

ICES Oceanography Committee  
ICES CM 2004/C:08, Ref. ACME

---

## Report of the ICES-IOC Working Group on Harmful Algal Bloom Dynamics (WGHABD)

---

5–8 April 2004  
Corsica, France

This report is not to be quoted without prior consultation with the General Secretary. The document is a report of an Expert Group under the auspices of the International Council for the Exploration of the Sea and does not necessarily represent the views of the Council.

International Council for the Exploration of the Sea  

---

Conseil International pour l'Exploration de la Mer

Palægade 2-4 DK-1261 Copenhagen K Denmark  
Telephone + 45 33 38 67 00 · Telefax +45 33 93 42 15  
[www.ices.dk](http://www.ices.dk) · [info@ices.dk](mailto:info@ices.dk)

## Contents

---

1	WELCOME AND OPENING OF THE MEETING .....	5
2	TERMS OF REFERENCE.....	5
3	SUMMARY AND CONCLUSIONS.....	6
4	TERMS OF REFERENCE IN DETAIL .....	8
4.1	Term of reference a: collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis.....	8
4.2	Term of reference b: review plans for the proposed Workshop on New and Classic Techniques for the Determination of Numerical Abundance and Biovolume of HAB-species .....	8
5	PRELIMINARY ORGANISATION .....	9
5.1	Preliminary list of participants to be invited .....	9
5.2	Background.....	10
5.3	Term of reference c: Review progress in computerized production of decadal maps from country reports, including revision of reports already in the database covering the last 10 years .....	12
5.4	Term of reference d: propose types of analysis that should be performed using the IOC-ICES HAE-DAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted.....	12
5.5	Term of reference e: review the report of the Workshop on Real-time Coastal Observing Systems for Ecosystem Dynamics and Harmful Algal Blooms (CM 2003/C:15).....	13
5.6	Term of reference f: review existing phytoplankton population dynamics models with particular emphasis on prediction of HAB events .....	13
5.7	Term of reference g: review biological loss processes of selected HAB species.....	14
5.8	Term of reference h: consider the environmental dynamics and impacts of individual phycotoxins and their metabolites enabled by new analytical technologies.....	15
5.9	Term of reference i: report and discuss new findings .....	16
5.10	Term of reference j: start preparations to summarise the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006.....	19
6	NATIONAL REPORTS.....	19
7	OTHER REPORTS.....	28
8	DRAFT RESOLUTIONS .....	28
9	WGHABD RECOMMENDATIONS .....	31
10	CONCLUDING BUSINESS.....	31
11	ANNEXES .....	32
	Annex 1 List of Participants.....	32
	Annex 2 Agenda of the meeting.....	35
	Annex 3 IPHAB Recommendations.....	36



# 1 Welcome and opening of the meeting

---

The ICES-IOC Working Group of Harmful Algal Bloom Dynamics (WGHABD) meeting was hosted by the University de Liège, STARESO, Corsica, France from 5–8 April 2004. Twenty -four scientists from 17 countries participated. The list of participants is presented in Annex 1. The meeting agenda is presented in Annex 2.

The meeting was opened by Dr. Anne Goffart from the University of Liege on the 5 April 2004 and the participants were introduced with respect to their names, institute, national affiliation and fields of expertise. The agenda was agreed and Eileen Bresnan and Pat Tester elected as Rapporteurs.

Being a joint ICES-IOC working group, the IOC every year announces the possibility for its Member States outside the ICES area to attend the WGHABD. Most years these participants are supported financially. In 2004 the IOC received applications to participate from Bangladesh, Bulgaria, China, Iran, Malaysia, Mexico, Morocco, Nigeria, Philippines, Tunisia, UAE and Uruguay. The applicants' CV's were reviewed by the IOC Secretariat, the WGHABD Chair and the Chair of the IOC Intergovernmental Panel on Harmful Algal Blooms (IPHAB). Participation and financial support was offered to the applicants from China, Egypt and Tunisia. The following attended; Kedong Yin (China), Snezana Moncheva (Bulgaria), Ons Daly Yahia-Kefi (Tunisia), and Amany Ismael (Egypt).

The Chair outlined the comments and review from the ICES Oceanography Committee relating to the WGHABD report from the 2003 meeting. The committee felt the report was well organized, informative and the meeting well attended. The proposed resolution for a workshop on new and classic techniques for the determination of numerical abundance and bio-volume of HAB-species in Sweden, August 2004) was supported and encouraged.

The Terms of Reference for 2004 were reviewed and adopted. A Term of Reference concerning starting preparations to summarise the distribution and number of harmful algal blooms in the North Sea for the period 2000-2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006 was added to the ToR.

## 2 Terms of reference

---

At the 90<sup>th</sup> Statutory Meeting (2003), Tallinn, Estonia the council approved the WGHABD (2004) Terms of Reference (C. Res. 2C07).

The **ICES-IOC Working Group on Harmful Algal Bloom Dynamics** [WGHABD] (Chair J. L. Martin, Canada) will meet in Corsica, France, from 5–8 April 2004 to:

- a) collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis;
- b) review plans for the proposed Workshop on New and Classic Techniques for the Determination of Numerical Abundance and Bio-volume of HAB-species;
- c) review progress in computerised production of decadal maps from country reports, including the revision of reports already in the database covering the last 10 years;
- d) propose types of analysis that should be performed using the IOC\_ICES HAEDAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted;
- e) review the report of the Workshop on Real-time Coastal Observing Systems for Ecosystem Dynamics and Harmful Algal Blooms (CM 2003/C:15);
- f) review existing phytoplankton population dynamics models with particular emphasis on prediction of HAB events;
- g) review biological loss processes of selected HAB species;
- h) consider the environmental dynamics and impacts of individual phycotoxins and their metabolites enabled by new analytical technologies;
- i) report and discuss new findings;

- j) start preparations to summarise the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006.

### 3 Summary and conclusions

---

Techniques for analysis and prediction of the population dynamics of HABs are not well developed and measures of species-specific growth rates and mortality rates are very difficult. Monitoring is an important aspect of HAB research and the WG needs to interact with monitoring programme designs and data interpretation. For example, more environmental data is often needed and sampling should be rationalised with local hydrography such as mixed layer depth, circulation patterns, frontal dynamics, etc. Historical data and time series data are important in looking for historical occurrences of HABs. Increase and decrease in population size is important to bloom dynamics.

The importance of the WG approach and focus on population dynamics of specific HAB species and not on phytoplankton ecology in general was emphasised. The economic, resource and environmental effects of HABs are included within the WGHABD. In addition, often phytoplankton ecology models are usually biomass, nutrient, and carbon cycling and in many cases, cannot define, explain or predict HAB dynamics. In the past we have had joint meetings with SSO and modellers to try and incorporate physics and HAB dynamics into the models.

The WG felt that the existing ToR were related and important to dynamics.

#### **Term of reference a**

Collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis

National reports were presented for Belgium, Canada, Denmark, Estonia, France, Germany, Ireland, Latvia, The Netherlands, Norway, Poland, Portugal, Spain, Sweden, the UK, and the USA. Maps were updated for inclusion to the decadal maps. Information for the database was provided in the required format.

#### **Term of reference b**

Review plans for the proposed Workshop on New and Classic Techniques for the Determination of Numerical Abundance and Bio-volume of HAB-species

This ToR remains unchanged. As it is not possible to secure funding for a Workshop in 2004, a decision was made to resubmit the resolution to ICES with the intention of holding the Workshop in August 2005. The objectives for the workshop will be to compare traditional methods of counting HABs using microscopic, molecular and other new techniques. It was emphasized that this Workshop would be an intercalibration exercise, as opposed to method development and would be restricted to approximately 24 participants who are currently using the identified methods.

#### **Term of reference c**

Review progress in computerized production of decadal maps including the revision of reports already in the database covering the last 10 years

Decadal maps are currently being updated manually. A new Decadal maps product which uses both ArcView and Flash software, and allows updating of maps from a MySQL database is being explored. The use of the MySQL database both in the new HAEDAT format and in the New Decadal Maps will open future technical options for linking these two datasets that will be studied during this year.

#### **Term of reference d**

Propose types of analysis that should be performed using the IOC-ICES HAEDAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted

At present information is entered into the HAEDAT dataset (which is in Access97 format). By next year there will be an electronic format (with the same information as previous forms) will be available for submission directly into the database. Monica Lion (IOC-IEO-SCCHA, Vigo, Spain) will prepare a list of potential problems for conversion of all the old historic records into the new form. Designated country delegates will go through these old records and identify discrepancies.

Although HAE-DAT has been underutilized in the past, it was used to generate economic loss information and display fishery closures. The WG suggested that the ICES WGSAM be given a subset of “clean” data for analyses.

### **Term of reference e**

Review the report of the Workshop on Real-time Coastal Observing Systems for Ecosystem Dynamics and Harmful Algal Blooms (CM 2003/C:15)

The Workshop, held in Villefranche (France) in June 2003, focused on the development and application of real-time observational technologies for coastal monitoring, with particular emphasis on harmful algal blooms. It was a successful, productive workshop that acknowledged issues associated with limitations with in-water and remote sensing for HAB dynamic studies and monitoring. It also provided the biologists and biooptical and physical oceanographers an opportunity to determine the needs for improving detection and resolution in the determination of observation technologies.

### **Term of reference f**

Review existing phytoplankton population dynamics models with particular emphasis on prediction of HAB events

Four presentations were made featuring: a fuzzy logic model of the Baltic and regional seas, influences of monsoons and oceanographic processes in Hong Kong waters, marine ecosystem and management advice (with a focus on HABs) for the Skagerrak / North Sea, and a synthesis of *Alexandrium* blooms in the Gulf of Maine.

Questions that arose from the presentations included: scientific and management questions requiring answers from modelling; the level of complexity; what information is required from both the modellers and biologists; the predictive capabilities of the models; knowledge and technical gaps that are impeding progress towards the quantification of phytoplankton.

A joint Theme session between WGHABD and WGPBI at ICES ASC in 2006 titled “Harmful Algal Bloom Dynamics: Validation of Model Predictions (possibilities and limitations) and status on coupled physical-biological process knowledge” was proposed.

### **Term of reference g**

Review biological loss processes of selected HAB species

Three presentations were made highlighting featuring predator-prey and selective feeding behaviours, niche dynamics studies. It was felt that loss processes are important to population dynamics, and at present there are only poor estimates of values. The WG proposed that it further study findings from relevant projects with the goal of recommending appropriate loss terms for models.

### **Term of reference h**

Consider the environmental dynamics and impacts of individual phycotoxins and their metabolites enabled by new analytical technologies

A comprehensive overview of environmental dynamics of phycotoxins and advances in analytical technologies was given. It highlighted instrumentation, trophic transfer, metabolic effects, microextractions, and future directions.

### **Term of reference i**

Report and discuss new findings

New findings were presented on:

- the detection of yessotoxins and *Prorocentrum minimum*.
- *Alexandrium* bloom dynamics for the Gulf of Maine, emphasizing the critical role played by numerical models in interpreting and analyzing observations
- a new digital system to record holograms of plankton *in situ* was described
- influences of monsoons and oceanographic processes on red tides in Hong Kong waters

### **Term of reference j**

Start preparations to summarise the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006.

Representatives from countries working in regions of the North Sea discussed meeting in Norway in September to compile data and determine the state of knowledge.

## 4 Terms of reference in detail

---

### 4.1 **Term of reference a: collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis**

National reports were presented for Belgium, Canada, Denmark, France, Germany, Ireland, Latvia, The Netherlands, Norway, Sweden, the UK, and the USA. Estonia, Poland, Portugal, and Spain, whose representatives did not attend the meeting, sent their national reports which were presented during the meeting.

Maps were updated manually for inclusion to the decadal maps and HAE-DAT forms were collected in electronic form.

The WGHABD has coordinated the collation of data for HAE-DAT since it began (<http://ioc.unesco.org/hab/data3.htm#1>). This also includes reports from countries where no representative participate in a WGHABD meeting in a given year. The IOC has taken the initiative to expand the HAE-DAT to global coverage. PICES is testing the format for 2003 events and will evaluate its experiences at a meeting in October 2004. IOC is working with its regional networks and HAB working groups to serve as a focal point for submission to HAE-DAT. At present this is on the ToR for the South American (FANSA) Group, the Caribbean Group (ANCA) and the North African Group (HANA).

The WG discussed the importance of the establishment of this database, its usefulness, and the importance of maintenance, updating and the formal extension to a global coverage. In order for HAEDAT to mature and develop into a stable and reliable source of data the WGHABD judges it necessary to streamline and formalize the mechanism for submission of reports. Therefore the WGHABD recommends that appropriate specific sources be identified for the information from each country. The WGHABD recommends that the appropriate officers of each sponsoring organisation contact their national members to nominate national focal points/individuals responsible for data submission to HAE-DAT. It is recommended that these focal points (for ICES Member States) are identified by March 2005. The WGHABD will continue to be responsible for the database and will review the submitted reports annually.

The HAE-DAT is unique and by moving the focus of the WGHAB in the ToR from collating the records to analysing them, the WGHABD will, to a much greater extent, benefit from HAE-DAT and it will facilitate the general use of the data in HAE-DAT.

### 4.2 **Term of reference b: review plans for the proposed Workshop on New and Classic Techniques for the Determination of Numerical Abundance and Biovolume of HAB-species**

The focus for study, potential participants, practical organization and possible publication output of the workshop were discussed thoroughly by all the participants at the working group. The WGHABD took into consideration Recommendation IPHAB-VI.4 (Annex 3 hereto) of the IOC Intergovernmental Panel on HAB which requests the WGHABD to consider the coordination of this workshop with the third international workshop on molecular probes. Sources of funding for this workshop were discussed and it was also noted that the WGHABD in 2003 anticipated a preparation period of 15 months once funding has been secured. The working group discussed the overall objective of the workshop and decided that it should focus on intercalibration of methods used for numerical abundance measurements of HAB-species. The experimental design of the intercalibration should be sufficiently robust to ensure strong statistics and significant results. A small number of focus species was preliminarily selected. To limit the scope of the workshop it was decided that biovolume measurements would be excluded from this workshop and could be dealt with in a separate future workshop devoted to this issue. It was also decided that the workshop should not be a training exercise, thus participants will be invited to cover the spectrum of methods currently used in HAB-monitoring and studies of HAB dynamics. The Organising Committee of the Intercalibration Workshop shall summarise the findings and conclusions in one or more manuscripts to be submitted for publication in an appropriate peer reviewed journal. The tentative date for the intercalibration workshop is August 2005. The workshop is planned to include 5 full working days. The ToR is to remain unchanged for the 2005 meeting of the WGHABD.



## 5 Preliminary organisation

---

Convenor: Bengt Karlson  
Co-convenor: Caroline Cusack  
Local organiser: Odd Lindahl

Organisation committee:  
Bengt Karlson, Sweden  
Caroline Cusack, Ireland  
Odd Lindahl, Sweden  
Chris Scholin, USA  
Don Andersen, USA  
Einar Dahl, Norway  
Per Andersen, Denmark

### 5.1 Preliminary list of participants to be invited

<b>Classic techniques</b>	<b>Proposed participant</b>
Utermöhl, Lugol fixation	Alejandro Clement (Chile) Mats Kuylenstierna, Lars Edler (Sweden)
Utermöhl, Formaldehyde fixation	Malte Elbrächter (Germany), Murielle LeGresley (Canada)
Filtering, Calcofluor staining	Per Andersen (Denmark)
Filtering, counting on semitransparent filters	Lars-Johan Naustvoll (Norway)
Filtering freeze transfer	Kevin Pauley (Canada)
Flowcam	Jennifer Martin (Canada)
SEM	Jim Ehrman (Canada)

<b>Molecular biological techniques</b>	<b>Proposed participant</b>
Whole cell technique with rRNA-probes	Allan Cembella (Germany), Linda Medlin/Katja Kerkmann (Germany), Don Anderson (USA), Lesley Rhodes (New Zealand), Laure Guillou (France), Caroline Cusack (Ireland)
Sandwich hybridization assay	Chris Scholin (USA)
Electro chemical detection of rRNA-probe stained	Linda Medlin/Katja Kerkmann (Germany)
Chemscan	Linda Medlin/Katja Kerkmann (Germany), Nyree West (France)
PCR-based detection	Anna Godhe (Sweden), Laure Guillou (France)
Quantitative PCR	Don Anderson (USA)
Antibodies	Tonje Castberg (Norway)

## 5.2 Background

Currently almost all HAB-monitoring for aquaculture is performed using classical microscopic techniques for determining abundance and biomass of HAB-species. Also, most studies of HAB dynamics use these techniques. New probe-based techniques show great potential for studying HAB dynamics with much higher resolution in time and space than previously available, making it possible to understand biological processes leading to HAB events in detail. However, validation of the new techniques in the field is limited.

Classic microscope based techniques are not standardised. Developments using filtering and centrifuging for fast sample throughput need to be intercalibrated with sedimentation chamber techniques. One example is the Calcofluor staining technique that is extensively used by some institutes for the identification of thecate harmful dinoflagellates. The problem of determining abundance of HAB-species that occur in low cell densities and still render shellfish toxic needs to be revisited. Also developments in computer aided microscopy for determining biovolume need to be intercalibrated with manual methods.

The HAB scientific community has been working towards the development of species- or strain-specific “probes” that can be used to label only the cells of interest so they can then be detected visually, electronically, or chemically. Progress has been rapid and probes of several different types are now available for many of the harmful algae, along with techniques for their application in the rapid and accurate identification, enumeration, and isolation of individual species.

With respect to applications on HABs in natural waters, the sandwich hybridization assay (SHA), as well as “whole” or intact cell assays using rRNA probes, have been used in field trials in several areas of the world, including both the east and west coasts of the US. (C. Scholin and D.M. Anderson, unpub. data), off the coast of Scotland (John *et al.*, 2002), and in several countries where *Pseudo-nitzschia* species cause ASP toxicity (C. Scholin, unpub. data). The most extensive field applications of PCR-based molecular probe technologies to HAB species are probably in the monitoring for *Pfiesteria piscicida* and other *Pfiesteria*-like species in the southeastern US.

One problem area has arisen with the application of both whole cell and SHA technologies to field populations – namely the agreement between cell counts made with different methods. For example, *A. fundyense* counts using an rRNA probe in the whole-cell format agreed to a variable extent with SHA analyses of the same samples from the Gulf of Maine (D. Anderson, unpub. data). At some stations and at some depths, agreement was excellent between the two methods, but for others, the SHA counts were 2 to 20X higher than the manual counts. It is possible that this discrepancy is due to grazing, perhaps resulting in the incorporation of *A. fundyense* cells and/or rRNA in fecal pellets or other detritus that was detected by the SHA, but not by the whole-cell method. Laboratory experiments, however, have not supported this hypothesis, so the reason for the discrepancy remains unknown.

In a similar manner, Allan Cembella reported at the ICES-IOC WG meeting (2002) on studies of *Alexandrium* populations off the coast of Scotland in which bright-field microscope counts of Utermöhl samples were consistently higher than whole-cell counts using species-specific oligonucleotide probes (John *et al.*, 2002). Here again, the discrepancies are significant – an order of magnitude or more. In this case, the differences are between probe-based, whole-cell counts and standard microscope counts, whereas in the Gulf of Maine data cited above, the differences were between the whole-cell probe approach and the SHA.

It is important to recognize the fundamental differences between these the different assay types that have been developed. For example, a successful whole-cell assay requires detection of molecules inside intact, recognizable cells and those molecules must: a) be accessible to the probe, and b) be of sufficient quantity to visualize that cell above background. Furthermore, the target cell must survive treatment from sample collection through processing and be visible to be counted. Results of a whole-cell assay are thus operationally defined – even if a target cell is present it may not always be detected with this approach. Furthermore, anything that causes cells to clump or otherwise be hidden (e.g., large quantities of particulate organic matter, fecal pellets) will affect results of a whole-cell assay, as shown for *Pseudo-nitzschia* by (Scholin *et al.*, 1999) and *Heterosigma* by Tyrrell *et al.* (2001). Similarly, results of cell homogenate assays are operationally defined. The basic concept of the SHA is to detect molecules freed from particulate matter, analogous to detection of algal toxins or DNA sequences in phytoplankton samples. Target cells, or even remnants thereof, need only survive the initial collection step. Successful detection of the target molecule then depends on: a) extracting the target molecule, b) a sufficient quantity of the target molecule to elicit a positive reaction, and c) minimal interference (signal suppression/enhancement) from the sample matrix.

With these considerations in mind, several possible explanations for the observed discrepancies in cell count estimates can be offered. For the whole-cell approach, cellular uptake of the probe may vary (independent of rRNA concentration) due to permeability differences, such as those associated with life history transformations or nutritional condition. Temporary cysts, for example, are readily formed by *Alexandrium* species when subjected to sudden mechanical or environmental stress (Anderson and Wall, 1978). This could lead to weakly stained cells, and lower counts. Alternatively, cells may be more prone to lysis under certain physiological conditions, reducing the number of cells observed by the whole-cell assay. These differences might be enhanced by the different processing and preservation procedures followed for the different probe-based assay methods. In particular, the formalin/methanol fixation used in some whole-cell assay may lyse more cells relative to the liquid nitrogen typically used in SHA assays. The extent of lysis may vary depending on physiological condition of cells, so counting differences might be expected in this regard.

The nutritional, temperature and light history of the cells may also have a significant effect on the results of the different assay methods. This again might reflect cell permeability differences, or the accessibility or structural form of the target rRNA in whole cells versus a cell homogenate.

Clearly, more work is needed before probe-based cell counts can be accepted as an alternative to more traditional approaches. The differences between these probes and classical methods and between different types of probe-based methods are significant and raise important questions about what actually should be counted, or is being counted, in research and monitoring programs focused on HAB species.

The workshop will have the following deliverables:

Through laboratory exercises, presentations, and discussions:

- 1) A comparison of traditional methods for concentrating, preserving, and counting common HAB species using light microscope techniques;
- 2) A comparison of molecular probe-based methods for cell enumeration with the traditional techniques;
- 3) Recommendations for further research and development efforts targeted at identified inaccuracies or deficiencies in the methods being evaluated;
- 4) Identification, where possible, of a reference counting method against which other methods can be calibrated;
- 5) Assessment of the usefulness and cost efficiency of the available numerical methods in routine monitoring.

Fifteen months of preparation between approval and the workshop are required and it is recommended that the Steering Committee members meet within 4 months of the approval of the workshop to plan the workshop in full detail.

The scope and objectives of the workshop are consistent with the objectives of WGHABD and GEOHAB.

#### The resolution:

A workshop on **new and classic techniques for the determination of numerical abundance and biovolume of HAB-species – evaluation of the cost, time-efficiency and intercalibration methods** will be held in Kristinneberg, Sweden, sponsored by ICES, IOC, GEOHAB, and EU with Dr. Bengt Karlson and Dr. Caroline Cusack as Conveners.

Priority:	ICES should take an active role in developing the implementation plan of the GEOHAB programme. The topic of intercalibration is relevant for GEOHAB and also fits well into ICES profile.
Scientific Justification:	Almost all HAB monitoring and dynamics studies are performed using classical techniques for determining abundance and biomass. New probe-based techniques show great potential for studying HAB dynamics and will make it possible to understand biological processes leading to HAB events. However, the validation of the new techniques is limited. Classic microscope techniques need to be compared with species and strain specific molecular probe methods as well as methods for preserving and concentrating phytoplankton. The goal is to produce scientifically based recommendations on choice of methodology for HAB-monitoring programmes.
Relation to Strategic Plan:	Implementation of the GEOHAB programme is relevant to the quantifying of human impacts on the on the marine ecosystem. The workshop will produce scientifically based recommendations on
Resource Requirements:	Conveners and lecturer's work time is required. Travelling and accommodation costs are needed for meeting participants. Laboratories, appropriate equipment and convenient access to coastal waters are required during the workshop. Technical support would be required for publication.
Participants:	Experts in relevant fields from around the world would be invited to participate.
Secretariat Facilities:	The Secretariat will be involved as normal in general professional and secretarial support, and the Secretariat should provide direct assistance during the workshop. The Secretariat might provide web space for the proceedings.
Financial:	Travelling support is needed for participants. Funds will be asked from IOC, EU, SCOR and other relevant organizations.
Linkages to Advisory Committees:	Harmful algal blooms are continuing issues in ACME.
Linkages to other Committees or Groups:	Support can be anticipated from the Baltic Committee, WGPE, and SGGIB.
Linkages to Other Organizations:	GEOHAB is sponsored by IOC and SCOR.

### **5.3 Term of reference c: Review progress in computerized production of decadal maps from country reports, including revision of reports already in the database covering the last 10 years**

Monica Lion, from the IOC-IEO SCCHA, Vigo, Spain, reported on the progress of the transformation of the present HAE-DAT software into a user-friendly format. With collaboration from Benjamin Sims from the UNESCO headquarters in Paris, the present HAE-DAT that runs under Access 97, is being transformed into a MySQL database. This will make the database on-line searchable and therefore it will not be necessary to download the complete dataset to the user's computer.

This new format will also allow the HAE-DAT forms to be filled in directly on-line, avoiding manual data input that, until now, has been done by the IOC-IEO SCCHA.

National focal points will have a user name and a password that will allow them to input new data and check old reports from their country. Country representatives will be responsible for dividing coastlines into section 100-200 km in length in order to update maps electronically with longitudinal/latitudinal information.

Catherine Belin (Ifremer, France) - who did not attend the meeting and whose data were presented by Monica Lion - submitted information concerning a new Decadal maps product which uses both ArcView and Flash software, and allows updating of maps from a MySQL database.

The use of the MySQL database both in the new HAEDAT format and in the New Decadal Maps will open future technical options for linking these two datasets that will be studied during this year.

### **5.4 Term of reference d: propose types of analysis that should be performed using the IOC-ICES HAE-DAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted**

Monica Lion (IOC-IEO SCCHA, Spain) reported problems and gaps of the present data included in HAE-DAT dataset.

One of the most significant problems with the present format for the dataset is the differences that exist in location information. This problem should be resolved with the introductions of the new HAE-DAT area codes that will replace the ICES area codes. There will also be standardization in the use of grades, minutes and seconds, in the latitude and longitude identification of the actual locations of actual events. The new HAE-DAT areas will represent coastal areas that are 100-200 km in length where they will be represented by a central dot that will be used as the focal point of reference for the different maps.

In addition, other problem areas that should be taken into account include the possible errors that may exist in the taxonomic information for a particular species that was involved in the event. This could include improper identification, the use of different synonyms, or the appearance of new species, reclassifications, and/or typing mistakes.

With the intent of improving the quality of the data, the new HAE-DAT format will include a drop down menu with a species list and names based on information in the updated IOC Taxonomic Reference List of Toxic Plankton Algae which will automatically link the genus or the species of microalgae with its taxonomic class.

Inconsistencies in information reported for toxin assay will also hopefully be remedied with the new format. And last, but not least, it was pointed out that there have been inconsistencies in the reporting of events in the past. Many past reports indicate the start and end of the bloom, where others report the dates quarantine levels were measured. This problem will be solved by the inclusion of two different fields in the new HAE-DAT form.

This new format will also allow the HAE-DAT forms to be filled in directly on-line, avoiding the manual data input that until now has been carried out by the IOC-IEO SCCHA. As soon as the new HAE-DAT form is ready to be checked, the IOC-IEO SCCHA will send country delegates a user name and a password that will allow them to input new data and check old reports from their country. They will amend, where possible, the old reports and fill in the new HAE-DAT area codes. During this verification period, country delegates will work with the IOC-IEO SCCHA and database experts from the IOC (UNESCO) and suggest improvements for the new forms.

Taking into account all the advantages of this new automated format, the old dataset revision process that had been agreed to during the last WGHAB meeting was delayed until this new format is operational. As soon as the new HAEDAT is operational, it will be available 'on line' initially for a verification period. During this time, access will be restricted to authorized personnel and, as soon as reports have been checked, there will be free access. Once the verification period has been completed, and the data quality improved, the WG has suggested that statistical analyses be initiated,

Given the extensive literature about the unverified paradigm of the "spread" and "increase" of HABs, the WGHABD concluded by formulating four key questions:

Can HAE-DAT data be used to describe:

- 1) the occurrence of new species;
- 2) the frequency of event occurrences;
- 3) the magnitude of harmful events;
- 4) the specific toxicity?

In order to answer these questions, the WGHABD proposes that the WGSEAM be asked to:

- 1) analyze a small subsample of the dataset that has been verified and based on these results the WGHABD will be informed as to which types of statistical analyses for which the dataset is suited;
- 2) identify appropriate tools for sporadic event analysis within HAE-DAT.

### **5.5 Term of reference e: review the report of the Workshop on Real-time Coastal Observing Systems for Ecosystem Dynamics and Harmful Algal Blooms (CM 2003/C:15)**

The IOC/ICES/SCOR HABWATCH workshop in Villefranche, France, in June 2003 included a total of approximately 90 participants. More than 20 plenary lectures were supplemented by practical demonstrations and tutorials that included remote and *in situ* sensing technology and data processing, measurement of inherent and apparent optical properties, application of taxon- and toxin-specific probes and development and deployment of operational oceanographic systems for oceanography. The focus of the workshop was on the development and application of real-time observational technologies for coastal monitoring, with particular emphasis on harmful algal blooms.

Three participants at the workshop, including two members of the HABWATCH Organising Committee, reported to WGHABD. The workshop was very successful in bringing together scientists from different disciplines concerned with real-time studies of HAB-dynamics and in introducing practical applications of many different techniques. Most of the tutorials were found to be productive and well organised. This advanced workshop represents the first concerted effort to integrate biooptics with HAB dynamics in a global scientific framework with emphasis on the end-users of such technology. Although some participants expressed the view that the focus on marine optics was disproportionate, it was generally acknowledged that at least the issues associated with current limitations of in-water and remote sensing for HAB dynamic studies and monitoring was well explored in this workshop. The biologists gained an appreciation of the physical and optical constraints inherent in the development of observational technology, whereas the biooptical and physical oceanographers were made aware of the requirements of biologists for improved detection and resolution of specific taxa in bloom events in real-time. It was apparent that no single technological approach could satisfy requirements for bloom dynamic studies and monitoring – the integration of multiple techniques is the obvious way forward. Information from this workshop including the programme abstracts and complete image files from the tutorial and demonstration sessions are available on the HABWATCH website. A special volume based on the plenary lectures will be published in 2005 in the series of UNESCO Monographs in Oceanography.

### **5.6 Term of reference f: review existing phytoplankton population dynamics models with particular emphasis on prediction of HAB events**

The rapid variability of this issue is so complex that it makes numerical modelling a useful tool for integrating/extrapolating physical-chemical-biological process knowledge and sporadic data into “continuous” 3D-space and time (quantification). It is crucial to take into account all the main driving forces on and processes around HABs. The main driving force is the varying climate/physics, in some areas fertilization and for some species predation/mortality. (In some areas pollution may be important).

A new version of a one-dimensional coupled biogeochemical-physical model was presented with validation from a set-up for the Skagerrak area (Station Släggö, Bohus coast). The physical and chemical conditions as well as total phytoplankton biomass in spring 2001 was well described by the model. Plans for implementation in the SMHI ecosystem model with four phytoplankton groups coupled to a 3-D physical model and some results from the fuzzy logic modelling of *Nodularia spumigena* in the Baltic from the EU-project HABES were presented.

Two 3D models were described, the *Alexandrium* model applied to the US east coast (*see also new findings*), and the NORWECOM model applied to the North Sea/Skagerrak/Norwegian coast. The importance of cysts together with ocean circulation, growth and mortality due to nutrient limitation were stressed by the *Alexandrium* model with a self-seeding and “propagatory” area in the Bay of Fundy in Canada postulated.

An overview of the ecosystem modelling approach and NORWECOM was discussed in detail. A simple view of an ecosystem approach is to consider the most important driving forces on, and the processes around, the ecosystem dynamics. The rapid variability of HAB processes is so complex that it makes numerical modelling a useful tool for integrating/ extrapolating physical-chemical-biological process knowledge and sporadic data into “continuous” 3D-space and time (quantification). It is crucial to take into account all the main driving forces on and processes around HABs. The main driving force is the varying climate/physics, in some areas fertilization and for some species predation/mortality. In some areas pollution may be important.

For most purposes, NORWECOM has been used in an area covering an extended North Sea (sometimes including the Bay of Biscay) with a horizontal resolution of 20 x 20 km. This resolution is too coarse to properly model processes within the Skagerrak, and for that purpose a nested version of the model is used. In the nested model, boundary values from the coarse North Sea model is used as input to a fine grid (4 x 4 km) model for the eastern North Sea, Skagerrak and Kattgat.

The circulation model is based on approximations from the three-dimensional, primitive equation, time-dependent, wind and density driven Princeton Ocean Model (POM) described by Blumberg and Mellor (1987).

At present NORWECOM only includes two functional groups (diatoms and flagellates), but work is ongoing to implement *Chattonella* spp. To introduce *Chattonella* as a single harmful species, the optimal parameterization of growth, vertical behaviour, mortality and maybe cyst dynamics are being examined, related to prognostic state variables already in the model such as: temperature, salinity, light, nutrients, turbulence, sedimentation/resuspension. This model has been used for real time forecast of the development (not the initialization) and decay (due to nutrient limitation) of observed blooms of *Chattonella*. The model is presently run operationally with daily updates of 7 days forecasts at the Norwegian Meteorological Institute, and the results are available at <http://moncoze.met.no>.

The presentations raised a number of questions:

- What are the main scientific and management questions to be answered by modelling approaches? In relation to this, how complex do we need to be in HAB dynamics/ecology? For example, is it worth the significant effort to parameterize all types of grazing losses, or are simplistic assumptions valid (e.g., mortality at 10% per day) acceptable, given the general uncertainty in the models thus far?
- What does the next trophic level need from us and us from them?
- Do available models have predictive capability?
- What knowledge and technical gaps are impeding progress towards quantification of phytoplankton?

It was concluded that:

- Modelling of single species needs to be embedded in coupled models of ocean physics and the main primary production, mainly to obtain the dynamics of nutrients and general state of eutrophication.
- The formulation of HAB dynamics should be made as simple as possible due to lack of quantitative process knowledge.
- Some predictive capability was demonstrated (what if scenarios, short term development).
- Much more effort is needed on model validation, including relevant observations.
- Knowledge of vertical behaviour is probably important for modelling success, limiting the complexity to parameterization accessible to experimental determination.
- Some formulation of growth and mortality is needed, including possible nutrient and light limitations and sensitivity to temperature (and possibly salinity and turbulence?).
- For operational purposes, hydrologic models are needed to predict river flows of freshwater. Together with flow–concentration relationships, this would provide nutrient loads. Direct atmospheric loads of nitrogen may also be important.
- Observations from satellites, ships and buoys will continue to be crucial input (assimilation/reinitialisation) for “correctly” initiating the HABs in the models. Such initiatives should benefit from the initiatives aiming at operational modelling
- More knowledge is needed on cyst dynamics of *Alexandrium*, and it is uncertain to what degree *Chattonella* spp form cysts,
- Question? Do we need a hydrosedimentary model to reproduce cyst sorting and possible movement and concentration of cyst beds? If yes, it would require annual modelling of sediment transport.

A joint (between the WGHABD and the WGPBI with invited contributions from GEOHAB) Theme Session at the ICES ASC in 2006 was suggested on: “Harmful Algae Bloom Dynamics. Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge” as in spite of large gaps of basic process knowledge around HAB dynamics, several 3D modelling initiatives are ongoing with respect to studying and predicting HABs. Therefore it is due time to couple the expertise of modellers and biologists to reveal the most urgent needs for better process knowledge to improve the predictability of models. The session aims at participation from 3D modellers and biologist interested in explaining why HABs occur, how they are initiated, how and why they develop in space and time, and why they decay.

### 5.7 Term of reference g: review biological loss processes of selected HAB species

The following abstracts outline the two presentations.

#### **Evidence of grazing on potentially ASP toxic diatoms from the genus *Pseudo-nitzschia* spp. by the naked heterotrophic dinoflagellate *Gyrodinium spirale***

*Per Andersen, Denmark*

High numbers of the heterotrophic naked dinoflagellate *Gyrodinium spirale* (5,200 cells.l<sup>-1</sup>) were observed during the declining phase of a *Pseudo-nitzschia* spp. bloom (maximum concentration > 1 million cells l<sup>-1</sup>) in October 2003 in Horsens Fjord, south western Kattegat. Observations of Lugol’s fixed samples revealed that, in many cases, the *G. spirale* cells present were deformed due to the presence of ingested *Pseudo-nitzschia* spp. inside the dinoflagellate. Fur-

thermore, bundles of *Pseudo-nitzschia* spp. in mucus were observed in the samples. The observation of ruptured cells of *G. spirale* releasing similar bundles of *Pseudo-nitzschia* spp. indicate that the free bundles of *Pseudo-nitzschia* spp. originated from *Gyrodinium spirale* present. The bloom of *Pseudo-nitzschia* spp. in Horsens Fjord terminated in October, where as blooms persisted in other localities where no *G. spirale* cells were observed) e.g. in the shallow Isefjord and did not terminate until December. The geographical and temporal variability in bloom dynamics of *Pseudo-nitzschia* spp. might to some extent be influenced by differences in grazing e.g. by heterotrophic dinoflagellates.

### **Copepodology for the phycologist with apologies to G.E. Hutchenson**

*Patricia A. Tester, USA*

*Heterocapsa triquetra* is one of the most common bloom forming dinoflagellates found in estuaries and near shore regions around the world. In order to bloom, *H. triquetra* optimizes a suite of factors including low grazing pressure, increased nutrient inputs, alternative nutrient sources, and favourable salinity and hydrodynamic conditions, as well as the negative factors of temperature-limited growth, short day lengths, and periods of transient light limitation. The prevailing environmental conditions associated its wintertime blooms are largely the result of atmospheric forcing. Low-pressure systems moved through coastal area at frequent intervals and are accompanied by low air temperatures and rainfall. Runoff following the rainfall events supplies nutrients critical for bloom initiation and development. *Heterocapsa triquetra* blooms can reach chl *a* levels  $>100 \mu\text{g} \cdot \text{l}^{-1}$  and cell densities between 1 to  $6 \times 10^6 \cdot \text{l}^{-1}$ . As the blooms develop, nutrient inputs from the river became insufficient to meet growth demand and *H. triquetra* feeds mixotrophically, reducing competition from co-occurring phytoplankton. Cloud cover associated with the low-pressure systems light limit *H. triquetra* growth as do low temperatures. More importantly though, low temperatures limit micro and macrozooplankton populations to such an extent that grazing losses are minimal.

### **5.8 Term of reference h: consider the environmental dynamics and impacts of individual phycotoxins and their metabolites enabled by new analytical technologies**

#### **Environmental Dynamics of Phycotoxins and Developments in Analytical Technology**

*Allan Cembella, Germany*

Phycotoxin dynamics in the marine environment is an important issue for the management and regulation of these toxins as dangerous natural contaminants in seafood for human consumption. The propagation of these compounds through marine food webs also has serious consequences for ecosystem stability and biodiversity, and contributes to social and economic losses of wild fish and shellfish stocks and their counterparts in aquaculture.

Phycotoxins in marine food webs are subject to concentration, “biomagnification” and metabolism after ingestion by pelagic and benthic grazers, including micrograzers (e.g. protists), macrozooplankton (e.g., copepods and herbivorous ichthyoplankton) and bivalve shellfish. The primary mechanism of toxin acquisition is via suspension- or “filter”-feeding directly upon the toxigenic plankton from the water column. Direct grazing upon epiphytic or epi-benthic toxic microalgae by herbivorous species, e.g. reef fish upon populations of the toxic dinoflagellate *Gambierdiscus* can lead to amplification of the toxicity associated with ciguatera fish poisoning (CFP) following human consumption of large carnivorous fish at higher trophic levels. Secondary accumulation of certain phycotoxins may also occur due to predation upon contaminated bivalve shellfish by gastropods, such as carnivorous whelks.

Biotransformation processes, typically in the digestive tract of planktivorous species, can have a profound effect on the molecular structure and hence the toxicity of ingested phycotoxins. In bivalve molluscs, where the phenomenon is best understood, ingestion of phycotoxigenic organisms is followed by digestion of the cells and liberation of toxins into the gut lumen where they are acted upon by digestive enzymes and other physico-chemical reactions. Time-dependent translocation of modified toxins from the hepatopancreas into other tissues, including the gills, mantle, siphon and foot are frequently observed in bivalve species. In the case of the PSP toxins, for example, liberation of the dinoflagellate toxins may result in epimerization (of  $\beta$ - to  $\alpha$ -epimers), conversion of the low potency N-sulfocarbamoyl toxins (C- and B-toxins) to highly toxic carbamate derivatives, and in a few species of clams, decarbamoylation of the carbamate toxins to decarbamoyl derivatives. These bioconversion processes cannot be strictly considered as detoxification mechanisms since certain catabolic reactions may in fact yield a massive increase in specific molar toxicity.

Oxidative biotransformation of the compounds associated with CPF from precursors such as gambieric acid and gambierol synthesized by the dinoflagellate to ciguatoxin derivatives in fish tissues is now well described. Similarly, metabolic conversion of derivatives of the yessotoxin (YTX) and pectenotoxin (PTX) complexes in bivalve molluscs, such as the production of pectenotoxin-2-seco acid (PTX2-sa) from PTX2 synthesized by dinoflagellates *Dinophysis* has also been recently described. The epibenthic dinoflagellate *Prorocentrum lima* typically synthesizes relatively water-soluble dinophysistoxin (DTX) derivatives, such as sulphated DTX4 and diol-esters, which are then subject to esterase degradation to more lipophilic (and potent) derivatives, including okadaic acid (OA) and acylated compounds.

Major advances in the detection and quantitation of phycotoxins have made possible the elucidation of biosynthetic and degradative pathways in causative and vector organisms, leading to a dynamic view of toxin transfer in marine food webs. The structural elucidation of saxitoxin, the first described phycotoxin, as a tetrahydropurine compound associated with paralytic shellfish poisoning (PSP) several decades ago was a major breakthrough in our understanding. Classical techniques of natural products chemistry applied to phycotoxin isolation include liquid-liquid partitioning,

fractionation guided by spectrophotometry (UV-visible) or fluorescence, and separation by open column (low-pressure) chromatography and thin-layer chromatography (TLC). The subsequent discovery of the causative agents of most of the major phycotoxin syndromes (e.g., PSP, DSP, NSP, ASP, azaspiracid poisoning, spirolide toxicity) have also been supported by bioassay guided fractionation, especially whole animal bioassays involving laboratory mice.

The rapid increase in the number of novel phycotoxins has occurred in parallel with the advent of new technological developments for toxin isolation and analysis. Indeed there is abundant evidence to suggest that this apparent proliferation of phycotoxins is primarily a reflection of our ability to access and apply sophisticated analytical methods to the detection and characterization of these compounds, even when present in trace amounts (sub-picomolar), and not an actual increase in the number of naturally occurring phycotoxins. For example, in Atlantic Canada prior to 1987, PSP toxicity was the only known toxin syndrome. In 1987, the occurrence of a major outbreak of amnesic shellfish poisoning (ASP) was rapidly linked to the presence of the neurotoxin domoic acid, identified as the causative agent by a combination of bioassay-guided fractionation, preparative and analytical high-performance liquid chromatography (HPLC), liquid chromatography with detection by mass spectrometry (LC-MS) and nuclear magnetic resonance (NMR). The subsequent implementation of high sensitivity tandem LC-MS/MS with high resolution NMR has now led to the discovery of a host of “new” toxins in Atlantic in various microalgal and seafood species, which now includes novel PSP and DSP toxins, PTX analogues and spirolides.

In the early 1980s, the introduction of HPLC coupled with on-line fluorescence detection (FD) made possible the analysis of the major PSP derivatives and certain of the DSP toxins (e.g., OA and DTX1) by employing post-column oxidation and pre-column fluorescence derivatization, respectively. For the first time, the systematic analysis of phycotoxin profiles from biological matrices was incorporated into routine procedures and widely applied. Unfortunately, these methods were limited by the availability of certified analytical standards for calibration and hampered by the fact that these chromatographic techniques do not yield *confirmatory* analyses (although often erroneously misused to do so!).

During the mid-1980s, the application of a number of alternative ionization interfaces for MS systems, including fast-atom bombardment (FAB), chemical ionization (CI), and electro-spray ionization (ESI) contributed to the rapid improvement in structural elucidation and quantitation of phycotoxins. Modern LC-MS systems are often multi-sector (MS/MS/MS), providing additional information from the fragmentation patterns. Perhaps the greatest technological advance in phycotoxin analysis was the introduction of atmospheric pressure ionization (API) and ion-spray (ISP) interfaces in the late 1980s. This made possible the analysis of virtually all of the relatively low molecular weight phycotoxins (almost all are <30,000 Da) from biological and seawater matrices. This innovation was accompanied by improvements in column technology, including new stationary phase material and development of microbore columns. Mass spectrometry offers significant advantages over other analytical and assay methods in that molecular weight and elemental composition (HRMS), as well as structural information (MS/MS) are provided. The technique may be applied to a wide range of analytes and the methods are both universal *and* selective. High sensitivity and excellent quantitation may be achieved even in automated analysis. The developments in LC-MS have contributed greatly to our understanding of the dynamics of novel and known phycotoxins, including their analogues and metabolites. Improvements in sensitivity are now to the point that several of the toxin groups can be quantified from individual cells after micro-extraction. Multiple toxin determinations (>30 analogues of various toxin groups) can be achieved in a single injection onto the column with multi-dimensional chromatography by LC-MS. Further innovations in separation technologies and sample preparation (e.g., micro-extraction) are expected to provide even better sensitivity, resolution and precision for phycotoxin analysis in the near future.

## 5.9 Term of reference i: report and discuss new findings

To support discussions of the status of coupled physical-biological models for HABs (ToR f) as well as the “New Findings” portion of the meeting, Don Anderson gave the following presentation on *Alexandrium* bloom dynamics for the Gulf of Maine, emphasizing the critical role played by numerical models in interpreting and analyzing observations.

A conceptual model of *A. fundyense* dynamics in the Gulf of Maine was proposed and is described in more detail in Anderson *et al* (submitted) and McGillicuddy *et al* (submitted). An important consideration that this model must accommodate is the general east to west flow of the Maine Coastal Current system and the mean alongshore flow of the Gulf of Maine. Average conditions therefore are, in effect, a one-way transport system that will move *A. fundyense* cells to the west and south towards Georges Bank, with the mean flow providing limited opportunity for those cells to circulate back into the northeast portion of the domain. Accordingly, if blooms begin with germinating cysts from specific seedbeds, as is hypothesized, the replenishment of those seedbeds with new cysts cannot be from the cells that have been transported away. The dilemma is that a system in which there is significant alongshore transport must also must have features that allow *A. fundyense* motile cell populations to accumulate and deposit cysts at the “upstream” end of the transport pathway.

In this context, the critical component is the population that develops near Grand Manan Island at the mouth of the Bay of Fundy (BOF), eastern Canada. It is well established that there is an eddy system that retains cells to the east of Grand Manan Island, resulting in exceptionally high *Alexandrium* cell concentrations, sometimes exceeding 60,000 cells<sup>-1</sup>. In the conceptual model, this area serves as the “incubator” for the region, with many cells remaining within the Bay of Fundy and completing their life history there, depositing new cysts at the end of the bloom season, and creating the seedbed at the mouth of the BOF that was mapped in the ECOHAB-GOM program (Anderson *et al.*, submitted).



This seedbed is a persistent feature that has been mapped on numerous occasions over the last 20 years. There are thus always benthic cysts present in the Bay of Fundy to initiate blooms in that area.

The BOF blooms are not completely isolated, however, as outflow from the Bay of Fundy will carry cells into the Maine Coastal Current system via its eastern branch, called the EMCC (Eastern Maine Coastal Current). This linkage has been depicted in surveys conducted by Martin and White (1988) and Townsend *et al.* (2001). As hypothesized by the latter authors, *A. fundyense* cells that enter the EMCC near the BOF do not initially flourish, due to the deep mixing and high turbulence of that water mass. However, as the cells are transported to the west, the water stratifies and allows growth (Townsend *et al.*, 2001). Model simulations of this *A. fundyense* population that include a nutrient dependence (McGillicuddy *et al.*, submitted) show nutrient limitation at the western edge of the EMCC in the mid-summer months. Based on laboratory studies (e.g. Anderson and Lindquist, 1985) nutrient limitation will result in the induction of sexuality in *A. fundyense* populations, leading to the formation of resting cysts that will then fall from the water column. Significant cyst accumulations offshore of Penobscot and Casco Bays are found in the general area where model results suggest nutrient limitation will occur (McGillicuddy *et al.*, submitted), and where Townsend *et al.* (2001) showed abundant populations of *A. fundyense* during large-scale surveys.

The *A. fundyense* populations that cause PSP problems in the western Gulf of Maine region have two possible origins. One is from motile cells delivered to the nearshore waters of the WGOM from the EMCC. The other is from the germination of cysts from both the inshore and offshore cyst seedbeds that have been mapped out in that region (Anderson *et al.*, submitted). The inshore cysts are not abundant, but may be responsible for the localized blooms and PSP outbreaks that occur within certain estuaries and sounds – as in Lumbo’s Hole and Cundy’s Harbor in northeastern Casco Bay. These blooms may be self-seeding as well as propagatory, analogous to the Bay of Fundy blooms described above, although the extent to which cells from these blooms are entrained into the WMCC is not known. The offshore cysts are another potentially important source of inoculum cells for the western Gulf of Maine, as a net shoreward flux of *A. fundyense* cells can arise from the joint effects of upward swimming and the time-dependent response of buoyant river plumes to fluctuating wind conditions (McGillicuddy *et al.* (2003). These are the cysts that presumably originated from the EMCC populations, which in turn originated from the Bay of Fundy “incubator” blooms. At least a one-year time lag is involved in this sequence, since the cysts that are deposited in the offshore Penobscot/Casco Bays seedbed from the EMCC would need to over winter before they would mature and be able to initiate blooms in the WMCC in subsequent years (Anderson 1980). The ultimate scenario is thus of cysts that germinate within the BOF seedbed, causing localized, recurrent blooms to the east of Grand Manan Island that are self-seeding as well as propagatory in nature, supplying cells that populate the EMCC. Some EMCC cells are entrained into western Maine waters, while others eventually deposit cysts offshore of Penobscot and Casco Bays. In subsequent years, these cysts serve as a seed population for the western Maine blooms that are transported to the south and west by the WMCC, causing toxicity along the coasts of western Maine, New Hampshire, and Massachusetts before they are either lost due to mortality or advected out of the region. Without the localized, incubator characteristic of the eddy system near Grand Manan Island, one would expect *A. fundyense* populations in the Gulf of Maine to diminish through time and the PSP problem to disappear. Since PSP has been a persistent problem in the region for a century or more argues for the effectiveness and stability of the conceptual model described here.

## References:

- Anderson, D. M. (1980). The effects of temperature conditioning on the development and germination of *Gonyaulax tamarensis* (Dinophyceae) hypnozygotes. *Journal of Phycology* 16: 166–172.
- Anderson, D.M., C. Stock, B. A. Keafer, A. Bronzino, D. J. McGillicuddy, M. Keller, B. Thompson, P. A. Matrai, J.L. Martin. (Submitted). Experimental observations and modeling of *Alexandrium fundyense* cyst dynamics in the Gulf of Maine. *Deep Sea Research II*.
- Anderson, D. M. and N. L. Lindquist (1985). Time-course measurements of phosphorus depletion and cyst formation in the dinoflagellate *Gonyaulax tamarensis* Lebour. *Journal of Experimental Marine Biology and Ecology* 86: 1–13.
- Martin, J.L., and A. White, (1988). Distribution and abundance of the toxic dinoflagellate *Gonyaulax excavata* in the Bay of Fundy. *Canadian Journal of Fisheries and Aquatic Sciences* 45, 1968–1975.
- McGillicuddy, D. J. Jr., D.M. Anderson, D.R. Lynch, and D.W. Townsend. (Submitted). Mechanisms regulating the large-scale seasonal fluctuations in *Alexandrium fundyense* populations in the Gulf of Maine. *Deep Sea Research II*.
- McGillicuddy, D.J., Jr., R.P. Signell, C.A. Stock, B.A. Keafer, M.D. Keller, R.D. Hetland, and D.M. Anderson (2003). A mechanism for offshore initiation of harmful algal blooms in the coastal Gulf of Maine. *J. Plankt. Res.* 25(9): 1131–1138.
- Townsend, D. W., Pettigrew, N. R. and Thomas, A. C., (2001). Offshore blooms of the red tide dinoflagellate, *Alexandrium* sp., in the Gulf of Maine. *Cont. Shelf Res.* 21, 347–369.

## Observations of shellfish toxins in Norwegian shellfish

Einar Dahl, Norway

The first confirmed occurrence of spirolides in mussels and plankton (from material collected in both 2002 and 2003) from Norway was observed in 2003. Analysis of Norwegian mussel extracts using liquid chromatography-tandem mass spectrometry (LC-MS/MS) according to Quilliam *et al.* (2002), revealed the presence of several spirolides. The same compounds were also found in algal samples dominated by *Alexandrium ostenfeldii*. One new spirolide, 20-methyl spirolide, was identified.

Two novel pectenotoxins, 36S-PTX-12 and 36R-PTX-12, were detected by LC-MS in a solid-phase extract from net hauls taken at Flødevigen, Norway, in June 2002 that were dominated by *Dinophysis acuminata* and *D. norvegica*. Analyses of shellfish extracts revealed that PTX-12 accumulated in Norwegian blue mussels (*Mytilus edulis*) and cockles (*Cerastoderma edule*), along with PTX-12 seco acids occurring as a complex mixture of diastereo-isomers. Analysis of algal cells picked from the net haul revealed that PTX-12 predominated in *D. acuta* and *D. norvegica*, whereas PTX-2 was the predominant pectenotoxin in *D. acuminata*.

Picked cells of *Protoceratium reticulatum* collected from five locations in Norway were shown by ELISA analysis to contain yessotoxins (YTXs). The production of yessotoxin (YTX) was verified by culturing followed by LC-MS analysis of one of the Norwegian isolates. This is the first report of the biogenic origin of yessotoxins in Norway. The sensitivity of the ELISA method made it possible to quantify YTXs in algal cultures, net-hauls, and in single cells of *P. reticulatum*. The cells picked from cultures and net-hauls contained 18–79 pg YTXs per cell. The sensitivity of this method makes it possible to search for other possible producers of YTXs.

### References:

Quilliam, M, Hess, P. and Del'Aversano, C. (2002). Recent development in the analysis of phycotoxins by liquid chromatography-mass spectrometry. pp. 383–391 in: *Mucotoxins and Phycotoxins in Perspective at the Turn of the Millennium, Proceedings of the Xth International IUPAC Symposium on Mycotoxins and Phycotoxins*. Edited by W.J.De Koe, R.A. Samson, H.P. van Egmond, J. Gilbert and M. Sabino. Wageningen, The Netherlands.

Tom Osborn presented a new digital system to record holograms of plankton *in situ* (due to Malkiel *et al.*) Each hologram provides a 3-dimensional snapshot of the particle field. They can resolve spherical particles as small as 5 micron and linear characteristics (such as setae) with diameters as small as micron. The movies of untethered individual copepods swimming and feeding held the audience spellbound. They are also able to reconstruct the 3D flow field in the sample volume. The ability to watch an individual, with sufficient resolution to see the motions of the feeding appendages of a copepod, combined with reconstructed flow field, and the distribution of preyparticles will have an enormous impact on studies of plankton of all types. Further details can be found at <http://www.me.jhu.edu/~lefd/shc/shc.htm> and in:

Malkiel, E., Sheng, J., Katz, J. and Strickler, J. R. (2003). The three-dimensional flow field generated by a feeding calanoid copepod measured using digital holography. *J. Exp. Biol.* 206, 3657–3666.

## Influences of monsoons and oceanographic processes on red tides in Hong Kong waters

Kedong Yin, China

Hong Kong waters in the northern part of the South China Sea experience seasonal oceanographic processes due to monsoon winds, and the Pearl River outflow. Several hundred red tides have occurred in Hong Kong waters during 1983–2001, and show a clear spatial and temporal distribution: Most (74%) occurred in northeastern semi-enclosed bays away from the Pearl River estuary with fewer occurring in the western estuarine-influenced waters. Most red tides (70%) occurred between December and May, with less in summer. Nutrient levels are high in the Pearl River estuary, whereas nutrients in the northeast semi-enclosed bays (Mirs Bay and Port Shelter) are generally low and cannot support high biomass red tides. This suggests that concentrating mechanisms exist promoting the formation of red tides in these northeastern waters, either by vertical migration or horizontal aggregation under suitable wind directions and speeds. East to northeast winds and a moderate wind speed ( $6 \text{ m s}^{-1}$ ) appear to be most favourable. The prevailing northeast monsoon winds in winter and spring result in downwelling. As a result, the residence time of waters in these semi-enclosed bays is longer and becomes more like a batch culture, allowing local inputs of nutrients and vertical migration of phytoplankton in shallow waters to play a dominant role in favour of local red tides. This may be one of the reasons why more red tides occur in winter and spring in semi-enclosed waters. During El Niño 1997–1998, the South China Sea warm waters appeared to flow against the southern China coast. Thus, the southern coastal waters had to be downwelling against the heavier South China Sea water at the bottom, which resulted in a trapping effect on the China coastal waters. This amplified the downwelling effect and hence, more red tides occurred in 1998. In summer, the southwest monsoon winds result in upwelling along the coast and high river discharge and rainfall cause an increased estuarine circulation in the Pearl River estuary and rapid outflow of the surface water from these semi-enclosed waters. As a result, the residence time of these waters decreases, and they may be analogous to semi-continuous or continuous cultures. This may explain why there are fewer red tides in summer.

The 6 most frequently-occurring species are (in descending order) *Noctiluca scintillans*, *Gonyaulax polygramma*, *Skeletonema costatum*, *Mesodinium rubrum*, *Prorocentrum minimum*, and *Ceratium furca*. In general, dinoflagellate red tides occur mostly in April when silicate is low whereas diatom red tides occur in June when Si increases due to the freshwater discharge. The deep oceanic water on the continental shelf is drawn into semi-enclosed bays during the upwelling and it is nutrient poor, which does not favour the nutrient acquisition in deep water by vertical migrating dinoflagellates.

### **Epimerization of PSP toxins**

*Bernd Luckas, Germany*

Bernd Luckas reported on problems concerning PSP analyses with the contribution “Epimerization of PSP-Toxins – A source of false estimation of PSP results”. In summary, it can be stated that the protocol usually applied for PSP determination consists of homogenization and extraction of a sample followed by direct injection of an aliquot of the micro-filtrated extract into the HPLC apparatus without any enrichment steps. However, PSP toxins are often present in very low concentrations, e.g. in water or media from algal cultivation. In such cases, a step to enrich the PSP concentration is required before chromatographic separation. To accomplish this enrichment, adsorption of PSP toxins onto a solid-phase extraction (SPE) cartridge, followed by elution of the toxins with dilute acetic acid, and evaporation of the eluate is recommended. Nevertheless, the application of SPE can lead to incorrect results regarding PSP content and profile, respectively. The evaporation of solutions with very low PSP concentrations results in low recoveries for PSP toxins due to degradation during this enrichment step. In addition, changes in PSP profiles in the course of evaporation of acid PSP containing solutions are observed. Specifically, the conversion of  $\beta$ - to  $\alpha$ -epimers results in a shift in the characteristic epimeric ratios. Such shifts in epimeric ratios are particularly likely to occur in very dilute PSP-toxin containing solutions, especially when subject to a change in temperature and/or pH. Though HPLC/FLD and LC/MS-MS methods normally applied for PSP determination give correct results reflecting the actual PSP toxin profile of a sample, the evaluation of data concerning PSP pattern of water samples or media for algal cultivation must be considered with care.

#### **5.10 Term of reference j: start preparations to summarise the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006**

The WG welcomed this TOR and established a task group with representatives from the North Sea countries (Sweden, Norway, Denmark, Netherlands, Germany and UK) to address this topic further. A meeting of this task group has been preliminary arranged for the 8<sup>th</sup>/9<sup>th</sup> September 2004 to allow members to assemble their data. The target species for examination selected for examination are *Phaeocystis*, *Alexandrium minutum*, *A. ostenfeldi*, *A. tamarense*, *Karenia miki-motoi*, *Chatonella* spp., *Noctiluca* spp. *Dinophysis acuta*, *D. norvegica*, *D. acuminata*, *Chysochromulina* spp., *Pseudonitzschia* spp., *Pseudogonyaulax*, *Heterosigma*, *Fibrocapsa*, *Dictyocha*, *Chaetoceros* spp. The team will examine the existence of trends in the occurrence of these species and if this could be due to anthropogenic forcing. The potential of the data to describe the dynamics of the natural harmful algal blooms including their natural variability will be studied. The use of the occurrence of harmful algae species as indicators of ecological quality will also be examined. The potential output from this study will be a scientific useful document which can take the form of an ICES general report and a deliverable product to the WG/HABD.

## **6 NATIONAL REPORTS**

---

### **Belgium**

Thus far, Belgium does not fund any HAB monitoring programme, because no commercial-level marine aquaculture is developed in Belgian coastal waters. Therefore, HAB monitoring is not considered a priority at the national level.

## Canada

### Salmon Mortalities:

#### West Coast

*Dictyocha speculum* was responsible for water discolouration and salmon mortalities in the mid-east Vancouver Island and west Vancouver Island areas when cell concentrations reached 960,000 and 1,500,000 cells l<sup>-1</sup> respectively during July/August. *Heterosigma akashiwo* caused mortalities on two separate occasions in the west Vancouver Island region when water became discoloured in the last week of July/first week of August (30,000,000 cells l<sup>-1</sup>) and again in late August through late September. *Chaetoceros concavicornis* (15,000 cells l<sup>-1</sup>) and *Chaetoceros convolutus* (10,000 cells l<sup>-1</sup>) were responsible for salmon mortalities in the east Vancouver Island between Sept. 18-21.

#### East Coast

Salmon mortalities were associated with a bloom of *Alexandrium fundyense* in the Grand Manan region of the Bay of Fundy that persisted through the month of September. Concentrations in excess of 400,000 cells l<sup>-1</sup> were measured and water discolouration was observed.

### PSP

Closures similar to most years occurred on the west coast (British Columbia) and the east coast (St. Lawrence, eastern Nova Scotia). Shellfish toxicity was higher than normal in the Bay of Fundy and extended into September and was associated with concentrations higher than 400,000 cells l<sup>-1</sup>. Western and southwestern Prince Edward Island had closures for PSP in June

### ASP

Domoic acid was detected in shellfish in the northern peninsula of Newfoundland in June and closures remain in effect.

## Denmark

In Denmark, as in 2002, no incidences of PSP or ASP were observed. DSP was observed in concentrations above regulatory limits locally in the Isefjord; in a few harvesting areas in the south western part of Kattegat as well as in Flensborg Fjord in the western Baltic. No DSP was registered in the major shellfish harvest areas in the Limfjord. DSP toxicity was observed in January-February as a result of low concentrations of *Dinophysis acuta* remaining after the autumn-bloom from 2003. During late summer and winter the DSP toxicity was most likely caused by *D. acuta*, occurring in rather low concentrations 100-400 cells /l. The toxicity continued into 2004 until February when the diatom spring bloom occurred. The timing and dynamics of the DSP incident in 2003 is very similar to that observed in 2002 in Danish waters.

High concentrations of *Pseudo-nitzschia* spp. were registered during summer in the Limfjord as well as in the Kattegat area, and harvesting areas were closed/opened under intensified monitoring. No ASP was observed during these blooms.

There were no fish kills caused by HABs during 2003. *Chattonella* spp. was registered in relative low concentrations during spring and early summer.

During summer high biomasses of phytoplankton (dominated by several *Ceratium* species and diatoms from the genus *Pseudo-nitzschia*) were observed. This became one of the triggering factors leading to oxygen deficiency in the major part of Kattegat. During the period with oxygen deficiency fish and invertebrate kills were observed.

## Estonia

Biomasses of cyanobacteria were surprisingly low in the Gulf of Finland along the Estonian coast in summer 2003. The excess phosphorus measured in winter would have favoured the formation of intensive summer blooms if the temperature conditions were appropriate. The weather in July was warm and sunny but the continuous upwelling along the Estonian coast was effective in preventing the development of cyanobacterial blooms. A moderate bloom was observed in the central part of the Gulf of Finland. In the beginning of August a very heavy *Nodularia* bloom was detected in the Väinameri area, especially south of Island Hiiumaa in the Kassari Bay. Cyanobacterial biomass was not measured but the accumulations were very dense and washed ashore over an area covering a number of kilometres. Toxin analyses of water taken on 4 August gave results between 28-220 µg l<sup>-1</sup> of nodularin. The *Nodularia* bloom was also detected west from Island Saaremaa.

## France

*Dinophysis acuminata* was observed in the Channel and Atlantic and *D. sacculus* in the Mediterranean. *Alexandrium minutum* was observed in Brittany and *A. catenella* and *A. tamarense* observed in the Thau Lagoon and Mediterranean. Research of DSP, PSP and ASP toxins in scallops (especially *Pecten maximus*, and sometimes *Chlamys varia*) from the main production areas was performed. Few samples were analysed at the beginning of the year in Seine bay, with more regular sampling since October in Seine Bay, St Brieuc Bay, and Quiberon bay. No toxins were observed. Specific studies on DSP toxins, with LC / MS-MS analysis, have showed (2002 – 2003):

- presence of AO + DTX3 in all affected sites, linked to *Dinophysis acuminata*,
- presence of DTX2 + DTX3 in the sites where *Dinophysis acuta* was observed,
- absence of DTX1, PTX2 and AZAs in every sites

## Germany

### North Sea:

No exceptional booms of *Phaeocystis globosa* (Prymnesiophyceae), or *Noctiluca scintillans* (Dinophyceae) were recorded. Local red water discolorations were reported, without any harmful effects. Cells of the potentially toxic dinoflagellate species *Alexandrium tamarense* and *A. ostenfeldii* as well as *Dinophysis acuminata*, *D. acuta* and *D. norvegica* occurred in low numbers. Toxin content of mussels were below critical values. Species of the potentially toxic diatoms *Pseudo-nitzschia*, e.g., *P. delicatissima*, *P. pseudodelicatissima* and of the *P. pungens-multiseriis* – complex were regularly observed but no harmful effect recorded. The same applies for the Raphidophyceae: *Fibrocapsa japonica*, *Heterosigma akashiwo* and *Chattonella* spp. have been recorded but no harmful effect observed.

### Baltic Sea:

Cyanobacteria were again reported in the Baltic Sea. The summer 2003 was characterized by stable weather conditions like sunny weather and mainly low wind strength, mainly from easterly and southerly direction. Therefore the accumulation process of the filamentous bluegreen species at sea surface was a more or less continuous process. Observed “algal fields” were of relatively big size. The origin of bluegreen development is usually more located in the central Baltic Sea and with Baltic current the accumulations are drifting into direction of the western Baltic Sea – a annually recurrent phenomenon as well as in 2003.

Depending on wind force and direction the bluegreens can reach the bathing waters. In 2003 the first turbidness of the water caused by accumulations of bluegreens, was detected by the coast guards end of July in the area of outer Lübeck Bight as well as of the Fehmarnsund. Within a few days observations during a helicopter flight showed clearly, that the south-easterly winds caused the drifting of bluegreen algal accumulations from Mecklenburg Bight directly into the outer and middle part of the Lübeck Bight. At that time some of the algal fields already reached the bathing waters of the outer part of Lübeck Bight. Within the next 10 days, accumulations were observed along the coast of Schleswig-Holstein up to Kiel Fjord. Flocky accumulations, which caused visible and conspicuous turbidness, were floating along the water current into the middle part of Kiel Bight as Flensburg Fjord.

During the weekend of 9–10 August the weather was still sunny, but combined with strong and squally wind from eastern direction, causing a mixing of the upper water column. A helicopter flight was conducted on August 11th. that showed a clear decrease in algal accumulations in the area of Lübeck Bight, but still some small algal fields were observed there. From other areas of coast no observations of accumulation were reported. On 14 August, the wind direction turned to the West and within a few days, the accumulations were destroyed completely.

Daily information was spread via the media to inform the public (current information also available under [www.algenreport.de](http://www.algenreport.de)). Bathing was prohibited on a few occasions for several hours per day or for 1-2 days only for a few beaches, for instance in front of Heiligenhafen and of Großenbrode, as a precaution, decided by local mayor and/or sea rescue team and/or health agency. Because of changing wind directions during the day the occurrence of “algal fields” was changing very fast, the prohibition lasted only that short time in general. Toxin analyses have not been performed on these samples.

### Southern and Central Baltic Sea:

Observations on a cruise from 25–31. July 2003 (Wasmund, IOW) and Toxin Analyses (Bernd Luckas)

During a cruise from Mecklenburger Bucht to Bornholm-Sea and Gotland-Sea 12 samples were taken from large blooms of Cyanobacteria – mainly *Nodularia*. Toxin analyses revealed contents of Nodularin in all 12 samples, ranging from about 150 to 880 µg Nodularin per liter filtrated seawater. Toxic effects have not been reported.

## Ireland

### Ireland – South West

Shellfish Toxicity, predominantly Okadaic Acid + DTX 2 was observed to occur mainly in the Southwest for 2003 (all *M. edulis*). There was a small amount of toxicity carryover from 2002 in samples from Bantry Bay during Jan 2003, however these levels were just below the regulatory limit of 0.16µg/g Total Tissue, and were observed to further decrease during February and March, with all areas in the South-West <0.01µg/g Total Tissue in April.

Levels of OA + DTX-2 were observed to gradually increase in Kenmare and Bantry Bay from May, with the first positive samples being recorded in Kenmare (Kilmakilloge and Tahilla) in June. This coincided with the observation of *Dinophysis* spp. in these areas, with levels increasing gradually throughout May and June to the highest levels recorded in July (1560 cells.l<sup>-1</sup> *Dinophysis acuminata* in Bantry Bay). This was followed by positive *M.edulis* samples observed in all parts of Bantry Bay in July – September, with the highest levels of OA+DTX 2 recorded in Outer Bantry Bay, Castletownbere (0.55µg/g Total Tissue) and Inner Bantry Bay, Snavel (0.44µg/g Total Tissue) in July. Low levels of *Dinophysis* spp. persisted in Bantry Bay throughout August – September. OA+DTX 2 levels were observed to gradually decrease from September, with the majority of areas in Bantry Bay re-opening in October, and remainder opening in November. No further positive samples were observed above the regulatory limit for the rest of 2003.

Generally the levels of Azaspiracids (AZA's 1,2,3) observed in the South West were low for 2003, with the highest levels observed in September in Cleandra, Kenmare Bay (0.1 µg/g Total Tissue) and Bantry Bay, Castletownbere (0.14 µg/g Total Tissue).

No PSP toxins above the regulatory limit were detected in samples from the Southwest with low levels of *Alexandrium* spp. observed.

The highest levels observed for *Noctiluca scintillans* for Ireland for 2003 were observed in Bantry Bay in August (9880 cells.l<sup>-1</sup>).

The highest levels observed for *Phaeocystis* sp. for Ireland for 2003 were observed in Castlemaine Harbour (16500000 cells.l<sup>-1</sup>) and Bantry Bay (1000000 cells.l<sup>-1</sup>) in April.

### DTX-3 Analysis

Retrospective DTX-3 analysis was conducted on samples predominantly from the Southwest. Highest observed DTX-3 levels were recorded for Castletownbere in August (0.52µg/g Total Tissue). This gave a combined total of OA+DTX 2 and 3 maximum of 0.95 µg/g Total Tissue.

Of all samples analysed for the presence of DTX-3, it was found that where DTX-3 was detected, it accounted for on average 38.7% of total DSP toxicity of the sample.

### Ireland – North West

Positive Bioassays with OA+DTX 2 levels just below the regulatory limit were observed in samples of *M.edulis* in Killybegs Harbour in July and August 2003. All other areas sampled were negative.

Highest levels of *Dinophysis acuminata* observed in Killybegs were recorded in June (240 cells.l<sup>-1</sup>). Highest levels of *Dinophysis acuminata* (680 cells.l<sup>-1</sup>) observed in the Northwest was in June in Inverin.

Azaspiracids in *M.edulis* samples were observed to be above the regulatory limit in Bruckless, Co. Donegal in September (max level observed 0.17µg/g total tissue) and Inverin, Co. Galway in October (max level observed 0.19µg/g total tissue). All other areas sampled were below the regulatory limit of 0.16µg/g. total tissue. No PSP toxins above the regulatory limit were detected in samples from the Northwest

PSP toxins were detected in *M. edulis* and *C. gigas* Cork Harbour in September and were Positive under AOAC Bioassay. *Alexandrium* spp. were also observed to present in the area during the same time period at a max. level of 15010 cells. l<sup>-1</sup>. Highest levels of *Alexandrium* spp. were recorded in Kinsale 19160 cells. l<sup>-1</sup>) in September.

### ASP Summary 2003

Highest levels of domoic and epi-domoic acid in the gonad tissues of *Pecten maximus* in offshore areas were observed in Wexford Ground (VIIa ICES Rectangle 33-E3) in June and November with max levels observed of 48.1µg/g domoic acid. In inshore areas, the highest levels observed in gonad tissues were in Portmagee Channel in December (50.2µg/g domoic acid) and levels were observed to be above the regulatory limit in Clew Bay South in June and July (24.9µg/g domoic acid).

For inshore areas the highest levels of Domoic acid in the remainder tissue were observed in Clew Bay South from April to July (max. 300µg/g Domoic acid in June). The highest levels observed in the total tissue were observed at the same time in the same area (144µg/g Domoic acid).

Whole flesh samples of *M. edulis* and *C. gigas* analysed for the presence of domoic and epi-domoic acid were all observed to below the regulatory limit of 20µg/g domoic acid).

## Latvia

Taxonomical HAB studies were performed in the frame of complex marine monitoring studies in the Gulf of Riga (Eastern Baltic Sea) conducted in 2003 according to the requirements of the Latvian national monitoring program and in line with the recommendations of Helsinki Commission. Eco-toxicological HAB studies were carried out in the frame of projects of Latvian Council of Science.

Phytoplankton samples were collected from January till December. The presence of HAB species, their abundance and the presence of hepatotoxins (in isolated cases) were detected.

No harmful algal blooms were observed in the Gulf of Riga (Eastern Baltic) in 2003. Totally ten HAB species representing 4 taxonomical groups were detected: 5 species of Cyanobacteria (mostly nitrogen fixing species), 3 - Dinoflagellates, 1 - Diatoms, 1 - Prymnesiophyta.

For most HAB species the maximal abundance was observed in summer (July – August), except *Prorocentrum minimum* and *Chaetoceros danicus*, which reached max values in autumn (October - November). According to density of cells maximum values showed representatives of the following genera *Aphanizomenon*, *Microcystis* and *Chrysochromulina* (small cells species); according to the HAB biomass (biovolume) prevailing genera were *Aphanizomenon*, *Nodularia* and *Dinophysis*. Nitrogen fixing cyanobacteria *Nodularia spumigena* and *Aphanizomenon sp.* reached the highest biomass in the open part of the gulf (210 mg/m<sup>3</sup> and 34 mg/m<sup>3</sup> respectively) while *Dinophysis acuminata* – in the southern part of the coastal zone (77 mg/m<sup>3</sup>).

Overall in 2003 HABs were less than previous years. The total phytoplankton biomass in the summer was near the parameters measured in years of low productivity (1999 and 2001) and lower than average long-term values. That could be explained by a decrease in annual nitrate and phosphorus concentrations linked with a simultaneous increase of silicate. Under average long-term values were also observed in the intensity of cyanobacterial blooms (mainly due to the windy weather and lasting water stratification, disturbing inflow of nutrients from deeper water layers). The highest biomass of *Aphanizomenon flos-aque* was observed at the end of July, but nevertheless its values were below the average indices in 1997–2002. No harmful or toxic events were observed.

Culture collections of HAB species were maintained in the laboratory. New strains of *Nodularia spumigena*, *Microcystis spp.* and *Anabaena spp.* were isolated from Latvian territorial waters and toxicological experiments with zooplankton organisms and fish larvae were carried out.

## The Netherlands

In 2003 there were only some potential threats of HABs in the Dutch North Sea. A bloom of *Phaeocystis* occurred, but due to the prevailing wind direction this did not result in any harmful effects. In the Dutch part of the Wadden Sea *Dinophysis acuminata* occurred for a brief period of time but no toxicity was observed.

### North Sea:

In May 2001 a massive *Phaeocystis*-bloom resulted in oxygen depletion in Lake Grevelingen. Strong indications are present that this bloom resulted in mussel mortalities in the Oosterschelde estuary (Louis Peperzak, HAB2002, Florida, USA). This triggered the monitoring in the Voordelta region (North Sea area West of South Western part of the Netherlands). RIKZ took water samples on a daily basis during high tide. The samples were analysed with use of a flowcytometer on a weekly basis for *Phaeocystis*-cell counts.

In week 16 the *Phaeocystis*-concentration was elevated to around 100 million cells per liter (comparable to 2001). Prior to the elevation of cell numbers the mussel cultivators were informed so that they could relocate their shellfish (if desirable). In the weekend of 19–20 April weather forecasts predicted winds coming from northern directions. This would blow the *Phaeocystis* bloom into the Estuaries and Lakes (with open water locks). The water locks were therefore closed, to avoid the bloom entering these areas.

Both microscopic and flowcytometer investigation of samples from both the Oosterschelde and the entrance of Lake Grevelingen on Thursday 24 April revealed that the colonies were rupturing into loose cells. This indicated the end of the *Phaeocystis* bloom. The water locks were opened and the potential harmful event was at an end.

### Wadden Sea:

Species of the potentially toxic diatoms *Pseudo-nitzschia*, e.g., *P. delicatissima*, *P. pseudodelicatissima* and of the *P. pungens-multiseriis* – complex were regularly observed but no harmful effects recorded. Cell counts ranged from 10,000-60,000 cells per litre over the period March through November, with the peak in September. These cell counts were only observed in the Wadden Sea, at this moment no data is available for the North Sea, since no active monitoring was carried out in this period. The presence of *Pseudo-nitzschia* species did not result in any toxic events.

*Dinophysis acuminata* was observed in the Dutch part of the Wadden Sea in July, at cell counts greater than 100 cells per liter. *Dinophysis acuminata* was present during the period June through November at background levels (<100 cells per litre). Cell levels reached a maximum of 500 cells per litre in June, but no toxin was reported from analyses of shellfish samples using the rat bioassay

## Norway

### ASP

For the first time ASP-toxin (domoic acid) was found in blue mussels (*Mytilus edulis*) above quarantine levels in Norway, although for a short period only. This episode was recorded at a monitoring station in mid-Norway in the end of May and lasted at most about three weeks.

### DSP

As usual DSP-toxins were recorded above quarantine levels at some monitoring stations in southern Norway, most frequently in January-March and November-December. The problems were about as "normal". In northern Norway, however, DSP-toxins were more common in 2003 than in average the recent years, even then less than in southern Norway. The toxicity of the mussels in the north was recorded in September-October.

### PSP

Also occurrences of PSP-toxins in mussels are recurrent problems in Norway. In 2003 the problems were small, except for a period in May-June in southern Norway, along the Skagerrak coast, when the mussels were toxic for about four weeks. The source could have been *Alexandrium tamarense* which was present in low numbers (up to 800 cells/L were recorded), but it was speculated if also toxic bacteria could contribute to the toxicity.

### AZA (Azaspiracids)

For the first time in Norway presence of azaspiracides in the mussels passed the quarantine levels. This happened in more or less the same period, September-October, both in southern and northern Norway, but not along the coastline between. We so far can only speculate on the potential source organisms to the problem.

## Poland

In Polish coastal waters, an increased number of *Nodularia spumigena* filaments occurred for the first time on 13 July, it was a month later than in the previous two years. The late occurrence of cyanobacteria was clearly connected with low water temperature at the beginning of summer (lower than 16°C). A sudden bloom of *Nodularia spumigena* was recorded on 17 July, when waters got warmer. This year two peaks of the bloom were observed: the first on 17 July with chlorophyll *a* concentration up to 19 904.8 µg/L, nodularin (hepatotoxin) up to 25 945 µg/L and 4 013 µg/g in water and lyophilized phytoplankton sample, respectively; and the second one on 24 July with chlorophyll *a* concentration up to 31.7 µg/L, nodularin up to 32.8 µg/L and 1134 µg/g in water and lyophilized phytoplankton sample, respectively. During the most intensive bloom all beaches were closed. However, people who did not refrain from swimming suffered from skin irritation.

That year the bloom was exceptionally intensive and the concentration of nodularin was the highest one ever recorded in these water bodies.

## Portugal

PSP was not detected in Portugal during 2003.

Persistent DSP outbreaks associated with *Dinophysis acuminata* and *D. acuta* on the northwest coast, mainly in Aveiro area, caused harvesting closures of *Mytilus edulis* and other mollusc bivalves from June to November.

There were two pulses of ASP toxins caused by *Pseudo-nitzschia* spp., also on the northwest coast at Óbidos lagoon in May (64550 cells l<sup>-1</sup>) and in July at Aveiro (42850 cells l<sup>-1</sup>). Domoic acid levels >20mg 100g<sup>-1</sup> were recorded in scallops.

## Spain

During 2003, DSP outbreaks caused by *Dinophysis* spp. constituted the main problem for shellfish exploitations in all regions on the Atlantic and the Mediterranean coasts of Spain. In Galicia, it was reported for the first time a bloom of *Lingulodinium polyedra* associated with closures for the presence of lipophilic shellfish toxins above regulatory levels.

### Andalucia

Atlantic coast: Suffered the usual chronic DSP events caused by *Dinophysis acuminata* (up to 8500 cell · l<sup>-1</sup>) that led to closures of very prolonged closures (March to July) of *Donax* clam harvesting.

Mediterranean coast: PSP outbreaks caused by *Gymnodinium catenatum* (up to 7 · 10<sup>3</sup> cell l<sup>-1</sup>) that led to closures of clams, scallops and mussels harvesting. Maximum toxin levels (974 µg 100 g) were detected in the warty Venus clam (*Acanthocardia tuberculata*). In addition, during 2003 there were also closures due to DSP toxins in clams (*Callista chione* and *Donax trunculus*) associated with proliferations of *Dinophysis acuminata* (up to 26000 cell l<sup>-1</sup>) in August.



## Catalunya

**PSP:** *Alexandrium minutum* that blooms recurrently between January and March reached maximum levels in May of  $2 \cdot 10^6$  cell  $l^{-1}$  and 269  $\mu g$  eq. STX  $\cdot 100$  g. This led to closures of natural banks of *Donax* clams in some embayments. Dense patches of *Alexandrium catenella* were reported in late August in Tarragona harbour, areas with no shellfish exploitations.

**DSP:** *Dinophysis sacculus* (up to 15600 cell  $l^{-1}$ ) and *D. caudata* (up to 1640 cell  $l^{-1}$ ) were responsible for diarrhetic shellfish toxin outbreaks affecting shellfish exploitations in the River Ebro Delta during one month in May and one week in August respectively. DSP toxicity of unknown origin was reported in oysters (*Crassostrea gigas*) in October.

**Icthyotoxic species:** *Gyrodinium corsicum*, that usually blooms between November and April, reached “red tide” levels ( $> 10^6$  cell  $l^{-1}$ ) between May and June. Fish in experimental tanks were saved by closing the intake of *G. corsicum* – containing water. High biomass blooms of non-toxic species (*Gymnodinium impudicum*, *Alexandrium taylorii*, *Calyp-trosphaera sphaeroidea*) occurred in the summer, causing social alarm and negatively affecting the tourist industry.

## Galicia

PSP outbreaks caused by *Alexandrium minutum* (up to  $3 \cdot 10^5$  cell  $l^{-1}$  and toxin levels of 900  $\mu g$  100 g) were reported in the Galician “Rías Altas” and in a restricted embayment inside the Ria de Vigo in June.

Very restricted and short-lasting domoic acid-outbreaks (up to 290 ppm) caused by *Pseudo-nitzschia* spp. occurred in 2003, except the persistent accumulation of domoic acid above regulatory levels in scallops during the whole year. Most of the mussel cultivation areas in the Galician Rías Baixas were free of domoic acid for the whole year. In contrast, DSP toxins produced first by *Dinophysis acuminata* (March to September, max. 2560 cell  $\cdot l^{-1}$ ) and afterwards by *D. acuta* (max. 5220 cell  $l^{-1}$ ) and to a lesser extent by *D. caudata* (September-October) caused very persistent closures that in some areas, such as Ria de Pontevedra, lasted for nine months.

*Lingulodinium polyedrum* usually blooms in the Northern “Rías Altas” overlapped with DSP producing populations of *Dinophysis*. But in 2003, *L. polyedrum* occurred in dense concentrations (even formed red patches, up to  $2 \cdot 10^6$  cell  $l^{-1}$ ) between June and September and for the first time (Arévalo *et al.* in press), closures due to high levels of lipophilic shellfish toxins were clearly ascribed to this species.

## Sweden

### Skagerrak and Kattegat

#### Harmful algae

No high biomass harmful algal blooms, such as *Chattonella*- or *Chrysochromulina*-blooms, occurred in the area in 2003. Dinoflagellates producing Diarrhetic Shellfish Toxins (DST) were found in the area all year. *Dinophysis acuminata* and *D. acuta* were observed with abundance above the warning limits (limits are 900 cells/l and 200 cells/l respectively). On a few occasions the Paralytic Shellfish Toxin (PST) producing dinoflagellates *Alexandrium tamarense*, *A. minutum* and *A. ostenfeldii* were observed with abundance above the warning limit (200 cells/l). The highest abundance of *A. tamarense* was 10,000 cells/l in the beginning of June at station Åstol.

The potentially harmful algae *Heterosigma* sp., *Phaeocystis pouchettii*, *Dictyocha speculum*, *Chrysochromulina* spp., *Pseudo-nitzschia* spp. and *Chattonella* sp. occurred in the area but in moderate abundance.

#### Algal toxins in blue mussels (*Mytilus edulis*)

From January to April and from September to the end of the year concentrations of DST (Diarrhetic Shellfish Toxins) were above the limit for marketing in some areas of the Swedish Skagerrak coast. It should be pointed out that in other areas mussels with DST content below the quarantine limit was harvested simultaneously, thus safe mussels were available most of year 2003. Paralytic Shellfish Toxins (PST) above the quarantine limit were not detected during 2003.

#### Baltic Proper

Substantial blooms of large cyanobacteria were observed in the Baltic from ca 10 of July to the end of August. *Nodularia spumigena* occurred in very high amounts. Satellite images showed strong surface accumulations in large parts of the Baltic proper. Maximum distribution was observed in mid July. Tourism was affected since swimmers were advised not to go into the water. Also the public found the cyanobacteria a nuisance. Nodularin was observed in the plankton by German scientists.

### Bothnian Bay

An off shore bloom was observed using satellites in September. Cyanobacteria are probably the organisms observed but no samples were obtained for microscopy.

## UK

### Northern Ireland

In general, 2003 was a quiet year for toxin producing microalgae in the twenty six Northern Ireland monitoring sites.

*Alexandrium* spp. were recorded on four occasions in 2003 but only reached very low numbers (max. 40 cells.l<sup>-1</sup>). No PSP toxins were detected.

Four species of *Dinophysis* (*acuminata*, *acuta*, *norvegica* and *rotundata*) were recorded in water samples taken during the year but were always below the regulatory level (triggering shellfish sampling) of 100 cells.l<sup>-1</sup>. A small number of isolated DSP episodes were recorded in shellfish during the year but were not coincident with any harmful microalgal species.

*Pseudo-nitzschia* spp. were present in 46% of samples reaching a maximum concentration of 128,160 cells.l<sup>-1</sup>. Toxicity, however, was confined to samples of scallops (*Pecten maximus*) where domoic acid levels reached a maximum of 328.27µg.g whole flesh.

No major blooms of phytoplankton were recorded during the year. However, large numbers of *Eutreptiella* spp. were recorded from Belfast Lough throughout May and again in August when counts reached 1,000,000cells.l<sup>-1</sup>. There were no reports of any associated harmful effects.

### England and Wales

*Alexandrium* spp. were very widespread this year, being recorded from eleven of the 23 sampling areas, with *A. minutum* occurring much more frequently than usual. Highest concentrations (*A. tamarensis*) were found in the Salcombe Estuary (1.4 million cells l<sup>-1</sup>) in June. A sample collected at Salcombe on 26 June contained *A. minutum* at 740,000 cells l<sup>-1</sup>, which coincided with PSPs in oysters of 52 µg 100g flesh (below the action level of 80 µg 100g flesh). There was only one other incident of PSPs, which occurred in cockles from the North-Kent coast in February 2003.

*Dinophysis* spp. were found in eleven sampling areas, but often only on one occasion and always at low concentrations. Highest concentrations (26 cells.l<sup>-1</sup>) were found at the offshore site at Blyth, Northumberland. These are much lower concentrations than normal as they often peak at 1,000 cells.l<sup>-1</sup> at this site. *Prorocentrum lima* was only found on one occasion (Fowey) at low concentrations. Atypical DSP responses (from flesh sampling) continued into October, particularly from samples collected in the Burry Inlet, Thames and Wash. Only on one occasion (Wash, Gat Channel) in early July, were *Dinophysis* spp. coincident with DSPs in a sample of cockles.

*Pseudo-nitzschia* spp. were found in 15 sampling areas but they never breached the 'investigative' level of 50,000 cells.l<sup>-1</sup>, when species identification is determined by Scanning Electron Microscopy (SEM). Highest concentrations were 28,000 cells l<sup>-1</sup> in a sample collected in the Fowey in August 2003. ASP toxins are most frequently found in samples of scallops from offshore fishing grounds, particularly in the Western Channel. In 2003, the concentration of ASPs found in flesh samples were lower than usual, with the highest concentration (3.88µg.g) being found in a sample of scallops landed at Plymouth in mid-September.

### Scotland

#### Coastal sites

Two blooms of *Alexandrium* spp. were observed during 2003, mainly concentrated in Orkney and Shetland. The first occurred in March/April with cell numbers reaching 4,000 cells.l<sup>-1</sup> in Orkney. The second occurred in July/August in Shetland and was associated with PSP toxicity in mussels from this area. The highest number of *Alexandrium* spp. observed to date by the Scottish monitoring programme was observed during this period (18,800 cells.l<sup>-1</sup>). Only two blooms of *Alexandrium* were observed on the West Coast of Scotland (max. 2,000 and 6,000 cells.l<sup>-1</sup>).

Moderate levels of *Dinophysis* species (*D. acuminata*, *D. acuta*, *D. norvegica* and *D. dens*) were observed this year from June to September (max number 4,000 cells.l<sup>-1</sup>). DSP was first detected in mussels in Shetland in April and by late June/early July had been detected in mussels from a number of sites from the East and West Coast of Scotland.

Two main types of *Pseudo-nitzschia* spp. blooms were observed during the year. The first was observed in March/April comprising of *Pseudo-nitzschia* 'delicatissima type' spp. (diameter <5µm). This was not associated with ASP toxicity in coastal areas. Blooms of *Pseudo-nitzschia* 'seriata type' spp. (diameter >5µm) were observed from July – September. ASP levels (>20µg.g<sup>-1</sup>) was detected in mussels at one coastal site on the West Coast during the year. High ASP levels were detected from *P. maximus* samples from a number of coastal sites along the West Coast.

*Pseudo-nitzschia* cells from 10 samples were analysed using Transmission Electron Microscopy to identify the cells present to species level. Seven species were observed; *P. australis*, *P. fraudulenta*, *P. pungens*, *P. cf. seriata*, *P. delicatissima*, *P. pseudodelicatissima* and *P. cf. heimii*.

A bloom of *Karenia mikimotoi* cells was observed in the Shetland Isles in August 2003. Water discolouration was observed at the sites effected and the maximum level recorded during this bloom was 18 ,000,000 cells.l<sup>-1</sup> from a surface water sample taken from 25<sup>th</sup> August 2003.

#### *Offshore areas*

PSP was detected in offshore *P. maximus* fishing grounds from the East Coast, Moray Firth, Orkney and Shetland during the summer but toxin levels fell to below the regulatory limit in most areas by September. Positive DSP results from whole scallops was detected by bioassay however subsequent chemical assays found little or no trace of the DSP toxins. ASP toxins continued to cause problems in all major offshore scallop fishing grounds throughout the course of the year with the highest level detected in the Northeast Coast.

#### **USA**

2003 was basically a “normal” year for HABs in much of the U.S., with several noteworthy or exceptional events.

#### **PSP**

Similar to previous years, Maine, California, Oregon, Washington, and Alaska all recorded PSP events during 2003. Both eastern and western Maine experienced closures due to high toxin levels (maximum of 33,785 µg STX eq./100g); in eastern Maine partial closures of the quahog fishery remain in effect into 2004. This occurred as a result of a late-season bloom that started in late September and continued into late October. In contrast to past years, Alaska experienced no human morbidity this year due to PSP poisoning. Areas in Washington state reported high toxin levels in late summer/fall- with mussels in Port Ludlow reaching a level of 1000 µg STX eq./100g and geoduck clams in Port Gamble reaching a new record of 3,414 µg STX eq./100g. By contrast, the last quarter of 2003 saw no geoduck PSP closures for the first time since 1995. The southern coast of Oregon saw elevated PSP toxin levels in late summer and fall and closed this area to recreational shellfish harvesting. In California, toxin levels were low through much of the area, with one exception. Mendocino county reported toxin levels peaking at 1600 µg STX eq./100g in September.

During 2002, the first Puffer Fish Poisoning (PFP) attributed to saxitoxin was reported in the U.S. (Florida). Since that time, the state has had a ban on the harvest of puffer fish from the Indian River Lagoon area. In 2003, puffer fish in this area were once again found to contain saxitoxin, presumably taken up by feeding upon small infaunal bivalves that were also found to contain saxitoxin. The presumed toxic organism is *Pyrodinium bahamense*. No human cases of PFP occurred in 2003.

#### **ASP**

ASP was recorded in California, Oregon, and Washington. Similar to last year, the outbreak in California was extensive, stretching from Los Angeles to northern California, but the toxin levels were not as high – with 140 ppm domoic acid being the highest number reported. Washington state experienced an ‘historic’ domoic acid bloom in September – for the first time a Puget Sound sample tested over the closure limit of 20 ppm. In the Northeast, domoic acid was detected in the tissues of several dead humpback whales in the Georges Bank area, and is now considered the most likely cause of the deaths of 19 of those animals.

#### **NSP**

NSP associated with *Karenia brevis* occurred in northwest and southwest Florida as well as in Texas. Fish kills and human respiratory irritation were reported along with the Florida blooms. These were all “normal” occurrences.

#### **Brown tide**

For the first time since 1985, there were no reports of brown tide this year.

#### ***Pfiesteria***

There were no reports of fish kills definitively attributed to *Pfiesteria* in North Carolina or Chesapeake Bay.

#### ***Prymnesium parvum* “Golden Algae”**

During 2003, Texas experienced toxic *Prymnesium parvum* blooms in 19 freshwater reservoirs and rivers. These blooms caused extensive fish kills – both natural and aquacultured fish.

## 7 Other reports

---

### Potentially Harmful Algae and HAB events along the Tunisian coast

Monitoring of harmful algae along the Tunisian coast is conducted by the Institut National Agronomique de Tunisie and concentrates on the following four main research themes: dinoflagellate/diatom taxonomy in neritic ecosystems (coastal area and lagoons); dynamics of phytoplankton community structure, abiotic factors influencing plankton distribution and taxonomy; and ecology of zooplanktonic community.

Research is focused particularly on the northern coasts of Tunisia. In the Bay of Tunis, phytoplankton samples were taken at monthly intervals between 1993–1995 and weekly during 2001. At other locations the sampling frequency is weekly or bi-monthly depending on the resource.

Investigations on phytoplankton in some Tunisian lagoons and coastal waters show;

- conditions of the Bou Grara lagoon (South of Tunisia) lead to monospecific blooms of *Gyrodinium cf aureolum* (*Gymnodinium* spp.) often associated with *Prorocentrum micans* and *P. minimum*. The first mass occurrence of *Gyrodinium* was recorded in 1991 and resulted in fish mortality.
- 16 potentially harmful algal spp. have been observed among the 300 taxa in the Bay of Tunis although no toxic events have been recorded.
- Cell densities of the potentially harmful algae are very high in some of the lagoons studied, particularly those with enclosed areas.

During 1995, extensive monitoring was undertaken where thirty-six sampling stations were sampled along the coast of Tunisia. In shellfish production areas, about 15 harmful algae associated with phycotoxins were observed e.g.. *Dinophysis* spp, *Prorocentrum lima* and *Alexandrium* spp.

### HANA: Harmful Algae of North Africa

The HANA network was established to promote scientific research and co-operation between North African countries in monitoring and management of harmful algal blooms and also to establish a link between the HANA region and international HAB programmes. The network has a number of aims;

- To improve scientific knowledge of physical and biogeochemical and physiological factors governing the development of HABs
- To create a database of HAB incidences
- Establish a directory of personnel working in the field of HABs and their areas of expertise
- Compile an inventory of regional publications relevant to HABs
- Promote exchange of information through regular workshops
- Promote capacity building for scientist and managers involved with HABs.

Currently the network includes members from Egypt, Morocco, Tunisia and Mauritania.

## 8 Draft resolutions

---

The **ICES-IOC Working Group on Harmful Algal Bloom Dynamics** [WGHABD] (Chair J. L. Martin, Canada) will meet in Norway, from 4-7 April 2005 to:

- a) Review the dynamics of toxin-producing phytoplankton and associated toxins in shellfish, related to phytoplankton abundance, and phytoplankton community structure with reference to HAB population dynamics. In 2005 the focus will be PSP toxin producing phytoplankton and associated toxins in shellfish (Canada, Spain, Scotland, US, Denmark;
- b) consider the status of knowledge concerning biologically active specific chemicals, their chemical nature, presence and production in algae and their effects on individuals and population dynamics as well as their impacts on ecosystems;

- c) bring new findings in phytoplankton population dynamics models, with emphasis on loss processes, to the attention of the WGHABD for discussion;
- d) review planning progress for the proposed Workshop on New and Classic Techniques for the Determination of Numerical Abundance and Bio-volume of HAB-species;
- e) continue summarization of the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and analyse for trends in these blooms over recent decades for input to the Regional Ecosystem Study Group for the North Sea in 2006;
- f) collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis;
- g) review progress in computerised production of decadal maps from country reports, including the revision of reports already in the database covering the last 10 years and the web interface;
- h) propose types of analyses that should be performed using the IOC-ICES HAE-DAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted;
- i) Discuss new findings that pertain to harmful algal bloom dynamics.

The WGHABD will report within 4 weeks (May 1) to the Oceanography Committee and ACME.

### Supporting information

Priority:	The activities of this group are fundamental to the work of the Oceanography Committee. The work is essential to the development and understanding of the effects of climate and man-induced variability and change in relation to the health of the ecosystem. The work of this ICES-/IOC WG is deemed high priority.
Scientific Justification:	<p>Term of Reference a) Review the dynamics of toxin-producing phytoplankton and associated toxins in shellfish, related to phytoplankton abundance, and phytoplankton community structure with reference to HAB population dynamics. In 2005 the focus will be PSP toxin producing phytoplankton and associated toxins in shellfish (Canada, Spain, Scotland, US, Denmark).</p> <p>Studies show that all phytoplankton populations, including HABs, have great interannual variations in bloom intensities. Analyses of total community structure for trends and patterns as well as physical and chemical parameters are necessary to advance the current knowledge of HABs. As longer datasets become available, it is becoming possible to determine impacts of occurrences, distributions, and amplitudes of HABs. It was therefore felt that the dynamics of HABs require different analyses approaches to data from long term studies.</p> <p>Term of Reference b) Consider the status of knowledge concerning biologically active specific chemicals, their chemical nature, presence and production in algae and their effects on individuals and population dynamics as well as their impacts on ecosystems.</p> <p>There is an increasing body of evidence showing the importance of specific chemicals excreted by phytoplankton species in processes directly regulating population dynamics. Such processes include inhibition of competing species for substrate, repression of grazers and even, in some cases, sensitivity to turbulence. The importance of these controls has largely been hindered by the stress put on toxins detrimental to seafood consumers. Since an international programme like GEOHAB stresses that toxins are not taken into account unless they directly influence the population dynamics, it appears necessary to review the evidence provided in this largely ignored literature.</p> <p>Term of Reference c) Bring new findings in phytoplankton population dynamics models, with emphasis on loss processes, to the attention of the HABDW for discussion.</p> <p>Modelling exercises aimed at understanding HAB population dynamics have suffered from poor estimates of biological loss terms. Review findings of relevant projects on biological loss terms (e.g. FATE) with the goal of recommending appropriate loss term for models. Plan joint meeting of WGHABD with the WGPBI in 2006.</p> <p>Term of Reference d) Review the plans of the intercalibration workshop for comparison of new and classical techniques for determination of numerical abundance and biovolume of HAB species.</p> <p>This workshop is a complex activity requiring algal cultures, field material and a variety of different methodologies and thus detailed planning is necessary for success. The WGHABD has prior experience in conducting intercalibration workshops (<i>in situ</i> growth rate measurements) and can help the SC with this process</p> <p>Term of Reference e) Continue summarization of the distribution and number of harmful algal blooms in the North Sea for the period 2000–2004, and analyse for trends in these blooms over recent decades for input to the</p>

	<p>Regional Ecosystem Study Group for the North Sea in 2006.</p> <p>The working group has established a task group with representatives from the North Sea countries (Sweden, Norway, Denmark, Netherlands, Germany and UK) to address this topic further. A meeting of this task group has been preliminary arranged for the 8<sup>th</sup>/9<sup>th</sup> September 2004 to allow members to assemble their data. The target species for examination selected for examination are <i>Phaeocystis</i>, <i>Alexandrium minutum</i>, <i>A. ostenfeldi</i>, <i>A. tamarense</i>, <i>Karenia mikimotoi</i>, <i>Chatonella</i>, <i>Noctiluca</i> spp., <i>Dinophysis acuta</i>, <i>D. norvegica</i>, <i>D. acuminata</i>, <i>Chysochromulina</i> spp., <i>Pseudonitzschia</i> spp., <i>Pseudogonyaulax</i>, <i>Heterosigma</i>, <i>Fibrocapsa</i>, <i>Dictyocha</i>, <i>Chaetoceros</i> spp. The team will examine the existence of trends in the occurrence of these species and if this could be due to anthropogenic forcing. The potential of the data to describe the dynamics of the natural harmful algal blooms including their natural variability will be studied. The use of the occurrence of harmful algae species as indicators of ecological quality will also be studied. The potential output from this study will be a scientific useful product which can take the form of an ICES general report and a deliverable product to the WGHABD.</p> <p>Term of Reference f)</p> <p>Collate and assess national reports and update the decadal mapping of harmful algal events for the IOC-ICES harmful algal database, HAE-DAT, on a regional, temporal and species basis.</p> <p>The work of collating the national HAE reports and building up HAEDAT and the associated maps is an activity which is unique to the WGHABD. HAE-DAT is not yet established enough to stand alone. A critical step forward is to make HAE-DAT operational with input from regions/countries outside the ICES areas as originally envisaged. In 2001-03 the aim is to include PICES and South America and Caribbean countries (via IOC/FANSA and IOC/ANCA) in HAE-DAT. It should be endeavoured to include HAE-DAT and the associated decadal maps as a contribution to GOOS, thereby embedding these activities in a permanent setting and securing continuity.</p> <p>Term of Reference g)</p> <p>Review progress in computerized production of decadal maps from country reports, including the revision of reports already in the database covering the last 10 years.</p> <p>The WGHABD feels it is important that the decadal maps be tied directly to the IOC-HAEDAT reports. Currently the decadal maps are produced manually with limited consistency and quality control. Procedures and techniques developed under this group may be applied to other regional activities (PICES, IOCARIB etc).</p> <p>Term of Reference h)</p> <p>Propose types of analyses that should be performed using the IOC-ICES HAE-DAT dataset and identify problems and gaps in this dataset that must be rectified before the analyses can be conducted:</p> <p>HAE-DAT is an extremely valuable dataset that has not been extensively utilised. The WG suggested the use of statistical analysis in the dataset trying to examine some key questions on the Dynamics of the HABs such as the possible increase in bloom frequencies and their distribution, the species involved in the harmful events, etc... After the revision of reports included in HAE-DAT, the database administrator will provide a subset of “verified” data to be checked with this objective.</p> <p>Term of Reference i)</p> <p>Discuss new findings that pertain to harmful algal bloom dynamics.</p> <p>The forum for presenting new findings has been an excellent tool for promoting the discussions about topics of general interest. There are obvious reasons to continue with this topic as a term of reference.</p>
Relation to Strategic plan:	This work is relevant to the quantifying of human impacts on the marine ecosystem,
Resource requirements:	None specific.
Participants:	The 2004 meeting attracted 24 participants, demonstrating the importance and interest of this Group within ICES and IOC.
Secretariat Facilities:	None
Financial:	None
Linkages to Advisory Committee:	The WG reports to ACME
Linkages to other Committees or Groups:	The WGHABD interacts with WGZE, WGPE, SGGIB, SGBOSV, WGPBI
Linkages to other	The work of this group is undertaken in close collaboration with the IOC HAB Programme. IOC should be consulted regarding ToR or discontinuation of the WG prior to the ASC.

Organizations:	There is a linkage to SCOR through the interactions of the IOC-SCOR GEOHAB Programme.
Secretariat Marginal Cost Share:	????

## 9 WGHABD recommendations

---

A joint (between the WGHABD and the WGPBI with invited contributions from GEOHAB) Theme Session at the ICES ASC in 2006 was suggested on: “Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge” as in spite of large gaps of basic process knowledge around HAB dynamics, several 3D modelling initiatives are ongoing with respect to studying and predicting HABs. Therefore it is due time to couple the expertise of modellers and biologists to reveal the most urgent needs for better process knowledge to improve the predictability of models. The session aims at participation from 3D modellers and biologists interested in explaining why HABs occur, how they are initiated, how and why they develop in space and time, and why they decay.

The WGHABD recommends that the appropriate officers of each sponsoring organisation contact their national members to nominate national focal points/individuals responsible for data submission to HAE-DAT. It is recommended that these focal points (for ICES Member States) are identified by March 2005.

## 10 Concluding Business

---

The Working Group thanked Anne Goffart for hosting the 2004 meeting and thanked Eileen Bresnan and Pat Tester for generously agreeing to act as Rapporteurs for the meeting. Balloting was held for the venue of the 2005 meeting of the WGHABD. The WG proposes to meet in Arendal, Norway from 4-7 April 2005 – to be hosted by the Institute of Marine Research, Flødevigen Marine Research Station, located in Arendal municipality, in southern Norway.

## 11 Annexes

### Annex 1 List of Participants

Name	Address	Phone/Fax	E-mail
Anderson, Don	Biology Department MS #32 Woods Hole Oceanographic Institution Woods Hole, MA 02543 USA	1-508-289-2351 (T) 1-508-457-2027 (F)	<a href="mailto:danderson@whoi.edu">danderson@whoi.edu</a>
Anderson, Per	Bio/Consult as, Johs, Ewaldsvej 42-44, 8230 Aabyhoej Denmark	45-86258111 (T) 45-86258173 (F)	<a href="mailto:pa@bioconsult.dk">pa@bioconsult.dk</a>
Balode, Maija	Institute of Aquatic Ecology University of Latvia 8 Daugavgrivas Street Riga LV-1048 Latvia	371-9471203 (T) 371-7601995 (F)	<a href="mailto:maija@hydro.edu.lv">maija@hydro.edu.lv</a>
Bresnan, Eileen	FRS marine Lab Victoria Road Aberdeen AB 119DB Scotland	44-1224876544 (T) 44-1224295511 (F)	<a href="mailto:Bresnane@marlab.ac.uk">Bresnane@marlab.ac.uk</a>
Cembella, Allan	Alfred Wegener Institute for Polar and Marine Research Am Handelshafen 12 27570 Bremerhaven, Germany	49-47148311494 (T) 49-47148311425 (F)	<a href="mailto:acembella@awi-bremerhaven.de">acembella@awi-bremerhaven.de</a>
Dahl, Einar	Institute of Marine Research Flodevigen Marine Research Station N-4817 HIS Norway	47-37059040 (T) 47-37059001 (F)	<a href="mailto:einar.dahl@imr.no">einar.dahl@imr.no</a>
Dahlin, Hans	EuroGOOS SMHI SE-601 76 Norrköping Sweden	46-114958305 (T) 46-114958001 (F)	<a href="mailto:Hans.dahlin@smhi.se">Hans.dahlin@smhi.se</a>
Daly, Yahya Kefi Ons	Institut National Agrono- mique de Tunisie 43 Avenue Charles Nicolle 1082 Tunis Tunisie	00-21671289431 (T) 00-21671799391(F)	<a href="mailto:dalyyahya.ons@inat.agrinet.tn">dalyyahya.ons@inat.agrinet.tn</a>
Elbrächter, Malte	Deutsches Zentrum für Marine Diversitätsforschung Forschungsinstitut Sencken- berg Wattenweerstation Sylt Hafenstr. 43 D-25992 Lis/Sylt Germany		<a href="mailto:melbraechter@awi-bremerhaven.de">melbraechter@awi-bremerhaven.de</a>



<b>Name</b>	<b>Address</b>	<b>Phone/Fax</b>	<b>E-mail</b>
Enevoldsen, Henrik	IOC Science and Communication Centre On Harmful Algae University of Copenhagen Øster, Farimagsgade 2D 1353 Copenhagen K Denmark	45-33134446 (T) 45-33134447 (F)	<a href="mailto:Henrike@bot.bu.dk">Henrike@bot.bu.dk</a>
Gentien, Patrick	CREMA Place du Séminaire BP 5 17137 L'Houmeau France	33-0546500630 (T) 33-0546500660 (F)	<a href="mailto:patrick.gentien@ifremer.fr">patrick.gentien@ifremer.fr</a>
Goffart, Anne	Université de Liège Aquapôle Quai Van Beneden 22 B 4020 Liège Belgium	32-43665076 (T) 32-43665102 (F)	<a href="mailto:A.Goffart@ulg.ac.be">A.Goffart@ulg.ac.be</a>
Ismael, Amany	Alexandria University Oceanography Department Faculty of Science Alexandria, 21511 Egypt	00-2035910142 (T) 00-2035457611 (F)	<a href="mailto:amany_3@yahoo.com">amany_3@yahoo.com</a>
Karlson, Bengt	Oceanographic Services Swedish Meteorological & Hydrological Institute (SMHI) Nya VArvet 31 SE-42671 Vastra Frolunda Sweden	46-31-7518958 (T) 46-31-7518980 (F)	<a href="mailto:Bengt.Karlson@smhi.se">Bengt.Karlson@smhi.se</a>
Lion, Monica	IOC-IEO Science & Communication Centre on Harmful Algae Bloom Instituto Español de Oceanografía Centro Oceanografico de Vigo PO Box 1552 36200 Vigo, Pontevedra Spain	34-986492111 (T) 34-986492003 (F)	<a href="mailto:vigohab@vi.ieo.es">vigohab@vi.ieo.es</a> <a href="mailto:monica.lion@vi.ieo.es">monica.lion@vi.ieo.es</a>
Luckas, Bernd	University of Jena Institute of Nutrition Department of Food Chemistry Dornburger Str. 25 07743 Jena Germany	49-3641949651 (T) 49-3641949652 (F)	<a href="mailto:bernd.luckas@uni-jena.de">bernd.luckas@uni-jena.de</a>
Martin, Jennifer	Fisheries & Oceans Canada Biological Station 531 Brandy Cove Road St. Andrews, NB E5B 2L9 Canada	506-529-5921 (T) 506-529-5862 (F)	<a href="mailto:martinjl@mar.dfo-mpo.gc.ca">martinjl@mar.dfo-mpo.gc.ca</a>
Moncheva, Suejana	Institute of Oceanology BAS Varna, 9000 PO Box 52 Bulgaria	00-359-52370485	<a href="mailto:suejm@mail.varna.techno-link.com">suejm@mail.varna.techno-link.com</a>

<b>Name</b>	<b>Address</b>	<b>Phone/Fax</b>	<b>E-mail</b>
Osborn, Thomas	Department of Earth and Planetary Sciences Johns Hopkins University 3400 N. Charles St. Baltimore, MD 21218 USA		<a href="mailto:Osborn@jhu.edu">Osborn@jhu.edu</a>
Poelman, Marnix	Wageningen-UR RIVO and RIKILT PO Box 77 4400 AB Yerseke Netherlands	31-113672314 (T) 31-113573477 (F)	<a href="mailto:marnix.poelman@wur.nl">marnix.poelman@wur.nl</a>
Silke, Joe	Marine Institute Galway Technology Park Parkmore Galway Ireland	35-3091730603 (T) 35-3091730470 (F)	<a href="mailto:joe.silke@marine.ie">joe.silke@marine.ie</a>
Svendsen, Einar	Institute of Marine Research PO Box 1870 Nordnes N-5817 Bergen Norway	47-55238500 (T) 47-55238584 (F)	<a href="mailto:einar.svendsen@imr.no">einar.svendsen@imr.no</a>
Tester, Patricia	NOAA 101 Pivers Island Road Beaufort, NC 238516 USA	1-252-728-8792 (T) 1-252-728-8784 (F)	<a href="mailto:Pat.Tester@noaa.gov">Pat.Tester@noaa.gov</a>
Yin, Kedong	Atmospheric, Marine & Coastal Environment (AMCE) Program Hong Kong University of Science and Technology Clear Water Bay Kowloon Hong Kong SAR China	852 2358-7329 (T) 852 2358-1559 (F)	<a href="mailto:kyin@ust.hk">kyin@ust.hk</a>

## **Annex 2 Agenda of the meeting**

### Monday, 5 April

- 09:00 Welcome; housekeeping issues  
Introduction of participants  
Review by the Oceanographic Committee  
Adoption of agenda/ Terms of Reference
- 09:45 ToR b: Review plans for the proposed Workshop on New and Classic Techniques for the determination of numerical abundance and biovolume of HAB-species (Bengt Karlson)
- 10:30 **Health break**
- 11:00 ToR c: review progress in computerized production of decadal maps from country reports, including the revision of reports already in the database covering the last 10 years (Henrik Enevoldsen, Monica Lion)  
**Discussion**
- 12:30 **LUNCH**
- 13:30 ToR d: Propose types of analyses that should be performed using IOC/ICES HAE-DAT dataset that must be rectified before the analyses can be conducted (Monica Lion, Henrik Enevoldsen, and others)
- 15:30 **Health Break**
- 16:00 ToR a: Collate and assess National reports and update the decadal mapping of harmful algal events for the IOC/ICES harmful algal database, HAE-DAT (Country Reps)
- 18:30 Adjourn for the day

### TUESDAY, 6 April

- 9:00 ToR f: Review existing phytoplankton population dynamics models with particular emphasis on prediction of HAB events (Bengt Karlson, Eileen Bresnan, Kedong Yin, Einar Svendsen)
- 10:30 **Health break**
- 11:00 ToR e: Review the report of the Workshop on Real-time coastal observing systems for ecosystem dynamics and Harmful Algal Blooms Proceeding have not been published but summary was submitted to ICES (CM 2003/C:15) – available at <http://www.ices.dk/reports/occ/2003/> (Don Anderson, Eileen Bresnan, and others)
- 12:30 **LUNCH**
- 13:30 ToR h: consider the environmental dynamics and impacts of individual phycotoxins and their metabolites enabled by new analytical technologies (Allan Cembella)
- 15:00 HANA network (Amany Ismael)
- 15:30 **Health Break**
- 16:00 ToR a: Collate and assess National reports and update the decadal mapping of harmful algal events for the IOC/ICES harmful algal database, HAE-DAT (Country Reps)
- 17:30 Report writing
- 18:30 Adjourn for the day

### WEDNESDAY, 7 April

- 08:30 ToR i: report and discuss new findings (Einar Dahl, Bernd Luckas)
- 09:45 Report on the Workshop on Future Directions in Modelling Physical Biological Interactions (Einar Svendsen, Tom Osborne, Eileen Bresnan)
- 10:30 **Health break**
- 11:00 ToR j: start preparations to summarize the distribution and number of harmful algal blooms in the North Sea for the period 2000-2004, and any trends over recent decades in the occurrence of these blooms for input to the Regional Ecosystem Study Group for the North Sea in 2006 (Eileen Bresnan, Allan Cembella, Einar Dahl)
- 12:30 **LUNCH**
- 13:30 ToR g: Review biological loss processes of selected HAB species (Per Andersen, Pat Tester). Discussion
- 14:30 Report writing
- 15:30 **Health Break**
- 16:00 Report Writing; subgroup and plenary sessions to be decided on 'ad hoc' basis
- 17:00 Adoption of sections of the report
- 18:30 Adjourn for the day

THURSDAY, 8 April

08:30 Adoption of all sections of the report  
10:30 *Health Break*  
10:45 ToRs for 2004; meeting location for 2004  
12:00 Meeting adjournment

### **Annex 3 IPHAB Recommendations**

#### **Recommendation IPHAB-VI.4**

##### **ICES-IOC Working Group on the Dynamics of Harmful Algal Blooms (WGHABD)**

##### **The IOC Intergovernmental Panel on Harmful Algal Blooms,**

**Noting** with satisfaction the progress of the ICES-IOC WGHABD 2000, 2001, and 2002;

**Recognizing** the valuable continued contribution of the WGHABD to the development of the IOC-SCOR GEOHAB Programme;

**Recommends** that the ICES-IOC WGHABD continue to exist within its own identity under the joint auspices of ICES and IOC;

**Recommends** that the IOC continue to encourage participation in the ICES-IOC WGHABD of experts in HAB dynamics from IOC Member States outside the ICES area;

**Noting with satisfaction** the timeliness of the plans of the WGHAB for a workshop on “New and classic techniques for the determination of numerical abundance and biovolume of HAB species”;

**Urges** IOC Member States to support such a workshop by providing funds and/or expert participants;

**Requests** the WGHABD to take note of the planned 3rd Workshop on Molecular Probe Technology for the Detection of Harmful Algae with a view to possibly merge the two planned activities.