



MARBENA

**Electronic conference on
'Genetic Biodiversity in Marine
Ecosystems: Measurement,
Understanding and Management'**

6 to 17 October 2003

Summary of discussions

An activity of:

The European
Marine Research
Stations Network
(MARS)



Organized by:

The Marine
Biological
Association (MBA)



Marine
Environment
Research Centre
(ENEA)



Centre for
Estuarine and
Marine Ecology
(CEME)



Flanders
Marine Institute
(VLIZ)



Supported by the European Commission under the Fifth Framework Programme and contributing to the implementation of the key action 'Sustainable Marine Ecosystems' within 'Energy, Environment and Sustainable Development'



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The topics and issues raised in the electronic conference have been presented to the EPBRS meeting “Genetic Biodiversity in Natural and Agricultural Systems” held on 20-24 November 2003 in Florence, under the Italian EU presidency. The powerpoint presentation is available at <http://www.vliz.be/marbena/fifthsummaries.htm>. This volume includes introductions and summaries of discussions. All the discussion points raised during the conference can be reviewed by registering onto <http://www.vliz.be/marbena>

This publication should be cited as follows:

Hiscock, K.; Vanden Berghe, E.; Appeltans, W.; van Avesaath, P.H.; Hummel, H.; Heip, C.H.R.; Mees, J., eds (2003). Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003. Flanders Marine Institute (VLIZ): Oostende, Belgium. vi, 55 pp.

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**General introduction
to the MARBENA project**

* General introduction to the MARBENA project

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Context

Ten years ago, in 1992, the Earth Summit was held in Rio de Janeiro. Rio produced the Convention on Biological Diversity that has now been signed by nearly all European countries and the European Union. Since 1992 many initiatives for research on biodiversity issues have been launched, the majority of them local, short term and terrestrial. Marine biodiversity research was long considered less urgent because the main problems were thought to occur on land. Long-term biodiversity research, i.e. for more than 3 years, is very difficult to implement, even at the national level. Some of the major obstacles are the national and European funding systems and also the lack of an internationally agreed methodology for the measurement of marine biodiversity and the choice of indicators for biodiversity.

In 1994, the European Network of Marine Stations (MARS, <http://www.marsnetwork.org>), a non-profit foundation incorporated in the Netherlands, was founded to cope with these obstacles. In 2000, the MARS-related initiative BIOMARE (Implementation and Networking of large-scale long-term Marine Biodiversity research in Europe, <http://www.biomareweb.org>), started. This concerted action, supported by the Fifth Framework Programme, aims at achieving a European consensus on the selection and implementation of a network of reference sites as the basis for long-term and large-scale marine biodiversity research in Europe, internationally agreed standardized and normalized measures and indicators for biodiversity, and facilities for capacity building, dissemination and networking of marine biodiversity research. Twenty-one institutes co-operate in the concerted action.

The BIOMARE concerted action is an important first step and will provide a framework for the implementation of marine biodiversity research on spatial and temporal scales that cannot be covered by traditional funding schemes. The next steps are of course the research itself and the subsequent transfer of its results to society. The rapidly growing interest in biodiversity, with Rio +10 (the Johannesburg UN meeting) and the next framework programme approaching, require a directed effort from the scientific community. What is needed as well is a broadening of the discussion to a wider range of subjects and to a wider audience by not only including more scientists of other disciplines (e.g. terrestrial biodiversity and biogeochemistry), but science managers and end users as well.

To define the issues at stake an electronic conference on marine biodiversity in Europe (M@RBLE, <http://www.vliz.be/marble>) was organized in October 2001. The objectives of the M@RBLE e-conference were to discuss the bottlenecks and their solutions in producing relevant knowledge and the implementation of this knowledge in policy, management and conservation;

* Please refer to this section as:

Heip, C.H.R.; van Avesaath, P.H. (2003). General introduction to the MARBENA project. Pp 2-6 in Hiscock, K. *et al.* (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

therefore contributing to the development of a network for (marine) biodiversity research in Europe. The results of the e-conference were presented at the meeting of the European Platform for Biodiversity Research Strategy EPBRS in Brussels, December 2-4 2001, and published as Vanden Berghe, E.; van Avesaath, P.H.; Heip, C.H.R.; Mees, J. (2001): Electronic conference on MARine biodiversity in Europe (M@rble): summary of discussions, 8-26 October 2001. Flanders Marine Institute (VLIZ): Oostende, Belgium. iii, 43 pp.

We believe that the present efforts, BIOMARE and M@RBLE, are an important start. However, more will be needed to support development and application of marine biodiversity research over a sufficient period of time to make the field mature and active on a truly European scale. The discussion on the issues at stake should not stop with the presentation of the results at one single meeting. Instead, the discussion should become a continuous process for at least as long as the EPBRS meetings are held, so that each EPBRS meeting receives a specific input from the field responding to the specific topic of that meeting. Starting from BIOMARE - that will produce a recommendation for a network of flagship and reference sites and a review of indicators - and M@RBLE - that produced through the e-conference and the link to EPBRS the first appearance of marine biodiversity on the EU policy scene - the next series of activities should be used to create a lasting network for marine biodiversity research in Europe. Such a network must adequately prepare and exploit the possibilities of the next framework programme and the European Research Area, must improve the infrastructure for marine (biodiversity) research and its accessibility and utilization by European scientists, and must increase the visibility of marine biodiversity issues for science managers, politicians and other end users, including the public at large.

Objectives of MARBENA

The objectives of the MARBENA project are:

- § To create the infrastructure for marine biodiversity research in Europe by creating a pan-European network of marine scientists, with strong links to the different stakeholders in Marine Biodiversity Issues, from the EU-EEA and the Newly Associated Nations, and that covers the European seas from the Arctic to the Atlantic, the Mediterranean and the Black Sea. This network must improve the science by cataloguing the existing expertise and infrastructure, by defining and prioritizing the issues at stake in terms of scientific knowledge, technological requirements and application to societal problems. It must provide an intellectually attractive environment for young scientists and a discussion forum for all. It must promote the European presence and the organization of international research programmes, and promote the discussion of their results and their application. It must provide the links between scientists and industrial companies willing to aid in technological development, between scientists and science managers and politicians and lead to better integration of research and a better insight in the 'market' of supply and demand of marine biodiversity information.

- § To create awareness on the issues at stake and enlarge the visibility of marine biodiversity research in Europe, the network must make the issues – the scientific questions and the relevance of the outcome of the scientific research – clear to a non-scientific audience, it must communicate with EU policy makers and politicians (presentation of marine biodiversity issues at the European Platform for Biodiversity Research Strategy meetings, presentation to the European Commission and European Parliament when requested), with global organizations and programmes such as several IGBP programmes (GLOBEC, LOICZ, perhaps SOLAS), DIVERSITAS and the Census of Marine Life initiative, national and other EU biodiversity platforms (e.g. the BioPlatform thematic network) and dissemination of information to the public at large.

Hereby, the project contributes to the European Research Area (ERA) initiative. Special effort will be undertaken to involve the stakeholders from the Newly Associated States (NAS) in the network.

For more information on the project and for the partners involved see <http://www.vliz.be/marbena>.

Overview of planned project activities

To achieve these objectives, MARBENA performs the following main activities:

A. To create a long-term infrastructure for marine biodiversity research

1. To develop a European Marine Biodiversity Network.

- § MARBENA will start by using existing information (e.g. the ESF and Diversitas Science Plan and the results from BIOMARE and M@RBLE amongst others) and by cooperating with existing European organizations, including the European Marine Research Stations Network MARS that through its member institutes has already played an active role in the development of marine biodiversity science.
- § MARBENA will open its activities and actively engage cooperation with any interested partner, including museums of natural history, universities and government laboratories.
- § MARBENA will establish a structural link with the BioPlatform.
- § One of the most important tasks will be integration with scientists of the Newly Associated States and a sufficient coverage of the marine areas at the periphery: the Arctic Sea, the Black Sea and – when possible – the Southern Mediterranean Sea.

2. To build a long term research infrastructure for the network MARBENA will provide the information and mechanisms for creating a solid basis on which the network can build:

- § By discussing research priorities and their implementation and coordination for the next five (or even ten) years and the ways of financing European-level research where needed, taking advantage of the new possibilities of the 6th framework programme and the European Research Area e.g. through complementation of national research.
- § By describing the market of ‘supply and demand’ of marine biodiversity information: who are the stakeholders and what is the information available and needed? Where are the gaps and what can we do about them?
- § By describing and publishing a catalogue of the research infrastructure existing and required (vessels, instrumentation, experimental facilities) and of taxonomic literature (floras and faunas, keys for identification), studying their accessibility to European researchers and prioritizing their development where necessary.
- § By promoting regional cooperation between different EEA and NAS countries focusing on regional problems and involving the regional end-users.
- § By promoting the possibilities for discussion between scientists, management and policy makers.

B. To create visibility for marine biodiversity issues in Europe

To enlarge the visibility of the marine biodiversity issues and therefore marine biodiversity research in Europe, MARBENA will work on publicizing these issues with the stakeholders and the public. This will be done by maintaining an active web site, by regular press releases, and by the publication of a newsletter, CD-ROM's and folders. MARBENA will link to other programmes of interest (DIVERSITAS, relevant IGBP-programmes, Census of Marine Life CoML etc.), to EU policy makers requiring information and support for implementation of e.g. the Water and Habitat

Directives, the European Environment Agency and to the ESF Marine Board as a representative of the national funding agencies.

1. To develop and maintain a web site where information and issues produced by the Marine Biodiversity Network will be easily accessible to stakeholders involved in marine biodiversity as well as the public at large. The website will be the main communication structure for the network of marine biodiversity stakeholder. The web site will have links to the MARS Web Site and to other web sites (BioPlatform, ESF Marine Board, EU Directorate of Research)

2. To organize Electronic conferences on selected themes

- § To provide relevant information on the Marine Biodiversity issue for use in the meetings of the “European Platform for Biodiversity Research Strategy” (EPBRS) connected with the EU presidencies. For this a close cooperation will be established with BioPlatform.
- § To discuss issues important for the establishment and maintenance of the Marine Biodiversity Network and the long term infrastructure for marine biodiversity research and the communication between researchers, management and policy makers.

3. To organize workshops, conferences and case studies

MARBENA will organize together with other partners a series of workshops on selected topics, discussion of four case studies on selected priority issues for four regions in Europe involving scientists, policy makers, industry and the public (including the press) and a major conference to finalize the project and create the conditions for the future existence of the network.

C. Involving the Newly Associated States

In this project special effort will be undertaken to include the scientists and through them the other stakeholders of the marine biodiversity research from the Newly Associated States in the network. For this we propose the concept of MARBENA Ambassadors, well known and respected scientists who are residents of the NAS, who will actively extend the network in these countries. Furthermore the 'Ambassadors' will discuss relevant biodiversity issues at the Electronic conferences.

The MARBENA electronic conferences

The MARBENA-project will organize a series of at least five e-conferences on selected themes.

Electronic conferences will be held before each of four European Platform for Biodiversity Research Strategy (EPBRS - see the BioPlatform website at <http://www.bioplatform.info>) meetings with the following objectives:

- § To raise a dialogue on the themes selected for the EPBRS meetings, involving a wide range of participants. These themes will be determined ad hoc in relation to the EU Minister Conference.
- § To prepare for the EPBRS meetings through this dialogue, involving both the scientific community and policy makers, specifically:
 - To identify current understanding on the selected themes.
 - To identify areas of uncertainty ('biodiversity information needs') on the selected themes.
 - To make provisional recommendations on research ('biodiversity research needs') on the selected themes for subsequent discussion at the EPBRS meetings.

- To provide background papers for the Platform meetings summarizing current understanding, areas of uncertainty and recommendations on research on the selected themes.

E-conference chairs are coupled with the EU presidency and organization of the EPBRS meeting: in order to reach participants from the nations that host the coming EPBRS meetings (Spain, Denmark, Greece, Italy), the chair of the respective e-conferences is conveyed to a scientist resident of these countries.

The fifth of these conferences ran for two weeks, from 6 to 17 October 2003. The topics and issues raised in the electronic conference have been presented to the EPBRS meeting “Genetic Biodiversity in Natural and Agricultural Systems” held on 20-24 November 2003 in Florence, under the Italian EU presidency. The powerpoint presentation is available at <http://www.vliz.be/marbena/fifthsummaries.htm>. This volume includes introductions and summaries of discussions. All the discussion points raised during the conference can be reviewed by registering onto <http://www.vliz.be/marbena>.

One additional e-conference on “The Future of Marine Biodiversity Research in Europe” will be organized independently of the platform meetings.

**Report of the conference on ‘Genetic
Biodiversity in Marine Ecosystems –
Measurement, Understanding and
Management’**

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*Welcome and introduction

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The Marine Biological Association (MBA) is pleased to organize this electronic conference together with the Flanders Marine Data and Information Centre as a part of the MARBENA series. The topics identified for this e-conference are targeted at informing the physical conference on “Genetic Biodiversity in Natural and Agricultural Ecosystems” being organized for 20-24 November 2003 and to be held in Florence. We have therefore, after discussion with colleagues in ENEA, the Marine Environment Research Centre, La Spezia, Italy (especially Dr Federica Pannacciulli) and the European Platform for Biodiversity Research Strategy (especially Professor Dr Isabel Sousa Pinto), therefore emphasized the impact of human activities on genetic biodiversity in the marine environment.

In recent years, powerful tools have become available to the marine science community to describe within-species diversity and address how human activities may be affecting genetic diversity in the seas. MBA staff undertake work on a wide range of studies that involve molecular genetic approaches. These include understanding marine viral diversity and how viruses affect marine phytoplankton populations; work on mating systems that generate patterns of biodiversity and understanding impacts of human activities on marine biodiversity via the EUMAR project of the EU 5th Framework. In conjunction with the Sir Alister Hardy Foundation for Ocean Science, genetic population structure of zooplankton is being investigated, and the MBA is also undertaking phylogenetic studies of intertidal organisms.

Whilst the sea yields most of its protein through harvesting of wild stocks, aquaculture increasingly supplies fin and shellfish as well as limited seaweed resources. Aquaculture is as close as we get in the marine environment to agricultural production. Aquaculture operations can lead to transplant (deliberate or otherwise) of individuals from elsewhere within the range of a species leading to change in spatial genetic population structure and the risk of hybridization. There are major concerns about escapees from fish farms contaminating local gene pools.

Understanding genetic biodiversity is also essential to understanding and managing natural marine ecosystems. Marine microbial ecology is now hugely dependent on molecular methods of assaying diversity. Genetic methods can tell us how connected or fragmented populations are and give insights into dispersal ranges – essential knowledge to inform siting and spacing of marine protected areas. Human impacts can also shape genetic biodiversity via selection: pollution-tolerant strains can arise at contaminated sites. Harvesting can reduce effective population size to the extent that only a few broadcasting individual contribute to the next generation.

We wish you all a productive and stimulating exchange of views.

* Please refer to this section as:

Hawkins, S.J.; Hiscock, K. (2003). Welcome and introduction. Pp 9 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

*Introduction to topic one “Measuring, managing and conserving genetic resources in natural marine systems” (I)

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Marine biodiversity is threatened by overexploitation, physical alteration, pollution, global atmospheric change and introduction of alien species. There is, therefore, a need to acquire knowledge about patterns and processes of marine biodiversity, in order to establish effective management plans. It is only by considering genetic diversity, too often neglected by stakeholders, and the processes that rule biological diversity, that a given plan will have long-term success.

In recent decades the introduction of more and more refined molecular and statistical techniques and the flourishing development of genetic labs all over the world has allowed the production of a great bulk of genetic data on a large number of marine taxa. Investigations have been mainly aimed at studying spatial (much less frequently temporal) genetic variability, measure within- and among-population genetic diversity, and estimating phylogeographic/phylogenetic relationships among populations/taxa. Available techniques allow the collection of information on several issues, such as gene flow, mixing of different gene pools, effective population size, inbreeding rate, genetic loss, the occurrence of recent bottlenecks, assignment of individuals to a population, the action of natural selection, gene introgression and hybridisation, taxonomic position. A large part of these data refer to “funding-attractive” species, such as commercial and endangered species, but a lot of work has also been carried out on many other taxa. Data gathered could be used to provide invaluable suggestions for the management and conservation of biological marine resources.

Connectivity and species’ genetic structuring

Connectivity has been defined by Palumbi (2003) as “the extent to which populations in different parts of a species’ range are linked by exchange of larvae, recruits, juveniles, or adults”. More and more studies are finding a number of exceptions to the equation “species with pelagic larvae” = “high dispersal” = “absence of genetic structuring”, reporting relatively high genetic differentiation even in marine species with potentially high dispersal. These results have been explained in terms of chemical-physical “barriers”, such as salinity or pollution gradients, presence of local eddies, gyres, current reversal, substrate heterogeneity, as well as biological factors, such as larval behaviour, selection on recruits, species interaction, etc. (e.g. Lee et al. 1994, Jones et al. 1999, Swearer et al. 1999, Luttikhuisen et al. 2003, Taylor & Hellberg 2003). From a structural point of view, the heterogeneity of marine systems determines moderate to strong genetic structuring. On the other hand, from an evolutionary perspective, different processes occur and determine population divergence and, ultimately, speciation.

* Please refer to this section as:

Maltagliati, F. (2003). Introduction to topic one “Measuring managing and conserving genetic resources in natural marine systems” (1). Pp 10-12 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

One of the problems of major conservation concern is habitat fragmentation mediated by human activities. This may provoke genetic loss in natural populations due to alteration or, ultimately, interruption of gene flow among the fragmented subpopulations and an enhanced inbreeding rate, as a consequence of critical reduction of the effective population size.

Furthermore, the marine domain includes habitats, such as estuaries, underwater caves, hydrothermal vents, deep sea trenches, which are easily spatially identifiable and which present peculiar abiotic and biotic characteristics. For instance, I have had experience with Mediterranean brackish-water environments, such as estuaries, lagoons, coastal ponds and canals, that are characterised by natural fragmentation, which is reflected in the genetic structure of most of the species living there. Common genetic characteristics of brackish-water species can be drawn from literature, namely low levels of within population genetic variability (bivalves seem to be an exception), high genetic divergence among populations due to isolation, and presence of a number of invertebrate sibling species (reviewed in Cognetti & Maltagliati 2000, Bilton et al. 2002).

Historical factors

Genetic patterns that we describe today are the result of contemporary and historical factors. In particular, dramatic changes determined by Quaternary events have been well documented in the terrestrial habitats of northern temperate regions (e.g. Hewitt 2000). Much less is known about the effects of Pleistocene periodical climate changes on marine coastal organisms. Reasonably, the marine coastal habitats affected by ice cover during the glaciations should have been affected by periodical extinction-recolonisation events. In the temperate regions, the marine areas never covered by ice have been affected by wide changes in sea level (-120/130 meters during the last ice age). Hence, marine coastal species should have faced ecological upsets due to "migration" of coastlines (see Graham et al. 2003). The understanding of the effects of Pleistocene climate changes on marine organisms of temperate regions could provide important assistance in making predictions on the effects that current rapid climate change driven by human activities will have on marine populations. Of course, this issue covers all hierarchical levels of biodiversity, but the contribution of historical factors in shaping patterns of marine genetic diversity could be addressed in this e-conference.

Genetic Monitoring (GM)

GM could be defined as an array of methodologies to assess the response of populations to environmental stress. From an applied perspective, it could also be meant as a tool to verify the effectiveness of a given environmental management plan at the lower levels of biodiversity (genes, individuals, populations). GM is aimed at determining reductions of within-population genetic variability (genetic loss) and alterations of among-population genetic diversity. The effects of environmental stress, pollution, or habitat fragmentation on natural populations are, for instance, problems that could be addressed with GM (e.g. see Maltagliati 2002). Rarely, environmental monitoring of the biological component focuses on population level. Ecotoxicological assays focus on cellular or individual responses to a given stress, whereas the analysis of the modifications of community structure following, for instance, the release of a given pollutant, addresses the problem at a level higher than population. The scarcity of methodologies focusing on population is a gap that scientists should fill, because this level is of great ecological importance, population being the unit of adaptation to a given environment.

This e-conference offers me the possibility to open a discussion on GM in order to define and apply effective methods of GM to marine environments. Furthermore, results of GM could be integrated with data on variation of abiotic environmental factors, as well as results from other disciplines, such as ecotoxicology, population dynamics and community structure. In addition, the

development of comparable and reliable statistical parameters representing several disciplines, including genetics, would be preferable.

Bearing in mind that most marine biodiversity is still unknown, I invite you to address some general questions on the points raised above, although, of course, many others may be raised by contributors.

- § What is the value of changes of marine genetic diversity at both within- and among-population levels?
- § Do historical factors matter, in order to predict the effect of climate change on genetic diversity of marine organisms?
- § Can available data on marine genetic diversity contribute to the planning of adequate environmental management, at least at a strictly local scale?
- § How should scientists working on marine genetic diversity direct their research to protect, conserve and manage natural marine resources?
- § Are there concrete possibilities to standardise methods of genetic monitoring and include them among the procedures of marine environmental monitoring?

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*Introduction to topic one “Measuring, managing and conserving genetic resources in natural marine systems” (II)

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As also mentioned by Ferruccio Maltagliati, most of marine biodiversity at ‘species’ and, even more, at intraspecific levels is still unknown. Obviously this is a severe handicap if marine (genetic) biodiversity is to be protected and managed. This problem is further exacerbated by the still quite pervasive belief that biodiversity in marine ecosystems is generally much less vulnerable to extinction caused by anthropogenic influences than biodiversity in terrestrial ecosystems. Yet compelling evidence indicates (1) that marine ecosystems are undergoing rapid and radical degradations (as suggested by symptoms such as coral bleaching, marine epidemics, algal blooms, invasive species, mass mortalities, collapsing fisheries, etc.) (Lubchenco et al. 2003), (2) that marine species may be at far greater risk of extinction than is generally assumed (Roberts & Hawkins 1998), and (3) that currently there is a continuous loss of local marine biodiversity due to local population declines and extinctions (e.g. Carlton 1993). Needless to say that if these latter two points hold at the species level, they definitely also apply to intraspecific genetic biodiversity. Unfortunately, irrespective of the system level considered, losses of marine biodiversity still remain largely unmeasured (McKinney 1998). This stands in sharp contrast to the large body of data documenting biodiversity losses in terrestrial ecosystems. It is therefore no surprise that marine conservation has received much less attention than the conservation of terrestrial biodiversity. A short scan of current conservation journals suffices to appreciate this point. Hence, no wonder that marine conservation has much less of a theoretical basis than terrestrial conservation (Allison et al. 1998) and therefore it has been tempting to implement experiences from the latter to the marine realm. Yet, in many aspects marine ecosystems differ fundamentally from terrestrial ones and thus require their own specific conceptual framework for conservation purposes (Carr et al. 2003). Fortunately, several recent major publications and research initiatives, some of which funded by the EU, are exactly aiming at providing such conceptual scientific framework for the conservation of marine biodiversity. A number of these publications and initiatives may be of particular relevance for participants of the present MARBENA electronic conference (the list is far from exhaustive !):

- § ‘The Science of Marine Reserves’ : a set of 17 papers, totalling 228 pages, aimed at providing a multidisciplinary approach to establishing marine reserves. The collection was published in February 2003 as a Supplement to ‘Ecological Applications 13(1)’ and is available as PDF at : <http://www.esajournals.org/esaonline/?request=get-static&name=s1051-0761-013-01-0001>
- § EU Fifth Framework research project ‘EUMAR’ (European Marine Genetic Biodiversity), a consortium of seven partner institutes from five countries, aiming at understanding the dynamics of marine genetic biodiversity in the European intertidal. The project combines

* Please refer to this section as:

Backeljau, T. (2003). Introduction to topic one “Measuring managing and conserving genetic resources in natural marine systems” (2). Pp 13-17 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

population genetic data (using a wide variety of molecular and quantitative genetic techniques) with demographic data to model the impacts of (anthropogenic) environmental change on marine intertidal genetic diversity in a number of key-species. More information can be found at : (<http://www.eumar.tmbi.gu.se/>)

- § EU Marine Biodiversity Cluster, an informal network of 16 EU-funded projects involving 166 partner institutes and organisations dealing with marine biodiversity research. More information is available at : <http://www.cordis.lu/eesd/ka3/cluster6.htm>
- § EU Sixth Framework Network of Excellence on Marine Genomics, a network involving 44 partner institutes in 16 countries ; the project will commence in January 2004 and more details about its aims and structure can be found in a PDF presentation available at : http://www.eurofaire.prd.fr/bibliotheque/pdf/ji080903/NoEMarine_Genomics.pdf
- § CORONA network (Coordination of Research On the Northern Atlantic), a US NSF-funded platform currently involving 118 researchers from 13 countries, promoting interdisciplinary research on the evolution of biota in the northern Atlantic Ocean, with particular emphasis on studies dealing with the effects of climatic perturbations on an ampho-Atlantic scale. The platform is open to any interested party and particularly welcomes links and input from European researchers. More information is available at : <http://www.biology.duke.edu/corona/>

The central idea underpinning the management and conservation of marine biodiversity is that one should understand the patterns and processes that govern the dynamics (i.e. origin, maintenance and loss) of marine biological variation. This in turn requires the consideration of at least five issues (Backeljau et al. 2001; paper available as PDF download at <http://www.vliz.be/marbena/fifthsummaries.htm>):

- § How do we measure and monitor biological diversity ?
- § Which part of this diversity is due to genetic factors (i.e. is evolutionarily relevant) ?
- § How is this genetic diversity structured in space and time ?
- § Which processes are at the base of these spatio-temporal patterns ?
- § What kind of operational concepts can be implemented to conserve and manage marine biodiversity ?

At first glance these issues appear trivial, yet a closer look at current practice and research focus shows that some issues need further indepth reflection. I would like to propose some of these for discussion here.

How to measure genetic biodiversity ? (cf. issues 1+2)

Currently the vast majority of marine genetic biodiversity studies involve analyses of supposedly 'neutral' (or nearly neutral) molecular markers to infer population genetic structuring, taxonomic differentiation and phylogenetic relationships. The power and relevance of this approach is beyond question. However, these markers usually have little to do with the genetic variation responsible for complex morphological, behavioural, and life-history traits that underly the (adaptive) responses and the functioning of organisms in changing environments (i.e. ecologically relevant traits). Disentangling the contribution of genetic and environmental effects in the overall phenotypic variety of such complex 'ecologically relevant' traits is the field of quantitative genetics, a field that appears to be far too much neglected in the conservation of (marine) genetic biodiversity (Storfer 1996). Unfortunately the relationship between neutral molecular variability and the (phenotypic) diversity of ecologically relevant quantitative genetic traits is still poorly understood.

Moreover in cases where both types of genetic markers have been compared, they usually provided a quite different picture of population structuring and differentiation. This leads to several issues for discussion, such as :

What are the merits, relevance and relative importance of quantitative genetic data for the measurement, management and conservation of genetic biodiversity in a time where genomics seems to be the central dogma ?

How should quantitative genetic data be combined or integrated with (neutral) molecular data (e.g. molecular and quantitative genetic data may respond differently to bottlenecks ; F_{st} and Q_{st} may show quite different pattern ; how to deal with reaction norms and adaptive phenotypic plasticity in terms of genetic biodiversity)?

How should consistently different, ecologically related phenotypes, between which current molecular data are unable to detect significant differentiation, be treated when assessing biodiversity? (e.g. different shell form of the periwinkle *Littorina tenebrosa* linked to lagoonal conditions, even though molecular markers do not reveal any consistent and significant differentiation from 'normal' *L. saxatilis*)

Are we not overemphasizing the ('neutral') molecules given that ecological separation (sensu lato) may be as, or even more, important than geographical separation in the evolution of species (e.g. Orr & Smith 1998 ; Morell 1999 ; Johannesson 2003)?

To what extent should epigenetic phenomena be considered as a relevant component of genetic biodiversity, given that these involve heritable variation without changing the order of nucleotides in the underlying DNA sequences ? (for some provocative ideas on this issue see Jablonka & Marion 1999)

How to account for temporal patterns and dynamics ? (cf. issue 3 and Ferruccio's point ii)

Evolution is essentially the change of allele frequencies (except in case of epigenetic phenomena !). Hence the time dimension in which allele frequencies vary appears fundamental when assessing effects of (anthropogenic) environmental change on genetic biodiversity. Yet :

How important and relevant really is this time dimension when interpreting and forecasting the dynamics of marine biodiversity ?

How accurately can impacts of past events on population genetic diversity be estimated and how precisely can impacts of current events be forecasted over time ?

How useful and reliable are 'time series' analyses based on historical collections without clearly defined reference points in time (e.g. what may be learned from comparisons between two, or more, sample points in time separated by many years and accordingly by several possible impact events whose individual and combined effects may hence become difficult, if not impossible, to distinguish) ?

Which operational concepts ? (cf. issue 5)

Usually biodiversity is measured in terms of taxonomic diversity. Unless a purely (phenetic) morphospecies concept is used, this approach entails also a measurement of genetic biodiversity. However, restricting the measurement of biodiversity to taxonomic levels, is by definition bound to obscure intraspecific genetic variation, even if it is precisely this latter that provides the "fuel for

evolution” (i.e. selection and drift can only act if there is variation). Hence restricting the management of marine genetic biodiversity to the “species level” will almost certainly result in important losses of evolutionarily relevant intraspecific variation. In recognition of this issue two operational concepts have been defined, i.e. Evolutionarily Significant Units (ESUs) [= historically isolated sets of populations that together encompass the evolutionary diversity of a taxon] and Management Units (MUs) [= sets of populations that are currently demographically independent] (Moritz 1994, 1996). In practice these operational concepts are defined on the basis of nuclear and mitochondrial molecular markers, such that ESUs should be reciprocally monophyletic for mtDNA haplotypes and at the same time differ significantly for allele frequencies at nuclear loci, whereas MUs only imply significant differences in allele frequencies, without considering a phylogenetic component. Hence ESUs are more inclusive than MUs. However the application of these operational concepts in conservation and management may be not free of critiques, e.g.

- § Given that ESUs and MUs refer to population inferences based on supposedly neutral markers, how well do they reflect “active” and ecologically relevant genetic diversity upon which selection is acting?
- § How adequate are these concept when dealing with phenotypic diversity for which (neutral) molecular markers do not provide a significant pattern of differentiation (see above the example of *Littorina neglecta*)?
- § Is it desirable and feasible to extend ESUs and MUs to include quantitative genetic components (including adaptive phenotypic plasticity), and if so how to do it?
- § To what extent are DNA-barcodes (e.g. Hebert et al. 2002) applied to e.g. species, ESUs or MUs sufficient ‘operational descriptors’ or ‘identifiers’ of (intraspecific) genetic diversity in view of genetic polymorphisms and quantitative genetic variation?

Epilogue

Many of the issues proposed above essentially deal with the main question whether the current scientific emphasis on ‘genomics’ is sufficient to describe, understand and even forecast the dynamics of (marine) genetic biodiversity in response to (anthropogenic) change. Perhaps this question can be used as some sort of “fils rouge” during the present MARBENA conference.

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*Introduction to topic two “Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Fisheries” (I)

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The marine realm is a theatre in which some very large scale selection experiments are being played out, with consequences that are ill-understood and potentially serious for the future health of our marine resources. The actors in this play are: (1) the fishery managers who set the rules by which fish are selected, (2) the fishers who apply the selective mortality, and (3) the fish stocks that undergo genetic change due to selective fishing. The precautionary principle places a responsibility on us to leave our resources in a state that can be utilised as fully by our descendants as by ourselves; if we continue on our present path, our descendants are not going thank us.

Here are some assertions about selective effects of fishing. They are intended to be provocative, to stimulate response from you over the next few days.

All fishing is selective. Most obviously the market attaches different values to fish of different sizes, and fishers try to catch sizes that give the greatest return. In addition fishery managers try to regulate fishing through technical measures such as net mesh sizes. The trait most evidently under selection is size-at-age. But there are many others, including size and age at sexual maturation, potentially important for productivity of fisheries.

The strength of selection is large. Selective fishing would be of little consequence if fishing mortality rates were small relative to other causes of death. However, the evidence is that most fisheries are fully exploited or over-exploited (FAO, 1998); fishing mortality rates are often two or three times greater than all other sources of mortality put together. The strength of selection on body size is large enough to be detectable within single year classes of North Atlantic cod, as the fish grow and selective mortality takes place (Sinclair *et al.*, 2002).

There is genetic variation in the traits under selection. It is not realistic to argue that genetic variation in the traits under selection is negligible. Over the last 20 years, the aquaculture industry has shown that it is quite feasible to bring about genetic change in growth and maturation of fish in captivity; this can only happen if these traits contain genetic variation. The heritabilities are in the range expected of traits closely related to fitness (Mousseau and Roff, 1987), even when estimated directly in the wild (Jónasson, Gjerde and Gjedrem, 1997).

Measurable changes in the traits are observable at least on a time scale of decades. Large phenotypic changes are taking place in major commercial fish stocks. These changes can, of course, be due to factors other than genetic change driven by selective exploitation. Nonetheless, the changes are often consistent in direction with evolution, and remain even when proximal

* Please refer to this section as:

Law, R. (2003). Introduction to topic two “Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Fisheries” (1). Pp 18-20 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

factors such as food supply and temperature are factored out (e.g. Rijnsdorp, 1993; Heino, Dieckmann and Gødo, 2003).

The traits that change under fishery-induced selection are important for the sustained productivity of exploited fish stocks. Most startling are the results of a laboratory selection experiment on the Atlantic silverside (Conover and Munch, 2000). Starting from the same stock, they culled either small individuals or large individuals from replicate populations; within four generations, the yield from the former was nearly twice that from the latter. Clearly we are not dealing with trivial effects on productivity caused by evolution.

Fisheries management does not consider the effect of fishery regulations on the genetic state of fish stocks. In fisheries institutes around the world, I am not aware of any systematic thought being given to these matters of selection and genetic change when proposals for regulation of fisheries are given to governments.

We need some radical rethinking about fisheries management. Typically regulations make use of minimum net mesh sizes, to ensure a period of growth before fish are caught. Other things being equal, this selects for slow growth, deleterious for productivity in the long term as the stock evolves. Concentrating mortality on large fish is also not necessarily sensible for management in the short term; if the minimum net mesh is set to catch fish before they reach maturity and if fishing mortality is large, the spawning stock biomass becomes seriously depleted, as in the case of North Sea cod. Arguably we need to put in place regulations that ensure low fishing mortality rates on large fish. Building up the spawning stock biomass is beneficial in the short term, and might also provide a selective advantage for fish carrying genes for faster growth, giving a benefit in the long term. These are preliminary ideas: we have much to learn about managing the genetic resources of our exploited fish stocks.

Of course, there is much missing in the brief comments above. Not least is the complete loss of gene pools when over-exploited fish stocks go to extinction. I hope you will point out things that I have missed and that you think are important. But overall, I think the case for a new—Darwinian—fisheries science is strong one. Right or wrong; do post your comments, ideas and results here over the next few days.

Specific questions about selective fishing to guide your thoughts (relevant comments outside these questions are also welcome):

- § How selective is fishing in reality? What methods of fishing are intrinsically more or less selective?
- § What traits are under selection? How strong is such selection?
- § How much additive genetic variation exists under natural conditions for traits under selection?
- § What evidence is there that fish stocks are undergoing genetic change as a result of fishing-induced selection? On what time scale does such evolution take place?
- § Does it matter? What, if anything, should managers of fisheries be doing about evolution caused by fishing?

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***Introduction to topic two “Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Aquaculture” (II)**

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Aquaculture production has been growing at 7.4% per annum in Europe for the past 10 years, over 55% of this being produced in the marine environment. Aquaculture production in Europe is expected to continue to grow at this rate and is likely to exceed 2.2 million tonnes by 2010 (New, 2001).

Today the genetics status of aquatic organisms used in aquaculture can be seen as continuum from effectively wild stocks on one hand to domesticated and genetically improved on the other. The wild stocks will include species that are captured in the wild and are ongrown for sale, such as Tuna and eels; offspring generated from wild broodstock which are stripped and the eggs hatched and ongrown in culture (eg. *Channos*, *Penaeid spp.*), and ranched species such as mussels and salmonids. There is still a debate as to whether any artificial propagation or disruption of normal reproductive patterns has a totally neutral effect on the genetic status of any species (Campton *et al.*, 1995). In species in which the whole life cycle has been closed so the broodstock come from farmed fish, the genetic effects can be marked. In Atlantic salmon, highly domesticated and genetically improved strains are being used and are under continuous development (Gjedrem *et al.*, 1991). In other species such as the seabass (*Dicentrarchus labrax*) and the gilthead seabream (*Sparus auratus*), which are at an earlier stage of development, the genetic status of the stock will be determined by the level and quality of the management of the stock to this point. Stock based on small founder populations that have suffered a ‘bottleneck’ will be prone to the effects of inbreeding and loss of variation through genetic drift. The world-wide trend is that more species are now being cultured, particularly tropical marine fish and shellfish. The time taken to develop a new farmed species is reducing as farmers build on the experiences gained from more established species. The difficulties in developing a sustainable farming system for any marine species should not be underestimated, as there are still major gaps in our understanding of the reproductive biology, nutritional requirements of larval stages, husbandry requirements and disease and health problems. Genetic management of newly farmed stock has normally been given a low priority until the industry has become established.

Genetic changes in captive populations

Fish farmers want to grow stocks that are tolerant of the artificial environment and efficient converters of the food they supply into high quality flesh. The availability of genetically improved fish strains is limited to a few salmonid species so most farmed marine species still effectively have wild genotypes. Population genetic studies show that there are genetically differentiated sub-populations of marine species, although the levels of differentiation (F_{st}) between populations of marine species are generally lower compared to freshwater fish. Information on population

* Please refer to this section as:

McAndrew, B. (2003). Introduction to topic two “Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Aquaculture” (2). Pp 21-25 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

structure is important as it helps to identify genetically isolated stocks that might display differences in commercial performance. Performance testing of genetically isolated stocks under farmed conditions has preceded a number of selective improvement programmes, differences in performance between wild populations being as much as 60% (Eknath *et al.*, ?).

Whether single or multiple populations are collected, the construction of the hatchery stock will have profound effects on subsequent genetic performance. Initially all broodstock will be collected from the wild either as juveniles or adults. Depending on how this is done and how many fish survive to breed a 'founder effect' will determine how close this population reflects the genetic potential of the wild stock. Hatchery populations often remain open for a number of years as new broodstock are introduced while the reproductive performance of the farmed fish improves or the offspring from the original parents starts to mature. Once farmed fish are available hatchery managers will try to use these as it greatly lowers costs, reduces the risk of importing disease and they believe they have some adaptation to the farm environment. The moment that hatchery populations become closed then there is a need to develop a genetically based broodstock replacement policy to avoid the genetic degradation of the stock. In farmed stocks without any genetic management the two main problems genetic drift and inbreeding are inversely proportional to the effective population size (N_e) of the hatchery stock. In randomly mated stocks, almost exclusively used in aquaculture, the larger the N_e the lower the risk of related mating and the loss of alleles through drift (Tave, 1993). Minimum N_e values have been suggested for randomly mated stocks by various sources (FAO, ?), around 50 for aquaculture stocks and several 100's for stocks used for natural enhancement. In many cases farmers do not practice even the basic management of N_e . The loss of performance caused by differing levels of inbreeding on commercial stocks of trout has been analysed by Kincaid (1983) and shows that sib-mating can reduce the yield of traits by an average of 25-30%.

Selective improvement of farmed animals and plants has resulted in an enormous increase in the productivity of these crops (Simms *et al.*, 1997). At this time, very little of the world's aquaculture production comes from improved strains. It is only recently that breeding programmes for Atlantic salmon have been implemented at an individual production company level. Aquatic organisms offer many challenges for those looking to improve their stock, not least the identification and pedigreeing of individuals, a prerequisite for a sustainable breeding programme. Aquatic organisms also have many attributes that can increase the rate of genetic gain not available to breeders of warm-blooded livestock. These include high levels of genetic variation, high fecundity and external fertilization; traits that enable breeders to construct complex breeding designs, yet maintain large family sizes, generating high selection differentials that can result in rapid rates of improvement per generation (Gjedrem, ?). The plastic phenotype of fish allied with the reproductive advantages mean that techniques such as chromosome set manipulation and sex-reversal can be used to generate unique genotypes as well as single sex and sterile populations.

Individual identification of most aquatic organisms to a single family is very difficult because physical tagging can only occur when the individual is large enough to be tagged; this requires on-growing in individual family units, widely used in salmonids, but for mass spawning marine species this is usually impractical. Genetic fingerprinting technologies, based on microsatellite loci, are now routinely used as a tool for parentage assignment in the management of farmed stocks. This genetic technology and the improvements in physical tagging that came with the use of electronically coded Passive Integrated Transponder tags (PIT tags) now forms the basis for a number of novel selective improvement programmes. I have personal knowledge of programmes for halibut, seabass, seabream and oysters. Such breeding programmes manage inbreeding and should result in long-term and sustainable improvement of these species for a number of commercial traits.

“Green fingered” genetic improvement using mass selection based on an individual’s phenotype has been widely practiced on fish farms, but rarely with the care needed to minimize non genetic sources of variation in the population being selected. Without this control the rate of genetic gain is often slow or non-existent. With good management but no assessment of genetic relatedness of selected fish the farmer runs the risk that they are restricted to a small number of families and so increases the risk of inbreeding. This problem is likely to be exacerbated in highly fecund marine species such as halibut, which requires the farm to maintain relatively few large individuals to generate egg production.

Although breeding programmes have not been widely adopted, genetic interventions that improve the performance of the farmed fish have been widely used. These include hybridisation between species in an attempt to obtain hybrid vigor or better performance characteristics. Single sex populations are widely used, usually the later maturing sex to avoid problems of maturation during the production cycle. Single sex fish can be produced either directly using hormones or indirectly by genetic techniques. The production of sterile individuals, usually triploid females, has also been widely used to avoid problems such as early maturation of fish, genetic interactions with native fish of the same species or exotic species establishing breeding populations. More recently transgenesis has been used to improve the growth characteristics of North American farmed Atlantic salmon (still experimental).

Main points for discussion are:

- § Generalizations on the genetic status of farmed stocks are impossible and will be determined by the stage of development and levels of genetic management used by the individual farm or company. There are advantages in assessing different stocks/populations to assess levels of variation present for commercially important traits. Genetic change is inevitable in hatchery reared commercial strains. Unintentional changes caused by poor management results in genetic drift and inbreeding with negative effects on the strain. Intentional change driven by scientifically based broodstock management or selective improvement programmes can result in real genetic gains.
- § Molecular markers are essential tools in the management of farmed stock used in assessment of levels of differentiation between populations or strains, assignment of offspring to parents, traceability of fish in production and market and confirming provenance of improved strains.
- § Genetic interventions are possible to generate single sex or sterile fish that might help to reduce the genetic impact of farm escapes. Research suggests that techniques being used can reduce the ability of these fish to resist stress and are therefore not favoured by farmers.

Genetic implications for wild stocks

Most of the large-scale aquaculture production or on-growing in the sea is carried out in net cages or in coastal lagoons and ponds. These systems are prone to accidental and malicious damage that can result in large numbers of fish escaping into the environment. Shore based pumped systems are more secure but are expensive to maintain and are usually restricted to broodstock and hatchery activities. Farming of a given fish species is often undertaken in the same environment as the native stock so there is the possibility of ecological and genetic interactions between farmed escapes and native fish. The potential problems of escapes from salmon farms in Europe, North America and Chile have been well-documented (Fleming *et al.*, 2003). Problems of competition between farmed and native fish for space, food and mates and possible transfer of diseases and parasites have all been cited. Atlantic salmon farming is now almost totally based on the use of non-indigenous

(originally Norwegian) and genetically improved strains so that any escapes breeding with native stock may disrupt local adaptation and homogenise any differentiation over large areas of the species range.

The problems of escapes of farmed marine species are likely to be different to those of salmonids. Species or population ranges are going to be very large, the size of the native breeding population is likely to be much larger. Even if there is some impact the difficulties in actually monitoring the level of escapes and any genetic change in the sea rather than individual rivers are likely to be great.

Patterns of differentiation also appear to be species specific and can be influenced by past and present hydrological patterns and the spawning behaviour of the species. Two important farmed species that overlap for most of their native range in Europe, seabass and gilthead bream have very different population structures. The bass has a high level of population differentiation over quite small geographic distances and shows different adaptations to marine and lagoon environments within the Mediterranean. The gilthead seabream appears to be effectively a single panmictic population over most of the Mediterranean. These findings are not intuitive considering the bass is a large migratory predator that should be able to disperse over great distances but appears to home to natal spawning areas (Ben, ?). There has already been large-scale movement of seabass from the Atlantic to the Mediterranean to supply broodstock for hatcheries. Escapes of these two species from a single farm (they are often grown in adjacent cages) would have very different implications for the respective native stocks.

At the present stage of development of most marine species the mass spawning techniques, high mortality and sensitivity of the larval stages mean that genetic interventions such as chromosome set manipulation, single sex and sterile populations have not been well developed. Problems of skewed sex-ratios in seabass and early maturation in seabass, halibut, turbot and cod compared to wild stocks suggest that culture conditions are inducing these problems. This will stimulate research into the development of single sex or sterile production systems that would also help to reduce the risk of genetic introgression between farmed and native fish. However, as genetic management is being incorporated at ever earlier stages in the development of new species then breeding programmes that selectively breed for late maturity might prove more efficient in the long-term.

Farmers tend to develop species with a high market price (e.g. halibut, grouper, marine shrimp) in order to recoup development costs more rapidly. The demand for these fish has probably reduced the population size of the native stock by overfishing. Demand for wild caught broodstock to supply hatcheries can further significantly increase the value of these fish and accelerate the reduction in population size. However, the continual decline in wild fisheries means that once abundant and cheap fish such as cod and haddock are now being actively developed as farmed species.

Main points

- § Marine fish farming using existing infrastructure will result in large and eventually widespread escapes of farmed fish.
- § Levels of escapes may be large in relation to the size and health of the native species.
- § Although the number of escapees may be estimated it may be difficult to assess the level of impact on the native population.
- § Restrictions on the use of only indigenous populations may help to reduce the impact of escapes but this is already too late for some species such as seabass and may be unnecessary for other species.

- § Farmers may resist any legislation to impose such restrictions, particularly if the characteristics of the local population put them at a commercial disadvantage.
- § Breeding companies that have developed improved strains would wish to supply fish to the largest possible market.
- § Genetic interventions that might reduce the risks of interactions between farmed and native fish are difficult to implement in mass spawning species and may also compromise farm performance.
- § Development of new species using broodstock from overexploited wild stock may put added pressure on these stocks because of the high value of such fish to the industry.

Please do communicate your views on any of the above points and more to the e-conference.

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* Introduction to topic three “ Effects of human activities in altering genetic characteristics and gene flow in the marine environment” (I)

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Besides the natural processes of expansion and contraction of the area colonized by a species over evolutionary times, sudden species-range expansions -often through human-mediated transport- have been observed at an increasing rate since the end of the 19th century. Since the original work by Elton published in 1958 (Levine & D'Antonio 1999; Williamson 1996), biological invasions have received a growing set of attention from the scientific community. Several international and national research programmes have been—and continue to be—carried out (e.g. SCOPE, GISP, (Mooney 1999); CIEM ITMO 'Introductions and Transfers of Marine Organisms) and more recently, a new scientific journal entirely devoted to the issues of biological invasions was founded in 1999 (“*Biological Invasions*”).

Tightly linked to human activities, biological invasions —besides catastrophic and chronic pollutions, new agricultural practices and overfishing—are one of the major threats for marine biodiversity and ecosystem stability (Carlton 2000; Occhipinti-Ambrogi & Savini 2003; Walker & Kendrick, 1998). One of the most devastating impacts of human activity on marine environments, particularly in coastal environments, is the introduction of foreign species, which may severely alter biodiversity of marine ecosystem. Although one of the main properties of biological invasions is the unpredictable nature of their appearance, invasion corridors are highly correlated to commercial shipping routes (Ricciardi & MacIsaac, 2000). For instance, shipping between USA and Great Britain may have caused the introduction of 20 % of the foreign species in British waters. Foreign species may also be capable of taking advantage of coastal water pollution (e.g., the introduced alga *Ulva fascicata* has supplanted the autochthonous species *Ulva pertusa* in polluted environments in Japan; (Morand & Briand, 1996)) or climate change (Stachowicz *et al.*, 2002).

The rate of marine biological invasions is ever increasing since the beginning of the 20th century (Gouletquer *et al.*, 2002; Reise *et al.*, 1999). Around 80 foreign species have been recently recorded on the coasts of the North Sea (Reise *et al.*, 1999; Wolff, 1999) and 140 along the Atlantic coast of Europe (Gouletquer *et al.*, 2002). The increasing interest in invasive species is not only due to the increasing rate of introduction of new species (due to an increasing trade volume and exchanges), but also to the fact that they may abruptly modify the interactions among species and affect the stability of the whole ecosystem (Levine & D'Antonio, 1999; Ruiz *et al.*, 1997; Simberloff & Von Holle, 1999). For example, non-native species may change the social behaviour of indigenous species, hybridize with the native species or induce extinction in the native fauna/flora. Introduced species have also major economical impact (Everett, 2000; Robinson, 1999) and have an effect on public health.

* Please refer to this section as:

Viard, F. (2003). Introduction to topic three “ Effects of human activities in altering genetic characteristics and gene flow in the marine environment” (1). Pp 26-30 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

Numerous conceptual frameworks and methodological outlines have been developed for the study of patterns and processes linked to the introduction of non-native species. These questions may be classified into three main research themes: (1) investigation of the history of the invasion processes and subsequent dispersal (e.g. date(s) and location(s) of the introduction(s); sources and vectors; pathways of colonization); (2) studies of the biological impact of the invaders and determination of the conditions that favor new species to colonize new territories (e.g. ecological consequences for the native community, invasion dynamics, life-history traits which may favor a foreign species); (3) predictions and control of the invasion process. Until recently, most of the work and much of the literature on invasion have dealt with ecology and population dynamics modelling. More recently, the evolutionary impact and the genetics of alien species has been emphasized (Grosholz, 2002; Lee, 2002).

Population genetics attempts to depict the historical and contemporary population structure and, in the longer term, to understand the processes resulting in evolutionary changes. Several authors have underlined the usefulness and lack of molecular data for the study of biological invasions (Holland, 2000; Lee & Bell, 1999; Ricciardi & MacIsaac, 2000). Until recently, invasive species represented a challenge for genetic analyses because non-equilibrium populations, genetically impoverished, often characterize such species. Some of the basic assumptions of classical population genetics model are thus violated (as, for instance, the relationships between F_{st} and Nm : Whitlock & McCauley, 1999). Genetic studies of invasive species should therefore go beyond the usual measurements of genetic differentiation to assess dispersal. Recent molecular tools (e.g. microsatellites, nuclear introns) as well as theoretical developments now provide the opportunity to address major issues as the origin of newly founded populations, the rate and pathways at which dispersal occurs. Assignment tests, based on multilocus genetic data and maximum likelihood calculations, are among the most promising statistical procedures (for a review see (Davies *et al.*, 1999)). Those tests provide a means of assigning individuals to particular sources and of assessing population structure as well as interpopulation dispersal (Pritchard *et al.*, 2000). Besides assignment tests, maximum likelihood calculations have been recently developed to take into account realistic assumptions about the migration patterns among local populations (Tufto *et al.*, 1998) or to test from recent expansion or bottleneck (Kimmel *et al.*, 1998; Luikart *et al.*, 1998; Saccheri *et al.*, 1999). Those theoretical developments attempt to elucidate contemporary gene flow and dispersal without relying on unrealistic migration model or assumptions of equilibrium that are clearly violated in many recently founded invading populations. As indicated by Ferriere *et al.* (2000), new theoretical frameworks and tests are available or under development to combine direct and indirect data to determine dispersal parameters (probabilities and distances). At a larger scale, the patterns and processes governing the geographical distributions of genealogical lineages at the intra-specific level may be analysed by phylogeography methods (Avice, 2000). Also, evolutionary forces responsible for the distribution of the genetic variability within a species are critical to understand and/or predict the evolution of populations. By determining the way genes are transmitted over generations, mating systems is one of the forces that plays a significant role in the distribution of genetic diversity within and between populations. Studies and estimation of mating systems parameters such as selfing rate, paternity, mate choice and level of inbreeding may be done using population genetics approaches. Finally, secondary contact of previously geographically isolated but not largely divergent taxa may lead to hybridizations and introgression events between non-native and native. This might promote important changes in the genetic architecture of native species (for example through allopolyploidy promoting reproductive isolation between native populations).

In non-native marine species, genetic studies have been recently used for:

- § to identify cryptogenic species or cryptic introduction events (e.g. *Carcinus*, (Geller *et al.*, 1997)) tracing the origin of the introduction and the dispersal routes of invaders and to test

hypotheses about founder events and/or recurrent introductions (e.g. *Crepidula fornicata* (Dupont *et al.*, 2003; Holland, 2001))

- § to assess dispersal strategies (gradual (stepwise) *versus* jump (long-distance and stochastic) dispersal), e.g. *Dreissena bugensis* (Wilson *et al.*, 1999))
- § to investigate the occurrence or the patterns of hybridization with native species (e.g. (Baumel *et al.*, 2001))

However, the data are still too scarce to carry out “meta-analyses” of these data and get comprehensive pictures, in order, for example, to determine if any life-history traits are common to successful non-native species. Also, whereas patterns of introduction and dispersal can be well described (with an appropriate sampling and using appropriate tools and models), the genetics and evolutionary processes are not so well investigated. In terrestrial animals, linkages between a reduced genetic diversity and behavioural changes (ex. ants *Linepithema humile*, (Tsutsui *et al.*, 2000)) or shift in mating system (Daehler, 1998) have been demonstrated. A further step, in marine non-native species, is to depict and understand the rates of change in the genetics of non-native species and associated life-history traits, the importance of natural selection in shaping the genetic architecture of genetically impoverished populations, the degree of hybridisation between native and non-native species (and thus the role played by exotic species in modifying the genetic architecture of native species).

In that context, many issues (about methods and data) can be addressed. Among them:

- § What are the limiting factors or the methods still to develop in order to investigate evolutionary genetics of non-native marine species?
- § Do evolutionary genetics studies elucidate traits that promote marine alien success?
- § How can ecological and evolutionary genetics approaches be correlated? Could theoretical modelling help?
- § Are there any peculiar genetic characteristics associated with marine invaders? What could be the strategy (model species, time and spatial scale to consider etc.) to develop?
- § How often are the introductions of non-native species associated with ‘founder events’ (reduced genetic variation)?
- § What are the expected/observed rate of changes in the genetic architecture and life-history traits of exotic species?
- § To which extent is the genetic diversity and structure of native-species impacted because of introduction of non-native species?

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***Introduction to topic three “ Effects of human activities in altering genetic characteristics and gene flow in the marine environment” (II)**

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During the two previous sessions of the Marbena e-conference on marine genetic diversity, some of the issues related to human alteration of gene flow and connectivity in marine environment have been partially addressed. It has been highlighted that species diversity is the result of a complex equilibrium between speciation processes and extinction rates. Quantifying the impact of human activities on genetic diversity requires knowledge of evolutionary processes and collection of a large amount of information on the genetic structure and differentiation patterns in marine species.

The classical model of speciation in evolutionary ecology is allopatric speciation (Palumbi, 1994). Allopatric speciation predicts that genetic differences accumulate during long periods over many generations when a species, with a wide continuous distribution, is broken up into populations isolated by extrinsic barriers that prevent gene flow. Changes in sea level, moving of tectonic plates, etc., are considered to be foremost drivers of speciation and divergence within marine species (Jackson & Cheetham, 1999) through the creation of physical/chemical barriers among species and populations. However, marine systems are often assumed to be continuous, and marine species to be able to disperse over large geographic ranges. Following the definition of allopatric speciation, this process in marine habitats has been considered slow, because of the absence of physical barriers to gene flow. Yet, the effective long-distance dispersal ability of species has rarely been quantified by direct measurements. Most data refer to indirect estimation based on genetic markers (Neigel, 1997). The development of sophisticated biochemical and molecular genetic markers, allowing an estimation of the genetic divergence among populations, showed that many of the widely distributed species are not cosmopolitan, but represent complexes of species (Knowlton, 1993). Some of them are difficult to identify on morphological bases alone (sibling or pseudo-sibling species). Moreover, recent studies have emphasised the role of ecological factors in sympatric speciation events (Johannesson, 2001), favouring rapid divergence at small spatial scales.

The paradigm that speciation must be rare in the marine environment is also contradicted by the high numbers of closely related species that occur in many marine habitats, ranging from coral reefs to the deep sea (Gray, 2001a, 2001b). Even those taxa with large marine populations, high fecundity and larvae that can disperse long distances often are composed of a large number of closely related species (Palumbi, 1994). Habitat heterogeneity is more evident in coastal environments and continental margins where most marine ecological studies are carried out.

Nevertheless, ecological and evolutionary studies have shown evidences of population fragmentation also in the pelagic environment, related to current circulation and gyres. Coastal

* Please refer to this section as:

Abbiati, M. (2003). Introduction to topic three “ Effects of human activities in altering genetic characteristics and gene flow in the marine environment” (2). Pp 31-34 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

areas are a complex mosaic of habitats, variously interspersed and interconnected. Human activities often modify natural patterns of coastal landscapes thus altering the level of isolation and connectivity among populations. This occurs both by the creation of corridors that allow/increase species dispersal, and by changing the degree of habitat fragmentation, thus affecting evolutionary processes. In the last centuries humans have become a major force capable of changing landscapes at a variety of spatial and temporal scales (Ayensu et al, 1999). This is especially true for coastal habitats, manifested in the worldwide trend towards coastal urbanisation. Indeed, it is important to stress to coastal managers and stakeholders that speciation did not only occur in the past, but it is an ongoing process. Population genetic studies have identified species that fall in a whole range of stages predicted by the speciation processes. Divergence among populations as detected by molecular tools reflects a process of differentiation that could be abruptly altered by human intervention shifting the degree of connectivity among populations.

Human enhanced fragmentation

Human induced habitat fragmentation and related impacts at both genetic and species levels received wide attention in terrestrial habitats for predicting the consequences of urban development, the design of natural reserves and restoration plans (Newman, 2000). Evidence of human induced fragmentation in the marine environment are rare, however. Continuous coastal shorelines can be interrupted by coastal cities and harbours, long jetties extending in the sea can modify the coastal circulation and affect the larval dispersal, and contamination or habitat exploitation could also lead to extinction of local populations. Only few studies have addressed the issue of fragmentation in marine habitats from the genetic point of view, and most of them deal with marine plants (Reusch, 2003; Valero, 2003).

Natural habitats with high levels of fragmentation could be dramatically altered by human induced changes in the patterns of fragmentation. Maltagliati has already provided examples of naturally fragmented habitats, focussing on brackish water bodies. Brackish waters are among the hot spots of evolutionary processes (Cognetti & Maltagliati, 2000), and land reclamation, often involving draining of wetland areas, chemical pollution and eutrophication events, can alter the mosaics and the connectivity among brackish-water populations, thus affecting their genetic structuring to the point of extinction of locally adapted populations.

Connecting naturally fragmented habitats

Along most coast rocky shores, alternation with sandy pockets and beaches, creates a mosaic of patches of varying size. In these heterogeneous environments, species with low dispersal often show interesting evolutionary patterns. Human intervention along the shores can affect genetic connectivity among populations and species. Nourishment of gravelled beaches with marine fine sand could allow settlement of soft bottom fauna in areas where it did not occur before. Even more dramatic is the development of coastal protection schemes using breakwaters, coastal walls and groynes. These structures introduce artificial rocky bottom 'islands' in areas where only soft bottoms naturally occur, and can cover areas of hundred of kilometres of sandy shores, as has happened in the Adriatic Sea. The establishment of artificial populations changes the metapopulation structure of the species and may increase gene flow among formerly isolated populations. These would affect mainly species with relatively short larval dispersal, which could use the artificial hard bottoms to propagate along the shores, following a stepping stone model. Increased connectivity among populations would have the opposite effect to fragmentation, reducing genetic heterogeneity in space and therefore reversing allopatric speciation.

Human transfer of adult specimens or propagules could determine dramatic changes in genetic diversity of formerly isolated population. McAndrew, in the introduction to Topic 2, has already

expressed concern about the potential impact on wild populations of species used in aquaculture (e.g. transfer of seabass specimens for the Atlantic to the Mediterranean). Furthermore, ballast water and fouling have been shown to be an effective transport mechanism for marine organisms across large biogeographic regions (Carlton, 1999). Studies on ballast water mainly focussed on alien species transfer and species invasions. However, shipping routs could also be seen as virtual corridors for the gene flow. Transfer of conspecific specimens among isolated Evolutionarily Significant Units, even if less obvious compared to species invasion, could represent a relevant threat to marine genetic diversity.

Sampling designs and statistical tools

In previous discussions, Zane has raised the question of the adequacy of experimental designs applied to population genetic studies. In recent years ecology moved towards more robust and rigorous experimental approach in analysing causal relationships between species distribution and environmental impacts (Green 1979, Hurlbert 1984, Underwood 1994, Benedetti-Cecchi 2001, Quinn & Keough 2001). Studies on the genetic structure of populations using biochemical and molecular tools can provide a picture of the level of divergence/distance among populations or species, however little can be inferred about processes leading to this picture. Speculations about the mechanisms that could have lead to the observed situation are common, but the efficacy of the proposed explanation is rarely tested. The resolution of genetic studies could be enhanced by development of experimental designs that take into account both the peculiarities of the molecular ecological analyses, in terms of type of data sets and overall costs (e.g. manpower, lab time, etc.), as well as the need for adequate replication in time and space with appropriate comparison with a reference conditions is needed to enhance the resolution of the genetic studies. These steps will benefit both of basic research and the development of a potential early warning methodology to be applied in environmental studies.

Early information on proposed human interventions on coastal landscapes (e.g deployment of off-shore platforms, reclamation of wetlands, isolation of coastal bays, building of coastal defence structures), could be used as large-scale experiments were apply the BACI (Before vs After/Control vs Impact) approach (Underwood 1994) to genetic impact assessments. The comparison of genetic patterns of target species in before the alteration with those after environmental change, would allow quantification of the consequences of human activities on species metapopulation structure and evolutionary processes. Of further relevance to the experimental design is the issue of the selection of molecular markers appropriate to the proposed task. In addition, we require the identification of statistical tools and indices sensitive enough to detect minor changes in allelic frequencies and genotypic patterns, to allow an indirect estimation of the connectivity among populations (Bossart & Prowell, 1998).

Understanding impacts of human activities on the genetic structuring of marine populations poses several question that need to be addressed:

- § are the available markers providing an unbiased indirect estimation of the effective gene flow among populations?
- § can molecular ecology provide experimental evidence of human induced changes in fragmentation/connectivity in marine species?
- § how could experimental designs be implemented to be applied in analysing spatio/temporal changes in species genetic structure?
- § could population genetics provide early warning tools to be used in the assessment of impact and in conservation of marine ecosystems?

Acknowledgements

I wish to thank T. Backeljau and L. Airoidi for the useful comments on this introduction. The ideas presented have been stimulated by discussions within the working groups of EUMAR and DELOS EU funded projects.

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Summaries of the discussions

***Summary of discussions on topic 1: Measuring, managing and conserving genetic resources in natural marine systems**

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Marine biodiversity is threatened by overexploitation, physical alteration, pollution, global atmospheric change and introduction of alien species. There is, therefore, a need to acquire knowledge about patterns and processes of marine biodiversity, in order to establish effective management plans. It is only by considering genetic diversity, too often neglected by stakeholders, and the basic processes that rule biological diversity, that a given plan will have long-term success. Within this context, more-and-more refined molecular techniques allow us to obtain an important insight into natural populations by collecting information on several issues, such as gene flow, mixing of different gene pools, effective population size, inbreeding rate, genetic loss, the occurrence of recent bottlenecks, assignment of individuals to a population, the action of natural selection, gene introgression and hybridisation, taxonomic position. Data gathered can be used to provide invaluable suggestions for the management and conservation of biological marine resources.

Some main themes were addressed in the conference. The first was about species' genetic structuring, connectivity among populations and habitat fragmentation. Ferruccio Maltagliati introduced this theme providing a definition of "connectivity" and starting a discussion on the influence on dispersal of barriers and life history traits, such as the presence of pelagic larval stages. He also raised the problem of the genetic effects that habitat fragmentation may produce on marine populations.

Thierry Backeljau expressed his concerns about the application to marine environments of the concept of habitat fragmentation, developed originally for terrestrial systems. He argued that often people are tempted to study patchy populations as a model for habitat fragmentation and that, in this respect, it is important to study the population subdivision in a relatively short time span. Care should be taken when interpreting data, because habitat fragmentation is not the only phenomenon determining population subdivision.

Ferruccio Maltagliati replied by providing two examples. The first was about the disruption of one-dimensional gene flow in a hypothetical coastal species with low potential for dispersal, due to the presence of an even minor construction on the coast. The second was about fragmentation of seagrass meadows that may reasonably provoke population subdivision of organisms associated.

* Please refer to this section as:

Maltagliati, F.; Backeljau, T. (2003). Summary of discussions on topic 1: Measuring, managing and conserving genetic resources in natural marine systems. Pp 36-42 in Hiscock, K. *et al.* (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

Tim Wyatt (Instituto de Investigaciones Marinas, Vigo, Spain) posed the question on the possibility of a temporal fragmentation, namely a sort of genetic discontinuity among cohorts of exploited stocks of fish. This could be provided by 1) the capture of the pool of high-ranking males that breed successfully and contribute to the progeny during spawning and 2) the exploitation of individuals whose presence is fundamental for the maintenance of the social structure because they are responsible of the control of the shoals for migration to spawning grounds.

Thierry Backeljau stressed the importance of studies that consider the impact of exploitation on breeder males. However, he expressed doubts on the analogies between the definitions of temporal and spatial fragmentation.

Andy Beaumont (School of Ocean Sciences, University of Bangor, Anglesey, UK) offered some comments on various aspects of the discussion. Firstly, he reported the importance of considering the effective population size in aquaculture, fisheries and conservation. Secondly, he argued that molecular biology advances have been mainly in methods that look closer at the DNA, rather than in increasing theoretical understanding of population genetics. This has produced a large temporal gap between the advent of a new genetic methods and their application for ecological or conservation purposes. Thirdly, he showed that micro-organisms and haploids are not taken into account when considerations on genetic diversity and conservation applications are made. Finally, he reminded us that human impacts may also homogenise populations, as happened with the European oyster.

Sebastian Holmes (Nederlands Instituut voor Onderzoek der Zee, Den Burg, Texel, Netherlands) argued that human-mediated transport of organisms exists and that it increases the levels of within population genetic diversity. In contrast, it decreases the degree of between population genetic diversity. He also stressed that the negative effects of inbreeding are overestimated. He separated the significance of within- and between-population components of genetic diversity and provided a terrestrial example where a recovery plan of a species has been successfully performed despite inbreeding. Furthermore, he argued that, from a metapopulation perspective, well connected (globally genetically homogeneous) populations that are locally genetically heterogeneous represent an evolutionary unstable strategy, in face of environmental variability. In contrast, poorly connected populations (globally genetically heterogeneous) but locally genetically homogeneous represent an evolutionary stable strategy.

Frédérique Viard (Station Biologique de Roscoff, Roscoff Cedex, France) agreed with this vision of inbreeding. She enlarged the concept stressing that, since inbreeding is common in nature, we should not blindly try to limit it in every situation, but we should assess the effects of increased relatedness on fitness. The analysis of bottlenecks or founder effects in natural populations may represent a useful tool to make reliable predictions for increased relatedness. Didier Aurelle (Centre d'Océanologie de Marseille, Station Marine d'Endoume, Marseille, France) joined the above ideas on inbreeding and introduced the importance of adaptive genetic variation at population or species level.

Keith Hiscock (Marine Life Information Network, Marine Biological Association of the UK, Citadel Hill, Plymouth, UK) stressed the importance to conservation of recovery, following decline, of a given species at a local scale, especially if the species is a key structural, key functional, or rare. He reported the different consequences of self-recruiting populations compared to populations recruiting from distant sources. He provided some examples of non-commercial invertebrates with different life history traits and population structure from British waters. He concluded by expressing the need of genetic investigation for those species in order to give insight on their conservation prospects.

Bella Galil (Israel Oceanographic and Limnological Research Ltd., Tel-Shikmona, Haifa, Israel) provided the example of a Mediterranean endemic reef-building gastropod with very low potential for dispersal. Given the distribution and life history traits of this species, she stressed the need of population genetic studies in order to shed light on its conservation status.

Another theme of the conference introduced by Ferruccio Maltagliati was environmental management and, in particular, genetic monitoring, meant as an array of methodologies to assess the response of populations to environmental stress. Genetic monitoring should be aimed at determining genetic loss and alterations of among-population genetic diversity. The effects of environmental stress, pollution, or habitat fragmentation on natural populations are examples of problems that could be addressed with genetic monitoring.

Lydia Ignaties (National Centre of Scientific Research “Demokritos”, Aghia Paraskevi, Attiki, Greece) argued that biodiversity mostly relies on phenotypic characters of species. This leads to mistakes due to problems associated with species identification and taxonomy. She supported, however, the efforts to set up methodologies of genetic monitoring.

Piero Cossu (Dipartimento di Zoologia e Antropologia Biologica, University of Sassari, Sassari, Italy) joined the idea of the importance of genetic monitoring in environmental management. He proposed the Inter Simple Sequence Repeats (ISSRs) markers as a quick tool for the first step of the procedure of genetic monitoring.

Frédérique Viard (Station Biologique de Roscoff, Roscoff Cedex, France) expressed her ideas on the fact that the concepts of Management Unit (MU) and Evolutionary Significant Unit (ESU) should be enlarged by including kinship, neighbouring size and mating system.

Adriana Zingone (Stazione Zoologica ‘Anton Dohrn’, Naples, Italy) with her interesting experience on phytoplankton species reported that coastal waters should assume greater conservation and protection relevance. In fact, coastal waters can be considered a sort of source of genetic diversity for phytoplankton organisms and they constitute, therefore, evolutionary grounds for many species. Human impacts on coastal waters may provoke the disruption of the phytoplankton ‘genetic diversity maximiser’ with a consequent drop of genetic diversity.

Recognizing the “units” to assess, chart and measure biodiversity, is pivotal for management and conservation purposes. Yet, the discussions on this issue clearly showed that there still exist a number of controversial points, leading to lively exchanges between several participants.

Thierry Backeljau emphasised the importance of going beyond the species approach in measuring biodiversity by considering infraspecific genetic variation for it is this infraspecific variation that provides evolutionary potential to organisms. In this context he referred to the practical value of concepts such as “Evolutionarily Significant Units” (ESUs) and “Management Units” (MUs) to chart infraspecific genetic variation in relation to evolutionary history. Yet he also emphasized the importance of quantitative genetic variation, in addition to molecular data and more “classical” characters. Nevertheless currently quantitative genetic analyses are seldom included in biodiversity research or in the application of ESUs and MUs (neither of these latter two concepts accommodate for quantitative genetics).

Part of these statements provoked reactions from Ferdinando Boero (Dipartimento di Scienze e Tecnologie Biologiche ed Ambientali, Università di Lecce, Lecce, Italy), Ward Appeltans (Flanders Marine Data and Information Centre, Flanders Marine Institute, Oostende, Belgium), Marco Casu (Dipartimento di Zoologia e Antropologia Biologica, University of Sassari, Sassari, Italy), Godtfred Høpner Petersen (Zoological Museum, University of Copenhagen, Copenhagen,

Denmark), Yves Samyn (Laboratory of Ecology and Systematics, Vrije Universiteit Brussel, Brussel, Belgium), and Stefano Mariani (Department of Biological Sciences: Molecular Ecology and Fisheries Genetics Laboratory, University of Hull, Hull, UK) who emphasized the central role of species as unit to measure and chart biodiversity. Ward Appeltans, for example, stressed the need for species lists as a tool to recognize biodiversity hot spots and endemisms. Although this point was not contested, Andy Beaumont and Thierry Backeljau remarked that (1) applying species concepts to several organisms remains problematic (e.g. in asexuals) and (2) that “species lists” often are very biased in the part of biodiversity they cover. Usually they include only “larger” metazoans and plants, and often they do not consider prokaryote, protist and meiofaunal diversity, even if this may be an important component of the overall biodiversity in a given ecosystem. Moreover, these groups of organisms are poor in phenotypic characters by which the organisms can be identified. Hence their recognition and diversity assessment is by necessity largely based on molecular genetic techniques.

This latter point, together with the earlier statement of Thierry Backeljau concerning the importance of infraspecific genetic variation, opened another discussion in which concerns were expressed about the perceived controversy between “phenotypically based” and “genetically based” approaches to biodiversity (e.g. Ferdinando Boero, Ward Appeltans). Everybody agreed that both approaches need each other and have to be integrated. This requires of course a good communication between researchers that may have very different backgrounds and interests (e.g. Ferruccio Maltagliati, Yves Samyn, Ferdinando Boero, Thierry Backeljau). Some participants felt that on this point there still is a long way to go (e.g. Ferdinando Boero), while others perceived this a lesser problem given that they precisely combined molecular genetic and phenotypic approaches (e.g. Stefano Mariani, Thierry Backeljau)

In a similar way, the discussion on species concepts arrived at the general conclusion, that there is no such thing as a generally applicable species concept, even if everybody recognized the relevance of delimiting species and this latter of course requires some interpretative framework under the form of some sort of species concept. Nevertheless, Godtfred Høpner Petersen did not endorse this latter point and stated that “species are” and “exist without definition as components in different food webs and energy flows”. Most other participants, however, seemed to attach importance on the consequences and scientific, as well as practical, implications of applying one or another species concept. Ferruccio Maltagliati remarked in this context that the discussion about species concepts is an old one and that each time new analytical tools are introduced to taxonomic research, there is some sort of initial euphoria related to the hope that the newer techniques will solve the old, long-standing problems. This applied to protein electrophoresis at the end of the 1960s and to PCR combined with DNA sequencing at the end of the 1980s. In both these cases it took several years before it became clear that no technique can be considered as Pandora’s box, i.e. each technique has its shortcomings and limitations with respect to measuring genetic variation and phylogenetic relationships. Hence no technique can solve all (taxonomic and phylogenetic) questions and problems (e.g. Andy Beaumont). As such molecular techniques have added to the species concepts problem, but have certainly not solved it (perhaps they even have aggravated it).

Yet this resulted in another concern of Ferdinando Boero and Peter Mill (The School of Biology, University of Leeds, UK), viz. that currently students, university curricula and funding agencies show little interest in “classical” (i.e. phenotypically based) taxonomic and ecological work. As a consequence, a lot of taxonomic expertise may be lost. However, Stefano Mariani and Thierry Backeljau were more optimistic about this, for they claimed that “classical” approaches are subject to a revival, both among students and funding agencies.

Perhaps the major take-home message from the discussion on taxonomy and species concepts, is that one might easily devote an entire conference to these issues, probably without arriving at a

general consensus. Yet this seems inherent to the very nature of biodiversity itself: it seems oversimplistic to try to look at biodiversity from one single point of view and hence the synergy between different approaches and conceptual frameworks is probably the best way of dealing with the measurement of biodiversity.

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***Summary of discussions on topic 2: Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Fisheries.**

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The marine realm is a theatre in which some very large scale selection experiments are being played out, with consequences that are ill-understood and potentially serious for the future health of our marine resources. The actors in this play are: (1) the fishery managers who set the rules by which fish are selected, (2) the fishers who apply the selective mortality, and (3) the fish stocks that undergo genetic change due to selective fishing.

The basis for this argument is as follows: all fishing is selective. Most obviously the market attaches different values to fish of different sizes, and fishers try to catch sizes that give the greatest return. In addition fishery managers try to regulate fishing through technical measures such as net mesh sizes. The trait most evidently under selection is size-at-age. But there are many others, including size and age at sexual maturation, potentially important for productivity of fisheries.

The strength of selection is large. Most fisheries are fully exploited or over-exploited (FAO, 1998); fishing mortality rates are often two or three times greater than all other sources of mortality put together. The strength of selection on body size is large enough to be detectable within single year classes of North Atlantic cod, as the fish grow and selective mortality takes place (Sinclair *et al.*, 2002).

There is genetic variation in the traits under selection. Over the last 20 years, the aquaculture industry has shown that it is quite feasible to bring about genetic change in growth and maturation of fish in captivity; this can only happen if these traits contain genetic variation. The heritabilities are in the range expected of traits closely related to fitness (Mousseau & Roff, 1987), even when estimated directly in the wild (Jónasson, Gjerde & Gjedrem, 1997).

Measurable changes in the traits are observable at least on a time scale of decades. Large phenotypic changes are taking place in major commercial fish stocks. These changes can, of course, be due to factors other than genetic change driven by selective exploitation. Nonetheless, the changes are often in a direction consistent with evolution, and remain even when proximal factors such as food supply and temperature are factored out (e.g. Rijnsdorp, 1993; Grift *et al.*, 2003; Heino, Dieckmann & Gødo, 2003).

The traits that change under fishery-induced selection are important for the sustained productivity of exploited fish stocks. Most startling are the results of a laboratory selection experiment on the Atlantic silverside (Conover & Munch, 2000). Starting from the same stock, they culled either

* Please refer to this section as:

Law, R. (2003). Summary of discussions on topic 2: Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Fisheries. Pp 43-45 in Hiscock, K. *et al.* (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

small individuals or large individuals from replicate populations; within four generations, the yield from the former was nearly twice that from the latter.

E-conference debate

Responses to the argument above mostly addressed the issue of how to manage fisheries, given that they evolve under exploitation. One view was that a substantial part of the marine realm should be designated as marine reserves (Mariani 9 October; 10 October), leaving natural processes to their own devices. There is much to recommend this, although the effectiveness of reserves clearly depends on the amount of gene flow across their boundaries and the strength of selection generated by fishing, relative to natural selection. [Theory suggests that the strength of selection generated by fishing can be at least an order of magnitude greater than natural selection in the reverse direction (Law, 2000, fig 3b).] Also, the merits of reserves have to be balanced against economic interests of communities dependent on fishing for their livelihoods.

Another issue was the extent to which the experimental results of Conover and Munch (2002) can be used make inferences about evolutionary management of stocks in the wild (Mariani 9 October; Kraak 10 October; 15 October). The life history of the species used by Conover and Munch was somewhat different to that of many major fish stocks; the work demonstrates the existence, but not necessarily the quantitative path of evolution under exploitation. It would be a great help to put in place experiments of a larger scale on commercially important fish species (Kraak 10 October).

Is it better to harvest smaller or larger fish for evolutionary management (Kraak 9 October; Mariani 9 October)? I suggested that there is some merit in maintaining a stock of large fish having relatively low fishing mortality rates on the basis that this helps to maintain the spawning stock biomass in the short term, and may generate selection for faster growth in the long term. However this argument runs counter to conventional view that fishing mortality should be concentrated on larger fish that have already achieved much of their potential growth. Clearly reducing fishing mortality on large fish will not do much good if it simply results in greater fishing mortality on smaller fish (Kraak 9 October; Law 11 October). There is a real need for research into this issue (Kraak 9 October; 15 October), guided by theory available from life-history evolution already in the literature (Law 11 October).

Another point raised in discussion was the erosion of genetic variability through small effective population size (Charrier 9 October). Whether there is general evidence that fishing causes small enough population sizes is open to question (Law 11 October). A major loss of variability requires dramatic genetic bottlenecks (Maltagliati 14 October), and populations driven to such small sizes by fishing may be on their way to extinction in any event. Complete destruction of the gene pool in this way is obviously of special concern.

Conclusion

The precautionary principle places a responsibility on us to leave our marine resources in a state that can be utilised as fully by our descendants as by ourselves. There is little systematic thought being given to these matters of selection and genetic change caused by fishing at the present time. If we continue on our present path, our descendants are not going thank us. Arguably we need a new — Darwinian — fisheries science.

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***Summary of discussions on topic 2: Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Aquaculture.**

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Having read all of the contributions to the conference it is clear that the community of scientists reached by the medium of electronic communication are fairly complacent about the potential effects of aquaculture on marine fish and shellfish. There were few direct comments on the subject of aquaculture.

Is there a need to worry about the potential effects?

As mentioned in the opening document aquaculture is the only sector of food production that is continuing to grow. In Europe and many other developed countries this is because fish is displacing other items from the diet, mainly on the basis of its healthy image. At present the world's aquaculture production is dominated by the production of freshwater carp species in Asia. This form of aquaculture is generally seen as environmentally neutral or possibly beneficial. Much of this production is actually from species that are non-indigenous to the countries in which they are grown.

However, the new growth in the aquaculture sector will be in the development of marine fish and shellfish because of their economic value and greater availability of suitable farm sites. The rapid and massive expansion of farmed marine shrimps and the devastation associated with the loss of mangrove habitat being a prime example of unsustainable aquaculture development.

It has been the problems of reproducing the broodstock and rearing the larval stages that has hindered the growth of the industry. However, the techniques applied to rearing seabream and seabass are now proving successful in other marine species such as cod and halibut so reducing the time taken to close the life-cycle of new species.

Once successful hatchery systems have been developed the actual number of fish in the industry may be many times that of the natural populations of a given species (eg Atlantic salmon production now nearly 1.2 million tonnes world-wide approximately a European wild catch <5000 tonnes). Widespread escapes under these conditions would eventually result in a homogenisation of allele frequencies over quite large areas of the species range. The seabass and sea bream industry standing crop (>100,000 tonnes) must be close to that of the total natural population size of both these species.

The inability of politicians to manage wild fish stock will mean that aquaculture will continue to produce more and more of the fish we eat. Farmed cod is already preferred by the premium hotel and catering trade because of its freshness, fillet quality and taste over wild fish.

* Please refer to this section as:

McAndrew, B.J. (2003). Summary of discussions on topic 2: Measuring, managing and conserving genetic diversity in aquaculture and fisheries. Aquaculture. Pp 46-48 in Hiscock, K. *et al.* (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

To date few restrictions have been imposed on the provenance of the fish being used in any form of aquaculture, recent legislation in the USA restricting the salmon industry in Maine to only use indigenous salmon strains is one of the first. This will put the local salmon farming industry at a distinct disadvantage, as these strains will perform much worse than the selectively improved strains used by farmers elsewhere in the world. Is this a long-term solution? Hatchery strains will change as they become domesticated unless managed as ranched stock.

The comments by Ferdinando Boero questioned the sustainability of the industry based on present feeding regimes. He likened the present process to us growing grass to feed sheep to feed these to wolves so we could feed lions. Fish are actually very efficient converters of food into protein compared to mammals and birds as they have a small metabolic cost. Substitution of fish meal and oil by vegetable protein and fat is feasible but marine fish cannot elongate or desaturate the fats into the omega 3's that we are being encouraged to eat because of their health giving properties. Most feed companies are trying to develop a standard that will result in 1kg of fish meal producing 1kg of farmed fish by controlled substitution. Fish meal is still relatively cheap and the cost of fish food is relatively expensive so the large feed companies will be happy to supply this sector over the price sensitive chicken and pig sectors. If we continue to loose our large carnivorous species what will eat the small prey species such as sandeels, capelin, anchoveta etc it may as well be turned into fishmeal if it is not polluted by PCBs.

The comments on the Conover and Munch work would not be surprising to people involved in selective improvement of fish. Properly controlled breeding experiments are showing good heritabilities for many traits important to aquaculture such as growth, body conformation and disease resistance. The fact that we are still dealing with essentially wild animals with relatively high levels of genetic variation and can impose high selection differentials, because of the high fecundity of fish, will ensure that these gains are likely to be maintained for many generations. Estimated improvement in growth performance of Norwegian salmon over 9 generations of selection is > 100%.

From the bullet points it can be read that aquaculture will continue to grow and the number of species being farmed will continue to increase. Improvements in husbandry, nutrition and genetics will result in improvements in the efficiency and sustainability of the industry. The potential impacts of aquaculture will be different to that of fisheries. Aquaculture could be used to mitigate the over exploitation caused by fisheries by releasing young fish to enhance natural stocks. This is already a widespread practice for salmonids and many freshwater species and is now being used in marine fisheries for lobster and Penaeid prawns. The availability of large number of animals for experimental purposes, particularly if they are from pedigreed stocks, is an important scientific asset to undertake genetic analysis of important fitness traits that have often been neglected in favour of a more molecular approach.

Aquaculture will result in escapees from most production units. This may result in the release of species that are non-indigenous and possibly invasive, such as the expansion of salmonids in South America. The use of indigenous species will mean that escapees will reduce possible differentiation between populations as gene flow increases between domesticated and wild populations. Avoidance of both these scenarios is possible by the use of single sex and/or sterile stocks that are relatively easy to produce, but are presently not acceptable to the industry.

Fish farms may also alter the marine environment at least locally because of the infrastructure, nutrients released and interactions with the same or other species. Some of the potential problems such as build-up of detritus are now actively managed by fallowing, predation by large carnivores (seals sealions) is being controlled by better net designs. But this is new industry and we have little

understanding of possible longterm effects that might appear. However the aquaculture industry is very sensitive to public opinion and, unlike fisheries, the larger more responsible companies will respond quickly to issues as they appear. If we continue to deplete our wild fish stocks and we want to continue to eat ever more fish then we will have to find the answers to the problems as they appear.

I recommend three particular papers that I believe are balanced assessment of the potential and problems of this new industry.

References:

- Naylor, R.; Goldburg, R.; Troell, M.; Beveridge, M.; Clay, J.; Folke, C.; Kautsky, N.; Lubchenco, J.; Mooney, H. & Primavera, J. (2001). Is Aquaculture A Net Loss? *Conservation Biology in Practice* Vol. 2, no. 4
- Naylor, R.L.; Goldburg, R.J.; Mooney, H.; Beveridge, M.; Clay, J.; Folke, C.; Kautsky, N.; Lubchenco, J.; Primavera, J. & Williams, M. (1998). Nature's subsidies to shrimp and salmon farming. *Science* Vol. 282, no. 5390, pp. 883-884
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*** Summary of discussions on topic 3 : Effects of human activities in altering genetic characteristics and gene flow in the marine environment.**

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Co-chairs introductions to Topic 3 focussed on two major aspects related to human alteration of gene flow in marine environment.

The first issue is biological invasions. Species-range expansions - often through human-mediated transport - have been observed at an increasing rate since the end of the 19th century. Biological invasions are one of the major threats for marine biodiversity and ecosystem stability. Several authors have underlined the potential usefulness, but lack, of molecular data for the study of biological invasions. Genetic studies could provide the opportunity to address major issues including the origin of newly founded populations, the rates of dispersal, and pathways through which dispersal occurs. Further contribution are to depict and understand the rates of change in the genetics of non-native species and associated life-history traits, the importance of natural selection in shaping the genetic architecture of genetically impoverished populations, and the degree of hybridisation between native and non-native species.

The second point addressed in the introduction dealt with human induced changes in the metapopulation structure of marine species caused by alteration of coastal habitats. Marine systems are often assumed to be continuous, and marine species to be able to disperse over large geographic ranges, yet the effective long-distance dispersal ability of marine species has rarely been quantified by direct measurements. Many marine species contradict this assumption, and show remarkable levels of genetic distinction over a range of geographic scales. Human activities can modify natural patterns of coastal landscapes, thus altering the level of isolation and connectivity among populations. They act both by the creation of corridors that allow/increase species dispersal, and by changing the degree of habitat fragmentation, thus affecting evolutionary processes. How relevant are these issues in the marine environment and how could population genetic studies provide insights into gene flow models that are relevant to marine species?

Several lines of discussions were followed during this topic and the two chairmen are grateful to all the contributors for their fruitful and stimulating discussions. From the 19 messages received during the three-days devoted to Topic 3, several important issues were addressed.

Firstly, the effects of human activities in altering genetic characteristics and gene flow in the marine environment are still to be quantified. In that context, the usefulness of population genetics is certain. However the time-scale needed to differentiate between 'natural' processes and anthropogenic effects is a critical point and the applicability of population genetics for short-term

* Please refer to this section as:

Viard, F.; Abbiati, M. (2003). Summary of discussions on topic 3 – Effects of human activities in altering genetic characteristics and gene flow in the marine environment. Pp 49-50 in Hiscock, K. et al. (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

management purposes is debated. There is still work to be done to formulate hypotheses that includes an *a priori* expectation as to the relevant scale to be considered and protocols useful for management purposes (see contributions by M. Abbiati, J.-P. Féral, Ferruccio Maltagliati). For instance, we cannot expect to see the same evolutionary and genetic patterns when considering climate change when investigating the sudden human-mediated introduction of a new species in a given habitat. Ferruccio Maltagliati also opened an interesting discussion concerning both the rate of evolutionary processes and the relevance of theoretical models (e.g. metapopulation) used primarily in the terrestrial environment (see also contributions by Frédérique Viard, J. P. Féral, M Abbiati).

Several contributors highlighted the need for extending our basic knowledge in systematics, taxonomy, phylogeography and species recognition (see the discussion line initiated by Keith Hiscock and answers from John Bolton, Sandra Duran and Frédérique Viard). This appears as a major issue concerning invasive species in particular when dealing with the concept of cryptogenic species (see Carlton 1996) and has implications for the establishment of species lists (e.g. list of threatened species) and management policy. For that purpose, as underlined several times not only in this topic but also in topic 1 and 2 of this conference, molecular and morphological analyses should both be carried out.

A third line of discussion dealt with the effects of climate changes (see contributions by Pierre Mathy, Boris Winterhalter, Keith Hiscock, Ferdinando Boero). Several programs (eg MarClim) and international councils or organizations (eg CIESM) have reported evidences of climate change on various biotas and some contributors provided specific examples. The genetic consequences of climate change at the species-level are however not straightforward (but see, for instance expectations about founder effects due to colonization of new habitats).

Many questions have been examined and many others are still to be discussed (see the questions addressed in the introduction by the chairmen). Obviously, marine communities and species are threatened by human activities. Also without a doubt, as with species in other environments, selective pressures (due to pollution), disruption of local populations, decreases in demographic size and geographical range of populations and the translocation of populations have implications on the genetic diversity and structure at the species level. All this is likely to affect species evolution. However, again without a doubt, the scientific community needs to define and formulate more specific and practical tools for management purposes and, at the same time, has to continue to extend its basic knowledge about marine species and communities.

*Final comments

Keith Hiscock

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I hope that you have all enjoyed and learned something from the MARBENA electronic conference on ‘Genetic biodiversity in marine ecosystems: measurement, understanding, management’. Although I feel something of a fraud overseeing a conference on genetics (I am a simple marine ecologist who likes whole organisms and understanding how they are distributed according to environmental conditions), I hope that we have managed to structure the conference in an informative and thought-provoking way.

I have learned a terrific amount particularly about the power that genetic studies have now and will have in answering questions about how humans are affecting the marine environment. The discussion about morphospecies and what we can learn about underlying biodiversity from molecular genetics was of fundamental importance in how we develop taxonomy (including in support of conservation) in the future. Although I agree with some later statements that morphological characteristics will continue to be the most practical tool for the field ecologist. I hope that the conference has given you some ideas about how studies of the genetics of marine organisms can help to identify those human activities that may most damage the marine environment. Those mentioned activities ranged from introduction of non-native species, through impacts of climate change and coastal construction to the effects of aquaculture and fisheries. ‘Connectivity’ and the role of human activities in breaking-down barriers has been a theme throughout the conference.

The concerns that pre-occupy agriculture on land – especially genetic manipulation/modification– do not seem to over-occupy marine biologists. However, significant worries about the impacts of fisheries have been raised – contributing to the now accepted view that fisheries are damagingly out of control and over-fishing may have permanent or very long-term adverse consequences. Some more discussion on genetic effects of fisheries and of aquaculture than our conference has given might be appropriate – commercial interests operate much more quickly than careful ‘academic’ research and before you know it, there is a problem.

Finally, my thanks to Federica Pannacciulli and Isabel Sousa Pinto for helping to determine topics that would inform the physical conference in Florence on “Genetic Biodiversity in Natural and Agricultural Ecosystems” being organised for 20-24 November 2003. Thank-you to the co-chairs for the three topics for providing such good outlines and for asking questions that provoked you into replying. Thank-you Ward Appeltans at the Flanders Marine Data and Information Centre for keeping the show on the road so effectively. And, especially, thanks to all who contributed their thoughts.

* Please refer to this section as:

Hiscock, K. (2003). Final comments. Pp 51 in Hiscock, K. *et al.* (eds): *Electronic conference on ‘Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management’ - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

Organisation and Statistics

* Organisation and statistics

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The conference was organized as a moderated bulletin board. Both the introduction to the themes and topics, and summaries of the discussions, were available on the Internet, (www.vliz.be/marben). Contributions to the conference were posted through a form on the web site. Contributions by non-moderators were flagged as 'non-moderated', until the forum administrator or moderator released them. For this purpose, they had access to a separate form, which allowed editing or deletion of messages.

A total of three separate themes were discussed in consecutive days (table 1). Discussions were guided by two co-chairs, each specialized in one of the topics. The themes had three days to run intensively, but discussions continued until the end of the conference. The co-chairs were responsible to open the discussion by making their opening statements and to follow up the discussion. They were also responsible to provide a general summary and synthesis of the discussion to include in this report.

An additional topic was raised for those who wanted to add messages of general issues on marine biodiversity, general aspects to the discussions held during this e-conference and to finalize the e-conference with a synthesis.

Date	Title	Chairs
6 October	"Introduction and overview "	S.J. Hawkins & K. Hiscock
I: 6-8 October	"Measuring managing and conserving genetic resources in natural marine systems "	T. Backeljau & F. Maltagliati
II: 9-10, 13 October	"Measuring, managing and conserving genetic diversity in aquaculture and fisheries"	B.J. McAndrew & R. Law
III: 14-16 October	"Effects of human activities in altering genetic characteristics and gene flow in the marine environment"	M. Abbiati & F. Viard
17 October	"Synthesis & Closure"	K. Hiscock

Table: 1. Time table: topic, themes and co-chairs respectively.

The basic flow of information of the conference was through the WWW. This was done to stimulate 'external' parties to participate in the discussion. To make sure the conference was widely known, mailing lists of several organizations and activities were used to invite all interested parties

* Please refer to this section as:

Vanden Berghe, E. (2003). Organisation and statistics. Pp 53-54 in Hiscock, K. *et al.* (eds): *Electronic conference on 'Genetic Biodiversity in Marine Ecosystems – Measurement, Understanding and Management' - Summary of discussions, 6 to 17 October, 2003*. Flanders Marine Institute: Oostende, Belgium.

to register. Access to the general pages of the conference, and to the summaries, is open to everyone. To be able to post messages and also to view posted messages, registration through a form on the web site was necessary. The requests for registration were handled individually; applicants were informed of successful registration in an e-mail. Once registered, access to the forum was possible by logging-in with user-defined username. The obliged login username aids in referring to the authors' details by linking to IMIS (Integrated Marine Information System), and in addition enables us to score participation during the course of the conference.

Statistics

Registered participants (includes 'marble' participants): 853

Registered participants to 'marble': 336

Number of countries: 60

Participants requesting summaries through e-mail: 576

Numbers of addresses on the circulation list: 1709

Number of messages: 79

Number of contributors: 30

Hits on marbena web site: 21,438 (from 1/10/2003 to 31/10/2003)

Hits on /cgi-bin/marbena.exe: 7,850

Hits on /marbena: 13,588, or approximately 3,068 html pages

Total number of pages requested: 10,918

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* For the sake of saving paper, the complete list of MARBENA subscribers is omitted and reduced to those who have contributed to the discussions.