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REPORT OF THE ICES-IOC WORKING GROUP ON HARMFUL ALGAL BLOOM DYNAMICS (WGHABD)

3-6 APRIL 2006 GDYNIA, POLAND

International Council for the Exploration of the Sea Conseil International pour l'Exploration de la Mer

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Executive Summary

The ICES-IOC Working Group of Harmful Algal Bloom Dynamics meeting was hosted by the Institute of Oceanography of the University of Gdansk, in Gdynia, Poland from 3–6 April 2006. 26 scientists from thirteen countries participated. This was a very successful meeting with a challenging set of terms of reference to deal with in the time available. Nevertheless, the group made 35 presentations under the terms of reference and this report is a summary of these presentations and discussions.

Over the three and a half days, the group dealt with:

- Progress in the detection of harmful algal blooms and their dynamics by remote sensing techniques.
- Reviewed the section on Phytoplankton Monitoring in the Report of the Joint FAO/IOC/WHO Ad Hoc Expert Consultation.
- Reviewed the outcome of the Workshop on New and Classical Techniques in Enumeration of Phytoplankton.
- Reviewed the progress and analyses that REGNS North Sea Group has made.
- Discuss new findings that pertain to harmful algal bloom dynamics.
- Reviewed the on-line format of HAEDAT submission form.
- Reviewed the structure and composition of the decadal HAE maps.
- Collated and assessed National reports.
- Discussed potential contributions to the ecosystem overview.

These are dealt with in detail in the report (Sections 5 to 15).

New Findings

Eight presentations were made by the group to report new findings in the area of HAB dynamics. These included the summary of a three year project on HAB oceanography in Ireland (BOHAB), The use of FlowcamTM in phytoplankton enumeration, A summary of Spirolides and Microcystin chemistry, The detection of Azaspiracid in crabs, a summary of the seasonality of *Dinophysis* in the Swedish Skagerak. Additionally, there were three notable, unusual blooms brought to the groups attention: *Alexandrium fundyense* in the Gulf of Maine, *Alexandrium* in the Bay of Fundy, and an exceptional bloom of *Karenia mikimotoi* in Ireland. More details of these presentations are given in the New Findings section of this report (Section 10)

The group also had the opportunity to have a half day joint session with the WGGIB (Working Group on GEOHAB implementation in the Baltic). Presentations by this group included reports on New Nodularin analogs in the Baltic, Phosphorus dynamics in the Baltic during a cyanobacterial bloom, satellite methods in Baltic system monitoring, Ecosystem effects and health hazards of Cyanotoxins, Nodularin concentrations in Southern Baltic sediments, mussels and Flounder, and latest news on BMAA neurotoxin in the Baltic Sea. These are reported in detail in the WGGIB report.

National Reports

One of the most useful functions provided by **WGHABD** is an annual opportunity for international delegates to compare international trends in HAB events. At the 2006 workshop twelve countries presented national reports for discussion, of which there are ten reported in this document. 2005 saw several noteworthy or exceptional HAB events.

• The **USA** experienced one of its most intense blooms of *Alexandrium fundyense*, which caused PSP all along the coast of New England. A second unusual HAB

was the bloom of *Karenia brevis* which lasted all through 2005 in the Florida area, killing fish, mammals and birds, and causing respiratory problems in humans.

- Farther north in **Canada**, it was a fairly unexceptional year with the usual round of closures experienced both on the east and west coasts from PSP.
- On the other side of the Atlantic, **Norway** did not record ASP toxicity even following a dense bloom of 16 million *Pseudo-nitzschia* cells/L recorded. Small events of DSP and PSP toxicity were recorded with levels exceeding the quarantine levels at only a few monitoring stations. AZA was detected for the first time in Crabs.
- **Estonia** recorded exceptionally low levels of Nodularia when compared to the annual trends,
- **Poland** also reported 2005 as being a "moderate" year for *Nodularia*.
- **Denmark** did not experience any PSP or DSP, however ASP was detected for the first time in mussels from Danish waters.
- The **UK** reported the presence of *Alexandrium* spp. presence in moderate levels in England at 1.8 million cells/L and low levels in Scotland > 1300cells/L. These did not coincide with PSP in shellfish. Low levels of *Dinophysis* were also reported. *Pseudo-nitzschia* were more widespread and persistent than previous years but the concentrations were not particularly high. The *Karenia* bloom that affected the Republic of Ireland did not extend into UK waters.
- Ireland did in contrast, however, experience a high number of HAB related problems in 2005. For the first time a major ASP event was recorded in both samples of *M.edulis* and *C.gigas* associated with a bloom of *Pseudo-nitzschia*. Extensive DSP toxicity was detected through the summer months in all areas and then quarantine levels of AZA, lasted right through the following autumn and winter months. Low levels of *Alexandrium* were detected through the summer without any toxicity, apart from a small PSP outbreak in Cork in June. Finally a bloom of *Karenia mikimotoi* wreaked havoc on the benthic and pelagic communities during June and July.
- The Netherlands reported a bloom of *Phaeocystis* in February which is earlier than normal, resulting eventually in a 10 million cell/L bloom. In May this peaked at 138 million cells/L. Moderate levels of *Dinophysis* were recorded which resulted in levels slightly above quarantine level of DTX-3 as recorded by LCMS.
- In **Spain**, a very intense *Gymnodinium catenatum* bloom (after 10 years of absence!) occurred from October to December affecting all the production areas in the Rías Baixas and some areas in the Northen Galician coast. Maximum *G. catenatum* concentration was 1.7·10⁵ cell·l⁻¹ and maximum level of accumulated toxins 4080 µg STXeq·100 g⁻¹ meat.

Full text of these National reports and the other Terms of Reference are given in the body of the report.

1 Welcome and opening of the Meeting

Following a welcome by the Chair, the Deputy Director of the Institute of Oceanography, Dr Hanna Mazur-Marzec, opened the meeting on the 3 April 2005. The participants were introduced with respect to their names, institute, national affiliation and fields of expertise. The agenda was agreed and Dr Pat Tester and Dr Eileen Bresnan elected as joint Rapporteur. The list of participants is presented in Annex 1. The meeting agenda is presented in Annex 2.

The Chair invited comments and review from the outgoing Chair of WGHABD, Dr Jennifer Martin relating to the WGHABD report presented to ICES Oceanography Committee from the 2005 meeting. She reported to the group that the oceanography committee felt the report was well organized, informative and the meeting well attended. While primarily concerned with HABs, the WG was not just addressing the dynamics but also more general areas of HAB science. An example of this was the co-ordination of the Intercomparison 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 (WKNCT) held in Kristineberg, Sweden 22–27 August 2005.

Being a joint ICES-IOC working group, the IOC in most years announces the possibility for its Member Countries outside the ICES area to attend WGHABD and offers travel support. In 2006 however, the IOC were not in a position to offer this support due to other demands on their budget. The IOC did support the intercomparison workshop (WKNCT). The IOC also supports the general aims of WGHABD, and continues valuable interaction regarding data collection and management of HAB data through the development of the HAEDAT database.

The Terms of Reference for 2005 were reviewed and adopted.

2 Terms of Reference

At the 92nd Statutory Meeting (2005), Aberdeen, Scotland, the Council approved the WGHABD (2005) Terms of References:

The ICES-IOC Working Group on Harmful Algal Bloom Dynamics [WGHABD] (Chair J.Silke Ireland) will meet in Gdynia, Poland, from 3–6 April 2006 to:

- a) Review progress in the detection of harmful algal blooms and their dynamics by remote sensing techniques and examining results from new sensors and algorithms as well as validation procedures used for HAB observations.
- b) Review the section on Phytoplankton Monitoring in the Report of the Joint FAO/IOC/WHO Ad Hoc Expert Consultation for Codex Alimentarius on Biotoxins in Bivalve Molluscs (Oslo 26 September 2004).
- c) Review the outcome of the WKNCT Workshop on New and Classical Techniques in Enumeration of Phytoplankton.
- d) Review progress towards the joint theme session between WGHABD and WGPBI for the ICES ASC in 2006 titled "Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge".
- e) Review progress and analyses that REGNS North Sea Group have done on datasets submitted by members of WGHABD (to meet in the interim).
- f) Discuss new findings that pertain to harmful algal bloom dynamics. Bring new findings in phytoplankton population dynamics models, with emphasis on loss processes, to the attention of WGHABD for discussion.
- g) Review the on-line format of HAEDAT submission form and evaluate the amendments made to update historical submissions and links to mapping.

- h) Review the structure and composition of the decadal HAE maps for the ICES region with special reference to clarifying the distinction between harmful algal blooms and the harmful affects that are reported on the maps. In particular, the registration of cyanobacterial blooms in brackish and marine waters should be revisited from the emerging perspective of their known toxicity and implicit harmful effects.
- i) 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).
- j) Discuss and report on potential contributions to the ecosystem overview of the advisory reports describing the quantity and quality of marine habitat and/or the health of the marine ecosystem, and to consider and report on potential indicators of significant change in these ecosystem attributes.
- k) Review and update sub-regional data tables and where necessary include new data (parameters) and/or existing data (parameters) updated where relevant. The data tables will be subject to thematic assessment to be undertaken at a REGNS thematic assessment workshop.

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 from sediment and climate studies 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 emphasized. The economic, resource and environmental effects of HABs are included within the WGHABD. In addition, phytoplankton ecology models often rely on 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 modellers to try and incorporate physics and HAB dynamics into models.

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

Term of Reference a) Review progress in the detection of harmful algal blooms and their dynamics by remote sensing techniques and examining results from new sensors and algorithms as well as validation procedures used for HAB observations.

Five presentations were made, including a review of current technology followed by presentations of data from Sweden, USA (2) and Canada.

Space – or airborne remote sensing of the sea, sometimes termed EO (Earth Observations) is often motivated with the aim of observing harmful algal blooms. Initiatives including the GMES (Global Monitoring for the Environment and Safety), the MERSEA program and its application to Operational Oceanography and HAB detection in real time will be reviewed. New satellites and sensors have become operational the last years, i.e., the MERIS sensor on the European satellite ENVISAT and the US satellites AQUA and Terra with the sensor MODIS. Older sensors include the SeaWIFS that has reached its end of life. Earth observations have limitations regarding HAB observations which include that only high biomass blooms are detected, and only surface water is monitored etc.

In general the only HAB-product available is chlorophyll a. Also cloud cover is a problem for the technique. New sensors with higher spatial and spectral resolution as well as new algorithms for data processing hold promise for resolving signals for HAB-species or algal groups, e.g., cyanobacteria. There is a great need to review the results from the new sensors and algorithms and the validations procedures used.

The conclusions of these discussions were:

- Use remote sensing data for detection and monitoring of possible harmful algal blooms is possible but should include verification observations with *in situ* data (traditional, buoys, SOOP, ferrybox, etc.);
- Combination of results with meteorological and oceanographic forecasting models to predict impact;
- Near real time monitoring is useful;
- It is vital to spread the information beyond the science community: to authorities, stakeholders and to the public and the information should be presented in a simple, understandable way.

Term of Reference b) Review the section on Phytoplankton Monitoring in the Report of the Joint FAO/IOC/WHO Ad Hoc Expert Consultation for Codex Alimentarius on Biotoxins in Bivalve Mollusks (Oslo 26 September 2004).

This report was in response to requests from countries for updated guidelines for biotoxin analysis methods and regulator thresholds and plankton monitoring

The draft report was distributed at last year's WGHABD for comments and Per Anderson presented a summary of the guidelines in the report.

Term of Reference c) Review the outcome of the WKNCT Workshop on New and Classical Techniques in Enumeration of Phytoplankton.

This workshop was a complex activity requiring algal cultures, field material and a variety of different methodologies, with the objective of providing valuable results on the comparison of different microscope based techniques and some advanced molecular techniques for the identification and quantification of harmful microalgae. The WGHABD was instrumental in initiating this process and established a steering committee. The steering group presented a summary of the report from the workshop and discussed its dissemination.

Term of Reference d) Review progress towards the joint theme session between WGHABD and WGPBI for the ICES ASC in 2006 titled "Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge".

An update on this theme session was provided by one of the co-conveners with input from the working group.

Term of Reference e) Review progress and analyses that REGNS North Sea Group have done on datasets submitted by members of WGHABD (to meet in the interim).

The REGNS study group has requested that the WGHABD prepare to provide data, information and indicators. A delegate from the WGHABD reported on the outcome of the REGNS meeting in May 2005 and on the progress of the assembly and analysis of the data.

Term of Reference f) discuss new findings that pertain to harmful algal bloom dynamics. Bring new findings in phytoplankton population dynamics models, with emphasis on loss processes, to the attention of WGHABD for discussion

The working group received nine presentations on new findings and events, from the participants. These included the Summary of a project on the Biological Oceanography of HABs in Ireland, A review of a *Nodularia* model, review of the FlowCam system in cell

enumeration, Spirolide Neurotoxins from *Alexandrium ostenfeldi*,. An exceptional bloom of *Karenia mikimotoi* in Ireland, The 2005 New England *Alexandrium fundyense* bloom, Azaspiracid in crabs, Dinophysis in the Swedish Skagerak and *Alexandrium* cell abundance in the Bay of Fundy.

Modelling exercises aimed at understanding HAB population dynamics have suffered from poor estimates of biological loss terms. Current knowledge on selected HAB loss processes (e.g., grazing, viruses, parasitism, and programmed cell mortality) is limited. Improved knowledge on the dynamics of these loss processes and their relative contribution is essential to improve models for HAB dynamics. Of the presentations received dealing with population dynamics of HABs there still was a concentration on the onset of blooms rather than their fate, there appears to be little information being generated on the loss processes.

Term of Reference g) The new online format of the HAE-DAT submission form was demonstrated and discussed. The suggestions from last year's discussions at WGHABD have been mostly incorporated and the dataset is in a format where better analyses can be conducted. This new format replaces the previous where data was entered manually into the HAE-DAT dataset (which was in Access97 format). This new electronic format (with the same information as previous forms) is available online for submission directly into the database. Monica Lion (IOC-IEO-SCCHA, Vigo, Spain) has gone through potential problems for the conversion of all the old historical records into the new form.

Term of Reference h) 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 Decadal maps are currently being updated manually. A new Decadal maps product which uses both ArcView and Flash softwares, 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. The capability of linking the maps has been and continues to be extended to additional countries. Most ICES member countries have provided divisions of coastlines and coordinates to enable the linkages. Further opportunities to develop the links will be explored inter-sessionally.

Term of Reference i) Collate and assess National reports and update the decadal mapping of harmful algal events for the IOC/ICES harmful algal database, National reports were presented for, USA, Germany, Denmark, Canada, Norway, Estonia, Latvia, Poland, Netherlands, Great Britain, Ireland, Spain. Maps were circulated for updating for inclusion to the decadal maps. Information was requested for input on the new online database in the required format for HAEDAT.

Term of Reference j) It was concluded that that the role of phytoplankton with regard to the ecosystem approach, is a far wider issue than could be addressed by WGHABD, whose main role is to investigate the dynamics of functional sub-group of phytoplankton. If the Oceanographic Committee wish any specific advice on this matter the group would be happy to discuss this inter-sessionally.

Term of Reference k) Dealt with under ToR e).

4 Term of Reference a)

4.1 Monitoring possible harmful algal blooms from satellite - review of recent developments and applications

Martin Hanson from the Swedish Meteorological and Hydrological Institute - SMHI Oceanographic Unit, Göteborg, Sweden opened this term of reference with a review of orbiting sensors and their application in the HABs area.

The advantages of Satellite sensing of HABs were described to include the fact that they offer a unique synoptic view of large areas of the oceans and provide regular monitoring (during cloud free conditions) repeat cycle about 0.5–6 days. They also show position of favourable blooming areas, ocean fronts upwelling, advection of water masses. (SST) and can improve model predictions. Other information such as inter-annual variability on extent, duration and occurrence can be mapped. When a HAB event has been confirmed with in situ measurement it is possible to monitor movements and development. In effect, remote sensing of HABs in combination with in situ monitoring and models result can be used as an efficient monitoring and prediction system.

However, certain disadvantages were also pointed out including HAB events must always be confirmed by in situ measurement, it is difficult to distinguish between species and also between other properties of the water; yellow substance, particles, etc. HABs usually occur in coastal waters where the rfesolution form satellites is poor, and

HABs are often present in too small amount to be detected but enough amounts to cause problems. Satellites rely on HABs being present in the surface layer, and also rely on cloud free conditions

A selection of applications using satellite derived data were presented:

Some notable applications included the Gulf of Mexico Harmful Algal Bloom Bulletin (Figure 4.1.1). In the Gulf area, harmful red tides (*Karenia brevis*) frequently occur and these can cause death to fish, birds and marine mammals. In addition eating shellfish from contaminated waters can cause NSP. Since 1999, Harmful Algal Bloom Bulletins have been issued by NOAA's National Centers for Coastal Ocean Science (NCCOS) to help coastal environmental managers Information and conclusions are based on satellite data (SeaWiFS), in situ measurements, models, wind observations Bulletins are issued frequently (once or twice a week)In South Africa a joint effort between the University of Cape Town, Marine and Coastal Management and the Benguela Current Large Marine Ecosystem (BCLME) programme provides near real time HABs information on a website (http://www.hab.org.za). The Benguela system is characterised by upwelling circulation along the entire west coast of southern Africa HABs are common, one or another dinoflagellate species and are associated with either high biomass or the toxicity of some species. Impact on both commercial and recreational interests, causing fish kills, contaminating seafood with toxins resulting in serious public health problems, or altering ecosystems.



Conditions Report

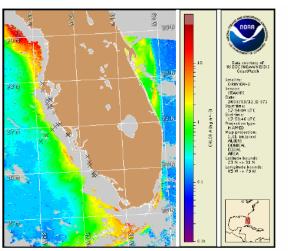
No impacts are expected in any Florida Counties this week. Due to current harmful algal bloom inactivity, bulletins are issued each Monday, until conditions warrant continuance of twice weekly bulletins.

Analysis

K. brevis was not present in any samples along the coast of Western Florida last week. Recent samples verified that patches of elevated chlorophyll at Boca Grande and Marco Island were due to nonharmful algae (FWRI). Discolored water is possible.

Chlorophyll levels remain elevated throughout much of the Lower Keys. No recent sampling is available. Samples from 3/1 (MML) indicated that K. brevis was not present on either gulf or oceanside of the Keys from Cudjoe Key to northeast of Marquesas Keys. Imagery from March 11 reveals the highest concentration of chlorophyll just west of Key West at 24°22.6'N 81°59.9'W. Sampling is recommended. Discolored water is possible from Key West to Marquesas Keys and along the oceanside Lower Keys.

- Stolz, Bronder



Satellite chlorophyll image with possible HAB areas shown by red polygon(s). Cell concentration shown as red squares (high), red triangles (medium), red diamonds (low b), red circles (low a), ora cles (very low a), green circles (present), and black "X" (not present).

Figure 4.1.1: Gulf of Mexico Harmful Algal Bloom Bulletin.

The Baltic Algal Watch System (BAWS) has been operational since 2002. This system produces daily map of extent of surface accumulation during bloom season. It is based on satellite data (NOAA-AVHRR and MODIS). An integrated web presentation for environmental managers and a public web site are available (www.smhi.se). The website provides information on bloom coverage, near real-time information, weather forecasts, model results, seatrack-web (Forecasts and dispersion of oil and chemicals, including surface accumulations of cyanobacteria) and incoming solar radiation.

4.2 Detection of harmful algal blooms and their toxins by *in situ* and remote techniques

Allen Cembella gave a useful overview of developments in the field of *in situ* and remote sensing of HABS

A wide array of emerging technologies have been developed within the last decade to specifically address the problem of detection and quantification of harmful algae and their toxins. Classical methods for cell enumeration and species identification are hampered by the requirements for a high level of taxonomic expertise, the time constraints imposed by tedious (and thus perhaps erroneous) manual counting and the difficulties of interpreting the results of point-source discrete sampling in long-term monitoring programmes. For example, discrimination of the various members of the genus *Alexandrium* by morphological criteria with microscopic methods generally involves careful study of diagnostic features (presence/absence and shape of the ventral pore; morphology of the apical pore complex) of individual cells, often with the aid of fluorochrome staining. Similarly, the diagnostic features of many species of the diatom genus *Pseudo-nitzschia* (e.g., arrangement and number of poroids on the valves) are at the limits of resolution of the light microscope and therefore misidentification at the species level is undoubtedly common.

From the perspective of phycotoxin assays and analysis, there are also urgent requirements for supplanting conventional whole-animal bioassays such as the AOAC mouse bioassay with more precise and refined methods for individual toxins or groups of toxins with a common mode of action. In both cases, it is desirable to move towards implementation of methods that offer real-time and continuous data on harmful taxa and their respective toxins for field deployment in monitoring programmes.

Deployable systems for bloom and toxin monitoring should be developed to fulfil three basic niches: surveillance, operational aspects and investigative requirements. Surveillance implies the ability to survey the target sites and regions of interest with the objective of providing information on early warning of impeding blooms, as well as on the subsequent stages of the bloom development cycle – including both qualitative and quantitative data. The operational elements refer to the function and integration of the technology into workable systems for field deployment, involving data acquisition and retrieval, use of the appropriate algorithms and calibration procedures, networking and reliability (e.g., with respect to biofouling), sensitivity, precision and accuracy of the measurements. As investigative tools, the systems should be amenable to incorporation into field-based research programmes on bloom dynamics and mechanisms of toxin propagation in marine food webs.

Field systems based upon general biooptical principles (fluorescence, spectral absorption, optical back-scatter, etc.) are best suited for detection and monitoring of high biomass HAB blooms, especially where the taxon of interest has a unique optical signature and where the bloom approaches monospecificity. By comparison, taxon- and toxin-specific methods will also work for low biomass blooms and complex assemblages where the target species may be dispersed and in low abundance. Among the taxon-specific probe methods, techniques based upon molecular targets at the cell surface (e.g. lectins, antibodies), in cell membranes, and moieties of intracellular proteins, nucleic acids and nuclear genes have proven to be most useful. As detection systems, many platforms, such as ELISA plate assays, epifluorescence microscopy, flow cytometry, and molecular approaches involving sandwich hybridization assays of extracted RNA and PCR-based methods have been successfully applied in the laboratory but most of these techniques have not been adapted for field deployment. Among laboratory methods, fluorescence in situ hybridization (FISH) coupled with epifluorescence microscopy, flow cytometry or solid-phase support cytometry (Chemscan) are among the most advance and widely employed for species discrimination. Operational fielddeployable systems are at present limited to the Environmental Sample Processor (ESP) developed at Monterrey Bay Aquarium, which employs a custom-printed oligonucleotide probe array to detect taxon- specific rRNA molecules by the principles of sandwich hybridization. An image of the resulting array is recorded using the CCD camera. An alternative system, developed from a prototype of a hand-held electrochemical detector (Inventus BioTech, Germany) is based upon detection of specific 18 rRNA sequences attached to a reporter probe and digoxigenin to generate an electrochemical signal. This system has been further developed to incorporate a multiprobe chip for simultaneous detection of 14 taxa in flow-through mode (Palm Sense, Germany); the intention is to incorporate this into a field system for cell-based detection (e.g. CytoBuoy, Netherlands).

In situ and portable ship board systems are particularly useful in monitoring and bloom dynamic studies in small-scale coastal embayments where the horizontal and vertical patchiness of the blooms renders satellite-based surveillance largely ineffective. Even the most advanced satellite-based optical sensors (SeaWifs, MODIS, etc.) have a minimal patch size of several hundred square metres and cannot provide data from below the surface, on cloudy days, or continuously. Aircraft-transported spectral sensors, such as CASI – compact airborne spectrographic imager – have been used successfully to map chlorophyll distribution and concentration with a spatial resolution of several metres with several meters of vertical penetration even in coastal waters, but again they cannot be used continuously or in poor weather. Such systems provide only limited taxonomic information based on spectral signature.

Developments in biooptical buoys, including for example the tethered attenuation coefficient chain sensor (TACCS) from Satlantic, offer the opportunity to continuously monitor the diffuse attention coefficient by spectroradiometric measurements – essentially a measure of ocean colour and the underwater light field. New hyperspectral sensors now provide more

information on the discrimination of phytoplankton pigment signatures as distinct from seston and CDOM but cannot resolve the profiles of individual species or in most cases even taxonomic groups (e.g. diatoms versus dinoflagellates) in complex assemblages without a dominating taxon. This technology is ideally used in conjunction with more cell-specific detection systems as a coordinated package.

It is now possible to conduct surveys of toxin profiles in the water column with on board liquid chromatography mass-spectrometry (LC-MS) systems. Underwater mass spectrometers do exist but thus far the marine toxins of interest cannot be detected with this technology. Nevertheless, such developments can be anticipated in the next few years.

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4.3 Regulation of alongshore Alexandrium transport in the Gulf of Maine USA

Don Anderson reported on developments in the use of AVHRR imagery rather than ocean colour imagery to detect conditions conducive to bloom development and transport in a Gulf of Maine *Alexandrium* event in 2005.

Many applications of remote sensing technology to HAB detection and tracking rely on ocean colour. There are locations, however, where HAB species do not make up a significant proportion of the plankton, and thus where pigment signatures cannot be used for detection. Alexandrium fundyense blooms in the Gulf of Maine are an example of this situation. Nevertheless, remote sensing technologies can be useful in management of paralytic shellfish poisoning (PSP) events – in this instance through a focus on sea surface temperature. In a recent paper by Luerssen et al (2005), relationships between satellite-derived sea-surface temperature (SST) patterns and the occurrence of PSP toxicity events caused by A. fundyense in the western Gulf of Maine were examined. Comparison between surface cell distribution patterns and SST images indicates that highest cell concentrations are associated with colder waters of the eastern segment of the Gulf of Maine coastal current (EMCC) and that frontal zones at the edges of the EMCC often act as boundaries to surface distributions. Surface thermal patterns can reveal enhanced connectivity between the EMCC and the western Gulf of Maine, suggesting transport linking A. fundyense cells in the EMCC to inshore areas of the western Gulf of Maine. Surface drifter data support such transport. Thirteen years (1990-2002) of toxicity data from eight monitoring sites along the coast of Maine and concurrent SST data show that in years of either large or very-reduced toxicity, a consistent relationship exists between the timing and strength of fronts, calculated from the SST data and taken as an

indicator of alongshore connectivity, and the occurrence and strength of toxic events. Years with weak fronts and/or fronts that become established relatively late in the summer growing season are years of the strongest toxicity events in western Gulf of Maine. Years of early and strong fronts are years with few and/or weak toxicity events. These results point to the utility of SST and other coastal observing system data for the monitoring and prediction of conditions linked to toxic events in coastal waters.

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4.4 HAB forecast for Karenia brevis in the Gulf of Mexico

Pat Tester described the utilisation of chlorophyll anomaly in the Gulf of Mexico to identify HABs of *Karenia Brevis*.

The HAB forecast for Karenia brevis in the Gulf of Mexico is based on ocean colour satellite imagery and is possible, in part, due to the lower backscatter of K. brevis compared to blooms of diatoms or Trichodesmium spp. (Carder and Steward 1985). Stumpf et al. (2003) suggested a simple chlorophyll anomaly might be useful to detect and monitor K. brevis. They developed a forecast model based on a 60-day running mean of chlorophyll when diatom and Trichodesmium spp. were not indicated. An anomaly is the difference in chlorophyll value per pixel between the image of interest and the 60-day running mean for the same pixel mean (lagged by 14 days to avoid the influence of a new bloom on the chlorophyll mean) (Figure 4.1.1). This was the first step in an early warning system to forecast K. brevis blooms in the eastern Gulf of Mexico and was available as an experimental product form 2001 to 2003 when the HAB forecast became operational on a regular basis with the frequency depending on the intensity of Karenia brevis blooms (http://coastwatch.noaa.gov/hab/bulletins). The bulletin also includes wind speeds and directions so potential surface movement of the blooms under the influence of the wind field can be judged. This forecast was evaluated (Tomlinson et al. 2004) and found to be accurate >83% of the time. There are plans to expand this forecast into the northern Gulf of Mexico in late 2006.

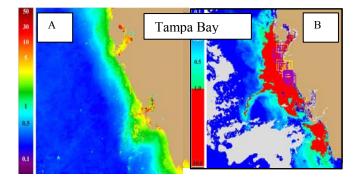


Figure 4.4.1: Harmful algal bloom forecast for *Karenia brevis* in the Gulf of Mexico is based on chlorophyll anomaly. A. 3 August–3 September 2001 Sixty-day running mean for each pixel in image. B) An anomaly is the difference in chlorophyll value per pixel between the image of interest and the 60-day running mean lagged by 14 for the same pixel in the image of interest. Anomalous chlorophyll values of 1 ug/L. are flagged in red. See Stumpf *et al.* 2003.

References

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4.5 Alexandrium measurements using remote sensing in Grand Manan Island in Bay of Fundy

Jennifer Martin showed an example where ocean colour imagery should be treated with caution. In 2003 highest cell counts of *Alexandrium* were not reflected in ocean colour (Modus or SEAWiFS) 888,000 cells/L in the next year with up to 4M cell/L still not able to tease out signal from ocean colour imagery

The presentation demonstrated with series of ocean colour images – areas of high chlorophyll that did not match where the high cell counts were recorded. It was noted that the variability between sea surface chlorophyll and high cell counts may not coincide due to mixing of the upper levels of the water column.

September 7, 2003 September 8, 2003 September 9, 2003 September 9, 2003 September 11, 2003 September 13, 2003 September 14, 2003 September 14, 2003

Figure 4.5.1: Chlorophyll concentrations in the Bay of Fundy 2003.

5 Term of Reference b)

5.1 Phytoplankton Monitoring in the Report of the Joint FAO/IOC/WHO Ad Hoc Expert Consultation for Codex Alimentarius on Biotoxins in Bivalve Mollusks (Oslo 26 September 2004).

Per Anderson gave a presentation on the section of the Report. This expert working group was made up of the following scientists who convened to draft guidelines for FAO/WHO/ IOC.

Expert Panel:

Dr Per Andersen, Bio/Consult, Denmark

Prof Tore Aune, Norwegian School of Veterinary Science, Norway

Dr Daniel G. Baden, University of North Carolina Wilmington, United States of America

Mrs Catherine Belin Ifremer Centre de Nantes, France

Prof Luis Botana, Univ. Santiago de Compostela, Spain

Mr Phil Busby, New Zealand Food Safety Authority, New Zealand

Dr Bob Dickey, US Food and Drug Administration, United States of America

Dr Valerie Fessard, French Food Safety Agency (AFSSA), France

Prof Lora E. Fleming, University of Miami, United States of America

Mr John Foorde, Marine and Coastal Management, South Africa

Dr Jean-Marc Fremy, French Food Safety Agency (AFSSA), France

Dr Sherwood Hall, US Food and Drug Administration, United States of America

Dr Philipp Hess, Marine Institute, Ireland

Dr Patrick Holland, Cawthron Institute, New Zealand

Dr Emiko Ito, Chiba University, Japan

Dr Tine Kuiper-Goodman, Health Canada, Canada

Dr Jim Lawrence, Health Canada, Canada

Mr David Lyons, Food Safety Authority of Ireland, Ireland

Dr Rex Munday, AgResearch, New Zealand

Prof Yasukatsu Oshima, Tohoku University, Japan

Dr Olga Pulido, Health Canada, Canada

Dr Michael Quilliam, National Research Council, Canada

Prof Gian Paolo Rossini, Universita di Modena e Reggio Emilia, Italy

Prof Michael Ryan, University College Dublin, Ireland

Dr Covadonga Salgado, Centro do Control do Medio Marino, Spain

Mr Joe Silke, Marine Institute, Ireland

Dr Gerrit I.A. Speijers, National Institute of Public Health and the Environment, the Netherlands

Dr Benjamin Suarez-Isla, Universidad de Chile, Chile

Dr Toshiyuki Suzuki, Tohoku National Fisheries Research Institute, Japan

Dr Andy Tasker, University of Prince Edward Island, Canada

Dr Hans P.van Egmond, National Institute of Public Health and the Environment, The Netherlands

Dr Phillippe J.P. Verger, Institut National Agronomique Paris-Grignon, France

Prof Takeshi Yasumoto, Japan Food Research Laboratories, Japan

Working group 3: was a sub-group tasked to draft guidelines on growing area management and monitoring

Working Group 3 Members

Per Andersen, Catherine Belin, Phil Busby (Chair), Henrik Enevoldsen, John Foord, David Lyons, Yolanda Pazos, Joe Silke, Covadonga Salgado

Task: Provide Guidance on Growing Area Management and Monitoring

1) Provide guidance on sampling methods for shellfish, including sampling depths, sample size, representative sampling, frequency of sampling

- 2) Provide guidance on the use of phytoplankton monitoring (strengths and weaknesses) as part of a shellfish marine biotoxin program
- 3) Provide guidance on indicator organisms for the different toxin groups
- 4) Provide information on the existence of biotoxin forming marine algae in various geographical regions of the world
- 5) Provide guidance on phytoplankton laboratories, accreditation, training of analysts, counting methods etc
- 6) Provide guidance on marine biotoxin management plans, including micro management
- 7) Provide guidance on environmental/hydrographic/oceanographic influences in growing areas
- 8) Provide guidance on sampling, including location of sample stations, sample size, use of indicator organisms, training of samplers, frequency of sampling, sample collection methods, transport of samples
- 9) Provide guidance on reporting, release and exchange of data

The Role of Micro-Algal Monitoring in Marine Biotoxin Management

Micro-algae (including planktonic and benthic organisms) are the primary source of biotoxins in bivalve molluscs. A marine biotoxin management programme should be described in a **marine biotoxin management plan (MBMP)**. The MBMP should include marine biotoxin action plans (MBAPs) for growing areas containing, for example, sampling strategy and requirements (frequency, sample size and composition), analyses to be carried out, and management action to be based on monitoring results and expert judgment.

Toxicity monitoring cannot be replaced solely by micro-algae monitoring. Information from micro-algal monitoring, especially if it is carried out regularly (for example weekly during harvesting), as part of a bivalve mollusc biotoxin management program, has particular strengths, including:

- Generally, observable concentrations of toxic micro-algae precede critical levels of toxins in bivalve molluscs and, therefore, allows management options to be considered, such as:
 - Precautionary closures:
 - Intensified monitoring or depth-specific sampling.
- Micro-algal monitoring can also help <u>focus</u> shellfish testing, for example on likely toxins, at the right location, at the appropriate time and when new toxin-producing species of micro-algae are found in an area.
- Micro-algal monitoring as part of an integrated biotoxin management program, is cost effective and operationally efficient.
- It may be used to investigate unknown, unusual or atypical toxic events.
- It may be used to provide information to set or use switching factors. These may activate associated management options.
- It may provide information not only on the onset of a toxic event but on the duration of any intensified management action.

Therefore, for early warning purposes and direct risk management activities it is recommended to have a program to monitor growing areas for species of toxin-producing micro-algae. The program should also include evaluation of other environmental conditions, for example wind, water temperature and salinity, which may suggest upwelling, stratification or mixing. These conditions may indicate that favourable conditions for a toxic event are developing.

However the weaknesses of such a system may include:

- Micro-algal observations may not accurately reflect the actual level of toxins in shellfish. In part this may be due to significant inter- and intra-species variability in toxin profile and toxin content for many micro-algal species even from the same area and over a short period.
- While micro-algae are the primary source of toxicity in shellfish, the toxins may remain in shellfish long after the toxic micro-algae are gone. Thus, the absence of toxic micro-algae cannot be taken as an indication that the shellfish are safe.
- Micro-algae are not always distributed uniformly in either time or space. "Patchy" distribution of micro-algae may make representative sampling difficult.
- The logistics of sampling offshore or remote areas, where scallops or clams for example are fished, may make micro-algal monitoring less cost effective.
- Special monitoring arrangements may be necessary to address the problems posed by benthic species of toxic micro-algae, for example *Prorocentrum lima*.

In conclusion, decisions made on the safety of shellfish can only be based on the direct measurement of toxins in shellfish flesh. However, an integrated shellfish and micro-algal monitoring programme is highly recommended to provide expanded management capability and enhanced consumer protection.

Furthermore, recent developments indicate that micro-algal monitoring coupled with operational oceanographic, meteorological, and remote sensing data, including modelling and other measurements may be used to base advice on the imminent onset of harmful events.

6 Term of Reference c)

6.1 A summary of the intercalibration workshop on cell counts held in Kristinaberg

Presented by Bengt Karlson, Caroline Cusack and Eileen Bresnan.

This workshop was held to compare traditional and novel methods for counting cells under controlled conditions, in order to attempt to identify the state of the art with regards to cell enumeration. The following objectives were set to be addressed by the workshop:

- Used Alexandrium ostenfeldii and A. fundyense;
- Examine traditional microscope methods for cell abundance to determine if traditional and molecular or new methods would provide comparable results;
- Is there one method that can be recommended?
- Used nine traditional methods, eight molecular methods and FlowCam, 100 ml used for all:
 - Utermohl's, settling bottle, counting chambers;
 - Molecular techniques used different versions and different personnel.

Bengt summarized findings

Generally the counting chambers with the small volumes were less reliable at low cell concentrations. At 500 cell/L results somewhat more consistent. Samples were fixed and sent to Canada to J. Martin's lab for FloCam analyses. In experiment 4 the numbers were in good agreement.

Problems observed:

- Time constraints;
- Limited sample volume;
- Aberrant cells from cultures observed;

- Some labs not perfect for molecular biological work;
- Not correct temp in incubation ovens;
- Some methods were not properly calibrated for the material used.

Did not test:

- Total phyto community;
- Biomass;
- Other species that target.

Results:

- Short report distributed on ICES website;
- Scientific article to Harmful algae;
- IOC Manuals and Guides A practical guide to quantitative phytoplankton analyses.

Conclusions: Successful:

- 24 participants from three continents;
- 18 methods for quantitative phytoplankton analysis were compared;
- Approx 1000 samples were made up and analyzed;
- Inter comparison focused on only one species A. f.

Financial support was acknowledged from IOC, BIM Ireland, and Marine Institute Ireland. SMHI Kristineberg Marine Research Station supported lodging.

Comments:

- A discussion of the workshop followed It was felt that the expectation was that traditional methods would not be good at identification of cells but would be better at enumeration. As it turned out this was not really the case, however, some the traditional methods were not as good at cell ID as expected. Some of the molecular methods were better at enumerating cells than expected;
- It was pointed out that one did not really have a big challenge in cell identification and that this was the best case scenario and any natural samples would have greater error;
- Overall the working group said that the exercise was successful. Some of the tests
 were under some constraints. Molecular techniques were difficult to set up –
 needed more lead time than microscope;
- The organizers were commended for their fine work in setting up and conducting a successful workshop.

7 Term of Reference d)

7.1 Update on the ASC theme session entitled "Harmful Algae Bloom Dynamics: Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge"

The Annual Science Conference will host a theme session jointly convened by Patrick Gentien (France) and Tapani Stipa (Finland). Dr Gentien provided information on this session for the working group.

In spite of large gaps of basic process knowledge around HAB dynamics, several 3-D 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 3-D modellers and biologists (including invited contributions from GEOHAB) interested in explaining:

- Why HABs occur,
- How HABs are initiated,
- How and why HABs develop in space and time,
- Why HABs decay

and to demonstrate:

- Existing 3-D modelling capabilities,
- Status on validation of such models,
- The need for observations (satellite and *in situ*).

The working group asked that a review of this theme session would be presented at the 2007 WGHABD.

8 Term of Reference e)

8.1 Review progress and analyses that REGNS North Sea Group have done on datasets submitted by members of WGHABD (to meet in the interim)

Einar Dahl presented an update on progress on datasets and analysis by REGNS Integrated Ecosystem Assessment of the North Sea – An ICES Pilot Project.

Objectives:

- 1) Look at ways in which the existing ICES structure (data centre & working groups) can input into the periodic production of regional integrated assessment.
- 2) To deliver a pilot Integrated Assessment (not advice) on the North Sea Ecosystem by September 2006.

North Sea – hope to get information from a variety of working groups – HABS related to eutrophication is the preferred data set for this assessment. Ended up with data from five different sources

- Plankton recorder, chl;
- Physical data;
- Fish landing by species;
- Sea birds;
- Fish assessment.

HAB data might go back to early 1980s and therefore care must be exercised in incorporating these data sets to ensure region specific artefacts are not incorporated. What is needed is REGNS gridded data for analyses and spatial and time series and for HABs most data is collected by regional programmes rather than on a wider geographical scale.

The presentation demonstrated specific areas were identified with PCA and mapped those regions that clustered together on PCA.

Conclusions: Clear gradients were evident in space and time. State changes were seen in 1988 but also in 1965 and 1979. The cause appears to be related to Sea water flux into the Northern

North Sea The weight given to the different parameters needs investigation – not all parameters are of equal ecological significance – expert input required. Sub-regions could be defined and the thematic assessment undertaken – now asking if HABS could fit in to the assessment. The working group felt that that the HAB data sets might be too short to be useful but perhaps some sources may have long data.

It is hoped that the value of undertaking this type of integrated assessment would be to help the future design of monitoring programs and the setting of realistic policy targets for management purposes

Lessons learned:

- Took time to understand the concept and definitions different user needs;
- Identify data sources was easy but not easy to access;
- Value action gap too many vested interests;
- Data gathering and checking not complete;
- New methods for analysis required still.

This work is not yet complete; however the preliminary overview assessment is the source of the existing information. A further meeting of the Regional Ecosystem Study Group for the North Sea (Chair: A. Kenny, UK) will meet at ICES Headquarters from 15–19 May 2006 to:

- a) Hold a workshop to evaluate and plan the finalization of the 2006 integrated ecosystem assessment for the North Sea, to be presented at the 2006 ASC;
 - review the outcome of the work of an intersessional correspondence group (sub-group of REGNS) with compilation and analyses of a comprehensive integrated data set for different aspects and components of the North Sea ecosystem;
 - ii) review the outcome of intersessional work on relating state variables of the ecosystem with human pressures according to themes (eutrophication, pollution, conservation, fisheries, climate, and management);
 - iii) prepare plans for finalization of the integrated ecosystem assessment which must take account of the relationship between the thematic human pressures assessments (in ii above) and the overview integrated assessment (in i) above);
 - iv) prepare for presenting the outcome of the integrated ecosystem assessment at the 2006 ICES Annual Science Conference;
- Advise on follow-up work to translate the experiences of REGNS in producing an integrated ecosystem assessment into a regular process in ICES of producing or contributing to the production of updated integrated assessments for the North Sea ecosystem;
- c) Based on the experience with the production of the 2006 North Sea integrated assessment; consider requirements that need to be taken into account in a design of a holistic monitoring of the North Sea ecosystem.

REGNS will report by 30 June 2006 for the attention of the Resource Management Committee, ACFM and ACE.

9 Term of Reference f)

9.1 New findings that pertain to harmful algal bloom dynamics:

9.1.1 BOHAB

Joe Silke and Caroline Cusack

Biological Oceanography of HAB species in Ireland

The National University of Ireland, Galway, Woods Hole Oceanographic Institute and the Marine Institute carried out a research programme entitled The Biological Oceanography of Harmful Algal Blooms (BOHAB) between 2003 and 2005. The primary objective of this work was to collect physical and biological information, which is associated with the generation of Harmful Algal Blooms (HABs) in Irish waters. Using this information the project worked towards the production of a predictive tool, which could potentially be used to mitigate the effects of these economically damaging events. The west of Ireland supports the bulk of Irish aquaculture activities. The two most important aquaculture areas in this region were chosen to carry out the detailed part of the research, Killary Harbour and Bantry Bay.

The Irish aquaculture industry has become a vital part of Irelands coastal economy since its initial development in the early 1970s. The diversity of sites used and the species farmed have also increased. The sector grew in output value from €37.2 million (26 500 tonnes) in 1990 to a peak in 2002 of €125 million (61 000 tonnes). Unfortunately there have been several protracted occurrences of Harmful Algae that have slowed down the development of the industry, and in some cases have been responsible for major mortality of stock and intoxication of shellfish.

HABs in Ireland have caused the greatest impact on shellfish farms, but have also been responsible for mortalities on caged finfish farms. Mussels, Pacific oysters, Native oysters, clams and scallops are the main shellfish species being produced in Ireland at present. Mussels, which are farmed using both suspended ropes (intensive) and bottom-culture (extensive), account for 80–90%, by volume, of annual shellfish production. Oysters (principally Pacific oysters) account for a further 10–15%. Shellfish farming is practised in every coastal county with the exceptions of Wicklow and Dublin. Shellfish species farmed on a smaller scale include abalone and purple sea-urchins.

From the point of view of shellfish producers who are on the front line of HABs, there is a requirement for a rapid and accurate response to the problems associated with algal blooms through a robust means of monitoring and management in order to protect their industry. In addition the prediction of closures due to HABs is important to schedule operational and marketing activities.

It is essential that these basic principles be in place for the development of the industry because the parallel objectives of providing consumer safety and development of the industry are top priorities. The emphasis on researching the means for early warning of the onset, intensity and duration of toxic events therefore are critical for to putting these management strategies in place, and this was the key component of this project.

The themes of the work proposed in this project were chosen to assimilate the knowledge obtained through the scientific aspects of monitoring programmes and oceanographic research, and to gather necessary information that may be used in the design of a predictive model of the onset, intensity and duration of a HAB event.

It was recognised that our understanding of oceanographic processes, in particular the physical processes which directly affect phytoplankton composition and dynamics, may be used to

develop early warning capabilities. For example, research over the last decade has shown that temperature of the water column taken at several depths is a good indicator to measure water and phytoplankton exchange events. The deployment of a simple thermistor string in certain southwest sites is therefore adequate to give fundamental information on potential HAB events. Further biological information on the uptake of toxic plankton by shellfish and the kinetics of toxicity were studied to understand the system so that a basic level of predictive modelling/forecasting can be achieved.

Building upon the BOHAB themes, a common research objective for producers and regulators should be a preventative approach to fish mortalities and shellfish toxicity. Real-time management could complement existing aquaculture and water quality monitoring programmes using agreed standard assessment methods, tools and indicators throughout the EU.

A holistic approach to the management of shellfish toxins has been used in Ireland in recent years. It incorporates the results of bioassay, toxin chemistry, phytoplankton presence, spatial and temporal factors such as high-risk time of year and status of neighbouring areas. This has been used to recommend voluntary action by the industry. The current management of harmful phytoplankton is, however, for the most part, put in place once a bloom is present in an aquaculture area. In some cases fish mortalities or toxic shellfish have already been harvested. Mitigation and prevention of the harmful effects rely on being in a position to predict the onset of a bloom.

One means of attempting this is to look at the relationship between climatic events, changes in phytoplankton composition, and the occurrence of harmful events.

Using this approach, a simple model of the onset of Shellfish Toxicity in the South West of Ireland has been attempted during BOHAB. This is based on the current and forecasted meteorology and time of the year. This model only applies to the SW due to the alignment of the bays in this area, and the fact that there are many years of phytoplankton and toxicity data to develop and test the model. It is, however, intended to research the potential for this approach to be adopted in other bays around Ireland.

From historical records, it can be seen that most of the toxic events in the South West have been preceded by an initial easterly wind, reverting back to the predominant south-westerly wind. When a population of potentially toxic phytoplankton are present, these conditions can allow the passage of these blooms into the bays of the southwest where they can cause toxicity problems. The coupling of these oceanographic events with the biological pathways of harmful algal events is the most promising approaches to predicting the onset of blooms. BOHAB has made considerable progress in assimilating data and studying the nature of HABs in Irish waters. Future studies can benefit through provision of essential baseline data and a conceptual framework to continue the task of generating a robust and accurate predictive model for HABs and improve the potential for aquaculture in Ireland's coastal waters.

9.1.2 Flowcam in Phytoplankton enumeration

Jennifer Martin

Advantages

- 1) When using a FlowCAM for phytoplankton enumeration the amount of manual labor in sample processing/handling is greatly reduced. The operator set up to begin a run, but only occasionally monitors it while the FlowCAM is running and detecting particles as pass they through the flow cell.
- 2) The FlowCAM provides a "non-biased" digital record of every particle within a specific size range (determined by the operator) for future analysis. With a traditional microscope, the operator looks as seeks out the particle of interest providing a biased count of a sample (depending on operator identification knowledge). With the data generated by the FlowCAM each particle is archived and if problems arise the data/images for each particle and/or cell can be reanalyzed at a later time. In the case of this intercalibration study, other species that were present in the sample could also be identified and enumerated, such as, *Dinophysis, Protocentrum, Ceratium*, heterotrophic dinoflagellates and some nauplii.
- 3) In addition to an image for each particle detected by the FlowCAM, the software provides instant image analysis on each detected and captured particle during a run. For example, particle length, width, equivalent spherical diameter (ESD), area-based spherical diameter (ASD), fluorescence (if applicable), time of flight, cells per image and aspect ratio are the primary data obtained. Given the data provided by the FlowCAM the operator can develop specific algorithms for values of interest such as bio-volume, depending on the needs of the application.
- 4) The FlowCAM is a portable. Even the bench-top version used in this intercalibration study has a relatively small foot-print and can be used in the lab or at sea on board ships. The image capture system prevents problems usually associated with vibration.
- 5) The FlowCAM allows for the visualization and or detection of a wide particle size range (1 um 1 mm). To detect across this large size range a series of objectives and flow cells would need to be used. Based on the size of the target organism to be detected in this inter-calibration study (*Alexandrium*), a 10x objective with a 100um depth flow cell was utilized. To examine smaller or larger particles with a sample more effectively other objectives and flow cell combinations would need to be utilized.
- 6) The FlowCAM allows for 3 different modes of detection. In this inter-calibration study, fluorescence detection mode was used. Fluorescence based detection allows for the detection and capture of fluorescent particles (containing either red or orange fluorescence indicative of chlorophyll and phycoerithrin). Depending on the application other detection modes either scatter detection or Auto-detection mode may be used.

Disadvantages/Drawbacks

- 1) In terms of cell identification it is very important to have the best focus possible, otherwise the images will be blurry and will be difficult to analyze.
- 2) Depending on the ecosystem that is being analyzed the phytoplankton cell size range may vary greatly. Each objective and flow cell size that can be used has a minimum and maximum cell size range that's possible (similar to the limitations of microscopy). Therefore, to best identify all phytoplankton within a given ecosystem, from 5 um particles to cells that are 100 um in size may require two FlowCAM runs one at 4x magnification using a 300 um depth flow cell and a 10x magnification using a 100 um depth flow cell. It all depends on what magnification is acceptable for cell identification. Therefore, method development at the beginning of a particular project is important. However, if cell identification is not necessary but particle/cell size is only one FlowCAM run may be necessary.

- 3) In situations were cold samples are to be analyzed in a high humid environment (deep CTD casts on board ship) there is a problem of water condensing on the flow cell that will prevent particle detection. Solution allow the sample to warm up prior to analysis not ideal, however, the sample could be preserved prior to manipulation.
- 4) When particle load to very high (detritus), as is riverine samples, there maybe more than one particle captured in a single field of view of the camera. Although, each particle will have different image analysis values, such as, particle length, width, ESD etc... the fluorescent value of both particles will be the same. Also it is possible to capture "non-fluorescent" material if the particle load (sediment) is too high if the instrument is detecting particles in fluorescent detection mode.

Type of training needed

- a) **perform task:** in order to perform the task very little training is needed to provide basic knowledge of sample running and analysis probably would require 1 day with guidance from a trained individual.
- b) **Troubleshoot and QC:** In order to troubleshoot the instrument a more experienced person (up to six months experience) would be more effective at troubleshooting the instrument. The company that manufactures the FlowCAM, Fluid Imaging Technologies provides over the phone customer assistance when needed.

Essential equipment: The FlowCAM, either the Bench-top or Portable Version

Cost per sample, consumables

Minimal and hard to calculate – the flow cells are the only disposable component to the instrument and only need to be replaced when broken or too dirty (something lodged in the flow cell). These can be made or purchased from Fluid Imaging Technologies. In some cases a single flow cell may last a few months if kept clean.

Handling time per sample

- a) **Processing** this will very greatly depending on the particle load in the sample and the flow at which the sample is processed (pump speed). When samples are concentrated such as in cultures or net tows the samples will need to be diluted however, if a sample is too dilute then a sample can be concentrated using nitex sieves. In this intercalibration study for the detection of *Alexandrium* the samples appear to be VERY dilute most of the time. In order to minimize sample manipulation each sample was processed at full strength and was NOT concentrated. In this case 20 mL were processed per sample and better results may have been obtained if sample concentration was performed. However, prior knowledge of each sample was not allowed. Due to the volume needed for processing and the flow cell size used the processing time for each sample was ~ 40 minutes. In most cases for natural field sample usually a processing time of ~20 minutes would be used.
- b) **Analysis time:** After a sample has been collected it does not need to be analyzed immediately as each sample is digitally archived. Usually, samples are processed in batch runs and then analyzed all together. Analysis time with vary depending on particle load in the sample, however, based on the low cell concentrations observed in each sample during this study analysis time ranged from 5–15 minutes per sample.

Sample throughput/person/day: Again depends on sample particle load, flow cell size, and flow rate. For this inter-calibration study between seven to nine samples were run per day. However, if processing time could change due to concentration the processing time would decrease and sample throughput would increase. Note, that only minimal monitoring of the FlowCAM is required during processing.

9.1.3 Spirolides and Micro-cystin Chemistry

9.1.3.1 Accumulation of double methylated Spirolides in mussels and oysters – a possible risk?

In the middle of the 1990s a new class of neurotoxins was discovered in aquaculture sites along the East Coast of Canada (Nova Scotia) [1]. These toxins were found to be so-called "fast acting" toxins causing death within several minutes when injected intraperitonally into mice. The molecular structure of those substances designated as spirolides consists of a spirolinked, tricyclic system of polyethers and a seven membered spiro-linked cyclic iminium moiety. First, spirolides were isolated from shellfish and later on from plankton samples [2,3]. In 2000, Alexandrium ostenfeldii as the causative organism for the production of spirolides was identified [4]. Whereas spirolides belonging to groups A-D are biologically active, the spirolides E and F, which could have been isolated only from shellfish, show no biological activity. Probably spirolides E and F are metabolites originating from a hydrolysis of the cyclic iminium moiety present in the spirolides A and B to an opened keto amine. The instability of the cyclic iminium function under enzymatic or acid conditions can be important with respect to an oral toxicity of spirolides. Especially the fact, that spirolides C and D, which wear two vicinal methyl groups at the cyclic iminium moiety, are enzymatic- and acidresistant, whereas spirolides A and B with only one methyl group are transferred to biologically inactive spirolides E and F under same conditions, is very informative. Obviously the presence of two vicinal methyl groups can prevent an enzymatic or acid hydrolysis of cyclic imines.

The Alfred-Wegener-Institute for Polar- and Marine Research (AWI) on Helgoland Island succeeded in cultivating a Danish strain of *Alexandrium ostenfeldii* (KO287). Subsequently its extracts were tested for spirolides by LC-MS/MS. It could be shown that strain KO287 contained only spirolides with two vicinal methyl groups. For a thorough study of the metabolisation of spirolides in shellfish, extracts from tissues of edible mussels were incubated with an extract of *Alexandrium ostenfeldii* (KO287) for several hours. In a subsequent measurement of the extracts from mussels' tissues by LC-MS, no characteristic differences in the spirolide profile compared to the profile of *Alexandrium ostenfeldii* (KO287) were obtained. Thus a metabolisation or hydrolysis of spirolides contained in *Alexandrium ostenfeldii* by enzymes present in mussels can be excluded.

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9.1.3.2 Co-Occurrence of microcystins and their demethylated variants – Only an analytical problem?

In summer 2005 phytoplankton and water samples of the lake "Senftenberger See", Germany, were analyzed with regard to occurrence of microcystins (MCs). Analysis was focussed on the microcystins MC-LR, MC-RR, MC-YR using an HPLC/UV method according to the European norm ISO 20179. In addition, the upper limit for MC-LR of 1.0 microgram MC-LR per litre in drinking water, given by the WHO, was considered.

To confirm the HPLC/UV results the LC/MS-MS coupling was applied. The samples were analyzed again. Although the same sample extracts and the same commercial MC standards were analyzed, the results obtained by mass spectrometric detection revealed lower concentrations of the microcystins MC-LR, MC-RR, and MC-YR in the sample extracts in comparison to the HPLC/UV determination. On the other hand, by LC/MS-MS, the corresponding demethylated variants of these microcystins could be detected additionally.

Hitherto chromatographic separation of these MCs was not achieved, because of the structural similarities of the MCs and their demethylated variants. Hence, MCs and their demethylated derivates co-eluate from HPLC column result in a peak comprising of both components, which can not be resolved by UV detection. Consequently, the use of mass spectrometric detection in SIM mode is necessary for correct MC quantification.

However, the unambiguously determination of demethylated microcystins is not only a need from analytical point of view. Here is only scarce knowledge about the toxicological relevance of these demethylated MC variants and studies with regard to the hepatotoxicity of these cyclic peptides are necessary. In addition, analytical methods for control of occurrence of microcystins in waters and aquatic organisms as food source have to be modified to guarantee valuable data for assessment the threat caused by microcystins in the environment.

9.1.4 Karenia mikimotoi Bloom

Report on an exceptional bloom in Ireland 2005

Joe Silke

A protracted bloom of *Karenia mikimotoi* was present in Summer 2005 along the northern half of the western Irish coastline. The onset of this bloom was identified in late May / early June. This event subsequently dissipated over the month of July and was succeeded by a bloom of the same species in the southwest in late July. The bloom was very intense (up to 3.7 million cells per litre) and resulted in discolouration of seawater and foaming in coastal embayments. Major mortalities of benthic and pelagic marine organisms were observed and a complete decimation of marine faunal communities was reported and observed in several locations. Deaths of echinoderms, polychaetes and bivalve molluscs were observed in County Donegal and Mayo, while farmed shellfish and hatchery raised juvenile bivalve spat suffered significant mortalities along the Galway and Mayo coasts. Reports of dead fish and crustacea were received from Donegal, Galway, West Cork and Kerry.

Karenia mikimotoi is one of the most common red tide causative dinoflagellates known in the Northeast Atlantic region, and is also common in the waters around Japan. Blooms of this species often reach concentrations of over several million cells per litre and these densities are often associated with marine fauna mortalities. Although cytotoxic polyethers have been

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extracted from cultures of the species, the exact mechanism of the toxic effect and resultant devastating damages yet remains unclear.

The visible effects following the mortalities included noticeable quantities of dead heart urchins (*Echinocardium cordata* L.) and lugworms (*Arenicola marina* L.) deposited on beaches. Several species of wild fish were also found washed up dead. The bloom coincided with a period of fine weather and tourists visiting the seaside were concerned about the safety of swimming in waters that were obviously harmful to marine organisms on this scale. A public awareness programme was mounted by the Marine Institute with several radio broadcasts, press releases and a website provided to give up to date pronouncements on the event.

While there have been several instances of *Karenia mikimotoi* blooms reported in Ireland over the past 30 years, this scale of mortalities associated with the 2005 bloom were not previously observed. Recording of the scale of this event was facilitated by satellite imagery from the MODIS platform while direct counts of the cells in seawater by the Marine Institute monitoring programme gave very useful information regarding the size and intensity of this event. The mortalities of marine organisms were documented from reports made by various observers and by Marine Institute field surveys. A full report was prepared and published by the Marine Institute

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9.1.5 New England Alexandrium fundyense bloom

Overview of the 2005 Alexandrium bloom in southern New England

Don Anderson

Blooms of the toxic dinoflagellate *Alexandrium fundyense* are responsible for outbreaks of paralytic shellfish poisoning (PSP) in the Gulf of Maine. This organism has been studied extensively, most recently through the five-year ECOHAB-Gulf of Maine program. This talk will first summarized ECOHAB-GOM data and the conceptual models that have been formulated to explain field and toxicity observations. Data were also presented on the extensive 2005 bloom that closed shellfish beds from central Maine to Massachusetts, including Nantucket Island, portions of Martha's Vineyard, and 40,000 km² of offshore federal waters.

Initial observations suggest that several factors contributed to the 2005 bloom. Abundant rainfall and heavy snowmelt substantially increased the amount of fresh water entering the Gulf of Maine at a critical phase of the bloom season. We hypothesize that this provided micro-nutrients, increased stratification, and augmented alongshore transport that led to high cell abundances and a broad, region-wide dispersal of the organism. Warm temperatures in western waters would have favoured *A. fundyense* growth in that region. In addition, several storms with strong winds out of the northeast occurred when cells were abundant and in locations where the wind-driven surface currents could advect them into Massachusetts Bay and keep them there, leading to high cell concentrations and toxicity. Another contributing factor may have been the high abundance of newly deposited cysts in western Gulf of Maine sediments, as documented in a fall 2004 survey. The cyst abundance in 2004 showed a 10-fold increase over an earlier 1997 survey. Post-bloom surveys in the fall, 2005 showed that cysts are present in very low abundances in the southern region, while the cyst abundance within the mid-Maine coast are reduced by about half (vs. 2004). The cyst abundance within

Massachusetts waters are quite low, and suggest that localized, in situ bloom development within Massachusetts Bay or southern waters is unlikely. On the other hand, there are still a significant number of cysts in western Gulf sediments – five times as many as there were in 1997, and only slightly less than were there in 2004, so there is potential for another regional event in 2006. Variability in cyst abundance may be an important factor in the inter-annual and inter-decadal bloom dynamics in the western Gulf of Maine.

In retrospect, the factors leading to the 2005 bloom were unusual in several respects (cyst abundance, runoff volume, major storms). A confluence of such factors should be viewed as a rare event - i.e., an event of similar magnitude is not likely in the near future. Nevertheless, we may have entered a "new regime" with more frequent and more intense toxicity in western Maine and southern New England, based on historical patterns and the high levels of cysts observed in the region. Superimposed on this "new regime", we should expect considerable inter-annual variability in blooms, driven by a variety of both small- and large-scale forcings.

9.1.6 AZA in crabs

Detection of Azaspiracid in crabs (*Cancer paguris*)

Einar Dahl

After recordings of DSP-toxins above quarantine levels in crabs (Cancer paguris) along the southern coast of Norway in 2002 a monitoring of algal toxins in crabs was set up along the Norwegian coast. In the autumn 2005 up to 277 microgram AZA-1 per kg brown meat was measured in crabs collected in northern Norway. Neither the source organism(s) nor the mechanisms for accumulations of the algal toxins in the crabs are known.

- AZA toxins were detected in crabs in northern Norway for the first time in autumn 2005;
- Up to 277 ug AZA-1 equivalents per kg brown mean measured. The quarantine level should be 80 (half the amount recommended for mussels);
- The source organism(s) is unknown contaminating *Cancer paguris*;
- In summer in southern coast will come up and eat blue mussels;
- On other coastal area do not know if crabs come up to intertidal to feed;
- This incident was on the mid coastal region on west coast;
- Industry did not suffer much with closure;
- Assay was by chemical method LC-MS;
- Will continue to monitor crabs.

9.1.7 Dinophysis in the Swedish Skagerrak

Bengt Karlson

- *Dinophysis* spp cell numbers 1990 2003 Swedish Skagerrak;
- Highest numbers in July;
- If we go to *D. acuta* there are too many missing data to provide good pattern. They are more abundant in the later months of the year (fall);
- D. acuminata is a spring / spring summer occurrence;
- D. norvegica is a spring summer with highest numbers July;
- D. acuta and D acuminata seem to be responsible for DSP not D. norvegica.

It was pointed out that it is quite important to determine *Dinophysis* to species because of the difference in toxicity and the difference in seasonality. It is generally held that *D. acuta* is the most toxic.

9.1.8 Alexandrium cell abundance in Bay of Fundy

Jennifer Martin

Cysts are abundance in central Bay of Fundy and there is central gyre in the Bay and cysts could be distributed throughout the Bay. There is regular monitoring at specific locations since late 1980s and at other locations from late 1990s.

An analysis of existing monitoring data was carried out to examine bloom characterizes, time of onset, duration, max concentration, number of blooms per year, extent of blooms and interannual trends to see if there are trends that allow prediction.

It was found that there is considerable seasonal variation however, blooms occur between about day 130 (May) and day 230 (October). It is not uncommon to have 3 blooms each summer but the timing of events is variable.

Peak counts are approx. 100,000 cells/L and the duration of presence of *Alexandrium fundyense* is inter-annually variable from 50–200 days but typically \sim 120 days with the first bloom is always lower that second bloom. It was also noticed that median max concentration decrease from offshore to inshore.

10 Term of Reference g)

10.1 HAEDAT, the Harmful Algal Event Data-Base of IOC-ICES-PICES

A review of the on-line format of HAEDAT submission form and evaluation of the amendments made to update historical submissions and links to mapping was presented by Monica Lion.

HAEDAT primary function is data archiving and can provide summaries of data (species, where, when, conc., effects etc), as well as information on which data exist and where. HAEDAT contains reports from 1987 to 2004 (1797 records) from the North Atlantic region, and as from 2000 from the North Pacific.

HAEDAT has been available since 1999 at http://ioc.unesco.org/hab. It is the ambition that HAEDAT will become the global database on harmful algal events and cover the North Atlantic (ICES), North Pacific (PICES), Caribbean (IOC-ANCA), South America (IOC-FANSA), Mediterranean (CIESM), North Africa (IOC-HANA). Areas where no mechanism for compilation has been identified include Australia, New Zealand, South East Asia, Central and Southern Africa.

Since the WGHABD meeting in 2005, HAEDAT has been transferred from desktop solution (Microsoft Access) requiring download by user to web-based solution (MySQL/PHP). The database has been 'normalised' (arranged into tables according to data types) and data has been harmonised. This has been a much more timing consuming task than first anticipated.

Web forms for online submission and update of records are now ready (format and layout otherwise unchanged), and data has been exported from the old to the new database. The online browser for records is ready with only minor adjustments to be made. The WGHABD revised the input form and found no need for revisions.

11 Term of Reference h)

11.1 Structure and composition of the decadal HAE maps

A further presentation was given by Monica Lion on the progress to review the structure and composition of the decadal HAE maps for the ICES region with special reference to clarifying

the distinction between harmful algal blooms and the harmful affects that are reported on the maps. In particular, the registration of cyanobacterial blooms in brackish and marine waters should be re-visited from the emerging perspective of their known toxicity and implicit harmful effects.

The major next step in the development of HAEDAT is the development of online searchable maps using a GIS-type interface. It is also the plan to continue to work on the extension to a full HAE information system, integrating data related to the events such as taxonomy (IOC Taxonomic Reference List), monitoring programmes (MON-DAT), HAB-MAP by ISSHA etc.

The existing ICES-IOC Decadal Maps provide information on presence of toxins or observations of mortalities (regardless of the level of toxicity). Modifications are provided by each country only as dots on maps. There are no data files.

In order to prepare for maps generated based on HAEDAT records, zones/grids have been defined/redefined for all countries, except Iceland, Belgium, Lithuania, Faroes & Greenland (Denmark). Each country has defined its appropriate zones. This implies that HAEDAT zones are now permanent to be able to compare one year to another.

In the working document for the development of the HAEDAT mapping function the following parameters have been chosen:

- **General information**: Nature of the event;
- Microalgae: Causative Species, Causative Genus, Causative Class;
- **Environment**: Resources affected, Syndrome, Toxin type, Toxin;
- **Location and date**: Year, Area code, Country, Regions (ICES, PICES, HANA etc.).

The template will build on MarBef Maps or similar systems. The WGHABD advised that a system that allows export of data to other GIS systems/products (Web Services Standards) should be sought (e.g. the functionalities provided with MapServer). Funding for the development of the mapping application is sought and success herein will decide when the mapping part of HAEDAT will be ready. The WGHABD agreed that each member of the WGHABD will explore with their respective GIS colleagues possibilities for partnership and sponsorship with IOC to develop the mapping application.

12 Term of Reference i): National Reports

12.1 U.S.A.

2005 was a year with several noteworthy or exceptional HAB events in the U.S., with many other areas having "normal" levels of bloom activity.

PSP. Similar to previous years, Maine, California, Washington, and Alaska all recorded PSP events during 2005. Unlike previous years, however, there was no PSP event in Oregon in 2005. The most noteworthy event was an extensive bloom of *Alexandrium fundyense* that occurred along the coast of southern New England from May to July 2005. The outbreak eventually closed shellfish beds from central Maine to Massachusetts, including Nantucket Island and portions of Martha's Vineyard, and resulted in the closure of 40 000 km² of offshore federal waters as well. The coastal *Alexandrium* bloom was exceptional in several ways: high toxin levels were measured farther south than ever before in New England; levels of toxicity in may locations were higher than previously observed at those stations; for the first time toxicity at some locations was above quarantine levels; cell concentrations far exceeded those observed in the coastal waters of southern New England in the past, and for the first time in the region the governors of Maine and Massachusetts officially declared the red tide to be a

disaster, clearing the way for federal assistance. This was not just a huge southern New England event, but Maine ranks it as the second worse outbreak in 30 years.

During 2002, the first Puffer Fish Poisoning (PFP) attributed to saxitoxin was reported in the U.S. (Florida – east coast). In 2005, for the first time, there were closures of Florida shellfish beds due to PSP toxins. There were no human cases of PFP in 2005.

ASP. ASP was recorded in California, Oregon, and Washington. Similar to the last three years, the outbreak in California was extensive, encompassing the entire coastline, with moderately high toxicity –290 ppm domoic acid being the highest reported (in lobster viscera). In Oregon, the south coast was closed most of the year to razor clam and mussel harvesting, and these areas remain closed in 2006. The north coast experienced closures in April and May and recorded the highest level of toxicity seen in mussels for the state of Oregon (128 ppm).

NSP. The other noteworthy regional bloom of 2005 was an extensive bloom of *Karenia brevis* that bloomed in southwest and northwest Florida. The southwest Florida bloom lasted through <u>all</u> of 2005 (beginning in early January and has continued into 2006). Fish, mammals, and birds were killed; humans experienced respiratory problems. The extensive and long-lasting *Karenia* bloom was a serious problem to the region, attracting significant media attention and highlighting the need to understand the nutrient sources that allow such a large bloom to persist in near-shore waters for so long. Texas also experienced a *Karenia brevis* bloom; in Corpus Christi approximately 12 million red drum fry were killed, and over 300 000 dead fish washed ashore near South Padre Island.

Brown tide. For the third time since 1985, there were no reports of brown tide this year in New Jersey or Long Island. Texas, however, experienced its first brown tide since 1997 – the causative organism was *Aureombra lagunensis*.

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

Cyanobacteria. Several cyanobacterial blooms occurred in Louisiana, including one in Lake Salvador where humans were affected.

Other Events. A *Cochlodinium* bloom occurred over a 20-mile area in New York state (Peconic Estuary). Possible toxicity for soft clams and oysters was reported, but not confirmed. There was also an extensive *Cocchlodinium polykrikoides* bloom in the Buzzards Bay region of Massachusetts, often in red water concentrations. There were, however, no harmful effects attributed to this bloom.

A *Karlodinium veneficum* bloom occurred in the Corsica River, Maryland (Chesapeake Bay tributary). 30 000–50 000 fish were killed.

12.2 Denmark

Per Anderson

In Denmark we experienced <u>no</u> cases of PSP or DSP in 2005. For the first time ASP was observed in concentrations above regulatory limits in Danish waters. The observations of ASP were done in three mussel harvest areas in the southwestern Kattegat during the period March-April. The maximum concentration observed was 32 mg/kg. The accumulation of ASP-toxicity coincided with a bloom of toxic *Pseudo-nitzschia seriata*. The ASP-toxicity was observed at relatively low concentrations (50 000 cells/l). Since this is below the current Danish regulation limit of 200 000 cells/l of *Pseudo-nitzschia seriata* the regulation guidelines with respect to this species have been changed.

High concentrations and biomasses of *Pseudo-nitzschia* spp. were registered during summer and autumn in the Limfjord area. Many harvest areas were precautionary closed or were opened under intensified monitoring, due to the risk of ASP-toxins in shellfish. No ASP-toxicity in shellfish was observed during the blooms. Exceptional blooms of *Dinophysis acuminata* and *D. acuta* were observed during summer and autumn in the Limfjord and precautionary closures were imposed on the shellfish harvest. However, no DSP-toxicity was observed in shellfish.

There were no reports on fish kills or recreational problems related to HABs in Danish waters for 2005.

12.3 Canada

Jennifer Martin

PSP

Atlantic Coast

The St. Lawrence Estuary experienced a normal year for closures of shellfish with four of the 12 HAE-DAT harvesting areas experiencing closures as a result of unsafe levels of PSP toxins. The highest level of toxins (5 819 µg 100g) was detected on 4 July in *Mytilus edulis*.

The Bay of Fundy experienced a more normal year (following two years of extreme highs) for toxicity. Levels of PSP toxins in *Mya arenaria* exceeded 1500 μ g STX eq. 100 g⁻¹ in southwestern New Brunswick. The Bay of Fundy did experience periods of low unsafe levels of toxicity during the winter months as a result of the highs from the 2004 bloom. The major part of the bloom persisted from June through August. The eastern portion of the Bay of Fundy (western Nova Scotia) had 1088 μ g/100g in blue mussels and 956 μ g/100g soft-shell clams. Southern Nova Scotia (Halifax region) had PSP toxin levels of 477 μ g 100 g in blue mussels.

The Newfoundland and lower Gulf of St. Lawrence regions did not have any closures due to marine toxins during 2005.

West Coast: There were a number of closures of shellfish harvesting areas due to unacceptable levels of PSP. Highest levels (3500 ug 100 g) were detected in sea mussels in the region north west of Vancouver

ASP

Domoic acid was measured at levels of $120 \mu g$ g in scallop digestive glands in the St. Lawrence Estuary.

Fish Kills

East Coast: There were no observed mortalities on the east coast.

West Coast: Salmon mortalities occurred following a bloom of *Heterosigma* at Broughton Archipelago (late July – early August) and Clayoquot Sounds (8 July). A prymnesiophyte and small flagellates (~2 000 000 cells L) caused fish mortalities in Quatsino Sound from 20–24 June. *Chattonella* (110 000 cells L) in Finlays Channel were implicated in fish deaths on 14–15 August. *Chaetoceros concavicorne* was responsible for mortalities in the Sechelt Inlet in late August.

12.4 Norway

Einer Dahl

ASP

There were no recordings of ASP-toxin (domoic acid) above quarantine levels in mussels along the Norwegian coast in 2005, even not after a very dense bloom of *Pseudo-nitzschia* in northern Norway where about 16 million cells/L was recorded.

DSP

As usual DSP-toxins were recorded above quarantine levels at some monitoring stations in southern Norway, while not at others. In total the problems due to DSP-toxins in southern Norway in 2005 turned out to be small, and where present, occurring mainly from October and onwards. In northern Norway, on the other hand, recordings of DSP-toxins in 2005, as in 2004, were extensive. Already from early in August *Dinophysis acuta* was common in the area and caused accumulation of DSP-toxins above quarantine levels in mussels. For the first time *Dinophysis acuta* was observed in bloom proportions in our northernmost county, Finnmark, and was associated with high levels of DSP in mussels (up to 1 600 microgram/kg), also causing a minor poisoning (5 persons) after consumption of mussels.

PSP

Also occurrences of PSP-toxins in mussels are recurrent problems in Norway. In 2005 these problems again were small, and concentrations of toxins in mussels exceeded the quarantine levels only at some few monitoring stations, and only for rather short periods. The problem may, however, occur all along the coast and tend to be more site-specific than the occurrence of DSP-toxins. In mid- and northern Norway *Alexandrium tamarense* had an unexpected, moderate, "winter bloom" in February–March, which led to temporary closure of mussel harvesting from commercial plants.

AZA (Azaspiracides)

For the third time in Norway presence of Azaspiracid in the mussels passed the quarantine levels. It happened at the monitoring station in the Finnmark county, closest to the Russian border, in May and July. At other stations in northern Norway smaller amounts of Azaspiracid were detected in mussels during autumn. More seriously, however, was detection of Azaspiracid in crabs in Norway for the first time (see under new findings).

Ichtyotoxic events

Chrysochromulina leadbeaterii was observed in the Trondheimsfjord (mid Norway) in April-May 2005 and associated with minor problems in fish farms (Atlanic salmon), as reduced appetite and a few dead fish. This is the southernmost observation of effects of Chrysochromulina leadbeaterii in Norway after it caused massive fish kills in northern Norway in May–June 1991.

12.5 Estonia

Andres Jaanus

The cyanobacterial maximum in the southern Gulf of Finland was recorded already in the second half of June, while the potentially toxic species *Nodularia spumigena* reached its measured maximum in the mid of July at the open gulf stations. As an average for the period from 15 June to 31 July, the biomass of N₂-fixing cyanobacteria (*Anabaena* spp.,

Aphanizomenon sp. and Nodularia spumigena) remained at the lowest level recorded since 1997 in the central Gulf of Finland (Figure 12.5.1).

In spite of the increased chlorophyll *a* concentrations and the enhanced abundance of some flagellates (*Pyramimonas* spp., *Chrysochromulina* spp.), no events of mass occurrence of any species were recorded during the year 2005.

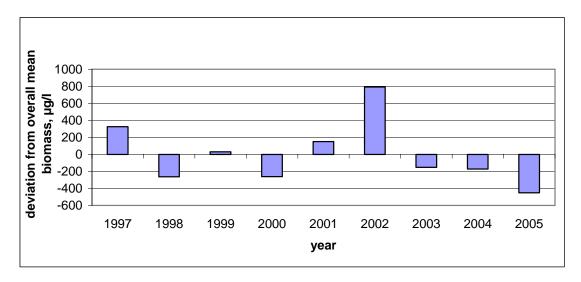


Figure 12.5.1: Relative biomass of N_2 -fixing cyanobacteria (μ g/l) along a cross-section of the central Gulf of Finland, Baltic Sea. The overall mean is calculated as an average of all observations during the period from 15 June to 31 July in 1997–2005.

Poland

Hanna Mazur

In the coastal waters of southern Baltic Sea hepatotoxin producing *Nodularia spumigena* occurred as soon as at the end of May. When weather conditions improved and water temperature reached over 20 °C, the bloom began to develop. In 2005, the most intensive *N. spumigena* bloom was recorded from 5 July to 14 July. Maximum cell number was 23.5 mln cell/L. The highest cell-bound nodularin concentration in seston was 3.9 mg/L and in freezedried phytoplankton sample 1.9 mg/g d.w. In water, the concentration of dissolved toxin during *N. spumigena* bloom ranged from 0.9 μg/L to 95.0 μg/L. Single filaments of the cyanobacterium in phytoplankton samples were present in Polish coastal zone till the mid September, but concentrations of nodularin in water were below the HPLC detection limit.

Generally, the bloom of toxic *N. spumigena* in Polish coastal waters was rather moderate in 2005. However, accumulation of nodularin was observed in blue mussels and flounder through out the whole summer season. Three incidents of cyanobacterial dermatitis were recorded.

On 28 June, there was one incident of fish kill in Puck Bay off Kuźnica (Hel Peninsula). It was accompanied by the presence of increased number of *Anabaena lammermanni* (potentially toxic). Single filaments of *Nodularia, Gleocapsa, Merismopedia, Synechococcus* were also recorded in the phytoplankton sample.

12.6 Netherlands

Marnix Poelman

Fibrocapsa japonica was not found in the Dutch waters in 2005.

Chatonela spp. was found in the North Sea in several samples. The cell densities detected up to 24*10³ cells / liter in June in only one sample. Next to that Chatonella spp. was found in three more samples in low cell numbers (100–300 cells / liter). Data on the period after July have not been processed yet.

Phaeocystis sp. came to high concentrations (1–2 million cells per liter) in February through, which is earlier then usually is observed. The cell concentrations from February on kept climbing up, which resulted in a bloom with cell number above 10 million cells per liter. In May the Phaeocystis sp. bloomed to a peak with cell numbers up to 138 million (!) cells per liter. After May the bloom declined to non-bloom format within 1–2 months time. No harmful events have occurred for as far as observed in relation to this Phaeocystis bloom.

Dinophysis acuminata was present in low cell numbers (below 100 cells/l) during the period of June through October. During the period August through mid October the cell numbers increased, with values ranging from 160 to 1100 cells per liter. A single week peak of 1100 cells per liter was observed in the Wadden Sea at the end of August. In general toxins are not found in shellfish in the summer period (with use of a rat bioassay), however with use of a LC-MS method Okadaic Acid and DTX-3 could be observed in maximum concentrations of respectively $30 \, \mu g/kg$ and $24 \, \mu g/kg$.

The DTX-3 presence in mussels is most likely a result of the co-occurrence of *D. acuta* next to *D. acuta* was observed in cell numbers not higher then 80 cells per liter. However, there have not been any food safety risks.

D. norvegica was only observed once in the Wadden Sea in mid august, the cell number was however very low, around 20 cells per liter.

12.7 Great Britain

Eileen Bresnan

Northern Ireland

In 2005 thirty five sites were sampled routinely on a fortnightly basis from Northern Ireland sea loughs. Alexandrium spp. was observed in low cell densities in 2005 (<100 cells .l⁻¹) and no PSP toxins were detected in shellfish samples.

Five species of Dinophysis (acuminata, acuta, norvegica, rotundata and fortii) were recorded in water samples taken during the year but at low cell densities. The maximum cell density observed was 640 cells l⁻¹ (mixed sample of *D.acuminata* and *D.fortii*) in Belfast Lough during mid July. Only one incident of DSP toxicity was recorded in shellfish during the year but was not linked with the presence of any known harmful microalgal species in water samples.

Pseudo-nitzschia spp. reached a maximum concentration of 69 960 cells 1^{-1} . Toxicity, however, was confined to samples of scallops (*Pecten maximus*). Domoic acid levels reached a maximum of 168.85 μ g/g whole flesh.

An extensive and intense bloom of *Karenia mikimotoi* occurred along the west coast of Southern Ireland in early June and dissipated through July. In Northern Ireland *K.mikimotoi* was recorded in our northern most sea lough, Lough Foyle, at 31 100 cells I^{-1} in mid-July. It was also recorded in the North Channel at 4550 cells I^{-1} .

England and Wales

Alexandrium spp. (PSP) were recorded from 16 of the 48 sampled areas in England and Wales. Highest concentrations were found at Brancaster on the north Norfolk coast (1.8 million cells/litre) in September. Alexandrium spp. were found regularly in samples collected from the Fal estuary from early May to late September with peak concentrations reaching 8800 cells/litre in June. Samples from Weymouth inner harbour also regularly contained Alexandrium spp from late March to mid October with a maximum concentration of 107 000 cells/litre occurring in July. None of these occurrences coincided with PSP's toxins being found in shellfish flesh. PSP toxins were only found on three occasions in 2005. Three samples of mussels taken from the Fowey estuary (Cornwall) during July contained PSP toxins up to 195μg / 100g flesh.

Dinophysis spp. (DSP) were found in ten sampling areas, but often only on one occasion and always at low concentrations. Highest concentrations (2000 cells/litre) were found at the offshore site at Blyth, Northumberland. *Prorocentrum lima* (DSP) were only found on three occasions, once in the Wash during April and twice in the Fleet Lagoon, Weymouth during August and always at low concentrations.

Pseudonitzchia spp. (ASP) were found in 31 sampling areas and appeared to be much more widespread and persistent than in previous years. However, they breached the 'investigative' level (50 000 cells/litre) and the action level (150 000 cells/litre) on only one occasion, in a sample from the Burry Inlet during May (310,000 cells/litre). ASP toxins are usually most frequently found in samples of scallops from offshore fishing grounds, particularly in the Western Channel. However, in 2005, samples of mussels and cockles from Milford Haven and samples of mussels from the Taw / Torridge, contained low levels of ASP toxins with the highest concentration (9.0 μ g/g) being found in a sample of cockles taken from Milford Haven in June.

Scotland

A *Phaeocystis* bloom observed in Shetland Islands and extended down the east coast of Scotland associated which affected farmed fish. Cell densities reached (8 million cells per litre (equated to 6µg chlorophyll).

Maximum number of *Alexandrium* spp. recorded was 1300 cells. Γ^1 along the east coast of the North Sea. *Dinophysis* spp. cell numbers were considerably reduced with cell maxima <1000 cells *per* litre. *Pseudo-nitzschia* showed the normal seasonal pattern of occurrence with blooms of *P. delicatissima* occurring in April while blooms of *P. seriata* type cells occur in the late summer/autumn time. Transmission electron micrscopy of *Pseudo-nitzschia* blooms showed the spring bloom to be dominated by *P. cf. delicatissima* while the summer/autumn bloom was dominated by *P. cf. seriata* and *P. cf. australis*.

The major *K mikimotoi* bloom that affected the republic of Ireland did not extend into U.K. waters.

12.8 Ireland

Amnesic Shellfish Poisoning (ASP)

For the first time in Ireland, a major ASP event was recorded in both samples of *M. edulis* and *C. gigas*, where levels were observed above the regulatory limit. Previously there had only been one recorded incident of ASP levels slightly above the regulatory in a sample of *M. edulis* in 2002 from Co. Donegal.

Typically levels were observed to be < Limit of Detection up until April. During April to mid-May, 35 samples were observed to be above the regulatory limit, predominantly *M. edulis* in

all Bantry sites, and in both M. edulis and C. gigas samples from Kenmare. Highest level observed was 444.9 μ g/g Whole Flesh. During this toxicity period testing was scaled up to analyse all samples from the SouthWest, Domoic Acid conc.'s. By the end of May all areas were re-opened, and testing reduced. Only one sample has been observed to be at the regulatory limit since, in June in M. edulis from Castletownbere (20 μ g/g Whole Flesh). A level of 4 μ g/g Whole Flesh was observed in samples of E. siliqua in early August from Gormanstown.

From August to September, dramatic increases in *Pseudo-nitzschia spp*. had been observed in Bantry and Kenmare, where cell counts were observed to be >1 000 000 cells/litre. Samples of *M.edulis* from these areas were analysed during this period and levels of Domoic acid typically observed were <LOD. From Mid Sept to Oct, there was a dramatic decrease in the numbers and distribution of *Pseudo-nitzschia spp.*, where very low levels were observed.

Diarrhetic Shellfish Poisoning (DSP)

Overall for 2005, (to end of October 05) the total number of all samples testing positive under DSP Mouse Bioassay was 15% (n = 2133) compared to 3.2% over the same time period for 2004, and 3.6% for 2001. A breakdown of percentage positives by species for *M. edulis* reveals 23.9% of samples tested positive (of 1130 samples) compared to the same period for 2003, 5.8% samples tested positive (n = 1122).

No cockle or Razor clam samples submitted and analysed were positive for DSP/AZA Toxicity during the same time period.

Whilst no samples of Oysters tested positive via bioassay, samples of *C. gigas* analysed showed the presence of Azaspiracids above the regulatory limit in samples from Donegal Harbour, Tra Eanach and Killala.

For the first time, DSP toxicity (total conc. present in the form of OA esters) was detected in samples of Clams (*S. solida, T. philipinarium*) from Galway and Sligo in July, highest concentrations observed was $0.27 \,\mu\text{g/g}$ total tissue (post hydrolysis) where corresponding positive bioassays were observed.

Positive DSP bioassays were first observed in samples from Galway at the end of May. Chemical analysis showed levels of Okadaic Acid equivalents present at vary levels above and below the regulatory limit in June, where Positive bioassays were observed in Donegal, Galway and the SouthWest. During July it was observed that further increases in Okadaic Acid conc. were observed in Donegal, decreases in OA conc. in samples submitted from Galway (except one site which showed increases in conc.), increases in OA conc above the regulatory limit in one site in Mayo, and on average remaining at the same conc. levels in the majority of sites in the SouthWest (two sites showed increases) as was observed for the region in June.

The predominant toxin observed in samples from the end of May to the beginning of August was Okadaic Acid, whereas from this time levels of OA decreased, where levels of DTX-2 increased to become the predominant toxin present in samples. The levels of DTX-2 peaked in early August and were observed to decrease throughout the remainder of August through to early November to below regulatory levels in the majority of samples submitted.

Hydrolysis extractions (to determine the presence of Okadaic Esters (DTX-3) were conducted on samples submitted from Sentinel Sites, where there was a discrepancy between bioassay and chemical results. The graph below illustrates the detection of Okadaic Acid esters present in samples from the end of May (predominantly Hydrolysed Okadaic Acid) reaching a peak in early August, and decreasing throughout Sept (predominant toxin observed was hydrolysed DTX-2) to levels below the regulatory limit in the majority of samples in October.

Azaspiracid Shellfish Poisoning Summary (AZP)

In September, levels of AZA's increased in a number of sites in the SouthWest above the regulatory limit (sites in Bantry, Kenmare & Castlemaine). Levels of AZA's increased during Sept in the NorthWest in a number of sites, but were observed to decrease in the West. Further sharp increases in AZA levels were observed in the SouthWest (in many sites, levels were > Upper Limit of Quantification (ULQ =0.9 μ g/g)) in October, whereas in the West & NorthWest no further increases were observed in AZA conc.'s; and remained at approximately the same levels as those observed for August. From July – December, AZA levels have remained consistently high in McSwynes Bay, where levels in a number of sites from the SouthWest were observed to decrease to levels below the regulatory limit in December particularly in all sites in Kenmare, whilst in Bantry Inner, AZA levels have been variable where in Outer Bantry sites AZA levels have been consistently high.

Paralytic Shellfish Poisoning (PSP)

During January–October, 227 samples were submitted for PSP analysis. All samples were negative via Jellet PSP Rapid Test Kit, apart from two *M. edulis* samples observed in early mid-June from Cork Harbour. These positive samples were re-analysed via AOAC PSP bioassay, where the maximum level observed was 66.12 μg STXeq100g⁻¹.

Levels of *Alexandrium sp.* were generally observed in low levels all around the Irish Coast throughout May–October (when compared to 2004), with the highest levels observed in North Channel, Kinsale, Oysterhaven, Loughras Beg and Greenore. The levels and distribution of *Alexandrium sp.* were observed to further decrease in September, and were observed to be mainly confined to the South & SouthWest at low levels in October. Samples of shellfish from these areas (except those from North Channel in June) tested negative for the presence of PSP toxins.

Other HABs recorded in Ireland in 2005 included *Noctiluca* and *Phaeocystis* in low numbers, and the Exceptional bloom of *Karenia* already reported in this report

12.9 Spain

Andalucia

ASP

Domic acid above regulatory levels were reported in the Andalusian west coast during April 2005 in different clam species (*Chamelea gallina*, *Donax trunculus*), associated to *Pseudo-nitzschia* populations. Maximum levels of 279720 cell/L and 48 µg DA · g⁻¹ meat. Similar events were observed in the Alboran Sea region (East Andalusian coast) in the same month. *Pseudo-nitzschia* populations there reached concentrations of 615,353 cell/L. Maximum levels of domoic acid were found in scallops (*Pecten maximus*)

Lipophilic Shellfish Toxins Toxins (OA, DTX, PTX, YTX)

Proliferations of *Dinophysis acuminata* (max. levels of 4200 cell/L) led to harvesting closures (DSP) in May-June in the coast of Huelva.

Basque Country

Data are reported for the first time on coastal waters and estuaries of the Basque Country (Bay of Biscay). Commercial exploitation of shellfish is not of importance in this area. However, harvesting of wild shellfish is a usual practice in the estuaries of Butroi, Oka and Bidasoa rivers during summer and autumn. Therefore, analysis of DSP, ASP and PSP toxins were performed in those estuaries (twice a year during the harvesting period). In addition, in order

to comply with the European Water Framework Directive, phytoplankton species composition and abundance were measured in 19 coastal stations and 12 estuaries (twice a year, in spring and summer).

In 2005, DSP, ASP and PSP toxins were not detected in shellfish harvesting areas. On the other hand, potentially toxic species were found in some coastal stations and estuaries. *Dinophysis acuminata* was detected in the Barbadun estuary (120 cells/L). *Pseudo-nitzschia* spp. was observed in 40% of the samples. The maximum concentration was found in coastal waters (6·10⁵ cells/L). In the outer Nervion estuary this diatom reached up to 3·10⁵ cells/L. *Prorocentrum minimum* was detected only in 7% of the samples. The maximum concentration was found in the Bidasoa estuary (8496 cells/L). *Karenia* sp. was detected in one coastal station (16 992 cells/L). Species harmless for human health but potentially icthyotoxic were also observed. *Chrysochromulina* spp. was found in 70% of the samples. The maximum concentration (3·10⁵ cells/L) was found in coastal waters. In the Oka estuary this haptophyte reached concentrations up to 1·10⁵ cells/L. *Heterosigma akashiwo* was detected in 2% of the samples, and its maximum was found in the Nervion estuary (21 240 cells/L).

Catalonia

ASP

During 2005 domoic acid was no detected on *Mytilus galloprovincialis* samples in the delta del Ebro region.

Lipophilic Shellfish Toxins (OA, DTX, PTX, YTX)

Four DSP events occurred in Alfacs Bay during 2005. Positive results in mousse bioassay were found in mussels (*Mytilus galloprovincialis*) in January, February, July, August and December, and in clams (*Tapes decussatus*) only in January. During these events *Protoceratium reticulatum*, *Dinophysis sacculus* and *Dinophysis caudata* were present at maximal cell densities of 1,600 cell/L, 120 cell/L and 400 cell/L respectively. DSP events occurred also in open waters in June, July and August affecting mussels and clams (*Donax trunculus*).

PSP

Cell densities of *Alexandrium minutum* exceeded alarm level in January, May and June in Alfacs Bay but PSP toxin concentrations (mousse bioassay) were below regulatory levels.

Fish-killers

Karlodinium spp. was present under alarm levels and reached maximum cell densities of 61 000 cells/L in June. Fish kills were not detected.

Galicia

ASP

A bloom of *Pseudo-nitzschia* spp. led to accumulation of domoic acid in shellfish, causing harevsting closures in May affecting the four Rías Baixas. From 27 April to 17 October *Pseudo-nitzschia* spp. were detected, mainly in the Rías Baixas, in concentrations around 100,000 cell/l. Maximum *Pseudo-nitzschia* concentration during the period of domoic acid detection was $5.8 \cdot 10^5$ cell·l⁻¹ and maximum level of domoic acid accumulated in mussels was $365 \mu g \cdot g^{-1}$ meat.

A precise taxonomical identification of the species was only made on some samples during the period of domoic acid detection in bivalve molluscs. The main species detected were *Pseudo*-

nitzschia australis and *P. fraudulenta*. Therefore, it is not possible to specify the time when the toxic cells disappeared.

The recurrence of *Pseudo-nitzschia* spp. blooms and the extremely slow detoxification process of domoic acid in scallops (*Pecten maximus*) caused the persistence of harvesting closures for this species in practically all the Galician coast. The harvesting of scallops was only possible in a small area of Ría de Arousa, during short periods of time, and only after evisceration according to Commision decision 2002/226/EC.

Maximum domoic acid levels detected in scallop were 663.9 μg·g⁻¹.

Lipophilic Toxins (OA, DTX, PTX)

During 2005 there were long lasting-closures due to the presence of lipophilic toxins in shellfish. Most of the mussel culture areas in the outer reaches of the Rías Baixas were closed in the beginning of the year, until mid-January, due to a bloom of *D. acuta* that persisted until mid December 2004. Between April and September, two blooms of *Dinophysis acuminata* caused bans that in some areas, like Ría de Pontevedra, were continuous due to the lack of time for depuration between succesive blooms (maximum *D. acuminata* cell concentration, $5.1\cdot10^3$ cell· Γ^1).

In the Ría of Ares-Betanzos (Northern Rías), *D. acuminata* caused closures between September and December (both months included; maximum *D. acuminata* cell concentration $3.2 \cdot 10^3$ cell·l⁻¹).

Finally, a bloom of *D. acuta*, that started in early October and remained until mid- December, affected most of the Galcician coast, with closures that persisted until early 2006 (maximum *D. acuta* cell concentration $1.8 \cdot 10^4$ cell·l⁻¹).

PSP

A very intense *Gymnodinium catenatum* bloom (after 10 years of absence!) occurred from October to December affecting all the production areas in the Rías Baixas and some areas in the Northen Galician coast. Maximum *G. catenatum* concentration was $1.7 \cdot 10^5$ cell·l⁻¹ and maximum level of accumulated toxins $4,080 \mu g$ STXeq· $100 g^{-1}$ meat.

Very localized bloom of *Alexandrium minutum* affected the Baiona embayment (Ria de Vigo) and the Rías Altas during short periods (a week) between May and June. Maximum concentration of *Alexandrium* cells was $411,345 \text{ cells} \cdot \text{L}^{-1}$ and maximum level of accumulated toxins $189 \, \mu g \, \text{STXeq} \cdot 100 \, \text{g}^{-1}$ meat.

Successive development and disappearance of *Dinophysis* populations during such a long period caused that in some areas, like in Ría de Pontevedra, shellfish harvesting was prohibited during more than nine months during 2005. It seems that blooms of *D. acuminata* were due to *in situ* growth whereas the population of *D. acuta* was initially advected.

13 Term of Reference j)

13.1 Contributions to the ecosystem overview

Discussions were held on potential contributions to the ecosystem overview of the advisory reports in particular to describe the quantity and quality of marine habitat and/or the health of the marine ecosystem, and to consider and report on potential indicators of significant change in these ecosystem attributes.

It was concluded that that the role of phytoplankton with regard to the ecosystem approach, is a far wider issue than could be addressed by WGHABD, whose main role is to investigate the dynamics of functional sub-group of phytoplankton. Therefore, the relevance of this ToR to our working group was questioned. In addition it was discussed that the quantity and quality of marine habitat and/or the health of the marine ecosystem would not be reflected by just using this functional group of phytoplankton, but would be better addressed by looking at biomass and composition of the complete phytoplankton community. It was suggested that the Working Group on Phytoplankton Ecology would be in a better position to tackle this ToR. If the Oceanographic Committee wishes any specific advice on this matter the group would be happy to discuss this inter-sessionally.

14 Term of Reference k)

14.1 Review and update sub-regional data tables for REGNS

Review and update sub-regional data tables and where necessary include new data (parameters) and/or existing data (parameters) updated where relevant. The data tables will be subject to thematic assessment to be undertaken at a REGNS thematic assessment workshop.

This was addressed under ToR e) (above).

15 Draft Resolutions

WGHABD will meet in Latvia in 2007. Please see the proposed Terms of Reference for the next meeting at Annex 3.

16 Recommendations

WGHABD recommends that officers of Iceland and the Faroe Islands identify national focal points/ individuals responsible for data submission to HAE-DAT, decadal maps and national reports. It should be emphasised that these reports should be submitted even if the delegates is unable to attend the meeting. It is recommended that these focal points are identified by December 2006.

It was proposed that delegates attempt to establish a group of interested parties to attempt to attract funding to develop the GIS capabilities of HAEDAT

The WG requests that the ICES Data management group be contacted to evaluate the potential for statistical analyses from the HAEDAT database.

Annex 1: List of participants

* = present only during the joint day with the WGGIB.

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Annex 2: Agenda

Monday, 3 April 2006

- 09:00 **Welcome**; Housekeeping issues; Introduction of participants; Review by the Oceanographic Committee / Assessment of Expert Working Groups; Adoption of agenda/ Terms of Reference
- O9:30 ToR a: Review progress in the detection of harmful algal blooms and their dynamics by remote sensing techniques and examining results from new sensors and algorithms as well as validation procedures used for HAB observations.
- 11:00 Morning break
- 11:30 ToR b: Review the section on Phytoplankton Monitoring in the Report of the Joint FAO/IOC/WHO Ad Hoc Expert Consultation for *Codex Alimentarius* on Biotoxins in Bivalve Mollusks (Oslo 26 September 2004).
- 12:30 Lunch
- 13:30 ToR d: Review progress towards the joint theme session between WGHABD and WGPBI for the ICES ASC in 2006 titled "Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge".
- 14:30 ToR e. Review progress and analyses that REGNS North Sea Group have done on data-sets submitted by members of WGHABD (to meet in the interim).
- 15:00 ToR k. review and update sub-regional data tables and where necessary include new data (parameters) and/or existing data (parameters) updated where relevant. The data tables will be subject to thematic assessment to be undertaken at a REGNS thematic assessment workshop.
- 15:30 **Afternoon Break**
- 16:00 ToR j discuss and report on potential contributions to the ecosystem overview of the advisory reports describing the quantity and quality of marine habitat and/or the health of the marine ecosystem, and to consider and report on potential indicators of significant change in these ecosystem attributes.
- 17:30 Adjourn for the day

Tuesday, 4 April

Location: Marine Station on Hel Peninsula

- 09:30 ToR c: Review the outcome of the WKNCT Workshop on New and Classical Techniques in Enumeration of Phytoplankton.
- 10:30 Morning Break
- 11:00 ToR f. Discuss new findings that pertain to harmful algal bloom dynamics. Bring new findings in phytoplankton population dynamics models, with emphasis on loss processes, to the attention of WGHABD for discussion.
- 12:30 Lunch
- 13:30 ToR f: New Findings Continued
- 15:00 Afternoon Break
- 15:30 TOR g. Review the on-line format of HAEDAT submission form and evaluate the amendments made to update historical submissions and links to mapping.
- 16:30 TOR h. Review the structure and composition of the decadal HAE maps for the ICES region with special reference to clarifying the distinction between harmful algal blooms and the harmful affects that are reported on the maps. In particular, the registration of cyanobacterial blooms in brackish and marine waters should be revisited from the emerging perspective of their known toxicity and implicit harmful effects.
- 17:30 Adjourn for the day

Wednesday, 5 April 2006

09:00 Joint presentations with ICES-IOC-SCOR Working Group on GEOHAB Implementation in the Baltic [WGGIB]

Report and discuss new findings on HABs and HAB modelling in the Baltic

Estimate the health hazard of cyanobacteria and dinoflagellate toxins to humans and review the concentrations of HAB toxins in the upper trophic levels of the Baltic foodweb

New nodularin analogues in the Baltic Nodularia spumigena and other environmental samples

- 11:00 Morning Break
- 11:30 Joint presentations continued:

Latest results from the Cyanobacteria ecosystem effects cruise in the Gulf of Finland 2005

Ecosystem effects and health hazards of cyanobacteria toxins in the Baltic – a review

- 12:30 Lunch
- 13:30 Report Writing / Tours of marine station
- 15:00 Terms of reference 2007
- 17:00 Meeting location for 2007
- 17:30 Adjourn For The Day
- 18:00 Dinner in local restaurant hosted by University of Gdansk.

Thursday, 6 April

- 09:30 ToR i. Collate and assess National reports and update the decadal mapping of harmful algal events for the IOC/ICES harmful algal database, HAE-DAT
- 11:00 Morning Break
- 11:30 Continue National Reports / Report Writing
- 12:30 Meeting adjournment

LUNCH

ICES-IOC SCOR Working Group on GEOHAB Implementation in the Baltic (WGGIB) continued: Thursday 6 April—7 April Friday

Annex 3: WGHABD proposed Terms of Reference 2006

The **ICES IOC Working Group on Harmful Algal Bloom Dynamics** [WGHABD] (Chair J. Silke, Ireland) will meet in Latvia in 2007 to:

- a) review outcome of the WKEUT workshop on Long term data sets and Eutrophication scheduled for the period 11-15 September 2006 in Copenhagen
- b) review outcome of the joint theme session between WGHABD and WGPBI at the ICES ASC in 2006 titled "Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge".
- c) review progress and analyses that REGNS North Sea Group have done and report on the second REGNS workshop to be held in Copenhagen from 15th to 19th May 2006. In particular the relevance of HABs in defining relationships between trends measured at the broad scale (the overview assessment) with those measured on a more localised scale (the thematic assessments).
- d) discuss new findings that pertain to harmful algal bloom dynamics. Bring new findings in phytoplankton population dynamics models to the attention of WGHABD for discussion.
- e) review the on-line format of HAEDAT system and developments made towards developing an integrated system and evaluate the amendments made to update historical submissions and links to mapping.
- f) review the structure and composition of the decadal HAE maps for the ICES region with special reference to clarifying the distinction between harmful algal blooms and the harmful affects that are reported on the maps.
- g) 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).

WGHABD will report by (date to be decided) for the attention of 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 AND RELATION TO ACTION PLAN:	Action Plan No: 1.1, 1.2, 1.5, 1.7, 1.10, 1.11, 1.12, 2.3, 2.9, 3.2, 4.11, 5.10, 5.13, 5.16, 6.1, 6.2, 6.3, 6.4, 8.1, 8.2, 8.4. Term of Reference a)	
	This workshop, a complex activity requiring long term data sets will provide valuable results on investigations into long term change and HAB dynamics. The WGHABD is most interested in the outcome of workshop It is appropriate that the WG evaluates the report from the workshop and promotes its dissemination. Term of Reference b)	
	Current knowledge on modelling HABs and HAB physical-biological processes is limited. Improved knowledge on the validation of these models and the status of coupled physical-biological process knowledge is essential to improve models for HAB dynamics. This joint theme session between WGHABD and WGPBI at the ICES ASC in 2006 titled "Harmful Algae Bloom Dynamics; Validation of model predictions (possibilities and limitations) and status on coupled physical-biological process knowledge" is of fundamental benefit to HAB science.	

SHARE:

Term of Reference c) The provision of integrated advice is a challenge; it is a process, which must be supported by methods, and tools that allow diverse sources of data and information on numerous pressure and state changes to be objectively and scientifically assessed. The REGNS study group has requested that the WGHABD prepare to provide data, information and indicators. A delegate from the WGHABD will attend the REGNS meeting in May 2006 and will report to the group in 2007 on the progress of the assembly and analysis of the data. Term of Reference d) 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. Term of Reference e) HAEDAT is an extremely valuable dataset that is only now becoming extensively utilised. There are developments on the technical end that allow users to mount their data and query it through the Internet. This system was demonstrated to WGHABD In 2006 In an almost complete version. It Is requested that the finished version be presented In 2007, and potential uses be identified. Term of Reference f) 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. HAEDAT has been improved in recent years and it would be desired that the maps be made more user friendly and adaptable. At the 2006 meeting it was requested that a joint project between institutes with active GIS departments be investigated to attract funding to allow the development of this functionality of the HAEDAT and HAE-MAPS be established. Term of Reference g) The work of collating the national HAE reports and building up HAE-DAT 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. PICES, South America, HANA and Caribbean countries (via IOC/FANSA and IOC/ANCA) are now included 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. RESOURCE The research programmes which provide the main input to this group are already REQUIREMENTS: underway, and resources already committed. The additional resource required to undertake additional activities in the framework of this group is negligible. PARTICIPANTS: The Group is normally attended by some 20-25 members and guests SECRETARIAT None FACILITIES: FINANCIAL: No financial implications LINKAGES TO There are no obvious direct linkages with the advisory committees ADVISORY COMMITTEES: LINKAGES TO WGHABD interacts with WGZE, WGPE, SGGIB, SGBOSV, WGPBI. OTHER COMMITTEES OR GROUPS: LINKAGES TO The work of this group is undertaken in close collaboration with the IOC HAB OTHER Programme. IOC should be consulted regarding ToR or discontinuation of the WG prior ORGANIZATIONS: to the ASC. There is a linkage to SCOR through the interactions of the IOC-SCOR GEOHAB Programme. SECRETARIAT **ICES** MARGINAL COST