

**Intergovernmental Oceanographic Commission**

Workshop Report No.195



# **INDICATORS OF STRESS IN THE MARINE BENTHOS**

Proceedings of an International workshop  
on the promotion and use of benthic tools  
for assessing the health of coastal marine ecosystems

Torregrande-Oristano, Italy  
8–9 October 2004

**UNESCO**

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Jointly organized by the Foundation IMC – International Marine Centre and  
the IOC – Intergovernmental Oceanographic Commission / Ocean Sciences  
Section of UNESCO

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## Preface

Coastal marine ecosystems are increasingly affected by environmental stress and degradation due to pollution and other anthropogenic factors. A large number of research programmes have addressed these problems within various coastal regions and have produced highly useful comprehensive data-sets on environmental and biotic conditions within these systems. A growing number of new tools and methods for assessing the health of these systems have emerged as well. The time has come now to provide an international framework for the assessment and use of common methodologies and tools as a pathway to greater capacity building, information exchange, and quality control and ultimately as a basis for improving our understanding of global patterns of ocean ecosystem health. The development of sensitive, reliable, and broadly applicable indicators of ocean health is relevant to Chapter 40 (Information for Decision-Making) of the Agenda 21 of UNCED (United Nations Conference on Environment and Development), which calls for sustainable use of the ocean environment.

In order to provide such a forum, an international workshop on “Indicators of Stress in the Marine Benthos”, jointly organized by the Foundation IMC – International Marine Centre ONLUS and the Intergovernmental Oceanographic Commission (IOC) of UNESCO, was held in Torregrande-Oriстано, Sardinia (Italy) October 8<sup>th</sup>–9<sup>th</sup> 2004. Key goals of the workshop were: (1) to exchange information on the scope and results of various programmes dealing with the development and application of environmental indicators for assessing and predicting the health of coastal ecosystems; (2) to promote the use of common approaches and tools for assessing the health of coastal marine ecosystems with applicable outputs to stakeholders and community end-users; and (3) to foster opportunities for the co-ordination of various international and national initiatives, as well as the integration of efforts across disciplines (e.g., the integration of results from field observations with related ecological and oceanographic modelling efforts). A major player in the organization of the workshop was UNESCO/IOC’s Ad-Hoc Benthic Indicator Group, established in December 1999 for the similar purpose of developing recommendations for globally applicable indicators and techniques to use in measuring the health status of marine benthic communities.

The workshop brought together international scientists and coastal managers in related fields including members of the UNESCO/IOC Ad-Hoc Benthic Indicator Group, the Benthic Ecology Working Group of the International Council for the Exploration of the Sea (ICES-BEWG), the LaguNET (Italian Network on Transitional Systems), the APAT (Italian Agency for Environmental Protection and Technical Services), and a number of other universities and research institutions. Several themes were addressed during the workshop including: (1) an overview of ongoing national and international programmes related to the topic of “Indicators of Stress in the Marine Benthos”; (2) presentations on results of individual projects that are providing new advances in the development of ecological indicators (including new field assessment techniques, diagnostic indices and data-analysis methods, modelling procedures, ecological attributes with corresponding thresholds for risk assessment, and response-to-disturbance classification schemes); (3) the development of common environmental indicators and their implications for marine and coastal ecosystem assessments; and (4) a discussion of ways to foster future collaboration among relevant initiatives brought by the participants to the workshop. These proceedings represent a compilation of the invited speakers relevant to the above themes and provide an important update on current efforts and capabilities in the use of marine benthic indicators for coastal marine assessments.

Paolo Magni  
on behalf of the Scientific Committee

### **The Workshop and the Proceedings**

About 40 participants were registered to the workshop; there were 15 invited speakers, all of them contributed to these proceedings. Central themes and unifying concepts that emerged from the workshop were also included in the Proceedings as Editorial Synthesis and Highlights.



Participants to the international workshop on “Indicators of Stress in the Marine Benthos”, jointly organized by the Foundation IMC – International Marine Centre ONLUS and the Intergovernmental Oceanographic Commission (IOC) of UNESCO, Torregrande-Oristano (Italy), 8–9 October 2004.

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Special thanks are due to the Director General, Ing. Renato Covacci, and the staff of IMC for their support and collaboration in the practical organization of this workshop, particularly to Ms. D. Marchi for logistic and secretariat assistance and to Mr. F. Angotzi for technical assistance and the layout of the proceedings.

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## Editorial Synthesis and Highlights

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### *Overall Synthesis and Some Unifying Concepts*

The workshop brought together scientists and coastal managers from different areas of the world involved in the development and application of environmental indicators for assessing and predicting the health of coastal ecosystems. Presentations and follow-up discussions focused on three general themes: development of new indicators and their implications for addressing critical coastal-research and management needs; review of relevant measurement, data-analysis, or modelling approaches; and overview of ongoing programmes related to the topic of “Indicators of Stress in the Marine Benthos,” a major emphasis of the workshop. The forum, which included two days of informal and lively round-table discussions, provided an opportunity to discuss ongoing initiatives in related fields from local to international perspectives and to exchange ideas on future directions regarding the use of environmental indicators.

One important theme that emerged was the recognition that a wide suite of tools, methods, and models would be best for such purposes rather than any one single indicator. It was also recognized that there is a wealth of existing biological and environmental observations originating from specific places and research programmes worldwide, and that there would be a tremendous advantage in bringing such information and resources together through collaborative efforts in order to provide consistent and comprehensive sets of indicators and related data for future regional to global comparisons. However, while consistent and globally applicable approaches are important, there also is a call for good regional models that provide a basis for understanding the natural states and unique properties of specific systems. Thus, there is also a need for adopting monitoring approaches that recognize and account for natural variations among various regions and ecosystem types.

The following is a list of these and other important unifying concepts that emerged from the meeting:

- Promote the use of weight-of-evidence approaches that bring together information from multiple indicators (e.g., including multiple biological endpoints as well as additional data on chemical, biogeochemical, toxicological, physical, and hydrographic conditions)
- Need for collaborative programmes to help develop consistent and comprehensive sets of indicators and processes for broad applications and comparisons (but at the same time

realizing that there are often major differences among various regions and ecosystem types, and thus that any single indicator may not work consistently across all scenarios)

- Need for better regional models of ecosystem health to help in evaluating status and trends in ecological conditions within the region and understanding whether observed changes are due to natural or human-induced sources
- Need for reliable and accurate taxonomy as a starting foundation for many benthic indicators
- Borrowing from models of freshwater ecology may serve as a useful analogue for marine applications
- Although the benthos was a major emphasis of the workshop, it is also important to think about other biological receptors that are easy to measure and may be more directly linked to public perception.

Several presentations also addressed the topic of what an ideal ecological indicator should be, with the term indicator in this case being a measure of some important attribute of the ecosystem (either biological, physical, geological, or chemical). There was general consensus that a good indicator in this context might be one that consisted of as many of the following features as possible (also see Fisher *et al.* 2001, Cairns *et al.* 1993):

- Conveys information that is meaningful and useful in decision-making with respect to the risk of concern
- Linkage to a conceptual stressor-response framework with corresponding thresholds signally the onset of conditions that may result in significant ecosystem degradation and thus require management action
- Amenable to measurement and preferably easy to measure
- High predictive ability (indicative of stress where stress should be occurring)
- Applicable over broad regions and environmental conditions
- Capable of surviving legal interrogation.

Other types of indicators were discussed and embraced including the use of various data-analysis techniques to assess change (e.g., basic statistical approaches; graphical methods; multivariate methods of classification and ordination; and diagnostic indices); the use of biological and oceanographic models that characterize the natural state and properties of a system, and thus provide a basis for detection of adverse change; and weight-of-evidence approaches (such as BENTIX, AMBI, and the Sediment Quality Triad) that combine suites of complimentary measurements as a basis for assessing current status and potential changes in environmental quality. Further highlights are summarized below.

### ***Specific Highlights***

An example of efforts to develop and evaluate indicators from a global (multi-regional) perspective was provided through presentations by the UNESCO-sponsored Ad-hoc Study Group on Benthic Indicators (<http://www.ioc.unesco.org/benthicindicators>), formed in December 1999 by the Intergovernmental Oceanographic Commission (IOC) of UNESCO. Following welcoming words by the workshop's co-organizers, P. Magni (IMC representative and member of the IOC Study Group on Benthic Indicators) and O. Vestergaard (IOC/UNESCO representative), the first three presentations were given by members of this international initiative. J. Hyland (NOAA, Charleston,

South Carolina, USA) gave an overview on the scope and activities of the committee, and J. Shine (Harvard University, Boston, Massachusetts, USA) presented results of the committee's recent efforts to look at organic-carbon content of sediment as an indicator of stress in the marine benthos. Macroinfaunal and TOC data from seven different regions of the world were examined to look for patterns of association consistent with conceptual-model predictions and to identify TOC critical points indicative of low to high risks of reductions in benthic species richness. R. Warwick (PML, Plymouth, England), member of the same IOC Study Group, also gave a presentation on the use of taxonomic distinctness ( $\Delta^+$ ) as an additional response variable for evaluating potential changes in the integrity of benthic communities in relation to anthropogenic disturbances. Desirable attributes of this and related measures of biodiversity that would be especially useful in the committee's work, as well as other similar programmes, include high sample-size independence, low sensitivity to "noise" in the data, robustness to influences of natural controlling factors (e.g., changes in salinity), and high sensitivity to detection of pollution impacts.

There were additional presentations and discussions that pertained to the topic of benthic-TOC relationships. D. Tagliapietra (CNR-ISMAR, Venice, Italy) presented the results of a large survey conducted in the Venice lagoon and showed how, and to what extent, the spatial patterns of benthic assemblages can be a function of organic matter. Tagliapietra stressed the importance of considering the degree of lability of organic matter and added that, particularly for transitional systems, the residence time seems to have a major effect on the "physiological" distribution patterns of species diversity. A proposal was then made by S. Guerzoni (CNR-ISMAR, Venice, Italy) to implement international protocols to foster the use of common methodologies (*i.e.*, TOC and organic-matter determination) and thus to help facilitate the comparison of results among different study areas. There was a general agreement on this matter. Besides the need for a better standardization of methodologies, the value of TOC as a screening-level indicator was recognized, in addition to its importance as a fundamental variable in the definition of the trophic state (and anthropogenic impact) of an ecosystem. Such efforts to standardize monitoring approaches should be applied to other variables and endpoints as well (not just the measurement of TOC).

An action plan resulted from this latter discussion. Specifically, P. Viaroli (Parma University, Italy), Tagliapietra, Guerzoni and Magni — who are all members of "LaguNet" (the Italian Network for Ecological Research in coastal lagoons and transitional waters, [www.dsa.unipr.it/lagunet](http://www.dsa.unipr.it/lagunet)) — proposed to test benthic-TOC relationships in samples from relatively similar systems, using historical datasets from coastal lagoons in Italy. The focus of such an exercise on a particular typology of coastal-marine systems might help to reduce variability in the data due to natural factors (e.g. estuaries *vs.* coastal sites, or oligotrophic *vs.* eutrophic systems), which could otherwise mask any potential effects due to anthropogenic factors. It was thus agreed to make such comparisons, based on several existing studies conducted in lagoons around Italy. As a first step, which was already started in December 2004 (Magni, Tagliapietra and Viaroli, pers. com.), each group of investigators will work autonomously on a volunteer basis, using their respective data sets. Following these initial independent analyses, there would then be an *ad-hoc* meeting among the participants to compare individual results and to discuss ways of merging the various datasets in support of a combined integrative analysis. Proper funds should be searched along the way, at both the ministerial and private level. Similarly, regarding other future efforts to coordinate results of individual research and monitoring programmes, it was proposed and agreed to have a data exchange among various participants in order to correct for potential methodological differences (e.g., sample size, sieve size, etc.) and to see whether meaningful patterns can be detected.

Other significant points and recommendations were made as well. With regard to comparing and predicting trends in benthic community structure, M. Scardi ("Tor Vergata" University, Rome, Italy) suggested that marine ecologists should be in touch with the freshwater scientific community

and take into consideration the significant experience they have gathered from these systems. H. Rumohr (IfM-GEOMAR, Kiel, Germany) also pointed out the need for developing and using regional biological models (such as the five-step benthic succession model for the Baltic Sea) as a management tool. Such models would provide a basis for understanding the natural state and properties of a system within a particular region and what to expect under disturbed conditions. À. Borja (AZTI Foundation, Spain) further underlined the need for recognizing and accounting for variations among different regions and systems. Borja, for example, pointed out the major differences that exist between northern and southern Europe. This point was also reinforced by Viaroli who described inherent differences between large and well-flushed estuaries vs. sheltered lagoons with high water retention times.

With regard to management applications, H. Rees (CEFAS, UK) gave an overview of criteria for evaluating scientific and management effectiveness of benthic indicators, which he presented as one of the current initiatives being addressed by the International Council for the Exploration of the Sea (ICES). Key criteria used in a preliminary ranking of several existing indicators are included in the list above. Rumohr presented a five-step succession conceptual model applied to the southern Baltic (as mentioned above) and highlighted the benefits of combining routine benthic sampling with sediment-imaging techniques. Rumohr, as a member of ICES and Chair of the Benthos Ecology Working Group (BEWG) of ICES, also introduced briefly the work of ICES and BEWG. ICES is the oldest organization that coordinates and promotes marine research in the north Atlantic. It acts as a focal point for a community of more than 1600 marine scientists from 19 countries around the north Atlantic. ICES plans and coordinates marine research through a system of committees, including more than 100 working groups, that cover most aspects of the marine ecosystem. The Benthos Ecology Working Group, a group of 20-25 scientists, is planning to hold a symposium on Marine Environmental Indicators in connection with the ICES "Study Group on the North Sea Benthos" in 2007. Hyland, Chair of the UNESCO/IOC *Ad-hoc* Study Group on Benthic Indicators, proposed that the 2007 ICES symposium be a forum for the presentation of any data and collaborations that may result from the present Sardinia workshop. Rees, co-convenor of the 2007 ICES symposium, also agreed to include any such developments in the symposium agenda. This idea was unanimously accepted by the workshop participants.

Presentations by À. Borja (AZTI, Spain) on the use of the AMBI Biotic Index, and by A. Zenetos (HCMR, Greece) on the use of the BENTIX index, were discussed in the context of the European Water Framework Directive (WFD 2000/60/EC), with this latter programme being described in more detail by C. Silvestri (APAT, Rome). AZTI and BENTIX were further compared in the presentation by G. Forni (Pavia University, Italy) who tested both approaches on benthic communities in the northern Adriatic Sea. Results showed that the two newly developed indices, based on the same principle (ecological identity of benthic species according to their response to pollution) produce somewhat similar results. However, there are discrepancies observed in the scoring of species and further restrictions to their use in certain environments. It was agreed that the differences should be resolved by collaboration of the two groups (AZTI and HCMR) and, if possible, that the two indices be integrated into a common tool in efforts to minimize confusion among scientists working in the Mediterranean area.

A. Marchini (Pavia University, Italy), using data on the distribution of hard-bottom assemblages in the Lagoon of Venice, described a new and innovative approach, the "fuzzy logic" model, as a potentially powerful tool for classifying different ecological sectors in such dynamic systems that are controlled by a complexity of natural factors and human influences. Furthermore, I. Karakassis (University of Crete, Greece) proposed new tools and prospects for environmental impact assessments in areas of fish farming in the eastern Mediterranean. Viaroli also proposed a biogeochemical approach to evaluate ecosystem functions and properties in coastal lagoons. Additional approaches included the use of thermodynamic and network-oriented indicators by P.

Vassallo (Genoa University, Italy) and community-structure models by E. Fresi and M. Scardi ("Tor Vergata" University, Rome, Italy). M. Zavatarelli (Bologna University, Italy) gave an overview of the Adriatic Sea ecosystem-modelling initiative, and discussed the application of a coupled physical/biogeochemical ecosystem model as a preliminary step to operational forecasting and climate-change studies of the Mediterranean Sea ecosystem.

Regarding broad-scale indicator testing at the European level, it was expressed that in spite of efforts of the European Environment Agency (EEA), various countries may not be ready yet to deliver large sets of data to EEA for such purposes. Consequently, pertinent results to date are based largely on smaller-scale case studies (e.g., Greek and Norwegian data sets). At the regional level, Zenetos on behalf of UNEP/MAP (United Nations Environment Programme, Mediterranean Action Plan) indicated that a current task of UNEP/MAP is the preparation of fact sheets on biological indicators, so as to issue guidelines on EQS to be used by Mediterranean countries. Initial results of the testing were presented with data from Greece, Turkey, Syria, Italy and Spain. A major future task will focus on additional validation with other data sets, which will require further collaboration among countries throughout the entire Mediterranean region.

There is also a need for a common set of indicators and monitoring approaches for use in shallow transitional waters (e.g., wetlands and coastal lagoons) that account for some of the unique properties of these systems. For example, one of the major goals of the EU Framework Water Directive (2000/60/EC) is to promote an agreed-upon and common approach to studies of biogeochemical processes as support for management and policy applications. Within this context, Viaroli made a brief presentation of the LaguNet network (also see above). LaguNet provides a forum for discussion and cooperation between researchers who are studying biogeochemical processes in lagoons, wetlands, and salt marshes along the Italian coasts. Key goals of LaguNet include conducting assessments of the quality of these systems, interacting with important stakeholders, and developing EU project-proposals either in Italy or in Europe (with Mediterranean EU partners as well as eventually eastern Europe and North Africa). An overarching goal of LaguNet is aimed at promoting common approaches to the study of biogeochemical processes in these ecosystems that can provide support to management or policy applications.

The LaguNet approach was also developed in Greece and Portugal, is in progress now in the Black Sea region, and will be further implemented in Spain. Overall, a product of this bottom-up networking would be a regional network federation, covering the Southern European Arc (SEANet). The LaguNet approach also has been used within the implementation plan of the Coastal module of the Global Terrestrial Observing System (C-GTOS). The goal here has been to characterize the basic stressor delivery system, namely the release of organic matter, phosphorous (P), nitrogen (N) and chemicals from watersheds into the adjacent ocean systems. A significant challenge is that shallow transitional waters and coastal lagoons are under the influence of multiple factors and have a great internal patchiness and heterogeneity, which can often bias the application of the most common indicator and indices of environmental quality and health status. Here, water-quality criteria that are suited for deep lakes and marine ecosystems cannot be used due to the shallow depth. Overall, the water-volume/surface-area ratio is of paramount importance in determining levels of ecosystem metabolism throughout benthic communities. Issues to be analysed cannot be resolved by considering only simple variables and linear relationships. Usually, the conventional trophic-status parameters and thresholds developed for deeper systems do not apply to coastal lagoons, the pelagic components being quantitatively less important than the benthic subsystem, especially when macrophytes become dominant. Under these circumstances, one should identify a set of basic benthic/sedimentary variables indicative of operative ecosystem properties and functions and that could be used for classification and quality-assessment purposes.

The approach of these latter networks at present is based mainly on the assessment of biogeochemical budgets of C, N, and P and does not consider benthic-community features. Viaroli proposed to start collaboration between LaguNet and IOC in environmental-assessment and data-exchange issues, in order to help fill some of these critical gaps. O. Vestergaard (UNESCO/IOC) indicated that the collaboration between IOC and LaguNet could help to increase methods of assessment. On behalf of UNESCO/IOC, Vestergaard also pointed out that one of the roles of IOC is to give support for data systems to developing countries. The importance of regional models was again stressed. The need for the development of a rapid-assessment programme also was expressed.

The workshop was a testimony of the importance of bringing scientists and coastal managers together in an international forum to promote open information exchange; to develop a better appreciation of the range in coastal environmental issues and corresponding approaches to addressing them in different parts of the world; and to reach consensus on solutions to some common problems, as well as the need to work collaboratively in the future (e.g., toward the development of consistent sets of indicators and protocols) to provide a stronger basis for understanding and predicting regional to global patterns of coastal-ecosystem health.

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## **Presentation Abstracts and Short Papers**

The following section is a compilation of abstracts and short papers corresponding to the various individual presentations given at the Sardinia workshop. Further information on the details of these programmes can be obtained by contacting the lead authors as indicated.

## **Developing Indicators of Stress in the Marine Benthos: the UNESCO/IOC Ad-Hoc Benthic Indicator Group**

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A special international committee of marine scientists has been set up under UNESCO's Intergovernmental Oceanographic Commission (IOC) for the purpose of developing global indicators of stress in the marine benthos. Benthic fauna are important components of coastal ecosystems, playing vital roles in detrital decomposition, nutrient cycling, and energy flow to higher trophic levels. Moreover, because of their existence within bottom substrates, benthic organisms are especially susceptible to sediment-associated stressors and may serve as sensitive indicators of environmental disturbances to the seafloor. The committee, referred to as the UNESCO/IOC Ad-Hoc Benthic Indicator Group (BIG), at present includes representatives from France, United States, Crimea, Greece, Italy, Japan, India, and Great Britain. Primary objectives of this initiative are: (1) development of reliable indicators, including key ecological attributes as well as data-analysis techniques, for measuring the health of the benthos in relation to human and natural stressors; (2) presentation of results in reports, publications, scientific meetings, and web sites to help promote wide usage; and (3) coordination with other similar international efforts to develop consistent and comprehensive sets of indicators of coastal ecosystem health for broad regional to global applications. Some desirable features to consider in the development and application of such indicators include: (1) linkage to a conceptual stressor-response framework with corresponding thresholds for management action, (2) ease of measurement, (3) ability to convey information that is meaningful to decision making with respect to the risk of concern, (4) applicability over broad regions & environmental conditions, and (5) high predictive ability (i.e., indicative of stress where stress should be occurring). The committee's work supports goals of IOC's Science on Ocean Ecosystems and Marine Environmental Protection (SOEMEP) program, aimed at developing environmental-health criteria and indices that can serve as early-warning signals of change in the quality of the world's ocean environment. The work also supports related goals of the various committee members and their parent organizations, all aimed at developing reliable indicators of coastal ecosystem health and determining potential changes in the condition of coastal resources in relation to human and natural disturbances.

## A Summary of Results of IOC-BIG Benthic-TOC Study

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While organic matter in sediments is an important source of food for benthic fauna, an overabundance can cause reductions in species richness, abundance, and biomass due to oxygen depletion and buildup of toxic byproducts (ammonia and sulphide) associated with the breakdown of these materials. Moreover, increasing organic content of sediment is often accompanied by other chemical stressors co-varying with sediment particle size. In the present study, synoptic data on the structure of macroinfaunal communities and total organic carbon (TOC) content of sediment were obtained from 951 stations representing seven coastal regions of the world: northern Black Sea (Crimean and Caucasian coasts); eastern Mediterranean Sea (Greece); North Sea (Ekofisk oil field); Firth of Clyde and Liverpool Bay, UK; Seto Inland Sea, Japan; Boston Harbor and Massachusetts Bay, USA; and estuaries along the southeastern USA. Macroinfaunal and TOC data were examined to look for patterns of association consistent with conceptual-model predictions and to identify TOC critical points corresponding to major shifts in the benthic data. Species richness, Hurlbert's  $E(S_n)$ , was selected as the primary response parameter. Results suggested that risks of reduced species richness from organic loading and other associated stressors in sediments should be relatively low at TOC concentrations < about 10 mg g<sup>-1</sup>, high at concentrations > about 35 mg g<sup>-1</sup>, and intermediate at concentrations in-between. Predictive ability across these ranges was high based on results of re-sampling simulation. While not a measure of causality, it is anticipated that these TOC critical points may be used as a general screening-level indicator for evaluating the likelihood of reduced sediment quality and associated bioeffects over broad coastal areas receiving organic wastes and other pollutants from human activities.

### Key Reference

Hyland, J., Balthis, L., Karakassis, I., Magni, P., Petrov, A., Shine, J., Vestergaard, O., Warwick, R., 2005. Organic carbon content of sediments as an indicator of stress in the marine benthos. *Marine Ecology Progress Series* (in press).

## **Taxonomic Distinctness as an Indicator of Stress in the Marine Macrobenthos**

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The effects of anthropogenic disturbances may result not only in a reduction in species richness, but also in the spread of species across the higher taxa. Both species richness and taxonomic spread are important attributes of biodiversity that should be given equal weight for environmental monitoring and conservation purposes. Newly developed biodiversity measures provide a means of comparing patterns of relatedness in community samples in the field, and monitoring changes in these patterns over space or time. They measure either the average distance apart of all pairs of individuals or species in a sample, traced through a taxonomic tree, or the variability in structure across the tree.

‘Average taxonomic diversity’ ( $\Delta$ ) is simply the average path length between every pair of individuals in a sample. ‘Average taxonomic distinctness’,  $\Delta^*$  is modified to remove some of the dependence of  $\Delta$  on the species abundance distribution and is more nearly a function of pure taxonomic relatedness of individuals. A special case is the use only of presence/absence information for each species,  $\Delta^+$ . The mean values of all three of these indices have been shown to be independent of sample size, i.e. the number of individuals in the case of  $\Delta$  and  $\Delta^*$ , and the number of species in the case of  $\Delta^+$ . Another aspect of the taxonomic structure is the ‘evenness’ of the distribution of taxa across the hierarchical taxonomic tree, or variation in taxonomic distinctness. The performance of these indices in relation to various kinds of disturbance to the macrobenthos is illustrated by three examples.

In Tees Bay, NE England, abrupt, detrimental changes coincided with a well-documented change in a variety of components of the North Sea ecosystem in the same period, termed a "regime shift". Traditional species diversity measures such as  $H'$  gave a false impression of improving environmental quality over this period.  $H'$  also failed to distinguish putatively impacted areas close to the estuary mouth compared with those more distant, despite clear differences in  $\Delta^+$  and  $\Delta^*$ .

The National Marine Monitoring Programme (NMMP) survey of macrobenthos at 67 estuarine and coastal sites around Britain has involved the combination of data of several different workers in the different regions, with variable sampling effort (number of replicates) at each site and using mesh sizes of either 0.5 or 1.0 mm for the extraction of the fauna from the sediment.

A comparison of the values of taxonomic distinctness from coastal and estuarine sites encouragingly shows no significant difference in the mean values of  $\Delta^+$ , in stark contrast to the more traditional species diversity measures which all show significantly lower values for the estuarine sites.

On the other hand, there is no clear relationship between these species diversity indices and metal levels in the sediment, whereas  $\Delta^+$  shows a clear linear trend of reduction with increasing metal levels.

Analysis of the United Kingdom Offshore Operators Association (UKOOA) database of macrobenthic and environmental data resulting from statutory monitoring programmes of offshore oil and gas installations in the North Sea shows that, although there are clear reductions in species

diversity as a result of oil and gas activities, there are also reductions in the taxonomic spread of species which would not have been detected if these novel taxonomic indices had not been applied. These effects are not confined to the immediate vicinity of the rigs but extend out as far as 10 km, which is the maximum distance sampled in most of the surveys. Since taxonomic spread is more directly related to functional diversity than is species richness, these novel measures might in fact be more important to consider in terms of the conservation of ecosystem functioning. Significant effects of oil and gas activity were detected in taxonomic distinctness indices based on simple non-quantitative species lists, and the use of such indices for monitoring purposes could result in a considerable saving in time, effort and cost.

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## A 5-Step Succession Model for the Baltic – A Future Management Tool?

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**Key words:** macrobenthos, monitoring, imaging, succession model, management tool.

The semi-enclosed, brackish Baltic Sea is characterized by steep horizontal and vertical, environmental and biological gradients. The present Baltic Sea is only about 8000 years old, and can be classified as a transitory successional stage between the more marine southern parts, and the almost limnic inner areas. The Baltic is still recovering from the „glacial trauma“.

The Baltic is a series of basins divided by sills. It has a strong salinity gradient from almost marine conditions to limnic waters. The Baltic needs inflows of marine water to maintain its integrity. This is hampered by the sills.

Eutrophication and hypoxia/anoxia are acute threats to Baltic biota, and the benthic communities registered are products of the short time available for succession, the sharp environmental gradients and the increasing anthropogenic influence.

Zoobenthos is a good indicator of environmental health. The Baltic biota are low in species diversity and the ecosystem is vulnerable to additional stress. Differences in successional sequences between basins and vertical zones (supra- and sub-halocline) are documented by Rumohr *et al.* (1996), and the changes in functional group diversity were published by (Rumohr *et al.*, 1996; Bonsdorff & Pearson, 1999).

Based on the experience we have today, it can be concluded that the combined use of sediment imaging techniques and routine benthic sampling will give a valid large-scale representation of the state of the benthic communities. It also provides a certain amount of predictivity, in that the spatially occurring clines of successional stages can represent temporal changes just as well (Pearson & Rosenberg, 1978; Bonsdorff & Pearson, 1999).

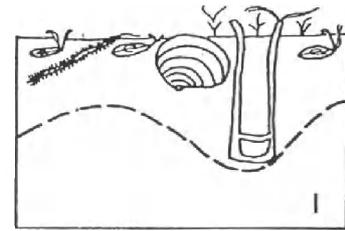
A conceptual model to analyze Baltic benthic successional stages was developed (Rumohr 1993; Rumohr *et al.*, 1996; similar to the Pearson-Rosenberg Model, 1978). The model was designed to evaluate monitoring data and as a management tool, and is a step forward in combining traditional data with modern imaging techniques (Rumohr, 1995).

The scientific basis for the model is traditional benthos data (since 1913) including HELCOM Monitoring data since 1986 and reports of fishers about anoxia and other peculiar events. Routine video and REMOTS/SPI recordings together with surface stills images were made since 1986 and formed the basis for this model.

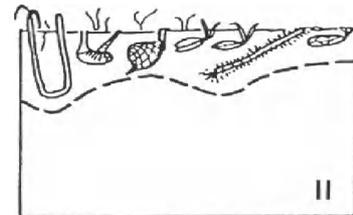
The conceptual model presented here divides the Baltic benthos into five successional stages (*sensu* Rumohr, 1993; the stages are virtually the same as in Nilsson & Rosenberg 1997 with opposite numbers) ranging from diverse communities to dead bottoms along the continuum from natural, diverse communities (stage I) to sediments completely devoid of macrofauna (stage V). The successional sequence is reversible at any point in time or space, and changes can be rapid, and stages in the development can be lost, coming further north in the Baltic.

The model based on the Baltic Sea benthos suggests 5 successional stages (Fig. 1):

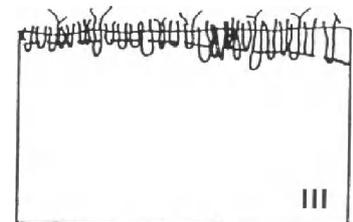
**Stage I:** a "climax-community" dominated by deep-burrowing species (Echinoderms, bivalves), and a deep (several cm's) RPD-layer (nowadays rare in the Baltic Sea),



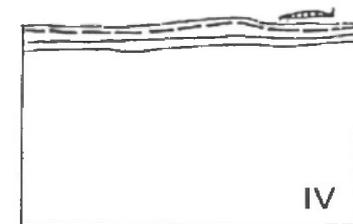
**Stage II:** first signs of stress, strong fluctuations, disappearance of echinoderms, decreasing species richness, increased productivity and biomass, and elevated RPD-layer (due to a natural lack of burrowers in much the Baltic Sea, this stage can be considered close to a natural "climax community"; Bonsdorff & Pearson, 1999),



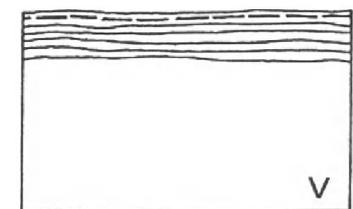
**Stage III:** dominated by small polychaete worms (or their ecological equivalents, such as oligochaetes, insect larvae, or even small molluscs), and the RPD-layer close to the sediment/water boundary layer. At this stage limited, more or less regular events of hypoxia may occur,



**Stage IV:** long periods of hypoxia and formation of bacterial mats (Beggiatoa) at the sediment surface, with no permanent benthic macrofauna (very common in the central Baltic Sea proper below the permanent halocline) but sometimes with vagile epifauna (Harmothoe spec.),



**Stage V:** complete absence of benthic communities due to long-term anoxia. Increased sedimentation rates and lack of bioturbation leads to lamination of sediments, and accumulation of organic material, nutrients and also pollutants. Sometimes vagile epifauna (Polyniods), when the bottom water is oxic.



**Figure 1.** Baltic succession model

Differences in successional sequences between the Baltic basins and vertical zones (supra- and sub-halocline) are documented (Rumohr *et al.*, 1996), and the changes in functional group diversity is also exemplified (Rumohr *et al.*, 1996; Bonsdorff & Pearson, 1999).

Based on the experience we have today, it can be concluded that the combined use of sediment imaging techniques and routine benthic sampling will give a valid large-scale representation of the state of the benthic communities that can be used for management purposes. It also provides a certain amount of predictivity, in that the spatially occurring clines of successional stages can represent temporal changes just as well (Pearson & Rosenberg, 1978; Bonsdorff & Pearson, 1999).

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## Do Benthic Indicator Tools Respond to All Impact Sources? The Case of AMBI (AZTI Marine Biotic Index)

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**Key words:** benthic indicators, AMBI, opportunistic species, sensitive species, environmental impact.

In the last years the interest on benthic indicators has increased dramatically, with a long list of new indicators proposed (see Diaz *et al.*, 2004, for a revision). One of them, the AMBI, was designed to establish the ecological quality of European coasts, investigating the response of soft-bottom communities to natural and man-induced changes in water quality (Borja *et al.*, 2000, 2003a). Hence, the AMBI offers a ‘pollution classification’ of a particular site, representing the benthic community ‘health’ (*sensu* Grall and Glémarec, 1997). The AMBI is based upon ecological models, such as those of Glémarec & Hily (1981) and Hily (1984). The theoretical basis is that of the ecological adaptative strategies of the r, k and T (McArthur & Wilson, 1967; Pianka, 1970; and Gray, 1979) and the progressive steps in stressed environments (Bellan, 1967; Pearson & Rosenberg, 1978; and Salen-Picard, 1983). Most of the concepts developed within the AMBI are based upon previous proposals, for example: (i) the species should be classified into five ecological groups (following several authors, such as Leppäkoski (1975), Glémarec & Hily (1981), and Grall & Glémarec (1997)); and (ii) with a scale introduced, from 0 to 7, based upon Hily (1984), Hily *et al.* (1986) and Majeed (1987).

However, the most novel contribution of the AMBI was the formula permitting the derivation of a series of continuous values (Borja *et al.*, 2000). Hence, taking into account the final objective of the proposal, several thresholds in the scale of the AMBI were established; those were based upon the proportions amongst the five ecological groups (see Fig. 2, in Borja *et al.*, 2000). These thresholds are coincident with the benthic community health proposed by Grall & Glémarec (1997) (see Table 1, in Borja *et al.*, 2000), whose sources can be found in Reish (1959), Bellan (1967) and Pearson & Rosenberg (1976). Further, the AMBI has been applied in the assessment of the ‘Ecological Status’, under the European Water Framework Directive (see Borja *et al.*, 2003b; 2004a; 2004b). In this particular case, these authors recommend the use of AMBI only as a part of a set of measures and indices (a multimetric approach), such as diversity, richness, etc., in order to minimize misclassification problems.

The AMBI has been verified successfully in relation to a very large set of environmental impact sources, including drill cutting discharges, submarine outfalls, harbour and dyke construction, heavy metal inputs, eutrophic processes, engineering works, diffuse pollutant inputs, recovery in polluted systems under the impact of sewerage schemes, dredging processes, mud disposal, sand extraction, oil spills, fish farming, etc. (see Table 1). On the other hand, the geographical areas where it has been applied extend over the Atlantic Sea, Baltic Sea, Mediterranean Sea, North Sea, Norwegian Sea, all in Europe, but also in Hong Kong, Uruguay and Brazil (see Table 1).

**Table 1.** Different impact sources and geographical areas for which AMBI has been applied in recent years. Key: p.c. = personal communication.

Impact Sources	Locations (Countries)	Seas	Author
Various sources along UK	(United Kingdom)		A. Miles, A. Prior (p.c., 2003)
Outfall and harbour	Brittany (France)		Borja <i>et al.</i> , 2003a
Engineering works (dyke)	Basque Country (Spain)		Borja <i>et al.</i> , 2000, 2003a
Sewerage works	Basque Country (Spain)	Atlantic Ocean	Borja <i>et al.</i> , 2000, 2003a; Gorostiaga <i>et al.</i> , 2004)
Harbour construction	Basque Country (Spain)		Muxika <i>et al.</i> , 2005
Submarine outfall	Basque Country (Spain)		Borja <i>et al.</i> , 2000; 2003b
Harbour and river inputs	Basque Country (Spain)		Muxika <i>et al.</i> , 2003
Various sources	Tejo estuary (Portugal)		M.J. Gaudencio (p.c., 2003)
Eutrophy	Mondego estuary (Portugal)		Salas <i>et al.</i> , 2004
River inputs	Guadalquivir (Spain)		AZTI (unpublished data)
Heavy metals	Huelva (Spain)		Borja <i>et al.</i> , 2003a
Estuarine inputs	Cádiz (Spain)		A. Rodríguez-Martín (p.c., 2003)
Various sources	(Morocco)		H. Bazairi (p.c., 2003)
Various sources	(Brazil, Uruguay)	Muniz <i>et al.</i> , in press	
Various sources	Latvia	Baltic Sea	V. Jermakovs (p.c., 2004)
Anoxia-hypoxia	Sweden		Muxika <i>et al.</i> , 2005
Dredging mud disposal	Sweden		S. Smith (p.c., 2003)
Various sources along Sweden	Sweden		M. Blomqvist (p.c., 2003)
Various sources in a lagoon	Smir (Morocco)	Mediterranean Sea	A. Chaouti (p.c., 2003)
Dredging in harbour	Ceuta (Spain)		Muxika <i>et al.</i> , in press
Diffuse pollution (mines, agriculture,...)	Almeria and Murcia (Spain)		Borja <i>et al.</i> , 2003a
Aquaculture cages	Murcia, Valencia (Spain)		AZTI (unpublished data)
Mining debris	Mar Menor (Spain)		L. Marin (p.c., 2004)
Submarine outfall	Catalonia (Spain)		M.J. Cardell (p.c., 2003)
Marina	Catalonia (Spain)		S. Pinedo (p.c., 2003)
Wastewater discharge in a lagoon	France		G. Reimonenq (p.c., 2003)
Inputs to a coastal lagoon	Adriatic Sea (Italy)		Caselli <i>et al.</i> , 2003
Various sources	Adriatic Sea (Italy)		Forni and Occhipinti Ambrogi, 2003
Industrial and urban pollution	Port of Trieste (Italy)		Solis-Weiss, <i>et al.</i> (in press)
Submarine outfall	Gulf of Trieste (Italy)		Solis-Weiss (p.c., 2004)
Various sources	Adriatic Sea (Italy)		R. Simonini (p.c., 2004)
Submarine outfall	Saronikos Gulf (Greece)		Borja <i>et al.</i> , 2003a
Aquaculture cages	3 locations (Greece)		Muxika <i>et al.</i> , in press
River inputs	Thames (United Kingdom)	North Sea	M. Davison (p.c., 2002)
Oil-based drilling muds (oil platforms)	11 locations (UK)		Muxika <i>et al.</i> , 2005
Impacts on sandy shores	(Netherlands)		S. Mulder (p.c., 2003)
Dredged sediment dumping	(United Kingdom)		H. Rees (p.c., 2004)
Ester-based drilling muds (oil platforms)	North Sea (Netherlands)		Borja <i>et al.</i> , 2003a
Re-opening of brackish lake to sea	Veerse Meer (Netherlands)		V. Escaravage (p.c., 2004)
Sand extraction	Belgium		Bonne <i>et al.</i> , 2003; Muxika <i>et al.</i> , 2005
Dredged sediment dumping	Hong-Kong (China)		Nicholson and Hui, 2003

Although the AMBI is particularly useful in detecting time and spatial impact gradient, its robustness could be reduced when only a very low number of taxa (1 to 3) and/or individuals are found in a sample. The same could occur when studying low-salinity locations (e.g. the very inner part of the estuaries), naturally-stressed locations (e.g. naturally organic matter enriched bottoms), or some particular impacts (e.g. sand extraction, some locations under dredged sediment dumping,

or physical impact). For problems associated with the use of AMBI, see Borja *et al.* (2004b), and the protocol for the use of AMBI contained in the free-ware software for its calculation ([www.azti.es](http://www.azti.es)). In the above mentioned particular cases Borja *et al.* (2004b) recommend the use of AMBI, together with other metrics, in order to obtain a more comprehensive view of the benthic community, being also recommended a more detailed analysis and discussion of the results.

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## The Use of BENTIX in Assessing Ecological Quality of Coastal Waters Across the Mediterranean

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**Key words:** EQS, Biotic indices, BENTIX, Mediterranean.

The Contracting Parties to Barcelona Convention at their 12th Meeting held in Monaco in November 2001 requested the MED POL Programme “To review and develop a set of marine pollution indicators, in cooperation with Blue Plan, EEA, UNIDO-ICS and other competent bodies and organizations” (UNEP/MAP, 2003a).

Based on Guidelines for the development of Ecological Status and Stress Reduction Indicators (UNEP/MAP, 2003a) and regional expert meeting, the proposed core set of indicators includes: Number of macrobenthic species, Shannon-Wiener diversity  $H'$  and BENTIX. The proposed ROAD MAP of UNEP/MAP at short term (2004-2006) includes a) developing methodology sheets (fact sheets) for the set in line with existing fact sheets developed by related organizations and b) undertaking a test procedure in a few Mediterranean countries.

This is an attempt to test BENTIX with data sets derived from:

- ❖ various geographic areas within the Mediterranean;
- ❖ coastal areas affected by different anthropogenic activities namely fishing; tourism, sewage, chemical effluent;
- ❖ the use of different sampling methodologies (sampler, mesh size, replicates).

In all cases results are compared with Ecological Quality Status assessments (EQS) according to Shannon-Wiener diversity (Zenetos & Simboura, 2001). BENTIX (Simboura & Zenetos, 2002) is a newly developed tool (based on macrozoobenthos of soft substrata) in assessing ecological quality status in accordance with the needs of the WFD.

The zoobenthic species are classified into three ecological groups and assigned a score from 1 to 3 according to their degree of tolerance or sensitivity towards pollution. The three ecological groups are:

- Group 1 (GI) includes species sensitive to disturbance in general.
- Group 2 (GII) includes species tolerant to disturbance or stress, whose populations may respond to enrichment or other source of pollution by an increase of densities (slightly unbalanced situations).
- Group 3 (GIII) includes first order opportunistic species (strongly unbalanced situations), pioneers, colonisers, and species tolerant to hypoxia.

Following many calculations, validation and testing with data from Hellenic ecosystems, an algorithm was developed giving different weight to the presence/abundance of each group:

$$\text{BENTIX} = \{ 6 \times \% \text{GI} + 2 \times (\% \text{GII} + \% \text{GIII}) \} / 100.$$

A classification system (Table 1) appears as a function of BENTIX including five levels of EQS in accordance with the needs of the WFD.

**Table 1.** Classification of EcoQ according to range of the Biotic Index

Pollution Classification	Bentix	EQS WFD	Bentix in physically stressed muds
Normal/Pristine	4,5 < BENTIX < 6	High	4 < BENTIX < 6
Slightly polluted, transitional	3,5 < BENTIX < 4,5	Good	3,0 < BENTIX < 4,0
Moderately polluted	2,5 < BENTIX < 3,5	Moderate	2,5 < BENTIX < 3,0
Heavily polluted	2 < BENTIX < 2,5	Poor	
Azoic	Azoic	Bad	

The Shannon-Wiener diversity index ( $H'$ ) has been widely used and tested in various environments. However, the use and interpretation of this index has been subjected to long debate. This index depends on sample size and effort and on habitat type and equally should refer to a standard sampling surface.

In Hellenic coastal Waters, based on the community diversity index, 5 classes of community health can be arbitrarily divided applying mostly to muddy sands or sandy muds marine benthic habitats (Table 2). The limits of these classes are somewhat arbitrary, and they are based on Hellenic literature and experience of the authors. However, it is further supported by literature in other Mediterranean areas.

**Table 2.** Ecological quality assessment in the Mediterranean Sea using the community diversity index ( $H'$ ). Source: EEA, 2004

Pollution Classification	$H'$ : Zenetos & Simboura, 2001	EQS based on $H'$	$H'$ : Simboura & Zenetos, 2002
Normal/Pristine	$H' > 4,6$	High	$H' > 5$
Slightly polluted, transitional	$4 < H' \leq 4,6$	Good	$4 < H' \leq 5$
Moderately polluted	$3 < H' \leq 4$	Moderate	$3 < H' \leq 4$
Heavily polluted	$1,5 < H' \leq 3$	Poor	$1,5 < H' \leq 3$
Azoic to heavily polluted	$0 < H' \leq 1,5$	Bad	$0 < H' \leq 1,5$

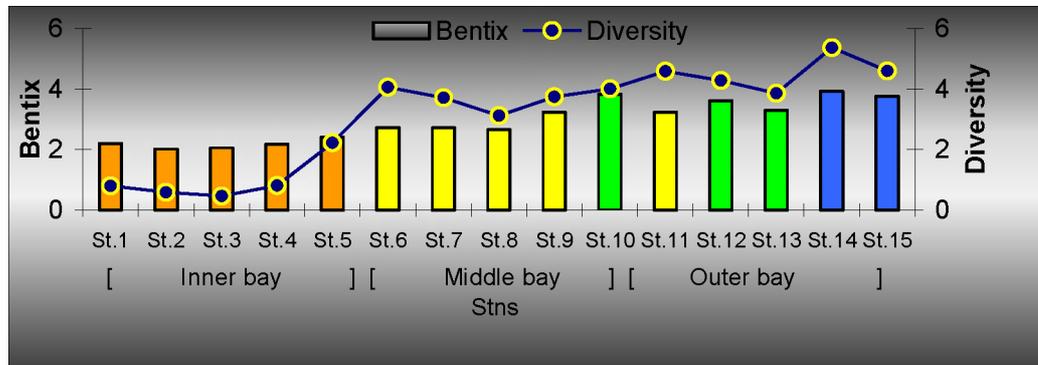
Ecological quality classes based on BENTIX and Shannon-Wiener diversity indices were not always consistent.

In **Saronikos** Gulf, receiving the sewage effluents of the Metropolitan city of Athens a Primary Treatment Plant started working in 1994. The benthic communities' ecological quality status is followed the years 1999 to 2004. As shown by Simboura & Zenetos (2002) the EQS is improving with the distance from the sewage outfall. A reference "high" quality status zone is limited in the more coastal areas.

In **E-SE Attiki** characterized by touristic development of the coastal zone, disturbance is attributed primarily to organic pollution (wastes of coastal villages, ports etc). The offshore areas of E. Attiki (Petalioi Gulf) are important fishing grounds for bottom trawlers. The EQS of the E, SE coasts of Attiki appears to be good to high. Bentix "behaves" best in assessing the EQS of coasts which are subjected to at least temporary effluent from the establishment of summer resorts. The impact of these temporal Land Based Sources (LBS) are not reflected in the community diversity albeit in cases of intense urbanization.

In **Izmir** Bay, mean values of the BENTIX and  $H'$  index are increasing from the inner towards the outer bay and so is EQS (Fig. 1). The poor quality of the inner Bay, which is subject to a

combination of pollution sources, is reflected in all parameters. Assessment by the two main indicators BENTIX and H' coincides in most cases 9 out of 15 (60%). In the innermost sites the actual EQS could be described as very polluted towards azoic since the area becomes azoic seasonally (no life present in St. 1 to St. 4 during the autumn and summer sampling).



**Figure 1.** Mean values of BENTIX and H' along a pollution gradient in Izmir Bay. Colours correspond to EQS classes as defined in the WFD (Source: Dogan, 2004).

orange yellow green blue

**Edremit Bay** is one of the most important fishery regions of the Turkish Aegean Sea. However, the innermost region of the Bay is partly affected by increasing human settlements (touristic resorts) and this is reflected in species richness of the zoobenthos (Albayrak, pers. com.). The EQS according to BENTIX appears to be moderate to high. The picture is quite different however when H' is considered. The assessments of the two indices coincided in 25% of all cases. In most contradicting assessments it is believed that BENTIX “behaved” best i.e. H' was insensitive to trawling activities. In other cases considering the species richness (S), it is believed that the EQS could be somewhere in the middle.

In **Augusta Bay**, qualitative and quantitative studies based on polychaetes and molluscs confirmed a degradation of the ecosystem between 1983 and 1985 (Di Geronimo, 1990). In a series of maps showing temporal trends in EQS as derived by BENTIX & H' no change in EQS was observed from 1984 to 1985, whereas BENTIX revealed a degradation of the shallower coastal sites (closer to LBS) and an improvement of the deeper stations.

In **Portman Bay** (Spain) the main stressor is dumping coarse metalliferous waste. The assessments derived by the BENTIX and H' did not match at all. As it appears none of the two indices was efficient in producing interpretable result but the simple parameter number of species. According to Marín-Guirao *et al.* (2005) the indicator species lists proposed by Simboura & Zenetos (2002) are based on organic pollution literature and therefore, its application in the case of purely toxic pollution was not successful.

In **Iskenderun Bay**, where the main stressor is a pipe line and a power plant, BENTIX produced similar results with Shannon-Wiener index in 60 % of the cases. The mismatching concerned mostly muddy stations where samples were collected with a dredge.

In **Banias**, a very impacted area along the Syrian coasts, results obtained by BENTIX and H' were inconclusive. With the exception of the most polluted station (site of sewage treatment plant) the assessment was always contradictory: polluted status according to H', high status according to BENTIX. This may be explained by two reasons: a) sampling was semi quantitative (dredges)

and b) most important the fauna was very poor and inadequately identified to species level. Out of 57 samples, 82 taxa were identified and of those only 56 to species level.

**Conclusively**, BENTIX appears to work successfully (different ecological quality classes corresponding to different stress) mostly in the eastern Mediterranean provided that a certain taxonomic effort is exerted (specimens assigned mostly to species level). Results were independent of mesh size used, but were misleading when based on semi qualitative data from dredges.

In any case, EQS assessments should be based on a combination of indices as the results may be misleading according to case (i.e. heavy metal pollution). Moreover, further development of this type of environmental tool requires the consensus of scientists in the assignation of species to a particular ecological group.

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## Testing Different Approaches for Quality Assessment Using the Benthic Community: Examples from the Northern Adriatic Sea

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**Keywords:** macrobenthos, soft bottoms, hard bottoms, biotic indices, fuzzy approach.

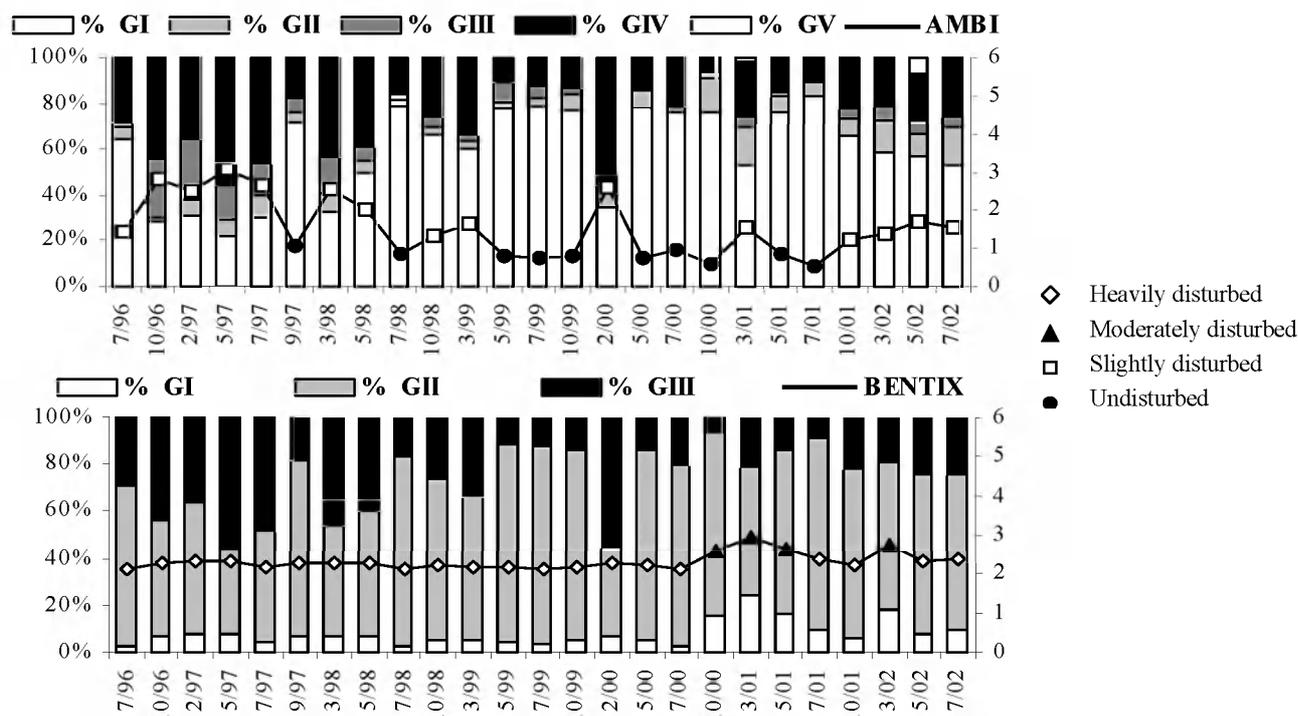
Habitat quality indicators for the marine coastal environment are in great demand following recent developments in the Water Framework Directive (WFD) which establishes that the ecological status of the water has to be determined by the condition of the biological elements including macrobenthos. Each member State should identify useful monitoring procedures for its water bodies and take into account the peculiar characteristics of its different coastal environments. In Italy, the Northern Adriatic Sea is an enclosed and very shallow sea, strongly influenced by the runoff of several rivers draining an intensively cultivated and inhabited area, resulting in a generally high trophic condition. Moreover these rivers create along the coastline a continuum of different environments, such as deltaic areas and bar-built estuaries. For this reason, we considered two different approaches: (i) the application of two biotic indices, from those indicated by WG COAST, to soft bottom macrobenthos data in the coastal environment, and (ii) the development of a fuzzy logic model based on hard bottom macrobenthos data in the lagoon environment.

The soft bottom samples were collected by Van Veen grab seasonally from July 1996 to July 2002, in six stations at different depths (3 m, 8 m, about 12 m), in two locations: Porto Garibaldi, closer to the Po river delta and Cesenatico, southward. The main chemical-physical parameters were weekly measured by the Oceanological Structure Daphne II of Cesenatico. Two biotic indices were applied on these data: the Ambi index by Borja *et al.* (2000) and the Bentix index by Simboura & Zenetos (2002).

The Ambi rates the shallow stations (3 m depth) as the most disturbed ones even though such disturbance, in most of the samples, is slight. The intermediate stations (8 m depth) and the deeper stations (about 12 m depth) are generally classified as slightly disturbed or undisturbed. On the other hand, the Bentix rates the shallow stations as the least disturbed, generally undisturbed. The intermediate and deeper stations mostly appear heavily disturbed with the exception of the intermediate southern station, which presents variable conditions, from moderately to heavily disturbed. The non parametric Spearman correlations between the two values of the indices and some chemical-physical and structural parameters have been calculated. The Ambi values are positively correlated with abundance and Simpson dominance, and negatively correlated with richness, diversity and evenness. In Figure 1, the Ambi and Bentix index variations are compared at the deeper station of Cesenatico. Most of the organisms sampled during the study period are assigned by the Ambi to the ecological group I, as is the Crustacean *Ampelisca diadema*.

For this reason, Ambi generally classifies the samples as undisturbed, or slightly disturbed in the case of high density of the Mollusc *Corbula gibba* (GIV). On the contrary, the Bentix index assigns *A. diadema* to group II (tolerant species) and *C. gibba* (a first order opportunistic species) to group III. Thus, most of the samples are classified as heavily disturbed, also due to the fact that groups II and III have the same weight in the index formula (Simboura & Zenetos, 2002). The quality level shifts to moderately disturbed when a slight increment in group I occurs.

In conclusion, the contrasting quality levels which emerge from the two indices appear mainly due to the different assignment of the species to the ecological groups, but also to the different number and weight of the ecological groups in the formula calculation. In all our stations, Ambi correlates better than Bentix both with the community structure and the chemical-physical parameters and seems more appropriate in describing the variations observed in our environment. As underlined by one of the Bentix authors (Simboura, 2004), the estuarine characteristics of this area, which hosts a low biodiversity with few species and high density, could represent a limit for the application of this index.



**Figure 1.** Variations of Ambi and Bentix indices at the deeper station of Cesenatico. The histograms represent the percentage of abundance in each ecological group (G), the line shows the index values. Symbols correspond to the disturbance levels listed in the legend (right of the graph).

A different approach has been implemented in some Northern Adriatic lagoons, using the sessile benthos living on hard bottoms such as wooden piles. A first proposal of classification of the Venice lagoon into six ecological sectors (Table 1), based on non-quantitative benthos abundance data and subjective knowledge, was presented by Occhipinti-Ambrogi *et al.* (1998). Marchini & Marchini (2004) used the fuzzy logic approach for the development of a model able to use those abundance qualitative data as input to identify an ecological sector out of the six previously put forward.

The fuzzy set theory, proposed in 1965 by L. Zadeh as a method for handling uncertainty, is particularly useful for processing vague expert knowledge and imprecise data. In the fuzzy systems a variable is not represented by a numerical value (e.g. phosphate concentration value), which has little meaning for a non-expert operator, but by an adjective that expresses a property of the variable (e.g. *limiting* or *high* phosphate concentration). The uncertainty is expressed by a membership function: an intermediate value of phosphate concentration is neither *limiting* nor *high*, but can assume partial membership grades for both the functions *limiting* and *high*. The concept of partial membership allows us to treat uncertainty as additional information instead of something undesirable to avoid: each variable is not represented by a single numerical value, but by several

adjectives that describe its properties. This approach is very similar to the ecologists' reasoning; furthermore, the fuzzy systems use logic rules (if...then) instead of complicated formulas. For example, the rules for the model of the Venice lagoon were directly obtained from the six definitions shown in Table 1: they were linguistic rules instead of mathematical expressions.

The application of the model in the Venice and Grado-Marano lagoons (Marchini & Marchini, 2004) and in the Sacca di Goro lagoon (Marchini & Occhipinti-Ambrogi, 2004) yielded results in agreement with the experts' assessments and with the literature information, showing that the fuzzy system was able to reproduce the human experience and could be used to interpret spatial differences and temporal trends within a lagoon.

In our opinion, there are three good reasons to recommend the fuzzy approach in the development of biotic indices of ecological quality: (i) a precise classification of the species into ecological groups is not required, so one species with intermediate behaviour can be both sensitive and tolerant at a certain grade and in this way uncertainty provides additional information instead of misclassification errors. Moreover, (ii) different expert judgments can be combined even if they are in disagreement because of the different responses of the species in different areas. Finally, (iii) an index can be built up without (arbitrary) weight coefficients and mathematical functions, but simply using expressions like «good species abundant and bad species scarce, then good water quality».

For these reasons fuzzy logic represents a promising tool for environmental quality evaluation, as it easily deals with qualitative information and allows to transform human experience and knowledge into an objective procedure. In conclusion, our work supports the use of macrobenthos for classification purposes and EcoQ assessments in marine and transitional waters, suggesting that models and indices that summarize expert knowledge can represent a useful management tool if site-specific information and historical knowledge are considered during validation and if the classification of the species into ecological groups is correct..

**Table 1.** Ecological sectors defined for the Venice lagoon by Occhipinti-Ambrogi et al. (1998) and used in the “if...then” form by Marchini and Marchini (2004) to create a fuzzy model.

ECOLOGICAL SECTOR	DEFINITION			
	marine species abundance	lagoon species abundance	oligohaline species abundance	total abundance
LAGOON MOUTHS	high	low	low	low
VIVIFIED LAGOON	high	low	low	medium
ROUGH EUTROPHIC	high	high	low	high
CALM EUTROPHIC	low	high	low	high
URBAN	low	high	low	low
ESTUARINE	low	low	high	low

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## Biogeochemical Processes in Coastal Lagoons: from Chemical Reactions to Ecosystem Functions and Properties

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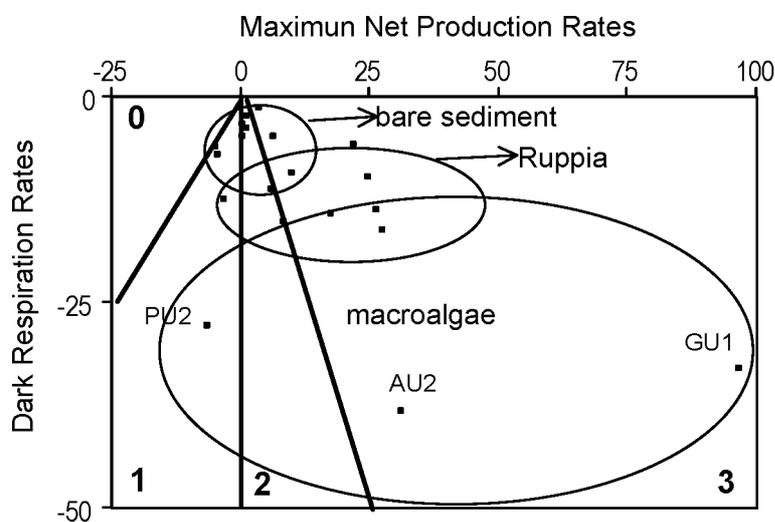
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**Key Words:** biogeochemical processes, benthic vegetation, ecosystem metabolism, buffer capacity.

Anthropogenic disturbances and natural factors often cause unpredictable non-linear responses in coastal lagoons. Important changes include the shift from seagrass to ephemeral seaweed communities, especially in sheltered habitats (Castel *et al.*, 1996). Here, biogeochemical reactions are related to primary production and decomposition processes and depend upon bioturbation by benthic fauna. Scaling up, the network of these components and reactions can be regarded as an ecosystem property which accounts for oxygen availability and for potential buffering functions - e.g. phosphate retention within the sediment, immobilisation of sulphides into the insoluble iron mono-sulphide and release of molecular nitrogen to the atmosphere (Golterman, 1995; de Wit *et al.*, 2001).

The net ecosystem metabolism basically results from primary production and microbial processes. At a first glance, it can be estimated with a simple mass balance of water, C, N and P and stoichiometric relationships, e.g. with the LOICZ biogeochemical model (Gordon *et al.*, 1996). Suitable descriptions of the ecosystem metabolism and its consequences are usually achieved with measurements of oxygen fluxes and processing over time and space (Viaroli & Christian, 2003 and references therein). Furthermore, the extent of oxygen production and consumption is given by an index resulting from the relationship between net maximum productivity, measured at saturating light, and dark respiration of the community (Fig. 1). The categorical classification of this index from autotrophy to heterotrophy provides a rapid assessment of the potential oxygen balance. It can also discriminate among different photoautotrophic conditions, including hyperautotrophy, as an abnormal oxygen production with respect to the biomass build up, and dystrophy, as the subsequent abnormal oxygen deficit which causes prolonged anoxia and the onset of anaerobic metabolism.



**Figure 1.** Representation of ecosystem metabolism with a trophic oxygen status index for lagoon with different benthic vegetation communities. Numbers represent the ecosystem metabolism categories with 0 = totally heterotrophic, 1 = net heterotrophic, 2 = net autotrophic, 3 = totally autotrophic. Units:  $\text{mmol O}_2 \text{ m}^{-2} \text{ h}^{-1}$ . For more details see Viaroli & Christian (2003).

The presence of labile organic matter combined with anoxia stimulates bacterial sulfate reduction, resulting in the production of toxic sulphide. Reactions of sulphide with iron represent an efficient buffering mechanism, which removes sulphide as insoluble FeS and FeS<sub>2</sub> (Heijs *et al.*, 2000; Azzoni *et al.* 2001; de Wit *et al.* 2001). The potential sedimentary buffering capacity of iron is usually assessed by determining reactive iron and its degree of pyritisation (Rozan *et al.*, 2002). To a first approximation, the buffering capacity of iron can be also estimated with the reactive Fe(II) to AVS ratio, with AVS (acid volatile sulphide) representing the iron quota precipitated as FeS (Viaroli *et al.*, 2004).

Phosphorus speciation depends mostly on reactions with calcium, carbonates, iron, aluminium, and humic compounds. Reactions of the calcium-carbonate-phosphate system control the formation of stable P-species and their strong retention within the sediment, whilst the iron hydroxide-phosphate-sulphide system has a weaker buffering capacity and regulates the short term cycling of phosphates (Golterman, 1995, Heijs *et al.*, 2000; Rozan *et al.*, 2002). Under oxic conditions reactive phosphorus is sequestered in different ways by ferric iron, e.g. through adsorption onto FeOOH, as Fe-P complexes, etc. Under persistent anoxic conditions the reduction of ferric iron favours Fe solubilisation with the concurrent phosphate release. The formation of the insoluble FeS and FeS<sub>2</sub> results in a drastic decrease of the P-retention capacity. Overall, iron-sulphur-phosphorus interactions may determine positive feedbacks for benthic vegetation, favouring the disappearance of phanerogams and the development of bloom forming macroalgae (see Heijs *et al.*, 2000; Rozan *et al.*, 2002 and references therein).

Nitrogen transformations within the lagoon ecosystems are primarily processed by benthic vegetation and microbial processes (Bartoli *et al.*, 2001), but are highly dependent on the element speciation (Herbert, 1999). In turn, nitrogen speciation is controlled by oxygen availability, and the different nitrogen species have different fates and follow different biogeochemical pathways. Benthic macrophytes take up and retain nitrogen at different degrees, depending on their life cycles and tissue recalcitrance. Usually, seagrass species tend to store nitrogen keeping at low levels benthic fluxes of ammonium and nitrate, as well as denitrification rates. By contrast, bloom forming seaweeds can induce wide pulses in nitrogen fluxes within the lagoon ecosystem. Benthic microalgae have a great influence on oxygen penetration and nitrogen transformations at the sediment-water interface, within a submillimetric sedimentary horizon. Here, the form in which N is returned to the water column is highly dependent upon oxygen availability.

Under oxic conditions ammonium can be oxidised to nitrate through bacterial nitrification. Part of this nitrate which diffuses to anoxic zones can be reduced to ammonium (dissimilatory nitrate reduction to ammonium-DNRA) or denitrified to gaseous N<sub>2</sub> and N<sub>2</sub>O, which are eliminated from the system (Herbert, 1999 and reference therein).

A tentative summary of essential elements and components, and associated properties and functions is reported in Table 1 (for details see Viaroli *et al.*, 2004 and references cited therein). A synthetic description of the most relevant properties depending on the different benthic vegetation types is also given in Table 2. Overall, we suggest that benthic vegetation, oxygen metabolism and coarse sedimentary biogeochemical indicators are sufficiently informative to be used for assessing environmental quality and to identify ecosystem properties and functions of coastal lagoons.

**Table 1.** Identification of essential quality elements and associated ecosystem properties and function for coastal lagoons. OM: organic matter, NP: net production rates of oxygen at saturating light (ecosystem level); DR: dark respiration rates of oxygen (ecosystem level); AVS: acid volatile sulphides (FeS and dissolved sulphides).

Quality element	Associated properties/functions
Benthic vegetation	Oxygen release, OM production, nutrient sink, sediment stability
NP versus DR	Ecosystem metabolism and stability
Sedimentary OM	Tendency of the system to consume oxygen and produce sulphides. Rates of oxygen uptake and sulphide production will depend upon OM lability
Sedimentary Reactive Iron	Buffering capacity towards sulphide and phosphate (weak)
Sedimentary AVS	Saturation degree of the buffering capacity towards sulphides
Sedimentary Carbonates	Buffering capacity towards phosphorus (strong)

**Table 2.** Ecosystem properties and functions associated to the different benthic vegetation types of coastal lagoons

	Microphytobenthos	Seaweeds	Seagrasses
<b>Oxygen</b>	Diffusion in the water mass and in the sediment	Stratification with surface rich water and bottom deficit. Pulsed availability	Oxygenation of the water mass. Radial loss by roots (ROL) in the sediment
<b>Sulphide</b>	Accumulation in the sediment	Production during biomass decomposing. Release into the water mass, dystrophy	Well buffered (depends on health status of vegetation)
<b>Nitrogen</b>	Losses due to coupled nitrification-denitrification	Pulsed assimilation and sudden release. Low denitrification	Retention by macrophyte biomass. Negligible denitrification

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## Effects of Fish Farming on Marine Ecosystems in the Mediterranean: New Tools and Prospects

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**Key words:** benthic communities, organic enrichment, disturbance, indicators.

Disturbance effects of fish farming on the seabed are well known for salmonids (Brown *et al.* 1987; Gowen & Bradbury 1987) as well as for the Mediterranean Sea bream and sea bass farming industry (Karakassis *et al.* 1998, 1999, 2000). These effects are in general similar to the model of macrobenthic succession along gradients of organic enrichment (Pearson & Rosenberg 1978). During the last 20 years, the fish farming industry in the Mediterranean has grown at an almost exponential rate facing strong local reactions by other users of the coastal zone. Three EU funded research projects namely MERAMED (Modelling environmental response to Aquaculture in the Mediterranean), AQCESS (Aquaculture and Coastal Economic and Social Sustainability) and MedVeg (Effects of nutrient release from Mediterranean fish farms on benthic vegetation in coastal ecosystems) have been carried out addressing the problem of environmental impacts of aquaculture. Results from these projects and reanalysis of data from different experiments pooled together have been used to derive quality indicators and to assess the reliability of benthic data as a means to assess environmental health of the sedimentary environment along with cost-effectiveness of the sampling protocols used. This type of data are very suitable for testing hypotheses on benthic indicators since they involve strong environmental gradients over relatively short distances thereby allowing for detection of impacts on benthic communities without the confounding effects of multiple gradients that could be present in large spatial scales.

Benthic samples were sequentially sieved through 1 and 0.5 mm sieves. The biomass fractionation ratio (biomass having passed through 1mm and retained only on 0.5 mm sieve/ total biomass > 0.5 mm) provides a simple and inexpensive method for assessing the effects of fish farming on benthic communities. The ratio was found to decrease consistently with distance from fish cages and particularly after 10m from the edge of the cages. The ratio at all the sites investigated was found to be significantly correlated with redox potential and distance. Multiple regression analysis showed that this index incorporates various components of sediment geochemistry (MD and TOC) as well as distance and feeding rates.

Sampling by means of different samplers, taxonomic resolution, and sieving size were compared in respect of information loss regarding the community structure as described through multivariate analysis as well as in respect of estimates of univariate metrics of macrofauna (Lampadariou *et al.*, submitted). The results clearly showed that information loss was relatively low as data were aggregated at higher taxonomic levels, particularly up to the level of family or even order. It was also found that the extra information gained by sieving samples through a 0.5 mm sieve did not improve the ability to distinguish the potentially impacted sites from the control stations.

Finally it was found that, a relatively large proportion of the available information concerning the community structure such as abundance, biomass or diversity is lost when sampling is carried out with corers. A cost/benefit analysis for the two sampling and the two sieving methods

showed minimal values for the van Veen samples (for both sieve fractions) at the family level, indicating that analysis at this level gives the best balance between precision of the results and decrease in taxonomic effort. However, if the time needed to sort the samples is included in the analysis, then, samples taken with corers using a 0.5 mm sieve and identified to families seems like a good compromise between precision and cost.

The overall TOC-E( $S_{10}$ ) relationship was very similar to the predictions provided by Hyland *et al.* (2005) regardless of sieve-size or sampler type. However, when samples were divided into coarse and fine sediment groups the curves obtained were substantially different among the two groups indicating that coarse sediment communities can maintain fairly high diversity even when TOC increases above 30 mg g<sup>-1</sup>.

In the framework of the AQCESS project, sampling was carried out across different communities (macro-, mega-, and ichthyofauna) at large spatial scales in search of subtle effects of fish farming zones at far fields affected by nutrient release. Although the analysis of macrofaunal samples showed no signs of environmental change (Karakassis *et al.*, 2004) it was found that large animals (such as demersal fish) with longer life spans are more likely to show response to subtle effects (Machias *et al.*, 2004; 2005). It has also been found that in oligotrophic waters fish farming wastes could have a positive effect on local fisheries.

Finally, in coarse sediments, such as those inhabited by *Posidonia oceanica*, the monitoring of benthic fauna could give a false impression of “health” showing high abundance, biomass and diversity although monitoring results regarding density of *Posidonia* meadows carried out in parallel to these benthic surveys showed severe effects on the health of the seagrass which is an important habitat for the Mediterranean.

All the above indicate that the paradigm of benthic succession described by Pearson & Rosenberg (1978) should not be used as the one and only measure of environmental health since it works well only with disturbance of macrofaunal communities in muddy sediments. Effects on coarse sediments as well as on different communities could vary considerably depending on the type of disturbance and the specific attributes of the biota.

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## Coastal Lagoons: Spatial Patterns of Benthic Assemblages and Bioindication

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**Key words:** benthic communities, labile and refractory organic matter, coastal lagoons, lagoon of Venice.

Coastal lagoons are shallow basins connected to the sea by one or more inlets. The relationships among tidal regime, dimensions of the inlets and basin morphology strongly influence water exchange with the adjacent sea, internal circulation and suspended solid transport. These characteristics make coastal lagoons a peculiar environment among marine coastal ecosystems.

Coastal lagoon are sedimentary environments especially along the microtidal and nannotidal coasts of the Mediterranean (Nichols & Boon, 1994; Barnes, 1995). As tides penetrate the lagoon, their energy is dissipated and suspended solids deposited. Lighter suspended matter sinks and accumulates at the more internal areas, forming sediments characterised by fine particles and high organic content. Often rivers flow into coastal lagoons, where particles suspended in the freshwater flocs and aggregate by salt contact generating local accumulation zones.

Across a coastal lagoon, there is a gradual departure from the marine environment toward habitats characterised by a high variability of environmental parameters and increasing levels of stress, for organisms that live there, caused mainly by organic matter accumulation and reduced water exchange.

The divergence from marine conditions are tolerated by a progressively lower number of species so that the number of species present in lagoonal assemblages typically decreases along the sea-land axis (Cognetti & Maltagliati, 2000). This spatial pattern is more evident in large microtidal coastal lagoons that are less subjected to stochastic fluctuations (Guelorget & Perthuisot, 1983; Barnes, 1994).

Many benthic indices of environmental quality include species diversity in their formulation and assign a direct relationship between diversity and environmental quality (for a review see Diaz *et al.*, 2004). If a diversity-based index must be applied to a coastal lagoon, it should take into account careful consideration of the natural spatial pattern of diversity that characterises this class of environment.

A quality diversity-based index that does not take into account the “physiological” relationships between number of species, water exchange, organic matter content and sediment texture will inevitably assign low quality values even to the secluded habitats of coastal lagoons which are in pristine condition.

During this workshop, we presented an exercise addressed to identify the relationships between diversity of macrobenthic assemblages, water exchange with the sea and organic matter in one of the largest Mediterranean coastal lagoons, the Lagoon of Venice.

A total of 140 samples (5 replicates each) were analysed, taken during May-June 2002 from sampling stations which were evenly distributed over the whole Lagoon of Venice, on subtidal shallows which is the most common habitat. As a measure of water exchange with the sea, we calculated the water residence time for every station. The measure of organic matter content was obtained by loss of weight on ignition at 350°C (LOI@350). This measure was preferred to other LOI at different temperatures because it gave the best correlation with elemental analysis of Organic Carbon conducted on 53 of the samples. Organic matter content, number of species, abundance and the diversity indices  $H'$ ,  $E(S_{10})$ ,  $E(S_{50})$ , calculated on numerical abundances, were analysed by regression methods and mapped using GIS.

Regressions showed negative relationships between species number (and diversity indices) with residence times, but gave less interpretable results with organic matter. Conversely, the visual comparison of distribution maps offered a good perception of the complementary spatial distribution of organic matter and species diversity. Numerical abundances did not show easily interpretable relationships.

Although the general scheme was applicable to the whole Lagoon of Venice, some differences emerged among the four lagoonal sub-basins. Some “anomalous” areas were identified, where species diversity was high with respect to high levels of sediment organic matter. These areas, some time ago, were vegetated marshes but now are submerged and the organic matter present in their sediments is made mostly by refractory remnants of old rhizomes. This fact suggests that not only the total sediment organic content should be taken into account, but also its degree of lability.

Many environmental quality indices are based on the diversity of macrobenthic assemblages. To apply these indices to coastal lagoons they should be in some way weighted, taking into careful consideration the “physiological” distribution patterns of diversity in such environments.

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## Use of Thermodynamic and Network Oriented Indicators for the Health Assessment of the Coastal Benthic Marine Environment

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**Key words:** thermodynamic analysis, network analysis, coastal zones, health assessment.

The application of a Thermodynamic and a Network oriented analysis on the micro and meiobenthic community at a wide number of coastal zones in the Mediterranean Sea allowed to assess different states of ecosystem health and to identify useful descriptors of the environmental quality employing a holistic approach. The analysis of micro and meiobenthic subsystem in terms of organic matter, bacteria, microphytobenthos and meiofauna reflected changes occurring in the trophic state of benthic ecosystems and provided a tool for comparison between different environments. The microbenthic loop is a major sub-system within the marine food chain and its role in affecting ecosystem function has raised increasing interest since it was first described in 1980 (Azam *et al.*, 1983). The microbenthic loop encompasses organic matter, bacteria, microphytobenthos, protozoa and meiofauna and the state of its structure and dynamics, have been recently proposed as sensitive indicators of the changes occurring in marine ecosystems (Boyd *et al.*, 2000). In particular, organic matter and bacteria are considered as powerful tools for assessing the trophic state and quality of the benthic marine environment (Vezzulli *et al.*, 2002). Temporal and spatial variability of the holistic indicators were evaluated using benthic measures collected at different times for different environments in the north western Mediterranean Sea (Ligurian Sea and south Adriatic Sea). The analyzed systems displayed great variability in respect to the organic matter load changing from extremely oligotrophic to hypertrophic conditions. Within this great variability the indicators considered in this study displayed different reciprocal behaviour let us supposing that it is by the comparative analysis of many indicators that the environmental state could be detected.

The performed Thermodynamic analysis is based on the determination of Exergy and Specific Exergy. The calculation of these indicators has been performed following Jørgensen's approach (Meyer and Jørgensen, 1979) that is based on the multiplication of the concentrations of different groups and the corrispective weighting factor based on Exergy detritus equivalent according to Marques *et al.* (2003). Exergy links the chemical energy of the various groups of the ecosystem to the information embodied in DNA. If it is necessary to consider only variations in structural complexity of the biomass Specific Exergy has to be taken in account and it is obtained by dividing the Exergy of the system for the total biomass (Fabiano *et al.*, 2004).

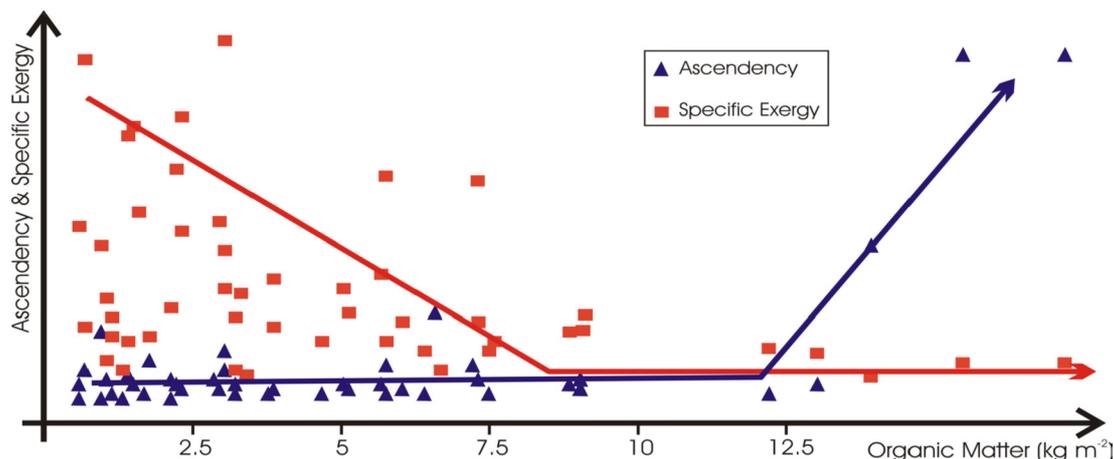
The analysis of networks of ecological trophic transfers is a useful complement to simulation modelling in the quest for understanding whole-ecosystem dynamics (Ulanowicz, 2004). In order to assess a complete network analysis of the considered systems a simulation of the micro and meiobenthic community has been performed using the dedicated software ECOPATH with ECOSIM 5.0 (<http://www.ecopath.org>). Systems that display increase in network indicators are generally considered systems in evolution with increasing functionality and efficiency in resources exploitation.

The calculation of the holistic indicators was made by using data on organic matter, bacteria, microphytobenthos and meiofauna collected from 84 stations in the southern Adriatic Sea continental shelf (Italian and Albanian coast) (Fabiano *et al.*, 2004) and 40 stations along the Ligurian coast (north-western Mediterranean) (Vassallo *et al.*, submitted). The first dataset is regarding an area that is considered oligotrophic with low human impact and anthropogenic pressure; on the other hand, along the Ligurian coast there is a wide range of human pressure with some of the considered stations placed in strongly eutrophic areas such as near the outlet of a city river or under an inshore fish farm installation.

In oligo-mesotrophic conditions Ascendency and Exergy increased at increasing Biopolymeric Carbon (BPC) concentration in the sediment. This trend is even more evident during spring when there is a strong detritus production with higher percentage of labile compounds that allow a great possibility of BPC exploitation confirming a strong and rapid benthic pelagic coupling for the micro and meio benthic community. In intermediate condition of trophic enrichment it resulted that both structure (i.e. Exergy) and functionality (i.e. Ascendency) of the micro and meiobenthic community increased at increasing resources availability. Nevertheless the micro and meiobenthic subsystem displayed the highest efficiency in resources exploitation when the resources availability was low displaying the highest values of biological complexity (Specific Exergy) and ability in exploiting and cycling resources detected as Finn Cycling Index (FCI) values. On the contrary these latter indices displayed a decrease in areas characterised by a stronger anthropogenic impact (i.e. near main urban centres) probably depending upon the origin and biochemical composition of the organic matter. This is intriguing and will need future investigations since specific Exergy and FCI might be considered as candidate indices for the assessment of health when the ecosystem is far below the hypertrophic conditions.

On the other hand if systems that we can consider reaching hypertrophic conditions are considered (Ligurian Coastal zones) different behaviours are detectable. The Ascendency displayed two different trends in function of Organic Matter (OM) load to the benthos: below  $10 \text{ kg m}^{-2}$  of OM we did not observe a significant increase in Ascendency values, while over this level we observed a rapid increase. This behaviour is related to the increase in the system activity due to an increase in the number and magnitude of fluxes that are directed to and from the detritus group. The Ex values increased with increasing OM concentrations confirming strongly the dependence of Ex on total biomass and in particular for these systems on detritus when calculated for the micro and meiobenthic subsystem. In contrast, specific Exergy showed an opposite trend and displayed a rapid decrease with increasing OM reaching values close to 1 for OM concentrations greater than  $10 \text{ kg m}^{-2}$ . Specific Exergy of 1 is only obtained in systems characterised by the absence of living biomass where only organic detritus is present. Therefore, the value of specific Exergy close to 1 observed in our study indicated a low structural complexity of the benthic system due to a progressive depletion of the living biomass at increasing OM concentrations.

For conditions with high trophic level Exergy resulted completely dominated by detritus contribution to the calculation and doesn't give any more significant information. If we consider on the same scale of organic matter load (Fig. 1) both Ascendency that represents the system functionality and specific Exergy that represents the system structural complexity it is clear that over a certain threshold of OM enrichment there is an uncoupling phenomenon characterised by a decrease in structural complexity (Specific Exergy) followed by a sudden increase in the functional component and activity (Ascendency) that could identify the buffer capacity of the microbenthic loop subsystem in tolerating organic matter enrichment.

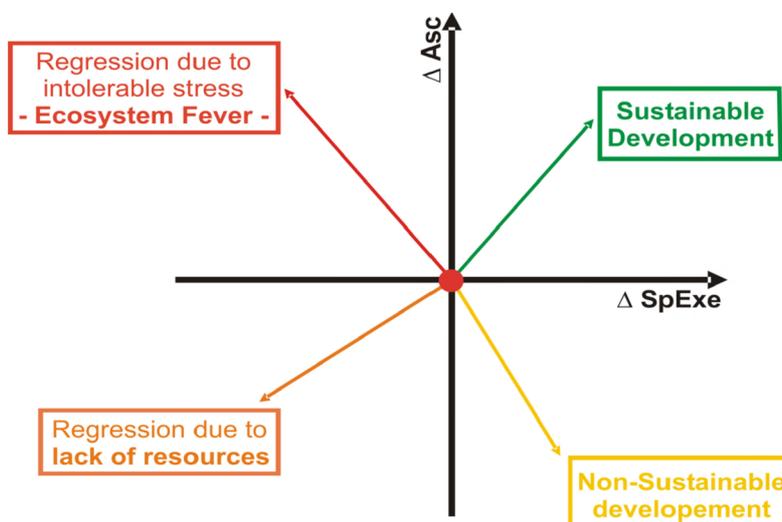


**Figure 1.** Ascendancy and Specific Exergy trend across the threshold of hypertrophy.

In conclusion, regarding the thermodynamic and network oriented analysis of benthic marine environment it is possible to summarize that the holistic approach allows a general point of view in assessing changes in structure and functionality of benthic marine environment; the comparison between different indicators permits a better comprehension of the reacting strategy of the considered system and that thus it is by the comparative analysis of more than one indicator that we can draw the best information on the health of the system. Following these conclusions a schematic approach for the health assessment of the benthic marine environment could be presented. It is based on the comparative analysis of variations of two holistic indicators that better represent variations in benthic environment: Ascendancy and Specific Exergy (Fig. 2). If in a spatial or temporal pattern we can observe a variation in the system that leads to an increase both in specific Exergy and in Ascendancy then we can reasonably suppose that the system is developing in the right way increasing its structure, consequently its complexity and functionality to support the development. Otherwise if only Exergy increases but Ascendancy decreases probably the system is just developing with a little delay in functionality increase or it is going toward a non-sustainable development where biomass and complexity are not supported by functionality increase.

Obviously if both the indicators are subjected to a decrease the system is going backwards probably due to a lack of resources. Finally if we can observe a strong increase in functionality and a contemporaneous decrease in complexity then we are attending a regression due to an intolerable stress that bring the system to try to remove the source of disturbance by increasing its activity but not using this activity to build new complex structure but dissipating energy and matter. That is what we call *ecosystem fever*.

**Figure 2.** Health assessment schematic approach for benthic coastal marine systems



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## **The Use of Benthic Elements in the European Water Framework Directive for Coastal Waters**

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**Key words:** water framework directive, ecological status, biological elements, intercalibration.

The new EC Water Framework Directive (Directive 2000/60/EC), which came into force in December 2000, requires the Member States of the European Union to establish ecological quality objectives and environmental quality standards for all surface waters including coastal waters. The overall purpose of the Directive is to achieve a “good water status” for all water body types by the year 2015, through a series of key actions that have to be undertaken by Member States to support the implementation of the Directive. This implies the identification of typologies, the description of reference conditions and the development of classification schemes through the knowledge of geomorphological, hydromorphological, physico-chemical and biological characteristics of water bodies. For this purpose, the European Commission agreed a “Common Implementation Strategy” (CIS), with the aim of developing common methodologies and approaches in the different countries, for a shared, uniform and coherent application of the Directive. A series of working groups (WGs) and joint activities, where technical experts and regulators from European Union Member States, Norway and some Accession States participated, were launched in summer 2001 to develop and test non-legally binding guidances. By the end of 2002 the “non-legally binding” guidance documents were approved by the EU Water Directors, for circulation and testing within the Member States, as the basis for carrying forward ongoing implementation work (Vincent *et al.*, 2002; Casazza *et al.*, 2003a, b; Silvestri *et al.*, 2003).

An important point resulting from the overall work of WGs was to consider some aspects and specifications of the Directive application, at the ecoregional level. Maritime areas were divided into three basic Ecoregions: Atlantic/North Sea; Baltic Sea; Mediterranean Sea.

The initial steps for the application of the Directive need to be developed using coherent and uniform technical bases, for all the different Member States and all water categories: definition of Typologies; identification of Reference Conditions; and development of Classification Systems.

The definition of typology lies in characterizing water bodies, based on the description of their geomorphological and hydrodynamic characteristics, and dividing them into specific and significant types. The aim is to produce as simple a physical typology as possible that is both ecologically relevant and practical to implement. The purpose of typology is to enable type specific reference conditions to be established. These then become the anchor for classification systems.

Reference conditions must represent the biological conditions (through the description of quality elements) of the system in its “best” ecological status, that is with the minor disturbance from human activities, to which assign the “high status”.

Such a reference is essential for the subsequent classification of the water body. The main problem in defining the reference conditions, in most of the European coastal regions, arises from the absence of areas without any anthropogenic impact. Nevertheless Member States agreed to base the choice of reference conditions on existing situations in each ecoregion, on the “best” available

ecological conditions. Moreover another problem, shared at EU level, is the lack of biological and chemical data in areas in a high environmental status, since historically marine monitoring programs have mainly concentrated on environmentally critical areas. In the case of the Mediterranean ecoregion, possible reference areas could be marine reserves and/or Marine Protected Areas (Silvestri *et al.*, 2003).

Classification must be accomplished through the analysis of different quality elements: biological, hydromorphological, chemical and physico-chemical. The Directive states clearly a hierarchy in the use of the different quality elements: biological elements (phytoplankton, aquatic flora, benthic invertebrate fauna) are fundamental while hydromorphological and physico-chemical elements are supportive parameters in the definition of the ecological status. Definitions of 5 ecological status classes (high, good, moderate, poor, bad) are reported in the Directive under "Normative definitions" (WFD Directive, Annex 5, 1.2); they are general indications on the different level of "alteration" from the reference conditions for each quality element. The Directive does not provide further specifications for classification: Member States have to establish their own appropriate classification systems. Common agreement on the conditions for the different status classes and above all the identification of the border between the "high" and "good" status and "good" and "moderate" status will have to be defined by intercalibration. The intercalibration exercise, that is the comparison at ecoregion level, of quality elements data available in Member States, in the selected sites (intercalibration network), is one of the normative requirements of the Water Framework Directive in its initial application. The intention here is to emphasise the role of such an exercise in the application of the Directive: the intercalibration must be used as a tool of adjustment, in the first phases of the implementation process, at EU level. The expected results concern: a) acquiring information on biological (and physico-chemical) quality elements data availability in the different Member States; b) verifying the coastal typology definitions and adequate assignment of the different sites to these; c) testing, where applied, the classification systems used by Member States.

The relevance of the biological elements for the assessment of coastal water quality represents one of the main innovative concepts introduced by the Directive. This legal requirement has highlighted serious gaps in the knowledge of biological communities structure and their ecosystems, in coastal waters of most EU countries: in fact monitoring programmes have historically been focused mostly on the analysis of chemico-physical parameters. As previously described (Vincent *et al.*, 2002), at the moment there are few suggested classification systems for coastal waters in the different European countries and anyhow none of these comply with the requirements of the Directive, both for the quality elements used and for the classification. For the Mediterranean ecoregion few systems have been proposed so far and, at the moment, they are being tested.

For the first biological quality element requested: phytoplankton, no classification schemes have been developed, the only proposed evaluation system (Vincent *et al.*, 2002) is the one used by IFREMER in the national monitoring programme REPHY ([www.ifremer.fr/envlit/surveillance/rephy.htm](http://www.ifremer.fr/envlit/surveillance/rephy.htm)), based on the presence of toxic phytoplankton species and algal blooms. This system is used on the French coastline, both on the north Atlantic and on the Mediterranean. Recently it has also been applied experimentally to the Basque coast (Borja *et al.*, 2004).

In regard to phytobenthos, an ecological evaluation approach based on different macrophyte species (both macroalgae and angiosperms) assigned to functional groups has been proposed by Orfanidis *et al.* (2001). Benthic plant communities of rocky substrates have been analysed for environmental quality evaluation along the Catalan coastline (Agencia Catalana de l'Aigua and CSIC 2002a, b).

For angiosperms on the basis of widely distributed, both spatially and temporally, research, studies and monitoring on *Posidonia oceanica*, some scientists from Euro-Mediterranean countries have recently formed (on a voluntary basis) a “Posidonia group” with the intention of evaluating the use of existing data and information to develop a possible quality classification of Posidonia meadows.

For benthic macroinvertebrates two possible classification methods have been proposed: the Biotic Index by Borja *et al.* (2000) and the Bentix by Simboura & Zenetos (2002).

The most relevant conclusions for the application of the WFD to coastal waters can be summarized as follows:

- 1) at present no reference networks of high status sites for coastal waters are known to exist within all Europe. To date most of monitoring programmes were focused on polluted areas rather than areas that would meet the Directive definition of high status. Data are not always available for all quality elements. Therefore, there is a need to start collecting data as soon as possible for the purposes of deriving biological reference conditions.
- 2) There is a lack of appropriate classification tools, to accomplish the requirements of the Directive but the existing classification tools may be suitable for testing by Member States. It is particularly important that information and knowledge, gained in some countries, are diffused and available tools need to be tested in other Member States, firstly within the same Ecoregion.
- 3) The intercalibration exercise is an important tool that should help to develop and test the correct application of the initial steps of the Directive, in particular for the interpretation of the normative definitions of the quality classes.

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## **Benthic Indicators: Criteria for Evaluating Scientific and Management Effectiveness**

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Case studies of the effects of human activities around the United Kingdom coastline were employed in order to evaluate a range of benthic indicators against scientific and management criteria governing their effectiveness. The number of species and the Shannon-Weiner diversity index produced the best overall performance, identifying impacts on the benthic macrofauna at all sites in the vicinity of known anthropogenic disturbance. Other indices were generally less effective even though some may have greater intrinsic potential to explain the causes as well as the occurrence of changes. In practice, all indices were dependent on the existence of adequate supporting environmental data and sound sampling design for effective interpretation. There is a need for versatility in the use of indicators of biological change, in order to compensate for the effects of local variability in natural and anthropogenic sources of disturbance. The adopted approach to performance evaluation appears to have practical value in meeting pressing environmental management needs and, with further refinement, may be suitable for wider application.

## **Assessing Environmental Quality In Benthic Ecosystems: the Role of Community Structure Models**

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Monitoring and assessing environmental quality is becoming a common task in marine biology and benthic communities are certainly among the most sensitive indicators. Their role has been clearly pointed out in many papers, as well as in the Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy (Water Framework Directive).

In spite of the emphasis on the role that communities play, however, no widely accepted methods are available as far as marine ecosystems are concerned, whereas several procedures (e.g. indices of biotic integrity) are available for freshwater ecosystems. In this framework, we will discuss the role of different approaches, ranging from very simple indicators to more complex procedures (e.g. models of community structure or comparisons with respect to reference sites).

In particular, we will focus on an application involving a community-based procedure that relies upon a very simple unsupervised modelling procedure. Our approach is aimed at identifying recurrent structures in benthic marine communities, as a preliminary step towards the assessment of the overall environmental quality of coastal ecosystems.

The large database which supported our work was collected by retrieving data from recent and mostly homogeneous sources consisting of lists of macrozoobenthic species and their abundances. We have been able to find such data for about 2200 sampling sites so far, but only in 673 sites the information about grain size and other sedimentological parameters was also available. This environmental information played a key role in determining the expected community structure, which was then compared to the observed one to obtain an estimate of the environmental quality.

In this framework we briefly discussed the role that methods based on Machine Learning techniques can play. Finally, the importance of the selection of a proper metrics for measuring the distance between observed and reference (or modelled) community structures were pointed out.

## **An Overview of the Adriatic Sea Modelling System and of the Adriatic Sea Ecosystem Modelling Developed within the Mediterranean Forecasting System Project**

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A modelling system for the Adriatic Sea has been built within the framework of the Mediterranean Forecasting system Pilot Project. The modeling system consists in a hierarchy of three numerical models (whole Mediterranean Sea, whole Adriatic Sea, Northern Adriatic Basin) coupled among each other by simple one-way, off-line nesting techniques, to downscale the larger scale flow field to highly resolved coastal scale fields. Numerical simulations have been carried out, under climatological surface forcing. Simulations were aimed to assess the effectiveness of the nesting techniques and the skill of the system to reproduce known features of the Adriatic Sea circulation phenomenology (main circulation features, dense water formation, flow at the Otranto Strait and coastal circulation characteristics over the northern Adriatic shelf), in view of the pre-operational use of the modeling system. The modeling system setup is described, and the simulation results for the whole Adriatic Sea and its northern basin are discussed in comparison with the observed climatological circulation characteristics. Unidimensional ecological modelling for the northern Adriatic Sea has been also carried out in view of the three dimensional implementation of the ecological model.