

**Introduction:
Micro-organisms, public health
and the environment**

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This chapter looks back at the results of the OECD conference on the “Environmental Uses of Micro-Organisms: An Overview of the State-of-the-Art and Implications for Biotechnology Risk/Safety Assessment”, as well as forward, to what this could mean for the future.

Outcomes of the OECD conference on the “Environmental Uses of Micro-Organisms”

The role that micro-organisms, in particular bacteria, play in the environment can hardly be overestimated. This is often summarised in the concept put forward in 1934 by Baas Becking (*italics by him*): “*Everything is everywhere, but, the environment selects*” (see De Wit and Bouvier, 2006). Ever since it was shown that there is also much more heterogeneity in the prokaryotic world (e.g. Torsvik et al., 1990), one could add: and there is much more bacterial diversity in the soil than we ever thought possible. One could also add: and much of this diversity is still unknown to us. The micro-organisms in soil perform a vast array of intricate biogeochemical processes without which life on earth would not be possible. It is hardly feasible that such complex multi-step processes are performed by single organisms. It stands to reason, and it has become obvious, that these processes are performed by consortia of organisms, working together (de Lorenzo, 2008; and Chapter 7). Is the time of pure cultures gone, then? No, it is not, not in the sense that it would no longer make sense to study the environmental role of individual species. This is, of course, particularly the case when the role is obvious, as it is for pathogenic species. But even in those cases, the role of the complex microbial environment around the pathogen is important, as is shown in Part I.

Man has turned the resources and processes in the macroscopic environment to his use, to start with in agriculture. It stands to reason that the agricultural use of micro-organisms has been among the first uses of the microscopic environment. Further study of the microbial environment has led to insights that can be used in a variety of applications, in biotechnology as well as in medicine. An underlying presumption is that we understand the ways of the microbial world. The contributions in this volume all relate to this underlying theme, that the microbial world can be used, but that its use should be prudent and sustainable. In our regulations, a particular case is made for the prudent use of modern biotechnology and genetically engineered micro-organisms. This use should be subsequent to a risk/safety assessment which takes into account the (potential) adverse environmental effects of the micro-organisms under their conditions of use. This risk/safety assessment is not straightforward, as we have seen, man’s knowledge of the microbial environment and the processes occurring in it is still far from complete or even satisfactory. The contributions in this volume provide a further underpinning of these introductory statements, which are summarised below.

Part I: Use of micro-organisms in agriculture

Two chapters in this part focus on the use of micro-organisms as biofertilizers and for phytosanitary purposes respectively. For biofertilizers, it is pointed out that there have been extensive developments since the 1995 OECD consensus document on this subject, which have resulted in a much better understanding of the processes that are ongoing in plant-bacteria interactions. This knowledge is already used for strain improvement by genetic engineering, and suggests that the use of transgenic organisms such as biofertilizers will be an area of multiple and diverse possibilities of action in the near future. Presently, there are some 100 companies involved in the development of growth-promoting micro-organisms, and some 500 products have been registered. Like biofertilizers, the use of micro-organisms as biocontrol agents has become of vital importance for agricultural practice. Farmers are challenged to grow more, but with less fertilizer, pesticides and fumigants and more sustainable practices, such as no-till, precision farming, biocontrol. To meet these challenges, all types of traditional and new

pest management technologies are needed. Transgenic biocontrol agents (BCAs) will need to be a part of these technologies. The development of transgenic BCAs has already been ongoing for more than two decades. These are examples for the development of strains and traits that can be incorporated into strains by biotechnological means, that can be used directly for agricultural purposes, and where knowledge of other soil functions may help but is not a direct necessity for understanding the way to proceed.

The application of transgenic strains to be released into the environment as biofertilizers and BCAs requires a thorough risk/safety assessment to be carried out to determine any potential adverse environmental impacts that these strains might have. The baseline for such risk/safety evaluations is the understanding that we have of environmental microbiology. Over the last two decades, our knowledge of processes that occur in the microbial environment has increased enormously (see also Chapter 15 and 16). The contribution on risk/safety assessment points out the necessity of improving our understanding of the normal operating ranges of micro-organisms, in order to interpret the environmental impacts that may be observed as the result of releases of transgenic biofertilizers and biocontrol agents, and indeed any micro-organisms that are released into the environment.

Part II: Use of microalgae for production purposes

There is a global need for energy, and a growing conviction that more energy should be generated in a renewable way. Algae, including photosynthetic cyanobacteria, offer ideal solutions to this problem because they can be cultivated year round, on non-arable land, in various wastewater streams or brackish to marine waters, alleviating the pressure on arable land and freshwater resources that would be exerted by crops grown for biofuel purposes. Many algal strains are suitable for producing renewable fuels (biodiesel, bioethanol and kerosene). They may also become a promising source of food and feed. The production of algae, in particular microalgae, has therefore become a focus area in biotechnology development.

Chapter 5 in this part presents an overview of developments in the production of microalgae. Emphasis is given to the need to understand algal taxonomy and the consequences for strain selection for production purposes. Also, an integrated approach and complete lifecycle analyses still need to be conducted to evaluate any potential large-scale environmental implications.

Chapter 4 and 6 point out that research on transgenic algae is rapidly expanding, while large investments stimulate research on transgenic algae. Within two years, applications for outdoor, confined (e.g. in raceway ponds) cultivation of transgenic algae are expected to be actual in Europe, and earlier than that in the United States. The successful application of large-scale production of microalgae, be it in (confined) releases or in contained use, will require a science-based approach to the risk/safety assessment. Approaches to this risk/safety assessment have already been discussed in a number of workshops in the United States, and their results are available for further discussion. The development of this risk/safety assessment may furthermore require, for instance in the United States, harmonisation of the existant regulatory oversight, which in the United States is included within laws.

Part III: Use of micro-organisms for bioremediation

Bioremediation involves the application of micro-organisms for the removal of contaminants from the environment. Bioremediation competes effectively with other remediation approaches, such as thermal desorption and incineration. The development of genetically engineered strains with enhanced biodegradability capabilities looks like a promising way for further innovation of this technology. At present, however, there have been very few reported examples where genetically engineered micro-organisms have been successfully released into commercial bioremediation. One of the main reasons for this is the lack of knowledge of the environmental risks and benefits that may be caused by releasing genetically modified organisms into a contaminated area. In this respect, non-specialist stakeholder support remains a crucial area that should be further improved, concomitant as part of the sustainable development of bioremediation.

The contributions in this part make it clear that with the evolving knowledge of micro-organisms and their roles in the environment, it will be possible to design bacteria for environmental use in a focused way instead of by trial and error.

Part IV: Use of micro-organisms in cleaning products

This part focuses on the emerging problems with the regulatory oversight of microbial cleaning products. The development of microbial cleaners has occurred over the past few years, relatively unnoticed by the field, resulting in a situation where a large number of products are on the market that are more or less successful in cleaning, but where knowledge of the actual composition of the product in terms of active ingredients is lacking. This has led to concerns and regulatory approaches in the United States and in the European Union. Although it is unknown whether any transgenic micro-organisms are present in these products – and this is expected not to be the case – the regulatory developments in this field may show parallels with as well as pitfalls for the developing regulatory oversight commercialisation of transgenic micro-organisms for other applications.

Part V: Environmental applications of microbial symbionts of insects

The use of micro-organisms in the control of disease transmission by insects has become very promising over the last few years. The use of *Wolbachia* symbionts for the control of dengue transmission by mosquitoes and fighting malaria with engineered mosquito symbiotic bacteria by “paratransgenesis” are two areas that are dealt with in this part.

For the case of controlling the transmission of dengue, no transgenic micro-organisms are used (yet), but it is an interesting and important example of how to deal with this type of approach in regulatory oversight and information of and co-operation with the general public. The case of paratransgenesis is a new – and to most people unexpected – way of gene delivery to insects, that is still in its infancy, but has a strong potential to become an important and valuable tool in public health. It would be important to anticipate how to deal with this in regulatory oversight. This probably requires a fuller understanding of the very specialised microbiomes in insects.

Part VI: Environmental risk/safety assessment of the deliberate release of engineered micro-organisms

The analysis of microbial populations in the environment is a major underlying theme of environmental microbiology. The continuously developing art of DNA sequencing has proven to be a powerful and indispensable method to unravel the genetic diversity of micro-organisms in the environment. In recent years, revolutionary next-generation sequencing technologies have become widely used in various microbiological disciplines, including microbial taxonomy and ecology. New views have evolved on the species concept of prokaryotes, including bacteria and archaea. The new techniques can be used as a comprehensive methodology for monitoring microbes in soil. Next-generation sequencing-enabled metagenomics should be useful and can be widely applied to modern microbiology and biotechnology. A good understanding of these techniques, and the interpretation of the results that are gathered by these means, are vital for performing meaningful and reliable risk/safety evaluations in this field.

The environmental use of micro-organisms offers a large number of extremely interesting and promising applications that may or will involve the use of transgenic organisms. Regulatory oversight will require rigorous risk/safety assessment, based on scientific knowledge about the role of micro-organisms in the environment. When the results of the OECD conference and the contributions in this volume are placed in the context of risk/safety evaluation, some questions arise. A major question in all debates on environmental risk/safety assessment, that is also prominent in the risk/safety assessment of releases of transgenic micro-organisms, is the question of what constitutes an “adverse environmental effect”. With the present knowledge of the microbial environment, this question requires careful consideration. The OECD conference and the contributions in this volume have shown approaches to acquire the necessary knowledge, and have highlighted the knowledge and experience gained already.

Prospects and potential new biosafety projects for the OECD

A major aim of the OECD conference on the “Environmental Uses of Micro-Organisms: An Overview of the State-of-the-Art and Implications for Biotechnology Risk/Safety Assessment” was to provide a state-of-the-art overview of the environmental uses of micro-organisms, focusing on concrete or expected developments in the field of transgenic organisms, as a support for the development of the work programme of the Sub-Working Group on Micro-Organisms of the Working Group on the Harmonization of Regulatory Oversight in Biotechnology (WG-HROB).

Based on the results of the conference, it can be concluded that there are two clear areas where research has progressed to an extent that use of transgenic organisms actually occurs or is forthcoming, and could be very important for further economic developments: the use of micro-organisms in agriculture and the use of microalgae for production purposes (see Parts I and II). Projects in these areas would fit very well with the OECD’s work on green growth and sustainable development. The specific issues that would be tackled in these projects would be the scientifically sound approach of risk assessment of environmental releases of transgenic organisms for these purposes.

The ideas that were put forward in the considerations on the use of micro-organisms in bioremediation (see Part III) are conceptually very important, as are the considerations on the analysis of microbial populations in the environment that is a major underlying theme in discussions on the environmental uses of micro-organisms (see, for instance, Part VI). These ideas would be a good basis for a conceptual document on how

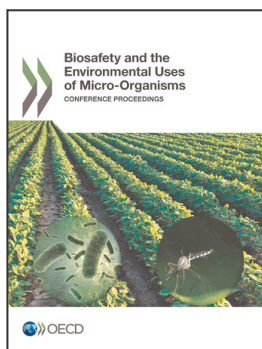
knowledge on microbial populations is gathered, and how this knowledge can be applied for predicting the behavior of soil micro-organisms, for optimising the (wanted) activity of micro-organisms in the soil (e.g. in bioremediation), as well as for optimising risk assessment of released micro-organisms. It has become clear that these underlying considerations are not always straightforward. The further development of these lines of thought, for instance in a guidance document, would be a very important complement to the projects mentioned above.

Environmental applications of microbial symbionts of insects are a specific niche within environmental microbiology that holds large promises for future developments, developments that could be very important from an economic as well as from a public health point of view. The scientific underpinning of the developments in these fields is not generally known even to all microbiologists, and they are not straightforward. A project to draft a conceptual document about the developments in this field could therefore be very important to help the development of regulation in this field proactively.

These considerations and the proposed projects will be further discussed in the Sub-Working Group on Micro-Organisms. The sub-working group will submit its conclusions to the WG-HROB, which will decide on whether and which project(s) will be initiated, while taking into account the available resources.

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