NPK) on transfer of 137 Cs and 90 Sr into crops. Bryansk Region, Novozybkov Tula Region, Plavsk Kaluga Region

   3.1. Effects of doses and types of nitrogenous fertilizers on transfer of 137 Cs intc crop.
   3.2. Effects of doses of phosphoric fertilizers on intake of 90 Sr into plants.

   The experiments will be conducted on oats, yellow lupine, potato.
   Evaluation of quality of feeding stuffs (diet balanced on microelements content) Bryansk Region, Novozybkov

Modelling

Modelling of radionuclides transfer from diet to animal products. Obninsk

Contribution of the Ukrainian Institute of Agricultural Radiology, UKRAINE and the Research and Industrial Association, PRIPYAT, UKRAINE

Soil-plant transfer and countermeasures

1. Influence of countermeasure on radionuclides transfer to the yield of oats.
2. Assessment of countermeasure efficiency depending on the biological peculiarities of plants

3. Investigation of radionuclides mobile forms in soil depending on the application of countermeasures

4. Influence of agrotechnical soil characteristics on radionuclide behaviour in soil and their transfer to plants.

Resuspension and rain splash experiment

Animal related studies

1. Investigation of radiocaesium availability from various types of forage.

2. Estimation of basic microelements contents in soil, forage crops, blood and milk of agricultural animals on the territory, contaminated with radioactive substances after the CHNPP accident.

3. Analysis and preparation of experimental field for 1994 experiments on the

assessment of countermeasure influence on microelements content in animals organisms.

"Hot particles"

In model experiments the processes of fuel particles destruction in soil will be studied depending upon physico-chemical soil properties, meliorants applied and characteristics of fuel particles.

Radioecology

Contract COSU-CT920017 The modelling and study of the mechanisms of transfer of radioactive material from terrestrial ecosystems to and in water bodies.

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Total Contribution by the Commission: 495.500 ECU
12 months *1/11/92* to *31/10/93*

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Description of research work

Contribution of the Institute of Freshwater Ecology

a) A joint study (with ECN, Petten; TYPHOON, Obninsk; Institute of Hydrometeorology, Kiev; Institute of Geology, Geochemistry and Geophysics, Minsk and LNETI, Portugal) of the processes involved in the delayed run-off of radiocaesium in dissolved form from peat bogs. Streams flowing from peat bogs into Kajanovskya lake in the Bryansk region of Russia will be sampled at regular intervals and analyzed for radiocaesium content, ammonia concentrations and major ion chemistry. Relationships between radiocaesium and major ions at different stream flow rates will be examined to identify the dominant release process in the bogs.

b) a study will be carried with ECN, Petten; TYPHOON, Obninsk; Institute of Hydrometeorology, Kiev and LNETI, Portugal to measure in situ K_D of radiocaesium in the sediments of Kajanovskya lake.

c) The IFE has carried out a statistical analysis of post Chernobyl radioactivity levels in trout, perch and pike in two UK lakes. More published data of fish radioactivity levels from the UK will be analyzed to increase the international database available for comparison of inter-site and inter-species differences. These data will be compared with similar radioactivity size data from the Netherlands, Norway, the Ukraine (Institute of Hydrobiology) and Russia (Severtzov Institute). In this way it should be possible to identify ecological factors, such as feeding habits, which determine the occurrence of the size activity effects.

d) The IFE will also play a consultative role in the experimental programme which is proposed at the Ukrainian Institute of Hydrobiology (UIH). This programme, in which the practical work is being undertaken by INETI and UIH, will study the factors affecting metabolic loss rates of radiocaesium from fish of different size.

e) Combined monitoring exercise with Ukrainian Institute of Hydrometeorology to obtain basic data on contaminated lakes in the Chernobyl area to aid the development of future programmes.

ECN contribution

Significant progress has been made in laboratory studies of radiocaesium mobility, yet in-situ studies in the W-European environment are hampered by the very low activities in aqueous compartments soon after the accident. Because it is our ultimate goal to predict the transport of radiocaesium through the environment, in-situ measurements of this radionuclide and its mobility controlling parameters are necessary to verify our predictions and to indicate additional factors affecting radiocaesium transport that should be considered in our models. The joint research within this project in contaminated CIS-environments is relevant not only because of the direct importance of radiocaesium remobilization but also because the levels of contamination allow us to study processes in more detail than is possible elsewhere.

The contribution of ECN focuses on the geochemical processes underlying the movement of radiocaesium in environments contaminated after the Chernobyl-accident in the CIS, or in W-European analogues to those environments. Following a combined field and laboratory approach, the research emphasizes the determination of reliable (in-situ and laboratory predicted) sediment/water distribution coefficients (K_D's). This parameter quantifies the interaction of the radionuclide with the solid compartment in sediments and soils and controls, therefore, its mobility.

The mobility of radiocaesium in CIS-environments varies not only between different sediments and soils, because of differences in the availability of illitic frayed edges, but is also affected by the chemical form in which the radionuclide was deposited (fuel particles/ionic) and subject to chemical changes within the environments of deposition. ECN research to date has focused on ionic radiocaesium inputs in W-European freshwater environments after the Chernobyl accident and has shown that post-depositional changes in the pore water chemistry of freshwater sediments may result in partial remobilization of sediment-bound radiocaesium. It was found that radiocaesium mobility was controlled by diagenetically-produced NH_4 in the sediment pore waters, which competes with the radionuclide for sorption sites on illitic clay minerals. It is believed that these reactions can soon be modelled quantitatively, which should allow is to "subtract" this sub-process from the more complicated set of processes controlling radiocaesium in CIS environments contaminated by both ionic and fuel particle-radiocaesium.

Laboratory and modelling work at ECN has shown that there is a strong interrelationship between the kinetics of radiocaesium sorption and the reversibility of the process. A slow uptake process is believed to represent migration of radiocaesium to illitic interlayer sites, from which the radionuclide is not easily released. The rate of this migration depends strongly on the major competing cation and is very relevant for the evaluation of the movement of radiocaesium in the aquatic environment and for estimating the risk of remobilization from soils or sediments. Present work within this project emphasizes the reversibility-question, its effect of K_D's, and how to deal with it in our transport models.

The behaviour of radiocaesium in areas with highly organic soils/sediments, which are being studied within this project, deviates from that in areas with mainly inorganic soils/sediments in that the radiocaesium availability remains high for many years and is reported to be dependent on divalent cations and Ph. These observations necessitate an investigation of the effects of organic functional groups on the retention of Cs by the highly selective frayed edges sites of illitic clay minerals. It has been reported that fulvic acids may enter and expand the interlayers of expandable clay minerals at low Ph values. It is not known to what extent organic acids can enter the illitic frayed edges and "compete" with either through a physical blocking of these Cs-selective sorption sites or by preventing the edges to collapse. Such organic interferences may also cause radiocaesium sorption and the reversibility of the interaction to be dependent on Ph. An investigation of the above hypothesis includes joint laboratory experiments with the CIS partners.

ECN works closely together on these subjects with the Institute of Freshwater Ecology (IFE Windermere, UK) the Scientific Production Association TYPHOON (Obninsk, Russia and the Ukrainian Hydrometeorlogical Institute (Kiev, Ukraine).

Contribution of KEMA.

field work

During the first year of the project, 1991/1992, 2 persons of KEMA have carried out field work (collecting fish, macroinvertebrates and water samples, processing samples) near the 30km zone. For the second year of the project sampling was carried out in November 1992 and is planned in April and the autumn of 1993.

interaction among partners

Field work is carried out in close cooperation with the "fish group" of Dr I Ryabov (Institute Animal Evolutionary Morphology and Ecology RAN, Moscow, Russia). The total group consists of 6 persons. Support is also given by Dr M.D. Lobakin of the Integrated Radioecological Expedition, Kiev, Ukraine.

ENEA-DISP Contribution

Radionuclides Transport and Resuspension processes

This joint research proposal on the quantification of radionuclides transport from the Pripyat and Dnieper Rivers to the Dnieper Estuary is aimed at:

1. Inter-comparison of sampling and measurement procedures (continuation 1991-1992 programme);
2. Assessment of radionuclide solid-liquid interactions and the quantification of the relative importance of the Chemico-physical parameters of water and suspended materials;
3. definition of criteria for identification of representative sampling points in large watershed-river systems.

The inter-comparison exercise will be conducted (April-May 1993) in the following 3 sites with different content of radionuclides and different concentrations of suspended materials:

- in the Pripyat river (30 Km zone);
- in the upper part of the Kiev Reservoir;
- in the lower part of the Kiev Reservoir;

The radionuclide solid-liquid interaction studies will be conducted during 1993 (April-May and in summer) in 3 rivers (Pripyat river near Chernobyl, Uzh river near Chernobyl and a tributary of Uzh river), characterized with different morphological scale, different hydrological regimes and different levels of Cs-137 contamination.

Water and suspended materials will be taken in different hydrological conditions to compare the results achieved in water bodies with different morphological scales. Bottom sediments will be taken at each sampling profile. Scientists from Ukrainian Hydrometeorological Institute of Kiev will participate in the ENEA-DISP research activities in Italy to compare laboratory and field methodologies.

Modelling

The ENEA Department of Environmental Analyses and Monitoring (ENEA-AMB) will collaborate in this project for the modelling aspect. The second year of activity, will be focused on the calibration of mathematical models describing, the transport of radionuclides in the river-reservoir systems (in collaboration with the Institute of Cybernetics of Ukrainian Academy of Sciences).

INETI Contribution

OBJECTIVES

1) Processes of remobilization of deposited radioactivity

Radiocaesium (and eventually radiostrontium) mobilization from soils and/or freshwater sediments, from the Chernobyl area, should be quantified. Laboratory leaching experiments using ion exchange techniques for a selective ion separation, or different displacement solutions, will be carried out.

2) Dynamics of accumulation of radionuclides in fish

The role of age and size on radionuclide accumulation and excretion by fish should be studied. The effect of different environmental parameters on the rate of excretion of radiocaesium will be investigated on different sized fish and the biological half-lives determined.

The feasibility of multiple tracer measurement on fish (Cs-137 from Chernobyl and Cs-134 from labelled food) will be assessed.

COLLABORATION

The studies concerning the remobilization of deposited radionuclides will be carried out in collaboration with the University of Leuven (ECP 2) and the Ukrainian Ministry of Chernobyl (Kiev).

The fish studies are carried out in close collaboration with the Institute of Hydrobiology of Kiev and the Ukrainian Ministry of Chernobyl (Kiev). There is also some collaboration with ECOPOLIS and Pripyat Association.

After the Chernobyl accident a relationship was observed in some fish species between radioactivity levels and fish size. This relationship did not occur in all species and a number of hypotheses have been developed to explain the phenomena. Among these are differences in metabolic rates of intake and excretion of the isotopes and ecological differences in diet and feeding rate. In previous work we have shown that radiocaesium excretion rates in brown trout are a function of size and ambient temperature, and that absorption varies with food type. In the planned work we will collaborate with LNETI, IFE, The Institute of Hydrobiology, Kiev and the Severtzov Institute, Moscow in the expansion of this work to study the environmental factors affecting excretion in different sized fish by other species.

To date there is no information in the literature concerning the variability of radiocaesium absorption by different sized fish. We thus intend to perform a laboratory study in Norway on the size effect on radiocaesium absorption from different food items. If available, our results will be compared with results from field experiments in the Chernobyl area.

Moreover, we have planned to perform field estimates for food consumption, and thus radiocaesium budgets, for selected age groups of a cypernid species from the Chernobyl area. This will give new information on the ecological differences between different size groups, which may be important determinants for the relationship between radioactivity and fish size.

Radioecology

Contract COSU-CT920018 Evaluation and development of decontamination strategies for a range of environmental situations and evaluation of their efficacy and other impacts.

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Description of research work

The Commission of European Communities has signed an agreement with the Republics of Russia, Belarus and Ukraine in order to set up a scientific collaboration about the aftermath of the Chernobyl accident. Among the other actions of this project, one of them is entitled "Decontamination Strategies".

The general aim of this action is to develop decontamination techniques, to obtain data on their efficacy and costs when applied on a practical scale. The results should, with the integration of previous studies in that field, form a sound basis for the development of practical strategies and policies for the decontamination in the event of a future accident. Actually the work is organised around 7 sub-projects:

- decontamination of urban surfaces,
- decontamination of agricultural soils,
- treatment of contaminated wood producing usable by-products,
- decontamination during food processing,
- study of self restoration of ecosystems,
- management and pre-treatment of residues,
- modelling and cost-benefit analysis.

It was decided to divide ECP4 in 7 sub-projects; they are listed below with a few remarks if necessary. A more detailed description of each sub-program is added in appendix.

subproject 1:

Urban decontamination: tests of techniques such as sandblasting, high pressure water hosing, use of reagents (clays, NH4NO3...), decontamination of freshly contaminated surfaces, management of wastes generated by these techniques.

subproject 2:

Soil decontamination: tests of techniques of soil scrapping (Decontaminating Vegetal Network), mapping of areas where these techniques may be applied, evaluation of the efficiency of different polymers in order to reduce dust and runoff.

subproject 3:

Treatment of contaminated wood. Study of techniques allowing to re-use contaminated wood by making usable products: gasification, pulp for paper, ethanol.. incineration of actual wastes.

subproject 4:

Food Processing: field studies of Sr and Cs uptake, migration and transfer from fodder to milk, investigation of decontamination while processing milk using European and Ukrainian technologies, study of meat decontamination, set up of a pilot installation.

subproject 5:

Study of self restoration and ecotoxicological impact of man made countermeasures, studies for different types of landscapes, elaboration of predictions up to the year 2010.

subproject 6:

Management of waste; presently, this topic is restricted to feasibility studies using the wastes actually generated in the other subprojects, such as residues coming from sand blasting in urban decontamination, rolls of grass and soil, organic byproducts of food processing.

subproject 7:

Modelling and cost-benefit analysis; definition of methodologies necessary to evaluate and establish decontamination strategies by cost-benefit analysis and multi-criteria analysis.

Radioecology

Contract COSU-CT920019 The behaviour of radionuclides in natural and semi-natural ecosystems (forests, marshes, heater, etc.)

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Description of research work

BACKGROUND AND OBJECTIVES

From the Chernobyl related investigation it has resulted that the radionuclides deposited in non-agricultural areas have a long-term impact on environment and/or on food chain. These non-agricultural areas, can be defined as "natural" and "semi-natural" on the basis of the alterations due to the human intervention. Natural ecosystems are those in which the flora and fauna are original and spontaneous and the vegetation has not undergone structural modifications by man. In the semi-natural environments, the flora and fauna are still native but the vegetation is greatly modified by human intervention; the relationships between biological forms have been modified.

The studies, carried out in natural and semi-natural ecosystems, show that the deposited radionuclides accumulate in some compartments of these ecosystems and are bioavailable for long time. The foods produced in semi-natural environments could contribute significantly to the long-term dose commitment to man. This is mainly attributable to the high levels of radiocaesium found in natural products (grasses, milk, berries, mushrooms, wild animals) and to the much longer ecological half-lives of Cs in these environments compared with agricultural systems.

Furthermore some compartments, like litter layers, could represent a transitory storage for deposited radionuclides which, in the long run, could be available to be transferred to the adjacent ecosystems.

Contrary to the agricultural ecosystems (characterized by high homogeneity of vegetations and of soil chemico-physical parameters) natural and semi-natural environments (forests, upland grasslands, pastures, swamps etc.) show high biotic diversity and heterogeneous values of soil parameters. For these characteristics the models, developed for describing the radionuclides behaviour in agricultural systems are not directly applicable to natural systems.

Further investigations are strongly necessary in order to fully understand the processes governing the radionuclides behaviour in these environments and to define the most suitable countermeasures.

In the 30-km zone around Chernobyl Nuclear Power Plant, about 50% of the land is covered by semi-natural environments: 36% of the area is covered by forests, 11% by meadows and 8% by swamps. The 60% of the wooded areas are formed by pine trees, deciduous trees (oak, white beech, birch alder) cover the remaining areas.

On the basis of these considerations, in the following are reported the objectives identified as relevant for the next year and for the activities started on 1992 are also indicated the collaborations with the C.I.S. Institutions.

- Role of Forest Litter on Radionuclides Migration Through Soil (RPII- Ireland). The role of the litter layers of the forests as delay barrier for the radionuclides transfer to the mineral layers of the soil will be studied. The rates of loss of

nutrients and radionuclides from forest litter will be measured and the physical, chemical and biological factors which regulate decomposition rates will be determined. The leaching of radionuclides from the litter layers will be assessed. To these studies collaborate the Moscow State University.

- Radionuclides availability and mobility in forest organic substrates (UCL-Belgium). The importance of the textural and chemical composition of the forest organic substrates will be assessed.

- Radionuclides Cycle in Forest Soil-Plant Systems (BfS-Germany). The mechanism of radionuclide uptake in different species of plants will be studied and transfer equations developed taking into account the influence of the soil characteristics. For this aspect, in 1992, a collaboration with Moscow State University and RIA "Pripiyat" has been established.

- Effects of biological 'fixation' and seasonality on radionuclide uptake by herbaceous plants of forest (ICSTM- United Kingdom).

  This study will investigate whether radionuclides are sequestered in the superficial layers of soil, directly by living biomass or by dead and decaying tissues. Furthermore, the seasonal effects on plant uptake will be studied.

- Processes Affecting Radionuclides Vertical Migration Along Meadow Soil Profiles and Transfer to Vegetation (ENEA-DISP- Italy).

  The physical and chemical factors which regulate the transfer of radionuclides along the soil profile, will be assessed in meadows and swamps. The role of the soil and vegetation characteristics will be assessed. The transfer factors from soil to different species of grasses will be evaluated. In these activities are involved the following C.I.S. Institutions: Moscow State University, Russian Research Institute of Agricultural Radiology, Ukrainian Research Institute of Agricultural Radiology and RIA "Pripiyat"

- Radionuclides Transfer to Wild Animals (SUAS-Sweden)

  The seasonal variation of radionuclides transfer from contaminated environment to wild animals will be evaluated. Wild boar (Sus scrofa) and roedeer (Capreolus) will be used as experimental animals. The Institute of Zoology, the Institute of Geography, the Institute of Nuclear Research of the Ukrainian Academy of Sciences and RIA Pripyat collaborate to this study.

- The long-term behaviour of radionuclides in semi-natural ecosystems (NIRP-Norway).

  In soil-plant-animal systems the ecological half-lives of radiocaesium will be determined. The effects of the site specific parameters on the persistence of caesium in these ecosystems will be evaluated. Further the contribution to the dose of food products from these environment will be assessed.

- Parameters affecting the caesium vertical migration and the external exposure (GSF-Germany)

  The influence of the deposition mode and of soil characteristics on the migration behaviour of radiocaesium in soil and on the resulting external exposure will be investigated.

INDIVIDUAL CONTRIBUTION OF THE PARTICIPATING ORGANIZATIONS

RPII, Ireland

Forests, and coniferous forests in particular have a high interception capacity for airborne materials. Much of the radionuclides released from Chernobyl accident were in aerosol form. Following the Chernobyl accident about 80% of the airborne radionuclides which passed over the coniferous forests of the Chernobyl area was intercepted by the forest canopy.

Subsequent washing by precipitation and natural needle fall resulted in the transfer of this contamination to the forest floor where it became incorporated into the upper layers of the soil profile. All elements associated with these organic layers of forest litter are mobilised by the processes of decomposition. Mobilised elements either migrate further down the profile, become bound to other soil components or are assimilated by the plants and fungi which feed off these soil layers.

The rate of decomposition in coniferous forests is relatively slow with the effect that in the case of Chernobyl radiocaesium the litter layers of the forest floor constitute a physical and biological barrier to the migration of radiocaesium through the forest profile. In addition this component of the forest ecosystem acts as a sink which slowly feeds the radiocaesium uptake of the other forest components (shrubs, trees, fungi, etc.). The process of decomposition of forest litter and the rate of release of radiocaesium from decomposing litter controls the cycling of radiocaesium within the forest. This study aims to measure the rate of decomposition and the radiocaesium budget of forest litter on a seasonal basis and observe the effects of physical and biological factors on these processes.

During 1992 the background work involved in setting up such a study was carried out, and the study commenced. A good working relationship was established with scientists from Moscow State University who have experience in similar aspects of research.

The work already carried out on both sides was compared and intercalibration exercises were carried out including laboratory visits to compare the techniques used in our different countries. The objectives of our study and a sampling programme were agreed. A site was selected at approx 7 km distance from the Chernobyl NPP and the necessary experimental equipment prepared and installed on site. In October 1992 a 12 month sampling programme was initiated.

For the coming year it is proposed to continue with the monthly sampling until October 1993. The final analysis of samples and the compilation of data will be carried out. It is

intended that joint publications are prepared with the scientists of Moscow State University. There are important links between this study and those of other (ECP-5) scientists involved in studies within the same forest ecosystem (eg. studies of radiocaesium transfer from soil to plant and plant to animal). Each of these studies will contribute to the understanding of the behaviour of radiocaesium within the forest ecosystem.

UCL, Belgium

The laboratories of the Catholic University of Louvain (collaboration between the Plant Biology, Nuclear Chemistry and Soil Sciences laboratories) are invested from 1989 in the study of the behaviour of radiocaesium in natural ecosystems and more particularly in forest ecosystems. Different studies have shown the importance of the textural and chemical composition of the forest soil layers on the radionuclide availability and mobility in forest organic substrates. The highest radiocaesium retention was observed in the hemi-organic layer.

Different specific soil components can interact on the radionuclides sorption or fixation (organic and/or mineral sites). UCL contribution is to identify the main source of radionuclides fixation in the Chernobyl forest soils and to test the reversibility and duration of these processes. Discussion will be conducted with the CEC and C.I.S. colleagues about the favourable or restrictive soil parameters for a good radioecological management of the contaminated area.

BfS, Germany

Radionuclides behave differently in natural ecosystems than in agricultural areas, which is expressed by the significantly higher activities in forest's plants and animals, compared to agricultural ones at same deposition rates. As forest ecosystems are important to man since they provide wood, paper, wild berries, mushrooms, game and recreation, it is of interest to analyze the behaviour of radionuclides in soils and uptake of these elements by plants.

The behaviour of radionuclides in soils depends on the properties of soil and of the nuclides. Forests are characterized by undisturbed soils with distinct horizons. The upper organic horizons show a different chemical-physical behaviour than the following mineral horizons. For this reason, important soil properties, such as texture, organic matter content or kd-value are to be measured separately for each horizon.

The transfer of radionuclides is dependent on plant and soil parameters. Considering the soil/plant transfer heterotrophic mushrooms and autotrophic green plants have to be distinguished. Since radionuclides are not distributed homogeneously in soil, it has to be analyzed in which horizon green plant are rooting and mycelium is living and to what extent nutrients are taken up from the different horizons. Transfer equations, including the soil properties, will be developed by using statistical methods.

Besides radiocaesium we have found high concentrations of the fission products cerium and europium in plant material. Because Ce-144 was the most frequent longlived radionuclide following the Chernobyl accident, cerium is taken into account to our

investigations. Also the behaviour of actinides in soils and their uptake by plants will be studied, as these elements are having very long half-lives and high dose factors. To summarize, our programme for the year 1993 can be described in the following topics:

1. Analyzing the distribution in forest soils of the radionuclides Sr-90, Ru-106, Cs-134, Cs-137, Ce-144, Eu-154, Pu-238, Pu-239-240 and Am-241;

2. Analyzing the content of the above reported radionuclides in different habits of mushrooms and autotrophic green plants;

3. Analyzing the soil properties for each occurring horizon;

4. Development of transfer equations for forest understorey plant species.

This research is carried out in collaboration with the C.I.S. colleagues in the 30-km zone around Chernobyl at the three sampling plots D1, D3 and K3 since 1992. Analog investigations will be accomplished at several sampling points in South Bavaria (only strontium, antimony and caesium).

ICSTM, United Kingdom

There is much evidence that 'fixation' within surface organic horizons of soils is of major importance to the cycling of radionuclides within the 30 km zone. However, although it is known that organic matter plays an important role in this fixation it is still unclear whether radionuclides are sequestered directly by living biomass or by dead and decaying tissues. In the former case 'fixation' would require efficient recycling within (mainly) microbial tissues; in the latter it is likely to result from humic-radionuclide complexation reactions. This study will attempt to distinguish between these two groups of processes and, further, to determine any seasonal effects on plant uptake due to variation in radionuclide availability across the spring-summer growth season.

Two experimental plots will be established within the 30 km zone in the spring of 1993. These will be used as sampling frameworks from which replicated, monthly samples of vegetation and soil will be removed. Radionuclide activities and stable element concentrations will be followed in individual plant species across the growth season. In addition, total soil radioactivities will be determined. Labile radioactivities and stable element concentrations in soils will be determined by the emplacement and sampling of ion exchange resin bags on a monthly basis. Organic complexation of radionuclides within monthly soil samples will be determined by gel filtration chromatography in the UK following sample preparation in the Ukraine. The expertise of Prof. Tikhomirov's group (MSU, Moscow) in this field will be invaluable. Direct biological fixation of radionuclides in fungal hyphae will be assessed by sampling microscopic hyphae from the litter layers of soils; radionuclide activities and stable element concentrations will be determined in these samples which may be identified in the autumn by the presence of fruiting bodies. Assistance will be sought in this connection from Professor Zhdanova's group at the Institute of Microbiology and Virology, Kiev. Results from this study will give a dynamic picture of radionuclide uptake by plants during one growing season in relation to soil biological activity and organic complexation processes.

ENEA-DISP, Italy

The results obtained in meadows after the Chernobyl accident showed that the deposited radionuclides remain in the superficial soil layers for long time. Particularly in the Chernobyl area, in well drained soil the radionuclides are mainly retained in the top soil layers. In wet area, in peat soil more mobility along the soil profile has been observed. The results obtained seem to show that the soil type and water content variability have a great influence on the radionuclides distribution pattern along the soil profile.

The aim of this project is to study radiocaesium and Sr-90 behaviour in natural and semi-natural meadows in order to assess the influence of the 'situ' dependent parameters (soil characteristics, vegetation, chemico-physical forms of the deposited radionuclides) on the radionuclides availability to root uptake and to migration along the soil profile.

During the first year of activity 4 sampling areas, characterized by different chemico-physical characteristics of soil and different ratio of fuel particles to condensation forms in the Chernobyl fallout, have been selected. In these areas the distribution along the soil profile of radiocaesium and others gamma-emitting radionuclides has been assessed. In the coming year the influence of the soil characteristics (cation exchange capacity, organic matter content, water content, exchangeable potassium and phosphorus, total N, P and K, Ca and Mg) on the Cs-137 and Sr-90 availability to root uptake and to migration along the soil profile will be assessed. To this end seasonal sampling of soil, soil solution and different species of vegetation will be performed.

The data obtained will be compared with those obtained in different scenarios located in Italy and in C.I.S. in order to:

- assess the main 'situ' dependent parameters, that influence the transfer processes of the radionuclides in these ecosystems;
- develop a model taking into account these parameters and their variability.

SUAS, Sweden

Uptake and accumulation of radionuclides in animals is of considerable importance in situations of environmental contamination. A substantial part of the radiation dose to humans will come from animal products consumed. The transfer of radionuclides through the chain soil-plants-animals to milk and meat from domestic animals is well documented but in wild animals living in semi-natural and natural environments much less is known. The present program is designed to study the uptake of radiocaesium in wild boar (Sus scrofa L.) and roedeer (Capreolus L.) by taking seasonal samples of animals living in the 30-km zone around Chernobyl. In close cooperation with soil and plant specialist the intake of the animals will be determined. Models will be constructed for calculation of the transfer of contaminants from soil through plants.

The studies during the first year has been performed in cooperation with the Institutes of Geography, Nuclear Research and Zoology of the Ukrainian Academy of Sciences, and RIA Pripyat. Some plant results have also been obtained from BfS (Germany).

The preliminary results from the first year show that most animals of both species have surprisingly low contamination levels in muscular tissue and internal organs. This is a reflection of the contamination level of forage plants eaten by the animals. The soil ingested by the wild boars rooting in the ground will be payed special attention as a potential source of radioactivity.

NIRP, Norway

The semi-natural ecosystem has shown to be an important contributor of dose to man. Although the intake of food produced in semi-natural ecosystems is not very high, it usually contributes significantly to the internal dose. For estimation of doses it is important to have knowledge concerning radiosensitivity,long term behaviour and degree of consumption of food products from these ecosystems. Determination of ecological half-lives is necessary for prediction of the long term behaviour of radioisotopes in the environment. By investigation of the soil-plant-animal nutrition pathway, it is possible to determine the transfer including aggregated transfer factors. the determination of bio-availability of radionuclides in different types of soils and plant species is also an important task. The main objectives of this study will be:

- to determine of ecological half-lives of radiocaesium in soil- plant-animal systems in three different semi-natural ecosystems, and identify of site specific parameters influencing the ecological half-lives;
- to estimate the dose contribution from food products from semi- natural ecosystems, and to determine their relative importance for the total dose using dietary studies;
- to characterize the physico-chemical forms of radionuclides in soil-vegetation systems. The availability of radiocaesium by sequential extraction and the mobility factors will be determined.

The project will be carried out in affected areas in Russia, Ukraine and Belarus. The project will require close cooperation with research institutes of agricultural radiology in all three republics.

The practical part of this work will include soil, vegetation and animal samples from all three republics in contaminated areas. This sampling sites are established from 1990 and 1991 and contains natural pasture, meadow and forest pasture. We will prefer pastures were activity levels in animals and animal products already have been determined.

In connection with determination of ecological half-lives and activity levels for wild animals will be established a strongly link with the colleagues involved in these studies.

GFS, Germany

Measurements of the external radiation exposure in air after the deposition of radiocaesium on grassland revealed systematically 40 % higher values per unit soil contamination for sites in the Ukraine as compared in Bavaria. This can be explained only by a slower vertical migration of radiocaesium in Ukrainian soils. A first inspection of the soil characteristics (clay, sand, pH), however, indicated no specific reason why the

radiocaesium migration should be slower in the Ukrainian soils. It is, therefore, conceivable that the physico chemical properties of the deposit, which also effect the migration of radiocaesium, were different in the Ukraine and in Bavaria.

The proposed collaboration between the GSF and the partners from the Ukraine shall contribute to the understanding of the observed difference of the migration of radiocaesium in soil in the Ukraine and in Bavaria and to study the resulting external exposures. The field work in 1993 includes the selection of various sites in the Ukraine and in Germany, in situ gamma-spectrometry and gamma dose rates measurements at these sites. At each site soil samples will be also taken with a core sampler and Cs-137 determined to obtain the depth profile. As far as not covered by the other groups in ECP-5, investigations on the speciation and fixation dynamics of radiocaesium will be carried out in the laboratory using soil samples from Germany and Ukraine. An intensive collaboration with the Ukrainian partners will be required.

International collaboration on the evaluation of the consequences of the accident at the Chernobyl Nuclear Power Plant

RISK MANAGEMENT

Risk Management

Contract COSU-CT920020 Real-time on-line decision support systems for off-site emergency management following a nuclear accident.

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Description of research work

1. Summary of Project and Objectives

This project is complementary to the development of RODOS (real-time on-line decision support system for off-site emergency management following a nuclear accident), which is being carried out within the framework of the CEC Radiation Protection Programme. The overall objectives are to integrate experience gained and techniques developed in the EC and CIS to further improve decision support systems for off-site emergency management and to develop systems that can be applied generally. In particular, the completion and evaluation of spatial and temporal data and information collected after the Chernobyl accident about radiological quantities, the implementation of early and late countermeasures and their effectiveness, the behaviour of the population, the resources required and the economic impact, would provide important input for validation studies with existing models and their improvement. A joint collaboration in these areas would enable the experience of all partners to be shared and will make optimal use of manpower and resources.

Complementary scientific cooperation with institutes in the CIS has already been started within the 1991/1992 research programme; it is aiming at

- the integration of the expertise available in the EC and the CIS in order to further develop models and to complete data sets for real-time on-line decision support systems using the prototype version 1 of RODOS as a platform,
- the evaluation of Chernobyl data to both improve and validate methods and models for decision support systems, and
- the implementation of RODOS in institutes of the CIS and its on-line operation under the real conditions of existing national meteorological and radiological monitoring systems.

Within the 1992/1993 project period it is planned to intensify and to extend the common research activities in order to achieve an effective scientific cooperation highly beneficial for all partners.

2. Research plans

Within the 1991/1992 research programme, four topics have been identified, on which common research work has already been started and will be continued in close cooperation mainly between the institutes indicated:

(1)Data assimilation and source term estimation

SCK/CEN Mol; Russian Research Centre "Kurchatov Institute", Moscow, SPA TYPHOON, Obninsk

(2)Protective measures - food-bans
NRPB; Ukrainian Institute of Agricultural Radiology, Kiev

(3)Radioactive contamination of water systems
KEMA, KfK; Institute of Cybernetics, Kiev, SPA TYPHOON, Obninsk

(4)Further development of decision support systems
KfK; SPA TYPHOON Obninsk, Institute of Cybernetics, Kiev.

Other important issues, such as the development of models for simulating the movement of people (evacuation , relocation) and decontamination will be considered in the contract period when either still confidential information will become available or results from the experimental collaboration projects (ECPs) emerge. Then, also other Russian, Ukrainian or Belorussian institutes might become involved.

3. Contributions from Participants

Detailed descriptions of the programme of work for each of the research areas in the project period 1992/1993 are attached. The principle working areas of each EC organisation and the responsible scientists are as follows:

SCK/CEN Mol

Acquisition and construction of a data base with early radiological measurements in the near field after the Chernobyl accident. Use of the data base for testing and improving data assimilation techniques, source term reconstruction methods and monitoring strategies.

NRPB

Improvement of agricultural countermeasures models by using the experience gained in CIS institutes and by evaluating radiological and countermeasure data bases already existing or still under construction.

KEMA

Development of model chains for short- and long-term prognoses of the effects on the aquatic part of the environment after an accidental release of radionuclides The catchment basin of the river Rhine will be used as a framework for application. A close cooperation with KfK will be established.

KfK

Improvement and extension of models for describing the behaviour in water systems, including dose assessments and countermeasure simulation (together with KEMA and NRPB). Incorporation of software into RODOS. Sharing of experience gained from the on-line operation of RODOS in institutes of the concerned CIS Republics. Evaluation of information on evacuation/relocation for improving and validating countermeasure simulation models.

4. Project management

At present, KfK is acting as coordinator for JSP 1 and for the proposal in hand. In parallel, KfK is coordinating parts of the RODOS project within the Radiation Protection Research Programme of CEC. Therefore, an effective communication and cooperation between all institutes involved in the development of RODOS will be achieved.

5. Data assimilation and source term estimation

Within the RODOS project several methods are under development to reduce the initial uncertainties about the radiological consequences in the environment following an accidental release. These methods rely on data assimilation techniques basically comparing early near-field (< 30 km) monitoring data with model predictions.

Suitable methods will allow to provide with an increasing number of monitoring data a more and more realistic and consistent picture of the environmental contamination together with an improved insight into the main accident parameters, such as source term, dispersion and deposition. Post-Chernobyl data from the surroundings of the site in the early phase of the accident will be used to test and improve data assimilation methods presently under development.

In order to achieve this goal, research work will concentrate on three main topics:

1. Preparation of a 'combined' data base system, in which the data bases from several CIS institutes will be linked. Given the project aims described above, data on the radionuclide inventory of the reactor core, all kinds of radiological measurements and information in dependence of space and time, and meteorological data from the early phase of the accident have to be included.

2. Development of means to support data exchange between the cooperating institutes, in particular between the CIS and CEC partners, such as E-mail or direct on-line access. Communication links should also be established for other JSP1 institutes (NRPB, KfK) for future validation studies with simulation models for agricultural countermeasures and relocation/evacuation, respectively.

3. Analysis and use of the combined data base for developing and testing of techniques for source term reconstruction and data assimilation methods.

Given the experience gained after the Chernobyl accident, information in the combined data base can also help to develop better monitoring strategies, especially in the near field during the early phase.

6. Protective measures - food-bans

The overall objective of this part of the JSP1 project in 1992/1993 is to obtain relevant information from the CIS, largely based on their experience with agricultural countermeasures following the Chernobyl accident, with the aim of generally improving the countermeasure models under development for RODOS and increasing their

applicability to Eastern and Western Europe. This information should include the following: the efficiency of the countermeasure in terms of activity and dose reduction; the economic consequences of countermeasures; resource requirements, including manpower and time required; doses to workers and behaviour of the affected population. Information and data obtained from CIS institutes would be incorporated into the countermeasures database of RODOS. Collaboration between CIS institutes and NRPB is already ongoing within the current JSP1 project. Emphasis has been placed so far on countermeasures relating to the banning of milk and milk products. Discussion with the CIS institutes on the nature and availability of data on agricultural countermeasures should be continued and extended to cover other potentially important agricultural foodstuffs.

During the contract period, research work will concentrate on the following topics:

1. Collection of relevant information for further improving agricultural countermeasure modelling; emphasis on milk and milk products.
2. Comparison of received information with other available data from non-CIS experience.
3. Generation of a database for milk and milk products and implementation in RODOS.
4. Collection of information on vegetable countermeasures and comparison with other data.

A close cooperation will be established to the partner institutes involved in ECP2, where data and experience on the effectiveness of food countermeasures exist.

7. Radioactive contamination of water systems

The aim of this JSP1 task is to develop a chain of models for assessing the doses to individuals and to the public resulting from contamination of water bodies after an accidental release of radionuclides. It has to describe all relevant processes such as run-off of radionuclide from watersheds, transport of radionuclides in large river systems including exchange with sediments and the radionuclide behaviour in lakes. Important exposure pathways which have to be taken into account are among others drinking water, groundshine and fish consumption. Once established the model chain will be implemented into the RODOS system.

Within the research programme of JSP1, emphasis will be given to the following topics:

1. Development of mathematical model chains for short-term and long-term predictions of contamination of rivers, lakes and storage reservoirs.

2. Modelling of wash-off and run-off processes in catchment areas as input to river and lake models.

3. Development of models for estimating doses, simulating countermeasures and quantifying their effectiveness and economic costs.

As starting point for a more generalised approach, the cooperating partners ,KEMA, KfK and TYPHOON, Obninsk, Institute of Cybernetics, Kiev, agreed to consider the Rhine river and its tributaries including the IJsselsmeer at first. The hydrological model chain starts with a run-off model, followed by a 1-dim. hydrological model, which provides the input for the lake model. In the overall chain, additional models describing flood events and dispersion near a special site are embedded. The corresponding 1-dim. and 2-dim. models will be provided by the institute of Cybernetics. TYPHOON will make available its run-off and flood models together with post-Chernobyl measurement data for model calibration and validation. Lake models are in use and under further development at KEMA and the Institute of Cybernetics. A biotic model for fish will also be provided by KEMA. KfK will collect hydrological data necessary for the models and post-Chernobyl measurements of radionuclide concentrations in water and on ground surface from the Rhine region for calibrating and testing of the various models. In addition, KfK will contribute work on models for assessing doses, countermeasures and economic costs.

A strong link to other projects will be established, in particular to ECP3 and the Radiation Protection Research Programme of DGXII.

8. Further development of decision support systems

Within the CEC Radiation Protection Research Programme, KfK is developing the hardware and software framework of RODOS and is integrating software products provided by the contractors. A first prototype version of RODOS is available, which contains models for near-range atmospheric dispersion, early emergency actions, dose calculations, food-bans, health effects and economic costs assessments. It is installed on workstations with the software framework as a transportable package to run with a standard based UNIX operating system and X-windows user interface.

TYPHOON is operating and further developing its Radio-ecological Analysis Support System (RECASS). It consists of a nested chain of atmospheric dispersion models (STOCH, EXPRESS, REGION, MESO, LOCAL) coupled to the Global Meteorological Network of the Northern Hemisphere, and some first models for calculating doses. It is installed on mainframe computers and PCs linked via Local Area Network.

A close cooperation has been installed on the development of hardware and software for decision support systems applicable in West, Central and East Europe. Within this collaboration, experience with existing models, the operation of RECASS and RODOS, the presentation of results and their integration in radiological and meteorological networks will be exchanged, and their further development will be commonly supported. In particular, atmospheric dispersion models, simulation models for countermeasures, dose models, models for assessing health effects (and economic costs) and the corresponding data sets will be improved and completed.

Within the present contract period,

1. the first prototype version of RODOS will be installed in the cooperating CIS institutes, i.e. SPA TYPHOON, Obninsk, and the Institute of Cybernetics, Kiev,
2. RODOS will be coupled to the national hydrometeorological and radiological information systems, and adapted to the national requirements and conditions by modifying/replacing specific models or data, and

3. geographical data bases and graphical systems for presenting results relevant for decision-makers will be further developed.
4. Development of simulation models for early emergency actions, such as sheltering and evacuation, and the corresponding dose modules.

The close cooperation with JSP2 will be continued during the 1992/1993 research programme.

Risk Management

Contract COSU-CT920021 Development and application of techniques to stablish intervention levels for use in nuclear accidents.

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Description of research work

Project Plan for 1993

The project is divided into 5 tasks :

A. Historical portrayal of countermeasures.
B. Investigation of social and psychological factors.
C. Conceptual framework for intervention.
D. Distribution of exposures in affected populations.
E. Decision-aiding system for establishing intervention levels.

TASK A: HISTORICAL PORTRAYAL OF COUNTERMEASURES

1. CONTRIBUTION OF CEPN (FRANCE)

1.1. General scope for 1992-1993 project

The second year of the project will be devoted to the completion of the historical portrayal for specific topics which have been identified as important during the course of the first year project. A particular attention will be given to the impact of previous experience with nuclear accidents, the iodine prophylaxy countermeasure and the impact of the IAEA project on the course of the post-accidental management. Specific and new developments will be focused on the most recent period (1989-1992) with the objective of better understanding the impacts of the social and political events as well as the various regulations adopted by the Republics. This regulatory dimension, combined with the results from the "social and the psychological factors" project (Task B) should allow to draw later a more complete picture about forces driving the acceptability of post-accidental countermeasures.

1.2. Detailed presentation of the project

In the assessment of effectiveness of the post Chernobyl accident management, the influence of non radiological factors was not performed in adequate scope and in details. The political, social and economic and regulatory factors existing in the country in the pre-accidental and post-accidental periods had considerably, and in some cases decisively, influenced the effectiveness, credibility and efficiency of the specific post-accidental countermeasures. The aim of the project is to produce a detailed analysis of these factors in order to make correct conclusions on justification, effectiveness and real cost of post-accidental countermeasures. Consequently, in this perspective further studies seem to be necessary in the following fields :

1.2.1. Completion of the historical portrayal during the first years after the accident

The work performed during the first year project allowed to draw a clearer picture of the main forces having driven the implementation of countermeasures and their acceptability by the population. However, some specific topics emerged in the course of the project to be further investigated :

- the importance of the past-experience related to nuclear accidents in the implementation of early countermeasures;
- the relatively poor information collected so far on the iodine prophylaxy countermeasures;
- the impact of the International Chernobyl Project conducted by IAEA with regard to the debate about the 35 rem concept and the relocation strategies.

1.2.2. Detailed analysis of the main political events of 1989-1992 having influenced the post Chernobyl situation

The fact that Chernobyl accident occurred in a period of considerable changes in political structure of the country as well as many social and political events put considerable influence on the post-accidental countermeasures and on their effectiveness. The influence of the following factors need to be further analyzed :

- glasnost and press liberalization;
- USSR Supreme Soviet elections;
- local elections in the affected regions;
- USSR disintegration and emergence of Community of Independent States;
- use of Chernobyl situation for political and other purposes by political parties and social groups.

The transition from the political and administrative centralization and control in all aspects of country's life to Perestroïka and Glasnost have created the conditions for an active and critical role of the public towards the decision making process. In this perspective, a detailed analysis of the factors having influenced public opinion and its forming by mass media need to be investigated. This analysis will be essentially focused on the political position about the nuclear secret, the lack of information due to the political censorship, the difference of scientific opinion about medical and biological effects of radiation amplified by the mass media and the insufficient information of the public on real situation in the affected areas, on real level of danger and on basis of countermeasures and on their objectives and expected results.

1.2.3. Detailed analysis of the impact of regulations on population behaviours and acceptability of the post-accidental situation.

Compensation systems implemented by regulatory decisions appear to have large negative impacts. Instead of pushing towards improvement of situations, compensations quite often lead to passive attitudes where people are trying to use contamination as a source of income. Globally the discussions pointed out the importance of the regulatory system imposed to the population with regards to the acceptability of the situations. This point was largely neglected so far and there is a need to start specific investigations on the issue. Furthermore such a perspective should be of prime importance for designing future regulations at the European level.

TASK B: INVESTIGATION OF SOCIAL AND PSYCHOLOGICAL FACTORS

1. CONTRIBUTION OF THE CENTER FOR RISK RESEARCH (SWEDEN)

The general objective of the project is the study of CIS people's reactions, attitudes and risk perceptions as related to ionizing radiation and the Chernobyl accident. The study planned for 1993 will be conducted within a framework of the investigation of social and psychological effects of the Chernobyl accident, and aim at a better representativeness of results, as compared to the successfully completed pilot study in Novozybkov in 1992.

The pilot study was completed according to plans and important results and experiences have been documented on the basis of this work. A questionnaire instrument has been tried out, and evaluated as appropriate and effective for data collection within CIS. Good working relations have been developed with Russian scientists and at this stage, when our work already has gained some attention, it is also possible to develop a wider network of contacts for collaboration and research exchange within the CIS. On the basis of the pilot study it is now possible to proceed with more thorough studies involving more subjects, and subjects from different types of areas with respect to contamination. The project in 1993 will use a refined questionnaire in the data collection, focus on comparisons of experiences and perceptions of subject living in different zones of radioactive contamination, including respondents who have stayed in contaminated areas and who have been relocated from one area to another. Measures and comparisons of the psychological effects of financial compensation due to radioactive contamination, to control for impact of distress and hardship in normal, everyday life will be included.

At this point in time very good working relations and contacts have been established which Russian scientists, in Moscow, Novozybkov and St Petersburg. A promising contact has been also established with researchers in the Ukraine, and contacts with researchers in Belarus are under development. The plan for the next year includes at least two of the CIS states in the project, which will make possible a comparison also of people's reactions to differently applied countermeasures, as utilized in the respective states. A minimum of 500 persons will be involved as respondents in each chosen area in the study. The method of sampling respondents within preselected focus groups will be employed to facilitate generalization of results.

As for methodology, the use of a questionnaire administered to a large number of respondents will be adopted the next year. This approach can without any difficulty be complemented with qualitative studies regarding results which need further exploration and a deepened analysis. The quantitative approach appears to be the most efficient mean, in time and money, to gain a sufficiently good orientation of degree and distribution of public reactions and risk perceptions, with respect to large groups of respondents, and as a necessity for meaningful statistical comparisons of groups.

It is also planned to initiate a series of research exchanges, to enhance the developing mutual understanding of theoretical backgrounds and used methodologies of those involved in the project, starting in connection with the organization of next year's work. Two kinds of exchanges are suggested ; theoretical seminars related to the planned and ongoing work, for example, seminars on social psychological research, risk perception, and methodological issues. These seminars would especially involve researchers already

contributing to the project. The second kind of seminars should focus on research procedures and involve occasional visits to Sweden and the Center for Risk Research, whereas the latter kind of seminars will beheld in the CIS, in close relation to the data collection.

2. CONTRIBUTION OF THE ROBENS INSTITUTE (UNITED KINGDOM)

The pilot phase of the JSP-2 TASK B included a field study in July 1992. The empirical data collected include background information on individuals, reported symptoms and a scaled stress measure, and risk perception measures. Preliminary analysis of these data have been made available by the end of the first year. The first task for the second year will be to conduct a full and detailed analysis of these data and to refine the measuring instruments in the light of what is learned.

The pilot data are restricted to a small area of the Russian Republic. In the second year of JSP-2 it would be necessary to expand the area of study to include people from the Belarus Republic and from the Ukraine. To this end further contacts with possible collaborators from these Republics would be sought and, in collaboration with the JSP-2 partners, further field work in all three Republics would be conducted. Such field work could be conducted. Such field work could be conducted in the first half of 1993.

As well as increases in the scale of the empirical work the study would be applied to further categories of respondent ; namely relocated populations and control populations resident some distance from the contaminated areas. In general the measures should fall into an experimental design having the aim of isolating, as far as possible, the impact of the accident and other contaminating factors.

3. CONTRIBUTION OF MUTADIS CONSULTANTS (FRANCE)

General objectives

The purpose of the project is to carry out a qualitative evaluation of the psychic and social damages derived from the Chernobyl accident and the countermeasures taken after the accident. This study can only be undertaken indirectly, after taking into account and interpreting the discourse held by the populations concerned. The actual psychological and social stakes can not be detected directly in the manifest discourse of the people interviewed, who are, whether consciously or unconsciously subject to individual or collective resistances. These resistances create an impenetrableness of the discourse regarding these stakes with the existence of unspoken comments which the analysis and interpretation (semiological, psychological, communicational, sociological) will allow to detect. This work will also take into account the distortions induced by the political and media discourses during or after the accident (particularly with the support of the study implemented within the framework of JSP-2 concerning the media discourse).

The inventory of the harm in the psychological and social fabric will be carried out at different levels. Through the analysis of the representations of the accident and of the countermeasures, this research will be focused on the detection of the reactional formations, the defences individually and collectively worked out to overcome the intensity of the traumatisms. In general, the study will attempt to underscore the possible,

unknown or unheard, paradoxical or counter-productive effects of the accidental and post-accidental measures, generated gradually through the usual mechanisms. It will also take into account the effects on the social bond particularly among the relocated populations.

Methodology

The investigation of the psychological and social dimensions requires a direct contact with the populations concerned (those living in the areas subject to countermeasures, those who have been relocated and those living out of these areas but in the immediate neighbourhood). It is intended to limit the study, at first, to a territory offering a certain linguistic and cultural homogeneity (Ukraine). This choice will allow to carry out a comparative work between data coming from the different areas set up within the framework of the countermeasures (including from out of these areas).

i) preliminary work on investigation

- inventory of the existing documentary data,
- setting-up of an interview guide,
- sampling (geographical, sociodemographic).

ii) investigation on the spot (tape-recording)

- carrying out of semi-directed interviews of the populations according to the sample,
- interviews of social notabilities involved (representatives, doctors, security staff, etc...),
- informal interviews in the institutional environment (hospital, maternity hospital, medico-social centre),
- meetings in urban and rural environment.

After their collection, the data gathered will be transcribed, then translated and will be subjected to analysis by an interdisciplinary group of academics and consultants (semiology, communication, psychology, psychoanalysis, sociology).

The analysis will be carried out according to the classical methods which will be completed by the following complementary methods :

- discovering of the word complexes and of the repetitive semantic series
- discovering of the frequent connections and of the constellations of significants.

TASK C: CONCEPTUAL FRAMEWORK FOR INTERVENTION

1. CONTRIBUTION OF RISØ (DENMARK)

Objective

The basic principles for intervention and intervention level setting will be analyzed and dose limits, doses to be compared with the intervention levels and realism in the dose assessment, will be thoroughly addressed. The analysis will elaborate on the *averted doses* by different countermeasures. The averted doses will be expressed by :

- dose rate,
- dose in a given year,
- lifetime dose,
- residual dose,
- surface contamination density,
- activity concentration in foodstuffs.

The results of the analysis will be expanded in a parameterized form including environmental and nuclide parameters. The relation between the intervention level of dose and operational intervention levels, e.g. dose rate or nuclide concentration, will also be expanded.

Application to foodstuff countermeasures and relocation

Normally, the doses from contaminated foodstuffs will not enter the deterministic region. Therefore the averted doses from the foodstuff pathway can be expressed as *averted collective dose* per unit mass of a given category of foodstuff. Intervention levels will be derived from the justification/optimisation principles and Soviet data bases. The data bases should include the following data for selected villages :

- distribution of ^{137}Cs-concentration in different foodstuffs, $m(C)$,
- efficiency of specified agro-technical countermeasures,
- realistic cost estimates of the countermeasures,
- realistic estimates of food cost.

Factors influencing the development and implementation of intervention levels for relocation will be averted individual doses, costs of relocation and length of relocation time. For selected Soviet villages the doses averted by relocation will be derived, provided that the following data will be available :

- external dose rate as a function of time,
- shielding factors for buildings,
- realistic cost estimates of relocation,
- contamination density of specific radionuclides,
- resuspension factors for the environment.

A reduction of an intervention level for a given protective measure (e.g. relocation, foodstuff restrictions) will increase the averted doses (increase benefit) but also increase the number of people involved and consequently the costs and social detriment from the countermeasure (increase costs). Correspondingly, an increase of the intervention level will decrease the averted doses (decrease benefit) and decrease the number of people involved (decrease costs).

The sensitivity of changing the intervention level expressed as a change in *averted doses* and *costs* can give the decision maker a picture of the costs of making a situation "more safe".

Based on Soviet data bases for distributions of contamination density, concentration in foodstuffs and external dose rate a sensitivity study of *differential/intervention level-cost/averted dose* will be made.

Risk perspective

One of the recommendations of the International Chernobyl Project was that "more realistic and comprehensive information should be provided to the Soviet public on the levels of dose and risk consequent upon their remaining in the contaminated areas concerned. These risks should be compared with risks experienced in everyday life and with risks from other environmental contaminants, e.g. radon and industrial emissions".

To support this recommendation the risks from the intervention levels derived in this project will be put into perspective in different, easily understandable ways.

TASK D: DISTRIBUTION OF EXPOSURES IN AFFECTED POPULATIONS

1. CONTRIBUTION OF THE NRPB (UNITED KINGDOM)

The primary aim of this proposal is to investigate the importance of considering the full distribution of doses to communities when reaching decisions on protective actions. This work will involve examination of the distribution of individual doses in communities contaminated by the Chernobyl accident, in collaboration with Institutes in the Republics of the CIS.

Three aspects of the work are identified:

- An assessment will be undertaken of measurements already made, and the doses estimated from them, to determine the extent to which these reflect the full distribution of doses received. The equipment used and exposure pathways taken into account will also be examined. The usefulness and scope for developing an additional programme of measurements for improving the determination of the full distribution of doses will also be explored.

- Experimental studies on 'hot particles' will also be carried out. The aim will be to predict the behaviour of the radionuclides in humans and to assess the appropriate doses per unit intake from inadvertent intakes of these

particles. These studies will be of value for improving the interpretation of chest monitoring and bioassay data, and for calculating the potential risks from hot particles to individuals living or working in the area. For 1992/93 consideration will be limited to the inhalation of hot particles an to hot particles obtained from a given site. In the longer term the work would be expanded to consider gastrointestinal absorption of the radionuclides after ingestion, their biokinetics after deposition in a simulated wound site, and to investigate the inter-site variability in biokinetic behaviour of the radionuclides.

. Using these studies, the importance of critical group doses in making decisions on protective actions will be explored. Issues examines will include :

- the variation between critical group and average doses,
- the size of critical groups,
- the potential for protecting critical groups directly using selective protective measures,
- the influence of probability of exposure to higher than average doses on decision making.

TASK E: Decision aiding system for establishing intervention levels

1. CONTRIBUTION OF DEMOKRITOS (GREECE)

During the first year of the JSP-2 project, a Decision Support System (DSS) for establishing emergency, intermediate and long term protective actions, with emphasis on the establishment of protective policy prior to the event of an accident has been developed. During the second year, a full-scope system will be designed. The final DSS will consist of procedural and methodological steps and corresponding software code which can be distinguished in five distinct modules.

a) Generation of Alternatives:

This module refers to the general format and the generation of alternative courses of action. Initially it has been determined that an alternative protection action will be modelled as consisting of number of discrete components each of which can take a number of values (discrete). This will result in a discretization of the space and time space with the ability to define a specific protective action for each element.

b) Generation of Attributes:

This module refers to the generation of an hierarchy of objectives and associated measures of effectiveness to measure the degree of satisfaction of each objective in the louder level of the hierarchy (attributes). The final set of attributes will cover health, environmental economic and social psychological consequences relevant to the implementation of specific countermeasures, and will be open to additions and/or substractions by the final user of the DSS.

c) Consequence Assessment:

This module will enable the evaluation of each alternative in all selected attributes, that is it will measure the impacts of each alternative in each and every of the attributes established in the second module. This module will depend on the models already developed.

d) Definition of Efficient set of Alternatives:

This module will select out of the set of all possible alternatives (the decision space) those that are efficient by rejecting all alternatives that are dominated by the efficient ones. An alternative is dominated by another if the latter is better than the former in all attributes. A dynamic programming approach will be followed to generated this efficient set of alternatives. This selection does not require any preference assessment and value-trade off among different attributes.

e) Preference Assessment Technique:

This module will consist of a computerized implementation of a multiattribute decision analysis technique generating though a Graphical User Interface (GUI) the utility function of a decision maker and assisting in this way the selection of the most preferred alternative out of those belonging to the efficient set.

International collaboration on the evaluation of the consequences of the accident at the Chernobyl Nuclear Power Plant

HEALTH CONSEQUENCES OF THE CHERNOBYL ACCIDENT

Health consequences of the Chernobyl accident.

Contract COSU-CT920022 Biological Dosimetry Including Cytogenetics.

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Total Contribution by the Commission: 223.000 ECU
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Description of research work

Introduction

The project has the aim of fostering collaboration between researchers in biological dosimetry in EC countries with their counterparts in the three CIS countries principally affected by the Chernobyl accident, namely Russia, Belarus and Ukraine. In the absence of reliable personal physical dosimetry, biological dosimetry is identified as an approach likely to assist in quantifying exposure of irradiated persons.

The project contains several components centred around the method of analysis for chromosomal damage in lymphocytes using the relatively new method of fluorescence in situ hybridisation (FISH) which permits selected chromosomes to be highlighted or "painted". This is particularly effective for detecting stable translocations. Scoring cells with FISH is much more rapid than the alternative method of G-band karyotyping and thus it is feasible to apply the method to blood samples from many persons.

The traditional method of biological dosimetry, as practised since the mid 1960s, is to examine peripheral blood lymphocytes for dicentric chromosomal aberrations. This approach works best soon (less than six months) after acute exposure to an external source. For internally incorporated radionuclides the interpretation of lymphocyte aberrations in terms of dose is often difficult because of the localisation of most nuclides to certain organs and tissues. Persons exposed to fall-out from the Chernobyl accident have received a complex exposure from both external and internal sources, the latter mainly via food. Much of the dose from incorporated radionuclides is due to isotopes of caesium. Fortunately for biological dosimetry purposes, these produce a more or less uniform exposure to the body as they accumulate in muscle.

Six years have elapsed since the accident. Much of the dose was received early on although some is still being delivered because of the environmental contamination. As time passes, the dicentric assay becomes quantitatively less reliable, mainly because of the turnover of lymphocytes which are said to have a half-life of about three years. This is, however, very variable between subjects. New experimental work initiated by the contract cannot rely solely on the dicentric assay but will be focussed on chromosomal endpoints that persist, ie. "stable aberrations". These are mutational changes that can pass successfully through cell divisions and thus should persist in the body on a timescale of many years.

There is a good infra-structure in CIS countries for biological dosimetry based on dicentric analysis. There is a data bank of results from several thousand persons comprising those heavily irradiated at the accident, liquidators and surveys of exposed communities, particularly of children. The more highly exposed subjects have been periodically sampled and this is ongoing.

Within the EC states there is also a long tradition of biological dosimetry by cytogenetics. A number of laboratories have now expanded on the conventional method with dicentrics to include the FISH method for stable translocations. The objectives in using this are to permit dosimetry in situations, like investigating old exposures, where the dicentric analysis is less accurate. In addition, the method is used

in research to investigate numerical relationships between unstable and stable forms of damage and to examine stable chromosomal lesions linked to neoplasia.

Details of the Contract

The first priority is to get technicians from the CIS participants trained in the FISH technique and equipped so that they are able to perform the analyses in their own laboratories. It should be noted that, prior to the EC contract, training had already commenced through an on-going bilateral agreement between Germany (Dr. Bauchinger's lab) and the CIS countries. Training within the EC project will complement this. Training will then lead on to a collaboration research project which will consist of two parts.

1. An in vitro study will be undertaken with specimens prepared in one laboratory and replicate slides distributed to the other CIS laboratories. Blood will be irradiated to a range of doses and scored conventionally for dicentrics and FISH translocations. In addition, G-band translocations will also be scored in one laboratory that has semi-automated karyotyping. This will a) give the participants some experience; b) produce dose response data; c) enable an intercomparison of laboratories performances; and d) produce important data on the relative numbers of lesions that can be identified, in their hands, by the different methods.

2. An in vivo study will involve approximately ten persons who had acute radiation syndrome in 1986 (but not treated in a way that would confound cytogenetics), and a similar number of persons currently working in the sarcophagus who have received exposures of several Gy protracted over a few years. Lymphocytes from all subjects will be sampled for dicentrics, FISH and G-band translocations. These subjects have been sampled for dicentrics regularly since their exposure and this is continuing. With the new data on the ratios of stable:unstable aberrations generated in the project, some comparisons with the earlier dicentric frequency will be made. The subjects will be followed over the next few years to plot temporal changes in aberration frequencies particularly since there is a need to establish how reproducible are the frequencies of the stable aberrations.

When the FISH technique is established in participating laboratories and experimental work described above is underway, it will be possible to commence dosimetry on cohorts registered in the epidemiology project - ECP7. This will, of necessity, take considerable time, although it should be remembered that the epidemiology will itself be a long-term study as cancers are awaited. Therefore, within the biological dosimetry project, frozen cell banks will be established to store material from the subjects until the laboratories are able to perform the analyses. This will circumvent problems of later availability of the subjects for sampling and avoid the need to work with cells that may be taken much later when neoplastic diseases could have developed and treatment is already in progress. Moreover, the stability of the aberrant cells, eg. their clonal expansion, is an unknown factor and samples should be taken and stored sooner rather than later. A frozen cell/DNA bank would also be an invaluable source of material for any new biological dosemeter that may be developed, and for fundamental research studies on DNA lesions related to cancer.

Participating laboratories in CIS countries

Russia — **Medical Radiology Research Centre** - Obninsk

Central Research Inst. of Roentgenology and Radiology - St. Petersburg

Scientific Centre for Haematology - Moscow

Belarus — **Institute of Genetics and Cytology** - Minsk

Institute of Radiation Medicine - Minsk

Ukraine — **Research Centre for Radiation Medicine** - Kiev

Research Institute of Medical Radiology - Kharkov

Persons from some of these laboratories have already been trained in FISH at GSF Neuherberg, Germany. Others still need training. All participants will collaborate to the experimental programme by scoring conventional dicentrics and FISH translocations. In addition a Cytoscan instrument at Obninsk will be used for banded translocations. All laboratories already have a data base of subjects examined by conventional cytogenetic dosimetry. These data will be available to the project and, as required, suitable subjects will be drawn from the data bases for the FISH study.

Participating laboratories in EC countries

National Radiological Protection Board - Chilton, UK

University of Leiden - Leiden, The Netherlands

National Scientific Research Centre, Demokritos - Athens, Greece

University of Tuscia - Viterbo, Italy

GSF - Forschungszentrum - Neuherberg, Germany

The EC participating laboratories will receive visiting scientists from CIS laboratories for training. In addition technicians and/or scientists from the EC labs will make visits to CIS laboratories for back-up technical assistance. As necessary, aliquots of blood specimens processed in the CIS laboratories for the experimental programme will also be sent to EC laboratories for parallel processing.

Health consequences of the Chernobyl accident.

Contract COSU-CT920023 Epidemiological Investigations Including Dose Assessment and Dose Reconstruction.

Coordinator Danish Cancer Society
Dep. Denish Cancer Registry
Rosenvaengets Hovedvej 35
DK-2100 COPENHAGEN
Tel. 45-35268866

Total Contribution by the Commission: 200.000 ECU
7 months 1/12/92 to 30/06/93

Participating Scientists

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NOT YET FINALIZED

Health consequences of the Chernobyl accident.

Contract COSU-CT920024 Patient Treatment

Coordinator Univ. Rotterdam - Erasmus
Department Radiobiology
Postbus 1738
NL-3000 DR ROTTERDAM
Tel. 31-104087766

Total Contribution by the Commission: 227.000 ECU
7 months 1/12/92 to 30/06/93

Participating Scientists

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Description of research work

Objectives of the Joint Study Project on Patient Treatment

The project aims primarily at a joint study on improvement of diagnosis and treatment of the effects of high dose accidental irradiation and is in particular directed at:

(i) evaluation of patients treated for acute radiation sickness after the Chernobyl accident;

(ii) the hemopoietic system, which belongs to the most radiation sensitive systemic organs, as well as

(iii) the skin, which is frequently affected since acute localized irradiation and external contamination are the most frequent radiation accident events. An existing group of collaborating researchers in CEC countries will collaborate with their colleagues in the three CIS countries principally affected by the Chernobyl accident, i.e., Russia, Byelorus and Ukraine. It is recognized that several of the CEC laboratories/institutes participating in this project have already bilateral ties with laboratories in the CIS.

Scientific background and proposed studies

(i) Patient evaluation

The Chernobyl accident offers substantial study material on high dose accident victims in the range of 1 to > 5 Gy exposures. Of 237 patients suffering from acute radiation sickness, 209 survived and were/are in the process of rehabilitation. It is proposed to use this wealth of patient material for an in depth investigation on the treatment of acute as well as late radiation damage, involving not only the immuno-hemopoietic system and the skin, but also the gastrointestinal tract, the kidney, the liver, the central nervous system and endocrine functions. A detailed protocol of investigation will be made in the initial phase of the project following a review of the patient material, that, in addition to existing data, might be suitable to establish dose-effect relationships for various types of organ damage. It is imperative that dosimetry be supplemented by up-to-date methodology and reanalysis.

(ii) Immuno-hematopoietic system

In the initial phase after accidental exposure, it is essential to distinguish as quickly as possible patients with sufficient residual hemopoietic capacity to survive with conventional supportive care and those which require intensive, sophisticated treatment in specialized centers, which may include treatment with hemopoietic growth factors, electively supported by (allogeneic) bone marrow transplantation.

a. **Improvement of diagnosis.**

Due to inhomogeneity of exposure in radiation accidents, physical dosimetry in general is insufficient to assess damage to cell renewal systems including hemopoiesis. Early after exposure, the changes in the leukocyte counts and differential counts may be of some assistance, but in addition chromosomal analysis is considered essential. Experimentally, the damage to the hemopoietic system should be assessed by measuring the hemopoietic response to defined doses of hemopoietic growth factors. Much experimental work is required in preclinical animal models to relate growth factor response magnitude to radiation dose and residual hemopoietic stem cell numbers. Although close to twenty growth factors are currently known and can be made available, very little is known about their use for this purpose, let alone the use of a combination of different growth factors.

b. **Prevention of clinical manifestation of radiation damage / improvement of therapy.**

Until recently, high dose accidental exposure was preferentially treated with allogeneic bone marrow transplantation, because in general, autologous bone marrow, even of persons at risk, has not been available. In principle, it is possible to cryopreserve autologous bone marrow of individuals at risk, but this has not been pursued for complex reasons. The results of allogeneic bone marrow transplantation in high dose radiations victims have been disappointing, which may have been predicted on the basis of insufficient knowledge of the actual dose of (inhomogeneous) radiation received. There are several ways to improve the results of bone marrow transplantation, which include the further development of suitable, non-toxic immunosuppressive conditioning regimens, transient allogeneic engraftment using purified stem cells and acceleration of allogeneic reconstitution by growth factor treatment, as well as rapid tissue typing to identify unrelated donors matched for major histocompatibility antigens.

Hemopoietic growth factor treatment alone may be advantageous in those patients that show an early response. Also here, it is unknown which combination of growth factors will be suitable for this purpose and much experimental work and clinical trials need to be done to identify the most optimal hemopoietic growth factor treatment regimen.

(iii) Radiation effects and their treatment after local exposure of skin and subcutaneous tissues.

The severity of radiation damage to the skin and subcutaneous tissues depends on the energy deposited in the tissues, and the dose has to be determined as accurately as possible in order to evaluate the prognosis and to manage the treatment. Experimental studies have been performed to specify the target cells involved in the pathological processes leading either to early or to late effects. Dose-effect curves have been established for the various endpoints (erythema, dry of moist desquamation, permanent ulcer or healing) from experiments in animal models and from observation in radiotherapy patients. Acute radiation damage to the skin is primarily a consequence of changes in the epidermis; the timing of the peak reaction is related to the kinetic organization of this layer. Late damage to the skin is primarily a function of radiation effects on the vasculature and on fibroblasts; this produces a wave of dermal atrophy after 16-26 weeks after which dermal necrosis develops at high doses. A second phase of dermal thinning is seen to develop after 52 weeks.

a. Improvement of diagnosis

The management of acute localized irradiation in patients after accidental exposure remains difficult to perform because the diagnosis of the extent of irradiated tissues and therefore the prognosis are doubtful insofar as the early reaction is rather weak and varying. Needs for earlier and more accurate diagnosis methods do exist either for the early inflammatory reaction (hyperemia, hyperthermia, oedema), for the secondary reaction (late erythema, ischemia, necrosis) as well as for the following fibrosis and sclerosis processes. NMR imaging and NMR spectroscopy, blood an lymph flow measurement and gamma-scintigraphy seem the best non-invasive methods to be developed in animals models before to be validated in man, both for diagnostic purposes and quantifying the therapeutic assays.

b. Prevention of clinical manifestation of radiation damage / improvement of therapy

Strategies for treatment will be directed either towards the reduction of inflammation and oedema, or towards the improvement of the vascular supply, trying to avoid the reperfusion injury: haemorrheological agents, anti-inflammatory and platelet anti-aggregant drugs, anti-adhesion molecules and inhibitors of iron-dependent lipid peroxydation will be assayed. Protocols for treatment will be developed by a joint effort of participants with emphasis on optimization of treatment during the acute and late phases in a way to reduce free radical production, and improvement of different methods to cover the combined lesions such as skin grafts, skin flaps and artificial skin used first as transitory dressing. The wound healing problem in irradiated skin is a further aspect of accidental overexposure. Trauma may be associated with accidental irradiation, but surgery may have to be carried out in irradiated areas to repair necrotic lesions. This will be evaluated in pig skin after irradiation with sources of various energy (to observe lesions at different depth) using a standard surgical wound, with or without different decontamination processes. Protocols for surgical management of radiological burns will be tested taking into account 1) the extent of the lesions according to the depth in the irradiated tissues 2) the delay for the intervention and 3) the technic used: skin graft, skin flaps, and combined surgical treatment. Furthermore, recombinant EGF will be tested in clinical situation of radiation burns, while superoxide dismutase (SOD) encapsulated in liposome is efficient in reducing radiation induced skin fibrosis after radiotherapy. Alternative source of SOD will be evaluated with other potentially active treatment such as: glutathione peroxidase (GPX), prostaglandins (PGE1 and prostacyclin) as anti-aggregating factors and cytokines (IFNG), pentoxifyllin, lazaroids, deferoxamine-Mn4 complexes. PAF inhibitors (BN52012) and antisense RNAs against growth factors receptors.

Collaboration

To meet the objectives of the joint study project, collaboration will include:

1/collaborative experimental research / experimental clinical protocols and data exchange;

2/exchange of data on radiation accident victims, their treatment and the outcome of the treatment;

3/a joint data base containing all possible relevant patient material;

4/exchange of trainees / scientists / equipment / reagents, resulting in technology transfer most likely to occur from west to east;

5/regular meetings on progress achieved;

6/development of common recommendations for the immediate health care of radiation accident victims;

7/the development of recommendations to be used eventually for a common network of expertise and assistance.

ANNEXES

ANNEX I: LIST OF RESEARCH CONTRACTS

ANNEX II: LIST OF SCIENTIFIC RESPONSIBLES

ANNEX III: GEOGRAPHICAL DISTRIBUTION OF CONTRACTS

Belgium

Switzerland

Germany

Denmark

Spain

France

Great Britain

ANNEX IV: LIST OF ACRONYMS AND ABBREVIATIONS

ADFAC	Association pour le development des facultés des sciences de l'université de Paris (FR)
ADPA	Association pour le development e la physique atomique (FR)
AECB	Atomic Energy Control Board, Ottawa (CND)
AECL	Atomic Energy of Canada Limited (CND)
AFPPE	Association Française du Personnel Paramédical d'Electrocardiologie, Paris (FR)
AIRM	Associazione Italiana di Radioprotezione Medice (IT)
AIRP	Associazione Italiana di Protezione contro le Radiazioni (IT)
ALARA	As Low As Reasonably Achievable
BFS	Bundesamt für Strahleschutz (DE)
CAATS/INSERM	Centre d'évaluation pour l'Assurance de qualité des Applications Technologiques dans le domaine de la Santé/Institut National de la Santé et de la Recherche Médicale, Cachan (FR)
CEA	Commissariat à l'Energie Atomique (FR)
CEC	Commission of the European Communities, Brussels (BE)
CEDHYS	Centre de Développement des Etudes et Applications en Hygiène et Sécurité, Paris (FR)
CEN/SCK	Centre d'Energie Nucléaire/Studie Centrum voor Kernenergie (BE)
CEPN	Centre d'étude sur l'Evaluation de la Protection dans le domaine Nucléaire, Fontenay-aux-Roses (FR)
CERN	Centre Européen de Recherche Nuclèaire (CH)
CETA	Centro di Ecologia Teorica ed Aplicata (IT)
CIEMAT	Centro de Investigaciones Energéticas, Medioambientales y Tecnológicas, Madrid (ES)
CIR	Centre International de Radiopathologie, Fontenay-aux-Roses (FR)
CNEN	Comitate Nazionale per la Ricerca e per lo Sviluppo dell'Energia Nucleare e delle Egnergie Alternative, Rome (IT)
CNR	Consiglio Nazionale delle Ricerche (IT)
CNRM	Centre National de Recherches Meteorologiques (FR)
CNRS	Centre National de la Recherche Scientifique (FR)
COSYMA	Code System Maria
CRSA	Centro Regionale per la Sperimentazione Agraria per il Friuli-Venezia-Guilia, Udine (IT)
CSTC-WTCB	Centre Scientifique et Technologique de la Construction (BE)
DG	Directorate General of the CEC
DIAS	Dublin Institute for Advanced Studies (IE)
DKFZ	Deutsches Krebsforschungszentrum (DE)
DMI	Danmarks Meteorologiske Institut (DK)
EAR	European Association of Radiology

EBHA	East Birmingham Health Authority (GB)
ECN	Energieonderzoek Centrum Nederland (NL)
EDF	Electricité de France (FR)
EBMT	European Bone Marrow Transplant Group
ECURIE	Early Exchange of Information in the Event of Radiological Emergency
EFOMP	European Federation of Organisations of Medical Physics, York (GB)
ENEA/DISP	Comitato Nazionale per la Ricerca e per lo Sviluppo dell'Energia Nucleare e delle Energie Alternative, Direzione Sicurezza Nucleare e Protezione Sanitaria, Rome (IT)
ENEL	Ente Nazionale per l'Energia Elletrica, Roma (IT)
ERPET	European Radiation Protection Education and Training CEC DG XI/A/1, Luxembourg (L) & DG XII/D/3, Brussels (BE)
EULEP	European Late Effects Project Group (BE)
EURADOS/CENDOS	European Radiation Dosimetry Group/Collection and Evaluation of Neutron Dosimetry Data (DE)
EUROMET	European Metrology
GRS	Gesellschaft für Anlagen und Reaktorsicherheit mbH (DE)
GSF	Gesellschaft für Strahlen- und Umweltforschung, Neuherberg (DE)
GSI	Gesellschaft für Schewerionenforschung mbH (DE)
IAEA	International Atomic Energy Agency (AT)
IARC	International Agency for Research on Cancer, Lyon (FR)
ICRF	Imperial Cancer Research Found (GB)
ICRP	International Commission on Radiological Protection (GB)
ICRU	International Commission on Radiation Units and Measurements (USA)
ICSTM	Imperial College of Techn. and Medicine Science, London (GB)
IFE	Institute of Freshwater Ecology, Ambleside (GB)
IFO	Istituti Fisioterapici Ospitalieri (IT)
IFS	Institut für Soziologie (DE)
IMPCOL	The Imperial College of Science Technology and Medicine (GB)
INFN	Istituto Nazionale di Fisica Nucleare (IT)
INRA	Institute National de la Recherche Agronomique (FR)
INSERM	Institut National de la Santé et de la Recherche Medicale (FR)
INTECHMER-CNAM	Institut National des Techniques de la Mer - Conservatoire National des Arts et Métiers-Cherbourg (FR)
INSTN	Institut National des Scienes et Techniques Nucléaires, Saclay (FR)
IOMP	International Organisation of Medical Physicists
IPSN	Institut de Protection et de Sûreté Nuclèaire (FR)
IRS	Integrated Radiological Services Ltd, Liverpool (GB)
ISH	Institut für Strahlenhygiene/Bundesamt für Strahlenschutz, Salzgitter/Neuherberg (DE)
ITE	Institute of Terrestrial Ecology, Grange-over-Sands (GB)

ITRI	Inhalation Toxicology Research Institute, Albuquerque, NM (USA)
ITRI/TNO	Instituut voor Toegepaste Radiobiologie en Immunologie, TNO Rijswijk (NL)
JRC	Joint Research Center of the CEC at ISPRA (IT)
KBFOC	Kaftens Bekampelse Fibigerinstitutet og (DK)
KEMA	Keuring Electrotechnische Materialen N.V. (NL)
KFA	Forschungsanlage, Jülich (DE)
KfK	Kernforschungszentrum Karlsruhe (DE)
KUL	Katholieke Universiteit Leuven (BE)
LNETI	Laboratorio Nacional de Engenharia e Tecnologia Industrial, Lisboa (PT)
MAFF	Ministry of Agriculture, Food and Fisheries, Lowestoft (GB)
MARIA	Methods for Assessing the Radiological Impact of Accidents
MLURI	McAulay Land Use Research Institute, Edinburgh (GB)
MRC	Medical Reserach Council (GB)
NEB	Nuclear Energy Board of Ireland, Dublin (IE)
NINA	Norsk Institutt for Naturforsknig (NO)
NIR	Niedersächsisches Institut für Radioökologie (DE)
NIRP	National Institute of Radiation Protection, Stockholm (SE)
NCSR	Democritos, National Centre for Scientific Research, Athens (GR)
NERC	National Environmental Reserach Council (GB)
NPL	National Physical Laboratory (GB)
NRPB	National Radiological Protection Board, Chilton (GB)
OECD/NEA	Organisation for Economic Cooperation and Development/Nuclear Energy Agency
ORNL	Oak Ridge National Laboratory, Knoxville, Tennessee (USA)
PTB	Physikalisch-Technische Bundesanstalt, Braunschweig (DE)
RADE-AID	Radiological Accident Decision Aiding System
RBE	Relative Biological Effectiveness
REM	Radioactivity Environmental Monitoring
RERF	Radiation Effects Research Foundation, Hiroshima (JP)
RIVM	Rijks Instituut voor Volksgezondheid en Milieu, Bilthoven (NL)
RPII	Radiological Protection Institute of Ireland (IE)
SCRPI	Service Central pour la Protection contre les Rayonnements Ionisants, Le Vésinet (FR)
SEPR	Sociedad Española de Protección Radiológica (ES)
SFEN	Société Française d'Energie Nucléaire, Paris (FR)
SFRP	Sociedad Española de Proteccion Radiologica (ES)
SRD	Safety and Reliability Directorate, Warrington (GB)
SUAS	Swedish University of Agricultural Sciences, Umea (SE)
TEPC	Tissue-Equivalent Proportional Counter

TNO	Nederlandse Organisatie voor Toegepast Natuurwetenschappelijk Onderzoek (NL)
UIR	Union International des Radioecologistes (BE)
UKAEA	United Kingdom Atomic Energy Authority, Harwell (GB)
ULB	Université Libre de Bruxelles, Brussels (BE)
US DOE	US Department of Energy, Washington DC (USA)
US EPA	US Environmental Protection Agency (USA)
US NCI	US National Cancer Institute, Bethesda (USA)
US NIES	US National Institute of Environmental Sciences (USA)
USL	Unità Sanitaria Locale, N° 7, Udine (IT)
VITO	Vlaamse Instelling voor Technologisch Onderzoek (BE)
WHO	World Health Organisation

European Communities — Commission

EUR 15238 — Nuclear fission safety programme 1990-94
Radiation protection research action 1992-94
Technical description of scientific projects

Luxembourg: Office for Official Publications of the European Communities

1994 — XI, 772 pp. — 16.2 × 22.9 cm

Radiation protection series

ISBN 92-826-6579-8

Price (excluding VAT) in Luxembourg: ECU 76

This book contains summaries of contractual work included in the 1992-94 period for:

Part I

'Radiation protection research action 1992-94', which is divided into three sectors:

A — Human exposure to radiation and radioactivity:
  (1) Measurement of radiation dose and its interpretation;
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B — Consequences of radiation exposure to man; their assessment, prevention and treatment:
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  (2) Non-stochastic effects of radiation;
  (3) Radiation effects on the developing organism;

C Risk and management of radiation exposure:
  (1) Assessment of human exposure and risks;
  (2) Optimatization and management of radiation protection.

Part II

'International collaboration on the evaluation of the consequences of the accident at the Chernobyl nuclear power plant', which is divided into three sectors:

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4 Effecten van straling op langere termijn en toxicologie van opgenomen radionucliden

5 Meting van straling en de interpretatie daar- van dosimetrie

6 Beoordeling van stralingsrisico s

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