Draft DG Sanco Document: Addressing the new challenges for risk assessment: – An industry perspective

Peter Campbell[1]

[1]Syngenta, Jealotts Hill research Centre, Bracknell, UK RG42 6EY

E-mail contact: peter.campbell@syngenta.com

1. Introduction

The following abstract summarises an individual perspective on the ecological risk assessment section of this draft DG Sanco Document "Addressing the new challenges for risk assessment", from Dr Peter Campbell who has been working as an ecotoxicologist within the Crop Protection industry for the last 15 years.

2. Discussion

This is a comprehensive overview of ecological risk assessment approaches and the high level goals of this paper such as reduced animal testing and development and promotion of high level ecological risk assessment expertise, should be commended. In addition, the following key recommendations are also well founded eg:-

1) The potential use and benefits of adopting higher-tier ecological risk assessment tools such as field or mesocosm studies and the more recent ecological modelling tools, which are essential within current precautionary pesticide regulatory framework.

2) The list of requirements for new innovative approaches, that should be met before such approaches can be used in regulatory ecological risk assessments.

3) The need for further research on how to deal with multiple stressors (ie chemical mixtures and/or environmental factors). However, what also needs to be considered is how such research findings can then be used within the context of a chemical registration framework, which can only be implemented at an individual chemical product level.

One of more holistic challenges for this documents and its audience, is the breadth of remit. Currently it encompasses both prospective and retrospective regulation as well as different types of potential chemicals contaminants (eg pesticides and general chemicals), where the issues and the available data sets are going to be very different. In addition, some of the statements within this document are not always relevant to some industry sectors eg "large number of untested or inadequately tested chemicals", does not apply to Pesticides sector. Therefore, one way to improve this document would be to consider restructuring the content to provide clear and specific advice to distinctly different situations eg Ecological Risk Assessment for Chemical Registration and Site Specific Ecological Risk Assessment, being 2 potential examples.

One of the ecological risk assessment challenges for the pesticide industry is extrapolation of risk assessment conclusions eg between species, between different environmental conditions and even between geographical regions. One of the tools recommended within this document to help with this issue is modelling (particularly more recent ecological and spatial modelling approaches), and indeed this recommendation should also be commended. In contrast, however, another recommendation for greater uncertainty analysis and application of appropriate uncertainty factors is not so helpful. For example, the use of the EFSA Bird & Mammal Guidance Document, which adopted such an uncertainty analysis approach, still results in low toxicity herbicides failing the risk assessment. In addition, the current practise of application of safety factors to the results of Mesocosm/pond studies now means there is no regulatory benefit to carrying out such studies. Consequently, we now miss a real opportunity to test, observe and understand potential in-direct and community level aquatic effects, both of which are highlighted within this document, as an area where more information is required. The key issue here is, there will always be uncertainty within ecological risk assessment and therefore can we really reduce the need for expert judgement, as recommended, without the continued application of large precautionary safety factors.

3. Conclusion

This is a very comprehensive review of ecological risk assessment, which makes some useful suggestions and recommendations to improve ecological risk assessment across the board and highlights key research needs. In particular, it usefully develops and promotes learning's and practises across different chemical sectors and from across different risk assessment purposes. From a pesticide industry perspective, which is relatively data rich and already utilises many of the recommended sophisticated risk assessment approaches, the focus on uncertainty analysis without any obvious direct benefits in terms of application of significantly reduced uncertainty factors is still a challenge. In addition, the drive to reduce the need for expert judgement in such a complex risk assessment arena is difficult to realise.

Effects of chronic radiation exposure on plant populations

Stanislav Geras'kin[1], Tatiana Evseeva[2], Alla Oudalova[1]

[1] Russian Institute of Agricultural Radiology and Agroecology, Obninsk, Russia
[2] Institute of Biology, Syktyvkar, Russia
E-mail contact: stgeraskin@gmail.com

1. Introduction

To understand effects of real-world contaminant exposure properly we must pay attention to what is actually going on in the field. However, for many wildlife groups and endpoints, there are no, or very few, studies that link accumulation, chronic exposure and biological effects in natural settings. To fill these gaps, the results of long-term field observations in the 30-km Chernobyl NPP zone, in the vicinity of the radioactive wastes storage facility (Leningrad Region), at radium production industry storage cell territory (the Komi Republic), in the Bryansk Region affected by the Chernobyl accident, and in the Semipalatinsk Test Site, Kazakhstan that have been carried out on different species of wild and agricultural plants are discussed here.

2. Materials and methods

In 2005-2007 seeds of crested hairgrass (*Koeleria gracilis* Pers.) were collected from four locations of the Semipalatinsk Test Site (Kazakhstan). Radiation background at the sites and activity concentrations of the most dose-forming radionuclides in the soil samples were measured. Absorbed doses to crested hairgrass were calculated. Squashed slides for cytogenetic analysis were prepared of coleoptiles (2-5 mm of length) of germinated seeds. In every slide, all ana-telophase cells (4800 – 11900 ana-telophases in 30-90 slides) were scored to calculate frequency of aberrant cells. Detailed description of methods used is given in [1].

To study biological effects in chronically exposed Scots pine (*Pinus sylvestris* L.) populations, six test sites were chosen in the Bryansk Region of Russia radioactively contaminated as a result of the Chernobyl accident. Pine cones were collected in autumns of 2003-2009. Activity concentrations of radionuclides in soil samples and cones were measured, and doses to the pine trees' generative organs were estimated. Aberrant cells were scored in root meristem of germinated seeds in ana-telophases of the first mitoses. The method of isozymic analysis of megagametophytes was used for an estimation of genetic variability in Scots pine populations. Five enzymatic loci (GDH, LAP, MDH, DIA, and 6-PGD) were studied in endosperms of the seeds collected in 2005. Detailed description of materials and methods used can be found in [2, 3].

3. Results and discussion

The STS is a unique place to study effects of chronic low dose rate exposure on non-human species over several generations. The wide range of plots different in levels and spectrum of radioactive contamination, an availability of plots with dominating contribution of particular types of radiation (α-, β-, and γ-radiations) to dose absorbed by plants and animals as well as specific climatic conditions provide a unique opportunity for studying long-term biological effects in chronically exposed ecosystems against the background of extreme environmental conditions. A study of crested hairgrass populations, a typical wild cereal for Kazakh steppe, showed that the frequency of cytogenetic alterations in coleoptiles of germinated seeds increases proportionally to the dose absorbed by plants [1]. Severe alterations of single and double bridges as well as laggard chromosomes contribute mainly to the observed cytogenetic effect. The agreement between findings from three years of study (2005-2007), different in weather conditions, suggests the leading role of radioactive contamination in an occurrence of cytogenetic effects.

The Chernobyl accident caused dramatic and long-term increases in ambient radiation doses to many forest environments. Sites still exist in the Bryansk Region of Russia, 25 years after the Chernobyl accident, where radioactive contamination significantly exceeds background. In the study reported herein, cytogenetic effects in Scots pine populations growing in the Bryansk Region have been investigated for 7 years [3]. There were no significant differences in frequencies of cytogenetic abnormalities, observed in the same study site from year to year. Thus, the effects observed can be regarded as quite robust and replicable over time. Aberrant cell frequency in root meristem of germinated seeds collected from these populations significantly exceeds the reference level and shows correlation with the dose absorbed. Combined with data from other our studies [1, 4-6], these findings indicate that an increased level of cytogenetic alterations is a typical phenomenon for plant populations growing in areas with relatively low levels of pollution.

An increase in mutation rate can affect the population genetic structure by producing new alleles or genotypes, and thereby has ecologically relevant effect. Alterations in the genetic make-up of populations are of primary concern because somatic changes, even if they lead to a loss of some individuals, will not be critical in populations with a large reproductive surplus. To analyze whether an exposure to radionuclides causes changes in population genetic structure, we evaluated frequencies of three different types of mutations (null allele, duplication and changes in electrophoretic mobility) of enzymatic loci in endosperm and embryos of pine trees from the studied populations. It is found that chronic radiation exposure results in the significant increase of total occurrence of enzymatic loci mutations [2]. In particular, frequencies of mutations for loss of enzymatic activity increase with the dose absorbed by generative organs of pine trees. Phenotypic variability in the exposed pine tree populations significantly exceeds the reference level and increases with dose absorbed by generative organs of pine trees. Moreover, the observed heterozygosity in pine tree populations at the radioactively contaminated sites is essentially higher then the expected one and increases with dose absorbed by generative organs of pine trees. Therefore, a high level of mutation occurrence is intrinsic for descendants of pine trees in the investigated populations, and genetic diversity in the populations is essentially conditioned by radiation exposure.

In the first year after the Chernobyl accident a significant decrease in reproductive ability of pines (reduction of seed mass and their number per cone, as well as increase in portion of abortive seeds) was observed at doses over 1 Gy. Eleven years after the accident this tendency still persisted. In 1997, the portion of abortive seeds from pine populations that had received doses of 10-20 Gy in 1986 significantly exceeded the correspondent reference level. The effect of radioactive contamination on reproductive ability of pine trees was also observed at the South Urals radioactive trail. Chronic exposure of pine trees at dose rates of 4.2-6.3 µGy/h resulted in a significant decline of seed mass, as well as an increase in the fraction of abortive seeds. At a lower dose rate of 0.8 µGy/h, the enhancement in percent of abortive seeds was not observed. In 2000-2001, decrease in pollen viability as well as increase the number of anomalous pollen grains in Scots pine populations from Bryansk region at dose rates of 1.8-5.4 µGy/h have been detected. In contrast to the results mentioned above, we failed to find any clear linkage between reproductive ability and doses absorbed by generative organs of pine trees [3]. So, the high mutation rate found in our study had no effect on the reproductive ability of the exposed populations.

An appearance of some standing factors (either of natural origin or man-made) in the plants' environment may activate genetic mechanisms, changing a population resistance to a particular stress. The response of a population exposed to low dose rate irradiation depends on both the type of organism and the biophysical properties of radiation. Contrary to the increased radioresistance of seeds from plant populations inhabiting contaminated territories described in [5], no significant difference in resistance to subsequent γ-ray exposure between seeds collected from the reference and exposed Scots pine populations was found in study [3]. Similarly, the seeds from the crested hairgrass populations that have been experiencing radiation exposure for more than a half century and are bearing the elevated levels of cytogenetic abnormalities do not show any reliable increase in resistance to the additional acute γ-ray exposure [1].

4. Conclusions

The effects of chronic exposure on living organisms and populations remain poorly explored, and represent a much needed field of research. Much more is to be elucidated in our understanding before we will be able to give an objective and comprehensive assessment of the biological consequences of chronic, low-level radiation exposures to natural plant and animal populations.

5. References

[1] Geras'kin SA, Oudalova AA, Dikarev VG et al. 2012. Effects of chronic exposure in populations of *Koeleria gracilis* Pers. from the Semipalatinsk nuclear test site, Kazakhstan. J Env Radioactivity 104:55-63.
[2] Geras'kin SA, Vanina JC, Dikarev VG et al. 2010. Genetic variability in Scots pine populations of the Bryansk Region radioactively contaminated in the Chernobyl accident. Biophysics 55:324-331.
[3] Geras'kin SA, Oudalova AA, Dikareva NS et al. 2011. Effects of radioactive contamination on Scots pines in the remote period after the Chernobyl accident. Ecotoxicology 20:1195-1208.
[4] Geras'kin SA, Dikarev VG, Zyablitskaya YeYa et al. 2003. Genetic consequences of radioactive contamination by the Chernobyl fallout to agricultural crops. J Env Radioactivity 66:155-169.
[5] Geras'kin SA, Kim JK, Oudalova AA et al. 2005. Bio-monitoring the genotoxicity of populations of Scots pine in the vicinity of a radioactive storage facility. Mutat Research 583:55-66.
[6] Evseeva TI, Majstrenko TA, Geras'kin SA et al. 2009. Estimation of ionizing radiation impact on natural *Vicia cracca* populations inhabiting areas contaminated with uranium mill tailings and radium production wastes. Sci Total Env 407: 5335-5343.

Designing an ecosystem approach for ecological risk assessment of radiation: a path forward for radioecology

F. Bréchignac[1], C. Bradshaw[2], S. Carroll[3], S. Fuma[4], L. Hakanson[5], A. Jaworska[6], L. Kapustka[7], I. Kawaguchi[4], L. Monte[8], D. Oughton[9], T. Sazykina[10], P. Strand[6]

[1]Institute for Radioprotection and Nuclear Safety, France
[2]Department of Systems Ecology, Stockholm University, Sweden
[3]Swedish Radiation Safety Authority, Sweden
[4]National Institute of Radiological Sciences, Japan
[5]Uppsala University, Sweden
[6]Norwegian Radiation Protection Authority, Norway
[7]SLR Consulting, Canada
[8]ENEA, Roma, Italy
[9]Norwegian University of Life Sciences, Norway
[10]Typhoon, Russia
E-mail contact: francois.brechignac@irsn.fr

1. Introduction

The protection of the environment from ionizing radiation has evolved as a major issue in the recent years to form a major topic for radioecologists who are now engaged in developing systems for the protection of the environment from radiation hazards. Ongoing developments are aimed at designing conceptual approaches and methods for Ecological Risk Assessment (ERA) of radiation delivered by radionuclides in the environment meant to aid decision making, especially with respect to situations of existing or future possible environmental contamination.

2. Problem formulation

Although an ideal ERA should focus on the structure and functions of communities and ecosystems, most current methods, due to ecosystem complexity and limited information, typically focus on radio-toxicity at smaller scales of biological organization such as physiological mechanisms of toxicity and the responses to toxicant exposure of discrete endpoints in individual organisms of a single species [1, 2]. Expanding from mammalian radiotoxicity studies, which have been used to support radiation protection of the human species, and from classical ecotoxicological studies of animal and plant test species, they are all based on several types of “reference organisms” designed to make use of dose-response relationships at the level of individual organisms. Whilst there is clear interest to exploit classical toxicological data, it is to be stressed that this approach to ERA employs a linear, reductionist and determinist paradigm that considers risks to each species to be independent of one another and determined by causal relationships inferred between input and output, such as dose and response [3, 4].

3. Discussion

One major shortcoming of such methods is therefore that they ignore interactions between species that are critical to the functioning of communities and ecosystems. As a result, they hardly can meet the general environmental protection objectives that have been set, in the vast majority of situations, at the population, communities and ecosystem levels. Indeed, the human radiological protection target is set at the individual organism level whereas it is most generally set at higher levels of biological organization for environment protection.

It is primarily for this reason that several areas of environmental management have already been engaged in working out “ecosystems approaches” of risk assessment [5] , like in halieutics for the protection of fish stocks in the oceans [6], or for the sustainable maintenance of biodiversity [7]. Indeed, perturbations induced by stressors within ecosystems cannot be fully grasped from an exclusive toxicological understanding of the stressor’s interaction with individual organisms. Such effects only act as triggers of perturbation which next propagate within ecosystems. Ecosystems are dominated by complex inter-population relationships featuring non-linear responses which can radically differ from the initial response observed within individual organisms. Inter-population relationships, such as predator-prey interactions, are also capable of mediating indirect effects by means of which the population actually exposed to the stressor may not be the most affected. This is particularly relevant when considering the long-term ecological effect of chronic exposure to toxicants, like radiation, where ecological damage may not be due to the direct radio-toxicological stress of radiation per se (upon individual organisms), but rather to the cascading effects among interacting populations within ecosystems as a result of differences in sensitivity to radiation.

There is clear justification therefore to address the rationale for developing a new wider approach for protection from radiation that is based on the ecosystem whilst building upon the current methods [8]. This strategy intends to focus on the inherent properties of ecological systems, particularly the dynamic interactions among system components that influence resistance to stressors, resilience of components to stressor effects, and delayed effects including translation of effects up or down trophic levels. The current focus on reference organisms of animals and plants, analogous to the reference man, has resulted in gathering data for a relatively narrow group of species. Moreover, these data are limited to organism-level endpoints, which cannot be translated effectively to systems-level interactions as shown in a number of multi-species experiments and in the field. Further, the list of species for which organism level data are available is too restrictive to allow for meaningful efforts to model dynamic interactions that are characteristic of ecological systems.

4. Research priorities and recommendations

4.1. Priorities for R&D

Moving towards an "ecosystem approach" for Ecological Risk Assessment of radiation requires to work along three main research priorities:

- Areas of emphasis for the systems-level research include detailing interactive responses to radiation exposure, propagation of effects, delayed effects, and resistance/resilience of ecosystems. Each of these could be designed to examine effects at a) population-, guild-, or community-levels, or b) systems functions such as primary productivity, decomposition, energy transfer, or nutrient flow.
- Additional research at the organism-level should be expanded to include representatives of trophic groups not currently included or understudied (e.g., decomposers). There should also be efforts to expand representation of taxa from multiple geographic regions to supplement the current dominance of data from northern temperate systems. Topical research that would be useful would be to develop better understanding of radiation effects that result in adaptation, acclimation, hormesis, and epigenetic effects.
- Field studies are needed to calibrate laboratory studies from both the systems- and organism-levels. In addition to the opportunities at Chernobyl and Fukushima (decidedly different in terms of ecological systems), studies should be undertaken in radionuclide mining areas. In each of these potential study areas, the investigative designs should be based on gradient analyses approaches and not some attempt to compare to "reference sites."

4.2. Recommendations for radiation protection

Recognizing that the ecosystem concept has been adopted in an increasing number of other situations, it is appropriate for radiation protection to move in this direction in order to improve the relevance of information coming to decision-makers. To that end, the following points should be considered:

- Promote the dialogue between environmental assessors and environmental managers (facilities operators, contaminated site managers, and other regulators) to increase the chances of improving the value of information flow (two-way dialogue).
- More integrated and functional endpoints to expand beyond the organism-level. This could also include consideration of additional indices that embed the existing and new endpoints (decomposition, primary productivity, etc.).
- Reference organism approach – improve to incorporate ecological functionalities, other ecological criteria, and reference species versus reference organisms, all aimed to facilitate an ecosystem approach. Better consideration of taxonomy, insects, bacteria, fungi to cover ecological functionality and to make it more accessible to people within different geographical areas, biomes.

5. Conclusions

Radioecologists should be engaged in efforts to promote consistency across the broad spectrum of ecological research and environmental management so that information can be leveraged from multiple efforts outside of the radioecology fields. In particular there should be efforts to coordinate work from chemical and various other stressors, as well as with theoretical ecologists involved in landscape ecology and systems modelling. The above-mentioned conclusions and recommendations arise from the output work of a Task Group under the International Union of Radioecology to be published in its reports series [8].

6. References

[1] Barnthouse LW 1993. Population-level effects. In: Ecological risk assessement. G.W. Suter, ed., CRC Press, Lewis Publishers, Boca Raton, London, New York, Washington DC, pp. 247-274.

[2] Suter GW, Bartell S 1993. Ecosystem-level effects. *In*: Ecological risk assessment, G.W. Suter, ed., CRC Press, Lewis Publishers, Boca Raton, London, New York, Washington DC, pp. 275-308.

[3] Bréchignac F 2003. Protection of the environment: how to position radioprotection in an ecological risk assessment perspective. Science of the Total Environment, 307: 35-54.

[4] Bréchignac F, Doi M 2009. Challenging the current strategy of radiological protection of the environment: arguments for an ecosystem approach. *Journal of Environmental Radioactivity* 100: 1125-1134.

[5] Apitz SE, Elliott M, Fountain M, Galloway TS 2006. European environmental management: Moving to an ecosystem approach. *Integrated Environmental Assessment and Management* 2(1):80-85.

[6] FAO (Food and Agriculture orgnization) 2005. Fisheries management. 2. The ecosystem approach to fisheries. FAO Fisheries Technical Guidelines n° 4, Suppl. 2.

[7] CBD (Copnvention on Biological Diversity) 2000. (http://www.cbd.int/decision/cop/?id=7148) COP Decision V/6, Annex B: Ecosystem approach.

[8] IUR (International Union of Radioecology) 2012. Towards an ecosystem approach for environmental protection with emphasis on radiological hazards. IUR Report n° 7, in press

Acknowledgement - The authors wish to thank the following scientists for their critical comments about the IUR draft report now in press that is reported on in this communication: D. Copplestone, C.M. Larsson and J. Pentreath (ICRP Committee 5), U. Kautsky (SKB, Sweden), S. Cormier (US EPA) and G. Smith (U.K.).

Why complexity matters: using ALMaSS for risk assessment of wildlife

Chris J. Topping

Department of Bioscience, Aarhus Universit, Grenåvej 14, DK-8410, Rønde, Denmark
E-mail contact: cjt@dmu.dk

1. Introduction

The primary focus of environmental risk assessment (ERA) for non-target organisms is on the direct effect of the toxicant or other stressor. However, it is clear that what are sometimes termed large-scale sources of variation are very important in determining the impacts [1]. However, these describe only part of the variation that occurs in the real world. Landscapes vary in structure, meaning that the field size and proximity to primary habitats will vary. There is climatic variation driving changes in phenology and behaviour, and there will be management changes in the proportion of crops grown; but also changes in how they are cultivated. All of these factors can affect the risk assessment. There is also another, difficult to observe, property of real systems, and that is the spatio-temporal dynamics associated with populations, climate, management, and ecology and behaviour, and the potential for feedback loops. These interactions can exacerbate impacts either via local feedback mechanisms e.g. multiple stressors, or by virtue of the spatial population dynamics. This uncertainty is normally considered as stochasticity and the factors are often incorporated into a single general term, utilizing a safety factor to account for uncertainty. This, however, robs us of both understanding and predictive power, since probability distribution can only be based on statistical expectations of past events, which do not necessarily account for interactions in the future. An alternative approach capable of dealing with these system properties is agent-based modelling (ABM).

2. ABMs and ALMaSS

Modern simulation models are capable of integrating many drivers and actors in space and time. Systems such as the ALMaSS system [2] of ABMs are capable of representing detailed farming operations on a large scale and integrating these with realistic models of animal populations, expressing each animal as an individual agent. These have a number of advantages over more general approaches. The first is that they are capable of representing complex dynamics in time and space. For example crops are sown, grow, harvested, and usually rotated whilst uncropped areas may develop natural seasonal vegetation dynamics and may also be subject to management (e.g. amenity grass). These changes are continual and often rapid and widespread, e.g. soil cultivation. The effect of these dynamics is therefore likely to be large, not taken into account in current risk assessment approaches, but can be incorporated in ALMaSS. Another key model attribute is the ability of agents to respond to the information they gather from their local environment. This automatically integrates many of the dynamics difficult to capture in traditional models, e.g. source-sink dynamics are emergent properties and do not need to be imposed.

ALMaSS models have been used for regulatory pesticide ERAs and in a number of research contexts e.g.: The spatial dynamics associated with risk assessment for a beetle species demonstrated that application of insecticide could have impacts outside of the treated area due to depletion effects caused by creation of source/sink dynamics [3]; Indirect effects can also be incorporated, as can interactions between management and animal behaviour [4]; Highly complex situations can also be managed, including issues such as chronic effects and genetic transmission of these [5]. An evaluation of the impact of an endocrine disruptor on field voles indicated that the major factors driving the population level impact were ecological not toxicological, i.e. they were related to interactions with the large-scale sources of variation [5].

3. Discussion

Simulation results of pesticide ERA in ALMaSS are unambiguous; the environmental context is often critical to the result. Thus simple approaches such as toxicity:exposure ratios or hazard quotients do not adequately capture system responses. In fact, considering ERAs as population-level exercises in realistic environments calls into question many of the precepts of regulatory risk assessment. Crucially, there is little ecology in an ERA, but examples such as the vole/endocrine disrupter study [5] demonstrate how important it is to consider context. Similarly, the fact that multiple stressors need to be taken into account, not just because of additive or synergistic effects but because the ecology of the system changes with more and varied stressors [6].

The need to detailed system descriptions are further supported by wider studies such as an evaluation of the importance of organic farming for six agricultural species [7], which demonstrated that insufficient precision in question definitions leads to imprecise and even misleading responses. In this case this was due to organic farming being comprised of many different farm operations, used in different contexts, and with animal responses to them being specific and also often context dependent. The relevance to pesticide testing should be clear.

The example of the organic farming modelling in ALMaSS and similar exercises such as evaluating the impacts of increasing the cropped area of energy maize in Germany [8] demonstrate the capability of handling complex questions and detailed environments *in silico*. There is a great deal to be gained from this approach, but it comes at a cost. This cost can be both in data requirements and developmental time.

Data requirements come from the need to accurately model animal behaviour and ecology, and from the need to have a detailed and realistic environmental representation e.g. topography, detailed farming practice, weather. However, much of this can be made readily available from local and national mapping schemes and the EU requirements for farmer record keeping. Hence, for much of the EU there is little to hinder environmental simulation.

Developmental time is another issue. ALMaSS itself has been under development for almost 15 years and individual models require substantial testing before they are deemed fit for use (e.g. [9]). The sheer volume of work required to develop a system such as this means that only large organisations with long-term funding could realistically undertake this. However, ALMaSS is now an open-source science project (see http://ccpforge.cse.rl.ac.uk/gf/project/almass/) and thus can be built on and developed if starting from scratch is not an option.

A potential ERA modelling strategy for the future might entail evaluating the impact of pesticides (new and old) against the background of current landscapes and agricultural practices. In this way the aim would be to improve, or at least not worsen the situation by the addition of new pesticides to the market. This requires a radical rethink of the way pesticide regulation is carried out and cannot be achieved overnight; but the option of making ecologically realistic assessments is there, and if we really want to be sure we are not harming our environment then we should take up this challenge.

4. References

[1] Perry, J.N., et al., 2010. *A mathematical model of exposure of nontarget Lepidoptera to Bt-maize pollen expressing Cry1Ab within Europe.* P Roy Soc Lond B Bio, **277**(1686): p. 1417-1425.
[2] Topping, C.J., et al.,2003 *ALMaSS, an agent-based model for animals in temperate European landscapes.* Ecol.l Model **167**(1-2): p. 65-82.
[3] Topping, C.J. and M. Lagisz, 2012. *Spatial Dynamic Factors Affecting Population-Level Risk Assessment for a Terrestrial Arthropod: An Agent-Based Modeling Approach.* Hum Ecol Risk Assess **18**(1): p. 168-180.
[4] Topping, C.J. and P. Odderskaer, 2004. *Modeling the influence of temporal and spatial factors on the assessment of impacts of pesticides on skylarks.* Env. Tox. Chem., **23**(2): p. 509-520.
[5] Dalkvist, T., C.J. Topping, and V.E. Forbes, 2009. *Population-level impacts of pesticide-induced chronic effects on individuals depend more on ecology than toxicology.* Ecotox. Env.l Safety, **72**(6): p. 1663-1672.
[6] Topping, C.J., et al., 2009. *The potential for the use of agent-based models in ecotoxicology*, in *Ecotox.Model.*, J. Devillers, Editor, Springer. p. 205-235.
[7] Topping, C.J., 2011. *Evaluation of wildlife management through organic farming.* Ecol Eng. **37**(12): p. 2009-2017.
[8] Gevers, J., et al., 2011. *Biodiversity and the mitigation of climate change through bioenergy: impacts of increased maize cultivation on farmland wildlife.* Global Change Biology Bioenergy **3**(6): p. 472-482.
[9] Topping, C.J., T.T. Hoye, and C.R. Olesen, *Opening the black box-Development, testing and documentation of a mechanistically rich agent-based model.* Ecol Model, 2010. **221**(2): p. 245-255.

Environmental sensitivity as a tool for the risk assessment of the use of nuclear energy

Carini F.[1], Barabash S.[2], Berkovskyy V.[3], Brittain J.E.[4], Chouhan S.[5], Eleftheriou G.[6], Iosjpe M.[7], Monte L.[8], Psaltaki M.[6], Shen J.[2], Tracy B. [9], Tschiersch J.[10] and Turcanu C.[11]

[1] Università Cattolica del Sacro Cuore, Via Emilia Parmense, 84, Piacenza, Italy
[2] EcoMetrix Incorporated, 6800 Campobello Road, Mississauga, ON L5N 2L8, Canada
[3] Division of Radiation, Transport & Waste Safety, IAEA, PO Box 100, 1400 Vienna, Austria
[4] Natural History Museum, University of Oslo, Norway
[5] Environmental Technologies Branch, AECL, Chalk River, ON, Canada
[6] National Technical University of Athens, 15780 Zografou, Greece
[7] Norwegian Radiation Protection Authority, Grini næringspark 13, Østerås, Norway
[8] ENEA, Via P. Anguillarese, 301, 00100 Roma
[9] Retired, formerly with Radiation Protection Bureau, Health Canada, Ottawa, Canada
[10] Helmholtz Zentrum München, Institute of Radiation Protection, 85764 Neuherberg, Germany
[11] Belgian Nuclear Research Centre, SCK•CEN Boeretang 200, B-2400 Mol, Belgium
E-mail contact: franca.carini@unicatt.it

1. Introduction

Approaches to the management of risk in radioecology have to take into account geographic, climatic, living and dietary habit differences, as well as ecosystem differences. The understanding of the factors of sensitivity of different environments, populations or geographic areas is important for scientists and policy makers to set priorities for the allocation of limited resources in the preparedness phase and also to improve emergency and post-emergency management. In particular, the identification of vulnerable environments will be valuable in planning the locations of new nuclear facilities.

A Task Group on Radioecological sensitivity was organized by the International Union of Radioecology (IUR) in 2007 on the basis of studies of the Radioecological Sensitivity Forum, 1998-2001. The objective was to discuss a standardization to represent the radiological state of the environment following accidental pollution and to develop a scale of radioecological sensitivity for use in emergency planning and preparedness. The work of the Group continued under the International Atomic Energy Agency (IAEA) EMRAS II Programme (Environmental Modelling for Radiation Safety), from 2009 to 2011, as Working Group 8 on Environmental Sensitivity. Three main categories of factors are of paramount importance in the decision process for the management of radiological emergencies: environmental, economic and social factors. The WG8 focused its studies on sensitive non-urban environments. The aim was to investigate which environments, which components of each environment, and which seasons of the year would be most sensitive to a major release of radionuclides. The concept of environmental sensitivity has been assumed by the participants to the WG8 as a -relationship among three elements: a set of effects or consequences A, an independent set of conditions B and a set of given stresses C. The exercises in this work were performed by accounting for such a definition of sensitivity. The overall aim was to aid the planning and response in case of emergency, as well as the long-term countermeasures following a nuclear accident.

2. Materials and methods

The models used in the exercises were CHERPAC [1], ECOSYS [2, 3], FDMT-RODOS [4], the Health Canada model, IMPACT [5], MOIRA-PLUS [6], NRPA box model [7] and NTUA 3D model [8]. The following environments located in different geographic areas were chosen for the analysis of sensitivity: temperate agricultural and alpine (Europe and Canada), coastal marine (Nordic seas, North-East Aegean Sea, Thermaikos Gulf Mediterranean Sea), temperate forest (Northern Saskatchewan and Ontario), freshwater aquatic (Norway, Italy, Northern Saskatchewan and Ontario) and Arctic (Northern Canada). Each environment was allocated the same initial single deposition (1000 Bq/m^2) of two long-lived radionuclides - ^{137}Cs and ^{90}Sr - and one short-lived radionuclide - ^{131}I. Deposition was considered under four different seasonal conditions, corresponding roughly to winter, spring, summer, and autumn. The end points of the exercise were the radionuclide concentrations in environmental media and food chain products leading to humans as well as the radiation doses to an adult, a 10-year old child and a one-year old infant who receive most or all of their food intake from the respective environments, during first, second, and 10th year following the deposition. Sensitivity analysis was then performed to ascertain which components of each environment are most responsible for ecosystem response and can thus lead to higher doses.

3. Results and discussion

Critical sensitivity factors were identified, regarded as responsible for the major radionuclide impacts on each environment, such as: high aggregated radionuclide transfer rates to mushrooms and berries and to game, high transfer factor of radiocaesium from contaminated feedstuff to lamb, long residence times, presence of permafrost, high mass interception factors and long biological half-times in lichens. For lakes and coastal environments: water depth, mean water retention times and sedimentation rates, ionic concentrations and ecological factors that influence the transfer of radionuclide to biota.

Sensitivity is time dependent. Calculations comparing the sensitivity factors of an alpine (Øvre Heimdalsvatn, Norway) and a lowland lake (Bracciano, central Italy) performed by MOIRA-PLUS show that the alpine lake is more sensitive to ^{137}Cs deposition than the lowland one, because of the low biomass, the low ionic concentrations and high runoff over frozen ground in spring. However, the persistent levels of contamination in the lowland lake give rise to continued high activity concentrations in biota, indicating its higher vulnerability in the long term.

Climate is another important factor of sensitivity. In northern food chains there is no milk production and little cultivation of leafy vegetables, that would lead to significant uptake of ^{131}I for children in temperate zones. Again, the impact of ^{90}Sr is expected to be less than that of ^{137}Cs, which is bio-accumulated in Arctic food chains.

There are also interactions between environmental sensitivity factors. For instance, the models used for the agricultural scenarios (CHERPAC and FDMT-RODOS) show the effect on the activity in animal products (e.g. milk) of the pasture activity decreases because of weathering; the activity in such food products increases again when feed is switched to harvested grass.

The doses to individuals and populations are not only associated with the environmental conditions but depend also on factors of social and economic nature such as the population living habits, food consumption preferences, and agricultural practices. A first comparison between model predictions shows that agricultural scenarios produces the largest doses for ^{137}Cs in most cases. However, ^{137}Cs in the Arctic environment can also produce significantly elevated doses. The doses in marine environments tend to be much lower, even in shallow coastal areas.

Results of calculations show that doses for the first year dominate the doses of the second and tenth year after deposition. Doses also depend on groups of age. For example in the agricultural environment CHERPAC predicts that the ingestion dose for ^{137}Cs is higher for adults than other age groups, while for ^{90}Sr and ^{131}I, the ingestion dose is highest for infants. The same applies to FDMT-RODOS, which predicts doses from ^{90}Sr higher for infants, as compared to adults or to 10 years old children.

4. Conclusions

The assessment of the radiological state of the environment following pollution depends on those pathways of highest environmental sensitivity, related to climatic area, as well as anthropic management, social and economic factors. The present study has confirmed the importance of the agricultural environment for the assessment of doses from an accidental release of long-lived radionuclide ^{137}Cs. A scale of radioecological sensitivity is under discussion for the further activities of the group.

5. References

[1] Chouhan S.L., N.W. Scheier, and S-R. Peterson. 2011. CHERPAC, an environmental transport code, and its predictions of environmental sensitivities in agricultural and forest ecosystems. Radioprotection, vol. 46, n◦ 6 (2011) S515–S520, EDP Sciences, DOI: 10.1051 / radiopro / 20116784s.

[2] Müller H, Pröhl G 1993. ECOSYS-87: A Dynamic Model for Assessing Radiological Consequences of Nuclear Accidents. Health Physics 64(3), 232-252.

[3] Sdouz G, Pachole M. 2006. Food chain data customization for decision support systems in Austria. Second European IRPA Congress on Radiation Protection, 15-19 May 2006, Paris, France, TA8, 1-4.

[4] Müller H., Gering F. and Pröhl G. (2006). Model Description of the Terrestrial Food Chain and Dose Module FDMT in RODOS PV6.0. RODOS(RA3)-TN(03)06. http://www.rodos.fzk.de/Documents/Public/HandbookV6/Volume3/FDM_Terra.pdf

[5] Jige Shen., Don Hart. (2009). IMPACT (Integrated Model for the Probabilitic Assessment of Contaminant Transport) User's Manual version 5.4.0.

[6] Monte, L., Brittain, J.E., Gallego, E., Håkanson, L., Hofman, D., Jiménez, A., 2009. MOIRA-PLUS: A decision support system for the management of complex fresh water ecosystems contaminated by radionuclides and heavy metals. Computers and Geosciences 35, 880-896.

[7] Iosjpe M., Brown J. & Strand P., 2002. Modified approach for box modelling of radiological consequences from releases into marine environment, Journal of Environmental Radioactivity, 60, 91-103

[8] Psaltaki M., Florou H., Trabidou G.,and Markatos N.C. « Modelling and assessment of pollutant impact on marine environments», 2nd WSEAS International Conference on Computer Engineering and Applications (CEA '10) Harvard University, Cambridge USA, (p.p.176-180, January 27-29, 2010).

The effect of radionuclide contamination of the Yenisei River on cytogenetic characteristics of aquatic plants

Alexander Bolsunovsky[1], Elena Muratova[2]

[1]Institute of Biophysics Siberian Branch, Academy of Sciences, Akademgorodok, Krasnoyarsk, 660036 Russia
[2]Institute of Forest Siberian Branch, Academy of Sciences, Akademgorodok, Krasnoyarsk, 660036 Russia

E-mail contact: radecol@ibp.ru

1. Introduction

The Yenisei River, one of the world's largest rivers, is contaminated with artificial radionuclides released by one of the Russian facilities producing weapons-grade plutonium (the Mining-and-Chemical Combine), which has been in operation for many years [1-5]. In addition to artificial radionuclides, effluents released by the Mining-and-Chemical Combine (MCC) were found to contain uranium isotopes. Another source of uranium for the ecosystem of the Yenisei River basin is the uranium enrichment facility situated at the Kan River – a Yenisei tributary. Thus, the Yenisei River continuously receives a wide range of radionuclides, both long-lived and short-lived, and, hence, the aquatic ecosystem of the Yenisei River is a unique environment that can be used to study the migration mechanisms of various radionuclides and their effects on living organisms. Aquatic plants are an important component of water ecosystems, which, owing to their ability to accumulate high levels of radionuclides, can be used in biomonitoring and bioremediation.

In previous studies [2-4], calculations were done to estimate internal dose rates to aquatic organisms of the Yenisei River near the MCC, taking into account high concentration factors of artificial radionuclides for aquatic organisms. The water moss (*Fontinalis antipyretica)* exhibited the highest capacity to accumulate artificial radionuclides; hence, it accumulated the largest artificial exposure dose among the study aquatic organisms [2]. However, all the samples of aquatic organisms examined previously were collected near the MCC discharge site, and just a few species of aquatic plants were subjected to analysis. Among the species that have not been investigated sufficiently is *Elodea canadensis*, which is abundant in the Yenisei River and which is traditionally used as a bioindicator in monitoring of aquatic ecosystems. Preliminary results of cytogenetic investigations of *Elodea* roots showed that at the MCC discharge site the occurrence of chromosomal aberrations in cells of *Elodea* was considerably higher than in the control areas [4]. However, plants growing in other parts of the Yenisei, including those with elevated uranium levels, have not been either analyzed for radionuclides or examined cytogenetically. As aquatic plants are very good concentrators of radionuclides, one would expect emergence of cytogenetic effects.

The purpose of the study was to assess levels of radionuclides and to evaluate the frequency of chromosomal aberrations in samples of submerged plants collected in different parts of the Yenisei River.

2. Materials and methods

Between 2001 and 2011, during the scientific expeditions, samples of water, sediment and aquatic plants were collected from the Yenisei River near the MCC and at various distances downstream, as well as in the mouth of the Kan River (Fig. 1). The Yenisei River section at the MCC receives artificial radionuclides and uranium isotopes released by the MCC; the Kan River receives uranium isotopes from the uranium enrichment facility. The control samples of aquatic plants were collected at positions upstream of the MCC, including positions at the city of Krasnoyarsk. The aquatic plants sampled belonged to the following species: *Fontinalis antipyretica, Batrachium kauffmanii, Myriophyllum spicatum, Elodea canadensis, Ceratophyllum demersum* and various *Potamogeton* species*.*

Activity concentrations of the γ-emitting nuclides in the samples of aquatic plants were measured on a γ-spectrometer (Canberra, USA) coupled to a hyper-pure germanium detector. Concentration of ^{32}P in some plant samples was calculated from the results of measurements of total β-activity on a UMo LB 123 instrument coupled to an LB 1238 detector (Berthold, Germany). Uranium concentrations in the biomass of plants were measured using a neutron activation method and mass spectrometry. Cytogenetic methods for studying aquatic plants have been described in detail elsewhere [4].

Figure 1. Diagrammatic map of the Krasnoyarsk Territory (Russia), showing villages and towns near which samples were collected. MCC - the Mining-and-Chemical Combine

3. Results and discussion

Detailed analysis of radioactive contamination of aquatic plants of the Yenisei River revealed large-scale contamination of aquatic plants as far as 250 km downstream of the MCC, indicating recent discharges of radionuclides from the Combine. About 30 radionuclides, including uranium and transuranium elements, were detected in the biomass of aquatic plants. Concentrations of radioactive phosphorus in the biomass of aquatic plants were the highest among radionuclides detected, and, hence, its concentration factor (CF) was also the highest (up to 220 000). The highest concentration factors of the major radionuclides were obtained for the aquatic moss *Fontinalis antipyretica* and *Potamogeton lucens*. The uranium CFs by the aquatic moss were the highest, reaching 60 000; the maximum uranium CFs by other species were several times lower. In *Potamogeton lucens*, activity concentrations of artificial radionuclides were somewhat higher in leaves than in stems. The plant samples collected upstream of the MCC discharge site contained only one artificial radionuclide, ^{137}Cs, with the activity levels not exceeding 5 Bq/kg.

Results of cytogenetic investigations of aquatic plants suggest that at the MCC discharge site and downstream the occurrence of chromosomal aberrations in ana-telophase and metaphase cells of the plants was considerably (up to 30%) higher than in the control areas. In the samples collected in the control areas (at Krasnoyarsk), which are not impacted by the radioactive contamination from the MCC, chromosomal aberrations did not exceed 6% [4]. Cytogenetic studies of *Elodea canadensis* samples collected at positions with elevated uranium levels (in the mouth of the Kan River) showed that the overall frequency of chromosomal aberrations in metaphase and ana-telophase reached 18%. This value is higher than that determined for the control area and comparable to the frequency of chromosomal aberrations in plants growing at the MCC discharge point. In the areas with elevated levels of uranium in the ecosystem, activity concentrations of artificial radionuclides were low. Thus, not only artificial radionuclides but also uranium concentrated in the biomass of aquatic plants can be responsible for cytogenetic aberrations observed in them.

4. References

[1] Bolsunovsky A. 2004. Artificial radionuclides in aquatic plants of the Yenisei River in the area affected by effluents of a Russian plutonium complex. Aquatic Ecology, 38 (1): 57-62.
[2] Sukovaty A., Bolsunovsky A. 2002. Exposure dose rates to aquatic organisms of the Yenisei River. In: Per Strand, Torun Jolle and Ase Sand (Eds.): Environmental Radioactivity in the Arctic & Antarctic. Norwegian Radiation Protection Authority, Norway, 337- 339.
[3] Bolsunovsky A., Sukovaty A., Pankov E. 2005. Radioactive contamination and exposure dose rates of aquatic organisms of the Yenisei River. Environmental Radioactivity in the Arctic & Antarctic. Norwegian Radiation Protection Authority, Norway, 138-141.
[4] Bolsunovsky A., Muratova E., Sukovaty A., Kornilova M. 2009. The effect of radionuclide and heavy metal contamination of the Yenisei River on cytogenetics of aquatic plant *Elodea canadensis*. Radioprotection, 44 (5): 83-88.
[5] Bolsunovsky A. 2011. Chemical Fractionation of Radionuclides and Stable Elements in Aquatic Plants of the Yenisei River. Environ. Sci. Technol. 45: 7143–7150

Planetary boundaries and chemical pollution

Cynthia A. de Wit[1]

[1]Department of Applied Environmental Science, Stockholm University, SE-10691, Stockholm, Sweden

E-mail contact: cynthia.de.wit@itm.su.se

1. Introduction

The planetary boundaries concept includes 9 boundaries for different global environmental variables within which humanity can operate safely: climate change, ocean acidification, stratospheric ozone depletion, nitrogen and phosphorus biogeochemical cycles, global freshwater use, land use, biodiversity loss, atmospheric aerosol loading and chemical pollution [1,2]. Using current scientific understanding, seven of these boundaries could be quantified. However, for aerosol loading and chemical pollution, it was not possible to quantify boundaries.

2. Results and discussion

Possible criteria that were identified for quantifying the global chemical pollution boundary were emissions, concentrations or effects on ecosystems of persistent organic pollutants (POPs), endocrine disruptors, plastics, heavy metals and nuclear waste. Due to the large number of chemicals in commerce, it is impossible to measure all possible chemicals in the environment. Another major stumbling block is lack of understanding of the effects of chemical mixtures on organisms, ecosystems and Earth system functions. Two complementary approaches were identified as possible ways to define a planetary boundary for chemical pollution: 1) focus on POPs with global distributions and 2) identify unacceptable, long-term, large-scale effects of chemical pollution on living organisms [1,2]. In the first case, data for only a few chemicals with POPs characteristics are available, for example, mercury, DDT and PCBs. In the second case, boundaries focussing on effects could be based on impacts on reproduction, immune systems and neurobehavior, particularly in sensitive species at sensitive life stages. However, it is currently difficult to link many chemicals to specific effects due to lack of toxicity data. One example of how this approach might work was the observed increase in neurodevelopmental disorders (autism, ADHD etc.) seen in children. A large number of chemicals are known to be neurotoxic in experimental animals and in humans, and five (methyl mercury, arsenic, lead, PCBs, toluene) are known to be toxic to human neurodevelopment. Thus, widespread exposure to low concentrations of many chemicals with known or suspected neurotoxic effects may have created a silent pandemic of neurodevelopmental disorders in children on a global scale [3].

3. References

[1] Rockström J et al. 2009. A safe operating space for humanity. Nature 461:472-475.
[2] Rockström J et al. 2009. Planetary boundaries: Exploring the safe operating space for humanity. Ecol Soc
[3] Grandjean P, Landrigan PJ. 2006. Developmental neurotoxicity of industrial chemicals. Lancet 386:2167-2178.

Dose-dependent effects induced by uranium at pH 4.5 in *Arabidopsis thaliana*

Eline Saenen[1,2], Nele Horemans[1], Nathalie Vanhoudt[1], Hildegarde Vandenhove[1], Geert Biermans[1,2], May Van Hees[1], Jean Wannijn[1], Jaco Vangronsveld[2], Ann Cuypers[2]

[1] Belgian Nuclear Research Centre, SCK•CEN, Biosphere Impact Studies, Boeretang 200, 2400 Mol, Belgium
[2] Hasselt University, Centre for Environmental Sciences, Agoralaan Building D, 3590 Diepenbeek, Belgium
E-mail contact: eline.saenen@sckcen.be

1. Introduction

To evaluate the environmental impact of uranium (U)-contamination, it is important to unravel the mechanisms by which plants respond to U-stress. It was already shown that U-exposure under normal laboratory conditions (pH 5.5) can disrupt the cellular redox balance and induce oxidative stress related responses in *Arabidopsis thaliana* plants [1]. However, U-speciation and as such its toxicity strongly depends on environmental parameters, such as pH [2]. In a previous experiment, we investigated the U-effects at different pHs. It seems that U is more toxic to *Arabidopsis thaliana* at pH 4.5 than at pH 7.5. The aim of this study is to analyse the biological effects induced in *Arabidopsis thaliana* exposed to different U-concentrations at low pH. We aimed to analyse growth responses and the antioxidative defence system of the plants.

2. Materials and methods

Arabidopsis thaliana plants were grown in a hydroponic system using a Hoagland nutrient solution with a pH of approximately 5.5. After 18 days pre-culture, plants were exposed to 0, 6.25, 12.5, 25, 50, 75 and 100 µM U for 3 days. The pH of the Hoagland nutrient solution was adjusted to 4.5. To retain the pH at a constant level, 500 µM MES (2-(N-morpholino)ethanesulfonic acid) and 500 µM TRIS (tris(hydroxymethyl)-aminomethane) were used. The pH of the nutrient solution was checked twice a day and adjusted if necessary. During the exposure time, a modified Hoagland solution was used with 1/80 phosphate solution [1]. After 3 days exposure, plants were harvested.

At harvest, leaf and root fresh weight was determined and samples were snap-frozen in liquid nitrogen and stored at -80 °C for further biological and molecular analysis. The photosynthetic activity of the youngest full grown leaf was determined. Samples for U-analysis were dried for at least one week at 70°C. For these determinations, roots were washed twice for 10 min in 1 mM $Pb(NO_3)_2$ and once for 10 min with distilled water to exchange surface-bound U.

Statistical analyses were performed using an ANOVA test in SAS 9.2. The ANOVA test was carried out separately for leaves and roots. Mean values of treatments were compared using Tukey's multiple comparison test. Transformations were applied when necessary to approximate the assumptions of normality and same error variance. If the assumption of normality was not fulfilled, a non-parametric Wilcoxon rank sum test was carried out.

3. Results and discussion

After 3 days exposure of *Arabidopsis thaliana* seedlings to U, the U-concentration in roots and shoots increased with increasing U-concentration added to the nutrient solution. However, the root-to-shoot transfer was small. These results are in agreement with previous studies that reported low root-to-shoot transfer at pH 5.5 [1].

A significant decrease in root and shoot fresh weight was observed after exposure to 50, 75 and 100 µM U (figure 1). The fresh weight of the roots exposed to 6.25 and 12.5 µM U significantly increased. The increased root fresh weight for lower U-concentration alludes to a hormesis effect as was observed before [1, 3].

The level of lipid peroxidation in the plant tissue was based on the thiobarbituric acid reactive compounds (TBA-rc) and is a measure for membrane damage and dysfunction. In the roots, there was only a slight increase in lipid peroxidation after U-exposure (not significant). A significant increase in lipid peroxidation was observed in the leaves of plants exposed to 25, 50 and 100 µM U (figure 2). These results are in agreement with previous studies, which reported an increased lipid peroxidation after heavy metal treatment [1, 4].

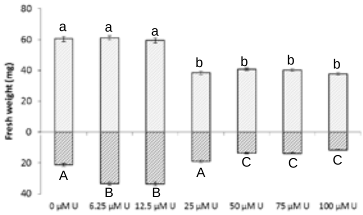

Figure 1. Fresh weight (mg) of Arabidopsis thaliana leaves (upper part) and roots (lower part) of plants exposed to different U-concentration during 3 days at pH 4.5. Statistical analyses were done separately for leaves and roots. Data points with different letters are significantly different (p<0.05).

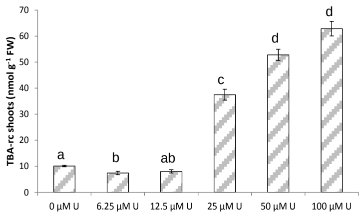

Figure 2. Level of lipid peroxidation, based on the amount of TBA reactive compounds, in Arabidopsis thaliana leaves after U-exposure during 3 days at pH 4.5. Values represent the mean ± S.E. of at least 5 biological replicates. Data points with different letters are significantly different (p<0.05).

One of the most important processes in plants is photosynthesis. In this process, plants generate carbohydrates and oxygen by using carbon dioxide, water and sunlight. The photosynthetic efficiency of *Arabidopsis thaliana* leaves was determined using a Dual-PAM-100 by measuring chlorophyll fluorescence in dark adapted leaves. *Arabidopsis thaliana* plants exposed to 25, 50, 75 and 100 µM U showed a faster increase in electron transport rate and in the energy that is used for photosynthesis (Y(II)). This increase indicates that more electrons in the chain were effectively used for photosynthesis. Due to the higher Y(II), less energy was dissipated, which in turn led to a decrease in the regulated energy dissipation. So it seems that under U-stress, *Arabidopsis thaliana* plants perform more efficiently photosynthesis.

On protein level, the enzymes of the antioxidative defence system were analysed to evaluate the importance of the cellular redox balance in *Arabidopsis thaliana* plants exposed to U. Generally, in the roots there was an increasing trend in enzyme activities with increasing U-concentration up to 50 µM U. In the leaves, there were no significant differences in enzyme activities.
Superoxide dismutase (SOD) constitutes the first line of defence against reactive oxygen species (ROS). The increased activity of SOD indicates an enhanced detoxification of superoxide ($O_2^{\bullet -}$) resulting in elevated hydrogen peroxide (H_2O_2) production. Catalase (CAT) and ascorbate peroxidase (APX) are responsible for the removal of H_2O_2. The increased activity of APX at 50 µM U could indicate an enhanced defence against H_2O_2. However, there was no increased CAT activity.
The increased activity of guaiacol peroxidase (GPX) could indicate an increased cell wall lignification as a defence reaction that limits the entry of toxic metals into the roots. We also observed an increased glutathione reductase (GR) activity in U-exposed plants. GR is important in the recycling of reduced glutathione (GSH) from oxidized glutathione (GSSG). GSH plays an important role in heavy metal responses: on the one hand, GSH is an important antioxidant, on the other hand, GSH is a precursor of phytochelatins. Phytochelatins are heavy metal-binding peptides that are important in the detoxification of toxic heavy metals. By an increased GR activity, plants could try to ensure sufficient reduced GSH levels that can be used in order to complexate U and thus decrease its toxicity.

4. Conclusions

Elevated U-concentrations at low pH can cause important morphological, physiological and biochemical effects in *Arabidopsis thaliana* seedlings. To further investigate these effects, metabolites important in the cellular redox balance will be analysed. Due to the increased GR activity, it seems that plants try to complexate U to phytochelatins. To investigate this hypothesis, phytochelatins will be examined.

5. References

[1] Vanhoudt N, Vandenhove H, Smeets K et al. 2008. Effects of uranium and phosphate concentrations on oxidative stress related responses induced in *Arabidopsis thaliana.* Plant Physiology and Biochemistry **46**(11):987-996.

[2] Ebbs SD, Brady DJ and Kochian LV. 1998. Role of uranium speciation in the uptake and translocation of uranium by plants. Journal of Experimental Botany **49**(324):1183-1190.

[3] Straczek A, Wannijn J, Van Hees M et al. 2009. Tolerance of hairy roots of carrots to U chronic exposure in a standardized in vitro device. Environmental and Experimental Botany **65**(1):82-89.

[4] Smeets K, Cuypers A, Lambrechts A et al. 2005. Induction of oxidative stress and antioxidative mechanisms in Phaseolus vulgaris after Cd application. Plant Physiology and Biochemistry **43**(5):437-444.

Agricultural land management options following large-scale environmental contamination

Hildegarde Vandenhove

Belgian Nuclear Research Centre, Biosphere Impact Studies, Boeretang 200, 2400 Mol, Belgium
E-mail contact: hvandenh@sckcen.be

1. Introduction

The accident at the Fukushima Dai-ichi Nuclear Power Plant has raised questions about the accumulation of radionuclides in soils and the transfer in the food chain. Numerous countermeasures (CM) were developed since the Chernobyl accident and applied on large scale. This presentation discusses CM strategies and their effectiveness and feasibility against the background of the Fukushima Dai-ichi nuclear accident and the agricultural areas affected.

2. Land management options

Selection of land management options should be based on a number of criteria: effectiveness; constraints on implementation; wastes generated and waste management options; doses received during implementation; side-effects; cost/benefit considerations; acceptance (stakeholder opinion). EURANOS [1] generated a critical compilation of agricultural countermeasures. Each contamination situation, however, requires a specific evaluation given differences in agricultural production system, ecosystem, culture, perception, ….

2.1. Removing contamination

Removal of the contaminated top soil (either directly or after application of a hardener [2]) is an efficient way for reducing transfer in the food chain but is unlikely feasible for large agricultural areas. A lot of waste is produced (for 5 cm topsoil removal, 500 m^3/ha) that has to be adequately disposed off. Exposure during remedial works can be high and fertile top soil is removed. In a patchy agricultural land-scape as in the Fukushima affected area, external dose is not decreased in the same proportion as the radioactivity reduction due to scatter from the surrounding non treated areas [3]. Notwithstanding, decontaminaton is the most applied management option for soils with radiactivity levels > 5000 Bq/kg radiocaesium [3].

Phyto-extraction is often put forward as a cost-effective ecological clean-up strategy. However, less than 0.5 % of ^{137}Cs is removed annually with the harvested biomass whereas 'removal' because of physical decay is 2.3 % [4]. Moreover, the contaminated biomass has to be disposed off.

2.2. Ploughing

Ploughing (shallow or deep ploughing, skim and burial ploughing) may reduce soil-to-plant transfer 1-10 fold and doses up to 80-95% [2] but may affect soil fertility and integrity and is not applicable on stony or slope soils. IAEA [3] reported dose reductions of 2.3 and 1.8 for deep ploughing and normal ploughing, respectively, in the Fukushima affected area. Ploughing has the advantage that no radioactive waste is generated and that it is less time consuming than soil removal.

2.3. Agrochemical countermeasures

Agrochemicals may be applied to arable soils and grassland to reduce plant uptake of radionuclides and hence to limit ingestion dose. Effectiveness of agrochemical CM is based on (1) either increasing the level of stable analogue nutrient (e.g. K for Cs) in soil and soil solution while guaranteeing a decreasing radionuclide:analogue ratio; or on (2) the fixation of the radionuclide in a less available form. The net effect of these changes depends largely on soil texture, clay mineral content and composition and base saturation. For example, K fertilizers will only effectively reduce Cs-uptake for soils with low K supply and low K-concentrations in the soil solution (< 20 µM) [5]. Reported efficiencies range from 0 to factor 5 decrease in transfer. Some amendments like aluminosilicates increase the soil caesium sorption pool. Effectiveness depends on the difference in caesium interception potential (RIP) between soil and amendment. Transfer reductions after application of aluminosilicates were factor two [5] to ten [6].

For Japanese soils caesium sorption characteristics are still largely unknown. Vandebroek et al. [7] reported that podzols, andosols and ferralsols, characterized by very low illite content, show the lowest RIP associated with high Cs transfer factor (TF), with log(TF) being negatively related with √RIP [8] (Figure 1). In the area affected by the Fukushima accident, brown forest soils (cambisols) and andosols dominate with patches of peaty soils, podzols and fluvisols or fluvic gleysols developing on alluvial [9]. In the area affected by the Fukushima accident, soils should be characterised in terms of RIP and potassium status in order to develop appropriate agrochemical countermeasure strategies, inclusive for permanently flooded soils.

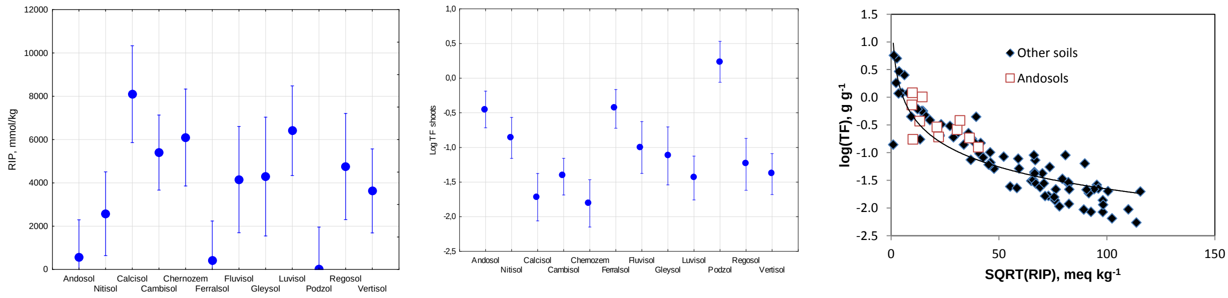

Figure 1: Radiocaesium interception potential (meq kg^{-1}) (left) [7], ryegrass rhizoplan transfer factor (TF) (g g^{-1}) (middle)for important soil groups. Right figure gives log (TF) in function of √RIP [9] for the same soils as discussed by [8].

2.4. Alternative land use

Japan has prohibited food production on soils with contamination levels in excess of 5000 Bq kg^{-1}. In such areas, industrial crops not used for food production may be an alternative. Energy crops (e.g. rape seed [10], short rotation coppice [11], …) or fibre crops [12] may be examples of valuable alternatives. The feasibility of the biofuel or fibre chain depends on radioecological, radiological, agro-technical and economic aspects but also on socio-political perception.

3. Conclusions

For optimizing agricultural management options, a good knowledge on agricultural practice and soil characteristics is required. Alternative land uses for areas were contamination levels are considered too high, need careful consideration. Planning for agricultural management options requires a holistic approach, considering radioecological, radiological, environmental, economic and socio-cultural and political aspects.

4. References

[1] EURANOS. 2011. Datasheets for countermeasure strategies in agricultural areas. http://www.euranos.fzk.de/index.php?action=euranos&title=products. Accessed 4 December 2011.
[2] Jouve A, Schulte E, Bon P, Cardot AL. 1993. Mechanical and physical removing of soil and plants as agricultural mitigation techniques. Sci Tot Environ 137 (1-3): 65-79.
[3] IAEA (International Atomic Energy Agency) Final report of the international mission on Remediation of large contaminated areas off-site the Fukushima Dai-ichi NPP. 7-15 October 2011, NE/NEFW/2011.
[4] Vandenhove H, Van Hees M, Van Winckel S. 2001. Feasibility of the phytoextraction approach to clean-up low-level uranium contaminated soil. Int J Phytorem 3(3): 301-320.
[5] Nisbet AF, Konoplev AV, Shaw G, Lembrechts JF, Merckx R, Smolders E, Vandecasteele CM, Lonsjo H, Carini F, Burton O. 1993. Application of fertilisers and ameliorants to reduce soil to plant transfer of radiocaesium and radiostrontium in the medium to long term – a summary. Sci Tot Environ, 137: 173-182.
[6] Vandenhove H, Cremers A, Smolders E. 2003. Potassium bentonite addition to soil reduces availability of cesium to plants. Eur J Soil Sci 54: 91-102.
[7] Vandebroek L, Van Hees M, Delvaux B, Spaargaren O, Thiry Y. 2012. Relevance of Radiocaesium Interception Potential (RIP) on a worldwide scale to assess soil vulnerability to ^{137}Cs contamination. J Environ Radioact 104: 87-93.
[8] Vandenhove H, Vandebroek L, Van Hees M, , Thiry Y. 2012. Relevance of Radiocaesium Interception Potential (RIP) on a worldwide scale to assess soil vulnerability to 137Cs contamination. J Environ Radioact 104: 87-93.
[9]European Soils (2011). http://eusoils.jrc.ec.europa.eu/esdb_archive/eudasm/asia/maps/JP2000_SO.htm, Accessed 4 December 2011.
[10]GOPA. 1996. Belarus: Study on Alternative Biodiesel Sources in Relation with Soil Decontamination. Bad Homburg, Germany; Project TACIS/Reg93, Contract WW 9204/0401/BOB.
[11]Vandenhove H, Thiry Y, Gommers A, Goor F, Jossart JM 2001. Short rotation coppice for re-valuation of contaminated land. J Environ Radioact 56: 157-184.
[12]Vandenhove H, Van Hees M. 2005. Fibre crops as alternative land use for radioactively contaminated arable land. J Environ Radioact 81(2-3): 131-141.

Priorities to improve ecological risk assessment for chemicals

Theo C.M. Brock

Alterra, Wageningen University and Research centre, P.O. Box 47, 6700 AA Wageningen, The Netherlands

E-mail contact: theo.brock@wur.nl

---

1. The definition of specific protection goals

An important problem formulation step in the ERA of chemicals is the definition of protection goals. The ecosystem services concept can be applied to operationalize and harmonize the generic protection goals formulated in legislative documents (as is recently done by EFSA for ERA of pesticides; EFSA 2010). For each ecosystem service potentially affected by chemicals, important taxa/functional groups need to be identified. To be sufficiently protective, and to keep the ERA procedures manageable, it is key to identify "potential vulnerable" representatives (focal species) of these taxa/functional groups on which the different risk assessment tiers should focus. From a risk manager and cost-benefit point of view, in many cases some (transient) effects of chemical exposure need to be accepted, particularly at or nearby the site of commercial application. An important prerequisite, however, may be that the acceptable impact is always reversible and that at larger distances no adverse impacts occur. This may require the development of a transparent spatial differentiation in specific protection goal options, including criteria to define specific protection goals for target sites of chemicals, mixing zones and non-target sites that require a higher level of protection. This again may require an optimal communication between risk assessors and risk managers and other stakeholders.

2. The definition of ecotoxicologically relevant exposure concentrations

Lack of a clear conceptual basis for the interface between the exposure and the effect assessment may lead to an overall low scientific quality of the risk assessment. This interface is defined as the type of concentration that gives an appropriate correlation to ecotoxicological effects, and is called by EFSA documents and Boesten *et al* (2007) the Ecotoxicologically Relevant Concentration (ERC). In ERA the ERC needs to be consistently applied so that field exposure estimates (e.g. PECs or measured concentrations) and effect estimates (e.g. Regulatory Acceptable Concentrations (RACs) or Environmental Quality Standards (EQSs)) can be compared as readily as possible. Key is that the type of ERC used to express the "C" in the PEC estimate should not be in conflict with the ERC used to express the "C" in the RAC/EQS estimate. ERCs may be different for substances that differ in toxic mode-of-action, for different populations of organisms, life stages of species, and so on. Considerations determining the choice of the ERC amongst others include (i) the environmental compartment (water, sediment, terrestrial litter layer, deeper soil layer) where the organisms at risk live or temporarily dwell, (ii) the bioavailable fraction in that compartment (e.g. pore water or total content of soil for a soil-dweller) and (iii) the time to onset of (maximum) effects to determine whether short-term or long-term exposures are relevant or whether latency of effects and/or trans-generational effects are relevant. In the near future the assessment of chronic risks of time-variable exposures to chemicals will become more important, for example for less mobile organisms exposed to chemicals that dissipate but are regularly emitted into the environment (e.g. biocides and pesticides) or for mobile organism that temporarily dwell at sites contaminated by persistent chemicals. An important question at stake is whether, and under which conditions, a time-weighted average PEC can be used in the chronic risk assessment and what should be the time-window of this TWA-PEC. In the near future many of the ERC problems might be solved by using toxicokinetic / toxicodynamic or population models and exposure scenarios specifically developed for "vulnerable" focal species (addressing the specific protection goals). Currently, an important question at stake is how to extrapolate the results of modelling (with models for a limited number of focal species) to the wider array of species potentially at risk and how to appropriately link these effect models with exposure scenarios/models (see e.g. Brock *et al*. 2010).

3. Calibrating/validating the different ERA tiers

Ideally, when many scientifically underpinned methods are available and costs are not a limiting factor, ERA's can be performed by applying the best available methods, which in the end may be computer simulation models that optimally integrate our current knowledge on factors determining the environmental fate and effects of chemicals. However, in practice ERA's are not based on an unlimited number of environmental fate and ecotoxicity data but on factors like pragmatism, costs and efficacy. This is the reason why in ERA tiered systems are developed (e.g. a tiered effect assessment scheme that includes the "standard test species-assessment factor" approach, the "species sensitivity distribution" approach, the “model ecosystem” approach and the “computer simulation model” approach). A tiered system as a whole needs to be: (i) appropriately protective, (ii) internally consistent, (iii) cost-effective, and (iv) address the problem with a higher degree of realism and complexity when going from lower to higher tiers (see e.g. Solomon *et al*. 2008). This also implies that a specific tier needs to be calibrated by comparing the results of that tier for a representative number of chemicals with the results of other tiers for the same chemicals. An important lesson learned from the past is that the consistency of the tiered approach needs to be re-evaluated every time new chemistries (with a novel toxic mode-of-action) come on the market. Consequently, a methodology to appropriately classify chemicals on basis of their (specific) toxic mode-of-action and read-across possibilities is an important priority for future research. The consistency of the tiered approach also needs to be re-evaluated when addressing mixture toxicity/multi-stress issues by chemicals. An important topic for future research will be how to prioritise the most important chemicals (environmental stressors) that contribute to chemical stress and how to extrapolate risk assessments for certain chemical combinations to that of other combinations.

4. Calibration/verification of computer simulation models

The ultimate goal of ERA is to provide knowledge that can be used to protect ecosystems (and their components) from chemical stress. To achieve this, further co-operation and communication is required between environmental chemists, ecotoxicologists and ecologists to develop computer simulation models that can be used in ERA. To date, it rarely occurs that e.g. environmental chemists/fate modellers organise workshops together with ecotoxicologists/ecologists although the main pillar of an appropriate risk assessment is the linking of exposure to effects. Ideally, laboratory tests, model ecosystem experiments and field observations should be performed in concert for developing decision support tools in the form of fate and effect models. An important pitfall in ERA remains the extrapolation of results of relatively simple model ecosystem experiments and computer simulation models to the diverse reality of the field (Solomon *et al*. 2008). In simple model ecosystem experiments (that contain a limited number of interacting populations) and in relatively simple food-web models (that model the interaction of a limited number of populations) the indirect effects of chemical-stress observed may be a caricature of reality, since not all essential feedback mechanisms that may dampen temporal chemical-stress in natural ecosystems will be captured in the simple models. On the other hand, the more or less exaggerated indirect effects observed in simple model ecosystems/ecosystem models may be highly clarifying, just as is the case with political caricatures. Nevertheless, it remains an important priority for future research to verify the outcomes of population and food-web models with results of model ecosystem that differ in complexity and to develop computer simulation models that allow the extrapolation of concentration-response relationships (including indirect effects and recovery) between ecosystems that differ in ecological complexity.

5. References

Boesten, J.J.T.I., Köpp, H., Adriaanse, P.I., Brock, T.C.M. & Forbes, F.E. (2007). Conceptual model for improving the link between exposure and effects in the aquatic risk assessment of pesticides. Ecotoxicology and Environmental Safety 66: 291-308

Brock, T.C.M., Alix, A., Brown, C.D., Capri, E., Gottesbüren, B.F.F., Heimbach, F., Lythgo, C.M., Schulz, R., Streloke, E. (Eds) (2010). Linking Aquatic Exposure and Effects: Risk Assessment of Pesticides, SETAC Press & CRC Press, Taylor & Francis Group, Boca Raton, London, New York. 410 pp

EFSA (2010). Scientific Opinion on the development of specific protection goal options for environmental risk assessment of pesticides, in particular in relation to the revision of the Guidance Documents on Aquatic and Terrestrial Ecotoxicology (SANCO/3268/2001 and SANCO/10329/2002). EFSA Journal 2010;8(10):1821

Solomon, K.R., T.C.M. Brock, D. De Zwart, S.D. Dyer, L. Posthuma, S.M. Richards, H. Sanderson, P.K. Sibley & P.J. Van den Brink (Eds) (2008). Extrapolation Practice for Ecotoxicological Effect Characterization of Chemicals, SETAC Press & CRC Press, Boca Raton, USA, 380 pp

Case study 1: prognostic – risk assessment of plant protection products

Frank de Jong[1]

[1] RIVM, P.O. Box 1, 3720 BA Bilthoven, The Netherlands
E-mail contact: frank.de.jong@rivm.nl

1. Introduction

Under the former EU directive 91/414, the (draft) Guidance Document on Terrestrial Ecotoxicology describes that the in-soil risk assessment for plant protection products (PPP's) mainly consist of testing of earthworms, functional tests with soil micro-organisms, additional testing with collembola and/or mites and a litterbag test for the higher tier. In 2011 the new directive EU 1107/2009 has come into force, and the guidance documents are being revised. The procedure of revising the guidance document starts with a definition of protection goals [1].

In parallel to this ongoing proces, a Dutch working group developed a proposal for the risk assessment of persistence of plant protection products in soil [2]. This working group tested the proposal using the public available data for four PPP's and one metabolite [3]. These elaborated examples are used to discuss the knowledge needed for a risk assessment following a tiered approach, to give a picture of the tools available at the moment and a possible need for additional tools.

2. Materials and methods

Data for the risk assessment were collected using the List of Endpoints and data from public literature. For some substances terrestrial model ecosystem studies were available, for other compounds field data were available. A tiered appoach was followed both for the exposure and for the effects (see Figure 1).

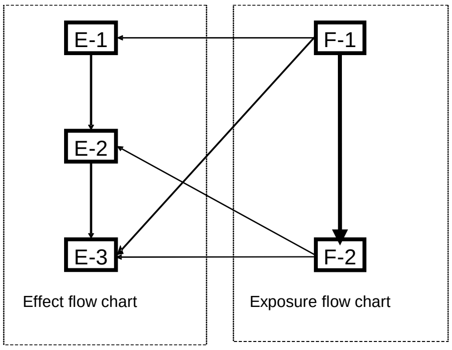

Figure 1: Tiered approach for exposure and effect.

For the first tier of the exposure flow chart a worst case concentration in the upper 5 cm of the soil was calculated. For the second tier the GeoPearl model was applied to predict a more realistic exposure concentration. For the effect flow chart EC50, EC10 or NOEC values were extracted from the available literature. Exposure was expressed as total soil content and as pore water content, in order to make a sound comparison between predicted environmental concentrations (PEC) and the regulatory acceptable concentration (RAC) possible.

3. Results and discussion

For the tiered approach for the ecological risk assessment a decission tree is proposed as summarized in Figure 2. In the first tier relevant species should be tested. For the selection of these tests a number of criteria such as taxonomy, sensitivity, availability of test methods, relevance or representativeness for the protection goal etc. are used. An appropriate assessment factor is applied to these data. In the second tier the uncertainty caused by limited number of species in the first tier is lowered, resulting in a lower assessment factor. In the third tier (semi) field studies take account of -among others- more realistic exposure conditions or recovery of populations of initially affected species within an acceptable time frame. The risk assessment can be conducted both with the total soil concent (tc) and the pore water concentration (pw).

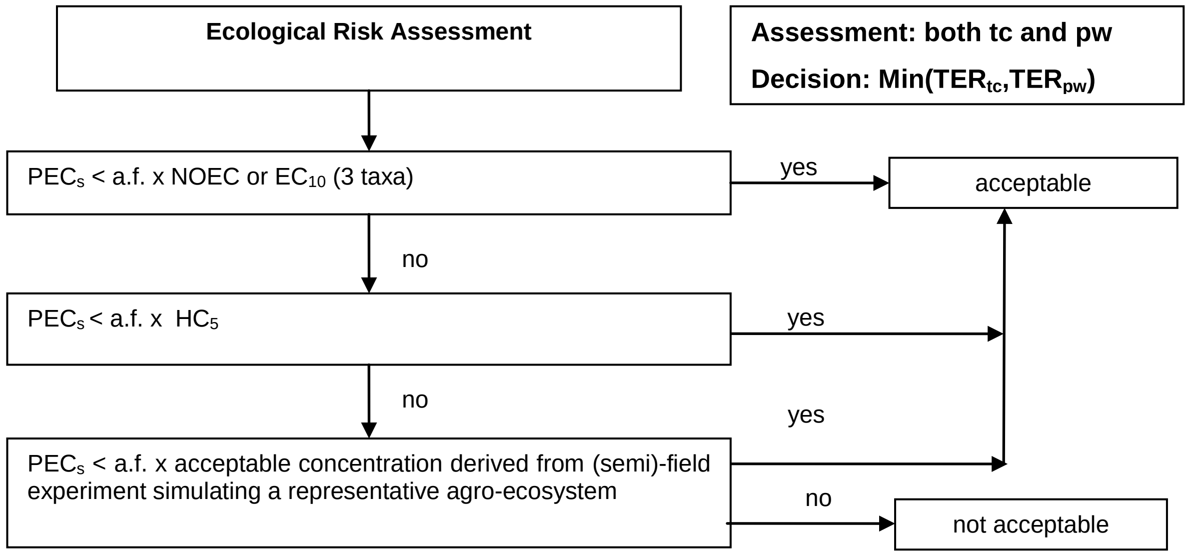

***Figure 2: Example of a decision tree for soil risk assessment*.**

In a tiered approach the lower tiers should be protective, as compared to the higher tiers. It is discussed whether sufficient knowledge (and methods) is available to ensure the basic principles of a tiered approach for in-soil risk assessment. The Dutch proposal clearly acknowledges the need for protection goals and a practical approach was followed to implement the protection goals into a risk assessment scheme. The relation between the protection goals used and soil health status is discussed.

4. Conclusions

The change of the in-soil risk assessment into a more ecological based approach results in a demand of a more diverse set of toxicity tests. The examples show that the available data do not allow an assessment for all tiers. It is worthwhile to study whether the methods developed for the assessment of soil health status could be used to fill the gaps.

5. References

[1] EFSA Panel on Plant Protection Products and their Residues (PPR). 2010. Scientific Opinion on the development of specific protection goal options for environmental risk assessment of pesticides, in particular in relation to the revision of the Guidance Documents on Aquatic and Terrestrial Ecotoxicology (SANCO/3268/2001 and SANCO/10329/2002). EFSA Journal 8 (10):1821. [55 pp.]

[2] Linden AMA van der, Boesten JJTI, Brock TCM, Eekelen GMA van, Horst MMS ter, Jong FMW de, Montforts MHMM, Pol JW. 2008. Revised proposal for the risk assessment of persistence of plant protection products in soil. Report 601712003. Bilthoven, The Netherlands. RIVM. 102 pp.

[3] Linden AMA van der, Boesten JJTI, Brock TCM, Eekelen GMA van, Horst MMS van, Jong FMW de, Montforts MHMM, Pol JW. 2008. Evaluation of the 2006 propsal for risk assessment of persistence of plant protection products in soil. Report 601712002. Bilthoven, The Netherlands. RIVM. 165 pp.

Are adverse effects of uranium mainly due to its chemotoxicity or its radiotoxicity?

Simone Al Kaddissi[1,2], Sandrine Frelon[1], Antonia-Concetta Elia[3], Alexia Legeay[2], Patrice Gonzalez[2], Frédéric Coppin[1], Daniel Orjollet[1], Virginie Camilleri[1], Karine Beaugelin-Seiller[4], Rodolphe Gilbin[1], and Olivier Simon[1].

[1]Laboratory of Radioecology and Ecotoxicology (LRE), Institute of Radioprotection and Nuclear Safety (IRSN), Bd 186, BP 3, 13115 Saint-Paul-Lez-Durance, France.

[2]Laboratory of Aquatic Ecotoxicology, University Bordeaux1/UMR CNRS 5805, Dr Peyneau square, 33120 Arcachon, France.

[3]Ecotoxicology Laboratory, Department of Cellular and Environmental Biology, University of Perugia, 06123 Perugia, Italy.

[4]Laboratory of Environmental Modelling (LME), Institute of Radioprotection and Nuclear Safety (IRSN), Bd 159, BP 3, 13115 Saint-Paul-Lez-Durance, France.

E-mail contact: simone.kaddissi@hotmail.com

1. Introduction

The ecotoxic profile of uranium (U) has not been studied extensively for non-human biota, particularly for aquatic invertebrates. The toxic action of U in organisms potentially originates from both its chemical and radiological properties, the latter depending on the specific activity of U isotopes and their associated energy radiation. However, information about distinguishing the hazardous effects of its chemotoxicity and radiotoxicity on aquatic organisms is scarce. Previous studies have demonstrated that U impacts the mitochondria of the crayfish *Procambarus clarkii* and causes oxidative stress [1,2]. The main aim of the current work was to identify the contribution of the chemotoxicity and the radiotoxicity to these U effects using transcriptional responses (*mt, sod(Mn), cox1, atp6, 12S*) and enzymatic activity as endpoints (SOD, CAT, GPx, GST). The opportunity was also taken to evaluate the sensitivity of the used biomarkers by comparing the impacts on the different biological levels of organization after short time of exposure to an environmental concentration.

2. Materials and methods

Adult males were acclimated for a month in synthetic water (which favours U biodisponibility) to laboratory conditions. Groups of crayfish were exposed for 4 and 10 days to 0 µg U/L (control) or either 30 µg/L of depleted uranium (DU) or ^{233}U which only differ from each other in their specific activity (DU =1.7×10^4 Bq.g^{-1}, ^{233}U = 3.57×10^8 Bq.g^{-1}) and hence in their radiotoxicity. U accumulation levels were measured in different organs (gills, hepatopancreas (HP), stomach, intestine, green gland, muscles, and carapace) whereas biological effects of the different types of U were evaluated only in the gills and the HP. In order to evaluate the radiotoxicity of both DU and ^{233}U, internal dose rates were calculated with EDEN-2.2 software in the HP. Gene expression levels were analyzed using quantitative real-time RT-PCR. *18s* was used as a reference gene; *sod(Mn)* was selected for its involvement in the antioxidant defence in mitochondria; *mt* was chosen for its role in metal detoxification and protection against oxidative stress; *12s* can be used as a marker of the abundance of mitochondria. *coxI*, and *atp6* were selected for their involvement in the mitochondrial metabolism. The enzymatic activity of CAT, SOD, GPX, and GST were evaluated in the cytosolic fraction of the HP and gills using the same techniques discribed in our previous work [2].

3. Main results and discussion

All organs accumulated significantly U after the waterborne exposure including the muscles which shows the necessity of assessing the potential U trophic transfer to predators. U accumulation was the highest in gills followed by the stomach,>intestine,>HP,>carapace,>green gland,>and finally the muscles. The high levels of U accumulation in the first three organs are probably due in part to U adsorption on the cuticle and the mucus. The potential additional radiotoxic effect of ^{233}U did not lead to a significant difference in accumulation levels in organs except for the carapace at T4. No explanation was obvious for this significant difference but the hypothesis of an adsorption on the carapace can be considered. This confirms the need to determine the relative U adsorption and absorption burdens .

The internal dose rate of the HP calculated after ^{233}U exposure was much higher (1.5 10^4x) than the one calculated after DU exposure. However, no significant effect of U on the studied antioxidants activity was observed in this organ. In gills of *P. clarkii*, the activity of CAT decreased (x3) after only 4 days (4d) of U exposure (Figure 1), indicating an early effect of U probably due to its chemotoxicity because no significant difference was observed between DU and ^{233}U exposures. Other defense mechanisms, not measured in this study could have been induced by U and contributed to its transitory effect on CAT. GST activity increased significantly only in gills of crayfish exposed to ^{233}U exposure conditions at day10 (Figure 1). This result can be linked to the radiotoxicity of U and demonstrates that the oxidative stress increased.

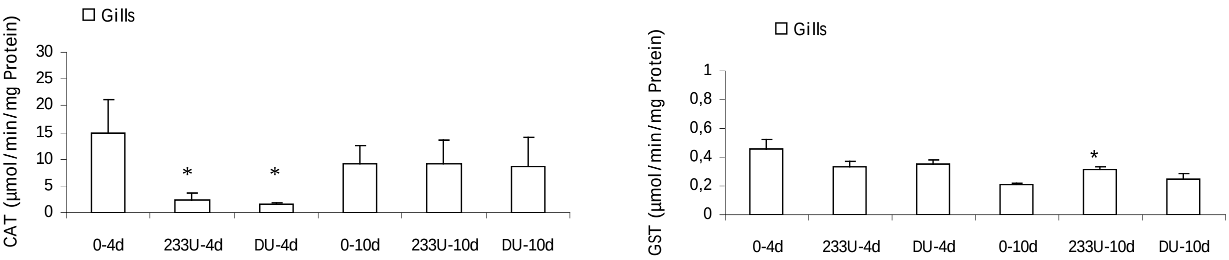

Figure 1: Enzymatic activities of catalase (CAT) (µmol/min/mg Prot, mean ± SEM, n=5 and glutathione S transferase (GST) (µmol/min/mg Prot, mean ± SEM, n=5) in of crayfish P. clarkii exposed 4 (4d) and 10 (10d) days to 0 (control) and 30 µg/L of depleted uranium (DU) or enriched uranium (^{233}U). (*) indicates significant differences between control and treated samples (ANOVA, P< 0.05).

U exposure led to a significant effect on the expression of the five studied genes in the HP and gills (Table 1). The expression of the mitochondrial genes (*12S*, *cox1* and *atp6)* were altered in presence of both contaminants, confirming that mitochondria were affected by the U exposure. Genes expression levels did not allow to distinguish a specific mechanism linked to the effect of ^{233}U since all genes had a same pattern of expression in presence of both types of U. However, higher alterations in *atp6* gene expression levels were observed in organs contaminated with ^{233}U than with DU. The radiotoxicity of ^{233}U could have amplified the effect in this case. The *mt* gene expression was initially repressed at 4d and then stimulated at the end of the experiment. It's possible that at 4d, a high basal level of metallothionein (MT) protein in gills could have compensated the U effect but was not sufficient and needed the *mt* transcription at day 10. Finally, *sod(Mn)* gene expression levels did not reflect an increase in endogenous ROS production in mitochondria. Even if DU and ^{233}U exposure led to different dose rates in the HP, it is important to note that most of the biological effects were not correlated to the radiological dose in this organ.

Table 1: Expression factors (EF) of the 5 genes studied in P. clarkii compared to the basal level of controls (n=5)

organs	Gills				Hepatopancreas			
time (days)	4		10		4		10	
Isotopes	^{233}U	DU	^{233}U	DU	^{233}U	DU	^{233}U	DU
12S	-2	-2	3	2	-5	-3	2,5	/
atp6	$\boxed{-36}$	-3	$\boxed{17}$	10	$\boxed{-6}$	/	17	17
Cox1	-2,5	-2	-2	-6	/	-2	/	/
Sod(Mn)	-2	-2	/	/	-2	/	/	/
mt	/	/	4	4	-4	-2	2	4

(/): Equal to control, (-): down-regulated, (+): up-regulated. Only EF≥2 or EF≤-2 are considered statistically significant.

4. Conclusions

Accumulation levels allowed to identify gills as the main target organ. The enzymatic activities of only CAT and GST in gills were modified after U exposure when compared to control and indicated an early generation of oxidative stress by U. The expression levels of the studied genes were strongly modified by U exposure and confirmed the damages on the mitochondria. In consequence, this endpoint shows a better sensitivity to U contamination than the enzymatic responses. Finally the tested endpoints showed that the adverse effects of U were mainly due to its chemotoxicity in our experiment since only the activity of GST in gills and the expression of *atp6* differed significantly after DU and ^{233}U exposure.

5. References

[1] Al Kaddissi, S., Legeay, A., Gonzalez, P., Floriani, M., Camilleri, V., Gilbin, R., Simon, O., 2011. Effects of uranium uptake on transcriptional responses, histological structures and survival rate of the crayfish *Procambarus clarkii*. Ecotoxicol. Environ. Saf. 74(7):1800-1807.

[2] Al Kaddissi, S., Legeay, A., Elia, C., Gonzalez, P., Camilleri, V., Gilbin, R., Simon, O., 2012. Effects of uranium on crayfish *Procambarus clarkii* mitochondria and antioxidants responses after chronic exposure: what have we learned?. Ecotoxicol. Environ. Saf. 78:218-224.

Analysis of bacterial diversity in a Chernobyl contaminated soil by pyrosequencing

N. THEODORAKOPOULOS[1], R. CHRISTEN[2,3], L. PIETTE[4,5,6], L. FEVRIER[1], F. COPPIN[1], A. MARTIN-GARIN[1], C. LE MARREC[7], C. SERGEANT[8,9], C. BERTHOMIEU[4,5,6], V. CHAPON[4,5,6]

[1] IRSN/PRP-ENV/SERIS/L2BT-Bât 186, B.P.3, Cadarache Center, F-13115 Saint-Paul-lez-Durance cedex, France
[2] Université de Nice-Sophia-Antipolis, UMR 7138, Systématique Adaptation Evolution, Parc Valrose, BP71, F-06108 Nice, France
[3] CNRS, UMR 7138, Systématique Adaptation Evolution, Parc Valrose, BP71, F-06108 Nice, France
[4] CEA, DSV, IBEB, SBVME, LIPM, F-13108 Saint-Paul-lez-Durance, France
[5] CNRS, UMR 6191, F-13108 Saint-Paul-lez-Durance, France
[6] Université d'Aix-Marseille, F-13108 Saint-Paul-lez-Durance, France
[7] ISVV, UMR 1219, Institut Polytechnique de Bordeaux/INRA, POB 50008, F-33882 Villenave d'Ornon,
[8] Univ. Bordeaux, CENBG, UMR5797, F-33170 Gradignan, France
[9] CNRS, IN2P3, CENBG, UMR5797, F-33170 Gradignan, France

E-mail contact : nicolas.theodorakopoulos@irsn.fr

1. Introduction

Chernobyl and recent Fukushima Daiichi nuclear disaster revives the importance of understanding the transfer of radioactive contamination in the environment and its ecological consequences. While some studies have been performed on higher organisms, only a few focused on bacterial communities. It is however well known that bacteria play an essential role in contaminant mobility in soils by lowering or enhancing their transfer to other compartments (e.g. water, plant, animals). Radionuclide (RN) contaminants might also exert toxic effects on bacteria hence inhibiting their role in the transfer. Thus, the objective of this study was to evaluate the impact of RNs contamination at the bacterial community level by the determination of its phylogenetic diversity. The study was carried out on soil samples coming from a trench of the Chernobyl exclusion zone, in which, contaminated soils, vegetation and other radioactive debris have been buried following the Chernobyl nuclear accident.

2. Materials and methods

The sampling site was the trench n° 22, which has been a pilot site for the study of RNs migration in soil for many years. It has been previously described in details [2]. Nine samples (number 1; 3; 4; 8; 10; 12; 13; 14 and 20), which consisted of sandy soils, were collected in and outside the trench. The determination of pH, water, ^{137}Cs and organic carbon contents, DNA extraction and bacteria isolation has been performed as described previously [1]. Metagenomic DNA extracted from soil samples was used as a template for PCR amplification with a couple of primers targeting the V3-V4 16S rRNA gene region. The PCR amplicons were sequenced on a Roche GS-FLX 454 sequencer (GATC, Germany). The 16S rRNA gene sequences were phylogenetically assigned according to their best matches to sequences of the Silva database and bacterial composition was profiled based on the abundance of each bacterial taxa. Statistical analysis was performed using the R software.

3. Results and discussion

Pyrosequencing-based analysis of 16S rRNA genes was conducted on RNs contaminated (1; 3; 4; 8; 10; 12) and non-contaminated control samples (13; 14; 20). Rarefaction curves were constructed by clustering sequences at 97% similarities. At this genetic distance, the curves reached saturation indicating that the analyses survey the full extent of taxonomic diversity. The results showed that the diversity detected in RNs contaminated soils is comparable to controls, demonstrating that a long term exposure to RNs did not lead to the decrease of bacterial diversity.

This huge diversity revealed with this method was illustrated by an average of 19,000 sequences per sample, with 963 genera and 39 phyla represented. The 4 most predominant phyla, detected in all samples, were *Chloroflexi*, *Proteobacteria*, *Acidobacteria*, and *Verrucomicrobia.* A statistical analysis (principal component analysis) was performed to evaluate the between-site variation at the phylum level. Results showed that contaminated and control samples were easily separated from each other on the first axis, which explained 31% of the total variability; suggesting the presence of specific bacterial community in contaminated soils. Some phyla such as *Chloroflexi* or *Thermotogae* were clearly more represented in control samples and may correspond to RNs sensitive species. The bacterial diversity of these samples had been previously estimated using a genetic fingerprint method (DGGE), which was not able to highlight such differences among sites [1]. Therefore our results emphasize the contribution of the pyrosequencing-based analysis compared to DGGE which only detected the predominant phylotypes.

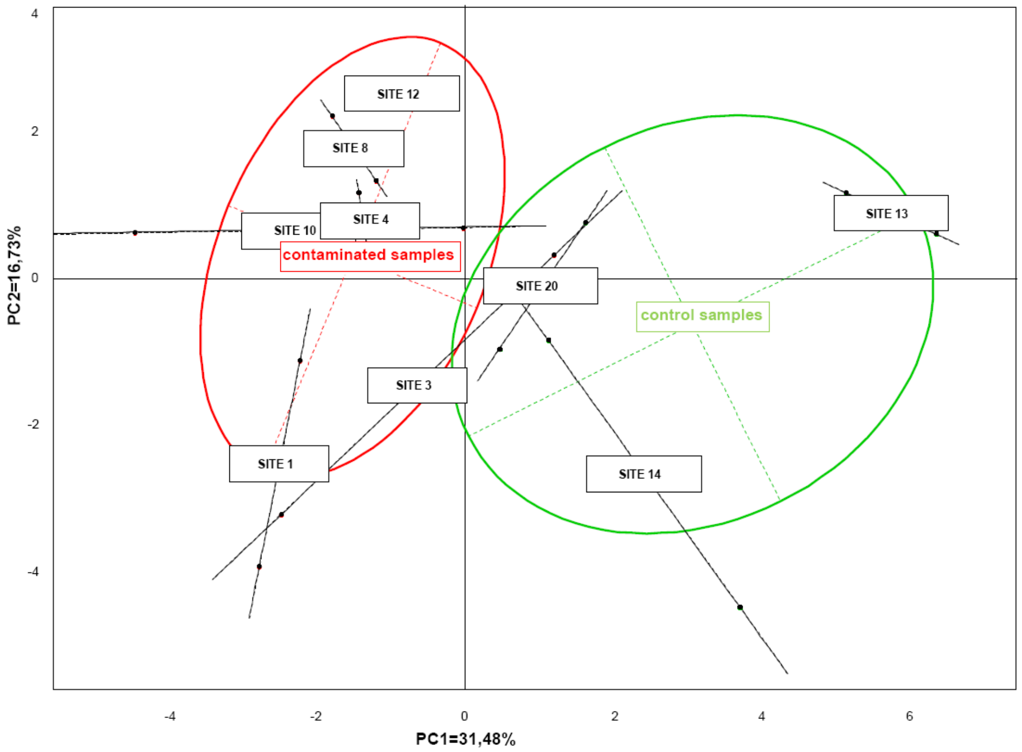

Figure 2: Graphical representation of the principal component analysis (the left red circle regrouped radioactive samples and the right green circle regrouped control samples)

4. Conclusions

These data demonstrated definitively that a long term exposure to RNs did not lead to the decrease of bacterial diversity as concluded from the DGGE analysis. Moreover, statistical analysis of the pyrosequencing data evidenced a distinction of bacterial community between contaminated and control samples, suggesting the presence of RN-adapted species in the contaminated samples. Future works will be devoted to improve the understanding of the implication of bacteria in RNs mobility in soils. The pyrosequencing data will be used for the selection of a bacteria model among a collection of 250 culturable isolates retrieved from these contaminated and control soils. This model strain will be further used in laboratory experiments designed to study interactions with some representative RNs of the trench (^{137}Cs, U, ^{90}Sr).

5. References

[1] Chapon V, Piette L, Vesvres MH, Coppin F, Marrec CL, Christen R, Theodorakopoulos N, Février L, Levchuk S, Martin-Garin A, Berthomieu C, Sergeant C. Microbial diversity in contaminated soils along the T22 trench of the Chernobyl experimental platform. Applied Geochemistry. (in press)

[2] Bugai D, Kashparov V, Dewiere L, Khomutinin Yu, Levchuk S, Yoschenko V. 2005.Characterization of subsurface geometry and radioactivity distribution in the trench containing Chernobyl clean-up wastes Environ. Geol., 47, pp. 869–881

A Planetary Boundary for Chemical Pollution

Miriam Diamond[1], Martin Scheringer[2], Sverker Molander[3], Rainer Lohman[4]

[1]University of Toronto, Toronto, Ontario Canada M5S 3G3
[2]ETH Zürich,CH-8093 Zürich, Switzerland
[3]Chalmers University, SE 412 96 Gothenburg, Sweden
[4]Graduate School of Oceanography, University of Rhode Island
E-mail contact: miriam.diamond@utoronto.ca

1. Introduction

Johan Rockström and co-authors[1] wrote a persuasive argument that human activity is exceeding the planet's biophysical boundaries. Rockström et al. went on to define 10 boundaries that we need to live within, or alternatively, face a radically different future. Chemical pollution was one of the 10 boundaries discussed, but the authors were unable to define the actual boundary.

The concept of a planetary boundary draws on earth systems science that builds on the identification of thresholds associated with global or continental scale control parameters that, if transgressed, may lead to vast disturbances possibly changing the conditions for human life in very uncertain ways. The approach of defining planetary boundaries draws on the research traditions related to concepts of self-regulation of living systems and complex system dynamics such as those articulated by C.S. Holling[2]. The concept also builds on and extends several well-known frameworks developed for addressing ecological/biophysical boundaries for human appropriation of natural resources and ecosystem functions/services, including "Limits to Growth" by Meadows et al.[3].

The case for a planetary boundary is best developed for climate change for which the boundary is the concentration of CO_2 (350 ppm, range 350-550 ppm). Beyond this concentration non-linear and unpredicable changes could occur to the earth's atmospheric and oceanic circulation patterns which, in turn, could dramatically affect global ecosystem form and functioning. Although there is uncertainty in the boundary, the science is relatively well developed. The limit for carbonate ion concentration in the surface ocean is another relatively well understood threshold, beyond which aragonite- and high-magnesium calcite-forming marine biota are considered to be imperilled. Other planetary boundaries for which the science is relatively well understood are stratospheric ozone depletion, atmospheric aerosol loading (although much debate remains concerning "safe" aerosol levels and impacts on the global climate system), phosphorus and nitrogen flows and global freshwater use. The science is less clear on the thresholds, but the impacts are clear for the planetary boundaries of land-system change (e.g., deforestation) and loss of biodiversity.

Unlike the other 9 boundaries, defining a boundary for chemical pollution will be challenging because the boundary for chemical pollution does not have a single variable, metric or indicator. Moreover, the boundary for chemical pollution may not be defined by a clear and non-linear "tipping point". Here we argue that a planetary boundary is still needed for chemical pollution. This need arises because adverse effects to ecosystem and human health from chemical pollution are being seen at "ambient" levels on a global scale.

2. Do we need a planetary boundary for chemical pollution?

We believe that a planetary boundary is necessary to address the local to global nature of chemical release and transport, and the myriad and often subtle adverse effects that are arising from single and multiple chemical exposures. Furthermore, populations are increasingly vulnerable to adverse effects from chemical pollution as a result of experiencing multiple stresses, i.e., the global system abutting against other planetary boundaries.

Examples of adverse health effects that militate for a planetary boundary for chemical pollution include learning disabilities and behavioural abnormalities in children exposed to neurodevelopmental toxicants such as lead and PCBs (both continue to be global problems!) and "newer" chemicals such as PBDE flame retardants[4]. Considerable evidence now points to adverse effects on the human male reproductive system associated with exposure to phthalate plasticizers (C1-4)[5]. We emphasize that adverse effects are being found in children at ambient levels of exposure – not high-end exposures. Ecological effects are being found of altered hormonal levels and reproductive success in birds and Arctic mammals[6]. As mentioned above, these effects can be amplified by the parallel occurrence of other stressors such as climate extremes, waters with depleted oxygen (cause by an overabundance of phosphorus and/or nitrogen), and depletion of stratospheric ozone.

While we study and attempt to regulate the chemical pollution, the rate of global chemical production is rising at about twice the rate of the global human population and global production is not waning[7]. Added to this increasing rate of production is the annual net accumulation of the total stock or inventory of chemicals, including those in landfills and storage in the technosphere[8]. A planetary boundary defines a finite limit on the total mass of chemicals anthropogenically-produced, stored, and released, above which adverse effects will occur.

3. How do we define a planetary boundary?

While we may agree on the need for a planetary boundary for chemical pollution, how can we define it? Its definition is complicated by numerous chemicals and numerous health endpoints. The discussion can quickly lead to the same vexing questions in the risk assessment sphere, i.e., how do we deal with mixtures? How do we incorporate, in a regulatory sense, the newest toxicological evidence? Questions of global equity also arise such as "do adverse effects experienced by an Arctic population necessitate a global planetary boundary?" How does one deal the fastest rate of chemical production now occurring in the rapidly expanding economies of the BRIC countries – Brazil, Russia, India and China?

We suggest starting the planetary boundary discussion by exploring the similarity with the "critical load" concept. A critical load is defined as the highest load that can be added to a system without causing a specified adverse effect in a sensitive population or ecosystem. A critical load is estimated by first choosing a sensitive toxicity endpoint in a sensitive organism, population or ecosystem, from which the corresponding emission rate is back-calculated. For the planetary boundary, the calculation should be done on a spatially resolved global scale with attention paid to ecologically vulnerable systems. Into the calculation we need to specify chemical emissions as tied to annual chemical production and inventory. Environmental chemical persistence, in addition to chemical persistence brought on by human stockpiling, must also be addressed.

Perhaps most challenging, we need to consider multiple chemical emissions rather than take a chemical-by-chemical approach. As a first step, we suggest calculating single-chemical critical loads or planetary boundaries for the few, relatively well understood high production volume chemicals for which we can gather sufficient information on physical-chemical properties, emission rates, inventory, environmental fate, exposure and toxicity. Considering lower production volume chemicals may be unnecessary.

4. Conclusions

In closing, we believe that it is imperative for environmental scientists to define a global limit to the total amount of anthropogenically-emitted chemicals entering our biosphere – a maximum amount under which we will avoid adverse effects to ecosystems and human populations. While defining a planetary boundary for chemical pollution will be emensely challenging, that should not deter us from trying. The alternative is passing to our children a world in which global chemical pollution increasingly circumscribes our health.

5. References

[1] Rockstrom J, et al. 2009, Planetary Boundaries: Exploring the Safe Operating Space for Humanity. *Ecology & Society,* 14 (2).
Rockström, J., et al. (2009), 'A safe operating space for humanity', *Nature,* 461 (7263), 472-75.

[2] Holling, CS, CR Allen. (editors) 2008 *Discontinuities in Ecosystems and Other Complex Systems.* Columbia University Press, New York, NY.

[3]Meadows DH, DL Meadows, J Randers, WW Behrens 1972. *Limits to Growth*, 1st Edition, Universe Books. Meadows DH, DL Meadows, J Randers. 1992 *Beyond the Limits*. Chelsea Green.

[4] Grandjean P and PJ Landrigan. 2006. Developmental neurotoxicity of industrial chemicals. *Lancet* 368 (9553): 2167-2178.

[5] Swan SH. 2006. Prenatal phthalate exposure and anogenital distance in male infants. *Environ Health Perspect* 114(2).

[6]Fernie KJ, JL Shutt, RJ Letcher, IJ Ritchie, DM Bird. (2009). Environmentally relevant concentrations of DE-71 and HBCD alter eggshell thickness and reproductive success of american kestrels. *Environ Sci Technol* 43(6): 2124-2130.

[7] Wilson MP, MR Schwarzman. 2009. Toward a new U.S. chemicals policy: rebuilding the foundation to advance new science, green chemistry, and environmental health. Environ Health Perspect. 117(8):1202-9.

[8] Diamond M, S Harrad 2009. The chemicals that will not go away: implications for human exposure to reservoirs of POPs. In: S.J. Harrad (Ed.) *Persistent Organic Pollutants: Current Issues and Future Challenges,* Wiley, Chichester, UK.

Persistent Organic Pollutants in Antarctica; System Input from Distant and Local Contaminant Sources

Susan Bengtson Nash[1], David McLagan[1], Darryl Hawker[1], Roger Cropp[1], Martin Schlabach[2]

[11]Griffith University, Atmospheric Environment Research Centre (AERC), Brisbane, Australia
[2]Nowegian Institute for Air Research (NILU), Kjeller, Norway
E-mail contact: s.bengtsonnash@grifith.edu.au

---

1. Introduction

Persistent Organic Pollutants (POPs) are ubiquitous toxic compounds that are incorporated into food chains with high efficiency. Polar Regions have long been established as receiving environments for POPs[1]. In order to manage environmental contamination by POPs in Antarctica, information regarding system input to this remote region is required.

The primary input of POP contamination to the Antarctic is expected to be via Long Range Atmospheric transport (LRAT)[1-2]. In addition, hydrospheric Long Range Environmental Transport (LRET) as well as *in-situ* chemical usage and to a lesser extent, migratory biota, must also be considered[3-4].

Our research has recently sought to provide information regarding the respective input pathways of POPs to the Antarctic region. Here we summarise our findings regarding long range atmospheric input; the implications of the unique oceanographic features of the Antarctic Circumpolar Current (ACC) for long range hydrospheric input of ionic perfluorinated compounds (PFCs); as well as local contamination from Casey station, an all-year research station in the Australian Antarctic Territory (AAT).

2. Materials and methods

A high-volume flow-through atmospheric sampler[5] was installed throughout 2010 approximately 3km upwind from Casey station. Atmospheric samples were analysed for organochlorine pesticides, polybrominated diphenyl ethers (PBDEs) and polychlorinated biphenyls (PCBs). Samples were analysed at the Norwegian Institute for Air Research (NILU), using methods described elsewhere e.g.[6]

We sourced 57 tissue samples from all trophic levels and from a broad geographical range for perfluorinated compound (PFC) analysis. Tissue samples represented Antarctic Krill (*Euphausia superba*), Adélie Penguin (*Pygoscelis adeliae*), Antarctic Petrel (*Thalassoica antarctica*), White-chinned Petrel (*Procellaria aequinoctialis*), Antarctic Fur Seal (*Arctocephalus gazella*), Weddell Seal (*Leptonychotes weddellii*) and Humpback Whale (*Megaptera novaeangliae*). Tissues were analysed for: Perfluorooctane sulfonate (PFOS); perfluorohexanesulfonate (PFHxS); perfluorooctanesulfonamide (PFOSA); Perfluorooctanoic acid (PFOA); perfluorononanoic acid (PFNA), perfluorodecanoic acid (PFDA); perfluoroundecanoic acid (PFUnA); perfluorododecanoic acid (PFDoA) and perfluorotridecanoic acid (PFTrA). Samples were analysed at the Danish National Environmental Research Institute (NERI) according to the methods of Bossi et al. 2005[7.]

Indoor dust, soil, and amphipod (*Paramoera walkeri*) samples were collected within Casey station perimeter and analysed for polybrominated diphenyl ethers as described in Bengtson Nash et al (2008)[8].

3. Results and discussion

3.1. Atmospheric Profiles

Atmospheric chemical profiles were dominated by hexachlorobenzene (HCB), polybrominated diphenyl ethers (PBDEs) and endosulfan I. These findings for HCB are in accordance with previous reports which have shown HCB to be a dominant compound accumulating in Southern Ocean food webs. The dominance of PBDE congeners BDE-206 and -209 was surprising as these heavier congeners are not expected to undergo long range atmospheric transport (LRAT). A local source is therefore proposed. The detection of endosulfan I is notable in light of its inclusion under the Stockholm Convention in April, 2011.

3.2. PFC accumulation in Antarctic biota Determined by Foraging Range and Antarctic Circumpolar Current

Two of fifty-seven tissue samples analysed in the current study revealed detectable concentrations of PFOS, namely liver of an adult Antarctic Fur Seal collected from sub-Antarctic Bird Island (2.0 ng/g w.w.) and pectoral muscle of a White-chinned Petrel collected from the sub-Antarctic Heard and McDonald Island region (1.2 ng/g w.w).

PFCs have not yet been detected in Antarctic biota that remain south of the Antarctic Circumpolar Current throughout their lifetime. PFCs have occasionally been detected in organisms that forage within and north of the ACC, namely Adelie and Gentoo Penguin and southern elephant seals from the Antarctic Peninsula[9-10]. This distribution of PFC contamination is congruent with the pattern expected via governance of the overturning circulation of the Southern ocean which results in export of surface waters to the north and replacement with older, currently PFC-free waters upwelling south of the ACC.

3.3 Stations as sources

The PBDE congeners BDE-206 and -209 dominated congener profiles around Casey station providing further evidence for a local source of these compounds as these heavier congeners are not expected to undergo long range atmospheric transport (LRAT).

4.0 Conclusions

These are the first results of atmospheric input of POPs to the AAT in over a decade. Further, we present the first audit of an Australian research base as a local emmitter of newly listed POPs and explain biota accumulation of PFCs in terms of species foraging ecology and the dynamics of the Antarctic circumpolar current.

It has recently been shown that climate change is beginning to mobilise global POP reservoirs[11-13]. Our findings therefore also provide a baseline for temporal monitoring of how input to this remote region stands to be impacted as global secondary sources are perturbed.

5.0 References

[1] Wania, F & Mackay, D 1993, 'Global fractionation and cold condensation of low volatility organochlorine compounds in polar regions', *Ambio*, vol. 22, no. 1, pp. 10-8.

[2] Racke, KD 2003, *Pesticide Science: Environmental Fate and Effects of Pesticides*, American Chemical Society, Washington D. C.

[3] Bengtson Nash, SM 2011, 'Persistent Organic Pollutants in Antarctica; Current and Future Research Priorities', *Journal of Environmental Monitoring*, vol. 13, pp. 497-504.

[4] Bengtson Nash, SM, Rintoul, SR, Kawaguchi, S, Staniland, I, Van den Hoff, J, Tierney, M & Bossi, R 2010, 'Perfluorinated Compounds in the Antarctic Region: Ocean Circulation Provides Prolonged Protection', *Environmental Pollution*, vol. 158, pp. 1985-91.

[5] Xiao, H, Hung, H, Harner, T, Lei, YD, Johnstone, GW & Wania, F 2007, 'A flow-through sampler for semi-volatile organic compounds in air', *Environmental Science and Technology*, vol. 41, pp. 250-6.

[6] Kallenborn, R, Christensen, G, Evenset, A & Schlabach, M 2007, 'Atmospheric transport of persistent organic pollutants (POPs) to Bjornoya (Bear island)', *Journal of Environmental Monitoring*, vol. 9, pp. 1082-91.

[7] Bossi, R, Riget, FF, Dietz, R, Sonne, C, Fauser, P, Dam, M & Vorkamp, K 2005, 'Preliminary screening of perfluorooctane sulfonate (PFOS) and other fluorochemicals in fish, birds and marine mammals from Greenland and Faroe islands', *Environmental Pollution*, vol. 136, pp. 323-9.

[8] Bengtson Nash, SM, Poulsen, AH, Kawaguchi, S, Vetter, W & Schlabach, M 2008, 'Persistent Organohalogen Contaminant Burdens in Antarctic Krill (*Euphausia superba*) From the Eastern Antarctic Sector: A Baseline Study', *Science of the Total Environment*, vol. 407, pp. 304-14.

[9] Tao, L, Kannan, K, Kajiwara, N, Costa, MA, Fillman, G, Takahashi, S & Tanabe, S 2006, 'Perfluorooctanesulfonate and related fluorochemicals in albatrosses, elephant seals, penguins and polar skuas from the Southern Ocean ', *Environmental Science and Technology*, vol. 40, pp. 7642-8.

[10] Schiavone, A, Corsolini, S, Kannan, K, Tao, L, Trivelpiece, W, Torres, D & Focardi, S 2009, 'Perfluorinated contaminants in fur seal pups and penguin eggs from South Shetland, Antarctica', *Science of the Total Environment*, vol. 407, no. 12, pp. 3899-904.

[11]Nizetto, L, Macleod, M, Borgå, K, Cabrerizo, A, Di Guardo, A, Hansen, KM, Jarvis, A, Lindroth, A, Ludwig, B, Monteith, D, Perlinger, JA, Scheringer, M, Schwendenmann, L, Semple, KT, Wick, LY, Zhang, G & Jones, KC 2010, 'Past, present and future controls on levels of Persistent Organic Pollutants in the global environment', *Environmental Science and Technology*, vol. 44, pp. 6526-31

[12] Ma, J & Cao, Z 2010, 'Quantifying the pertubation of persistent organic pollutants induced by climate change', *Environmental Science and Technology*, vol. 44, pp. 8567-73.

[13) Ma, J, Hung, CLH, Tian, C & Kallenborn, R 2011, 'Revolatilization of persistent organic pollutants in the Arctic induced by climate change', *Nature*, vol. 1, pp. 255-8.

Acknowledgement - The Australian Government Department of Sustainability, Environment, Water, Population and Communities (DEWSPaC) provided funding for the air sampling components whilst station PBDE analysis was performed under the Australian Antarctic Science (AAS) project 3115