



The role of environmental biotechnology in exploring, exploiting, monitoring, preserving, protecting and decontaminating the marine environment

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In light of the Marine Strategy Framework Directive (MSFD) and the EU Thematic Strategy on the Sustainable Use of Natural Resources, environmental biotechnology could make significant contributions in the exploitation of marine resources and addressing key marine environmental problems. In this paper 14 propositions are presented focusing on (i) the contamination of the marine environment, and more particularly how to optimize the use of biotechnology-related tools and strategies for predicting and monitoring contamination and developing mitigation measures; (ii) the exploitation of the marine biological and genetic resources to progress with the sustainable, eco-compatible use of the maritime space (issues are very diversified and include, for example, waste treatment and recycling, anti-biofouling agents; bio-plastics); (iii) environmental/marine biotechnology as a driver for a sustainable economic growth.

Introduction

The most recent international strategies to re-launch global (bio)economy consider the marine environment as the last frontier. Baseline scenarios identify successful trends for high technology marine sectors that operate in a truly global market place. Such a fast developing and diversifying maritime industry can seriously threaten the marine environment.

Environmental biotechnology may provide important knowledge and tools that will help to protect the resource base upon

which marine-related economic and social activities depend. Environmental biotechnology can play a significant role in addressing marine environmental problems. These are summarized in the following paragraphs grouped in five focus areas.

Early warning systems to foresee marine threats (natural and anthropogenic)

The EU has adopted several environmental directives, strategies, recommendations, and agreements aimed at protecting the marine environment and its resources.

The EU Marine Strategy Framework Directive (MSFD; 2008/56/EC [1]) includes 11 qualitative descriptors for determining good

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environmental status (GES). Descriptors 5 (includes harmful algae blooms), 8 (contaminants and their effects), 9 (contaminants in seafood) and 10 (marine litter) are intimately related; however, their assessments often follow different approaches and are based on unrelated technologies. Biotechnology can provide a bridge to harmonize procedures and optimize resources for MSFD monitoring programs as well as to implement nearly real time early warning systems for natural and anthropogenic threats to the marine environment.

MSFD focuses on biological endpoints with ecosystem health at the center of regulation and management decision-making. Thus, the MSFD requires criteria and methodological standards to allow consistency in approach in evaluating the extent to which GES is being achieved [2]. Establishing criteria and methods to determine GES is therefore a priority challenge for basic research, aimed at establishing solid foundations to achieve harmonized assessment and monitoring procedures.

More recently, chief research efforts have been addressed toward an omics approach for the diagnosis of environmental syndromes due to, for example, pollution and climate change in marine biota and ecosystems [3–5]. In this case, biomedical advances are followed more closely by environmental and marine scientists but much remains still to be done. For instance, the use of biomarkers; a biomarker, or biological marker, refers to any characteristic, which can be measured and serves as an indicator of some biological state or condition. Molecular biomarkers measuring gene expression alterations (e.g. microarrays) after chemical exposure are to-date the front line of research in marine ecotoxicology [6–9]. We need sequence information for relevant pollution sentinel species that could be employed for the design, fabrication and commercialization of oligonucleotide high-density microarrays. One weak point in transcriptomic studies is the lack of information on the organismal/environmental relevance of alterations in gene expression profiles. High-throughput transcriptomic studies need to be assessed together to functional endpoints in order to link molecular mechanisms with phenotypic alterations. These functional endpoints should be also high throughput such as proteomics and metabolomics, and although the application of proteomics and metabolomics in the marine environment is still in its beginning, – omic studies are already in progress for marine flora, fauna and microorganisms [6–13]. Interestingly, – omics biomarkers represent a continuum of cellular responses to chemical exposure and to multiple sources of environmental stress, and provide linkages to mechanisms of cell injury/cell death or carcinogenic transformation [14].

Overall, improving mechanistic understanding, determining natural variability and baseline values, standardizing sampling and analytical procedures, integrating biomarkers among them and with chemical endpoints and relating biomarkers to ecological effects are issues of major concern for implementing biotools (biomarkers and omic diagnosis data sets) for the MSFD [15]. Criteria and methodological standards are urgently needed to allow consistency in the biomarker + omic approach for evaluating the extent to which GES is achieved [16], as well long-time series that relate pollutant exposure to effects on organisms and ecosystems at long-term scale [17].

Learning from biomedical sciences to speed up the development and use of advanced biomarkers and high-throughput technologies

suitable to foresee marine threats will provide scientists, environmentalists and decision-makers with up-to-date early warning systems for the monitoring of marine chemical pollution and its effects.

MSFD descriptors 8 and 9 deal with the environmental risk assessment of chemical contaminants. Likewise, the Water Framework Directive (WFD; 2000/60/EC [18]) and the Registration, Evaluation, Authorization and Restriction of Chemicals (REACH; 2006/1907/EC [19]) deal with the compliance of environmental quality standards established for chemical substances at European level. Chemical monitoring can be done with a combination of chemical technology and biosensors. Two major FP7 calls were recently launched on the development of biosensors for the marine environment for continuous monitoring of priority pollutants, emerging contaminants and biotoxins (funded projects: BRAAVOO and SMS). Chemical biomonitoring, that is, methylmercury contamination can also be done by measuring the behavior of fish in response to an external stimulus; in this case, the response of the fish is the biomarker. This is a very promising area where research is still very scarce.

Whole cell biosensors are detection tools based on a live bacteria that can sense a signal (of interest is the presence of a petroleum hydrocarbon), and deliver an output response that can be detected and quantified using a suitable detector device. Biosensors can be very useful for a fast and cost-effective first-line screening of the presence of particular pollutants [20–22]. Although biosensors are not aimed at substituting analytical techniques such as gas chromatography or high-pressure liquid chromatography, they can be attractive complementary tools to detect pollutants *in situ* in a cheap and flexible way, and do not need heavy and expensive equipment. Importantly, biosensors respond to the amount of the pollutant that is bioavailable (available to the cells), while chemical methods detect the total amount of the compound present (bioavailable and not bioavailable), which may overestimate the real risks in terms of toxicity [23]. Therefore, biosensors can be very useful for measuring the ecotoxicity of contaminants, as well as for monitoring bioremediation processes. For example, a whole-cell biosensor based on an engineered *Escherichia coli* bacterial strain has been successfully shown to be useful in field tests for the detection of arsenic salts in groundwater [24].

Remediation of marine oil spills

In the recent BP's Deepwater Horizon incident in the Gulf of Mexico, it was the first time that large-scale release of dispersants (COREXIT 9500) was attempted in the deep subsurface [25] directly at the wellhead at a water depth of about 1500 m. The experience collected through monitoring and dispersant use in this incident [26] is very valuable in designing new products for subsea use that address the problem of dissolved hydrocarbon gases released as the oil is moving upwards. It was the first time that a cloud of micro-droplets was monitored several kilometers from the well-head at a depth of about 1000–1300 m. Fortunately, such accidents are rare and highly unlikely to occur, however, given the recent interest in oil and gas present in the Eastern Mediterranean Sea, the potential of a well blowout accident during exploration at depths 2000–3000 m is high enough to force policy makers and end users to have suitable contingency plans in place. Based on the experience of the Deepwater Horizon accident, it is

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