

Environmental Science and Technology Department. Annual report 1992

Jensen, A.; Nielsen, Gunnar Gissel; Gundersen, V.; Nielsen, O.J.; Østergård, Hanne; Aarkrog, A.

Publication date:
1993

Document Version
Publisher's PDF, also known as Version of record

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Citation (APA):

Jensen, A., Nielsen, G. G., Gundersen, V., Nielsen, O. J., Østergård, H., & Aarkrog, A. (1993). Environmental Science and Technology Department. Annual report 1992. (Denmark. Forskningscenter Risoe. Risoe-R; No. 680(EN)).

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Risø-R-680(EN)

Environmental Science and Technology Department Annual Report 1992

**Edited by A. Jensen, G. Gissel Nielsen, V. Gundersen,
O.J. Nielsen, H. Østergård and A. Aarkrog**

**Risø National Laboratory, Roskilde, Denmark
March 1993**

Abstract

Through basic and strategic research, the Environmental Science and Technology Department aspires to develop new ideas for industrial and agricultural production thus exerting less stress and strain on the environment.

The department endeavours to develop a competent scientific basis for future production technology and management methods in industrial and agricultural production. The research approach in the department is predominantly experimental.

Selected departmental research activities during 1992 are introduced and reviewed in seven chapters: 1. Introduction, 2. The Atmospheric Environment, 3. Plant Genetics and Resistance Biology, 4. Plant Nutrition and Mineral Cycling, 5. Chemistry of the Geosphere, 6. Ecology and Mineral Cycling, 7. Other Activities. The department's contribution to national and international collaborative research programmes is presented in addition to information about large research and development facilities used and managed by the department. The department's educational and training activities are included in the annual report along with lists of publications, publications in press, lectures and poster presentations at international meetings. The names of the scientific and technical staff members, visiting scientists, Post. doctoral fellows, Ph.D. students and M.Sc. students are also listed.

ISBN 87-550-1891-2

ISSN 0106-2840

ISSN 0906-8090

Risø 1993

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1 Introduction

1.1 The Department of Environmental Science and Technology

Research Objectives

The department strives to improve the scientific basis for the development of new ideas for the future production and management methods in industrial and agricultural production.

Through basic and strategic research, the department aspires to develop methods for future industrial and agricultural production thereby exerting less stress and strain on humans and the environment.

Approach

The department expertise spans a wide range of subjects including atmospheric chemistry, chemical kinetics in the liquid and gas phase, geochemistry, biogeochemistry, geochemical modelling, hydrochemistry, analytical chemistry, process chemistry, organic synthesis, plant molecular biology, plant pathogens, plant genetics, population biology, methods for plant breeding, plant nutrition, biological interaction, nutrient cycling, marine and terrestrial ecology, radioecology, trace metal ecology and ecotoxicology. Research and development activities are concentrated in the following areas:

- The atmospheric environment
- Plant genetics and resistance biology
- Plant nutrition and nutrient cycling
- Radioecology
- Trace metal ecology and ecosystem studies
- Geochemical processes, chemical analysis and waste treatment
- Industrial use of radiation and dosimetry
- Organic synthesis, sensors and molecular design

The results of research and development are disseminated to companies, institutions, organizations and public authorities through scientific publications, research reports, lectures and posters at scientific – and other professional meetings, personal communication with collaborators and through teaching courses at Universities.

The research and development activities in the department are planned for three years and are reassessed each year. The department regularly evaluates possibilities for improving the scientific contribution within the main field of research, and examines promising new research areas.

The research activities are mainly funded directly from the government or from National Science Research Councils, but national and European research programmes, private foundations and commercial contracts make a substantial contribution to the total budget of the department.

1.2 The Atmospheric Environment and Air Pollution

The atmospheric research aims to establish a firm scientific basis for sensible and effective legislative measures to reduce industrial and agricultural impact of air pollution on the atmospheric environment.

The effect of human activity on the global atmosphere has become evident during the last decades. For example, the tropospheric ozone concentrations have increased by a factor of three in remote unpopulated sites and photochemical smog can be detected very long distances away from densely populated areas.

Changes in the atmospheric composition and chemistry influences the climate, and deposition of airborne pollutants can give rise either to direct injuries to plants, or to indirect damage through bio-accumulation in ecosystems. To model and predict changes in the atmospheric

composition, a better understanding of the chemical processes in the atmosphere is needed. If research is to operate successfully in environmental policy making, critical loads for different air pollutants need to be established for natural and semi-natural ecosystems.

The research activities in the department include basic atmospheric chemistry, gas kinetics, transport and dispersion of air pollutants (especially nitrogen compounds), carbohydrates, VOCs, PANs, PAHs, CFCs, HCFCs and HFCs, and the effect of air pollution on trees and crop plants.

The research in atmospheric chemistry and air pollution involves a number of aspects: 1) Determination of the sources of air pollution. 2) Determination of primary and secondary pollutants in rural areas to assess the effects of atmospheric processes. 3) Laboratory studies of chemical mechanisms of relevance to atmospheric chemistry in the gas phase, and in the liquid phase. 4) Combustion kinetics where direct kinetic studies of important radical species are carried out in the presence of various additives such as O_2 and NO . 5) Field atmospheric chemistry, including PAH from city traffic. 6) The uptake of nitrogen dioxide and nitric oxide through the leaves of barley. 7) The effect of ammonia vapour and ammonium sulphate particles on the Norway spruce.

1.3 Plant Genetics and Resistance Biology

The department aims to develop a scientific basis for breeding crop plants with new and stronger resistance to diseases and with improved nutrient efficiency. Crop plants highly resistant to diseases and efficient in nutrient uptake, are a prerequisite for the low input plant production of the future. Plants highly resistant to diseases with less nutrient demand will reduce a number of environmental problems related to high yield plant production.

An understanding of the interaction between plant species, as well as the interaction between plants and pathogens, can be enhanced by the identification of changes in genetic variability under different environmental conditions. The use of genetic variability, related to attributes of eco-

nomical and environmental importance in crop improvement, can be greatly enhanced by the use of genetic markers in the selection process. A large part of the research effort within plant biology and genetics is devoted to the use of genetic markers. The research in this field relies on the command of a number of techniques such as RFLP, RAPD, QTL, genetical analysis and cytogenetic techniques.

The department works to establish genetic linkage maps of the barley genome, based on RFLP and RAPD markers. The markers are then successfully used to map loci for agronomical important traits, such as disease resistance, grain quality and yield.

In 1991, mapping of the Norway spruce and cotton genome was initiated to establish similar linkage maps based on RAPD markers, thereby enabling us to be able to map loci for productivity, disease resistance and quality of products in these two species.

The actual cytological position of a gene loci is necessary for the transformation of a crop species with a specific gene from an alien species. For this purpose, the department apply *in situ* hybridization of DNA probes with chromosomes to identify the physical position of a gene loci.

The storage protein, hordein in barley grain, is poor in lysine, and the department is active in improving the nutritional quality of barley protein by increasing the lysine content.

Different aspects of plant-pathogen interactions are studied. 1) Chemical defence compounds, such as callose, peroxidase and chitinase are studied in barley and rape seed as well as various phenolic compounds in barley. 2) Genetic resources of species specific resistance in barley to powdery mildew and leaf stripe disease are studied. 3) We try to elucidate the very specific interaction, based on gene to gene recognition in the barley powdery mildew system.

In population biology, experiments and theoretical studies of interactions between populations are important in understanding the ecosystems. The research involves interrelationships between crop plants and weeds, and between crop plants and their pathogens. The possible exchange of genes between related species and the competitive abilities of crop plants are investigated. The

risk of gene flow from genetically modified cultivated plants into wild natural relatives and the effect of new genes transferred to natural plants are assessed. This type of research is needed to evaluate the risk to the environment from transgenic varieties.

Interactions between crop plants and weeds are studied, and a synergistic effect of corn cockle on wheat was demonstrated in preliminary experiments. Both total biomass and seed production of wheat increased significantly in the presence of corn cockle.

1.4 Plant Nutrition and Mineral Cycling

Research in plant nutrition and mineral cycling aims to provide basic scientific information for a better understanding of the mechanisms and processes involved in the root-microbe symbiosis in relation to carbon, nitrogen and phosphorus. It also aims to provide basic scientific information about the processes involved in the turn-over of carbon, nitrogen and phosphorus in agro ecosystems.

In addition, the research will provide information to improve the agricultural management practice, reducing the losses of nutrients (especially nitrogen) from plants to the atmosphere, and from the root zone to the ground water, streams or lakes. The transfer of plant nutrients, particularly nitrogen and phosphorus, through the Soil-Plant-Air-Continuum is being investigated.

The research in plant nutrition and mineral cycling has four aspects. 1) Research on symbiotic nitrogen fixation, which highlights the exchange of compounds between the host plant and the microsymbiont across the membrane interface. 2) Studies of the interaction between VA-mycorrhiza, plant roots and the rhizosphere with special emphasis on the role of VA-mycorrhiza in phosphorus uptake by crop plants. 3) Studies on turn-over of nitrogen and phosphorus from crop residues and farmyard manure. 4) The biological transfer of nitrogen from legumes to non-legumes.

These research activities are essential for the future development of a new low input plant production strategy based on biologically fixed nitrogen and phosphorus uptake mediated by VA-mycorrhiza. To reduce the loss of plant

nutrients due to surface run off, or losses of nutrients to the atmosphere or leakage to the ground water, highly effective assimilation and recycling of nutrients are required.

1.5 Chemistry of the Geosphere

In geochemical modelling, the JENSEN programme has been expanded and improved and a systematic study of humic acids, with respect to flocculation and de-flocculation processes was performed. These processes were studied in relation to concentrations of sodium, calcium, chloride, protons and rare-earth metals in soil solutions.

The presence of inhomogeneities in geological formations may have a profound influence on the path of flowing ground water. In the department computer simulations are used to estimate uncertainties to be expected in the transport of ground water and contaminants through a heterogeneous geological media.

In marine geochemistry, rare and precious metals are studied in marine sediments. Most marine ferromanganese phases contain anomalously high amounts of metals not usually found in terrestrial environments. One of these heavy metals, tungsten, was studied in detail. The interesting new phenomena, black smokers at the mid-ocean spreading-ridges were studied. These hydrothermal vents may supply significant amounts of metals to the ocean floor thus influencing the global metal budget.

Radiometric methods for inspection of the sea floor are important in environmental research. Radiometric techniques, in the form of integrated instrument platforms and new techniques for the recovery of samples, are under consideration for exploring marine sediment resources.

The retention of heavy metals by clay minerals has been studied in an effort to develop techniques to eliminate the hazard of heavy metals in municipal sludge and industrial waste. The smectic minerals, montmorillonite in particular, have a large capacity for cation exchange; bentonite (a clay high in montmorillonite) was used for the experimental work. Growth experiments with Italian rye grass were used to test the accessibility of heavy metals in the clay after treatment.

The department contributes to the scientific basis for developing the wet oxidation technique to treat soil and waste water polluted with organic compounds and heavy metals. New methods are being developed for the conditioning and treatment of soil, sludge, sewage and other waste products from industrial productions contaminated with heavy metals. The wet oxidation of wheat straw was studied as a pre-treatment to enzymatic treatments. Wet oxidation readily solubilizes lignin in straw and the product is susceptible to enzymatic treatment.

In analytical chemistry the electrothermal vaporization unit linked to the ICPMS instrument is in operation and a Ph.D. project was initiated to develop new methods to eliminate interference from chloride on As and Zn analysis. The department is presently involved in developing a new BCR lichen standard, and fast and reliable analytical screening methods for production control in the Danish agro-industry.

1.6 Ecology and Mineral Cycling

Research within ecology and mineral cycling aims to improve the scientific basis and understanding of the impacts of environmental pollution on marine and terrestrial ecosystems with emphasise on natural and semi-natural ecosystems. Further the research aims to establish better knowledge about transport and the fate of pollutants by the use of stable and radio isotopes and tracemetal techniques.

To study the possible transfer of radio-nuclides from the soil to humans, the uptake of radiocesium by grain and vegetables was examined in the Nordic countries along with ecological half-lives of radiocesium in the terrestrial environment contaminated by regional fallout from the Chernobyl accident.

A dynamic radioecological model, simulating the transfer of radiocesium through the soil-grass-lamb foodchain, was developed. The model is based on field observations in the Nordic countries and thus reflects the possible transfer of radiocesium to humans via lamb meat.

The total amount of ^{90}Sr in the debris from Chernobyl may be underestimated significantly, because the standard analytical technique does not include insoluble ^{90}Sr . If the insoluble ^{90}Sr in the soil becomes more soluble with time, due to weathering, the doses from ^{90}Sr close to Chernobyl may remain higher for a longer period than otherwise expected.

Radioactive tracer techniques are used as bioindicators for marine monitoring programmes for radionuclides and heavy metals in projects studying metal toxicity to fish and Baltic mussels. Survival of rainbow trout exposed to Cu^{++} was very dependent on temperature and salinity at the time of exposure, as well as changes in salinity after the exposure.

Radionuclides, discharged from La Hague in France, are used to trace the advection and dispersion of water in the European Coastal Current from the English Channel to the entrance to the Baltic. Results from these and previous Atlantic studies, provide very important knowledge about the origin of the water masses entering the domestic Danish waters. The European Coastal Current water carries much higher loads of nutrients and pollutants than that from Atlantic waters.

The Ecology Section also develops strategies and methods for decontaminating radioactive contaminated areas. In 1992, the efficiency of different decontamination methods was tested in the town of Pripjat near Chernobyl and the results were, in general, encouraging.

The indoor aerosol deposition was studied by using neutron-activatable tracers as substitutes for airborne contaminants, such as bacteria, fungi and radon progeny which are attached to ambient aerosols. The process of aerosol deposition is an important factor for aerosol concentration and thereby for potential health hazards in buildings.

1.7 Organisation

The Environmental Science and Technology Department includes approximately 135 scientific and technical staff members, and the department is organized in four sections with Dr. Arne Jensen as Department Head.

1. Ecology Section,

Head: Dr Asker Aarborg.

2. Chemical Reactivity Section,

Head: Dr Ole John Nielsen.

**Dr Arne Miller is in charge of the Risø
High Dose Reference Laboratory.**

3. Chemistry Section,

Head: Dr Vagn Gundersen.

4. Plant Biology Section,

**Head of plant nutrition: Dr Gunnar Gissel
Nielsen.**

**Head of plant genetics: Dr Hanne
Østergaard.**

**Dr Vagner Haahr is in charge of growth
chambers, greenhouses and the experi-
mental farm, Dyskærgaard.**

**The Environmental Science and Technology
Department was formed in March 1990, and
includes the former Agricultural Research De-
partment, part of the former Chemistry Depart-
ment, and the Ecology Section from the former
Ecology and Health Physics Department.**

2 The Atmospheric Environment

2.1 Introduction

The effect of human activities on the global atmosphere has become evident during the last decades. For example, the tropospheric ozone concentrations have increased by a factor of three in remote areas and photochemical smog can be observed thousands of kilometres away from populated areas. An understanding of atmospheric changes requires a detailed characterization of the composition of the atmosphere as well as an understanding of physical and chemical processes. The atmospheric research of the department has three aspects: The first is the determination of the sources of air pollutants. The second is the determination of primary and secondary pollutants in rural areas to assess the influence of atmospheric processes. The third is the laboratory investigation of chemical mechanisms in all three phases relevant to atmospheric chemistry.

2.2 The Gas Phase

The work in the gas phase was carried out in several different projects: Fundamental chemical kinetics, halogenated compounds, CFC substitutes and related compounds, nitrogen and sulfur species, including aromatics and NO_x reduction chemistry.

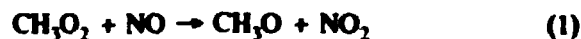
Fundamental Chemical Kinetics

Alkyl peroxy radicals are key reaction intermediates in the atmospheric oxidation and low temperature combustion of every hydrocarbon. Recognition of the crucial role played by these species has led to a significant research effort to elucidate the kinetics and the mechanisms of the reactions of these species. Kinetic studies usually involve monitoring the change in concentration of these species using their strong absorption in ultraviolet, 200–300 nm. Absolute absorption cross-sections are then needed to determine the concentrations of these peroxy radicals.

Despite a large number of studies, there are still significant uncertainties associated with absolute values of the absorption cross-sections

of alkyl peroxy radicals. For example, the absorption spectrum of CH_3O_2 radicals was measured in 16 different studies. While all studies agree on the overall shape of the spectrum, there are significant differences in the reported absolute values of $\sigma(\text{CH}_3\text{O}_2)$. These differences reflect the fundamental experimental difficulties associated with the absolute calibration of low concentrations of these reactive species.

The conventional approach to this problem was to conduct two experiments. First, the absorption of the peroxy radical in question is measured. Then, in a separate experiment, the yield of radicals in the system is determined using actinometry, either by following the loss of the photolytic precursor, or by monitoring the appearance of a product. A new approach to this problem has been found in which the peroxy radical is converted stoichiometrically into an alkyl nitrite by reaction with NO. For example for methyl peroxy radicals:



Alkyl nitrites absorb strongly in the UV. Hence, as peroxy radicals are converted into the nitrite the initial UV absorption, attributable to the peroxy radical, is replaced by absorption by the nitrite. From a single experiment it is, therefore, possible to relate the absorption cross-section of these two species. By varying the monitoring wavelength, the isobestic point, where the absorption cross-section of the peroxy radical is equal to the sum of those of the nitrite and NO_2 , is readily determined.

Once the isobestic point has been established, the problem of measuring the absorption cross-section of the peroxy radical is simplified to measuring that of the nitrite and NO_2 at that wavelength. Alkyl nitrites are, in general, stable compounds under ambient conditions and are readily synthesized in high purity. Hence, measurement of the UV spectra of these compounds can be performed with great accuracy using conventional UV spectrometers.

Provided that conditions are chosen to ensure stoichiometric conversion of peroxy radicals into the nitrite, this approach offers a significant advantage over conventional methods, with nitrite as an internal standard.

This new approach was explored, using our pulse radiolysis system, to study the absorption spectrum of methyl peroxy radicals. The results were impressive and have been published, see Fig. 2.2.1 (Wallington *et al.*, 9.1c).

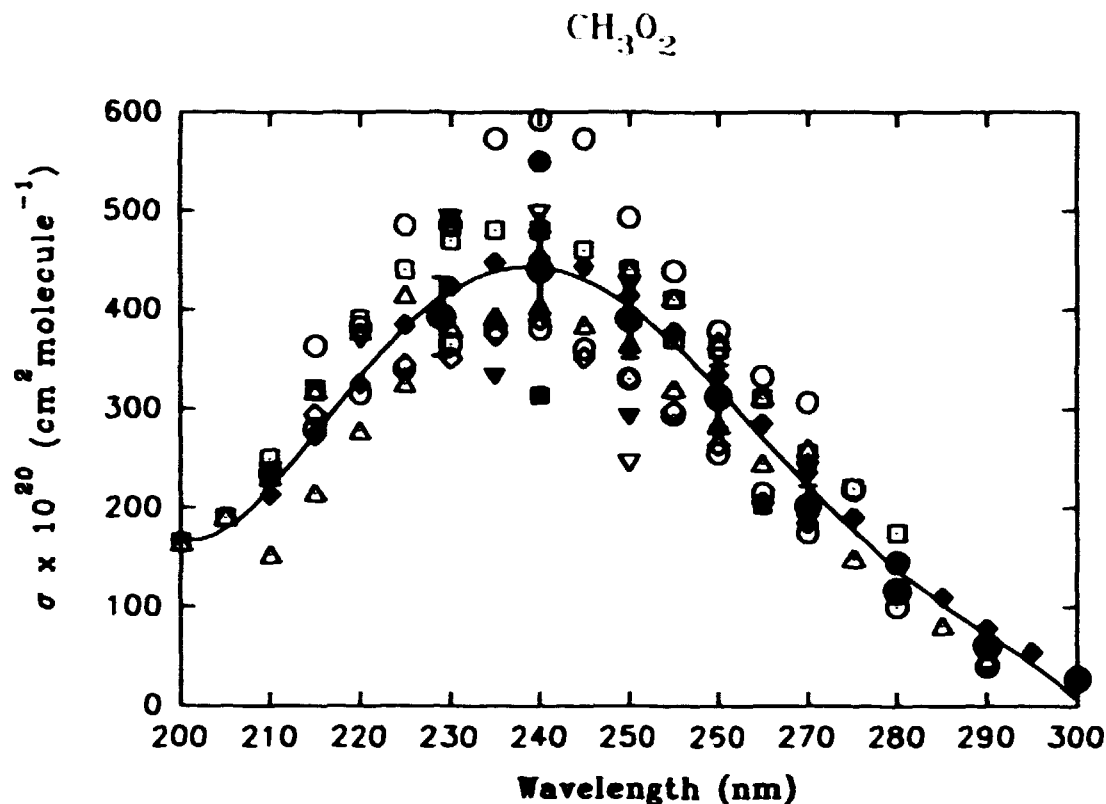


Fig. 2.2.1. Sixteen different experimental determinations of the UV absorption spectrum of the methyl peroxy radical, CH_3O_2 . The Riso data are indicated by enlarged filled circles.

While there is now much information concerning the chemistry of alkyl peroxy radicals containing 1 or 2 carbon atoms, there have been relatively few studies of larger peroxy radicals. The largest peroxy radical, in terms of the number of carbon atoms, for which spectroscopic and kinetic data are available, is the neopentyl peroxy radical, $(\text{CH}_3)_3\text{CCH}_2\text{O}_2$. Two previous independent studies of this radical, using flash photolysis of neopentane/ Cl_2/O_2 mixtures, distinctly showed nonsecond order decay of the UV absorption following the flash. From these studies, however, two very different mechanistic conclusions were drawn. As part of a collaborative study between Riso National Laboratory and Ford Motor Company on the atmospheric chemistry of peroxy radicals, a new experimental study of the neopentyl peroxy radical was performed. The pulse radiolysis technique was used to measure the UV

absorption spectrum and the self-reaction kinetics of the $(\text{CH}_3)_3\text{CCH}_2\text{O}_2$ radical. The products of the self-reaction were determined using an FTIR spectrometer coupled to an atmospheric reactor. The results clearly support the conclusion from one of the previous studies (Wallington *et al.*, 9.1a).

Halogenated Organic Compounds in the Troposphere

The STEP project called HALOCSIDE, involving 9 European laboratories, is concerned with the kinetics and mechanisms of oxidation of halogen-containing compounds under atmospheric conditions. This project was extended for one more year in 1992. The aim of this study is to determine the tropospheric sinks for these compounds, to provide the basis for calculation of the

ozone depletion potential for released halogenated species.

A fundamental study of the reactions of OH radicals with a series of chloroalkanes over the temperature range 295–360 K was performed. Surprisingly, high activation energies were found for the reaction of OH with chloropropanes. There is evidence that chlorine substitution in alkanes has two different effects on the reaction with OH radicals: a) an inductive effect which decreases the reactivity for H-atom abstraction extending at least to the β -carbon and b) a mesomeric effect increasing the reactivity at the α -carbon. In the reaction of these chloroalkanes with OH radicals, the first effect might be dominant, resulting in an overall decrease in the rate constants and an increase in the activation energies compared to n-alkanes with the same number of CH_2 -groups. It cannot be ruled out that an increase in bond dissociation energy at the β and/or γ carbon explains part of the increased activation energy, but it is unlikely to be the main reason. Further support for this suggests that for exothermic chlorination reactions, bond dissociation energies are less important, because the extent of bond breaking in the transition state is small. This might, to some extent,

also be the case with OH radical reactions (Markert and Nielsen, 9.1b).

The part of this work concerned with the spectra and kinetics of halogenated peroxy radicals is being performed in collaboration with Dr T. Wallington, Ford Motor Co., USA. Several halogenated compounds, and a series of fluorinated methyl peroxy radicals (CH_2FO_2 , CHF_2O_2 and CF_3O_2), were investigated. All three radicals, namely the spectra, kinetics and mechanisms were studied for their self-reaction. Their UV spectra are shown in Fig. 2.2.2. The substitution of H atoms with F atoms has little or no effect on the shape and magnitude of the absorption spectra. The F atom substitution, however, moves the absorption maximum to progressively shorter wavelengths by approximately 15 nm per F atom.

There are no previous studies of the products of the self-reaction of fluoromethyl peroxy radicals with which to compare our present results. The self-reaction of e.g. CF_3O_2 radicals can proceed via two possible pathways:

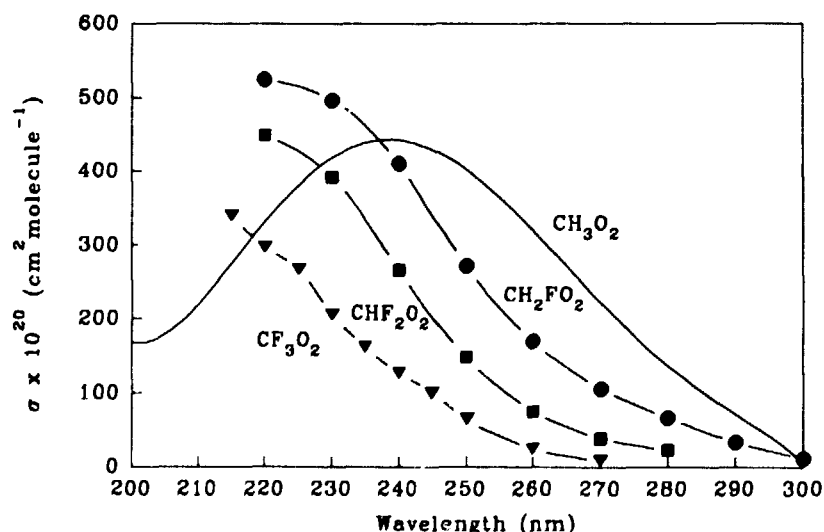
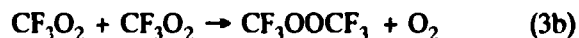
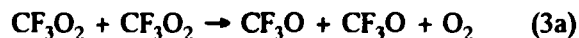
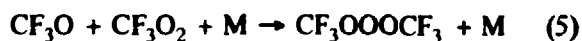


Fig. 2.2.2. UV absorption spectra of fluorinated methyl peroxy radicals obtained at Risø, and compared to that of CH_3O_2 .

The absence of any product with IR features, attributable to the peroxide CF_3OOCF_3 , shows that (3b) is not an important reaction. The absence of the peroxide with essentially 100%

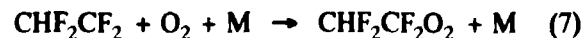
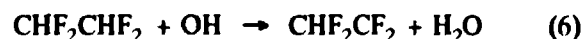
yield (in terms of carbon) of the trioxide, shows that the self-reaction of CF_3O radicals (reaction (4)) does not compete effectively with reaction (5) for CF_3O radicals:



The fact that the $\text{CF}_3\text{OOOCF}_3$ product accounts for essentially all the CF_3O_2 radicals, formed in the FTIR reactor, has interesting ramifications for the kinetic data obtained using the pulse radiolysis system. Each CF_3O radical that reacts via reaction (5) consumes a CF_3O_2 radical. In the kinetic experiments, the decay of CF_3O_2 is used to derive the rate constant for reaction (3). Hence, the observed rate constant for reaction (3) may be an overestimate of the true rate constant. Interestingly, within the combined experimental uncertainties, the observed self-reaction rate constants for all three fluorinated methyl peroxy radicals are indistinguishable (Wallington *et al.*, 9.1b), (Nielsen *et al.*, 9.1a, b).

CFC Substitutes

By international agreement, industrial production of chlorofluorocarbons (CFCs) will be phased out. Hydrofluorocarbons (HFCs) are one class of potential CFC substitutes. Prior to large scale industrial use, it is important to establish the environmental impact of the release of HFCs into the atmosphere. Following release, HFCs will react with OH radicals in the lower atmosphere to produce fluorinated alkyl radicals which will, in turn, react with O_2 to give peroxy radicals. For example, in the case of CHF_2CHF_2 (HFC-134):



There is relatively little information available concerning the atmospheric fate of the fluorinated alkyl peroxy radicals.

As part of a collaborative study by Risø and Ford on the atmospheric fate of HFCs, an experimental study of $\text{CHF}_2\text{CF}_2\text{O}_2$ was conducted. CHF_2CHF_2 (HFC-134) is one of the two isomers of tetrafluoroethane, the other being CF_3CFH_2 (HFC-134a) which is a proposed substitute for CFC-12 in automotive cooling systems. The spectrum and kinetics of the peroxy radical derived from HFC-134a have previously been studied at Risø.

The ultraviolet absorption spectrum, and kinetics and mechanism of the self-reaction of $\text{CHF}_2\text{CF}_2\text{O}_2$ radicals were studied in the gas phase at 298K. Two techniques were used; pulse radiolysis UV absorption, to measure the spectrum and kinetics and long path-length spectroscopy FTIR, to identify and quantify the reaction products. Absorption cross-sections were quantified over the wavelength range 220–270 nm. At 230 nm, $\sigma_{\text{CHF}_2\text{CF}_2\text{O}_2} = (3.2 \pm 0.5) \times 10^{-18} \text{ cm}^2 \text{ molecule}^{-1}$. This absorption cross-section was used to derive the observed self-reaction rate constant for the reaction, $\text{CHF}_2\text{CF}_2\text{O}_2 + \text{CHF}_2\text{CF}_2\text{O}_2 \rightarrow \text{products}$, $k = (2.7 \pm 0.6) \times 10^{-12} \text{ cm}^3 \text{ molecule}^{-1} \text{ sec}^{-1}$. Measured UV transients were not corrected for possible complications caused by the formation of CHF_2O_2 and HO_2 radicals. The only carbon-containing product observed by FTIR spectroscopy was COF_2 . The carbon balance was, within our experimental uncertainty, 100%. Hence, the oxidation of HFC-134 in the atmosphere will not give HF in contrast to HFC-134a (Nielsen *et al.*, 9.1c).

Organonitrogen and Sulphur Atmospheric Transformations

A four-year CEC-funded collaborative study with the University College Dublin, concerning the reactions of OH radicals and Cl atoms with a wide variety of compounds, was concluded by the writing of a final report to the CEC (Nielsen *et al.*, 9.3).

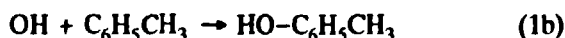
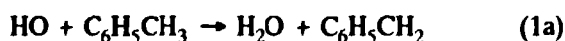
Much recent interest has centred on the role of natural organic sulphur compounds in atmospheric sulphur chemistry. The CH_3S radical, and possibly the CH_2SH radical to a lesser extent, are important intermediates in the oxidation of naturally occurring sulphur species in the atmosphere. We have continued our collaboration with Dr C. Anastasi's group at the University of York (UK) and have determined the rate constants for the reaction of CH_2SH radicals with O_2 , NO, and NO_2 . The results have been published (Anastasi *et al.*, 9.1).

In the daytime the OH radical is the most important oxidizing species in the troposphere; during the night the NO_3 radical plays a similar role. In spite of the fact that NO_3 plays an important role in the troposphere, the chemistry of this species is not sufficiently well examined. For example, there are large gaps in the knowledge

of the reactions of NO₃ with 1,3-butadiene and with isoprene, the latter being the most important non-methane hydrocarbon in the troposphere. Few studies on the product formations of these reactions occur in the literature. The rate constants for the NO₃ reaction with 1,3-butadiene and isoprene are not well established despite numerous determinations. The rate constants for the reaction of NO₃ radicals with a series of 7 dienes: 1,3-butadiene, isoprene, 2,3-dimethyl-1,3-butadiene, trans-1,3-pentadiene, cis-1,3-pentadiene, trans,trans-2,4-hexadiene, and 1,3-cyclohexadiene were measured at 295 K. The rate constants were obtained using the absolute technique of pulse radiolysis combined with kinetic UV-VIS spectroscopy. The results have been published (Ellermann *et al.*, 9.1).

Oxidation of Aromatics (STEP 0007 C(MB))

The tropospheric oxidation of benzene and other aromatics is initiated by reactions with OH radicals. In the case of toluene, the reaction with hydroxyl radicals was found to proceed via two different pathways:

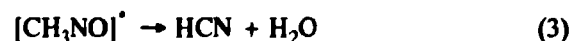


The ultraviolet spectra of C₆H₅CH₂ and HO-C₆H₅CH₃ were recorded in the range of 210–300 nm. The overall rate constant $k_1 = k_{1a} + k_{1b}$ was determined by direct kinetic studies, while the branching ratio, $k_{1a}/(k_{1a}+k_{1b})$ was derived from the observed radical yields. The radical species produced in the reactions (1a) and (1b) exhibit great differences in structure and chemical reactivity. The decay rate of the radical C₆H₅CH₂ in the presence of oxygen is fast, while HO-C₆H₅CH₃ was found to react much more slowly with oxygen, as in the case of HO-C₆H₆ which we have studied previously. The reaction of benzyl radicals with molecular oxygen gives rise to the formation of a peroxy radical, C₆H₅CH₂O₂ which has an absorption spectrum with a maximum at 260 nm. Work is in progress to study the kinetics of benzylperoxy radicals, *e.g.* the self-reaction and the reaction with NO.

Combustion Kinetics (JOUE-0036-C(JR))

The CEC JOULE programme is concerned with Kinetic Evaluation for Combustion using Experimental and Numerical Expertise (KEENEX). Our contribution to this European collaboration is concerned with kinetics and spectroscopy of free radicals of importance in combustion processes. The experimental technique is based on pulse radiolysis combined with time-resolved ultraviolet and infrared spectroscopy.

For the first time, we have recorded the UV-spectra of a number of important radical species, *e.g.* HC=O, CH₂OH, CH₂CH₂OH and CH₃CHOH. Based on these spectra, we have been able to carry out direct kinetic studies of these radical species in the presence of various additives, *e.g.* O₂ and NO. The chemistry of nitrogen oxides in combustion processes is very complex and the development of De-NO_x technologies is of great importance. "Reburn" of exhaust gases from power-plants is one of the methods employed to reduce NO_x emissions. The chemical reaction mechanisms, however, are very complex. In particular, it has been difficult to account for the formation of unwanted products such as the very poisonous HCN gas. We have studied the kinetics and mechanism of the reaction of methyl radicals with nitric oxide which may give rise to the formation of HCN under typical "reburn" conditions. The kinetics of methyl radicals was studied by time resolved infrared spectroscopy and the rate constant was found to be strongly dependent on the total pressure. This implies that the formation of a "hot adduct" can be stabilized by collisions with bath gas molecules. The reaction mechanism may be visualized in terms of the reactions (1) – (3):



The reactions so far, have only been studied at room temperature. The formation of HCN via reaction (3) was studied by time resolved infrared spectroscopy. The fraction of methyl radicals,

which are converted into HCN, amounts to only a few percent at $T = 298$ K. At higher temperatures, it seems very likely that reaction (3) may become the dominant reaction channel.

In addition to the pulse radiolysis experiments, we also employed time-resolved infrared spectroscopy in studies of slow thermal reactions of various nitrogen oxides, NO_x with suitable additives, which may bring about the conversion of nitrogen oxides to molecular nitrogen, $\text{NO}_x \rightarrow \text{N}_2$.

2.3 The Liquid Phase

The initiation reaction of O_3 decomposition in acidic solution is different from that in alkaline solution. In acidic solution, the initiation of the chain decomposition is ascribed to the ozone dissociation/recombination reaction. The creation of chain-propagating radicals (OH and HO_2) proceeds via a reaction with water in a heterogeneous process on the surface of the reaction vessel. A better understanding of the initiation process was achieved, when acetic acid is employed as a radical scavenger. In a study of concentration effects, pH, temperature and H_2O_2 formation and consumption, the rate of the initiation reaction and the activation energy were determined. From the same system, a rate constant for the reaction of ozone with undissociated hydrogen peroxide was found. A study of the reaction of O_3 with H_2O_2 indicates that both initiation and termination of the radical chain are surface reactions. Initiation of O_3 decomposition by H_2O_2 (a homogeneous initiation) leads to a surface to volume effect of the O_3 decay, which is not the case with thermal initiation. This is interpreted in terms of a heterogeneous initiation and termination of the process. The rate of initiation (termination) is proportional to r^2 of the vessel, which indicates diffusion of radicals to and from the surface area (Sehested *et al.*, 9.1).

The rates of the oxidation reactions of ferrous compounds with ozone were measured in acidic solution by a stopped-flow technique. An intermediate complex, FeO^{2+} (the ferryl ion) was found. Several rate constants were determined, and a mechanism for the disappearance of the complex, involving OH and HO_2 radicals, was suggested. This mechanism is supported by com-

puter simulations of all the results obtained (Løgager *et al.*, 9.1).

The SO_4^- and NO_3 radicals are important oxidizing agents in the aqueous phase of the atmosphere. The rate constants for SO_4^- reaction with HNO_3 and NO_3^- and the NO_3 radical reaction rate with HSO_4^- and SO_4^{2-} , respectively, were determined. An equilibrium between SO_4^- and NO_3 radicals was established. The equilibrium constant at pH 3.5–4 (SO_4^{2-} and NO_3^-) was found to be $K = 0.5$, which, by extrapolation to zero ionic strength, became $K = 0.28$. This constant and the ΔG_0 's for the solutes are used to calculate the reduction potential of the couple $\text{NO}_3/\text{NO}_3^-$ to $E_0 = 2.45 \pm 0.5$ V. The same reduction potential can be obtained from the equilibrium reactions in 1 M acid (HSO_4^- and HNO_3) (Løgager *et al.*, 10).

Using our high pressure technique, the formation of peroxyxynitrous and peroxyxynitric acids from $\text{HO} + \text{NO}_2$ and $\text{HO}_2 + \text{NO}_2$, respectively, was studied. These peracids are important constituents in the atmosphere. Their rates of formation and decay over the pH range 2–12 were determined. The former is stable in acid and the latter in alkali. The pK's of the acids and their respective spectra were also determined. Work is in progress to elucidate the mechanism of the decay at various pHs.

A study on the exchange between water and the OH radical was performed by mass-spectrometric measurements of the isotopic distribution of oxygen developed from H_2O_2 in the irradiated sample. The work was carried out in collaboration with Aarhus University.

The work on high-temperature (up to 300°C), high-pressure (up to 15 MPa) aqueous radical chemistry resulted in the determination of rate constants and activation energies of $\text{e}_{\text{aq}}^- + \text{OH}$, $\text{e}_{\text{aq}}^- + \text{H}$, $\text{e}_{\text{aq}}^- + \text{H}_2\text{O}_2$ and $\text{H} + \text{OH}^-$, $\text{H} + \text{OH}$ and $\text{Fe}^{2+} + \text{H}_2\text{O}_2$ and $\text{OH} + \text{NH}_3$. The work was carried out in collaboration with Studsvik Material AB, Sweden, and Saclay, France (Christensen *et al.*, 10), (Hickel and Sehested, 9.1).

Kinetics of reaction of primary water radicals with pyridine, 3-methylpyridine, 3,5-dimethylpyridine, 2,6-dimethylpyridine and 2,4,6-trimethylpyridine, as well as spectral and kinetic characteristics of the transients formed, were studied by pulse radiolysis. The pK_a values of OH adducts and to what extent the pyridinyl radicals formed, were determined. The work continues by

studying several chlorinated aromatic compounds with special emphasis on the peroxy radicals, as these radicals are essential in the oxidation process. The compounds are important pollutants in water and waste waters. The work is being performed in collaboration with the University of Vienna, Austria (Solar *et al.*, 10).

2.4 Field Atmospheric Chemistry

City Traffic PAH Profile

Air pollution by polycyclic organic matter (POM) including polycyclic aromatic hydrocarbons (PAH) has been investigated for many years, one reason being that several of these compounds are carcinogenic.

Within DCAR (Danish Center of Atmospheric Research), the Section of Chemistry started a traffic PAH-project in January 1992. The project provides PAH data from different locations in the initial phase of the introduction of catalyst-equipped cars. Another aim of this project is to determine the total, as well as the individual,

contributions from gasoline- and diesel-driven cars.

In order to estimate the total contribution from traffic, one needs to assess the composition of PAH from traffic and other sources. So far, the PAH composition in car exhaust has been evaluated from measurements of the exhaust from individual cars or from measurements in traffic tunnels. None of these methods are perfect. The PAH composition in the exhaust differs from car to car and is dependent on driving conditions, fuel, lubricant and engine conditions. Thus a new approach has been developed to assess the PAH composition from traffic sources. PAH levels were determined at two close measuring points, a busy street and a city park area. The difference in levels between the two measuring points is caused by higher emission levels of city traffic sources. The PAH composition from city traffic sources can easily be calculated from these differences. Fig. 2.4.1 shows the PAH composition from city traffic sources, the PAH composition in fifteen 24-hour samples from the busy street, and in the same number of samples from the city park.

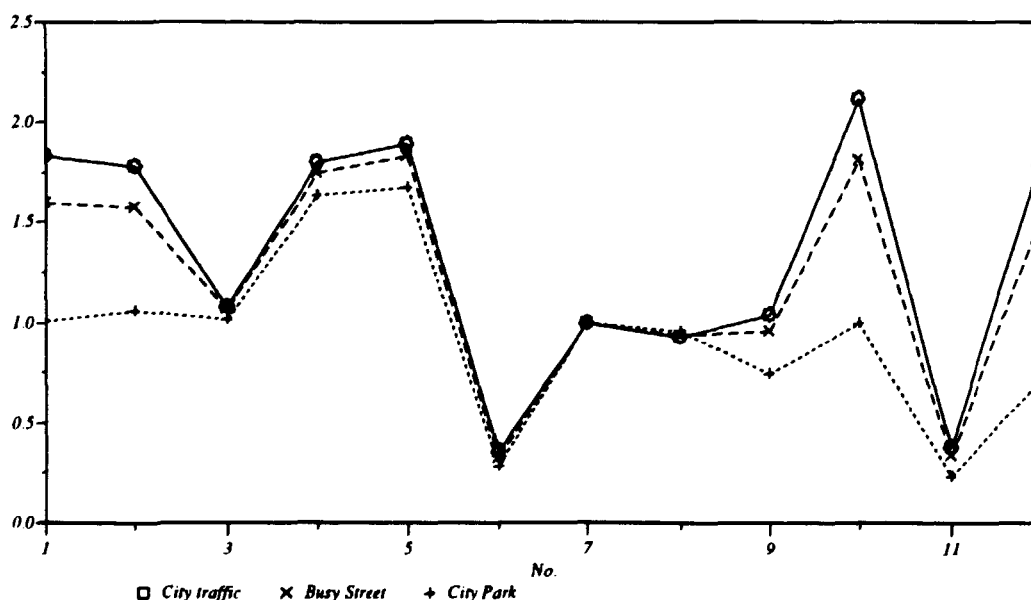


Fig. 2.4.1. The PAH composition (relative to benzo(e)pyrene) from city traffic sources, in a busy street and in a city park area. Identity of the PAH: 1. Benzo(ghi)fluoranthene + benzo(c)phenanthrene, 2. Cyclopenteno(cd)pyrene, 3. Benz(a)anthracene, 4. Chrysene + triphenylene, 5. Benzo(b)- + benzo(j)- + benzo(k)fluoranthene, 6. Benzo(a)fluoranthene, 7. Benzo(e)pyrene, 8. Benzo(a)pyrene, 9. Indeno(1,2,3-cd)pyrene, 10. Benzo(ghi)perylene, 11. Anthanthrene, 12. Coronene.

As one can see from Fig. 2.4.1 benzo(ghi)fluoranthene + benzo(c)phenanthrene, cyclopenteno(cd)pyrene, benzo(ghi)perylene (BghiP) and coronene (Cor) are emitted in relatively high amounts from traffic sources. This was also observed by means of factor analysis in a recent study of the PAH pollution at an international airport. The ratios of benzo(ghi)perylene to benzo(e)pyrene (BeP) and the Cor/BeP ratio in car exhaust gases have been slightly underestimated in earlier studies. The city traffic difference values show the BghiP/BeP ratio to be 2.1 ± 0.4 ($\pm 2\sigma$) and the Cor/BeP ratio 1.8 ± 0.4 . The earlier values were based on literature values and measurements from traffic tunnels. The previous values were 2.0 and 1.5, respectively, and the deviations were not significant. Hence, it was a good approach. The project is supported by the Danish Ministry of the Environment. The airport results are presented in a paper in press (Nielsen *et al.*, 10b).

Comparison of PAH and Mutagens

Aerosols and gas phase samples were collected at two locations at the international airport of Copenhagen and analyzed for PAH and mutagenic activity in the Ames' *Salmonella* strains TA98 and TA98NR. The distribution of PAH and mutagenic activity between particles and gas phase varied with season. The particulate fraction was the most important. Only for the lighter PAHs, such as phenanthrene and anthracene, a considerable part was found in the gas phase. The distribution was dependent on the ambient temperature and probably the concentration of soot particles. Winter samples contained approx. 10% of the total TA98+S9 activity and < 10% of the total TA98-S9 activity were in the gas phase. Summer samples contained 30-50% of the total TA98+S9 activity and 10-20% of the total TA98-S9 activity were in the gas phase.

The detected levels of pollutants were comparable to the general levels detected in the suburbs of Copenhagen. The average amount of benzo(a)pyrene at the airport terminal was 2.5 ± 0.7 ng/m³. The sources of PAH at the airport were a mixture of traffic and other sources, including local sources of heating and long-range transport. The air traffic was not an important source. Compared to the level of PAH, the level

of mutagenic activity was higher in long range transported particulate matter. This suggests a formation of mutagenic compounds during transport. Neither the ratios of TA98-S9 to PAH and TA98-S9 to TA98+S9, nor that of TA98-S9 to TA98NR, suggest air traffic as being an important source of mutagenic nitroaromatics. The results have been described in a paper submitted for publication and at an international symposium. The work was done in collaboration with Institute of Toxicology (Aagaard Nielsen and Nielsen, 9.2).

Indian Summer Photochemical Long-Range Transport

Summer long-range transport of photochemical oxidants from the European Continent to southern Scandinavia has been observed several times. These episodes are characterized by increased levels of ozone, peroxyacetyl nitrate (PAN) and visibility-reducing and strong acidic aerosols.

A similar episode was observed in the autumn of 1992, 21-27 Sept. see (Fig. 2.4.2). The transport of PAN, ozone and H₂O₂ was associated with increased levels of SO₂ indicating long-range transport of pollution from Continental Europe. The levels of photochemical oxidants did not follow the local amount of global radiation. The week of 21-27 Sept. had unusually mild weather, caused by warm winds from the south. Part of the period (22-23 Sept.) was characterized by strong winds (9-11 m/s). Most interestingly the transport was interrupted during the night of 23 Sept. by a cold front passing from the south-west. In the late morning of the 24 Sept. the wind turned to the south again and increased to 5 m/s, the temperature increased from 13 to 19°C, and the levels of photochemical oxidants again increased. In the last days of the period 26-27 Sept. the wind speed was low (approx. 3 m/s). The weather was cloudy and the global radiation low, showing that the photochemical air pollution was not caused by local formation. The period ended with a cold front coming from the north and passing Zealand on the night of the 27 Sept. After this on the 28 Sept. winds from the east brought cleaner air from southern Finland to the measuring site at Risø.

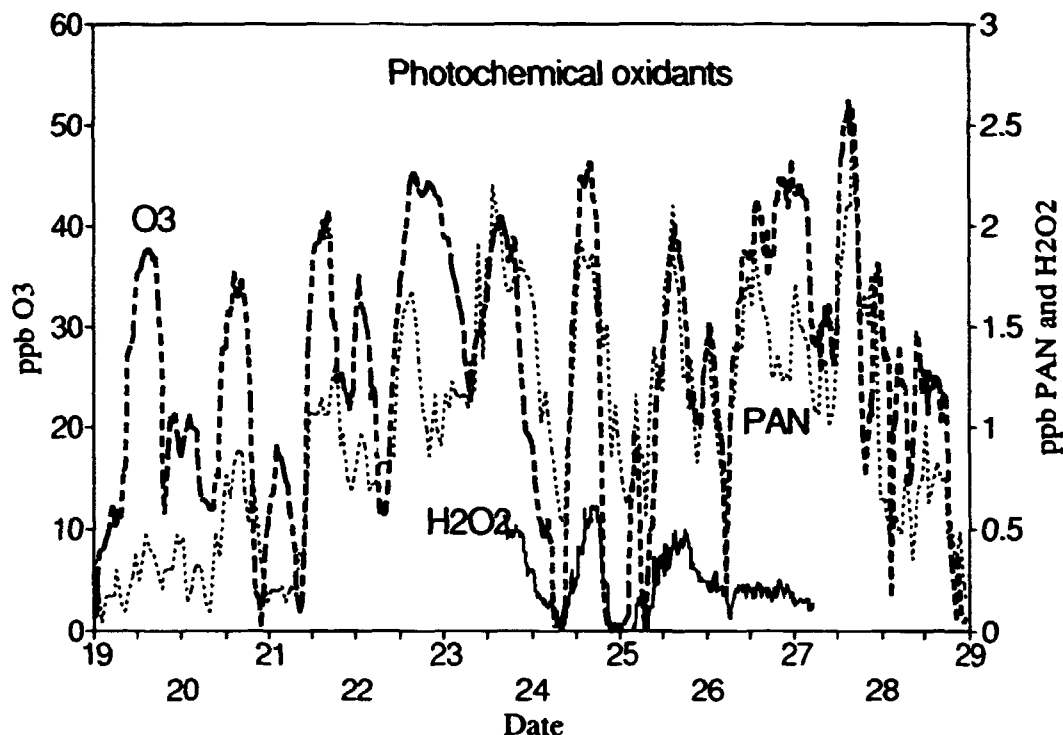


Fig. 2.4.2. The levels of the photochemical oxidants, PAN, ozone and H_2O_2 before, during (21–27 Sept.) and after Indian summer photochemical long-range transport.

In most cases NO_x in rural and agricultural areas represents the oxidation products of NO and NO_2 , i.e. the sum of HNO_3 , PAN, NO_3 , $2 \cdot N_2O_5$ and organic nitrates. The NO_x proportion of volatile NO_y was found to follow the levels of photochemical oxidants, e.g. O_3 : O_3 (ppb) = $(57 \pm 4) \cdot (NO_x/NO_y) + (18 \pm 10)$; $r = 0.61$, $p < 0.001$.

The major nitrogen components in the long-range transported polluted air were PAN, HNO_3 and particulate nitrate. On the night of 26/9, the three compounds made up more than 70% of the total NO_y .

The ratio between the formation rate of PAN and that of ozone depends on at least four factors:

1. The ratio between NO_2 and NO
2. The temperature
3. The hydrocarbon composition
4. The interaction of chlorine radicals.

Despite this, a positive correlation between PAN and ozone was observed:

$$O_3 \text{ (ppb)} = (8.6 \pm 1.8) \cdot \text{PAN (ppb)} + (28 \pm 6);$$

$$r = 0.70, p < 0.001.$$

The intercept of approx. 30 ppb, corresponding to the background ozone level was most interesting. The correlation only included the daily maximum values to avoid interference from depletion processes, e.g. decomposition by NO and by deposition. The possible differences in hydrocarbon composition in local and continental sources did not affect the correlation, as it was not affected either by local cases, long-range transport or both.

This investigation was performed within DCAR under the leadership of the Section of Chemistry as the Danish contribution to EUROTRAC-TOR. The investigation was supported by the National Strategic Environmental Project (SMP) and the National Agency of Industry and Trade. The results have been presented at the EUROTRAC-TOR workshop and at the DCAR Annual Meeting (Nielsen *et al.*, 9.5d, e).

Seasonal Atmospheric Composition of Nitrogen Compounds

The critical load concept is used to evaluate atmospheric pollution by nitrogen and other compounds. The non-critical use of this concept

does not take into consideration the great variations in their occurrence and composition. A winter and a summer period have been compared. In the winter period the levels of nitrogen oxides were high, typically around $80 \mu\text{gN/m}^3$ and the ozone levels close to zero. In the summer period the levels of nitrogen oxides were 7 times lower than those in the winter period, while the ozone levels were normal, in the range 30–35 ppb.

The composition of nitrogen compounds was also quite different. In the winter experiments, NO was the dominant species followed by NO_2 . HNO_2 was also observed. The composition was similar to that observed in urban street air. In the summer, NO_2 was the dominant species followed by HNO_3 and particulate nitrate. The proportion of the latter was 5 times higher in the summer than in the winter, while the proportion of NO was 8 times lower.

The ratio between volatile NO_2 and NO was much higher in the summer than in the winter period, showing that the amounts of transformation products of NO_2 are higher in the summer than in the winter:

August – September: $\text{NO}_x/\text{NO}_2 = 0.7 \pm 0.1$

January – March: $\text{NO}_x/\text{NO}_2 = 0.16 \pm 0.02$

Changing levels, as well as the composition of nitrogen oxides may occur within a few hours. An example was observed on 23 January 1992. In the morning and at night the levels of NO were high ($40\text{--}130 \mu\text{gN/m}^3$) and those of ozone zero. The ground inversion layer was in the lower 100 m. At noon, the inversion layer broke up and the polluted air at ground level was mixed up with fresh air containing ozone and low concentrations of nitrogen oxides. Thus at noon, ozone increased and NO and NO_2 decreased; NO to almost zero and NO_2 from 15 to $5 \mu\text{gN/m}^3$. In the late afternoon a new ground inversion layer was built up and stabilized at a height of 80–100 m. At 4 p.m. the ozone levels were zero again, with the levels of nitrogen oxides increasing.

This investigation was performed within DCAR under the leadership of the Section of Chemistry as the Danish contribution to EUROTRAC-TOR. The investigation was supported by the National Agency of Industry and Trade. The results are presented in a paper in

press in the Proceedings of the EUROTRAC Symposium (Nielsen *et al.*, 9.2b).

2.5 Uptake of Nitrogen Dioxide through the Leaves of Barley

Nitrogen dioxide (NO_2) and nitric oxide (NO) are present in the atmosphere primarily as a result of fossil fuel combustion, while other nitrogen compounds, such as nitrous oxide (N_2O) and ammonia (NH_3), are mainly biogenic. NO is rapidly oxidized to NO_2 , which subsequently reacts with atmospheric constituents to form peroxyacetyl nitrate (PAN), nitric acid (HNO_3), nitrate (NO_3) and dinitrogen pentoxide (N_2O_5). NO_3 and N_2O_5 are later converted to HNO_3 . NO_2 , PAN and HNO_3 are removed from the atmosphere by dry deposition.

Several investigations have shown that gaseous nitrogen compounds can be deposited on leaf surfaces and incorporated in the plant tissue. Nitrogenous gases can be divided into three groups:

- 1) highly reactive gases such as HNO_3
- 2) gases whose deposition is controlled by stomatal resistance such as NH_3 , NO_2 and PAN
- 3) gases that exchange slowly with plants such, as NO and N_2O

Recent investigations show that NO_2 and NO, as well as NH_3 , can also be taken up through the cuticle, although the amounts are usually much smaller than those taken up through stomata.

Most investigations of plant uptake of NO_2 were performed as short term experiments with concentrations much higher (300–3000 ppb) than those normally occurring in background areas. In rural areas of Denmark, average concentrations of NO_2 are 4 ppb, and in cities 20–30 ppb.

To determine foliar absorption of NO_2 in spring barley (*Hordeum vulgare* L.) under realistic concentrations, a fumigation experiment was carried out in open-top chambers. The uptake of nitrogen dioxide was determined by means of the ^{15}N -dilution principle. Also investigated was, whether barley plants can compensate for insuf-

ficient supply of nitrogen in the growth medium by increased absorption of NO_2 from the air.

The experiment showed that there was an absorption of N from fumigation amounting to 5–6% of the total N in plants. In addition, 3–5% was taken up from other sources, probably also of gaseous origin. The total absorption from gaseous sources in the NO_2 -fumigated plants might add up to approximately 10%. The absolute absorption from aerial sources was proportional to the top weight. In plants grown at low N-supply level, there was a slightly higher relative contribution from NO_2 , but the amounts absorbed from NO_2 were less than those in the plants supplied with optimal N to the growth medium.

The experiment is described in detail in (Jensen and Pilegaard, 10).

2.6 Effects of Air Pollution on Plants

Forest Decline and Ammonia

The strong symptoms and the many dying Norway spruce trees, particularly on nutrient poor sandy soils in Jutland show that the forest decline type called "røde rødgraner" (red Norway spruce) continues to be a serious problem. We investigated the ammonium hypothesis of Nihlgård, stating that the trees are over-fertilized with nitrogen in the form of ammonia vapour and ammonium sulphate particles derived from animal manure and slurry. The ammonium load is highest in Jutland which has a large population of domestic animals. We are over-fertilizing small Norway spruce trees in pots with ammonium nitrate or urea, in amounts up to ~ 800 kg N per ha. After the second season, many needles turn brown and drop off; some of the trees die. The trees contain a very unbalanced set of free amino acids, where arginine and ornithine are increased 10–20 fold and are found to be present in almost gram amounts per kg of needles. We hope that these amino acids can be used in needle tests in the field to diagnose N-over-fertilization, as it has been difficult to use standard leaf analyses of N, P and K and other nutrients for this particular purpose.

In some of our controls with balanced fertilization there are also trees (specific clones) with browning needles that eventually drop off. These needles are more reddish and the symptoms look more like those of the "red Norway spruce". This indicates that another hypothesis of spruce decline could be true, namely that Norway spruce will not tolerate mild winters and that they actually require a mean January temperature of -2°C .



Fig. 2.6.1. Very good growth is the initial response of overfertilization with nitrogen in young Norway spruce.

3 Plant Genetics and Resistance Biology

Methods for identification, characterization and quantification of genetic variability are essential to many basic and applied research programmes in the agricultural and environmental sciences. Exploitation of genetic variability, related to traits of economic and environmental importance in crop improvement, can be greatly facilitated by the use of marker-assisted selection. In particular, the new methods for identifying molecular markers may help in understanding the genetic basis of many agronomical traits which are of a quantitative nature and governed by the interaction of many Quantitative Trait Loci (QTLs) impossible to assay independently. An understanding of the regulation and expression of genes and the characterization of the gene products, the proteins, is important for developing plants with improved characteristics, e.g., a better quality of grain proteins or a higher level of resistance to pathogens.

Studies of plant population biology are necessary for predicting the consequences of using new genotypes and new species in agriculture. In this context, we are studying gene introgression from crop plants to their wild relatives in relation to risk assessment of the release of genetically modified plants.

An understanding of interactions between species of plants, as well as between plants and pathogens, can be greatly improved by identification of changes in genetic variability under various environmental conditions. For these purposes, a large part of our research is devoted to the establishment of genetic markers, construction of genetic/physical chromosome maps, and analysis of genetic diversity in populations. With these tools, the mechanisms of plant-pathogen interactions are studied with the aim of describing those gene products which are important for resistance or virulence. Further, the evolutionary processes imposed by interactions between crop plants and wild plant species or pathogens are being studied.

3.1 Gene and Chromosome Mapping

Linkage Map of the Barley Genome for Qualitative and Quantitative Traits

In modern agriculture optimal grain yield and quality have to be obtained under conditions with a minimal use of fertilizers and pesticides. We think that these goals can partly be obtained by means of plant breeding. More genetic information is needed, however, to exploit new advanced breeding methods. We try to establish genetic linkage maps of the barley genome based on RFLP and RAPD markers. The markers are then used to map loci for agronomically important traits such as disease resistance, and grain quantity and quality. Some of these traits are easily scored and simply inherited. The mapping of loci, which harbour genes for these characters, is important when attempting to isolate a particular gene to obtain information about its gene product and eventually to manipulate the gene. In general, genetically favourable traits that are easily scored, will rapidly be incorporated into new varieties. As breeding progresses, however, we have to deal with traits that are more and more difficult to score and analyze. This category includes most quantitative traits. These are often controlled by genes at several loci, the basic function of which is unknown. The use of the increasing number of phenotypically "neutral" DNA markers (RFLP and RAPD) produces more detailed linkage maps. Combined with statistical methods, this has made it possible to map and identify a number of loci for quantitative traits.

Linkage maps of the barley genome were constructed based on the analysis of chromosome-doubled haploid offspring lines from two crosses, 'Vogelsanger Gold' × 'Alf' and 'Vogelsanger Gold' × 'Tystofte Prentice'. Lines from both crosses have been grown in replicated field experiments to record a number of quantitative traits.

Chromosome 1 Chromosome 2 Chromosome 5 Chromosome 6 Chromosome 7

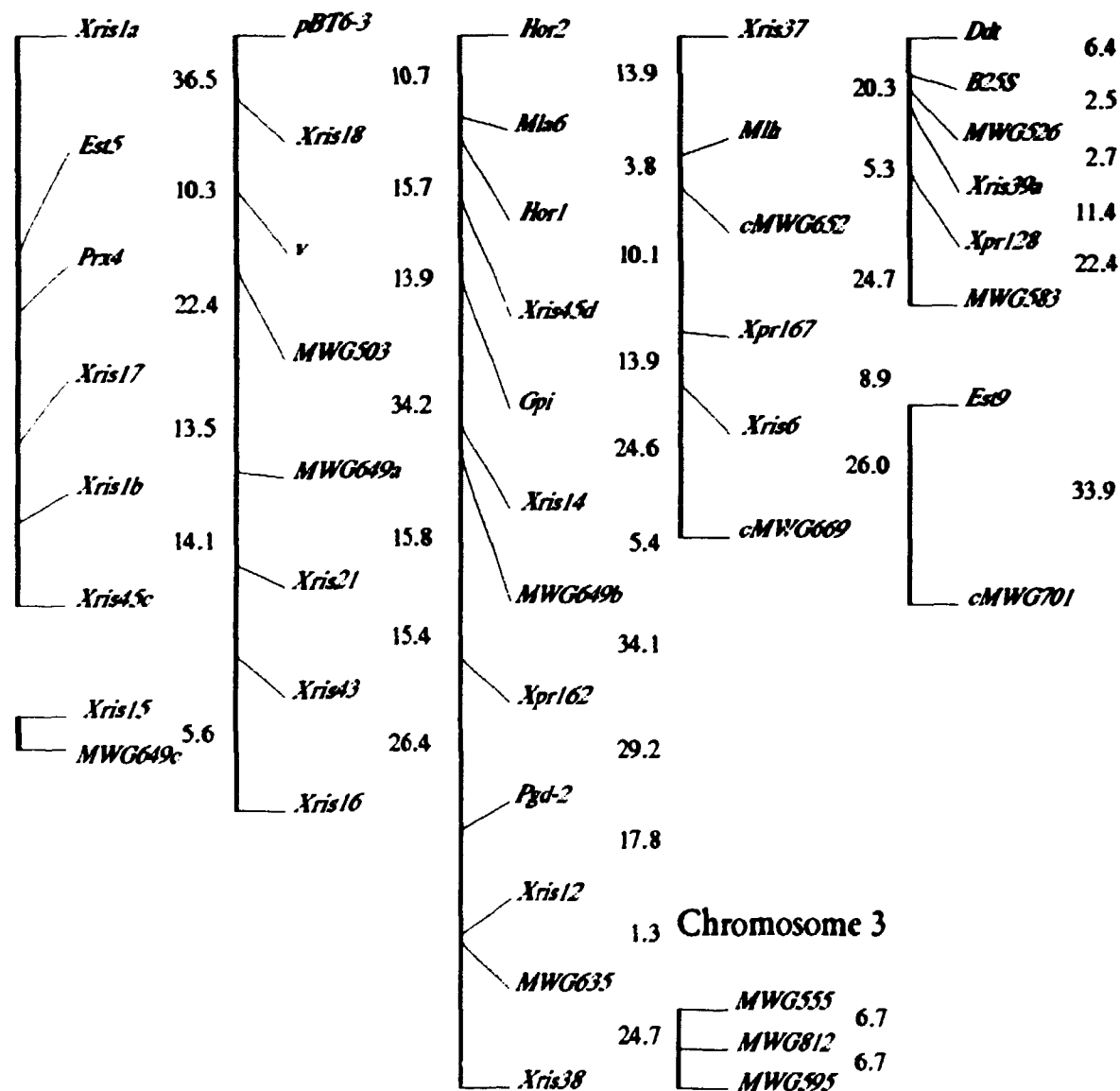


Fig. 3.1. Preliminary linkage map of barley based on conventional markers and RFLPs in chromosome-doubled haploid lines from the cross 'Vogelsanger Gold' x 'Tystofte Prentice' (distances between loci are in centimorgans).

The first cross, 'Vogelsanger Gold' × 'Alf', has until now been scored for 66 markers. Fourteen of these are conventional markers, including a leaf stripe-resistance locus and the "Laevigatum" powdery mildew-resistance locus *MiLa*. Both loci have been localized to chromosome 2. Forty-six of the markers are RFLP markers and 6 are RAPD markers. Linkage analysis of the segregation of the 6 RAPD markers indicates that this technique can be used in barley. Thus, we will be able to increase the number of markers considerably in a relatively short span of time, since the production of RAPD markers is much faster than the production of RFLPs.

Sixty-four of the 66 markers, so far studied, were localized to chromosome. More markers were mapped to chromosomes 2, 5, and 6 than to the other barley chromosomes. Some of the chromosome 2 data are under publication (Giese *et al.*, 10). Significant associations to grain yield have been found with markers on chromosomes 2, 3, 5, 6 and 7 identifying QTLs on these chromosomes. Our data suggest that the effect of the QTL alleles may not be additive. This influences the recombination estimates. Therefore, adequate transformations of the yield data are important. Furthermore, we have indications that linkage between two QTLs may occur in the repulsion phase (*i.e.*, the effect of alleles in two QTLs affecting the same quantitative trait goes in opposite directions in the two parents). So far, this has caused unsolved calculation problems.

The second cross, 'Vogelsanger Gold' × 'Tystofte Prentice', has been scored for 53 markers, 8 conventional ones and 41 RFLPs. The markers are distributed equally on 6 of the 7 barley chromosomes. Chromosome 4 has only 1 marker.

In the latter cross, the concentration and accumulation of phosphorus and nitrogen are also studied. The morphological marker *v* (2-rowed / 6-rowed), located on chromosome 2, has shown correlation with the variation of the concentrations of N and P in grain and straw with the exception of N% in straw. The correlation can be explained by variation in grain and straw yield. An association between N% and P% in straw, and an RFLP marker on chromosome 5, suggests that one or two QTLs are located on this chromosome. Further, a putative QTL for N% in grain is linked to a RFLP marker with an as yet unknown location. A preliminary analysis of grain-yield, thousand kernel weight, and

heading date based on data from two-year yield trials showed that, in some cases, the same QTL could be detected in both years. In other cases, a significant interaction between year and QTL was found.

Cytogenetics of Barley and Related Species

Knowledge of the physical linkage maps of organisms, indicating the cytological positions of gene loci on each chromosome, is a necessary prerequisite for a future, directed exploitation of modern plant breeding techniques, such as the transformation of one species with a specific gene from an alien species. *In situ* hybridization of DNA probes to chromosomes is a valuable tool for identifying the position of gene loci cytologically. To improve the resolving power of the method, the synchronization of mitoses, the making of preparations, and the hybridization technique have been optimized. Using a wheat rDNA probe (pTA71) on chromosome preparations of cultivated barley, it has been possible to detect 10 hybridization sites, 4 large ones corresponding to the NORs on chromosomes 6 (61) and 7 (51) and 6 minor ones, the two larger of which are located intercalarily in the short arms of chromosome pair no. 5 (11). The same rDNA probe was hybridized to meiotic metaphase I in pollen mother cells of *Hordeum marinum* ssp. *gussoneanum* (4x) × *Secale cereale* (rye) hybrids having up to 7 *H. marinum* ring bivalents. It identified a large and a smaller hybridization site located similarly in the same bivalent. This further strengthens the idea of an autopolyploid origin of *H. marinum* ssp. *gussoneanum* (4x) (Linde-Laursen *et al.*, 10).

Giemsa C-banding produces patterns of transverse stripes, "C-bands", on the chromosomes of plant species. This has been used to identify chromosomes and chromosome arms of species of the Triticeae (Poaceae), *e.g.*, cultivated barley and wheat. *Psathyrosachys* is an Asiatic genus rather closely related to barley (*Hordeum*). The Giemsa C-banded karyotypes of 2 taxa of *P. fragilis* and 2 taxa of *P. lanuginosa* not previously studied cytologically, showed a close overall resemblance to each other and to those of *P. juncea* (Russian wild rye) and *P. fragilis* (earlier investigated), indicating a close relationship. The genus *Elymus* contains many species with characters of interest for agriculture. The

Giemsa C-banded karyotypes of 3 South American *Elymus* species showed a wide variation, indicating that they are not very closely related.

Application of RAPDs for Genome Mapping in Norway Spruce and Cotton

Worldwide construction of genetic linkage maps is carried out on important forest trees. In 1991, mapping of the genome of Norway spruce (*Picea abies*) was initiated to study genetic mechanisms and to search for linkage between molecular markers and QTLs of practical importance such as health, productivity and wood-quality. The use of RAPD markers appears, at present, to be by far the most effective method. The segregation and recombination analysis for identifying linkage groups can be performed at the haploid seed endosperm – the megagametophyte. Up to now, ~ 100 PCR-amplification products have been identified as possible Mendelian segregating loci. The aim is to map about 200 marker loci.

The possibilities of establishing a molecular marker system in cultivated species of *Gossypium*, i.e. cotton, based on the RAPD technique, were surveyed. Twelve lines of *G. hirsutum* (tetraploid), 1 line of *G. barbadense* (tetraploid) and 1 line of *G. arboreum* (diploid) were screened for polymorphisms. The plant material originated from India and Thailand. Nineteen oligonucleotide primers were tested in preliminary experiments. The results indicated virtually no polymorphism within lines of *G. hirsutum*, low levels of polymorphism between lines of *G. hirsutum* and moderate levels of polymorphism between the 3 species of *Gossypium*. The use of the polymorphic RAPD bands as RFLP probes revealed a banding pattern consisting of low copy sequences, but did not show any polymorphism between the lines tested.

3.2 Proteins of Importance for Stress Resistance and Quality

Barley Peroxidases

Peroxidases are involved in many biological processes in plants. They are, 1) tissue-specific, developmentally regulated, or modulated by environmental stress factors, and 2) involved in

the biosynthesis of the cell walls, in the regulation of the auxin level, and in response to microbial attack. Barley peroxidases are studied at the biochemical, physiological, genetic and molecular levels to delineate the biological function of the individual enzymes.

The cloning of genes, which code for the barley peroxidases, aims at a characterization of sequences that specify the tissue-specific expression and are sensitive to stress factors. Well-characterized genes will be used to construct vectors for genetic transformation of the model plant *Arabidopsis thaliana* which is currently being used because the transformation of barley is still difficult.

Two new members of the BP 2 peroxidase gene family were isolated and sequenced. The nucleotide sequences of λ Ppx6.2 and λ Ppx6.3 isolated from barley line NK 1558 are 98.8% identical with λ Ppx6.1 (Theilade and Rasmussen, 9.1). The latter originates from a cross between the varieties 'Villa' and 'Hiprol'. The promoter in all three genes showed a putative abscisic acid-response element, 5'-GTACGTGTC, 115 kb upstream from the start codon.

A genomic clone encoding the barley powdery mildew-inducible peroxidase, pcD1311, was isolated and is now being analyzed by nucleotide sequencing (Rasmussen *et al.*, 10b). The isolation and the preliminary sequencing results were established during an M.Sc. programme (Quarcoo, 11.2). A full-length clone coding for the barley β -hordothionin, known to be toxic to microorganisms, was isolated (Rasmussen, 10). The inhibitor of insect α -amylase CMA has also been cloned (Rasmussen and Johansson, 9.1). Both of the latter proteins are located in the endosperm ~ 15 days after flowering and thus adding to the number of proteins which could have a protective role during seed germination.

Callose Synthesis in Barley Leaves

Barley and other plants respond to pathogens or physical injury by forming cell wall appositions (papillae) in the epidermal cell layer between the plasma membrane and the cell wall. The main component in the papillae is the polysaccharide callose, which consists of 1,3- β -linked glucose units. Callose is synthesized by the plasma membrane-bound 1,3- β -D-glucan synthase from the substrate, UDP-glucose.

Plasma membranes were concentrated by partition in a polyethylene glycol 3350/Dextran T-500 aqueous polymer two-phase system. The plasma membrane-bound callose synthase was dependent on uridine-diphospho-glucose (apparent $K_m = 0.39$ mM) and was activated by Ca^{2+} , digitonin, cellobiose and polyamines. The enzyme was inhibited by uridinediphosphate and uridinetriphosphate. Glucano-hydrolase digestion of the *in vitro* product showed that it was a 1,3- β -linked polysaccharide. Following inoculation with powdery mildew, *Mlo* mutants of barley are known to produce larger papillae at an earlier stage than those in the mother varieties. It was shown that this was not due to *de novo* synthesis of callose synthase (Pedersen, 11.1). The activation of the synthase could be an effect of changes of the Ca^{2+} concentration in the cell. Non-denaturing polyacrylamide gel electrophoresis separated the callose synthase from most other plasma membrane proteins. Sodium dodecylsulfate gel electrophoresis of the proteins in the callose synthase activity-stained zone revealed six dominant polypeptides of M_w s 36, 52, 54, 60, 70 and 94 kDa, respectively. The polypeptide profiles of the barley callose synthase are in agreement with the results of previous attempts to isolate the polypeptide(s) involved in callose synthesis in dicot plants (Pedersen, 11.1).

Quality of Barley Grain Protein

The storage protein (hordein) from barley grain is poor in the essential amino acid lysine. The nutritional quality of barley may be improved by an increased synthesis of proteins rich in lysine. Five high-lysine barley mutants and a control were yield-tested in a field experiment. The mutants are being analyzed for the contents of lysine and other amino acids. Protein Z is a barley seed storage protein with a favourable lysine content. We are aiming to modulate the barley seed protein composition, thus balancing the nutritional quality of the grains. This is done by increasing the amount of protein Z by two different approaches. In one, we try to duplicate locus *Paz4* by classical genetic methods. In the other, we isolate protein Z genes and attempt to increase the number of gene copies by genetic transformation. Both procedures imply that the increased number of gene copies will lead to an

increase in protein Z production and lysine content in the barley grain. The work is carried out in collaboration with Dr B. Eggum, Foulum, and Dr J. Heigård, DTH.

A system to produce transgenic barley is still lacking. Gene analysis is, therefore, carried out in the model plant *Arabidopsis thaliana*. The promoter from a protein Z gene is dissected and inserted in a Ti vector in front of the GUS reporter gene. Roots of *A. thaliana* were infected by *Agrobacterium tumefaciens* carrying the vector construct. This will facilitate the identification of organ-specific elements in the protein Z promoter. Besides the impact on seed quality, protein Z is of evolutionary and functional interest. A gene coding for a new plant serpin, protein Zx, consists of two exons interrupted by a 971 basepair intron. Protein Zx is 79% identical with protein Z4 and approximately 30% identical with members of the super family of mammalian serine proteinase inhibitor (Rasmussen, 10).



Fig. 3.2.1. *Arabidopsis thaliana*

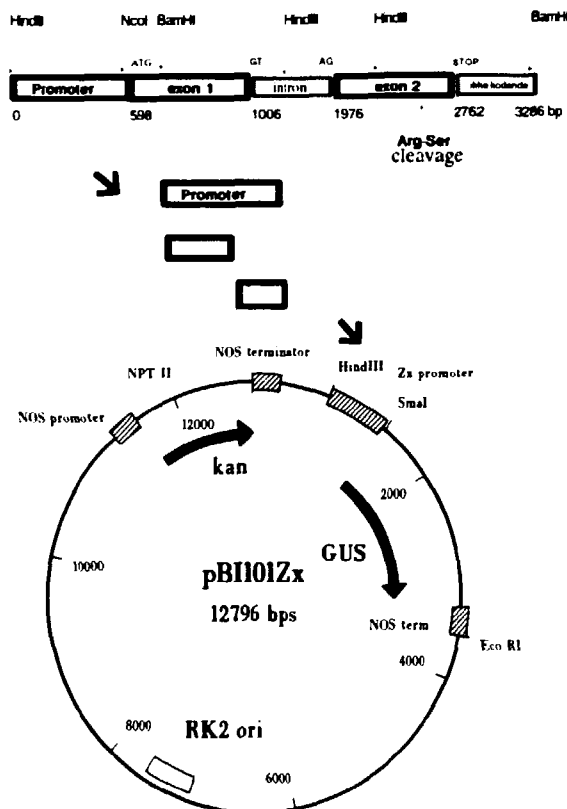


Fig. 3.2.2. Strategy for promoter analysis of protein Z.

3.3 Gene Dispersal in Relation to Risk Assessment

Interaction between GMPs and the Environment

Release and marketing of genetically modified plants (GMPs) demand an assessment of the risks of gene transfer from these plants to other plant species by hybridization. Presently, transgenic oilseed rape (*Brassica napus* L.) is being released in various field experiments (Madsen and Jørgensen, 9.1). Therefore, introgression between oilseed rape and the wild species *Brassica campestris* and *Brassica juncea* is being studied within the Danish Biotechnology

Research Programme: "Risks in relation to release of gene-modified plants". The project is carried out in collaboration with the firms Mari-bo Seed and Monsanto Europe.

Gene Dispersal from Oilseed Rape to *Brassica campestris*

Field experiments with mixed populations of the two species were carried out. Seventy plants of *Brassica campestris* (representing four Danish collections) were interspersed in oilseed rape fields. Seed production was 0-950 seeds per *B. campestris*-mother plant. The seeds germinated easily. Five hundred offspring plants were examined by enzyme electrophoresis and 90% of these were hybrids, as revealed by enzyme patterns. The hybrid status of these plants was confirmed by morphology, RAPD profiles and by chromosome counts. Natural hybridization between oilseed rape and *B. campestris* was thus found to occur frequently.

Interspecific hybrids were also identified in a natural population of *B. campestris* from Zealand by morphology, enzyme electrophoresis, RAPD technique and chromosome number. In this material, the frequency of hybrids was 27%. From 20 Danish populations of *B. campestris*, individuals have now been collected for analysis of introgression from oilseed rape.

Gene transfer from oilseed rape (genome AACC, $2n=38$) to *B. campestris* (genome AA, $2n=20$) in natural environments is dependent on the ability of the hybrids to backcross to the wild species. Therefore, mixed populations of hybrids and *B. campestris* were analyzed. The hybrids formed from 0-750 seeds each. Some of the resulting plants were similar to *B. campestris*; their chromosome number being $2n=20$, and they were found to be fertile. The remaining offspring plants had chromosome numbers varying from $2n=21$ to $2n=29$.

Laboratory crosses were carried out to identify the parts of the oilseed rape genetic material which are transferred to *B. campestris*. Fifty-two genotypic combinations were examined. They revealed that more hybrids were produced in the cross *B. napus* (♀) × *B. campestris* (♂) than in the reciprocal combination. The hybrids were backcrossed to the wild species. The offspring are being characterized using chromosome-specific RAPD markers.

Gene Dispersal from Oilseed Rape to *Brassica juncea*

Preliminary results from mixed populations of oilseed rape and *Brassica juncea* indicate that the species hybridize in the field.

Six transgenic oilseed rape lines, having transgenes providing tolerance to the herbicide "Roundup" (glyphosate resistance) were crossed with *B. juncea* to obtain hybrids. The hybrids are backcrossed to the wild species. Transfer of the transgene to F_1 and BC_1 plants will be verified by PCR with a primer specific to the transgene. Sixty-nine crosses between *B. juncea* and the 6 transgenic oilseed rape lines were carried out. The results from 34 of the crosses (some of the offspring have yet to be harvested) showed that the cross-compatibility is ~ 10 times greater when *B. juncea* was used as the female parent than that in the reciprocal cross. Seeds harvested from the latter combination are often without embryos.

To identify which of the 9 chromosomes from the C-genome of oilseed rape are transferred to the BC_1 plants, 22 RAPD-primers, found to mark specific C-chromosomes were tested on our plant material. Twelve of these RAPD-primers produced 18 markers on 8 of the 9 C-chromosomes in the 6 transgenic oilseed rape lines, in the male sterile lines (11-7060 and 11-7094) and in the varieties 'Line' and 'Topas'.

3.4 The Barley/Powdery Mildew System

Chromosome "walking" in the Barley Powdery Mildew Fungus

The specific interaction between barley and the obligate parasitic fungus, *Erysiphe graminis* f.sp. *hordei* (barley powdery mildew), is only understood at the genetic level as conforming to the gene-for-gene hypothesis. The function of the specific avirulence/virulence genes in the fungus and of the resistance genes in the plant, determining whether a compatible or an incompatible interaction will take place, is unknown. One way to identify the individual genes, is to clone them and subsequently to characterize them at the molecular level. Knowledge of the genetics facilitates a chromosome "walking" strategy to iso-

late the specific genes. The present project is attempting to isolate an avirulence gene in the fungus by this means.

A prerequisite for the chromosome "walking" strategy is the construction of dense genetic maps based on molecular markers such as RFLPs or RAPDs. A linkage map of the fungal genome has now been developed. It includes 45 RFLP markers and 18 RAPD markers. The closest linkage between a marker and an avirulence gene is 9cM. Estimation of the genome size of the fungus, based on the frequency of random linkage between two arbitrary markers, is in the region of 50 Mb. An estimation, based on dot blots using a single copy sequence arrives at a similar estimate. This corresponds to genome sizes reported for other obligate parasitic fungi.

An attempt was made to insert the entire fungal genome into YAC vectors, which may contain up to 1 Mb of foreign DNA. 700 YAC clones were constructed and examined for the presence of fungal mildew DNA. The YAC clones have inserts in the range of 50 to 200 kb and will be further characterized and manipulated. Based on the genome estimates, a recombinational distance of 1cM should correspond to approximately 20 kb. A more dense chromosome map with markers that link more closely to an avirulence gene is still required in order to develop probes which can successfully select YAC clones containing such a gene.

The genome contains a large amount of repetitive DNA which may hamper the "walking" strategy. A repetitive element was sequenced and shown to resemble SINE (short interspersed repetitive element) identified in mammals and thought to have arisen by virus-aided reverse transcription (Rasmussen *et al.*, 10a). The element is present in very high copy numbers in the fungus and is dispersed throughout the genome. Two similar elements with different sequences were identified, one of which is found in very close proximity to the β -tubulin gene. The elements may increase the variation in the fungus by unequal crossing over and by interrupting coding sequences. Primers from the predominant SINE were designed and have proved useful as a tool in the identification of positive YAC clones.

To verify that an avirulence gene has indeed been cloned, a transformation system is necessary. Constructs, containing reporter genes and

fungus promoter regions, were designed. No background activity of the reporter genes was found in the fungus. The only feasible way to transform an obligate parasite is to use particle gun acceleration. This will be attempted.

Genetics of Virulence in the Barley Powdery Mildew Fungus

A thorough knowledge of the genetics of the pathogen, as well as that of the host, is essential for understanding the dynamics of the powdery mildew disease. Four mildew isolates, two 'Laevigatum'-virulent and two avirulent ones, were crossed and three progeny populations tested for segregation in virulence. On 'Laevigatum' resistant barleys carrying the resistance gene *Mla*, the segregations were close to a 1:1 ratio indicating the presence of a single virulence locus with two alleles, *VLa* and *ALa* (Jensen and Jørgensen, 10). These data combined with data from crosses of 'Laevigatum'-resistant and -susceptible barley lines, strongly suggest that the 'Laevigatum' host-pathogen interaction is conferred by a single gene pair in the host and one pair in the pathogen in accordance with the gene-for-gene system.

Simultaneous testing of the mildew progeny isolates showed 1:1 segregation ratios in virulence loci *Va1*, *Va3*, *Va10*, *Va12*, *Vat*, *Vra*, *Vg*, and *Vh*. None of these eight loci showed any sign of close linkage with locus *VLa*.

Mlo Aggressiveness of the Barley Powdery Mildew Fungus

A laboratory-derived Mlo-aggressive powdery mildew isolate, a non-aggressive one and two near-isogenic barley lines, with and without powdery mildew-resistance gene *mlo5*, were used as a model system to study the effect of the Mlo aggressiveness of the pathogen and the Mlo resistance of the host. The powdery mildew aggressiveness was expressed as an approximately 5-, 75-, and 1,000-fold increased ability to successfully infect subsidiary, short, and long epidermal plant cells, respectively, supported by a reduced latent period. The Mlo resistance was expressed by a reduced, a strongly reduced, and a virtually unsuccessful infection in the three cell types, respectively. The latent period was also substantially extended. The number of mildew

colonies, formed per 100 viable conidia applied, was about 25 on non-resistant barley. On Mlo-resistant barley, the figures were 0.05 for the non-aggressive isolate, and 3 for the aggressive isolate. The ability of the Mlo-aggressive isolate to grow on non-resistant barley was not significantly reduced (Andersen and Jørgensen, 9.1).

The discovery, characterization and exploitation of Mlo resistance in barley in Europe, and the possible durability of Mlo resistance were reviewed (Jørgensen, 10d).

Population Studies of the Barley Powdery Mildew Fungus

Changes in frequencies of virulence genes in the aerial population were recently described mathematically by various models in collaboration with Dr Mogens Hovmøller, The National Institute of Plant Protection, and Dr Lisa Munk, The Royal Veterinary and Agricultural University (e.g., Hovmøller *et al.*, 9.2a, b, 10a, b). The most recent model (Hovmøller *et al.*, 9.2b) tries to answer the question: Can virulence genes decrease in frequency in ways other than by direct selection against them? The answer is - yes. In the case of negative gametic associations (gametic disequilibrium) between two virulence genes, strong selection favouring one will result in a decrease in the frequency of the other. The models predict that this situation occurs when varieties with different resistance genes are grown in sequence over a large area. Actual survey data confirm the prediction.

Multigene Families of Powdery Mildew Resistance Genes in Locus *Mla* in Barley

Experience and data, accumulated over several years, suggest that some of the named genes at locus *Mla* on barley chromosome 5 are closely linked to "additional" mildew-resistance genes. One more case was recently disclosed in the barley variety 'Sultan'. Progeny from a cross between 'Sultan' which possesses gene *Mla12* and the susceptible variety 'Pallas', showed that 'Sultan' possesses two closely linked, semi-dominant resistance genes, *Mla12* and an "additional" one that is tentatively designated *Mla(Em2)* (Jørgensen, 9.1b). Other data, from a genetic analysis of the virulence genes present in two powdery mildew isolates imply that the barley

variety 'Welam' with gene *Mla9* and lines 'Iso 26 R (Long Glumes)' and 'Line A-7' both with gene *Mla7* each possess one or two new "additional" resistance genes closely linked to the named *Mla* genes.

A compilation of these and former data (Jørgensen, 9.1b) indicates that there are at least 12 multigene families each comprising a named *Mla* gene and one or more closely linked "additional" mildew-resistance genes at locus *Mla* in barley. This may explain why genes at locus *Mla* have frequently been selected by plant breeders as promising sources of resistance.

Agronomical Traits of Mlo Resistant Barley

Until 1992, seventeen barley varieties with the effective Mlo resistance to powdery mildew were approved on the European variety lists and several new ones are now under development. The 17 varieties have been traced back to four different Mlo donors. Ten of these were studied for presence of disease attack and for yield in field experiments over a 2-year period (Jensen, 9.2). The Mlo varieties were highly resistant to powdery mildew, but considerable variation was found in their resistance to barley leaf rust and scald. Yield losses in untreated, compared to fungicide-treated Mlo-resistant varieties, were correlated with the degree of leaf rust resistance.

3.5 Barley Leaf Stripe

Two series of chromosome-doubled (DH) lines of spring barley were tested for resistance to leaf stripe and to powdery mildew ('Laevigatum'-resistance) to further clarify the inheritance of these resistances.

Both DH populations segregated in a 1:1 ratio for leaf stripe and powdery mildew reaction. In addition, the combined segregations of both traits showed that the two resistance genes are linked with recombination percentages of 20.2 and 19.0, respectively, in the two populations. Linkage of these genes to RFLP markers, with known chromosomal location show, that they are located on barley chromosome 2 (2 I) (Giese *et al.*, 10).

Tests with leaf stripe isolates from various countries showed that the leaf stripe-resistance gene from 'Vada' was effective against all but one isolate, from Syria. Moreover, a test of se-

veral varieties with a high level of resistance to leaf stripe showed interactions among varieties and isolates thereby proving the existence of other sources of resistance than that in the cultivar 'Vada' (Thomsen, 10).

3.6 Interaction between Crop Plants and Weeds

It is generally assumed that weeds reduce the production of crop plants. This is not always the case, however, as our studies of mixtures of wheat and corn cockle (*Agrostemma githago*) demonstrate. The effect of corn cockle on wheat was studied in pots in a growth chamber (Søgaard and Doll, 9.1) and in the field, in collaboration with Dr Bodil Søgaard, Royal Veterinary and Agricultural University. Nine wheat plants were grown to maturity with 0, 3, 6 or 9 corn cockle plants in pots. Both total biomass and seed production of wheat significantly increased in the pots containing corn cockle. Further, there was a clear influence of corn cockle on wheat within the pots containing both species. Wheat plants, sown in the same hole as a corn cockle plant, were 20 to 50% larger than wheat plants standing alone.

A clear effect of corn cockle on wheat has also been observed in the field. A significantly higher wheat grain production was obtained when 25 or 50 seeds of corn cockle were sown with wheat at normal density (400 seeds per m²).

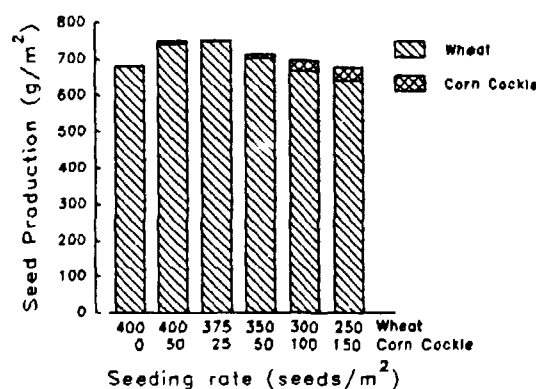


Fig. 3.6 Seed production of wheat grown in a mixture with corn cockle in the field.

These results illustrate that a crop plant may react positively to the presence of weed. Corn cockle was previously found to be a difficult weed, because the relatively large, poisonous seeds were harvested together with the seeds of the cereal. It is possible that wheat and corn cockle have had a close interaction for a long period of time, during which this weed plant has coevolved with cereals.

3.7 Seeds for Educational Purposes

The Plant Biology Section offers seed samples of barley and other plant species for teachers in biology. The samples illustrate Mendelian gene-segregations and the effect of ionizing radiation on various plant species. In 1992, a total of 1417 individual "experiments" were distributed.

4 Plant Nutrition and Nutrient Cycling

The objective of the projects in this research area is to provide a better understanding of those biological and chemical processes in the soil-plant-atmosphere environment, which are important for plant nutrition. The processes must be utilized and managed in such a way that nutrient uptake by plants is optimized. This will lead to a reduced requirement for energy-consuming fertilizers and to a smaller loss of nutrients to the aquatic and atmospheric environments.

Mutualistic symbioses between plant roots and microorganisms are directly involved in the nutrient uptake by plants. Nitrogen is provided by *Rhizobium* bacteria, fixing atmospheric nitrogen in symbiosis with legumes. The soil-plant transport of mineral nutrients, especially phosphorus, is mediated by vesicular-arbuscular mycorrhizal (VAM) fungi in symbiosis with most plant species.

The general soil microflora is of fundamental importance to the mineralization of nutrients contained in plant residues and in farmyard manure. Special attention is directed towards

processes involved in the cycling of nitrogen, which is a major nutrient source for plants, but also a risk to the environment by leaching as nitrate and losses as volatile nitrogen compounds.

The study of these research topics will provide the basic information needed for increasing the economic and ecological sustainability of plant production.

4.1 Nitrogen in Soil and Plants

The aim of the present research is to elucidate the intricate biological, physical and chemical processes involved in the cycling of nitrogen (N) in the soil-plant-air-system. At present, the main emphasis is on the cycling of N in crop residues, farmyard manure and mineral fertilizers. Plant materials and organic manure labelled with ^{15}N are used to study the dynamics of mineralization-immobilization turnover of the organic N in soil and the uptake of mineralized N by subsequent crops.



Fig. 4.1.1. Crop residues are an important factor in maintaining soil organic matter and recycled residue nutrients contribute to the nutrition of succeeding crops.

Dynamics of Pea Residue N turnover

In order to optimize the utilization of crop residue N by subsequent crops and to reduce the losses of N to the aquatic and atmospheric environments, it is necessary to improve our knowledge on the dynamics of the turn-over of plant residue N in soil.

The dynamics of ^{15}N -labelled mature pea residue N turnover in soil were studied in two experiments, over a 3-year period, in unplanted soil in the field. During the initial 10 days of

decomposition the organic- ^{15}N in soil declined with 14–28%. Simultaneously, 14–22% of the ^{15}N label was immobilized in the microbial biomass, accounting for 66–100% of the increase in the biomass N. During the following years, the microbial biomass ^{15}N declined faster than the total residual organic ^{15}N . The pool of potentially mineralizable ^{15}N , determined by an anaerobic incubation procedure, varied with time in a way which was similar to the biomass ^{15}N , but the size of the potentially mineralizable ^{15}N pool was much smaller than the biomass ^{15}N pool.

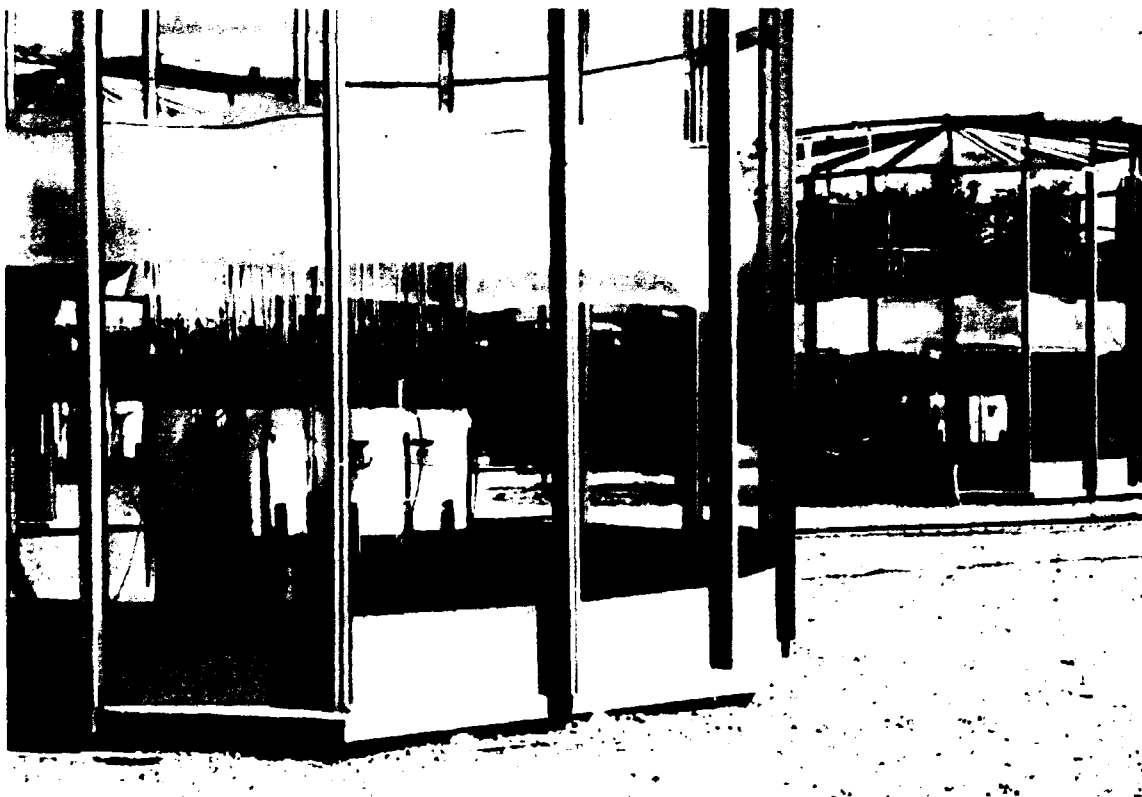


Fig. 4.1.2. Absorption of NO_2 by plants is studied in open-top chambers using ^{15}N isotope techniques.

After 90 days of decomposition, 10% of the ^{15}N input was unaccounted for, but unaccounted for ^{15}N increased to 20–30% after 2 to 3 years of decomposition in unplanted soil. Nitrogen unaccounted for was probably lost by denitrification, since the soil had high nitrate concentrations during the growing and autumn months.

The two experiments were carried out with either ground or chopped residues. The results indicated that the residue particle size may significantly influence the initial turnover of pea residue N.

Labelling of Sheep Manure with ^{15}N

A sheep was fed with ^{15}N labelled ryegrass hay over a period of 9 days to obtain ^{15}N -labelled animal manure. After this period of feeding, the total faeces N contained 3.70 atom % ^{15}N excess, compared with 4.52 atom % ^{15}N excess in the feed. The easily decomposable faeces N fraction was less labelled (2.89 atom % excess) than the slowly decomposable fraction. The ^{15}N enrichment of inorganic N, formed after incubation of the faeces in sand, did not change signi-

ificantly during 32 weeks of decomposition. 20–25% of the faeces N was water-soluble, and this N fraction had a higher ^{15}N enrichment than the total faeces N, indicating that a part of the water soluble N was non-digested in digestible feed N. The major part of this fraction was organic N, which was resistant to decomposition. The faeces contained only small amounts of NH_4 , with a ^{15}N enrichment similar to that of the mineralized N. The labelled faeces, N obtained after a few day's feeding, could be divided in two N pools: A decomposable N fraction (57%) with a ^{15}N enrichment as the first mineralized N and a very slowly decomposable N fraction (43%) with a ^{15}N enrichment as the feed.

Plant Residue Decomposition and Phosphorus Availability

Decomposition of crop residues in soils are influenced by the content and availability of mineral nutrients essential for microbial growth. It has been suggested that deficiency of mineral nutrients is partly responsible for slow decomposition of crop residues in soil. It is well known that incorporating cereal straw into a soil may reduce the N available to a succeeding crop, because soil N is temporarily immobilized in the soil biomass. A similar effect on P-availability may occur since the concentration of P in cereal straw is low compared to the P concentration in the microbial biomass. The availability of P may be a rate-determining factor for the decomposition of cereal straw, but experimental evidence is rather scarce. Investigations on the effects of P supply on the rate of mineralization of wheat straw have been initiated.

Nitrogen Accumulation in Winter- and Spring-Types of Cereals and Oilseed Rape

A crop rotation experiment was initiated in 1985 to measure the N uptake by winter- and spring-types of barley, oilseed rape, wheat and rye, grown in this sequence in field plots receiving increasing amounts of N fertilizer. By comparing the amounts of N accumulated by winter- and spring-types, the potential of winter crops reduction of N-leaching was determined.

Preliminary results were reported (RISØ-R-624), indicating that winter types of wheat and barley require 40–60 kg N/ha more than the

corresponding spring types for optimum yields. Since the area covered with winter crops (mainly winter wheat) has increased by almost 400,000 ha in recent years, it can be calculated, that the total amount of N required by crops has increased by about 20,000 t N. The leaching of N from arable land presumably has been reduced by a similar amount, since the total supply of N-fertilizer has been almost constant for several years.

4.2 Root-Microbe Symbioses

Symbiotic associations between plants and soil microorganisms are of major importance to plant nutrition and consequently to the sustainability of plant production. Our research in this field involves the physiology and ecology of the legume-*Rhizobium* symbiosis and of vesicular-arbuscular mycorrhizas, symbioses between plants and fungi.

Symbiotic Nitrogen Fixation

The symbiosis between legumes and rhizobia bacteria enables the plant to grow on the nitrogen fixed by the bacteria as the sole nitrogen source. The bacteria inhabit characteristic nodules on the roots of the host plant. Within these nodules, the bacteria are dependent on the plant for a supply of carbon as a source of energy for their metabolism. In return, the bacteria supply the plant with nitrogen which they fix into ammonia from atmospheric N_2 .

The establishment and maintenance of an effective symbiosis between the plant and the microorganism are based on an extensive molecular communication between the two. Signals and metabolites are continuously being exchanged between the symbionts to regulate the process. Nitrogen-fixing bacteroids, in mature legume nodules, are enclosed in a plant-derived peribacteroid membrane (PBM). The two symbionts are thus separated, not only by the PBM but also by a compartment, the peribacteroid space (PBS), which lies between the PBM and the bacteroid outer membrane. Signals and nutrients exchanged between the symbionts have to pass the PBM, PBS, and the bacteroid membrane. We have demonstrated an exchange of amino acids and organic acids across the mem-

brane interface in pea root nodules (Rosendahl *et al.*, 1991). The PBS may be involved in regulating this exchange of metabolites.

We have devised techniques to isolate the PBS from pea root nodules. Our analyses of the proteins in the PBS, demonstrate that it contains a unique set of proteins some of which are glycosylated. Our results indicate that the PBS contains a specific isoform of the enzyme malate dehydrogenase and a tissue specific isoform of aspartate aminotransferase. Both are enzymes of key functions in the carbon metabolism and may be involved in the observed exchange of amino acids and organic acids.

Mycorrhizas and Plant Nutrition

It is desirable to reduce the agricultural use of P fertilizers, which are produced from limited resources of rock phosphates. High P contents in arable soils increase the risk of eutrophication of the aquatic environment, and P fertilizers contain small amounts of toxic heavy metals, especially cadmium.

Vesicular-arbuscular mycorrhizas (VAM), common symbiotic associations between plant roots and soil fungi, contribute to an efficient utilization of soil and fertilizer P. The formation of VAM, however, is impaired by high soil-P levels. Reduced P fertilizer may, therefore, be expected to be counterbalanced by a concurrently increased P supply via the VAM fungi.

A field experiment was carried out to determine the amount of P fertilizer required to obtain maximum growth of mycorrhizal and non-mycorrhizal barley, subterranean clover, flax and leek. The soil contained 20 mg kg⁻¹ soil of 0.5 M NaHCO₃-extractable P, ~ 55% of the average level in Danish arable soils. Plots were supplied with superphosphate at 0, 75, 150 or 300 kg P ha⁻¹, and half of each plot was fumigated to eliminate the VAM fungi. Oil-seed rape, a non-host, was included as a control for treatment effects unrelated to VAM fungi.

The growth of non-mycorrhizal clover, leek and flax responded markedly to increasing P levels, whereas mycorrhizal plants were much less affected by fertilizer level (Fig. 4.2.1). Growth of barley and oil-seed rape was unaffected by P fertilizer both in fumigated and unfumigated soil. The effect of VAM corresponded to as much as 100 and 150 kg P ha⁻¹ in leek and

flax, respectively, and VAM colonization was obviously crucial for the maintenance of near-maximum growth of several crop species at moderately low levels of plant available soil P.

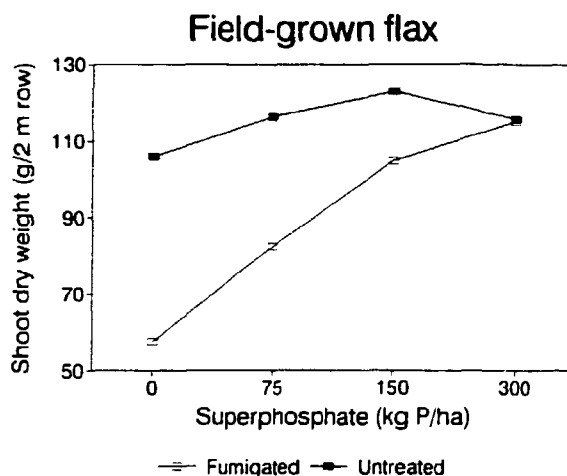


Fig. 4.2.1. Phosphorus response curves for non-mycorrhizal (fumigated soil) and mycorrhizal flax (untreated soil) harvested 63 days after sowing.

We have developed a novel method to directly measure the relative contribution of VAM fungi and roots to total P uptake by plants. Dual labelling with ³²P and ³³P was used in combination with two labelling compartments: One was a root-free hyphal compartment with ³²P-labelled soil, the other was identical, but contained both roots and hyphae in ³³P-labelled soil. The contribution of the roots was measured as the difference between amounts of ³³P and ³²P in the plant.

The hyphal transport of ³²P differed markedly between three isolates of VAM fungi in association with cucumber, but these differences were not reflected in the uptake of ³³P from the compartment also containing roots. A low ³²P transport by the hyphae was associated with a marked increase in the ³³P uptake by the roots, whereas the transport of ³²P and ³³P was similar in the association with the most efficient fungus. This represents the first demonstration of a marked influence of VAM fungi on P uptake by roots. Feed-back regulation via local changes in P concentrations of root cells or root tissues may be responsible for the observed interactions.

The fungus with the most efficient P transport was used in a similar experiment under field conditions, with pea as the host. The results

confirmed that the micro-symbiont may be responsible for the major proportion of P uptake.

It is unknown to what degree interfungal differences in P transport capacity are reflected in the efficiency of populations of VAM fungi producing similar levels of root colonization. This was investigated in collaboration with researchers at the University of Western Australia. Subterranean clover was grown in undisturbed soil cores from five pasture sites. A mesh bag with ^{32}P -labelled soil was inserted in the centre of each core. The hyphal transport of ^{32}P into the plants and the level of root colonization were measured four weeks after sowing. The levels of root colonization ranged from 56 to 88%, while the hyphal uptake of ^{32}P differed markedly between the five soils: 2, 168, 191, 386 and 1593 Bq per meter of colonized root. These results indicate that some VAM populations are not very efficient; consequently there seems to be a scope for management of field populations of VAM fungi towards an improved P transport efficiency.

Role of Mycorrhizas in Nutrient Transfer from Legume to Rubber

The effect of vesicular-arbuscular mycorrhizal (VAM) fungi on ^{15}N and ^{32}P transfer from neighbouring donor legume (*Pueraria phaseoloides*) to receiver tree plants (*Hevea brasiliensis*) was studied in a specially designed three compartment container using a sterilized Oxisol (Fig. 4.2.2). Plants were contained in nylon bags allowing the passage of VAM hyphae, but not roots and which were separated in the middle by a soil bridge. Another tube in the legume root compartment served as the isotope labelling compartment. ^{15}N and ^{32}P were applied to part of the split-root system of donor plants 13 weeks after planting, and the transfer of nutrients to receiver plants was examined under mycorrhizal and non-mycorrhizal conditions, from live, dying (killed) or shaded plants.

In a short-term harvest, ^{32}P was not detectable in *Hevea* and ^{15}N was detected in trace amounts in some VAM-inoculated replicates.

In a longer-term harvest, the infection of receiver plant roots averaged 73%. Mean root dry weights of mycorrhizal receiver plants were significantly higher than those of the non-mycorrhizal controls, indicating that the fungus

markedly affected plant development. Shoot dry weights of receiver plants were increased by VAM as were their N or P contents. Such greater levels of N and P in the plants suggest that VAM exhaust the soil for mineralized soil N and P.

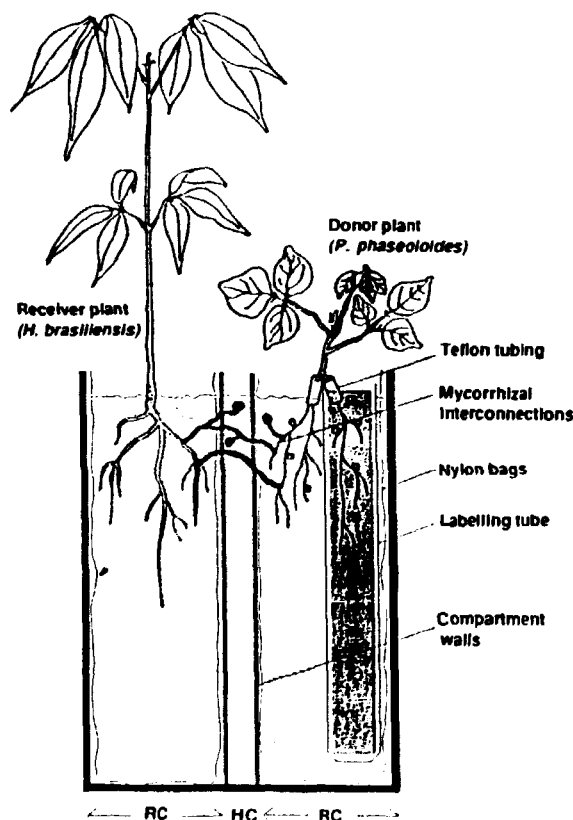


Fig. 4.2.2. Experimental system for measuring ^{15}N and ^{32}P transfer from *Pueraria phaseoloides* to *Hevea brasiliensis* (rubber).

For all treatments, the ^{15}N recovery in donor plants averaged 41% and 34% of the ^{15}N added, for VAM-inoculated and uninoculated plants, respectively. The amounts recovered in receiver plants were small and averaged 0.05% and 0.03% for mycorrhizal and non-mycorrhizal plants, respectively. The amount of N transferred to *Hevea* averaged 0.1% of the *Pueraria* N, irrespective of VAM. Nitrogen transferred from *Pueraria* to rubber never exceeded 0.4% of total N in rubber. As expected, the amounts of N transferred from killed donor plants were more than twice as high as those transferred from live or shaded plants.

The results did not suggest a significant role for VAM in the "direct" transfer of nutrients from a legume to its neighbouring receiver plant, despite the high level of root colonization achieved. This would imply that the transfer of nutrients is occurring by "indirect" means via mineralization of legume plant residues and exudation by roots during normal growth.

5 Chemistry of the Geosphere

5.1 Geochemical Modelling

Humic Acid Behaviour

A systematic study of humic acid behaviour in solutions containing varying amounts of NaCl and CaCl_2 was performed, in which the residual concentrations of sodium, calcium and humic acid, in solution after centrifugation from a flocculate, were determined. From these data the amounts of flocculate formed, as well as the amounts of Na and Ca ions bound to the flocculated, could be determined.

Contrary to the commonly accepted belief that flocculation increases with an increase in ionic

strength, the observation that a calcium-ion induced flocculate is wholly or partly redissolved, when additional amounts of NaCl are added, indicates that competitive specific ionic interaction mechanisms are likely to operate. The observed release of protons with added cations supports the proposed mechanism.

It was possible to make a semi-empirical model describing the flocculation process as seen from Fig. 5.1.1. The model assumes that the calcium ion coordinates two sub-units of the multifunctional humic acid thereby "glueing" them together. This process continues when additional amounts of calcium are added making larger and larger aggregates, which finally may flocculate.

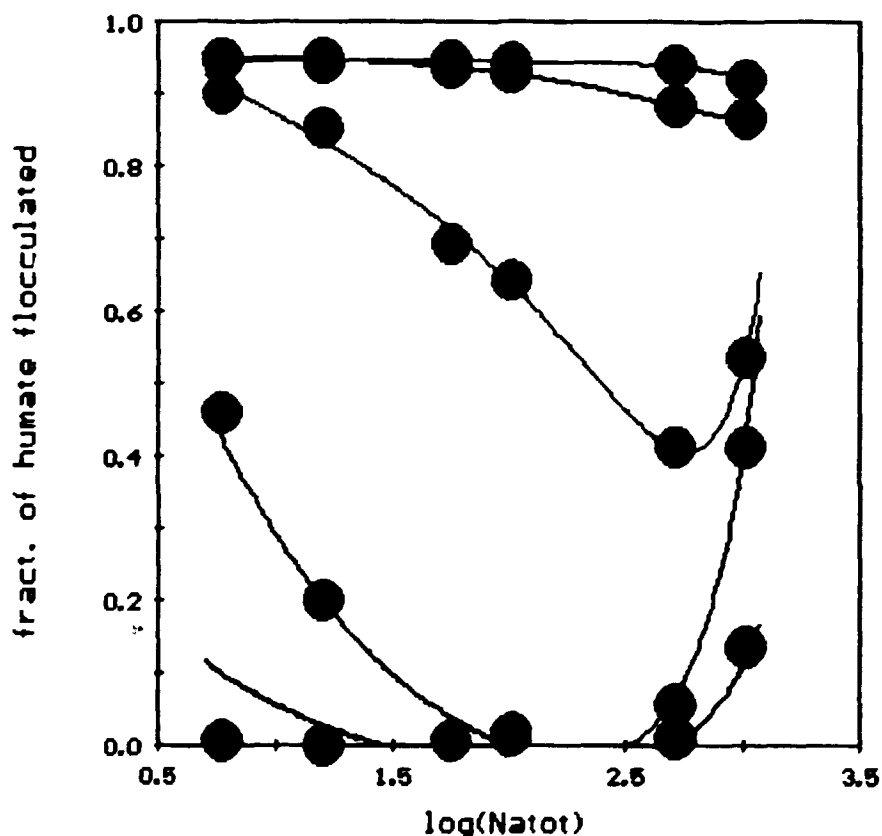


Fig. 5.1.1. Experimental points and modelled response curves displaying the variations in flocculation of added humic acid and NaCl at different total concentrations of calcium in the system. From bottom to top Catot : 0.0018, 0.0052, 0.010, 0.049 and 0.099 mol/litre.

With added sodium ions alone no flocculate is formed, but co-ordinatively bound calcium may be displaced by the sodium ions giving rise to de-flocculation and de-aggregation processes which are observed as dissolution phenomena.

Trace amounts of rare-earths are strongly coordinated to humic acids even in solution, whereby the humic acids may form a vehicle for transporting selected trace elements with flowing ground water. The easy flocculation of humic acid by the addition of simple salts, where the strongly bound trace elements follow the precipitate, points to a possible geochemical engineering potential in taking advantage of these observations.

The Uncertainties of Ignorance

It is well known that the presence of inhomogeneities in a geological formation may have a profound influence on the paths actually followed by flowing ground water. In practice, it is impossible to identify with certainty numbers, positions, sizes and properties of inhomogeneities found in extended geological formations through which contaminated ground water may possibly flow, hence the word ignorance.

As a substitute for extremely expensive field experiments, computer simulations are used for estimating the uncertainties to be expected in the transport through heterogeneous media of ground water plus contaminants. The uncertainties considered, are not of the type usually ascribed to experimental errors, but refer to the insufficient characterization of the media in which water moves, *i.e.* mere lack of knowledge.

It is obvious that when contaminated water moves through an uncharacterized heterogeneous medium, which as a limiting case could be described as cracks in a solid phase, the actual path followed by a single volume unit cannot be traced either theoretically or experimentally. The overall behaviour of the system may, however, be described by effective diffusion and dispersion coefficients with their associated uncertainties. In this case the migration of contaminants can be estimated by calculations, valid for homogeneous media, using the effective diffusion coefficients and their associated uncertainties as input parameters.

The diffusion "experiments" were performed in both two- and three-dimensional calculations. The numerical analogue of the diffusion equation, to be used in heterogeneous media, had to be developed and tested. The resulting programme kept the mass-balance within a few percent, even after hundreds of iterations, and was considered satisfactory for its purpose. Experimentally the positions of a given number of inhomogeneities in the medium were determined by means of a random generator, and the calculation of diffusion, away from a given source, started. For each configuration, the effective "porosity" of the medium was calculated by summing over the "experimental" grid. After a given time, the concentration profile in the medium was calculated and the effective diffusion coefficient calculated as an approximation to the theoretical expression valid for the homogeneous case.

With a given number of inhomogeneities, the above experiments were repeated 30 times and the means of the effective porosities and effective diffusion coefficients, with their associated uncertainties, calculated.

By systematically varying the number of inhomogeneities in the medium, it was possible "experimentally" to derive a relation between the effective diffusion coefficients and the effective porosities, which in the three-dimensional case, displayed an almost linear dependence ranging from the diffusion coefficients of the two selected media. The associated uncertainties had a maximum for effective porosities around 0.5, where inhomogeneities may become matrix and vice versa.

In the two-dimensional case a more complicated behaviour was observed, and it was concluded that the neglect of diffusion, over or under an obstacle, is unrealistic and will result in false conclusions.

The dispersion "experiments", in which diffusion is neglected and only convective transport considered, are similarly made in both two- and three-dimensional systems.

Convective laminar flow around obstacles is amenable to calculations in two-dimensional systems by conformal mapping. By a particle-tracing technique, it can be demonstrated that laminar flow in a two-dimensional, inhomoge-

neous medium invariably gives rise to a longitudinal dispersion effect, but not to any transversal dispersion effects. In inhomogeneous media, convective laminar flow will not give rise to any dispersion effects, but obviously diffusion processes will still operate.

Unfortunately, no simple mathematics are available to treat laminar flow in three-dimensional heterogeneous media, but it is expected that the additional possibility to change direction, velocity and pathlength must give rise to large dispersion effects in all dimensions.

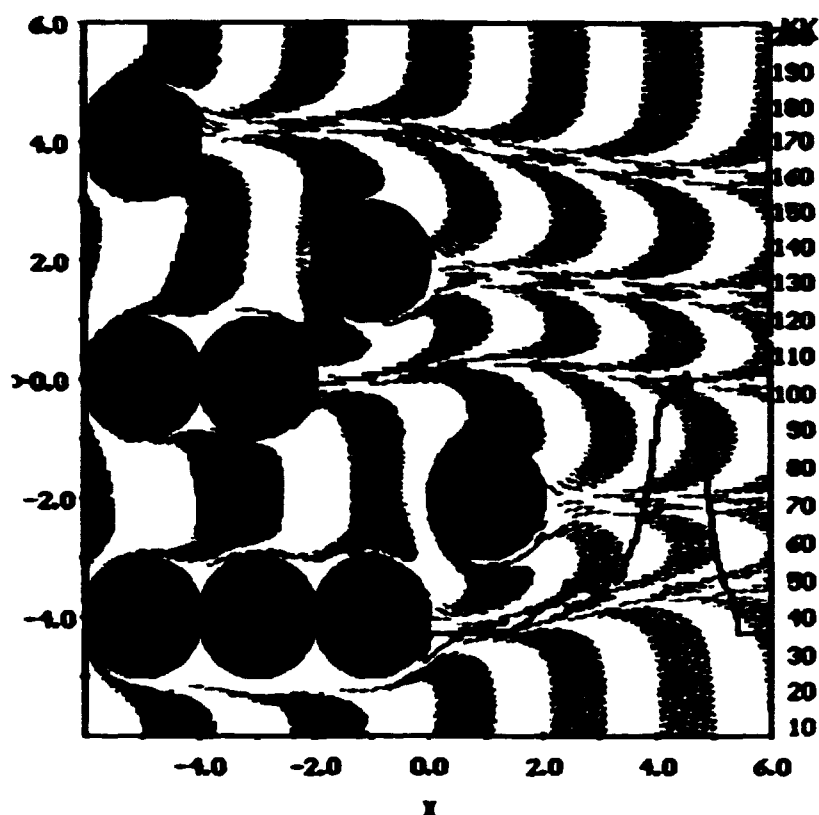


Fig. 5.1.2. An example of the modelling of the distortion of a concentration front by two-dimensional laminar flow around inhomogeneities. The equidistance between the numbering of the streamlines at the right demonstrates the lack of any transversal dispersion. The concentration distribution in branch 6 integrated from streamlines No. 30 to 170 as shown by the continuous curve demonstrates the longitudinal dispersion effects of the introduced inhomogeneities.

By simplified calculations of transport through a lattice of inhomogeneities in three-dimensions, a mechanism demonstrating the effective transversal dispersion of a solute was demonstrated. The calculations can be improved and uncertainties ascribed to the coefficients. The coupling between the longitudinal and transverse dispersion coefficients, which must exist, has not yet been characterized.

5.2 Applied Geochemistry

Methods

The inspection of the upper layers of the seafloor has developed into an important topic of environmental research. Radiometric techniques in the form of integrated instrument platforms and new approaches for the recovery of samples, are

under consideration. As regards the former possibility, towed analytical tools may be used. We have made a contribution in developing a radio-metric sonde (together with the Risø Electronics and Mechanics Department and Roskilde University Center). These, and literature data on other probes (X-ray fluorescence, neutron), were incorporated into a book chapter of the volume CRC Handbook of Geophysical Exploration at Sea (Kunzendorf, 9.1).

The determination of rare earth elements (REE) in samples from the geosphere was previously a major domain of instrumental neutron activation analysis. A relatively new technique, with great potential, is ICP-MS which makes possible the analysis of all of the 14 elements of the REE group. In cooperation with the analysis group, a study was initiated to measure REEs in geological samples. Dissolution procedures were studied and results of the methods were verified by certified standard materials (Kunzendorf *et al.*, 9.5).

A very important factor in the study of metal fluxes into marine sediments is the rate of deposition of sediment particles. Only through realistic sedimentation rate figures is it possible to determine metal accumulate rates on the seafloor. We have implemented known gamma-spectrometric technology to systematically determine ^{210}Pb activities in spade corer samples from Danish offshore areas. The pilot study results were presented at a Nordic meeting (Kunzendorf, 9.2b).

Terrestrial Geochemistry

Danish activities within research on mineral resources were the subject of a one-day meeting at the University of Copenhagen. An overview on marine mineral resources including both shelf and deep-sea deposits was given (Kunzendorf, 9.2a).

Within the framework of a finished energy research project the method of fission track analysis (age dating technique) was reconsidered and an investigation was carried out to establish a new interpretational model (Jensen *et al.*, 9.1b). The data of the energy research project were also published (Hansen *et al.*, 9.1b).

Marine Geochemistry

Most marine ferromanganese phases contain anomalous amounts of metals which are usually not found in their terrestrial counterparts. One of these heavy metals is tungsten which, compared to its content in seawater, is enriched by several orders of magnitude. Some ferromanganese concretions have contents exceeding 100 ppm. The question is, therefore, why and how the large oxyanionic species (WO_4^{2-}) enters the appropriate phases?

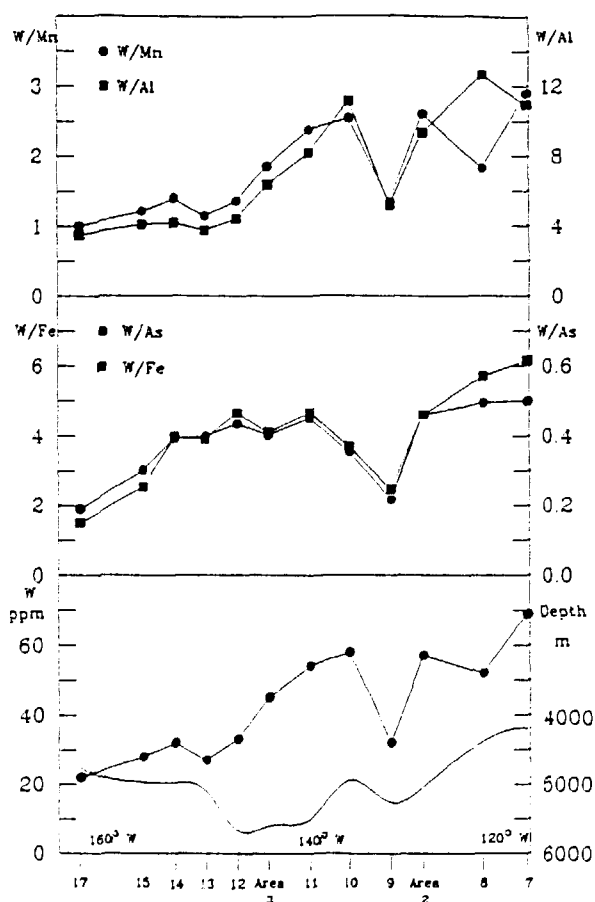


Fig. 5.2.1. Selected W-to-metal ratios in manganese nodules along an E-W transect at 42° S. Ratios W/Mn, W/Fe and W/Al are in units of 10^{-4} .

We have investigated ferromanganese samples (nodules and crusts) along an E-W transect at 41° S in the Pacific. Tungsten distributions found along the transect, could be explained by a mechanism involving adsorption of W onto ferromanganese phases influenced by the northward flowing Antarctic Bottom Water. In this case, the highest W contents were observed in the central parts of the current which are also believed to carry larger amounts of dissolved oxygen. Furthermore, some indication exists that there is a transport of W from the East Pacific Rise to the adjoining deeper basins, suggesting significant element fluxes from the hydrothermal centers. The deposition of tungsten in the marine ferromanganese phases is, therefore, explained as a combination of co-precipitation with Fe/Mn oxyhydroxides from seawater and submarine hydrothermal activity (Kunzendorf and Glasby, 9.1).

The rare earth elements (REE) are thought to represent a group of chemical elements that may be used to predict redox variations in the marine environment. By following their occurrence in normal marine sediments, from the near-shore marine realm to the deeper parts of the oceans, one is able to observe a significant change in the Ce anomaly (expressed as the Ce/La ratio). Waters are increasingly depleted in Ce far away from land and towards the deeper parts of the ocean. Surface sediments show similar REE distribution trends the fundamental difference being that there are sedimentary deposits on the seafloor which show a marked positive Ce anomaly. Literature and our own data have been combined in an overview study (Kunzendorf, 9.2c).

Some of the most interesting new phenomena on the deep ocean floor are black smokers at the mid-ocean spreading ridges. These hydrothermal features may intermittently supply significant amounts of metals to the ocean floor, thereby having an imprint on global metal budgets. We have participated in these studies and have described some of the fundamental facts on black smokers (Kunzendorf and Rose-Hansen, 9.4).

5.3 Geochemical and Applied Technology

Marine Environmental Technology

Several project proposals were channelled through the Baltic Marine Cooperation (BMC), a joint effort of Roskilde University Center and Risø and also CAT. Risø input has mainly been in the form of participating in international meetings.

Heavy Metals Pollution

The retention of heavy metals by clay minerals has been thoroughly studied. The smectic minerals, in particular montmorillonite, have a large capacity of cation exchange, a property which can be related to their structure. Experiments were carried out with bentonite, a clay with a high content of montmorillonite, whose capacity for the absorption of different metals was shown to decrease in the order Cu>Zn>Ni>Cd.

Laboratory work was carried out with a piece of equipment simulating a continuous pipe autoclave. Reaction time was typically 30 minutes and temperatures varied from 140° to 279°C. After the treatment, leaching tests were performed at pH 5 and 3. It was shown that the fixation increased with the amount of added Na₂CO₃ or NaHCO₃, the effect of the former being slightly greater than the latter. The order of retention was Ni>Cu>Zn>Cd. Only 1.2% of nickel but 35% of cadmium were leached under similar conditions (H₂SO₄ at pH 3.0-3.1). This probably reflects the adaptation of the ion to the silicate lattice. By substituting Ca(OH)₂ for Na₂CO₃, the ability to retain heavy metals became neutralized during the autoclave treatment. This is again ascribed to a conversion of the montmorillonite complete structure by reaction with Ca(OH)₂.

Many soils contain smectic clay minerals in amounts sufficient to ensure metal fixation by direct autoclaving. In other cases, such as industrial waste or municipal sludge, it is necessary

to add an adequate clay. Cadmium being one of the most hazardous pollutants is not satisfactorily fixed by silicate lattices. In such a case, a more promising approach is to transform the pollutant into its sulphide, which can be done hydrothermally in the autoclave by adding a small amount of sulphur under anoxic conditions. The disposal of sulphide-bound metals into the environment under oxidizing conditions presents problems. When the oxygen concentration is sufficient the ubiquitous *Thiobacilli* will convert the sulphides into the more soluble sulphates. Therefore, dumping sites must be chosen with special reference to long-term anoxic conditions.

The preliminary pot experiments with Italian rye grass have proved useful in the evaluation of the accessibility of metals in soil.

Investigations have been carried out on scrap-iron refuse from the Danish Steel Works, to remove its content of heavy metals.

Contact is being established with a Danish organisation, which is charged with the detoxification of flue gases from municipal incineration plants. The chemistry section may act as a consultant.

Wet Oxidation of Organic Material

Research

Basic research on the aqueous oxidation of formic acid and formate has been carried out at 260°C, 2MPa of O₂. The formate is converted to bicarbonate whereas formic acid, besides oxidation, decomposes by at least two different routes, namely a dehydration or a decarboxylation. In particular the second one is dependent on the reactor vessel used. It is catalyzed by a mixture of oxides of stainless steel components. The presence of CH₃COOH or CH₃CHO promotes the decomposition of HCOOH by way of both decarboxylation and oxidation. In any case, formic acid is a relatively short-lived intermediate in the wet-oxidation process.

In view of the widespread dislike of polyvinylchloride (PVC) in municipal waste, we have attempted an alternative approach to its decomposition. The wet oxidation combined with alkaline hydrolysis yields water soluble products.

Experiments were carried out at temperatures from 180–260°C and reaction times of 8–24 minutes. The chloride liberated provides information of the rate constants. Considering the measured Cl⁻ and the Chemical Oxygen Demand (COD) values, we find hydrolysis and oxidation processes to be interdependent. The main products are Cl⁻ and CO₂, the remainder comprising a range of water-soluble compounds, a small Cl-free residue and a measurable amount of H₂.

A selection of polyolefins and other paraffins has been studied with respect to the mechanism of their decomposition by wet oxidation. Virtually all hydrocarbon was decomposed into water-soluble compounds and CO₂. Analysis of the products indicates that the decomposition proceeds mainly by fragments of one or two C-atoms at a time. The yield of different products was dependent, to some extent, on the type of starting material. A reaction mechanism has been proposed to explain the intriguing occurrence of significant amounts of hydrogen in the gas phase.

The wet oxidation process of wheat straw was studied as a pre-treatment to the enzymatic break-down of cellulose to glucose. Wet oxidation readily solubilizes lignin from straw and the products are prepared for enzymatic hydrolysis. The wet oxidation process was optimized with respect to oxygen pressure, reaction time, base addition and temperature. The enzymatic hydrolysis was studied by combining two enzymes, a cellulase and a cellobiase (both obtained from Novo Nordisk). An optimum action time and concentration of enzymes gave a 100% conversion of cellulose to glucose.

Development

In 1990, Risø was contacted by a Canadian company with six different pollution problems to be investigated. One of these resulted in the setting up of a large experimental programme for the destruction of phenol in process effluent. The wet oxidation was found to be extremely efficient for the purpose. Under optimal conditions, the reaction time was less than 10 minutes for a total conversion of 15000 ppm phenol. The pH was found to be the most critical parameter. Three detailed reports have been prepared, but so far, they are classified.

As a result of the investigations the NKT-Risø process has now been sold to the Canadian company, who were in urgent need of the technology.

5.4 Chemical Analysis

Research in chemical analysis is guided by the needs of the projects planned, introduced, and completed through cooperation between Risø and other Danish and foreign institutions.

Environmental and agricultural investigations are often dependent on inorganic trace analysis, while geological and material research often involve unusual elemental components presenting analytical difficulties. For such purposes the laboratory's instruments for Atomic Absorption Analysis (AAS), Ion Chromatography (IC), High Performance Liquid Chromatography (HPLC) and Inductively Coupled Plasma Mass Spectrometry (ICPMS) are excellent. The methods for trace analysis can provide reliable data of concentrations in the ppb range or lower, and measure absolute quantities of picogram on a wide range of heavy toxic or essential elements.

These procedures are also used to serve Risø Large Facilities and, as far as possible, Danish industry, especially when exceptional requests are presented. This happened several times in 1992, because of a growing interest in buying special chemicals (e.g. "red mercury") or metals (e.g. osmium, cesium) at very high prices for resale in western Europe. The ICPMS analytical method easily exposed the truth about "red mercury", which was found to be a worthless chemical. Also the claimed isotopic composition of pure metals was certified or invalidated.

ICPMS offers the possibility of using enriched non-radioactive isotopes as trace elements as practised in the project under FLAIR, where first pigs, and later humans, received food with small added amounts of enriched isotopes of ^{25}Mg , ^{67}Zn , ^{70}Zn , ^{65}Cu and ^{74}Se . The uptake and excretions were followed through the analysis of hundreds of samples of blood, faeces and urine processed at the Research Department of Human Nutrition and the National Institute of Animal Science. Concentrations of individual elemental isotopes in a sample may be derived from the

total amount of the element, determined by non-mass-spectrometric method and the measured isotopic ratios. Alternatively, ICPMS measurements of isotopes were converted into concentrations using a software programme, PESOISO. Both methods were used, but the latter benefited from the high sensitivity of ICPMS technology. During 1993, final reports will be presented in addition to the preliminary reports, containing e.g. Round Robin results already published by the Management Committee.

The use of ICPMS in geological investigations gives an added advantage of establishing the relationship of the concentrations of all 14 rare earth elements. An example was presented at the 2nd Danish Symposium in Analytical Chemistry. One of the main concerns when evaluating ICPMS procedures is to find ways to eliminate interferences from e.g. combination ions. When analyzing sediments for rare earth elements, Ba is often present in high concentrations giving rise to formations of BaO interfering with ^{151}Eu and ^{153}Eu . We found it possible to correct for BaO by running Ba standards and samples simultaneously and measuring the ratios $^{135}\text{Ba}/^{151}\text{BaO}$ and $^{137}\text{Ba}/^{153}\text{BaO}$.

A M.Sc. thesis work was designed to try other ways of eliminating interferences by using the newly installed ETV accessory. Chloride can combine with Ar to interfere with ^{75}As and Cl_2 interferes with ^{70}Zn . Because As is monoisotopic and ^{70}Zn is an enriched isotope used in the FLAIR project, both interferences are serious. Even when HCl is avoided as a reagent in the pre-treatment of the sample, Cl might still be present. In using ETV, the idea was either to find a combination of temperature profile and modifier so that the chloride evaporated before As and Zn appeared in the plasma, or to hold Cl in the furnace until all As and Zn had been measured. Preliminary results are promising in some instances.

One of the goals of the FØTEK project was to find a fast analytic method suited for a production line control of raw nutrition products from the Danish agro-industry. Where toxic or other un-wanted inorganic elements are concerned, an obvious choice is ICPMS because of the favourable combination of high sensitivity, fast operation, and multi-element analysis. The combined

efforts of the National Food Agency of Denmark and the section of Chemistry are in the first stage of evaluating a method to find the relationship between element concentration and sample type, e.g. blood, liver, muscle etc. The procedure was tried on reference material from BCR and NIST to establish detection limits and accuracy.

European standardization work calls for much standard reference material, and BCR are presently making a lichen standard. We participated in the preliminary work – Round Robin results reported in 1992 – as this standard will be valued in several current projects in the department and because ICPMS is most convenient for such work. After a short pause due to financial constraints the final analysis programme has just been announced and the section of Chemistry invited to participate.

Close relations between the few Scandinavian laboratories equipped with instruments for ICPMS are maintained by annual user-meetings. This year's two-day meeting was successfully held at Risø in October, with expert participants from a factory in England supplying an exchange of experiences among users.

6. Ecology and Mineral Cycling

6.1 Copper Contamination Affects Osmoregulation in Rainbow Trout

We exposed rainbow trout to Cu^{++} at levels ranging from 0.1 to 0.8 ppm in brackish water and subsequently transferred them to either clean fresh, brackish or sea water. If this was done at a constant temperature (13°C), the result was a drop in plasma Na^+ by up to 8% in fresh water (after 0.8 ppm), which was not seen similarly in brackish or sea water. When the temperature was lowered to 4°C following the Cu^{++} treatment, a drop in plasma Na^+ in fresh water was still seen with a lack of Cu^{++} effect in sea water. This time, however, a small but significant rise in plasma Na^+ was also seen in the fish that remained in clean brackish water. In general, these results showed how the same initial Cu^{++} poisoning of rainbow trout can have very different physiological effects, depending on further ambient salinity and temperature. Similarly, if trout are treated with 0.8 ppm Cu^{++} in fresh water rather than in brackish water, then half of them will die within 3 days.

We have also attempted to study the biochemical mechanism responsible for these effects. The gills are the main site of osmoregulation in fish. We have assayed the formation and turnover of gill surface membranes by measuring the incorporation of ($1\text{-}^{14}\text{C}$) acetate into various gill phospholipids. We could show the same effect of Cu^{++} , whether the fish remained in brackish water or were transferred to fresh water; in both cases there was an enhanced relative incorporation of ^{14}C -activity into phosphatidylcholine. In seawater, previous copper treatment had apparently induced a marked phospholipid degradation.

The incorporation of ($1\text{-}^{14}\text{C}$) acetate into gill phospholipids at 5°C was not in any case influenced by prior Cu^{++} treatment. This may be because a drop in temperature had lowered the rate of membrane lipid turnover, thereby either protecting the gill membrane against any prior damage in contaminated water, or, on the contrary, preventing any function of biochemical

repair mechanisms that might otherwise have been active. We cannot tell directly from our results whether damage or repair is represented by the changes we see in gill lipid metabolism.

In conclusion, we can say that both plasma Na^+ and gill lipid metabolism were influenced by Cu^{++} treatment, but there was no evident correlation between them.

6.2 Annual Crops

A project, dealing with the uptake of radiocesium from soil in annual crops in the Nordic countries after the Chernobyl accident, was carried out. This was done in the framework of NKS-RAD3 (Nordic Nuclear Safety research programme on Radioecology in agricultural ecosystems).

Barley, potato, carrot, cabbage and pea were selected as suitable representatives of Nordic annual crops.

In the Nordic countries the ^{137}Cs deposition varied between 0.1 and 200 kBq m^{-2} , the lowest figure seen in Iceland and the highest in parts of Sweden. In Sweden, Finland and Norway the distribution was very uneven, whereas in Denmark, the Faroes and Iceland, levels were low and more homogeneously distributed.

The uptake of radiocesium from arable soil through the roots to edible parts of annual crops, is generally very low in the Scandinavian countries. Observed ratios_(plant-soil) (OR) ranging between 10^{-5} and $10^{-3}\text{ m}^2\text{ kg}^{-1}$ seem to be the norm in the Nordic countries. The general findings were that levels, after the first year, decreased considerably in the agricultural ecosystems due to the absence of fresh direct fallout and the fast and strong fixing of cesium in most soil types. Agricultural practices inhibit uptake and especially resuspension, by deeper placement of the contaminated surface soil. Only in regions with very special soil types, increased OR values were observed.

Table 6.2.1 below gives an example of the differences in uptake from soil between the Nordic countries.

Table 6.2.1. Observed ratios ($m^2 kg^{-1}$) for potatoes in the different Nordic countries.

	1986	1987	1988	1989	1990
Denmark	0.00016	0.00011	0.00008	0.00009	0.00007
Faroes	0.0076	0.0018	0.0042	0.0035	
Finland	0.0002	0.00005	0.00005	0.00004	0.00004
Norway	0.00054				0.00054
Sweden	0.005	0.002	0.0007	0.0008	

The most striking result is the high rate of uptake on the Faroes, where it is even higher than that on the peaty sites in Sweden. This is especially evident in 1988 and 1989, because of a shorter ecological half-life in Sweden.

Reliable ecological half-life for content of radiocesium in the treated species, cannot be calculated from the material available. An estimate might be approximately 5–10 years in the period from 1987 and until the present time. It is obvious that there is a difference between both the place and the species regarding the ecological half-life. Therefore the study of ecological half-lives on that level might be a subject for future study.

The content of radiocesium is lower in grains of barley than that in the vegetable species. Carrots have a lower uptake to the edible parts compared to other vegetable species, where parts

other than the root are used. These patterns of uptake correspond well with the general assumption.

Because of the low uptake from soil in annual crops, the most important pathway of radiocesium transfer from annual crops to man, is through direct fallout. Therefore after an event like the Chernobyl accident, the season of the year when the event took place is a central factor in determining the transfer to man. As the Chernobyl fallout came at the beginning of the growing season, direct contamination was of minor importance as regards contamination of annual crops. The effect of the direct fallout on winterbarley was seen in Denmark and Finland in 1986 (Fig. 6.2.1), where the decrease from 1986–87 was due to absence of direct fallout in 1987. No evidence of this was seen in Sweden, as springbarley was used in these experiments.

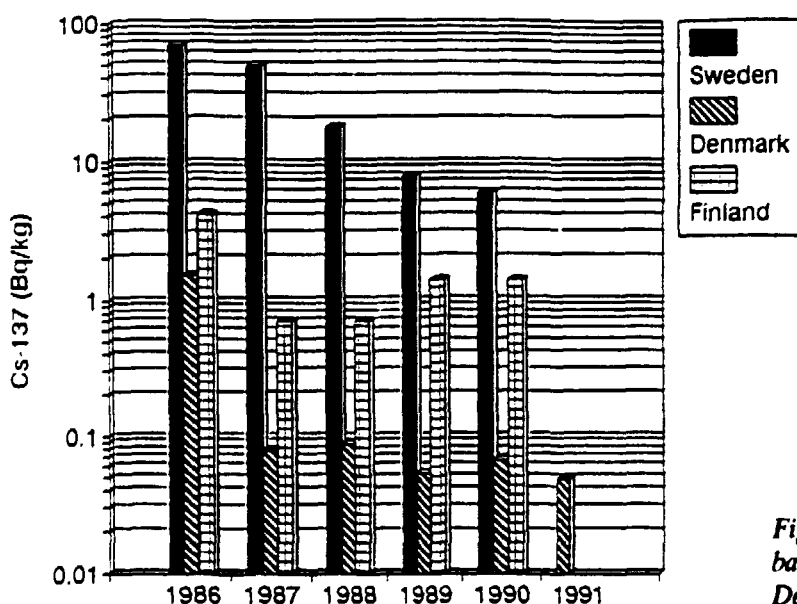


Fig. 6.2.1. The content of ^{137}Cs in barley in Norway, Sweden and Denmark in that order

6.3 Modelling the Transfer of Cs-137 to Lamb

A dynamic radioecological model, simulating the transfer of Cs-137 through the soil-grass-lamb foodchain, was developed in connection with a Nordic project in the framework of NKS (Nordic Nuclear Safety Research). The model is based on observations made in 1990, of the radiocaesium levels in soil, grass and lamb from the Nordic countries and thus reflects the Scandinavian ecosystems used for sheep farming and the farming practices.

The model is based on first-order differential equations where the transfer of Cs-137 between the model compartments is based on rate constants. The model uses time steps of one day. The schematic structure of the model is shown in Fig. 6.3.1.

The model incorporates an atmospheric compartment, a soil compartment, two grass compartments (one for the plant surfaces and one for the internal parts of the plant), and two lamb compartments (one for the gastro-intestinal tract and one for the meat). The transfer processes are indicated by arrows in Fig. 6.3.1. The atmosphere, soil and grass compartments are associated

with a surface area of one square metre ($1 \cdot 1 \text{ m}^2$). Physical decay is taken into account for all the model compartments.

The model assumes an initial contamination of the surface soil of 1 kBq m^{-2} . From the soil compartment, a fraction of the activity is re-suspended into the atmospheric compartment, which is limited by an atmospheric mixing height of 1000 m. The simulation of the resuspension process is based on a resuspension factor of 10^{-9} m^{-1} . From the atmospheric compartment, the activity is transferred to the ground through the application of a generic deposition velocity of 0.023 m s^{-1} representing both wet and dry processes. The deposition is split between a fraction, intercepted by vegetation surfaces and the rest, which is returned to the soil compartment. From the soil compartment, the bio-available activity is reduced through the application of a removal process corresponding to a fixation halflife of 1.5 year. Root uptake from soil to grass is governed by two rate constants, based on concentration factors of the soil-to-grass transfer. The activity, on the external surfaces of the grass, is transferred to the soil through a rate constant corresponding to a weathering halflife of two weeks.

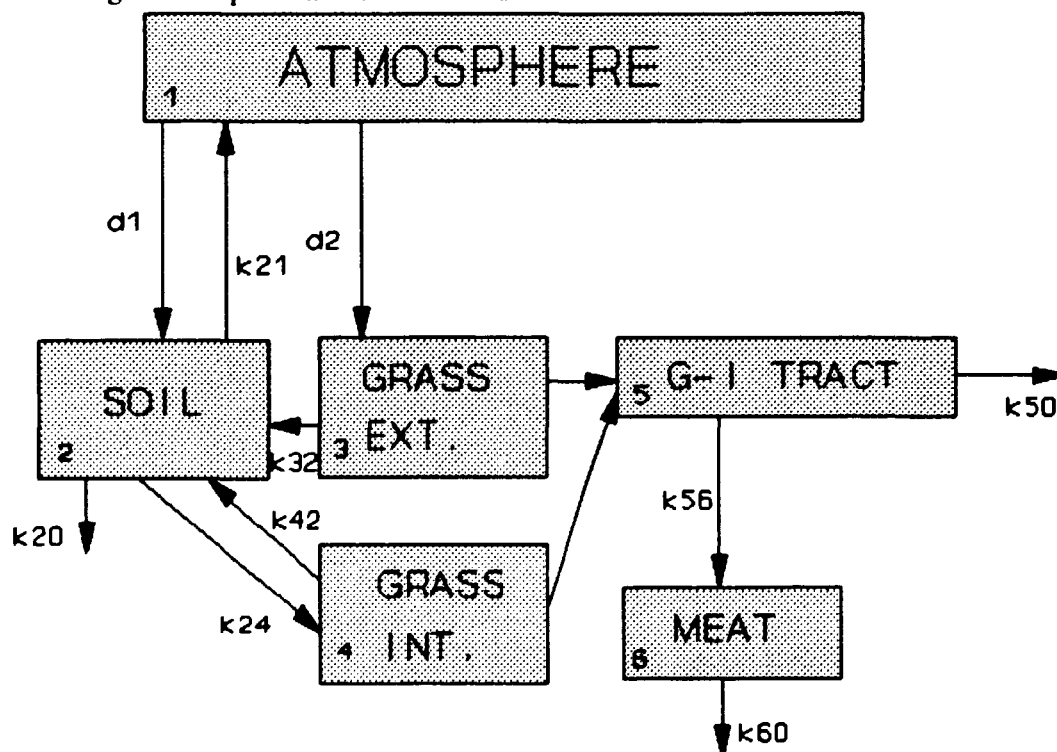


Fig. 6.3.1. Schematic structure of the dynamic model for the transfer of radiocaesium through the soil-grass-lamb foodchain. The transfers between the model compartments are indicated by the arrows.

Fig. 6.3.2 shows the time course of the radiocaesium concentrations in grass and lamb from a simulation for organic soil, where the growing season starts 90 days from the birth of the lambs; 10 days later the lambs are sent on

pasture and slaughtered when they are 250 days old. The resulting concentrations at the time of slaughter are about 50 Bq kg⁻¹ dry weight for the grass and about 30 Bq kg⁻¹ for the meat.

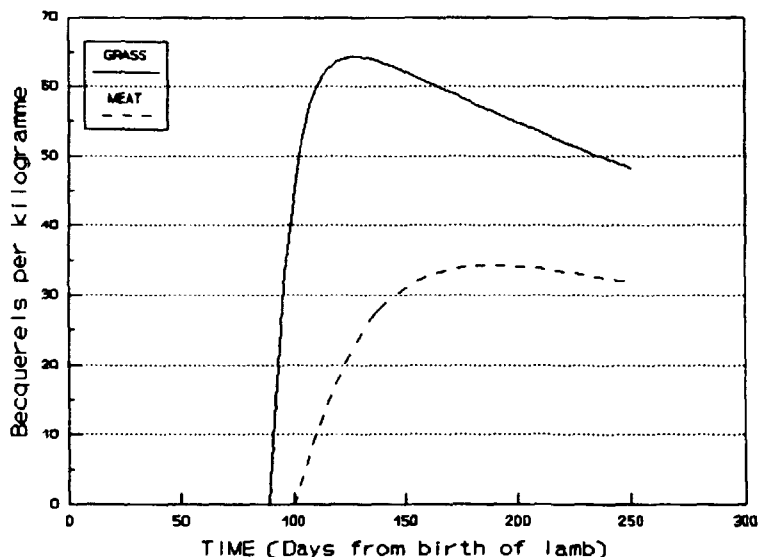


Fig. 6.3.2. Results of model calculations of the transfer of Cs-137 from organic soil (1 kBq m⁻²) to grass (dry weight) and lamb.

6.4 Impact of Insoluble ⁹⁰Sr in Chernobyl Debris on Inventory Estimates

The radiologically most important long-lived radionuclides, after the Chernobyl accident, are ¹³⁷Cs and ⁹⁰Sr. Their relatively high fission yields, the long half lives (~30 years) together with their biochemical similarity with potassium and calcium respectively, are the main reasons for their importance.

The Chernobyl accident released in the order of 100 PBq ¹³⁷Cs (10¹⁷Bq), but the estimate of the ⁹⁰Sr release is less precise. Various sources have reported ⁹⁰Sr releases to be 1 to 2 orders of magnitude less than that of the ¹³⁷Cs release. In the Chernobyl 4 reactor, the inventories of the two radionuclides were 2.6·10¹⁷Bq ¹³⁷Cs and 2.2·10¹⁷Bq ⁹⁰Sr. The ⁹⁰Sr/¹³⁷Cs ratios were thus 0.84. This ratio, however, is not observed in the fallout from the accident, because ⁹⁰Sr is less volatile than ¹³⁷Cs. Moreover, the environmental samples showed a decreasing ⁹⁰Sr/¹³⁷Cs with distance from the reactor. The reason for the

varying estimates of the ⁹⁰Sr release may be due to these variations, together with the fact that ⁹⁰Sr requires a relatively complicated radiochemical analysis for a reliable determination. This analysis is normally started with an extraction of the activity from the soil samples with hydrochloric acid. The procedure has been sufficient for most environmental contaminations with ⁹⁰Sr e.g. for global fallout from nuclear weapons testing. But in case of the Chernobyl debris, this method seems inadequate closer to the accident site. To obtain the total content of ⁹⁰Sr in soil samples collected within 100 km from Chernobyl NPP, a fusion with sodium carbonate is necessary.

This was recently observed in an intercomparison exercise between Risø and 3 CIS-laboratories, which analysed soil samples collected 15 and 5 km from the Chernobyl NPP. While ⁹⁰Sr/¹³⁷Cs in the soil, analysed by the normal procedure with acid extraction only showed a mean of 0.24, the mean ratio in the fused samples was 0.42.

The official CIS estimate of the ⁹⁰Sr inventory from the accident is 8 PBq. If this estimate

was based on the normal analytical procedure for ^{90}Sr , however, the figure may be too low. This may not have any immediate radiological consequence because the ^{90}Sr not accounted for in this estimate, is unavailable for plant uptake due to its insolubility. However, if the ^{90}Sr becomes more soluble with time due to weathering or other geochemical processes, the doses from ^{90}Sr in the near-zone around Chernobyl may decrease less rapidly than otherwise expected. Table 6.4.1 below shows our estimate of the present inventories of total ^{90}Sr and ^{137}Cs at various distances from Chernobyl. The ^{137}Cs inventory within the 100 km zone was based on an integration of the power function shown in Fig. 6.4.1.

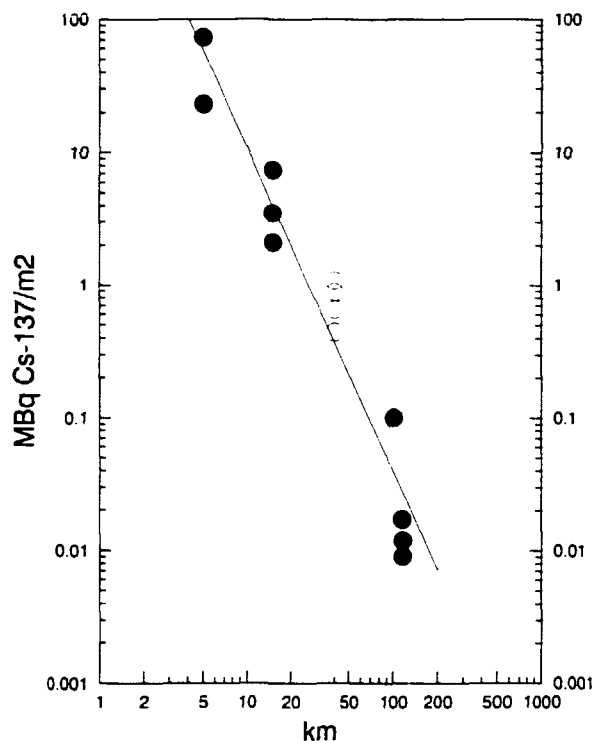


Fig. 6.4.1. The deposition of Cs-137 in the 100 km zone around Chernobyl NPP follows a power function: $\text{MBq Cs-137/m}^2 = 3100 (\text{km})^{-2.45}$.

Table 6.4.1. Estimated inventories of ^{90}Sr and ^{137}Cs deposited from the Chernobyl accident.

Distance in km from Chernobyl/NPP	Area in km^2 of zone	^{90}Sr inventory in PBq	^{137}Cs inventory in PBq	$^{90}\text{Sr}/^{137}\text{Cs}$
0-100	7900	6.7	16	0.42
100-600	275000	4.7	18	0.26
600-1000	500000	0.6	7	0.09
1000-10000	$75 \cdot 10^6$	0.2	53	0.004
TOTAL		12.2	94	0.13

6.5 Radiotracers in Coastal Oceanography

Risø is taking part in a 3-year EEC project under the Marine Science and Technology programme (MAST), where radionuclides discharged from La Hague are used to trace advection and dispersion of water masses in the European Coastal Current from the English Channel to the entrance to the Baltic. Time-series of radionuclide measurements in water samples are being performed.

Results from the Kattegat reveal an interesting variability in the origin of the inflowing saline bottom water. The origin of the water masses entering the inner Danish Waters is important because the European Coastal Current water carries much higher loads of nutrients and pollutants than those from the Atlantic Water. Fig. 6.5.1 shows the measured concentrations of ^{99}Tc in the inflowing bottom water in the Kattegat from two stations: DMU-413 ($56^{\circ}40'\text{N}$, $12^{\circ}07'\text{E}$) and DMU-925 ($56^{\circ}08'\text{N}$, $11^{\circ}10'\text{E}$).

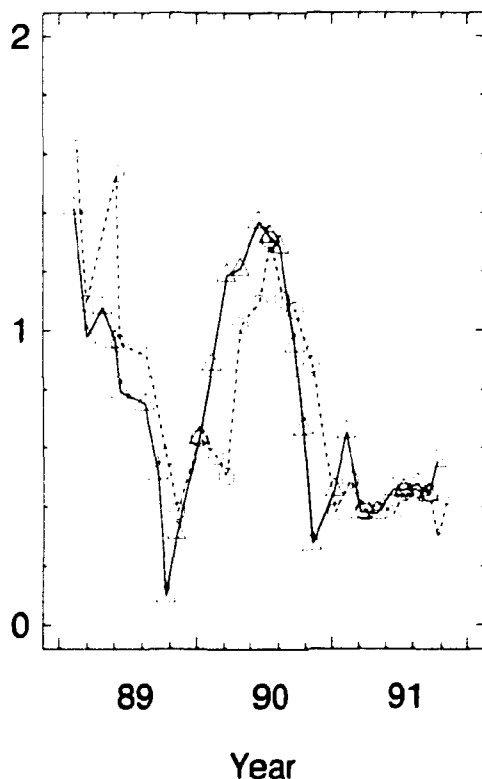


Fig. 6.5.1 Technetium-99 concentrations at two locations in inflowing Kattegat bottom water 1989-1991.

The relatively high levels of ^{99}Tc , i.e. a clear La Hague signal, indicate inflow of water from the European Coastal Current during the first part of 1989. This fits with a breakthrough of nutrient-rich waters from the German Bight to the Kattegat in early 1989. The next breakthrough of ^{99}Tc in 1990 could not be correlated with inflowing nutrients.

The measured ^{99}Tc concentrations in the German Bight and the Danish Waters have been compared with La Hague discharge data to obtain Transfer Factors (TF). The calculations are based on 6-month average values of measured ^{99}Tc concentrations and discharges m months before the sampling, where m is the assumed transport time from La Hague to the sampling point. As an example, a TF value is calculated by averaging measured values from Borkumriff (January-June 1991), and dividing them with the average discharge m month earlier (January - June 1990). The TF values in Table 6.5.1 are the average \pm SD for these n half-year values. This reduces the sensitivity to a correct estimate of the transport time m . As the unit for the transfer factor, $\text{Bq m}^{-3} / \text{TBq yr}^{-1}$ is equal to $\text{yr} / 10^3 \text{ km}^3$, it represents the number of annual discharges found in 10^3 km^3 . The reciprocal value, TF^{-1} , then expresses the number of 1000 km^3 which an annual discharge has been diluted at the sampling point.

The water transport through the Kattegat can be compared to a conveyor belt transporting high salinity water towards the Baltic in a bottom layer which is gradually mixed up in the low salinity surface layer leaving the Baltic. From salinity data, it has been estimated that on average, $1450 \text{ km}^3 \text{ yr}^{-1}$ enters the Kattegat from Skagerak via the bottom layer, whereas $1920 \text{ km}^3 \text{ yr}^{-1}$ leaves the Kattegat via the surface layer. The difference is due to the fresh water surplus leaving the Baltic. Comparing these water transport figures with the TF^{-1} values of Table 6.5.1 it is indicated that 15% and 12% of the La Hague discharge passes the Kattegat's bottom and surface layer, respectively. These figures may be compared to a previously calculated Transfer Factor from Sellafield to the Kattegat, $0.015 \text{ Bq m}^{-3} / \text{TBq yr}^{-1}$, indicating that 2% of the Sellafield discharge passes Kattegat.

Table 1: Transfer Factors. TF between La Hague German Bight and Danish Waters. *m*: assumed transport time (months), *n*: number of half-year TF values averaged, TF: Bq m⁻³ / Tbq yr⁻¹, TF⁻¹: 10³ km³ yr⁻¹.

From La Hague to:	m	n	TF ± SD	TF ⁻¹
Borkumriff	12	4	0.34 ± 0.14	
Elbe-1	12	3	0.31 ± 0.11	
German Bight mean	12	7	0.32 ± 0.12	3.1
Utsira, s.w. Norway	18	10	0.086 ± 0.029	12
DMU-413, bottom	15	6	0.098 ± 0.034	
DMU-925, bottom	15	6	0.105 ± 0.040	
Kattegat, bottom mean	15	12	0.101 ± 0.036	10
DMU-413, surface	20	6	0.067 ± 0.031	
DMU-925, surface	20	6	0.061 ± 0.034	
Kattegat, surface mean	20	12	0.064 ± 0.031	16
Klint, Kattegat coastal	20	8	0.069 ± 0.042	14
DMU-444, Arkona, surface	20	6	0.019 ± 0.006	54

How representative are these figures for other coastal discharges? This question may be answered by the oceanographic models, if they are able to simulate the measured radionuclide concentrations from the discharge values. A comparison of the transport pathways from the two discharges to the Kattegat, gives a reasonable explanation on the higher transfer from La Hague, where the discharge follows a coastal current directly to the Kattegat. The Sellafield discharge to the Irish Sea is first transported to

the North Sea via a coastal current around Scotland, whereafter a fraction is crossing the North Sea and entering the Kattegat. The two figures probably represent minimum values for the transfer to the Kattegat of discharges to the Coastal currents on the two transport routes, *i.e.* at least 2% of a conservative discharge on the coasts of Scotland and east England and at least 15% of the discharge to the coastline between Cherbourg and Denmark will probably reach the Kattegat.

7 Other Activities

7.1 Decontamination of Urban Surfaces in the Town Pripyat near Chernobyl

In June an expedition was sent to the Chernobyl area in order to test the efficiency of different methods of decontamination of radioactive contaminated surfaces in the urban area.

This was done in collaboration with the IAEA-EC international CHECIR laboratory situated close to the Chernobyl 30 km zone. The project was sponsored by CEC.

In order to facilitate in situ measurements, a specially adapted road vehicle was constructed.

A bus originally created for transport of furniture was modified for use as a mobile laboratory. The driver's cabin was mounted with 4 extra seats and on top of the truck, an extra room was built for emergency sleeping.

A power system that could provide the equipment as well as the measuring system with 220 v power, was installed in the mobile laboratory.

The mobile laboratory was then equipped with tools and chemical, for decontamination purposes and transported to Kiev.

In Kiev, a CIS-team joined the Danish team to go to the town of Pripyat. Pripyat is situated a few km from the damaged Chernobyl reactor and was evacuated during the Chernobyl accident.

Four different methods were tested in Pripyat:

- a) high pressure washing
- b) NH_4NO_3 - treatment
- c) wet sand-blasting
- d) treatment with clay material.

The decontamination factors i.e. the relations between the contamination density before and after decontamination were identified on brick walls and asbestos-type roofs by beta measurements, and on the walls, for calibration purposes by shielded gamma measurements.

The results for the different methods are shown in the graphs in the following Figs (7.1.1-7.1.6). The graphs show the accumulated reading of the beta-instrument before and after decontamination for the different points at which the surface was measured as a function of the accumulated number of points.

The first test was done with a KEW high pressure washer 150 bar and hot water (60°C). The decontamination factor, i.e. was found to be 1.65 with a 95 % confidence interval of 1.33 to 1.97.

Fig. 7.1.1 shows the results.

NH_4NO_3 treatment was tested by spraying a 0.1 M NH_4NO_3 solution onto a wall. The wall was saturated with the solution and after 30 minutes it was flushed with water. The results are shown in Fig. 7.1.2. The decontamination factor was found to be 1.13, with a 95 % confidence interval from 1.06 to 1.19.

Cold water, with a pressure of 65 bar fed by a turbo nozzle, was used to decontaminate an asbestos-type of roof with readily available equipment. The results are shown in Fig. 7.1.3. The decontamination factor was found to be 1.86 with a 95 % confidence interval from 1.72 to 2.01.

Wet sand-blasting was performed on a wall using carborundum sand. The cumulative distribution of readings before and after decontamination is shown in Fig. 7.1.4. The decontamination factor was found to be 2.19, with a 95 % confidence interval of 1.80 to 2.59.

A treatment with clay material was made by covering the surface with a clay emulsion and subsequently, after drying, removing the clay by vacuum cleaning. This treatment was done twice on the same wall. Fig. 7.1.5 shows the results after the first treatment and Fig. 7.1.6 shows the results after the second treatment.

The decontamination factor after the first treatment was found to be 1.08 with a 95 % confidence interval from 1.00 to 1.15. After the second treatment the decontamination factor was found to be 1.21 with a 95 % confidence interval of 1.12 to 1.31.

Fig. 7.1.1. The cumulative sum of the readings of surface contamination density before and after decontamination by high pressure (150 bar) warm water hosing using a turbo nozzle as a function of accumulated points.

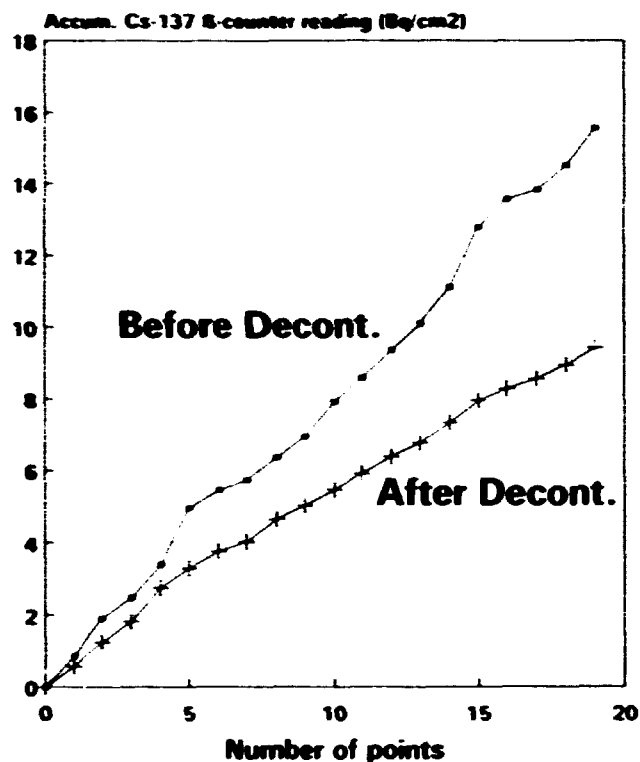
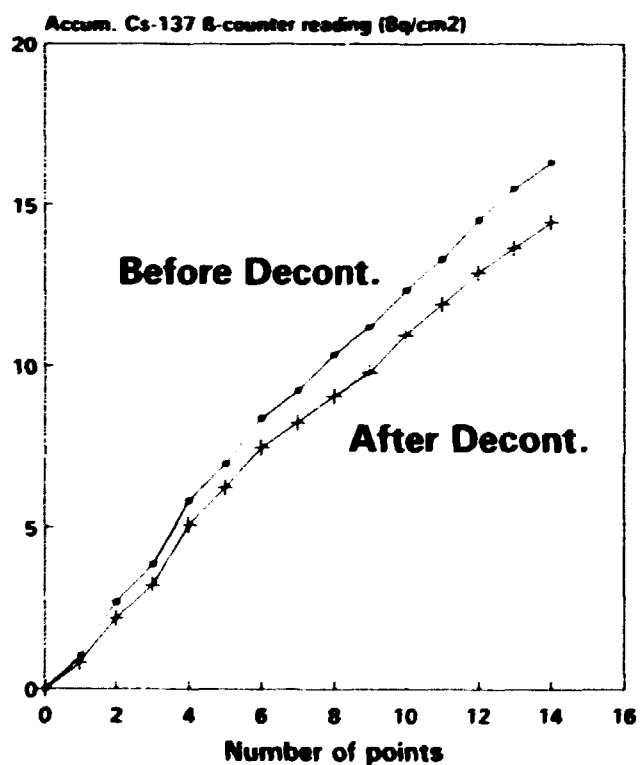


Fig. 7.1.2. The cumulative sum of beta counter readings of surface contamination density on a sandstone wall before and after decontamination using a NH_4NO_3 treatment as a function of accumulated points.



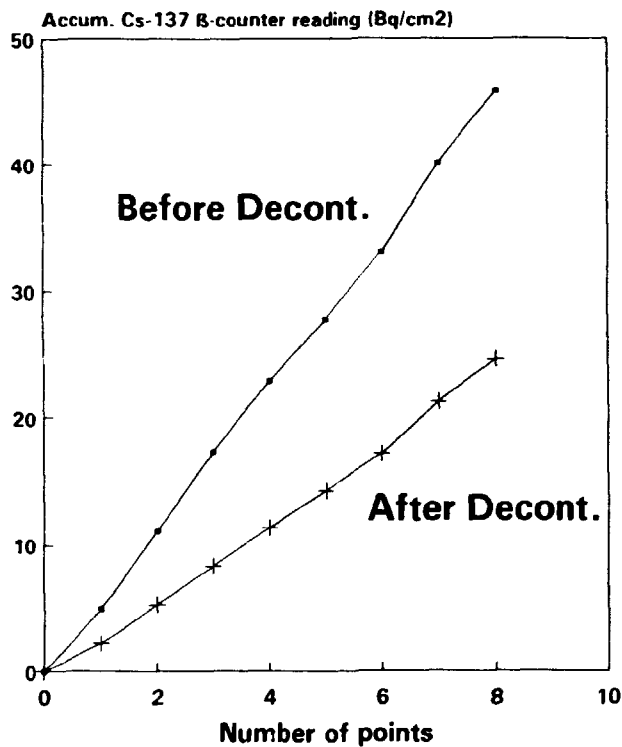


Fig. 7.1.3. The cumulative sum of the beta counter readings of surface contamination density on an asbestos type roof before and after decontamination by high pressure (65 bar) cold water hosing using a turbo nozzle as a function of accumulated points.

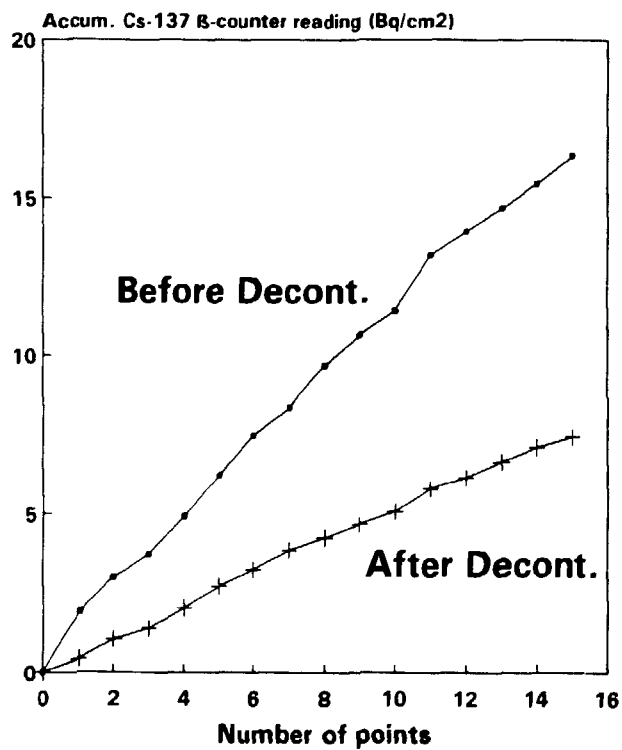


Fig. 7.1.4. The cumulative sum of the beta counter readings of surface contamination density on a sandstone wall before and after decontamination by wet sandblasting as a function of accumulated points.

Fig. 7.1.5. The cumulative sum of beta counter readings of surface contamination density on a sandstone wall before and after decontamination by clay, as a function of accumulated points.

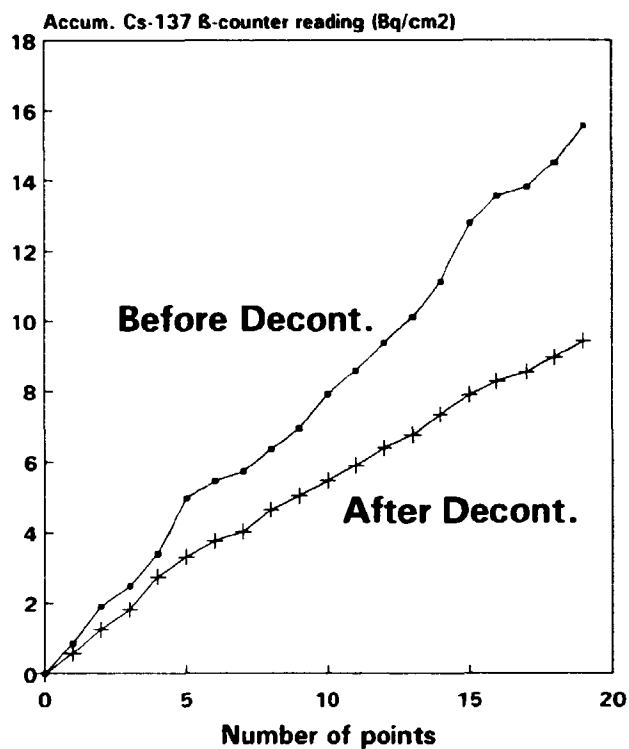
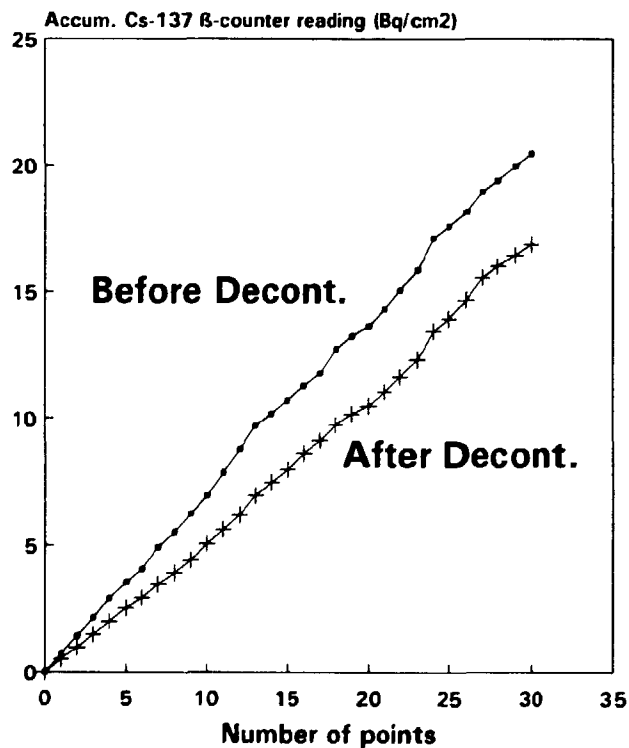


Fig. 7.1.6. The cumulative sum of beta counter readings of surface contamination density on a sandstone wall before and after decontamination using a double clay treatment, as a function of accumulated points.



For calibration purposes, the efficiency of decontamination was also measured using a shielded gamma-spectrometer facing the surface to be investigated.

Gamma spectrometry was performed both before and after decontamination on one of the treated walls, so that the beta and gamma measurements could be compared.

The gamma measurement showed a decontamination factor of 1.4, when the result obtained by beta counting was 1.7.

Decontamination testing on wall surfaces, 6 years after contamination, showed that the most efficient of the investigated methods was sand-blasting. For beta counting, a DF factor of more than two was found using this method on a brick wall. The sand-blasting, however, creates relatively large quantities of solid waste compared to the other investigated decontamination methods.

High pressure water, fed through a turbo nozzle, was also found to be an efficient means of decontamination giving less solid waste compared to that of sand-blasting. Decontamination factors measured with beta counters were found to be 1.7 for 60°C hot water at a pressure of 150 bar. For a clean asbestos-type of roof, a decontamination factor of 1.9 was found, using cold water at a pressure of 65 bar.

Least efficient were the chemical methods, NH_4NO_3 and clay having decontamination factors ranging from 1.1 to 1.2. These methods, however, are believed to be much more efficient on newly contaminated surfaces.

The decontamination factor measured by beta counters was 1.7, when the same factor was found by gamma spectrometry to be 1.4. The larger beta factor is due both to the shape of the contamination profile through the uppermost layer of the wall and to the shielding of the beta particles.

7.2 Indoor Aerosol Deposition Studies Using Neutron-Activatable Tracers

The indoor environment can contain a variety of airborne contaminants, such as bacteria, fungi and radon progeny, which exist in discrete size categories or are attached to ambient aerosol.

Such contaminants, when inhaled, present a potential hazard to the occupants of a dwelling. The ingress of artificial radionuclides into buildings present both an inhalation dose to the occupants and an external gamma dose, when those radioactive particles are deposited on indoor surfaces.

The process of aerosol deposition is an important factor for aerosol concentration and thereby the potential health hazards. To yield relevant data, a detailed particle size-specific deposition study has been undertaken, examining a variety of conditions of building occupancy.

A full-scale deposition experiment involves establishing a well-mixed tracer aerosol in a suitable room and monitoring the aerosol concentration decay by sequential filter sampling of the air. If the air exchange rate in the room is measured, the rate constant for aerosol deposition can be calculated by subtracting the air exchange rate constant from the observed aerosol decay rate.

Aerosol Generation and Labelling

A technique of particle labelling, dispersion and detection was developed in collaboration with Imperial College, UK. Porous silica particles are supplied by Phase Separations Ltd. in a variety of monodisperse size distributions. By agitating the particles in a solution of a suitable tracer, tracer ions become bound to the particle surfaces in a uniform manner. Typical labelling yields are of the order of 5 mg of tracer per gram of silica.

The neutron activatable tracer presently used is Dysprosium-164. Dysprosium has many advantages as a tracer: it is present in very low concentrations in nature and it has a high activation sensitivity. Different filters for aerosol sampling have been investigated for low background radiation after activation, *i.e.* a low sodium content. To minimize the filter surface, high pressure pumps are used. This enables us to use a smaller filter to achieve a lower background radiation when irradiated.

The labelled particles are dispersed using a Palas RBG-1000 powder dispersion generator. To equilibrate charges which build up on the particles during aerosolization process, a 150 MBq beta source is positioned at the generator outlet.

In the experiments carried out to date, mono-disperse particles of two sizes were used. A TSI 3300 Aerodynamic Particle Sizer, APS, were employed to measure the particle size distributions. The particles were found to have mean aerodynamic diameters of 2 and 4 microns, respectively.

Results

Full scale deposition experiments, using surrogate aerosol, were carried out in two houses: a single-story typical Danish dwelling, of double brick construction with a good standard of weather-proofing and a two-storey test house at the British Building Research Establishment's site

at Garston, (BRE). In both series of tests, the air exchange rate in the experimental rooms was measured by monitoring the decay rate of sulphur hexafluoride tracer gas.

Experiments were carried out for the two particle sizes in both furnished and unfurnished rooms. Fig. 7.2.1 shows the decay in particle concentration during an experiment. The slope of the curve yields the deposition constant for the room.

The average deposition constant from the two tests are largely consistent; the results from the Danish series are shown in Table 7.2.1. The results show an increasing deposition with size and increased surface. This illustrates both the dominant influence of gravitation in this size range and the influence of diffusion.

Deposition Test 1.5

Fig. 7.2.1. Time decay curves for 2 micron aerosols in an unfurnished room at BRE. The y-axis is the count rate for a filter divided by the amount of air sucked through the filter. The upper curve shows the result of an activation in the reactor at Risø and the lower represents an activation in the UK.

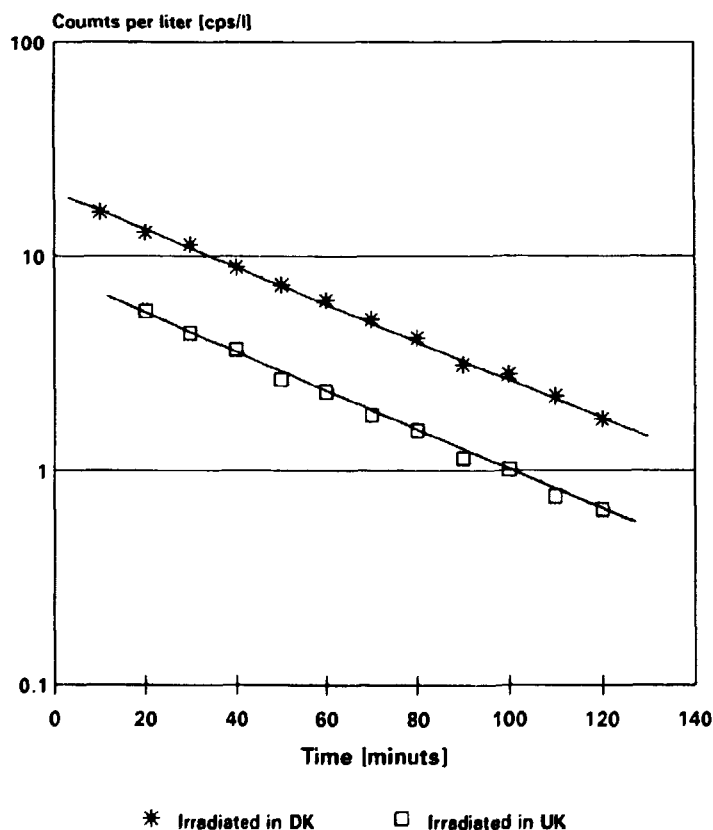


Table 7.2.1. Deposition constant for room with and without furniture.

	Particle size	Deposition constant	Mean deposition velocity
Unfurnished	2 μm	0.84 h^{-1}	0.015 $\text{cm}\cdot\text{s}^{-1}$
—	4 μm	1.51 h^{-1}	0.027 $\text{cm}\cdot\text{s}^{-1}$
Furnished	2 μm	0.95 h^{-1}	0.017 $\text{cm}\cdot\text{s}^{-1}$
—	4 μm	2.10 h^{-1}	0.038 $\text{cm}\cdot\text{s}^{-1}$

Calculations of deposition velocities were made with models developed to predict the deposition of radon progeny. Both convective and turbulent airflows were considered. A comparison of the experimental deposition velocities from the two houses and a model prediction are shown in Fig. 7.2.2. In general the 4 micron particles deposited as predicted by the models, but the 2 micron particles had a larger deposition velocity than predicted for both flow types.

This experimental technique for determining indoor aerosol rates will be adapted to other building occupancy parameters which might alter indoor particle deposition. An effort to produce submicron particles is carried out using Indium as tracer. Extension of the technique to measure surface-specific deposition velocities is also a new research topic. These labelled particles are the basis for a recent Ph.D. project.

Convective deposition dT from -3.0 to 3.0

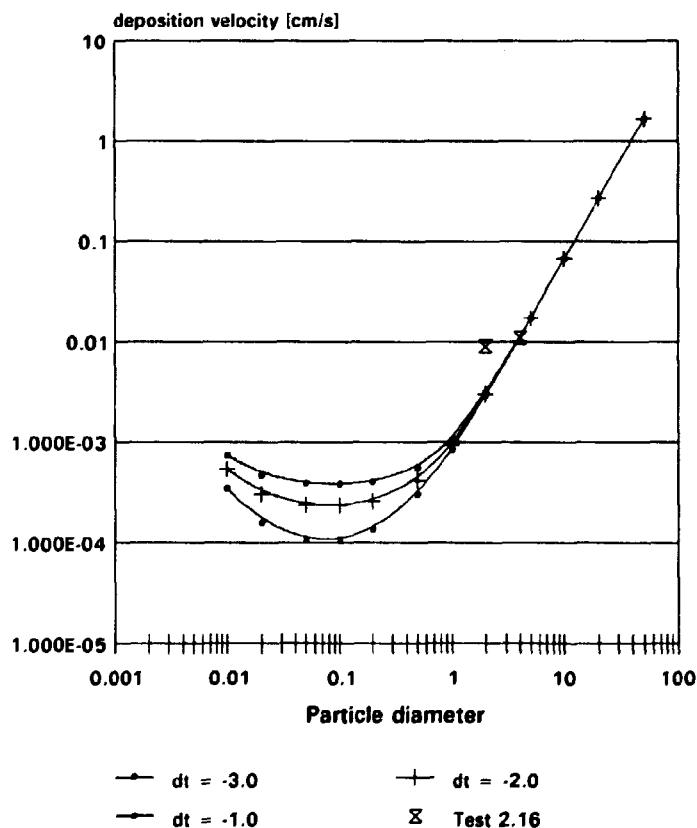


Fig. 7.2.2. Predicted deposition velocity as a function of particle diameter. Curves are shown for wall temperatures 1, 2 and 3 degrees cooler than air. Two experimental values are also plotted.

7.3 Photochemistry and Photophysics

The general scope of the activity has been expanded considerably. In particular, a new Excimer-laser, operating at wavelengths of 193 and 248 nm, was installed. The laser was interfaced with the existing experimental flash photolysis setups, and a new setup for flash photolysis in the gas phase is under construction. This expansion has been possible through considerable support by the Danish Natural Science Research Council.

Most of the previous studies are being continued. They all concern investigations of the potential energy surfaces of electronic excited states or free radicals of organic molecules of fundamental and applied interest. All studies were carried out in close collaboration with Dr F.W. Langkilde, Danish Engineering Academy. Studies on polyenes and related compounds are being continued. In particular, studies were done on specially synthesized hexatriene which is "locked" at the central position and therefore unable to isomerize. Theoretical work on the triplet state of stilbene was undertaken to understand the previously experimental results. Both these projects were carried out in international collaboration with laboratories in Italy (Professor G. Orlandi) and The Netherlands (Drs A.M. Brouwer and H.J.C. Jacobs). An extensive review article has been accepted for publication ("Molecular Conformations in Electronic Excited States", in *Structures and Conformations of Non-rigid Molecules*, 1993).

A study of intramolecular proton transfer was completed in collaboration with the Institute of Physical Chemistry of the Polish Academy of Sciences (Dr A. Mordzinski). This work is being continued and studies of molecular switches, based on intramolecular proton transfer, initiated.

An article of fundamental interest in atmospheric and combustion chemistry, on experimental and theoretical results on the benzyl radical, is almost completed (in collaboration with the Industrial Chemistry Research Institute, Poland, Dr K. Bajdor and Professor G. Orlandi, Bologna). These studies are being continued and expanded, and collaboration with Dr Torben Lund, Roskilde University, has been established. In this context, we have also initiated theoretical ab initio calculations on one of the large vector processors

(Convex 3840 in Århus) thanks to support by the Danish Natural Science Foundation.

A Ph.D. project (J. Frederiksen) on studies of fluorescence in macrocyclic compounds e.g. in molecular switches, is being continued in collaboration with CISMI, Copenhagen University (Professor K. Bechgaard) and the Institute of Chemistry, Copenhagen University (Dr N. Harriet).

Collaboration with Dr O.S. Mortensen, Odense University, in the field of resonance Raman spectroscopy is being continued.

7.4 Risø High Dose Reference Laboratory (HDRL)

In late 1991, the Risø High Dose Reference Laboratory was accredited by DANAK, the Danish Accreditation Scheme. Since the accreditation was granted, the following accredited services have been carried out:

- | | |
|---|------|
| 1. Irradiation at cobalt-60 gamma-cell of samples or dosimeters | : 12 |
| 2. Irradiation at 10 MeV accelerator of samples or dosimeters | : 4 |
| 3. Issue of calorimeters | : 5 |
| 4. Issue of dichromate reference dosimeters | : 7 |
| 5. Issue of FWT-60 reference dosimeters | : 6 |
| 6. Measurements of facility parameters | : 3 |
| 7. Measurements of dose distribution in irradiated products | : 58 |

The reports and certificates of the HDRL have been received by its customers as important to the quality control primarily of radiation sterilized products.

Through membership of the sterilization committee in DMDA (Danish Medical Device Association) we have taken part in the work of creating a new European Standard for Radiation Sterilization. The standard (prEN 552) is written by the European Standard Organization, CEN. In this standard, the need for traceable dose measurements is stressed, which is exactly the main aim of the HDRL.

The International Standard Organization, ISO, is also preparing a new standard on Radiation

Sterilization. The two standards are very similar, but in one area a disagreement exists: The definition of "Sterile" differs in the two standards, therefore, different radiation doses may be used, depending on which standard is followed.

7.5 Synthesis

The commercial activities have increased steadily and are now one of the major activities in the group. They are mainly concerned in the preparation of chemical compounds with diagnostic applications and radioactive labelled compounds for medical research.

A major project with a Danish firm, concerned with the development of new products with special applications, is being negotiated and is expected to start in 1993.

The synthesis group is taking part in a co-operative project FØTEK (Fødevarer-Teknologi-Projekt), by preparing non-radioactive labelled compounds, *i.e.* toxins and anabolic steroids.

Among other non-commercial activities, in a cooperation between DMU and Risø, are the syntheses of isotopically labelled compounds for studying the degradation of sulphonated soaps in the environment, and the building-in of chlorinated monomers into humic acids used for migration studies.

Synthesis of surface-active compounds for sensor studies, and of surface-bound cyclic metal complexes for electrochemical studies, initiated by MODECS, has resulted in student projects as part of the group's educational activities.

7.6 MODECS

In 1992, MODECS arranged a one-day meeting on Tribology and a two-day meeting on Sensors in Medicine and The Environment, both with internationally renowned specialists as the main speakers. Both meetings were very successful.

Topics for two meetings in 1993 have been selected and are, at present, being planned. The first topic is "Plastics of the Future, the Environmental and Energy Aspects.", and the other is "Molecular Recognition and Self-organizing Systems".

The MODECS initiated Ph.D. on "Collective Phenomena of Functional Molecules on Surfaces" is progressing well, in both the theoretical and experimental aspects. At present, the work is concerned with the thermodynamical characterization of the collective phenomena, including counter-ion associations.

8 Large Facilities

8.1 10 MeV Linear Electron Accelerator

During 1992, an accident happened at one of the Danish irradiation facilities, making this facility unable to irradiate products mainly for sterilization, and an old obligation to serve as back-up was effectuated.

The intention was to irradiate products for a few weeks only, but the problems at the commercial facility proved more difficult to solve than expected, and extended product irradiation was actually carried out in a 2-shift operation and on weekends for approximately 6 months. This extra work was made possible through an extra effort by all personnel involved.

8.2 Pilot Plant for Wet Oxidation

The heart of the plant is a 2000 meter long pipe reactor which makes continuous treatment of suspensions possible under alkaline, neutral or acid conditions with oxygen at 280°C and 100 bar. The plant has been used to implement the Risø-NKT-wet oxidation process on a semi-industrial scale.

8.3 The RIMI Field Station

Risø's Integrated Environmental Project (RIMI) is an interdisciplinary project studying pathways, processes and effects on terrestrial ecosystems of nitrogen compounds derived from human activities.

RIMI is managed by the Ecology section and involves scientists from the Environmental Science and Technology Department, as well as scientists from the Meteorology and Wind Energy Department, and the Optics and Fluid Department at Risø.

The RIMI station is used in the Danish contributions to the EUROTRAC projects BIATEX (managed by the Ecology section), TOR (managed by the Chemistry section) and TRACT (managed by the Department of Meteorology and Wind Energy). A number of projects within the SMP Center for Air Pollution Processes and Models make use of the field station. One of these projects, concerning the atmospheric composition of nitrogen compounds, is managed by the Chemistry section and performed in collaboration with the National Environmental Research Institute.

The RIMI station also serves as a reference station for air pollution monitoring in Copenhagen by the National Environmental Research Institute.

8.4 Dyskærgaard, the Experimental Farm

Dyskærgaard has 120 hectares of arable land, and is managed by the Plant Biology Section. The experimental farm is partly used for scientific field experiments, and partly for grain and beef production.

During the growing season a herd of Hereford cattle graze the sea-shore meadows and salt marshes along the Roskilde Fjord for nature management purposes.

In 1992, a project was initiated to implement Computer Aided Farming (CAF) techniques for Local Resource Management (LRM) in farming. The project includes application of the most advanced technology for differentiated, on demand, fertilizer application, and differentiated, on demand, use of crop protection chemicals. Partly because of this project, the department acquired a new combine-harvester (Dronningborg, Jumbo D 8700) equipped with an advanced computer system, including a Global Positioning

System (GPS) and instrumentations for yield measurements. Further, a portable GPS-system is in the process of being interfaced with red and infrared light sensors, and a new spectroradiometer is under development to automatically obtain information about biomass and plant vitality on an areal basis. A preliminary version of the new instruments will be tested during the growing season of 1993.

8.5 Open Top Chamber Facility

In collaboration with the National Environmental Research Institute and the University of Copenhagen, the department operates a large Open Top Chamber Facility situated at Risø. The facility consists of 19 large chambers (3.5 meters in diameter) and 20 small chambers (1.5 meters in diameter). The small chambers are equipped with lysimeters for water and nutrient balance studies, and for measuring nutrient leaching from the soil profiles.

The Open Top Chambers are used for short- and long-term studies of the effects of air pollution on trees and crop plants.

Both the common control system for the OTC-facility, and the common data logging system have been reconstructed during 1992.

8.6 Risø Ecological Risk Assessment Facility, RERAF

The RERAF-facility, which is being developed in the Environmental Science and Technology Department, will become one of the most advanced systems for conducting genetic, physiological, ecophysiological, biogeochemical and ecological experiments. Natural physical, chemical and biological conditions can be simulated with good approximation. It will be possible to study the biological interactions between different plant species, symbiotic relations between plants and rhizobia and VA-mycorrhiza, and the effect of plant pathogens. The facility will also be well suited for experiments with model ecosystems to validate models.

RERAF will embody 11 growth chambers (4 x 6 meters) planned as north-facing greenhouse cubicles, each equipped with an advanced climate control system which will not only control the amount of light, temperature, and humidity inside the growth chambers, but will also be capable of simulating the diurnal cycle for all three parameters in a near natural manner. The chambers will have mobile light roofs with Hg high pressure lamps, giving a maximum photon-flux of approximately $1000 \text{ } \mu\text{mol m}^{-2} \text{ s}^{-1}$. All C_3 -plants will therefore be light saturated. The chambers will be certified for experiments with radioisotopes and transgenic organisms.

The growth chambers can be supplied with either atmospheric air or special mixtures of gasses such as nitrogen, oxygen, carbon dioxide or air pollutants such as ozone, NO_x and SO_x .

Each of the eleven growth chambers will have a 3 meter deep root compartment with separate temperature controls. The growth medium can be homogenized soil in lysimeters, or monoliths of soils, or nutrient solutions in a continuous flow or recycling system. The system can be used for sand culture or for culture where the plants are grown in artificial rooting media, such as rockwool perlite or polyethylene beads.

The RERAF-facility has planned to combine the latest developments within analytical chemistry and computer technology, with advanced plant growth facilities, where separate root and shoot compartments are being used. In this way, it is possible to operate and independently control experimental conditions of both above- and below-ground parts of model ecosystems.

With the use of the RERAF-facility it is possible to perform fully integrated ecophysiological experiments with higher plants, where climatic conditions, water consumption, assimilation and exchange of nutrients, as well as metabolic gasses, can be controlled and monitored continuously or within short time intervals.

Besides being suited for experiments with higher plants, the facility is also suitable for integrated experiments with submerged plants, macro algae, benthic algae, planktonic algae, and bacterial cultures. Further, the RERAF-facility can be used for experiments with transgenic plants and their relationships with herbivore insects.

The RERAF-facility will be the ultimate tool in a wide range of basic and applied research tasks related to plant species or plant strains. Their germination, ontogenetic development, growth, nutrient demands, and tolerance towards physical, chemical, biological and/or climatic stress can also be studied.

Using the RERAF-facility it will be possible to conduct fully integrated multi-parameter experiments, where whole plants or plant materials are cultured under climatic and nutritional control over long periods of time. The biological material can be tested under steady state or transient conditions, or under conditions simulating the fluctuating natural environment. The climatic and nutritional control will be either by feedback from analytical equipment or from computer models.

9 Publications

9.1 Refereed Journals and Books

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- Nielsen, O.J., J. Sehested. Atmospheric chemistry of HCFCs and HFCs studied at Risø. Kemisk Forenings årsmøde, Odense (DK), 18 Jun 1992.
- Nielsen, S.P. A compartmental model for the coastal waters of the North-East Atlantic. MAST-052C 5th Meeting, Petten (NL), 16-18 Sep 1992.
- Nielsen, S.P. An assessment of the CS-137 inventory in the Baltic Sea using the Evans box model. Seminar on the inventory of artificial radionuclides in the Baltic Sea. 7. Meeting of the group of experts on monitoring of radioactive substances in the Baltic Sea (MORS), Helsinki Commission, Helsinki (FI), 1-5 Jun 1992.
- Nielsen, S.P. Summary of Danish activities relevant to CRESP. CRESP task group meeting, La Spezia (IT), 7-9 Oct 1992.
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- Nielsen, T., A.H. Egeløv, G. Hilbert, C. Lohse, L. Sørensen e. Indian Summer long-range transport of PAN, nitric acid, nitrate and other photochemical products. Annual Meeting of the Danish Center for Atmospheric Research (DCAR) on Nitrogen, Copenhagen, November 11, 1992.
- Nielsen, T., A.H. Egeløv, G. Hilbert, C. Lohse, L. Sørensen f. Composition of NO_x. EUROTRAC Symposium '92 on Photooxidants: Precursors and products, Garmisch-Partenkirchen, March 23-27, 1992.
- Nielsen, T., A.H. Egeløv, G. Hilbert, C. Lohse, L. Sørensen g. Occurrence and composition of atmospheric nitrogen compounds. Annual Meeting of the Danish Center for Atmospheric Research (DAR) on Nitrogen, Copenhagen, November 11, 1992.
- Pagsberg, P. Reactions of atom-radical and radical-radical reactions studied by pulse radiolysis and infrared diode laser spectroscopy. Department of Physical Chemistry. Medical Academy, Wrocław (PL), 3-10 Oct 1992.
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- Petersen, L. The potential and limitations of back-crossing as an introgression tool. Norges Lantbruksuniversitet, Honne (NO), 17 Jan 1992.
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- Roed, J. Resuspension collected by rainsamples in different height. VAMP 4th RCM conference, Vienna (AT), 2-6 Mar 1992.
- Rosendahl, L. Exchange of metabolites across the peribacteroid membrane of pea root nodules. Lund Universitet. Institut for Fysiologisk Botanik, Lund (SE), 30 Jan 1992.
- Rosendahl, L., B.U. Jochimsen. Characterization of proteins in the peribacteroid space of pea root nodules. In: Proceedings from the 9th International Congress on Nitrogen Fixation, Cancun (Mex), 6-12 Dec. 1992.
- Sehested, J., T. Ellermann, O.J. Nielsen, T.J. Wallington. UV-absorption spectrum, and kinetics and mechanism of the self reaction of CHF₂CF₂O₂ radicals in the gas phase. CEC/EUROTRAC atmospheric chemistry discussion meeting, Leuven (BE), 21-24 Sep 1992.
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- Skov, E., H. Wellendorf. Application of RAPD for high-density genome mapping and marker-aided selection in Norway spruce. Conference on molecular biology of forest trees, Carcans-Maubuisson (FR), 15-18 Jun 1992.
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- Solgaard, P. Erfaringer med ICP-MS. Elemental analysis seminars 1992, Solna (SV), 9 Jun.
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- Theilade, B. Cloning characterization and RFLP mapping of the barley peroxidase BP 2A gene. Nordic postgraduate school in plant pathology, Tisvildeleje (DK), 2-9 Feb 1992.
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9.6 Patents

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10 Papers accepted for publication

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- Christensen, H., I. Jakobsen. Reduction of bacterial growth by a VA mycorrhizal fungus in the rhizosphere of cucumber. Biol. Fertil. Soils
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- Giese, H., A.G. Holm-Jensen, H.P. Jensen, J. Jensen. Localization of the 'Laevigatum' powdery mildew resistance gene to barley chromosome 2 by use of RFLP markers. Theor. Appl. Gen.
- Hansen, H., A.G. Olsen, P. Rosenkilde. The effect of Cu²⁺ on osmoregulation in Rainbow trout (*Oncorhynchus Mykiss*) assayed by changes in Plasma salinity and Gill lipid Metabolism. The Science of the Total Environment.
- Hovmøller, M.S., L. Munk, H. Østergård a). Observed and predicted changes in virulence gene frequencies at 11 loci in a local barley powdery mildew population. Phytopathology.
- Hovmøller, M.S., L. Munk, H. Østergård b). Observed and predicted changes in virulence gene frequencies in a local population of *Erysiphe graminis* f.sp. *hordei*. IN: "Durability of Disease Resistances (ed. Th. Jacobs & J.E. Parlevliet).
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- Jensen, E.S., K. Pilegaard. Nitrogen dioxide absorption by barley in open-top chambers. New Phytologist.
- Jensen, H.P., J. Helms Jørgensen. Genetics of 'Laevigatum' resistance and virulence in barley and powdery mildew. Barley Genetic Newsletter 21.
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- Johansen, A., I. Jakobsen, E.S. Jensen a). Hyphal transport by a vesicular-arbuscular mycorrhizal fungus of N applied to the soil as NH₄ or NO₃. Biology & Fertility of Soils.
- Johansen, A., I. Jakobsen, E.S. Jensen c). External hyphae of vesicular-arbuscular mycorrhizal fungi associated with *Trifolium subterraneum* L. 3. Hyphal transport of ³²P and ¹⁵N. New Phytologist.
- Jørgensen, J. Helms a). Genetics of powdery mildew resistance in barley. Critical Reviews in Plant Sciences.
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- Nielsen, T., A.H. Egeløv, G. Hilbert, C. Lohse, L. Sørensen c). Composition of NO_x. In: EUROTRAC Symposium'92 on Photooxidants: Precursors and products. EUROTRAC International Scientific Secretariat, Garmisch-Partenkirchen.
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- Sommer-Larsen, P., T. Bjørnholm, M. Jørgensen, K. Lerstrup, P. Frederiksen, K. Schaumburg, K. Brunfeldt, K. Bechgaard, S. Roth, J. Poplawski, H. Byrne, J. Anders, L. Eriksson, R. Wilbrandt, J. Frederiksen. A Molecular Switch Involving Large Conformational Changes. A Theoretical Study, Molecular Crystals and Liquid Crystals. The Science of the Total Environment.
- Thomsen, S. B.: Byggestrøbesyge med særligt henblik på resistensforhold. M.Sc. Degree Thesis, The Royal Veterinary and Agricultural University and Risø National Laboratory, p. 105.
- Turgeon, G., H. Bohlman, L.M. Ciuffetti, S.K. Christiansen, G. Yang, W. Schäfer, O.C. Yoder. Cloning and analysis of the mating type genes from *Cochliobolus heterostrophus*. Molecular and General Genetics.
- Turgeon, G., S.K. Christiansen, O.C. Yoder. Mating type genes in ascomycetes and their imperfect relatives. Proceedings of the Holomorph Conference, 4-7 Aug. 1992, Newport, Oregon.
- Wilbrandt, R., F.W. Langkilde. Molecular Conformations in Electronic Excited States. In: Laane, J. ed. Structures and Conformations of Non-rigid Molecules.

11 Education

11.1 Ph.D. Thesis

- Johansen, A.*, 1992. Hyphal transport and depletion of soil N by *Glomus intraradices*, a VA-mycorrhizal fungus. Ph.D. Thesis, Risø National Laboratory and University of Copenhagen, 115 p.
- Pedersen, L.H.*, 1992. Callose Synthesis in Barley Leaves. Ph.D. Thesis, Risø National Laboratory and Technical University of Denmark, 82 p.
- Rasmussen, U.*, 1992. Biochemical and molecular studies of a pathogen-induced chitinase in *Brassica napus* L. ssp. *oleifera*. Ph.D. Thesis, Risø National Laboratory and Royal Veterinary and Agricultural University, Copenhagen, 67 p.

11.2 M.Sc. Thesis

- Halken, T.*, 1992. Udfældning af humussyrer. (In Danish). The Technical University of Denmark and Risø National Laboratory, Roskilde.
- Lyngkjær, M.F.*, 1992. Koblingsanalyse og genkortlægning i bygmeldugsvampen (*Erysiphe graminis* f.sp. *hordei*) ved brug af Random Amplified Polymorphic DNA (in Danish). The Royal Veterinary and Agricultural University and Risø National Laboratory, 86 p.
- Quarcoop, E.A.*, 1992. Genes encoding two barley peroxidases induced by the powdery mildew fungus, *Erysiphe graminis* f. sp. *hordei*. The Royal Veterinary and Agricultural University and Risø National Laboratory, p. 62.

11.3 B.Sc. Thesis

- Johansen, B.I., K. Petersen.* 1992. Particulate organic nitrates. (In Danish). Danish Engineering Academy and Risø National Laboratory, Roskilde.
- Jørgensen, L.R.* 1992. Measurements of NO₂ by the KI method and O₃ by the PA method. (In Danish). Danish Engineering Academy and Risø National Laboratory, Roskilde: 173 p.

11.4 External examiners

- Andersen, A.J.* censor in plant nutrition at The Royal Veterinary and Agricultural University.
- Engvild, K.C.* censor in plant physiology at the University of Aarhus and at The Royal Veterinary and Agricultural University.
- Giese, H.* censor in molecular biology at the University of Aarhus and molecular plant pathology at The Royal Veterinary and Agricultural University.
- Haahr, V.* censor in plant breeding at The Royal Veterinary and Agricultural University.
- Jørgensen, J. Helms* censor in plant breeding and plant pathology at The Royal Veterinary and Agricultural University.
- Jørgensen, R.B.* censor in plant tissue culture and molecular biology at the University of Aarhus.
- Linde-Laursen, I.* censor in genetics at The Royal Veterinary and Agricultural University.
- Rasmussen, S.K.* censor at The Royal Veterinary and Agricultural University.
- Skou, J.P.* censor in plant pathology at The Royal Veterinary and Agricultural University.

11.5 Title of honours

- J. Helms Jørgensen*, adjunct Professor at the Royal Veterinary and Agricultural College.
- I. Linde-Laursen*, silvermedal from The Royal Danish Academy of Sciences and Letters.

12 Exchange of Scientists

- Anastasi, Dr C.*, University of York, England (1 week).
- Bajdor, Dr K.*, Industrial Chemistry Research Institute, Poland (3 months).
- Barthiewicz, M.Sc. E.*, Agricultural and Teachers University, Siedlce, Poland (1 week).
- Barton, Geophysicist K.*, University College Galway, Ireland (2 months).
- Brouwer, Dr A.M.*, University of Amsterdam, Holland (4 weeks).
- Carroll, D.P.*, University College Dublin, Ireland (1 month).
- Christensen, Professor H.*, Studsvik Energiteknik AB, Sweden (3 weeks).
- Daniellou, Stud. V.*, INRA, Centre de Grignon, France (1 week).
- Douka, Dr K.*, National Center for Scientific Research "Demokritos", Athens, Greece (2 months).
- Getoff, Professor N.*, University of Vienna, Austria (3 weeks).
- Hickel, Dr. B.*, CNRS, Saclay, France (1 week).
- Ikram, Dr. A.*, Microbiol. Division, The Rubber Research Institute of Malaysia (RRIM) (2 months).
- Jakobsen, I.*, Visiting Research Fellow at The University of Western Australia (2 months).
- Joner, Stud.dr.scient., E.*, Agricultural University of Norway (5 months).
- Jungkamp, Tim*, University of Kiel, Germany (1 week).
- Kamens, Professor R.M.*, North Carolina University, USA. (2 weeks).
- Kelly, C.*, University College Dublin, Ireland (2 weeks).
- Keszthelyi, T.*, Technical University, Budapest, Hungary (3 months).
- Kläning, Dr. U.*, Aarhus University, Denmark (2 weeks).
- Koch, Stud. G.*, Justus Liebig Universität Gießen, Germany (1 week).
- McLaughlin, Dr. W.L.*, National Institute of Standards and Technology, Gaithersburg MD, USA (4 weeks).
- Mellor, Dr. R.B.*, EC fellowship, Göttingen, Germany (3 months).
- Mod Ali, N.*, Nuclear Energy Unit, Malaysia (3 weeks).
- Mordzinski, Dr. A.*, Polish Academy of Sciences, Poland (6 weeks).
- Ngamchuen, Dr. R.*, Kasetsart University, Department of Agronomy, Thailand (8 months).
- Pearson, Dr. J.N.*, School of Agriculture, The University of Western Australia, Perth (2 months).
- Pecchioni, Dr. N.*, Sperimentale per la Cerealicoltura, Fiorenzuola d'Arda, Italy (1 week).
- Polikarpov, Academician G.*, Inst. of Biology of the Southern Seas Academy of the Science of the Ukraine, Sevastopol, Ukraine (1 week).
- Pozolotina, Dr. V.*, Institute of Industrial Ecology, Ekaterinburg, Russia (2 month).
- Ramirez, T.*, Instituto de Ciencias Nucleares, Quito, Ecuador (1 month).
- Ratajczak, Dr. E.*, University of Wrocław, Poland (6 weeks).
- Sidebottom, Dr. H.*, University College Dublin, Ireland (1 week).
- Simpson, Dr. V.J.*, University of York, England (1 week).
- Solar, Professor S.*, University of Vienna, Austria (3 weeks).
- Walker, Dr. M.*, National Institute of Standards and Technology, Gaithersburg MD, USA (1 week).
- Wallington, Dr. T.*, Ford Motor Co., Dearborn, USA (2 weeks).
- Wille, U.*, University of Kiel, Germany (1 week).
- Yamamoto, H.*, Japan Atomic Energy Research Institute (1 year).
- Zhai, Dr. X.*, China Institute of Atomic Energy, China (4 months).
- Zlobenko, B.*, Institute of Geochemistry and Physics of Minerals, Ukrainian Academy of Science (1 month).

13 Main cooperative Projects

Risø project coordinators are marked with *
Principal investigators are marked with **

Radioactive tracers in the Greenland Sea. Part of the international Greenland Sea Project. *H. Dahlgaard*.

Studies of the Transport of Coastal Water from the English Channel to the Baltic Sea using Radioactive Tracers. Marine Science and Technology Programme (MAST), CEC. Cooperative project including Risø National Laboratory; Federal Maritime and Hydrographic Agency, Germany; Netherlands Energy Research Foundation; Commissariat à l'Energie Atomique, France Ministry of Agriculture, Fisheries and Food, Lowestoft, UK; and Centre de Spectrométrie Nucléaire et de Spectrométrie de Masse, CNRS, Orsay, France. *H. Dahlgaard***, *S.P. Nielsen*.

Nordic Radioecology Cooperation (NKS/RAD). RAD1 - Education, Quality Assurance, Methodology; RAD2 - Aquatic Radioecology; RAD3 - Agricultural Ecosystems; RAD4 - Natural Ecosystems. *A. Aarkrog*, *H. Dahlgaard***, *S.P. Nielsen*.

Radioecology of seminatural ecosystems (CEC). Cooperative project including Risø National Laboratory, (DK), Nuclear Energy Board (IR) (Coordination), Institute of Terrestrial Ecology, Merlewood (UK), Swedish University of Agricultural Science, Uppsala (S), University of Thessaloniki (G). *A. Aarkrog*, *S.P. Nielsen**.

Exemption of radiation sources and practices from regulatory control for disposal into the marine environment. International Atomic Energy Agency, Vienna. *S.P. Nielsen*.

Pilot project on radioactive environmental problems in the River Ploucnice. Cooperative project with Danish Hydraulic Institute and Water Quality Institute. *S.P. Nielsen*.

Effects of countermeasures on radiation doses to critical groups from contaminated food after a release of radioactive material from Ringhals Power Station. Cooperative project with Vattenfall AB and the Swedish Radiation Protection Institute. *H. Yamamoto and S.P. Nielsen*.

Risø's integrated environmental project (RIMI). *O.J. Nielsen*, *T. Nielsen*, *K. Pilegaard**, *P. Solgaard*.

Uptake of Nitrogen compound by above ground plant parts. Collaborator: National Environmental Research Institute DK. *K. Pilegaard**.

Biological monitoring of heavy metals. Collaborator: Laboratory of Environmental Research Institute. Science and Education, The Technical University, Lyngby. *K. Pilegaard**.

EUROTRAC-TRACT, Transport of pollutants over complex terrain. *K. Pilegaard**.

EUROTRAC-BIATEX. Biosphere/Atmosphere Exchange of Pollutants. *K. Pilegaard**.

EC-project on Deposition in Urban Areas. Collaborators: UKAEA, Harwell, GSF, Munich. *J. Roed*.

EC-CIS Project on Countermeasures. CHECIR. Main collaborators: Cadarache, France; IGPM, Kiev. *J. Roed**.

EC-project on Urban Decontamination. Collaborators: UKAEA, Harwell, Catholic University, Leuven, Glasgow University. *J. Roed**, *K. Andersson*.

EC-project on Indoor Deposition. Collaborator: Imperial College, London. *J. Roed***.

EC-project on Weathering and Resuspension. Collaborators: UKAEA, Colchester, GSF, Munich. *J. Roed**.

EC-project on Development of Skim and Burial Plough. Part of RESSAC; Main collaborator: Cadarache, France. *J. Roed***.

EC-Project on Deposition of artificial radionuclides, their subsequent re-location in the environment and implication for radiation exposure. Collaborators: GSF, Munich; NRPB England and Imperial College, London. *J. Roed**, *Chr. Lange*.

NKS-project on Waste Created by Clean Up of an Urban Area. Collaborators: Helsinki University, SIS Oslo, SSI Sweden. *J. Roed*.

NKS-Project BER 3. Countermeasures. Collaborators: SSI, Stockholm, SIS, Oslo, STUK, Helsinki and Swedish University of Agricultural Science, Uppsala. *J. Roed*.

IAEA Project on Validation of Model Parameters VAMP. *J. Roed***.

SNF-project on Cretaceous-Tertiary boundary studies by Copenhagen University. *H. Kunzendorf**.

Trace element studies in sediments from Skagerrak-Kattegat area. Collaborators: Geological Survey of Denmark and University of Gothenburg, Sweden. *H. Kunzendorf**, *P. Solgaard*.

Baltic Marine Cooperation. Marine environmental studies in the Baltic Sea. Collaborators: Roskilde University Center and Danish Road Laboratory. *H. Kunzendorf**.

DCAR-EUROTRAC-TOR. Collaborators: National Environmental Research Institute, Meteorology and Wind Energy Department. *T. Nielsen***.

FLAIR Concerted Action No 10. The Measurement of Micronutrient Absorption and Status. Danish collaborators: Research Department of Human Nutrition and National Institute of Animal Science. *P. Solgaard**.

FØTEK. Collaborators: Levnedsmiddelstyrelsens Centrallaboratorier and others. *P. Solgaard**.

DCAR-Traffic PAH. Collaborators: National Environmental Research Institute, Institute of Toxicology, DK-Technics, Meteorology and Wind Energy Department. *T. Nielsen***, *P. Solgaard*.

NMR-Photochemical oxidants. Danish Collaborators: National Environmental Research Institute, Meteorology and Wind Energy Department. *T. Nielsen**.

SMP-Nitrogen-Project. Collaborators: National Environmental Research Institute, University of Odense. *T. Nielsen***.

Treatment of wastes. Collaborators: NKT Research Center and Canadian Occidental. *E. Sørensen**, *A.B. Bjerre*.

Colloid Behaviour. A Joint CEC project within the MIRAGE group. *B. Skytte Jensen**.

Uncertainties in the Modelling of Migration Phenomena. A joint CEC project within the MIRAGE group. *B. Skytte Jensen**.

MODECS. Molecular Design of Chemical System. An R&D-forum for industries and institutions interested in the mentioned topic. *B. Skytte Jensen***.

EC-project on Atmospheric chemistry of halogenated compounds. 9 European laboratories. *O.J. Nielsen*.

EUROTRAC-LACTOZ. Laboratory Studies of Chemistry Related to Tropospheric Ozone. *O.J. Nielsen*.

Ford Motor Company collaborative project on CFC substitutes, *O.J. Nielsen*.

NATO Collaborative Research grant Denmark *R. Wilbrandt**, Italy *G. Orlandi*, The Netherlands *A.M. Brouwer*, *H.J.C. Jacobs*, *J. Lugtenburg*.

Investigation of MOLSWITCH compounds. CSMI University of Copenhagen, *K. Bechgaard* and *Risø R. Wilbrandt**.

Danish Natural Science Research Council, project on the characterization of potential energy surfaces of molecules and radicals. Risø *R. Wilbrandt**, Royal Danish School of Pharmacy *F.W. Langkilde*, Odense University *O.S. Mortensen*.

Danish Natural Science Research Council, project on atmospheric chemistry. Copenhagen University and National Environmental Research Institute. *O.J. Nielsen*.

FUOTRAC-TOR, Tropospheric ozone research. Danish collaborator: National Environmental Research Institute. *T. Nielsen**.

EUROMET. Collaboration with National Physical Laboratory, UK on calibration and measurements of ionizing radiation. *A. Miller*.

IAEA. Research agreement no 6919: Reference dosimetry for industrial Electron Accelerators. *A. Miller*.

IAEA. Research agreement no 6612: Methods for measurement of absorbed dose and dose distribution at 200-1000 keV electron beams. *A. Miller*.

IAEA. Research Contract no 5508/R1/RB: Development of educational computer programme for simulation of radiation-induced chemical kinetics. Final report. *E. Bjerghakke*, *O. Lang Rasmussen*, *P. Kirkegaard*.

Assessment of air pollutant effects on growth, quality and metabolism of agricultural crops. Subcontract to NERI EEC collaboration. *K.C. Engvild*.

EC project on the ammonium hypothesis of spruce decline. Subcontract to the National Forest and Nature Agency. *K.C. Engvild*.

Computer Aided Farming (CAF). Collaborator: Foulum Research Station, Research Centre of Agriculture, Skejby. *V. Haahr*, *A. Jensen*.

Mycorrhizal fungi and saprophytic soil microbes. Collaborator: B. Söderström, University of Lund. *I. Jakobsen*.

Nutrient transport in VA mycorrhizas. Collaborators: L.K. Abbott, A.D. Robson and J.N. Pearson, The University of Western Australia with Grains Research and Development Corporation, Canberra. *I. Jakobsen*.

Mikrobielle processer i rodzonen i relation til planternes forsyning med næringsstoffer 1988-1991 (Microbial processes in the root zone in relation to supply of plants with nutrients). Collaborators: Seven Danish research laboratories. *I. Jakobsen**, *A. Johansen*.

Evaluating the role of legume cover crops in the nitrogen nutrition of rubber 1989-1993. CE project under The International Scientific Co-operation Programme. Collaborator: The Rubber Research Institute of Malaysia, Kuala Lumpur. *I. Jakobsen*, *E.S. Jensen**.

Kombineret halmnedbrydning og efterafgrødedyrkning 1989-1992 (Combined straw incorporation and cultivation of catch crops 1989-1992). Collaborators: I. Thomsen and B.T. Christensen, Government Research Station Askov, Danish Research Service for Plant and Soil Science. *E.S. Jensen*.

Nutritional therapy of patients with liver cirrhosis 1990-1992. Collaborator: K. Nielsen, Medical Department A, Rigshospitalet, Copenhagen. *E.S. Jensen*.

Danish Strategic Coordinated Environmental Research: Programme: Influence of crop residue particle size and spatial distribution in soil on the mineralization-immobilization turnover, plant uptake and losses of nitrogen. 1992-1996. Collaborators: Danish Centre for Root Zone processes, nutrient losses from agro-ecosystem and processes in riparian areas. *E.S. Jensen*.

Research programme: Sustainable Agriculture Project: Cycling of nitrogen from animal manure and plant residues. 1992-1996. Collaborator: Department of Plant Nutrition and -Physiology, Danish Research Service for Plant and Soil Science. *P. Sørensen*, *E.S. Jensen**.

Exchange of metabolites across the peribacteroid membrane in legume root nodules 1989-1992. Collaborators: M.J. Dilworth and A.R. Glenn, Murdoch University, Western Australia. *L. Rosendahl*.

SJVF project "Plant-microbe interactions". Collaborators: University of Aarhus, University of Lund, University of Stockholm, University of Minnesota, University of Western Australia. *L. Rosendahl*.

Compound exchange in symbiotic nitrogen fixation. Collaborator: B.U. Jochimsen, University of Aarhus. *L. Rosendahl*.

EC-project "Characterization of the peribacteroid space from 3 types of legume nodules". Collaborator: University of Geneva. *R.B. Mellor, L. Rosendahl**.

Interaction between crop plants and weeds. Collaborator: B. Søgaard, The Royal Veterinary and Agricultural University. *H. Doll*.

Efficiency of partial mildew resistance in barley and correlation with secondary metabolites in the leaves. Collaborator: B. Søgaard, The Royal Veterinary and Agricultural University. *H. Doll**.

Linkage map of the barley genome. Collaborators: The Royal Veterinary and Agricultural University; the Danish plant breeding stations: Ahd, Sejet, Pajbjerg; European and American barley geneticists. *H. Giese, J. Jensen, L. Petersen, S.K. Rasmussen*.

Molecular genetics of *Erysiphe graminis* f.sp. *hordei*, the causal agent of barley powdery mildew. Collaborators: Danish Plant biotechnology Research Center; The Royal Veterinary and Agricultural University; J. McDermott, ETH Zentrum, Zurich. *H. Giese, M. Lyngkjær, L. Borbye, M. Rasmussen, S. Christiansen*.

Genetic variation between cotton varieties. Collaborator: The Royal Veterinary and Agricultural University. *H. Giese, B. Kolling*.

Genetic markers for embryogenesis in barley. Collaborators: University of Copenhagen; D.

Bowles, Leeds University, U.K. *H. Giese, E.T. Larsen*.

QTLs (Quantitative Trait Loci) in barley. Collaborator: The Royal Veterinary and Agricultural University. *J. Jensen, B. Kjær*.

Improvement of protein quality in barley. Collaborator: B.O. Eggum, Foulum Research Station; J. Hejgaard, Technical University of Copenhagen. *J. Jensen, S.K. Rasmussen*.

Evaluation of cereal genetic sources in the Nordic Gene Bank, 1990-. Collaborators: Nordic Gene Bank, Alnarp, Svalöf AB, Sweden; Hankkija, Finland. *J.H. Jørgensen*.

New sources of powdery mildew resistance genes in barley, 1989-. Collaborators: Three Danish cereal breeding companies. *J.H. Jørgensen, H.P. Jensen*.

Risk assessment of genetically modified plants. Collaborators: A. Buchter-Larsen, Maribo Seed; J. Fredshavn, The Royal Veterinary and Agricultural University; F.B. Christiansen, Aarhus University; S. Mark, National Forest and Nature Agency. *R.B. Jørgensen*, H. Østergård*, T. Mikkelsen, L. Landbo*.

Recombination estimates and physical/chemical distances on the chromosomes. Collaborator: The Royal Veterinary and Agricultural University. *I. Linde-Laursen, H. Giese, C. Pedersen*.

Polyploid *Hordeum* species. Collaborators: R. von Bothmer, Department of Crop Genetics and Breeding, The Swedish University of Agricultural Sciences, Svalöv, Sweden. *I. Linde-Laursen*.

Barley peroxidases. Collaborators: Aarhus University; Danish Plant Biotechnology Programme. *S.K. Rasmussen, B. Theilade*.

Genetics of resistance to barley leaf stripe. Collaborator: B.J. Nielsen, Plant Protection Institute, Lyngby. *J.P. Skou, V. Haahr*.

Linkage map of Norway spruce. Collaborator: H. Wellendorf, The Arboretum, Hørsholm. *E. Skov*, H. Giese*.

Population biology of airborne pathogens on cereals. Collaborators: M. Hovmøller, Bent Nielsen, Research Centre for Plant Protection, and L. Munk, The Royal Veterinary and Agricultural University. *H. Østergård, J.H. Jørgensen, H.P. Jensen.*

Oil Seed Rape Initiative. Collaborators: O. Rasmussen, University of Aarhus, B. Eggum, Foulum Research Station; J. Schjørring, The Royal Veterinary and Agricultural University; P. Ulvskov, Research Centre for Plant Protection, *H. Giese.*

14 Guest Lectures

The list includes only guest lectures from foreign countries

Dr K. Bajdor, Warsaw, Poland: "Resonance Raman study of the benzyl radical".

Dr W.M. Bartczak, Institute of Applied Radiation Chemistry, Łódź, Poland: "Recent computer experiments in different subfields of radiation chemistry: track effects, scavenging, charge recombination in inhomogeneous media".

Geophysicist K. Barton, University College Galway, Ireland: "Geochemical and Geophysical methods in environmental research in Ireland".

Dr F. Brouwer, University of Amsterdam, Holland: "Light-induced electron transfer in multi-component electron donor-acceptor systems".

Dr M. Foryś, Agricultural and Teachers University, Siedlce, Poland: "Thermal electron mobility and electron capture processes".

Dr A. Grabowska, Institute of Physical Chemistry of the Polish Academy of Sciences, Warsaw, Poland: "Strongly fluorescent phototautomers of bipyridyl-diols and their precursors. Symmetry conditions for efficient radiative process".

Dr C. Hackett, Scottish Crop Research Institute: "The use of normal mixture models to map quantitative trait loci in barley".

Dr J. Kaleciński, W. Trzebiatowski Institute, Wrocław, Poland: "NO₃ radicals in aqueous systems – optical investigation".

Professor R.M. Kamens, North Carolina University, U.S.A.: "Transformation of PAH and formation and stability of nitro-PAH".

Professor R.M. Kamens, North Carolina University, U.S.A.: "Gas/particle distribution of semi-volatile organic compounds".

Professor R.M. Kamens, North Carolina University, U.S.A.: "Aerosol Chamber".

Professor R.M. Kamens, North Carolina University, U.S.A.: "Acid Aerosols".

Dr G. Koch, Justus Liebig Universität, Giessen: "Genetic variability of barley powdery mildew".

Dr W.L. McLaughlin, National Institute of Standards and Technology, Gaithersburg, USA: "Polymeric fluorenes clad in fluorenes: Radiation effects on fiberoptic properties".

Dr A. Mordzinski, Institute of Physical Chemistry, Warsaw, Poland: "On the bottle-neck in the ground state repopulation of some electronically excited H-bonded molecules".

Dr P. Panta, Institute of Nuclear chemistry and Technology, Warsaw, Poland: "Gas phase dosimetry connected with radiation processing of industrial flue gas by electron beam installed at INChT and Power Station EC Kaweczyn".

Dr N. Pecchioni, I. Sperimentale per la Cerealicoltura, Fiorenzuola d'Arda, Italy: "RFLP analysis of highly polymorphic loci in barley".

Professor E. Ratajczak, Institute of Chemistry and Physics, Medical Academy, Wrocław, Poland: "Kinetics of CH₃ radical reactions studied by pulse radiolysis combined with infrared diode laser spectroscopy".

Professor R.N. Schindler, Institut für Physikalische Chemie, Universität Kiel, Germany: "The Chemistry of NO₃ Radicals in the Gas Phase".

Dr E. Szajdzirska-Pietek, Institute of Applied Radiation Chemistry, Łódź, Poland: "Reactivity of hydrated electron towards acceptors bound to cationic micelles. Pulse radiolytic investigation".

Dr D. Whisson, Department of Agriculture, Glen Osmond, Australien: "Extreme pathogenic variability in *Rhynchosporium secalis*: Possible mechanisms".

Dr Z. Zimek, Institute of Nuclear Chemistry and Technology, Warsaw, Poland: "Laboratory and demonstrations facilities for flue gas treatment by electron beam installed at InChT and Power Station EC Kaweczyn".

15 Committee Memberships

15.1 National

- Aarkrog, A.* Danish National Council for Oceanology.
Danish reference group for CEC's MAST programme.
Danish reference group for CEC's Radiation protection Programme.
AMAP's Oceanographic research Group.
- Dahlgaard, H.* Greenland Sea Project, Danish Working Group.
- Giese, H.* The Danish Agricultural and Veterinary Research Council.
Board of The Institute of Forest and Landscape.
Danish Society for the Conservation of Nature, Committee for Environmental Issues.
- Gissel-Nielsen, G.* Vice chairman of The Danish Agricultural and Veterinary Research Council.
Board of the Danish Academy of Technical Sciences.
Board of the National Research Service for Soil and Plant Sciences.
Danish Society for the Conservation of Nature, Committee for Environmental Issues.
- Jørgensen, J.H.* Dansk Genbanknævn.
Koordineringsgruppen for resistens og virulens i korn og komsygdomme.
- Miller, A.* Danish Medical Device Association (DMDA). Sterilization committee (formerly DUFO).
- Nielsen, O.J.* The National Committee for The International Geosphere-Biosphere Programme (IGBP).
- Pilegaard, K.* Danish Centre for Atmospheric Research (DCAR). Working Group on effects. (Chairman).
- Skou, J.P.* Danish Society of Plant Pathology (chairman of Committee of Nomenclature).
- Solgaard, P.* Dansk Standardiserings Råd. S-168-Vandundersøgelse, FaU 213.
- Østergård, H.* Programme Committee for Biotechnology Programme "Risk of introducing genetically modified plants into the environment" (head of programme).
Danish reference group for OECD's "Security in Relation to Biotechnology".

15.2 International

- Aarkrog, A.* IAEA CRP MARDOS (Chairman).
IAEA VAMP programme. (Acting chairman of terrestrial group).
CGC for CEC's Radiation Protection Programme.
Reference group NKS-RAD Programme.
EF MARINA MED programme. (Chairman of the datagroup).
Programme Committee for International Conference on "Environmental Radioactivity in the Arctic and Antarctic". Kirkenes, Norway, Aug. 1993.
Programme Committee for International Symposium on "Radiological aspects related to remediation and restoration of radioactive contaminated sites". Antwerp, Belgium, Oct. 1993.
Editorial board of Journal of Environmental Radioactivity.
- Dahlgaard, H.* Baltic Marine Environment Protection Commission, Helsinki Commission (HELCOM). Group of Experts on Monitoring of Radioactive Substances in the Baltic Sea (MORS).
Marine Radiocology Working Group (MARECO), International Union of Radioecologists (IUR). (Working Group Leader).
Working Group on a Nordic participation in the World Ocean Circulation Experiment (WOCE).
Nordic Nuclear Safety Project (NKS), Radioecology programme (RAD). (Coordinator).
- Engvild, K.C.* Member of Journal Committee of 'Physiologia Plantarum'.
- Gissel-Nielsen, G.* OECD committee for plant/soil/microbial interactions.
- Jakobsen, I.* Management committee for COST Action 810: The role of VA-mycorrhizae in transformation of matter in the soil and their importance for plant nutrition and plant health.
- Jensen, J.* International coordinator, barley chromosome 5; International Barley Nomenclature Committee.
- Jensen, B. Skytte.* Working Groups within the EEC R&D Programme Management and

- Storage of Radioactive Waste (MIRAGE), in the subcommittees CHEMVAL, COCO, and Natural Analogues.
Member of WP Geological Disposal of Radioactive Waste. The Editorial Board of "Waste Management".
- Jensen, E.S.* Editorial Board of 'Plant and Soil'.
Sectionboard for Soils and Fertilizers, Scandinavian Association of Agricultural Scientists.
- Jørgensen, J.H.* Editorial board of 'Euphytica'.
International coordinator for disease and pest resistance genes in barley.
Working group on cereal disease resistance biology, Scandinavian Association of Agricultural Scientists.
Working group cereal, Nordic Gene Bank. (Chairman).
Working group barley, European Coordinated Programme on Genetic Resources.
Coordinating Committee, International Barley Genetics Resources Network.
- Kunzendorf, H.* Editorial Board of 'Marine Mining'.
Member of the planning group for a new Technology Center for Marine Minerals.
Reviewer of Marine Chemistry, Marine Geology, Marine Mining, Mineralium Deposita and Talanta.
- Linde-Laursen, I.* Editorial Board of 'Plant Systematics and Evolution'.
- Miller, A.* American Society for Testing and Materials: Subcommittee E10.01: Dosimetry for Radiation Processing.
Radiation Physics and Chemistry (Pergamon Press): Editorial Board.
Radiation Sterilization (Israel): Editorial Board.
Int. Meeting on Radiation Processing, Beijing, 1992: Programme Committee.
- Nielsen, O.J.* COST-611 Working Party 2 Steering Committee, Atmospheric and Photochemical Processes.
- Nielsen, O.J.* COST-611 Concertation Committee on Physico-Chemical Behaviour of Atmospheric Pollutants.
- Nielsen, S.P.* International Committee on Radionuclide Metrology.
Articles 35 and 36 of the Euratom Treaty (Environmental Monitoring).
Baltic Marine Environment Protection Commission, Helsinki Commission (HELCOM).
Group of experts on Monitoring of Radioactive substances in the Baltic Sea (MORS).
- Nielsen, T.* The board of Nordic Society of Aerosols (NOSA).
- Roed, J.* The Group on in-situ measurements (ICRU).
IAEA and EEC VAMP Project Urban Group. (Chairman).
The Fuel Cycle Safety Group (OECD).
SCOPE RADPATH.
- Skou, J.P.* Section for Plant Protection (Chairman), Scandinavian Association of Agricultural Scientists.
- Wilbrandt, R.* International Organization Committee on the Conference of Time-resolved Vibrational spectroscopy, Berlin, Germany.
International Organization Committee on the Conference of Time-resolved Vibrational Spectroscopy, Tokyo, Japan.
International Organization Committee on the Conference on the Spectroscopy of Biological Molecules, Athens, Greece.
International Organization Committee on the Conference on the Spectroscopy of Biological Molecules, York, England.

16 Seminars Organized

The sixth Nordic Radioecology Seminar, Tórshavn, Faroe Island. 14–18 Jun 1992. (*H. Dahlgård*).

5th Nordic ICP-MS User Meeting, Risø, 26–27 October 1992. (*P. Solgaard*).

12th Danish-Polish Meeting on Radiation Chemistry. Section of Chemical Reactivity, 20–24 May (*O.J. Nielsen*).

RFLP course for Danish Plant Breeders. Section of Plant Biology, 18–22 May, 1992. (*H. Giese*).

DNA-methods to assay genetic variation in relation to gene mapping and population studies, 23 April, 1992. (*H. Østergård*).

Population biological aspects of risk assessment, 21 October, 1992. (*H. Østergård*).

17 Personnel

The list also includes short term employees

Head of Department

Arne Jensen

17.1 Scientific Staff

Aarkrog, Asker
Andersen, Arne J.
Andersson, Kasper
Bjergbakke, Erling
Bjerre, Anne Belinda
Christiansen, Solveig Krogh
Dahlgaard, Henning
Doll, Hans
Engvild, Kjeld C.
Fenger, Jørgen
Giese, Henriette
Gissel Nielsen, Gunnar
Gundersen, Vagn
Hansen, Heinz H.
Hansen, Knud B.
Holcman, Jerzy
Holm, Ulla
Haahr, Vagner
Jakobsen, Iver
Jensen, Hans Peter
Jensen, Erik Steen
Jensen, Jens
Jensen, Bror Skytte
Johansen, Anders
Juhl, Lone Finnerup
Jørgensen, Jørgen Helms
Jørgensen, Rikke Bagger
Kläning, Ulrik
Kunzendorf, Helmer
Linde-Laursen, Ib
Lis Værgman
Lynggård, Bent
Miller, Arne
Nielsen, Torben
Nielsen, Ole John
Nielsen, Sven P.
Nilsson, Karen
Pagsberg, Palle
Pilegaard, Kim
Qiangjiang, Chen
Rasmussen, Eshen Lindegård

Rasmussen, Søren Kjærsgård
Roed, Jørn
Rosendahl, Lis
Schedsted, Knud
Sillesen, Alfred Heegaard
Skou, Jens Peder
Solgaard, Per
Sørensen, Emil
Wilbrandt, Robert
Østergård, Hanne

17.2 Technical Staff

Andersen, Margit Elm
Andersen, Fritz
Andersen, Bente
Andersen, Lis Brandt
Andersen, Esther
Bitsch, Gunnar
Brandstrup, Oda
Brink Jensen, Merete
Baade-Pedersen, Pearl
Christensen, Tove
Christensen, Hanne Bay
Clausen, Jytte
Corfitzen, Hanne
Djurdjevic, Stanko
Dyrgaard Jensen, Lone
Ebling, Elise
Femqvist, Tomas
Gade, Poul
Green, Jytte
Hansen, Ina
Hansen, Carl
Hansen, Elly
Hansen, Jakob Egede
Hasselbalch, Finn
Henriksen, Ebbe
Holm-Jensen, Anne Grethe
Hougaard, Henrik
Ibsen, Elly
Jensen, Ellen Møller

Jensen, Karen Mandrup
 Jensen, Hanne
 Jensen, Christina Falk
 Johansen, Torben
 Jørgensen, Vibeke
 Jørgensen, Ole
 Jørgensen, Ole Emil
 Karlsen, Aage
 Kjølhed, Alice
 Kristiansen, Bo
 Larsen, Erik Engholm
 Larsen, Fritz
 Larsen, Inge Merete
 Larsen, Hanne Egerup
 Larsen, Ingelis
 Lilholt, Ulla
 Lindskou, Fini
 Madsen, Michael Birch
 Meltofte, Liselotte
 Nielsen, Jette Bruun
 Nielsen, Svend
 Nielsen, Ivar Rønn
 Nielsen, Vagn Aage
 Nikolaisen, Anette
 Olsen, Anette
 Olsen, Inge
 Olsen, Svend K.
 Petersen, Susanne
 Prip, Henrik
 Rasmussen, Charlotte
 Ravnskov, Sabine
 Schmidt, Lars Christian
 Sillesen, Anerikke
 Sørensen, Poul
 Vestesen, Hans
 Vinther, Lis
 Vinther, Niels
 Wojtaszewski, Hanne
 Yu, Yixuan

17.3 Office Staff

Andersen, Annie
 Bay, Kirsten
 Bækmark, Anni
 Frandsen, Anette
 Jakobsen, Inger
 Komerup, Berit
 Kristensen, Ingrid
 Krogh, Helle

Larsen, Aase Neve
 Madsen, Ruth
 Nielsen, Margit
 Petersen, Lis

17.4 Ph.D. Students

Borbye, Lisbeth
 Christiansen, Jens H.
 Frederiksen, John Myhre
 Johansen, Anders
 Justesen, Annemarie
 Jørgensen, Birgitte Lund
 Kjær, Birgitte
 Landbo, Lars
 Løge, Christian
 Larsen, Else Toftdahl
 Løgager, Tine
 Markert, Frank
 Mikkelsen, Thomas
 Olsen, Allan Gylling
 Pedersen, Carsten
 Pedersen, Lars H.
 Petersen, Lene
 Rasmussen, Merete
 Sehested, Jens
 Skov, Elise
 Strandberg, Morten
 Sørensen, Peter
 Theilade, Bodil

17.5 M.Sc. Students

Behrens, Anette
 Frello, Stefan
 Halken, Thomas
 Hansen, Kit Ramshøj
 Kolling, Bent
 Lyngkjær, Michael
 Quarcoo, Eric
 Sørensen, Lisbeth Dahl
 Thomsen, Susanne Brun

17.6 B.Sc. Students

Danielsen, Thomas
 Johansen, Brynjalvur
 Jørgensen, Lene

Klock, Jørgen
Kofoed, Pernille
Krarup, Helene
Petersen, Klaus
Simonsen, Tina
Thomsen, Solgerd

17.7 Apprentices

Birkebæk Larsen, Nille
Hassel, Maja
Jensen, Christina Falk
Jensen, Lene
Jensen, Thomas Nygaard
Jespersen, Charlotte
Korgaard, Charlotte
Lauritzen, Lisbeth
Møller, Margit
Møller, Margit
Olsen, Su
Olsen, Susanne Oxholm
Pedersen, Dorte Juul
Schønning, Liza
Thomsen, Susanne

18 Acronyms

AA:	Atomic Absorption
AAS:	Atomic Absorption Spectrometry
AMAP:	Arctic Monitoring & Assessment Programme
BaP:	Benzo(a)pyrene
BCR:	Community Bureau of Reference
BcP:	Benzo(c)pyrene
BghiP:	Benzo(ghi)perylene
BIATEX:	Biosphere-Atmosphere Exchange of Pollutants
BIOMOV5:	Biospheric Model Validation Study
BMC:	Baltic Marine Cooperation
BRE:	Building Research Establishment
CAT:	Center for Avanceret Teknologi
CEC:	Commission of European Communities
CEN:	European Standard Organization
CGC:	Management and Coordination Advisory Committee
CIS:	Commonwealth of Independent States
CISMI:	Center for Interdisciplinary Studies of Molecular Interactions
Cor:	Coronene
CRESP:	Coordinated Research and Environmental Surveillance Programme (under NEA)
DANAK:	Danish Accreditation Scheme
DCAR:	Danish Centre for Atmospheric Research
DF:	Decontamination Factor
DH:	Chromosome-Doubled Haploid
DIA:	Denmark's Academy for Engineering
DMDA:	Danish Medical Device Association
DMU:	Danmarks Miljøundersøgelser
DNA:	Dioxyribonucleic acid
DTH:	Technical University of Denmark
EC:	European Communities
EEC:	European Economic Communities
ETV:	Electro Thermal Vaporisation
EUROTRAC:	European Experiment on Transport and Transformation of Environmental Relevant Trace Constituents of Anthropogenic and Natural Origin.
FLAIR:	Food-Linked Agro-Industrial Research
FØTEK:	Fødevare-Teknologi-Projekt
GMP:	Genetically Modified Plants
GRECA:	Group of Experts on accident Consequences (under NEA, OECD)
GSF:	Gesellschaft für Strahlen – und Umweltforschung
GSP:	Greenland Sea Project
HDRL:	High Dose Reference Laboratory
HELCOM:	Helsinki Commission
HPLC:	High Performance Liquid Chromatography
IAEA:	International Atomic Energy Agency
IBSS:	Institute of Biology of the Southern Seas (USSR)
IC:	Ion Chromatography
ICRU:	International Commission on Radiological Units
ICPMS:	Inductively Coupled Plasma Mass Spectrometry
IEPA:	Inst. of Ecology of Plant and Animals
IFE:	Institute for Energy Technology Norway
ISO:	International Standard Organization

IUR:	International Union of Radioecologists
KEENEX	Kinetic Evaluation for Combustion using Experimental and Numerical Expertise
LACTOZ:	Laboratory Studies of Chemistry Related to Tropospheric Ozone
MARDOS:	IAEA CRP on "Sources of Radioactivity in the Marine Environment and their Relative Contributors to Overall Dose Assessment from Marine Radioactivity"
MARECO:	Marine Radioecology Working Group (under IUR)
MARIA:	Methods for Assessing the Radiological (Impact of Accidents. (Research programme under CEC)
MARINA-MED:	Overall Radiological Impact on Population of Member States of Natural and Man-made Radionuclides Present in the Mediterranean
MAST:	Marine Science and Technology Programme (under CEC)
MODECS:	Molecular Design of Chemical Systems
MORS:	Group of Experts Monitoring of Radioactive Substances in the Baltic Sea.
NEA:	Nuclear Energy Agency (under OECD)
NIST:	National Institute of Standards and Technology
NKA:	Nordic Liason Committee for Atomic Energy
NKS:	Nordic Nuclear Safety Project (RAD)
NLVF:	Norway Agricultural Research Council
NMR:	Nordisk Ministerråd
NOR:	Nucleolus Organizer Region
NPP:	Nuclear Power Plant
OECD:	Organisation for Economic Cooperation and Development
OR:	Observed Ratios
PA:	4-Pyridyl Aldehyde
PAH:	Polycyclic Aromatic Hydrocarbons
PAN:	Peroxyacetyl Nitrate
PBM:	Peribacteroid membrane
PBS:	Peribacteroid space
PCR:	Polymerase Chain Reaction
PMC:	Pollen Mother Cell
POM:	Polycyclic Organic Matter
QTL:	Quantitative Trait Loci
RAD:	Radioecology Programme (under NKS)
RADPATH:	Biochemical Pathways of Artificial Radionuclides (under CEC)
RAPD:	Random Amplified Polymorphic DNA Technique
REE:	Rare earth element
RESSAC:	The Consequence of a Major Nuclear Accident on the Surrounding Environment: Soil and Surface Rehabilitation (under CEC.
RFLP:	Restriction Fragment Length Polymorphism
RIMI:	Risø Integrated Environmental Project
RVAU:	The Royal Veterinary and Agricultural University
SCOPE:	Scientific Committee on Problems of the Environment (under International Council of Scientific Unions: (ICSU)
SINE:	Short Interspersed Repetitive Element
SJVF:	The Veterinary and Agricultural Research Council
SMP:	Strategisk Miljøforskningsprogram
TOR:	Tropospheric Ozone Research (under EUROTRAC)
TRACT:	Transport of Pollutants over Complex Terrain
UKAEA:	United Kingdom Atomic Energy Authorities
VAM:	Vesicular-Arbuscular Mycorrhiza
VAMP:	Validation of Model Project (under IAEA)
WOCE:	World Ocean Circulation Experiment
YAC:	Yeast Artificial Chromosome