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

10–13 March 2008

Galway, Ireland

ICES
International Council for the Exploration of the Sea

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Executive Summary

Highlights

- Reviewed HABs and Aerosolized toxins and methodologies used to monitor them
- Decided on and arranged a joint session with WGPBI in 2009
- Presented an example of modelling activities in Scotland (*K. mikimotoi* bloom 2006)
- Discussed developments in *Verrucophera* and *Chatonella* blooms
- Collated and Assessed 12 National Reports
- Discussed the development of HAEDAT and Decadal Maps
- Reviewed the IOC Harmful Algae website to make it more dynamic and links to WGHABD
- Discussed the utility of Mesocosm studies and recent activities in this area.

New Findings

There were 14 presentations that described new findings made by the group and they are synopsized in this report. These reports are extremely useful means of disseminating ongoing projects and very recent developments. They often lead to productive collaborative arrangements between scientists meeting at the working group, and this was the case at this years working group.

The reports included:

- An interesting comparative study of spirolides and PSP toxins in strains of *Alexandrium ostenfeldii* from the North Sea and the Baltic Sea
- A bloom of *Peridinium quinquecorne* and co-occurrence of illness in bathers in Sweden, which is the first implications of this species being harmful.
- Identification of toxic and benign species of *Alexandrium* in Scottish waters
- A synopsis of the recent GEOHAB activities
- A report that described the dynamics of a high density thin layer of *Dinophysis acuta* off the coast of Ireland.
- Developments of gene probes for *Pseudo-nitzschia* and *Dinophysis* species in the PHYTOTEST project.
- A description of unusual lipophilic marine toxins profiles obtained using passive sampling (Spatt) in Irish waters
- New Findings on *Dinophysis* in Spanish waters and the use of Spatt samplers in their detection
- A study of these Spatt methods and evaluations and method development of solid phase adsorbents for phycotoxins in the marine environment
- Rapid Enzyme Linked Immunosorbent Assay for the detection of Domoic Acid
- The news of a culture of phytoplankton collected in 2007 from the North Sea that is producing Azaspiracids in culture. This is the first report of in vitro AZA production by phytoplankton.
- Update on operational forecasting of Alexandrium blooms in the Gulf of Maine, USA

- Culturing and toxicity of North American *Dinophysis*. A first report of toxicity of *in vitro* culture of *Dinophysis*
- *Alexandrium fundyense* bloom dynamics in relation to total phytoplankton community and environmental variables in the Bay of Fundy

1 Welcome and opening of the Meeting

The **ICES-IOC Working Group on Harmful Algal Bloom Dynamics** (WGHABD) meeting for 2008 was hosted by the Marine Institute in Galway Ireland from 10 to 13 March 2008. On behalf of the Marine Institute the working group Chair Joe Silke opened the meeting and welcomed the participants to Galway. The agenda was agreed and Dr Eileen Bresnan and Dr Pat Tester were elected as joint *rapporteurs*. Individual sessions also had session leaders and working *rapporteurs* appointed to share the workload.

29 Scientists representing 14 countries participated in the meeting. The list of participants is presented in Annex 1. The meeting agenda is presented in Annex 2. The meeting was very successful and with a full agenda of challenging and diverse terms of reference. Over the course of the 4 day meeting the group made presentations on 32 topics and this report presents a summary of, these and subsequent discussions. Along with ICES, the IOC are joint organizers of WGHABD, and provide valuable interaction regarding data collection and management of HAB data through the development of the HAEDAT database and its linkages to HAB-MAP. As coordinators of the Intergovernmental Panel on HABs, the participation of IOC in WGHABD forms an important linkage between the working group and this panel. The IOC in also take responsibility to promote the working group among IOC Member Countries outside the ICES area to attend WGHABD and some years is in apposition to offer travel support. In 2008, there were no attendees from outside the ICES area apart from Dr Adriana Zingone from Italy who was hosted by the Marine Institute to attend and present a review of aerosolized toxins and methodologies to monitor them.

WGHABD facilitates interaction between scientists working in diverse areas of HAB science and monitoring and provides a useful forum for interchange of useful terms of reference on various approaches to HAB research. The present working group was established in 1994 following a study group on the Dynamics of Algal Blooms, established two years earlier; however its origins go back further into the 1980s and evolved from other study groups within ICES. The group is an important forum for ICES and IOC to review and discuss HAB events and to provide advice and updates on the state of HABs in the region on an annual basis.

In the opening session the Chair, Joe Silke (Ireland) gave a summary of the presentation of the WGHABD 2007 report to the parent Oceanographic Committee (OCC) at the ASC meeting in Helsinki. The report was very well received and feedback indicated the report was well organized, informative and the meeting was well attended. The 2007 summary report of the Oceanography Committee (OCC) picked up on some topics from the WGHABD '07 meeting. These included items from the WKEUT review, which reported long-term changes in phytoplankton linkages to anthropogenic nutrient enrichment, and that WKEUT found some changes in phytoplankton community structure linked to the North Atlantic Oscillation Index. OCC also listed in its report the signs of declining phytoplankton biomass reported along the Norwegian coast and the spatially and temporally extensive bloom of *Karenia mikimotoi*, which was reported from Scotland in 2006. In light of the valuable information contained in the decadal maps, the OCC recommended that these should be made available on the ICES website together with the original data source if possible. WGHABD discussed this recommendation and the archival of the data is already underway through the developing IOC HAEDAT database that can be linked from the ICES website without any difficulty. Further updates on the developments at HAEDAT and HAB-Maps are discussed in this report.

A proposal to have a joint session with the WGPBI at their 2009 meeting was discussed in the opening session and in one of the Terms of Reference. This session was proposed by the group last year with an aim to mutually inform the two working groups of the priorities in the study the environmental interactions regarding HABs dynamics and to support modelling aspect of HAB research. As one of the strengths of the WGHABD is the interaction between monitoring programme managers, research scientists and data analysts this joint session was deemed appropriate to pursue. There are practical aspects of physical-biological interactions that can be developed jointly, for example, environmental data are often needed in modelling HAB events and sampling could be aligned with local hydrography such as mixed layer depth, circulation patterns, frontal dynamics, etc. Historical data and time-series data are also important in looking for historical occurrences and trends of HABs. Increase and decrease in population size is important to bloom dynamics and modelling HABs. The proposed joint working group was seen as a productive means to explore these areas.

WGHABD recognized that phytoplankton ecology models are usually based on biomass, nutrient, and carbon cycling and in many cases cannot define, explain or predict HAB dynamics. Past joint meetings with modellers have tried to incorporate physics and HAB dynamics into the models and liaison with WGPBI and other working groups will be furthered in coming years.

The group discussed developments in the generation of the new PEP WG during the year. The group remained firm that as the work of the WGHABD remains focused on the dynamics of harmful algae and toxin production and there was still a requirement for a group concerning phytoplankton ecological issues. The group also felt that the success of WGHABD and its continued development was attributed to the applied focus of the group. Many governments have a direct interest in harmful algae as a result of the implementation of the EU shellfish directive and development of national aquaculture industries. The focus of the ASC theme session in 2008 was discussed and delegates were asked to consider the opportunity to present at this session. A group presentation was proposed, however, there were no obvious topics from WGHABD put forward appropriate to this session. While the importance of the microbial loop in marine ecology was acknowledged, it was felt that this topic was very academic with a specialist group of participants and audience for the outputs and advice. There still remains a 'gap' in the provision of advice to ICES in the field of general phytoplankton ecology. In Europe, current drivers such as the Water Framework Directive and the forthcoming European marine strategy means that there is a requirement for information about the ecological role of phytoplankton in the marine ecosystem particularly in relation to water quality.

One activity that occurred during the Working group meeting was a request for advice originating in the occurrence of a *Dinophysis* bloom and presence of okadaic acid in Texas waters which caused recall of shellfish in the area An inquiry came to the WGHABD on Monday afternoon 10/3/08 to determine if finfish were safe to eat during this *Dinophysis* bloom. The strong consensus of the group was "yes fish are safe". Okadaic acid is only known to cause problems in Shellfish and as it is highly lipophyllic would in any case sequester in the liver, ovaries. Recommendation went back to Texas that fish are OK from the assembled group of experts.

The WG felt that the existing ToRs were related and important to dynamics and the Terms of Reference for 2008 were reviewed and adopted.

2 Terms of Reference

At the 94th Statutory Meeting 2007, Helsinki, Finland the Council approved the WGHABD 2008 Terms of References as follows:

The **ICES-IOC Working Group on Harmful Algal Bloom Dynamics [WGHABD]** (Chair J. Silke Ireland) will meet in Galway 10–13 March 2008 to:

a) Review and discuss HAB events related to aerosolized toxins and the methodologies used to monitor them

b) WGHABD to generate key questions for HAB models intersessionally, submit to the PBI group for their consideration with the potential for a ToR for the 2008 PBI meeting and a view to holding a joint meeting of both groups in 2009.

c) To discuss the requirements for and, if agreed, plan the preparation of a draft outline an ICES Cooperative research report on new findings and developments relating to the distribution of phycotoxins and metabolites and recent findings on the distribution of HABs and toxin producing phytoplankton species in the ICES area.

d) Review developments in knowledge pertaining to the development of *Verrucophora* and *Chattonella* blooms

e) 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.

f) fCollate and assess National reports (Country Reps)

g) Review the progress on intersessional updating and inputting data in the IOC-ICES-PICES HAEDAT database and developments made towards developing an integrated system.

h) Review the structure and composition of the decadal HAE maps for the ICES region with special reference to the linkage between the decadal maps and the HAEDAT database and the need for new maps for specific algal species.

i) Review intersessional work to generate a website to electronically archive past reports of the WGHABD and to facilitate intersessional work carried out by the group.

j) To investigate and discuss the possibility of comparative studies that will use mesocosms to explore the dynamics of HABs subject to eutrophic pressures.

3 Term of Reference a)

3.1 Review and discuss HAB events related to aerosolized toxins and the methodologies used to monitor them:

During recent years, there have been increasing reports of respiratory irritations affecting sunbathers in coastal waters of Europe. The victims, sometimes in large numbers (up to 100), have required medical assistance. These events have been associated with toxic aerosols derived from epibenthic microalgae of the genus *Karenia* and *Ostreopsis* that eventually colonize the water column. Current gaps in scientific knowledge are multiple, and concern: a) The hydrodynamic conditions that lead to the detachment of seaweeds and other substrates where the microalgae are attached leading to their resuspension in the water column; b) The seasonality of these microalgae and the environmental conditions promoting their numerical increase; c) The appropriate protocols to monitor epibenthic microalgae populations, whether attached or loose in the water column, and the toxic aerosols derived from them; d) The complex mechanisms underlying the passage of toxins from the whole cells to the irritator aerosols. It was identified that the group could compile the available information of these events, derived actions, and gain from the years-experience on respiratory irritation syndromes related with *Karenia brevis* events from the Florida coasts. In addressing this area of HAB Dr Adriana Zingone from the Stazione Zoologica Anton Dohrn in Naples Italy was asked to present information to the working group on July 2005 Genoa echoers report of fever, red eyes, wheeze after swimming in the sea where *Ostreopsis* was identified

3.1.1 Blooms of *Ostreopsis* species in the Mediterranean area

Reported by Adriana Zingone

Blooms of benthic toxic microalgae have been reported from the Italian coasts since 1998. Their impact has become a major concern starting from 2005, when about 200 people who had spent the day on a beach in Genoa were hospitalized with symptoms of fever, conjunctivitis, and respiratory problems. In Apulia in 2004, several people reported cases of dermatitis and temperature alteration after swimming in coastal waters during blooms. Blooms of the species have been reported in many other coastal Italian and Mediterranean areas and normally during summer. Two different species are present in the area, *Ostreopsis* cf. *ovata* and *O.* cf. *siamensis* (Penna *et al.*, 2005), their taxonomic position still warranting clarification. The former species is the one involved in the harmful outbreaks. Strains genetically similar to the Mediterranean ones are only known from the Brazilian coasts, which are the only other site in the world where *Ostreopsis ovata* has been associated with harmful events. *Ostreopsis* species grow mainly as an epiphyte of macroalgae but can colonize hydroids and other benthic organisms as well as the inorganic substrate. They form at times thick mucus beds that cover the substrate and eventually detach and disperse in the water column. The toxins belong to the group of palytoxins. The most abundant toxin in *O.* cf. *ovata* has been recently described as Ovatoxin-a (Ciminiello *et al.*, 2008). In the phases of bloom collapse, it is probably released into the water and delivered onto the coast through the aerosol. Italian researchers have established a network to allow for quick spread of information, technique intercalibration, and collaboration (http://www.bentoxnet.it/index_en.htm). Many research questions are open on these kind of HABs in the Mediterranean area, including the taxonomic identity and the life cycle of the species, the mechanisms of impact on benthic fauna and on the trophic web, the structure and toxicological properties of ovatoxin-a, the role of bacteria in the production of mucus beds and toxins, the interannual variability of the outbreaks in relation with meteorology and hydrography and the possible increase of these phenomena under changing environmental conditions.

References

Ciminiello, P., Dell'Aversano, C., Fattorusso, E., Forino, M., Tartaglione, L., Grillo, C., Melchiorre, N. 2007. Putative palytoxin and its new analogue, Ovatoxin-a, in *Ostreopsis ovata* collected along the Ligurian coasts during the 2006 toxic outbreak. J. Am. Soc. Mass Spectrom., 19: 111–120

Penna, A., M. Vila, S. Fraga, M. G. Giacobbe, F. Andreoni, P. Riobó and C. Vernesi, 2005. Characterization of Ostreopsis and Coolia (Dinophyceae) isolates in the Western

Mediterranean Sea based on morphology, toxicity and internal transcribed spacer 5.8S rDNA sequences. J. Phycol., 41, 212–225.

4 Term of Reference b)

4.1 WGHABD to generate key questions for HAB models intersessionally, submit to the PBI group for their consideration with the potential for a ToR for the 2008 PBI meeting and a view to holding a joint meeting of both groups in 2009

Current knowledge of modelling HABs and HAB physical-biological processes is limited. Improved knowledge of the validation of these models and the status of coupled physical-biological process knowledge is essential to improve models for HAB dynamics. WGHABD wish to pursue this by interdisciplinary work with WGPBI and development of joint ToRs and potential joint WG sessions in the coming years. This was discussed and the logistics of hosting this session was kindly facilitated by the Spanish delegates who offered to host the meeting in Huelva in Andalucia Spain in 2009. Contact was established with the Chairs of the WGPBI group and the session was confirmed.

The 2009 meetings of both groups will be held simultaneously and both groups will join for one combined session day. A number of models that could form the basis for discussions were proposed:

Presentation of selected Harmful Algal Bloom (HAB) models for short term forecasts

- *Alexandrium* model for the Gulf of Maine
- *Karenia* model for the Bay of Biscay
- Cyanobacteria (*Nodularia-Aphanizomenon)* model for the Baltic
- *Verrucophora* model for the Skagerrak-Kattegat-North Sea area
- Florida red tides model for the Gulf of Mexico

Other topics for discussion included:

- Mathematical tools and techniques in HAB-modelling
- GLOBEC developed tools and their application to HAB Modelling.
- Further presentations as identified by the WGPBI ---
- Key characteristics for HAB-organisms for modelling – What are the HABs characteristics that can be described by equations (mortality rates, seed beds, germination rates, upwelling, variable growth rates, windforcing, thin layers)
- Addressing problems specific to particular areas
- Model resolution

General Themes for Discussion

- how do we incorporate biological processes into physical models
- Grazer control of HABs…. Key Characteristics
- Bio- optical modelling
- Information about the GEOHAB modelling workshop - Justification

4.2 Individual based modelling of the dinoflagellate *Karenia mikimotoi* in Scottish coastal waters

P. A. Gillibrand and K. Davidson

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During summer of 2006, a large bloom of the marine dinoflagellate *Karenia mikimotoi* appeared in the continental shelf waters to the west of Scotland. The bloom was detected both by harmful algae coastal monitoring programmes and in satellite imagery, both sources of data indicating that the bloom progressed northward along the western Scottish coast, past the Orkney and Shetland Isles and into the North Sea. In Orkney, measured concentrations exceeded 3 million cells per litre in August; elsewhere concentrations were lower but reached 1 million cells per litre around the islands of Skye and Lewis.

In order to investigate the factors that may govern bloom transport the progression and development of the bloom was simulated using a lagrangian plankton transport model coupled to a three-dimensional baroclinic hydrodynamic coastal ocean model. The numerical approach consisted of a coupled hydrodynamic-lagrangian random walk model, whereby the algal cells are represented by numerical 'particles' which are subjected to coastal physical processes of advection and diffusion, and which also are ascribed biological characteristics such as growth, mortality and behaviour. By releasing and then tracking the movement and properties of many tens of thousands of particles, the development and transport of the bloom was simulated. Individual particles are transported by three-dimensional flowfields derived from a hydrodynamic model of the region.

For shelf-wide study, we used archived daily-mean flow and temperature fields from operational simulations of the Medium Resolution Continental Shelf (MRCS) hydrodynamic model performed by the UK Meteorological Office. The model has a horizontal spatial resolution of *ca* 6 km and is forced by modelled wind stresses from the UKMO Unified Model. Archived velocity fields, which are available from water surface to seabed at 5 m depth intervals throughout the model domain, are daily-mean values and do not include tidal currents. Daily three-dimensional flow and temperature fields for the UK continental shelf were obtained for the period June – December 2006.

The velocity fields were linked to a particle tracking model, which uses virtual "particles" to represent fixed numbers of *K. mikimotoi* cells. The particles are then advected by the velocity field and mixed by horizontal and vertical eddy diffusion to simulate transport of the cells.

Algal growth and mortality were simulated by the stochastic generation and removal of particles respectively, with algal growth treated as a function of temperature and mortality is a function of local cell density and vertical velocity shear (Gentien *et al.*, 2007).

The model study demonstrated the feasibility of simulating the transport and growth of *K. mikimotoi* blooms using a coupled model platform, but also demonstrated that cells cannot be treated simply as passive particles and that the model must include more sophisticated representation of cell biology, including equations for growth, mortality and phototaxis.

Reference

Gentien, P., Lunven, M., Lazure, P., Youenou, A., Crassous, M. P. 2007. Motility and autotoxcity in Karenia mikimotoi (Dinophyceae*). Phil. Trans. Roy. Soc. B,* doi:10.1098/rstb.2007.2079.

5 Term of Reference c)

5.1 To discuss the requirements for and, if agreed, plan the preparation of a draft outline an ICES Cooperative research report on new findings and developments relating to the distribution of phycotoxins and metabolites and recent findings on the distribution of HABs and toxin producing phytoplankton species in the ICES area

The WG discussed the requirements for the production of an ICES cooperative report. The WG were informed of a UK report that will be generated during 2008 examining the causal links between HABs and eutrophication. Information on a proposal for a SCOR Working Group on 'Land Based Nutrient Pollution and the Relationship to Harmful Algal Blooms in Coastal Marine Ecosystems" was circulated. The WG expressed concern that there was no representation from WGHABD or feedback to IOC-IPHAB on this WG and identified a candidate to attend.

The WG produce a number of outputs on an annual basis such as new findings, national reports, HAE – DAT. Recent developments in the identification of species and new phycotoxin syndromes in the ICES area as well as the requirements of aquaculture managers was considered to necessitate the generation of a report summarizing this information. Rather than consisting a series of distribution maps the WG felt that it would be pertinent to approach this report on a regional basis e.g. North Sea, Baltic, Northern US etc.

The WG felt that two activities would be appropriate:

1) The WG would review the UK report on the causal links of HABs to be produced during 2008 as well as the output from the proposed SCOR working group on HABS and eutrophication. The requirement for the WG to produce an additional report on the causal links between HABs and eutrophication will be assessed after reviewing these outputs.
2) A descriptive report detailing the phycotoxins and species distributions in the ICES area will be produced. This report would take a regional approach and consider the species and toxins of concern in different areas. The WG discussed the work involved in generating this report and asked for five volunteers to form an editorial committee to drive this forward. It was decided that this committee would meet for an additional day after the WGHABD meeting in 2009 to discuss the generation of this report. This committee is made up of delegates from Canada, Germany, Sweden, Scotland and Germany.

6 Term of reference d)

6.1 Review developments in knowledge pertaining to the development of *Verrucophora* and *Chattonella* blooms

Bengt Karlson (with contribution from Per Andersen, Denmark, and Lars Naustvoll, Norway)

Verrucophora farcimen is a flagellate belonging to the class Dictyochophyceae (Edvardsenet al, 2007). It has previously been known as *Chattonella* aff. *verruculosa.* Fish are affected by *V. farcimen* showing symptoms similar to anoxia. Fish gills are clogged by the algae and mucus is produced. It is unknown if toxins are involved. No toxins have been found during blooms in Norway. Large blooms of *V. farcimen* were observed in the Skagerrak area for the first time in 1998. Blooms have occurred in the area also in years 2000, 2001, 2004, 2006 and 2007. Mortalities of wild fish (*Belone belone,* Garfish) were observed in year 1998. In that year about 350 tonnes of farmed salmon died in Southern Norway and in 2001 about 1100 tonnes of farmed salmon died in the same area. In year 2006, farmed fish was affected in the Danish part of the Southern Kattegat. About 18 tonnes of *Salmo trutta* died in a land-based fish farm. In 2007, about 48 tonnes of *Salmo gairdneri* died in a Danish fish farm in the same area. Fish alive after the first few days of the incident survived the following period, although the observed concentrations of *Verrucophora* increased from 200.000–500.000 cells per litre to more than 9 million cells per litre from week no. 11 to week no. 13.

The blooms in years 1998 and 2000 developed in the Southern North Sea and were transported into the Skagerrak and the Kattegat by currents. These blooms were observed mainly in April-May. From 2001 and onwards blooms have mainly occurred in early spring shortly after the diatom spring bloom and the blooms seem to start in the Kattegat. *Verrucophora farcimen* obviously competes well with diatoms. An early hypothesis was that the organism may be an introduced species but reanalysis of preserved samples show that it was present in the area in 1993. In culture experiments with *V. farcimen* no resting stages have been observed. *Verrucophora farcimen* now seems to be well established species in the area and occur year around. Future fish mortalities due to *V. farcimen* are to be expected, especially among farmed fish.

Preliminary advice to fish farmers based on experiences 1998–2007

- Risk for damage to fish at 250 000 cells l^{-1}
- High risk for damage to fish at 500 000 cells l^{-1}

Reference

Edvardsen, B. *et. al* 2007. *Verrucophora farcimen* gen. et sp. nov. (Dictyochophyceae, Heterokonta)—a bloom-forming ichthyotoxic flagellate from the Skagerrak, Norway, J. Phycol. 43, 1054–1070 2007.

7 Term of Reference e)

7.1 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.

7.1.1 Characteristic profiles of spirolides and PSP toxins in strains of *Alexandrium ostenfeldii* from the North Sea and the Baltic Sea

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It is well known that *Alexandrium ostenfeldii* of different provenience is able to produce spirolides. The molecular structure consists of a spiro-linked, tricyclic system of polyethers and a seven-membered spiro-linked cyclic imine moiety. Especially spirolides belonging to groups A-D and G are biologically active, and moreover, spirolides C, D and G, which wear two vicinal methyl groups at the cyclic minimum moiety, are resistant towards enzymes and acids. Obviously, the presence of vicinal methyl groups in the cyclic imine moiety can prevent an enzymatic or acid hydrolysis to a keto-amine. Thereby, the observed spirolide pattern highly depends on the marine region. Strains from *A. ostenfeldii* in Canada (Nova Scotia) generally produce spirolide A, C, D and 13-desmethyl spirolide C.

However, *Alexandrium ostenfeldii* KO287 isolated in 1986 from Limfjord, Denmark, produced 13 19-didesmethyl spirolide C, spirolide G and 13-desmethyl spirolide C, i.e. only spirolides with two vicinal methyl groups, whereby the same profile of spirolides resulted as it could be observed in phytoplankton containing *Alexandrium ostenfeldii* analysed by LC-MS/MS during a research cruise in 2000 across the North Sea.

Therefore, incubation experiments by feeding mussels (*Mytilus edulis*) and oysters (*Crassostrea gigas*) with *Alexandrium ostenfeldii* KO287 were performed to elucidate the metabolization of those spirolides. The subsequent measurements of the extracts from shellfish tissues by LC-MS/MS revealed no characteristic differences in the spirolide profile compared to the profile of *Alexandrium ostenfeldii* KO287. Thus a metabolisation or hydrolysis of the spirolides produced by KO287 in mussels and oysters can be excluded.

Other neurotoxins with regard to *Alexandrium* species were PSP toxins, and in 1992, it was published that *A. ostenfeldii* from the Baltic Sea (Limfjord, Denmark) emerged as a potent PSP producer. In this context, cultures of several strains of *A. ostenfeldii* isolated in the NE regions of the Baltic Sea (Gulf of Finland) were checked concerning the presence of spirolides and PSP toxins, respectively. Surprisingly, those strains produced no spirolides, but higher concentrations of PSP toxins. In addition, the cultures of those strains from brackish waters showed better growth with higher salinity and the greatest PSP content per cell was observed under nitrogen limited growth conditions. These findings could be interpreted that PSP production may be a way for nitrogen fixation.

In 2006 mussels (*Mytilus edulis*) from Norsminde Fjord (Western Baltic Sea) were tested positive for contamination with PSP toxins, whereby the toxin profile was composed only by GTX 2, GTX 3 and STX, i.e. similar to the PSP pattern produced by the strains of *A. ostenfeldii* from the NE regions of the Baltic Sea. The control of phytoplankton from Norsminde Fjord results in the assumption that *A. minutum* could be the causative organism. However, all strains of A. *minutum* ever investigated exhibited constant PSP toxin profile characterized exclusively by the presence of GTX 1 and GTX 4 as well as GTX2 and GTX 3. Therefore, incubation experiments with mussels *(Mytilus edulis*) and both *A. minutum* CCMP-113 and *A. ostenfeldii* AOTV-A4 were performed.

The experiments were accompanied by effective analytical methods for toxin determination, which allow monitoring all changes in toxin compositions. Especially, the stability of the most toxic PSP toxins as the gonyautoxins and saxitoxin could be demonstrated. In addition, it could be confirmed that the accumulation of the PSP toxins is associated with epimerization of ß- to more stable α- epimers, i.e. the formation of the more toxic GTX 2 from GTX 3 and GTX 1 from GTX 4, respectively.

In the mussels from Norsminde Fjord the dominance of GTX 2 over GTX 3 could be observed, whereas GTX 4 and GTX 1 were not detectable. These results allow the conclusion that *A. minutum* can be excluded definitely as the source for PSP toxin accumulation of those mussels from Norsminde Fjord.

Summarizing it can be stated that the marine dinoflagellate *Alexandrium ostenfeldii* abundant in North Sea and Baltic Sea is a potent producer of marine biotoxins. Both groups of neurotoxins spirolides and PSP toxins can be generated. Characteristic toxin profiles with dominance of highly toxic and stable compounds are observed, which can be transferred in the marine food chain.

7.1.2 *Peridinium quinquecorne*:

Bengt Karlson, SMHI, Sweden

Figure 1. Location – Askimsbadet (beach) at Askim (Suburb of Gothenburg, Sweden) Photographs by Ann-Turi Skjevik.

6 June 2007: Female swimmer bathing at the beach at Askimbadet outside Gotenburg (Figure 1) noticed feeling of paralyzation, numbness, stinging needles and burning sensation in skin. Visit to hospital – medical doctor suspected allergic reaction. Symptoms remained next day. Also other swimmers were affected but less so.

Phytoplankton sampling:

8 June	
Euglenophyte	2 400 000 cells l^{-1}
Peridinium quenquecorne	1 500 000 cells l^{-1}
12 June	
Euglenophyte	130 000 cells l^{-1}
Peridinium quenquecorne	60 000 cells l^{-1}

Species known from Asia, Spain and Norway, (E. Dahl, pers. comm.)

7.1.3 Identification of toxic and benign species of *Alexandrium* in Scottish waters

Liz Turrell FRS, Aberdeen Scotland

Contamination of shellfish with paralytic shellfish poisoning (PSP) toxins and spirolides (SPXs), produced by *Alexandrium* species, poses a continual threat to the sustainability of the Scottish aquaculture industry (Figure 2).

Routine light microscopy (LM) analysis of water samples from around the Scottish coast has previously identified *Alexandrium* Halim (Dinoflagellata) as a regular part of the spring and summer communities. However, LM analysis using Lugol's fixed material prevents the identification of these cells to species level and does not provide any information on the toxicity of the phytoplankton populations. To understand the complexity of shellfish toxin events in Scottish waters a deeper knowledge of the distribution of *Alexandrium* species and the toxicity of *Alexandrium* populations is required.

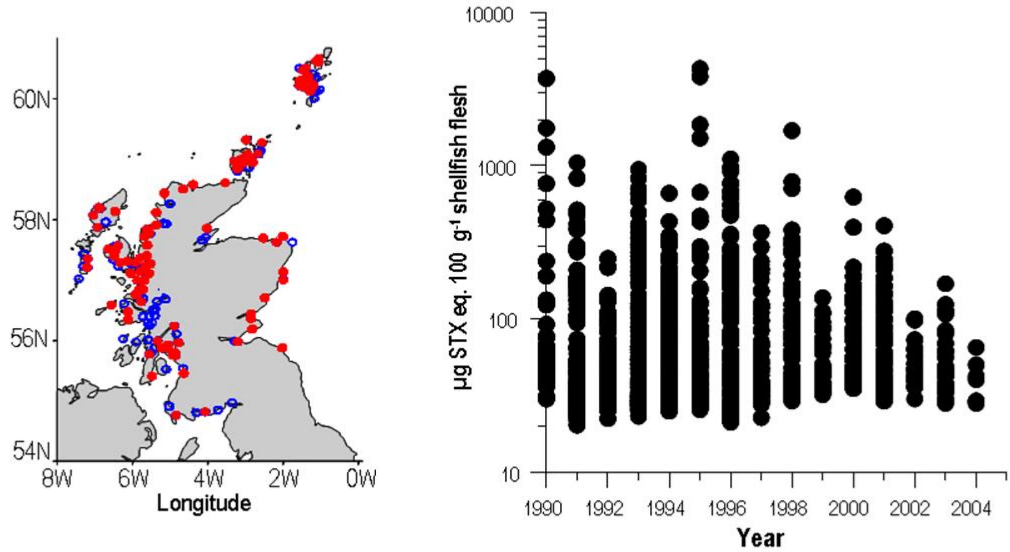

Figure 2. Shellfish in Scottish waters contaminated with PSP toxins: Continual problem to the aquaculture industry.

In this study, Alexandrium cells isolated from sediment and water samples obtained from Scottish waters were established in laboratory culture. Using the cal plate dissections four species of *Alexandrium* were identified: *A. tamarense*, *A. minutum*, *A. ostenfeldii* and *A. tamutum*. The identification of these isolates was confirmed by molecular characterization based on their LSU sequence. In addition, molecular characterization and phylogenetic analysis was able to show the presence of two strains of *Alexandrium tamarense*: Group I (North American Ribotype) and Group III (Western European ribotype) as defined by Lilly *et al.* 2007. This associates the *Alexandrium tamarense* isolates from Scottish waters with both PSP toxin production (Group I) and non-toxin production (Group III).

Subsequently, laboratory cultures of the Alexandrium isolates were assessed for PSP toxin production using the Jellet Rapid Test (JRT) with PSP toxin profiles determined using a newly developed hydrophilic interaction chromatography-linear ion spectrometry method. The production of SPXs was determined using liquid chromatography with mass spectrometry (Figure 3).

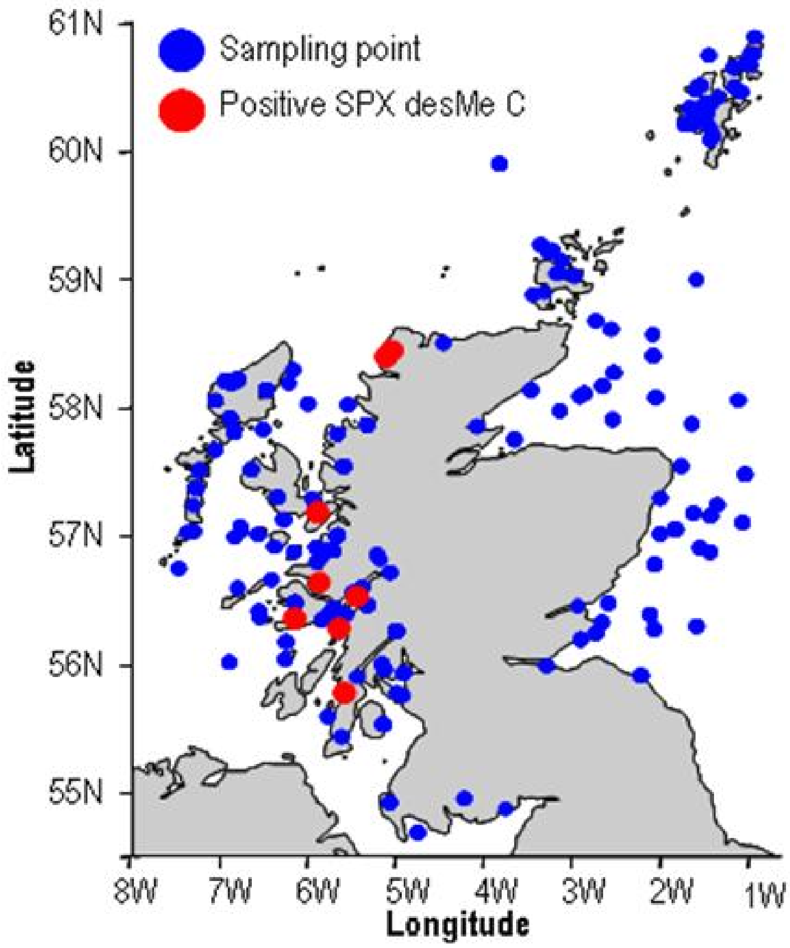

Figure 3. SPX des MeC detected in spring 2004: Detected in cultured mussels and Pacific oysters on the West coast of Scotland.

A. tamarense (Group I) isolated from the Scottish east coast showed a complex array of toxins (ca. 2000 fg STxdiHCl eq cell^{-1}) with the major toxins being saxitoxin, neosaxitoxin, gonyautoxin-4 (GTX-4), GTX-3, and C2. Lower concentrations of corresponding epimeric C and GTX toxins were observed. *A. ostenfeldii* isolated from offshore sediments produced low quantities of the PSP toxins (saxitoxin and neosaxitoxin (ca. 100 fg STxdiHCl eq cell^{-1})) and spirolide desMethyl-C (ca. 3 fg SPX des MeC-cell-1) in addition to other spirolide analogues during exponential growth. A. minutum, A. tamutum and A. tamarense (Group III) did not produce toxins under the growth condition used in these experiments.

Historically, it was considered that *Alexandrium* species found in Scottish waters produce potent PSP toxins while *Alexandrium ostenfeldii* has previously been suspected as a possible source of PSP and SPX toxicity in Scottish waters. This study confirmed the ability of *A. ostenfeldii* to produce a limited range of PSP toxin analogues and SPXs and has highlighted the presence of both PSP toxin producing and benign species of *Alexandrium.* The consequences for the shellfish industry and public health will now need to be considered.

Reference

Lilly, E. L., Halanych, K. M., Anderson, D. M. 2007. Species boundaries and global biogeography of the Alexandrium tamarense complex (Dinophyceae). Journal of Phycology, 43, 1329–1338.

7.1.4 GEOHAB activities

Robin Raine, Chair, GEOHAB Scientific Steering Committee, The Martin Ryan Institute, National University of Ireland, Galway, Ireland

The Global Ecology and Oceanography of Harmful Algal Blooms (GEOHAB IOC SCOR) Programme, endorsed by the Scientific Committee on Oceanic Research (SCOR) and the Intergovernmental Oceanographic Commission (IOC) of UNESCO, is an international programme aimed at fostering and promoting cooperative research directed toward improving the prediction of harmful algal bloom events. GEOHAB is a programme of international cooperative research on HABs in marine and brackish waters. The GEOHAB Scientific Goal is to improve prediction of HABs by determining the ecological and oceanographic mechanisms underlying their population dynamics, integrating biological, chemical, and physical studies supported by enhanced observation and modelling systems. The GEOHAB Mission is to foster international cooperative research on HABs in ecosystem types sharing common features, comparing the key species involved and the oceanographic processes that influence their population dynamics. The efforts by GEOHAB fall under three categories: national/regional studies; targeted research and the four core research projects on HABS in Upwelling systems, Eutrophic Systems, The Stratified Environment and finally, Coastal Bays and Fjords.

Activities over the last year have seen the development of cooperative work through Asian GEOHAB. A first meeting organized by Dr Ken Furuya with participants from a number of Asian countries including Japan, China, and Korea took place over 2 days after the GEOHAB Scientific Steering Committee in Tokyo in March 2007. This was followed a year later by a much larger meeting in Nha Trang, Vietnam in February 2008 involving over 10 countries. A report on the second meeting will be published by GEOHAB in advance of the next international HAB conference in Hong Kong next November. The stage is now set for future cooperative studies on HABs in the region.

One targeted research project has been endorsed by GEOHAB over the past year which was the NORCOHAB cruise organized by Allan Cembella (AWI, Bremerhaven) investigating HABs in the North Sea (see contributions by Cembella and Luckas in this report).

The Core Research Projects (CRPs) continue apace, with meetings through the year of the committees of both the Upwelling Systems and Eutrophication CRPs. A special publication of progress in Oceanography is being prepared by the Upwelling group which should be in the press before the Hong Kong HAB conference. The HABs in the stratified environments CRP has been implemented through cooperative research cruises in Spain, France and Ireland mainly through the HABIT programme. For an example outcome of this activity see the article on the *Dinophysis* bloom off southern Ireland in 2008 described in this report.

Future developments of GEOHAB activities include a workshop on modelling biophysical interactions relevant to the development of HABs. This workshop, which will be organized with similar forma to the HABWATCH workshop, should take place in 2009. For further information contact Dennis McGillicuddy (WHOI). An agenda item for the next SSC meeting is to start a new CRP on epibenthic harmful dinoflagellates, a coverall term which includes the near bottom dwelling, epiphytic and sand-dwelling species. The global problems caused by the ciguatera toxin producing dinoflagellates, and *Ostreopsis* among many others, demands that progress is made on the biogeography, substrate preference and methodology associated with these HAB species.

7.1.5 Transport of a high density thin layer of *Dinophysis acuta* off the coast of Ireland

Robin Raine, The Martin Ryan Institute, National University of Ireland, Galway, Ireland

A ten day research cruise took place on board the Irish National Research vessel *Celtic explorer* at the end of July 2008. This cruise was principally associated with the HABIT (EU FP6 project) and involved scientist groups led by Raine (NUI Galway), Gentien (IFREMER, Brest), Reguera (IEO, Vigo) and Fernand (CEFAS) from participating countries. Studies using fine scale sampling devices such as the in situ particle profiler and fine scale sampler of IFREMER revealed the presence of a sub surface thin layer 2–3 m thick, of *Dinophysis acuta* which contained cell densities of 10 000–55 000 cells per litre (Figure 4). The layer was located approximately 5 metres above the subsurface fluorescence maximum. Small-scale spatial surveys indicated that the patch of *D. acuta* had dimensions of ca. 3 km diameter, which was of a scale smaller than the tidal excursion, which is typically 6–7 km in this region. The patch was located within the seasonal coastal jet, which flows westwards along the south coast of Ireland, and is promoted by the interactions of tidal mixing and stratification as the water depth shallows towards the coastline (Fernand *et al.*, 2006). The main part of the fieldwork involved tracking the patch westwards within the coastal jet, which has a net alongshore speed of 7 km per day.

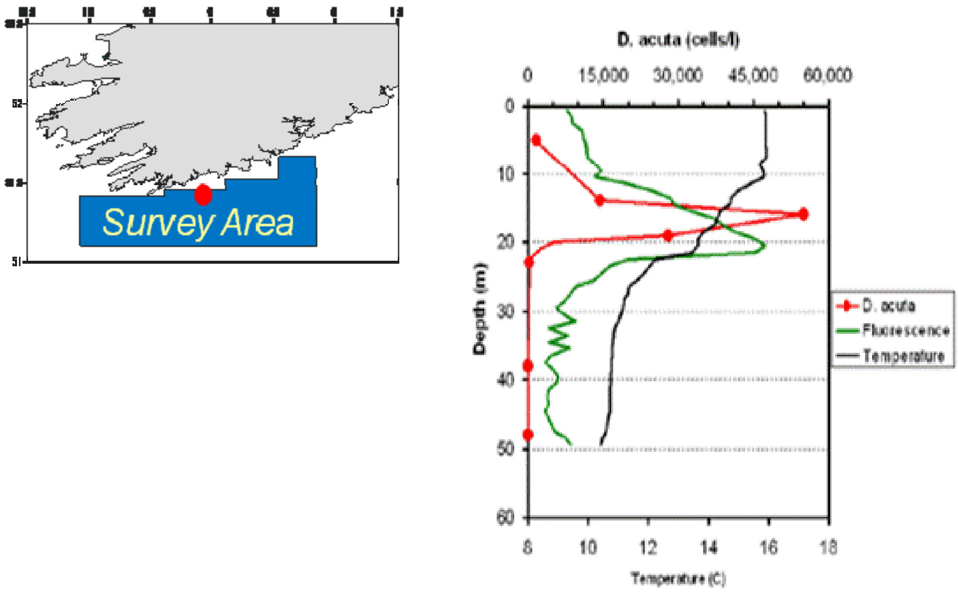

Figure 4. Subsurface layer, 5 m thick, of Dinophysis acuta at ca. 16 m depth.

The outcomes of these particular studies therefore included:

- the horizontal scale of the patch of *D. acuta* was smaller than the (daily) length scales of tide and thus similar events and features will be very difficult to find and study in the future.
- the movement of the patch followed the coastal jet at the predicted rate over a period of six days; at the end of the cruise the patch was located only 5 km away from predictions of the coastal jet path, which did not include any windforcing effects.
- the phytoplankton monitoring data at coastal stations in the region did not reveal any significant (>200 cells per litre) cell densities of *D. acuta.* This adds to the accumulating information of HAB populations existing offshore which are not always picked up in coastal monitoring datasets. This highlights the requirement for offshore HAB observatories, particularly when windforcing can under correct conditions blow these populations onshore within 2–3 days, as is the case in southwestern Ireland.

7.1.6 Phytotest

Siobhan Kavanagh[1] , Majella Maher[1], Rafael Salas[2], and Joe Silke[2]

[1]National Diagnostic Centre National University of Ireland Galway
[2] Phytoplankton Unit, Marine Institute, Ireland

PHYTOTEST is a recently completed 3-year research and development project funded through the Irish Marine Institute Strategic Research Programme in Advanced Technologies as part of the National Development plan 2000–2006. This project involved collaboration between the National Diagnostics Centre at NUI Galway and the Marine Institute. The objective of the PHYTOTEST project was the development of nucleic acid tests (primarily real-time PCR assays run on the Light-Cycler® (LC®) platform) for the identification of key toxic phytoplankton species (*Dinophysis* and *Pseudo-nitzschia* species) in Irish waters. The final year of the project involved a technical transfer of these assays into the MI, to act as a support for the phytoplankton monitoring service.

***Dinophysis* species real-time PCR assay**

As part of the assay evaluation process, PCR analysis of wild samples containing Dinophysis cells was carried out. Samples from the MI phytoplankton-monitoring programme were selected for testing, as they are routinely examined by light microscopy for the presence of HAB species. Fifty-five Lugol's iodine preserved samples from the 2006 MI phytoplankton-monitoring programme were tested with the assay. Thirty-three of these samples were reported to contain Dinophysis species cells based on microscopic analysis, with cell numbers in the range of 1–8 in 25 ml. Twelve samples contained only 1 cell of either D. acuta or D. acuminata. The real-time PCR assay detected the presence of Dinophysis species in all samples reported to contain Dinophysis cells. The real-time PCR assay identified D. acuta or D. acuminata in DNA extracts from all samples reported to contain either species by microscopy. The assay detects down to 1 D. acuminata cell and 1 D. acuta cell in 25 ml of preserved wild sample.

An advantage of Hybridization probe technology is the potential to detect and distinguish between two or more species with a single probe set, through the application of melt-peak analysis. Specific-melt peaks were observed for D. acuta and

D. acuminata with the real-time PCR assay from the four samples reported to contain both species by light microscopy, illustrating that this assay simultaneously detects and discriminates between species, even when occurring at small numbers. There was no detection of Dinophysis in the Dinophysis negative MI samples. A specificity panel assay, including a range of phytoplankton species commonly found in Irish waters was tested against the assay and no cross-reactivity was observed.

The development of these probes and their success in detection with high specificity is a useful development in future studies of Dinophysis distributions and for the future development of in situ autonomous detection of this species which causes huge disruption to the Irish shellfish industry.

***Pseudo-nitzschia* species real-time PCR assays**

As with the *Dinophysis* species assay, evaluation of the *Pseudo-nitzschia* species assays was performed using wild samples that had been examined for the presence of *Pseudo-nitzschia* cells by light microscopy. Fifty-eight Lugol's iodine preserved samples from the 2006 MI phytoplankton-monitoring programme were tested with each of the *Pseudo-nitzschia* species (*P. australis*, *P. fraudulenta*, *P. delicatissima*, *P. pungens*) real-time PCR assays. Forty-one of the samples contained *Pseudo-nitzschia* species cells, with numbers ranging from 40–3889 cells.l^{-1} in 25 ml preserved sample. In addition to these samples, seven Lugol's iodine preserved samples were received for testing from Dunstaffnage Marine laboratory. All seven samples contained *Pseudo-nitzschia* species cells, with numbers varying from 31–2750 cells in 25 ml.

There was 100% detection of *Pseudo-nitzschia* species in the forty-eight samples containing *Pseudo-nitzschia* cells from 2006. Two samples produced melt peaks at 53 ^{0}C with the *P. pungens* assay, indicating the presence of *P. multiseries* cells. The real-time PCR assays identified the presence of two or more species in 78% of the samples. Greater than half of the 2007 phytoplankton monitoring programme samples tested were also seen to contain two or more *Pseudo-nitzschia* species, in support of the reported co-occurrence of *Pseudo-nitzschia* species from the literature (Hasle *et al.*, 1996, Cusack et al, 2004). There was no detection of *Pseudo-nitzschia* species in the seventeen samples that were negative by microscopy for *Pseudo-nitzschia*.

Implementation of PHYTOTEST

Since July 2007, the technology transfer phase of the project has gotten underway with the purchase and installation of the LC® 480 instrument and training of MI Phytoplankton Unit staff in real-time PCR and nucleic acid extraction methodologies. Initial testing of the assays with positive controls indicated that the real-time PCR assays for *Dinophysis* and *Pseudo-nitzschia* species are working successfully on the LC® 480 instrument. The specificity and limits of detection for these assays were also verified. A performance evaluation of the assays for the identification of the relevant toxic species in wild samples was performed between the MI (LC® 480 instrument) and at the NDC (LC® 2.0 capillary machine) as part of a validation of the real-time PCR assays. It is expected that testing of samples from the monitoring programme will be carried out by MI Phytoplankton Unit staff after March 08

7.1.7 Unusual lipophilic marine toxins profiles obtained using passive sampling in Irish waters

Elie Fux[1, 2] and Philipp Hess[1]

[1]. Marine Institute, Biotoxin Chemistry, Oranmore, Ireland

[2]. Dublin Institute of Technology, School of Chemistry and Pharmaceutical Sciences – Dublin 8 – Ireland

A passive sampler suitable for the detection of lipophilic marine toxins was developed in New Zealand and referred to as solid phase adsorption toxin tracking (SPATT) [1]. Recently, we have shown the applicability of the technique in mesocosm experiments and developed an efficient extraction technique for subsequent LC-MS detection [2].

We now report some examples of SPATT samplers that were obtained as part of the monitoring of lipophilic marine toxins in shellfish production sites on the West Coast of Ireland. Analyses of the passive samplers using a UPLC-MS/MS method [3] detected okadaic acid (OA), dinophysistoxin-2 (DTX2), pectenotoxin-2 (PTX2), pectenotoxin-2 seco-acid (PTX2sa), azaspiracid-1 (AZA1), azaspiracid-2 (AZA2), yessotoxin (YTX) and spirolide-13-desMethyl-C (SPX-13-DesMe-C).

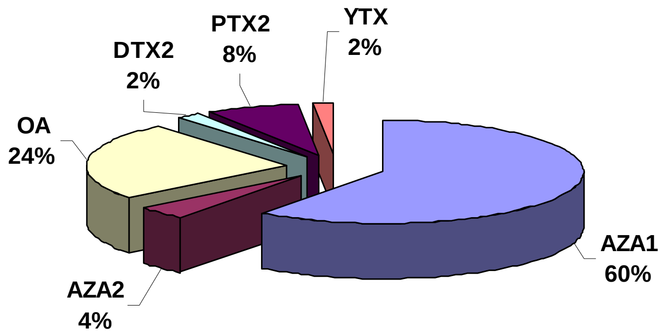

Figure 5. Toxin profile obtained in passive sampler deployed in the Northwest of Ireland from the 31/8/05 to the 8/9/05.

Toxin profile in station 1 – Northwest of Ireland

The SPATT consisted of 3 g of polymeric resin HP-20 and was immersed in the water for seven days (from 31/8/05 to 8/9/05). During this period, 17.7 μg of toxins accumulated in the SPATT. The toxin profile is shown in Figure 5.

AZA1 accounted for 60% of the detected toxins and was also the predominant toxin that was detected in mussels from the production area (data not shown). AZA2 was also detected at lower levels (4%). In addition, traces of DTX1 were also detected slightly above the limit of detection (LOD = 4 ng/g of resin based on OA response factor). A rise in YTX concentration was observed in the SPATTs during the sampling period in the absence of known YTX producer and coincided with quantifiable amounts of YTX in the transplanted mussels. Phytoplankton sampling indicated that the numbers of *Heterocapsa sp*, *Prorocentrum balticum/minimum* and *Prorocentrum dentatum* rose at the same period as YTX in the SPATTs. Analysis of cells of *Lingulodinium polyedrum* from Ireland by ELISA already detected low amounts of YTX in the past (Clarke, 2001 – Personal Communication). The origin of YTX in the present study should be further investigated as none of the above organisms is known to produce YTX and none of the known YTX producing organisms (*Protoceratium reticulatum* [4], *Lingulodinium polyedrum* [5] and *Gonyaulax spinifera* [6]) were detected in the water.

Toxin profile in station 2 – Southwest of Ireland

The SPATT consisted of 3 g of polymeric resin HP-20 and was immersed in the water for seven days (from 21/9/05 to 5/10/05). During this period, 8.5 µg of toxins accumulated in the SPATT. The toxin profile is shown in Figure 6 and consisted principally of toxins produced by *D. acuta* in Ireland (OA, DTX2 and PTX2). In addition, AZA1, AZA2 and SPX-13-desMe-C were also detected.

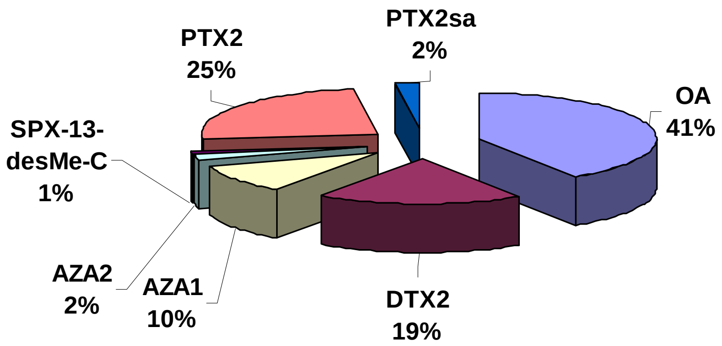

Figure 6. Toxin profile obtained in passive sampler deployed in the Southwest of Ireland from the 21/9/05 to the 5/10/05.

Conclusions

This is the first report that unambiguously identified the occurrence of SPX-13-deMe-C and YTX in Ireland. The combination of the SPATT and the UPLC-MS/MS method allowed for the detection of numerous lipophilic marine toxins. In addition to the sensitivity provided by these techniques, toxin profiles can be obtained very rapidly. We have shown that microgram amounts of toxins can accumulated in the SPATT within 12 hours {Fux, Marcaillou, *et al.*, #4407}and that excellent recovery of toxins can be obtained using a 25 minutes elution with methanol [2]. Analysis of one sample by UPLC-MS/MS only requires 6.6 minutes [3].

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7.1.8 New Findings on Dinophysis –Spatt samplers

Beatriz Reguera, IEO Vigo Spain

Beatriz Reguera (Spain) reported on an unusual moderate peak of a small (<40 µm) *Dinophysis* sp. in the Galician Rías Baixas during 2006 and 2007. The peak in 2006 appeared in mid June before the usual spring (May-June) proliferations of *D. acuminata*. This small *Dinophysis*, that belongs to the *D. acuminata* complex, and is labelled as *D. acuminata* or as *D.* cf *acuminata* in different parts of the world, has a very regular oval contour and can be easily distinguished, on morphological basis, from the local *D. acuminata* strains that resemble the original description of *D. acuminata* Claparède et Lachmann. Its morphology agrees with the description of *Dinophysis ovum* Schütt. LC-MS analyses of net-hauls rich on this species and of single-cell isolates (by micropipette), showed a toxin profile composed of only okadaic acid, and no traces of DTXs and PTXs. For the first time, molecular analysis of mitochondrial DNA were carried out in an attempt to find regions of the genome that showed a higher variability, within *Dinophysis* spp., than the ribosomal DNA. The results (Raho *et al.*, accepted) showed that the sequence of the mitochondrial cytochrome C carboxilase (cox 1) of the two "species" presented a much higher variability, and is, therefore, a more suitable region of the genome for discrimination between *Dinophysis* spp.

Information was also presented on a full year monitoring, by means of passive samplers (SPATT *sensu* MacKenzie 2004) of extracellular lipophilic toxins during the *Dinophysis* season. Simultaneously, estimates of cellular toxin content from plankton concentrates and from single cell isolates at the same station were obtained. All analyses were carried out by liquid chromatography coupled to mass spectrometry (LC-MS). Discrepancies in toxin profile and content between estimates from both kinds of samples (picked cells *vs.* plankton concentrates) and the seasonal variability of toxins adsorbed by the SPATT resins suggest that extracellular toxins are most abundant after the blooms' decline and that an important proportion of the toxins can remain in the water, off the cells, attached to organic aggregates weeks after the producing cells have disappeared. The use of SPATT was not efficient, as an early warning tool, for the detection of *Dinophysis* cells. Nevertheless, it was good to warn, 3 weeks in advance, on the onset of shellfish harvesting closures (Pizarro 2008).

References

MacKenzie, L., Beuzenberg, V., Holland, P., McNabb, P. and Selwood, A. 2004. Solid phase adsorption toxin tracking (SPATT): a new monitoring tool that simulates the biotoxin contamination of filter feeding bivalves. *Toxicon* 44: 901–918.

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7.1.9 Evaluations and method development of solid phase adsorbents for phycotoxins in the marine environment

Liz Turrell, FRS, Aberdeen Scotland

Solid-phase adsorption toxin tracking (SPATT) was recently developed to facilitate monitoring of lipophilic shellfish toxins (LSTs) in shellfish harvesting areas (Mackenzie *et al.*, 2004). SPATT for LSTs was founded on the observation that when low levels of toxin-producing algae were present in the water column significant amounts of toxins were released in seawater. A lag between detection of released toxins adsorbed onto porous synthetic resin, phytoplankton peak cell densities and highest toxin concentrations in shellfish was demonstrated, suggesting that SPATT technology could be a useful predictive tool for the onset of a toxic event.

In this study, we sought to further evaluate adsorbents that could be applied to SPATT for LSTs and additionally to hydrophilic phycotoxins including paralytic shellfish poisoning (PSP) toxins and domoic acid (DA), associated with amnesic shellfish poisoning (ASP).

Previously, a synthetic adsorbent resin DIAION® HP20, in the form of spherical beads was used for the adsorption of LSTs from seawater (Mackenzie *et al.*, 2004). In this study, a further resin, SEPABEADS® SP700, produced by the Mitsubishi Chemical Cooperation was also tested for its capacity to adsorb LSTs from seawater. The adsorption and recovery of a range of LSTs (incl. okadaic acid, dinophysistoxins, yessotoxins, azaspiracids, pectenotoxins and spirolides) from seawater using HP20 and SP700 was investigated. Results demonstrate that the SP700 resin was superior to HP20. For example, after 4 hours incubation ca. 60% of available okadaic acid was adsorbed onto the SP700 resin compared to 30% for the HP20 resin with recovery of the toxins using methanol.

Optimized SPATT bags, containing SP700 resin beads as the adsorbent, were deployed weekly (from 2005) in conjunction with sampling of phytoplankton at Loch Ewe in Scotland. SPATT extracts were analysed by LC-MS using a multi-toxin analysis for LSTs.

During the monitoring period OA and PTX-2 were detected in the SPATT sachets in the absence of known causative phytoplankton, *Dinophysis* spp., in the water column. Demonstrating that toxins can be present in the water column for weeks when cells are not detected. However, concentrations of OA and PTX-2 increased prior to detection of *Dinophysis acuminata* suggesting that SPATT does have potential as an early warning system. Crucial questions will be to determine if the toxins are actively liberated by healthy cells or are associated with the demise of the cells.

The causative organism of AZAs was previously recorded as the dinoflagellate, *Protoperidinium crassipes* (James *et al*., 2003). This was ascertained by picking a large number of cells of *P. crassipes* from a sample of phytoplankton taken while shellfish were contaminated with AZAs, and analysing the cells for AZAs using LC-MS. In this study, AZA-1 was not detected in SPATT sachets before or immediately after highest peak cell densities of *Protoperidinium* spp. were observed. Highlighting that SPATT may provide a better indicator of possible shellfish contamination than phytoplankton monitoring using *Protoperidinium* spp. as an indicator species for toxicity. Indeed doubts exist within the scientific community on whether this dinoflagellate species is the main producer of this group of toxins and new findings presented to this group by Professor Allan Cembella show that AZAs are vectored through the marine environment with a previously unidentified thecate organism ‘Azadineum’ being the progenitor of this toxin group.

Using laboratory-scale experiments, we investigated the removal of two PSP toxins (neosaxitoxin, NEO and saxitoxin STX) from seawater using computationally designed polymers (CDPs). An ethylene glycol methacrylate phosphate (EGMP) based polymer was able to adsorb both PSP toxins completely. To optimize toxin recoveries, a variety of extraction procedures were examined and a protocol developed enabling the recovery of 97% of NEO and 92% of STX from the CDP. An additional CDP and a variety of adsorbents (activated glass beads, resins and zeolites) were assessed for the adsorption and recovery of DA from seawater. The best adsorbent was found to be Amberlite®XAD761 which demonstrated nearly 100% binding of available DA.

Extensive field trials in European waters will now be progressed to determine if SPATT (for LST, PSP and ASP toxins) has potential as an early warning system for shellfish contamination through EC Collective Research SPIES-DETOX (Contract No. 030270–2).

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7.1.10 Rapid Enzyme Linked Immunosorbent Assay for the detection of Domoic Acid

Reported by Pat Tester and co-authors, National Ocean Service, NOAA

Domoic acid (DA) is a potent toxin produced by bloom-forming phytoplankton in the genus *Pseudo-nitzschia* which is responsible for causing amnesic shellfish poisoning (ASP) in humans. ASP symptoms include included vomiting, diarrhoea, and in more severe cases confusion, loss of memory, disorientation and even coma or death. This paper describes the development and validation of a rapid sensitive enzyme linked immunosorbent assay (ELISA) test kit for detecting DA. The assay gave equivalent results to those obtained using standard HPLC and FMOC-HPLC methods. It has a linear range from 0.1 to 3 ppb and was used successfully to measure DA in razor clams, mussels and phytoplankton. The assay required approximately 1.5 h to complete. The kit has a standard 96-well format where each row of eight wells is removable and can be stored until needed. The first two wells of each row serve as an internal control eliminating the need to run a standard curve each time. This allows as few as 3 and as many as 36 duplicate samples to be run at a time. Other assays which use standard curves generally require half the 96-well plate to be run at a time requiring that a sufficient number of samples be acquired before the assay can be preformed. In cases where only a few samples at a time are accumulated, the current format eliminates this need for a minimum number of samples, thereby allowing real-time sample processing and avoiding any degradation of DA which can occur during storage. The assay did not cross-react significantly with glutamate, kainic acid, epi- or iso-DA. The relatively low cost, accuracy, and rapid processing time offered by this assay make it useful to environmental managers and public health officials for monitoring DA concentrations in environment samples.

7.1.11 The Poseidon Expedition: Hunt for the source of azaspiracids

Reported by A. Cembella with contributions from B. Krock, U. Tillmann and U. John from the Alfred Wegener Institute for Polar and Marine Research, Bremerhaven, Germany

The NORCOHAB P-352 cruise aboard the R. V. Poseidon (Kiel) was conducted during early summer 2007 to study the coastal oceanographic processes and mechanisms underlying the dynamics of key toxic bloom species and the biogeographical distribution of their toxins in the water column. The cruise was endorsed by GEOHAB, under the auspices of the Core Research Project on HABS in Fjords and Coastal Embayments. The cruise transects were from Bremerhaven, Germany across Dogger Bank with detailed sampling initiated along the Scottish east coast from the Firth of Forth to the Shetland Islands. For comparison, sampling also conducted within the Norwegian Current on the south coast of Norway and then along the north coast of Denmark. In addition to primary and secondary transects, drift stations were sampled over several days on the Scottish and Danish coasts to obtain time-series data within a given water mass. Standard physical oceanographic parameters (temperature: ˚C, salinity: psu, σt) plus current velocity were supplemented with bio-optical measurements with multiple profiling fluorosensors and various passive optical profilers (for turbidity and diffuse attenuation), including a hyperspectral radiometer.

Our working hypothesis was that bloom dynamics of our key toxic species in the North Sea are regulated at least as much by “top down” factors such as grazing by copepods and protists than by “bottom up” factors, including light, temperature and nutrients. One of the related major objectives was to apply liquid chromatography coupled to a highly sensitive triple-quadrupole mass spectrometer (LC-MS/MS) to identify and quantify phycotoxins at trace levels throughout the cruise trajectory. With this equipment on board we were able to respond directly to relevant findings on the composition and concentration of toxin is various size-fractions. Plankton assemblages were analysed from various depths and size-fractions in near-real time (i.e. between stations) and from on-board grazing experiments. The analytical instrumentation provided a gold-mine of information, allowing us to characterize many toxin derivatives (of spirolides, yessotoxins, saxitoxin/gonyautoxins, domoic acid, okadaic acid/dinophysistoxins, azaspiracids) from particulate fractions from the water column at sub-picomolar concentrations. Despite the rather low concentrations of putative causative organisms, phycotoxins were found at most stations around the perimeter of the North Sea from plankton samples. Such sensitive analytical methods proved crucial to the dynamics determination of the occurrence and fate of toxins in various components of the planktonic foodweb.

Azaspiracids are a group of lipophilic algal toxins, at first mistakenly associated with the diarrhoetic shellfish poisoning (DSP) syndrome. Azaspiracid poisoning initially occurred in 1995 after eight people in the Netherlands became ill after consumption of mussels from the Irish west coast. Symptoms of the affected persons were nausea, vomiting, severe diarrhoea and stomach cramps - typical for DSP. However, the only DSP toxins in the mussels were okadaic acid and dinophysistoxin-2 and at very low concentrations, which could not explain the observed intoxications (McMahon and Silke 1996). Azaspirazid-1 (AZA-1) was isolated and structurally elucidated from shellfish as the causative compound of AZP (Satake *et al.*, 1998). Other structural variants of AZA-1 were subsequently detected and isolated from shellfish (Ofuji *et al.*, 1999; Ofuji *et al.*, 2001; James *et al.*, 2003). To date over 30 AZAs are known, either completely or partially described, of which AZA-1, AZA-2 and AZA-3 have been previously found in plankton samples (Furey *et al.*, 2003).

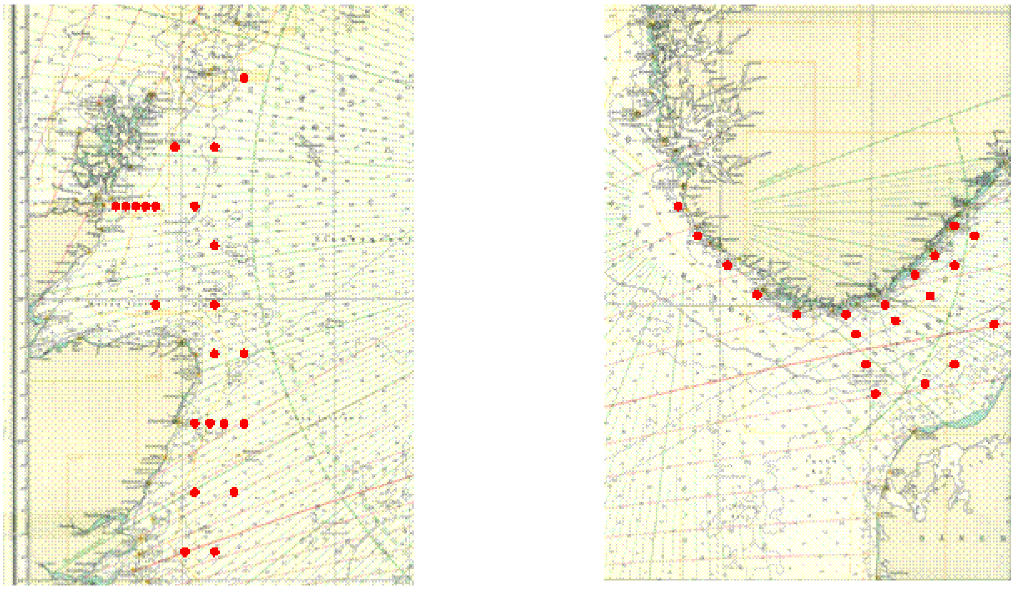

Figure 7. Stations in the North Sea where azaspiracids were found (filled circles) and not found (open circles) in size-fractionated plankton samples by LC-MS analysis during June–July 2007.

One highlight of the *Poseidon* cruise was the on-board determination of azaspiracids in plankton samples and the subsequent isolation and culture of the causative organism of azaspiracid poisoning (AZP) (not *Protoperidinium* as previously reported James *et al.*, 2003). The compound AZA-1 was first measured in size-fractionated plankton net tow (20 µm mesh size) material by a highly sensitive triple quadrupole-linear ion trap LC-MS/MS (Figure 1). Highest amounts of AZA-1 were later found in the 3 – 20 µm size-fraction from Niskin bottle casts. From this fraction, a large number of crude cultures were established by serial dilution. After 8 weeks of growth under controlled conditions, the cultures were screened for the presence of AZAs. From one crude culture containing AZA, a small dinoflagellate was subsequently isolated by microcapillary pipette and brought into pure culture. The isolated strain produces AZA-1, AZA-2 and a thus unknown isomer of AZA-2.

The proximal source of azaspiracid (AZA) is a small photosynthetic thecate dinoflagellate of approximately 12 – 15 µm long-axis diameter and 8 – 11 µm width, which was found at stations with high AZA-1 concentrations. The AZA-producing dinoflagellate has now been described morphologically and with reference to molecular phylogenetic characteristics for several genes (ssu rDNA, lsu rDNA, *cox*, ITS). The major azaspiracid component (AZA-1) was almost ubiquitous in the North Sea, but its abundance varied significantly. In the western North Sea, AZA-1 was most abundant along the southern Scottish coast, but further north around the Orkney and Shetland Islands, AZA-1 was only detected at trace levels. In general, AZA-1 concentrations were higher in the eastern North Sea with highest levels found in the southern Skagerrak.

The producing organism and its toxin components are under further investigation to determine the relationship with the AZP toxin syndrome and biogeographical distribution within the plankton.

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7.1.12 Update on operational forecasting of *Alexandrium* blooms in the Gulf of Maine, USA

Don Anderson, Woods Hole Oceanographic Institute USA

A coupled physical/biological model of *A. fundyense* population dynamics in the Gulf of Maine has been described in several recent publications (e.g. McGillicuddy *et al.*, 2005; Anderson *et al.*, 2005b). The model is initiated from large-scale maps of cyst distribution, with germination rates parameterized through laboratory experiments. Likewise, the growth of the resulting vegetative cells is regulated by light, temperature, and salinity, again parameterized using laboratory cultures. The physics of the system are well represented by a Regional Ocean Modeling System (ROMS) model for the Gulf of Maine, nested within two larger models – HYCOM (Hybrid Coordinate Ocean Model, maintained by the NRL and U. Miami) and the ROMS for the Mid-Altantic Bight and the Gulf of Maine, maintained by Rutgers and UCLA.

Two types of forecasting capabilities were discussed at this meeting – 1) Long-term (interannual) empirical forecasts and 2) synoptic forecasts (days to weeks).

Long-term and interannual forecasts

Forecasts of this nature are being made on the basis of two factors: 1) annual maps of *A. fundyense* cyst abundance; and 2) long-term trends in PSP toxicity in shellfish. The major development in this regard is the high abundance of *Alexandrium fundyense* cysts observed in GOM sediments in late 2007 (see Figure 1). Our numerical model of *A. fundyense* dynamics (e.g. McGillicuddy *et al.*, 2005) is initiated from maps of cyst abundance in the prior year. At this time, we are testing the hypothesis that cyst abundance is a first-order predictor of regional bloom magnitude in the Gulf of Maine. Our studies have generally demonstrated a correlation between the abundance of *A. fundyense* cysts in a mid-coast Maine "seedbed" and the overall magnitude of the bloom in the western Gulf of Maine and waters further to the south and west. Note that this does not mean that the cyst abundance correlates with levels or extent of PSP toxicity (though that is sometimes the case), as it is possible to have a lot of cells in offshore waters that are not delivered to shore (and the toxin monitoring stations) by ocean currents (influenced by wind and other factors).

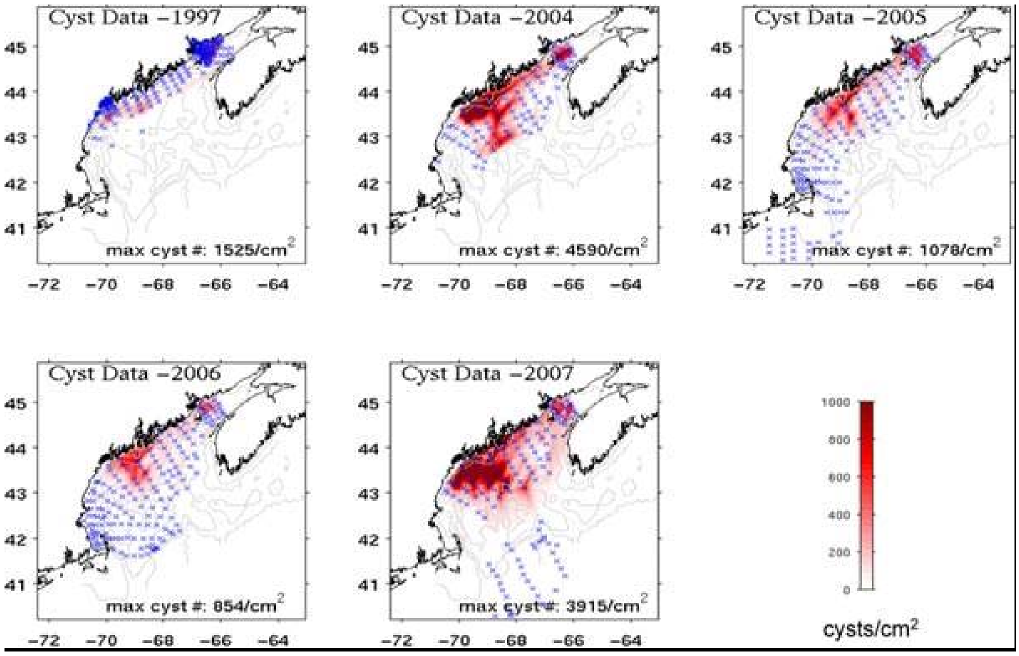

Figure 8. Maps of Alexandrium fundyense cyst abundance (cysts cm-2) in surface sediments of the Gulf of Maine for 1997, 2004, 2005, 2006, and 2007. (D. M. Anderson, unpub. data).

Figure 8 shows a map of the *A. fundyense* cysts that were in surface sediments of the Gulf of Maine in late 2007. That map is attached here, along with maps for 1997, 2004, 2005, and 2006. Considerable interannual variability in cyst abundance is evident. Note several features of the cyst data. First, the large increase in cyst abundance that occurred sometime between 1997 and 2004. Our modelling studies (He et al, in press) suggest that the very high cyst abundance off mid-coast Maine in late 2004 was the major cause for the extensive 2005 "red tide" in the western Gulf and southern New England (Anderson *et al.*, 2005), although northeast storms and high river run-off were supporting factors. Note that the cyst abundance in surface sediments actually decreased by about 50% after the 2005 bloom, leading to a 2006 bloom that was quite large, but not as extensive as the one in 2005. Cyst abundance fell again in 2006, but as seen in the 2007 map, is now very high again. In fact, the 2007 cyst abundance is about 30% higher than it was in 2004.

What does this suggest about the 2008 bloom season? Given the working hypothesis that the size of the mid-Maine cyst population is a predictor of regional bloom magnitude, this would argue that the 2008 bloom could be a significant one. We are not sufficiently confident to issue a firm forecast, but we have notified the management community and other scientists that there is a significant possibility of a large regional bloom in 2008, based on the high cyst abundance and the general relationships between cysts and cells observed in three of the four years for which we have data. This forecast is also consistent with an apparent upward trend in overall PSP toxicity in the western Gulf of Maine region that began in late 2003 and 2004, following a decade or more of low toxicity years (unpublished data).

Synoptic forecasts

On a short-term level, the *Alexandrium* population dynamics model is being used to provide weekly forecasts of bloom distribution and cell abundance. These runs are posted on a website and notices of the latest results are posted to the *Northeast PSP* listserver that has several hundred subscribers, including state and federal managers and others interested in news and information about PSP outbreaks in the region. In past years, these results have proven to be very valuable to managers; as otherwise, their information is predominantly limited to nearshore shellfish toxicity measurements. The model runs provide perspective on what might be occurring offshore, and upstream. See http://omgrhe.meas.ncsu.edu/Redtide/Redtide_07/ for an example of the announcement, and model run from one period in 2007.

Based on the general accuracy of these model simulations compared to cruise observations, and the value of the data to managers, NOAA National Ocean Service has plans to implement an operational forecasting system for *A. fundyense* in the Gulf of Maine. This would be based on the numerical models described above. These HAB Forecasts would be developed through a collaboration of NOAA with state, local, and federal managers and will be designed to assist the managers in their responsibilities in monitoring and management for public health and reduced economic impact. The proposed effort involves transitioning current research efforts to an operational capability. A specific approach is not predetermined by NOAA, but is based on the unique nature of the bloom ecology for a particular region. The approach will be driven by the results from regional and local HAB research project and the monitoring tools that have been developed.

The NOS HAB Forecast System for a region will incorporate a variety of environmental data types and HAB data, operating models, and analysing the data, much as is done for severe weather forecasts. Local analysts will be trained to interpret data in a way analogous to local weather forecasters, and will be co-located with local HAB experts (possibly at state environmental or fisheries offices). NOAA would provide data integration capabilities, support in developing training, transition of research models into operations, run these models operationally, and conduct skill assessment. National datasets, such as satellite imagery, would be processed and incorporated into the capability as appropriate. In addition, new monitoring tools (e.g. toxin probes, HAB sensors) would be evaluated for their utility and implemented, also as appropriate.

Unfortunately, the 2009 US federal budget is not yet decided, and the preliminary figures that have been circulated do not include funds for this operational forecasting effort. Unless changes are made during the budget process, it will be at least another year or two before the operational forecasting capability for *Alexandrium* blooms in the Gulf of Maine will be implemented.

A relevant development is that funding has been obtained to deploy the Environmental Sample Processor in the Gulf of Maine. This instrument developed by Chris Scholin at MBARI (Scholin *et al.*, in press) samples water and conducts all of the filtrations and other manipulations needed to enumerate *A. fundyense* cells in situ. The first few years of deployment will be developmental in nature, but the goal is to deploy this instrument and others to provide real-time data on *A. fundyense* cell abundance and distribution that can be assimilated into the numerical model runs and used to improve forecasts.

In summary, modelling efforts and our conceptual understanding of *A. fundyense* dynamics in the Gulf of Maine have progressed to the point where we can make both long- and short-term forecasts of bloom magnitude. It is also possible to provide near real-time maps of potential cell distributions along the coast, working from an annual cyst map from the preceding fall. Through data assimilation techniques, these latter forecasts could be made even more accurate once remote, automated cell detection of *A. fundyense* becomes a reality.

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7.1.13 Culturing and toxicity of North American *Dinophysis*

Don Anderson, Woods Hole Oceanographic Institute USA

For over one year, a culture of Dinophysis acuminata from North America has been maintained, using a ciliate and a cryptophyte as food, as described in recent publications by Korean workers (e.g. Park *et al.*, 2006). High cell densities (>12 000 cells ml-1) have been achieved, and sufficient biomass accumulated to allow toxin measurements. Cells were harvested and sent to collaborator Philipp Hess at the Irish Marine Institute, where they were analysed using liquid chromatography-mass spectrometry (LC-MS). Analyses were conducted using a Waters HPLC coupled to a Micromass Q-TOF Ultima (quadrupole – time-of-flight hybrid) equipped with a z-spray ESI source. Peaks having retention times of okadaic acid (OA) and dinophysis toxin 1 (DTX1) were detected in each sample. Peak identity was confirmed by comparison with OA and DTX1 standards. We now have unequivocal evidence of DTX1 in the Dinophysis sample based on: 1) Retention time in two-dimensional chromatography (<0.05 min shift); 2) Mass spectral comparison through ion ratios (<2.5 % difference); and 3) Mass spectral comparison through mass accuracy (<5 ppm). For OA, we have similar retention time evidence, but toxin concentrations were too low to allow spectral comparison through ion ratios or mass accuracy. The DTX1 concentration was approx. 0.14 pg/cell and the OA concentration 0.01 pg/cell. This is the first report of toxin production in cultured Dinophysis.

7.1.14 *Alexandrium fundyense* bloom dynamics in relation to total phytoplankton community and environmental variables in the Bay of Fundy

Jennifer L. Martin, Murielle M. LeGresley and Fred H. Page

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Phytoplankton abundance and a number of physical and chemical variables have been monitored at four locations in the Bay of Fundy at varying weekly to monthly intervals since 1987. Through the time-series, *Alexandrium fundyense*, the organism known to produce paralytic shellfish poisoning (PSP) toxins, has occurred annually at all sampling sites. The date at which *Alexandrium* first appears in samples varies from mid-April to late June with the mean and median date of the first appearance of *A. fundyense* varying by only a few days between stations. The date of the maximum concentration of *Alexandrium* cells varies by about 30 days between stations and years with the maximum cell counts occurring earliest at the inshore estuarine station (June 20–24) and latest at the offshore station (July 15–22). The annual maximum concentration of *Alexandrium fundyense* varies from 140 - 23 000 cells cells•L^{-1} in the estuarine station to 2.2 x 10^3 - 5.0 x 10^6 cells cells•L^{-1} at the offshore site. The mean and median concentrations increase from inshore to offshore during most years. The duration of *A. fundyense* blooms ranged from 42 to 205 days between sites and years. The mean (median) duration of the bloom is 114 (112) days. *A. fundyense* blooms vary interannually with some blooms consisting of 1 to 3 events per year.

During most years and at all stations, *A. fundyense* cells were minor components (<50%) of the total phytoplankton community. This phenomena seems to be apparent for most sampling days except during rare years such as 1989, 2003, 2004 and 2007 when *A. fundyense* concentrations exceeded 1.0 x 10^5 cells•L^{-1} and when it was observed at these high concentrations it comprised 50–80% of the total phytoplankton community. Organisms that coexisted varied between years and locations. At concentrations >5.0 x 10^5 cells•L^{-1} the total phytoplankton community was often >75% *A. fundyense*. When comparing *A. fundyense* concentrations to total dinoflagellate cell densities during periods where maximum cell densities were observed, *A. fundyense* was not a major component (60–95%) of the dinoflagellate community during most years unless there were >1.0 x 10^5 cells•L^{-1}. *A. fundyense* coexisted with *Guinardia delicatula, Eutreptia, Scrippsiella* and *Mesodinium* at most locations during 2007 and a suite of other organisms during other years.

Preliminary results suggest that during years with colder February and March temperatures the *A. fundyense* cell numbers are elevated. Additionally, concentrations exceeding >7.5 x 10^4 cells•L^{-1} appear to be linked to reduced nitrate levels. Results from the twenty year dataset indicate that high *A. fundyense* cell densities are required to make any linkages with environmental or physical factors.

8 Term of Reference f)

8.1 National reports

8.1.1 Canada National Report

Jennifer Martin, DFO Canada

West (Pacific) coast: Salmonid Mortalities

During 2007, mortalities to the salmonid industry occurred on Canada's west coast due to a number of different HAB species. During May, *Chrysochromulina* spp. and *Dictyocha speculum* occurred in Quatsino Sound at concentrations exceeding 5.0 x 10^5 cells L^{-1} and mortalities occurred around May 20.In late May/early June, *Noctilucas scintillans* was implicated in mortalities on the east side of Vancouver Island. *Heterosigma akashiwo* was linked to fish kills on the west side of Queen Charlotte Strait between July 31and August 2 and again on the east side of Quadra Island in mid-August when cell densities were 1.8 x 10^6 cells L^{-1}.

PSP

PSP toxins were detected at levels about regulatory levels along the coast of British Columbia with the highest level detected in sea mussels (1700 µg STXeq /100g) at Ballet Bay.

East (Atlantic) coast: Salmonid Mortalities:

No salmon mortalities associated with HABs were detected in 2007 along the Atlantic coast.

Marine toxins

PSP toxins were detected in *Mytilus edulis* (4368 µg STXeq /100g) and the hepatopancreas of lobster, *Homarus americanus*, (54 µg STXeq /100g) in Miramichi Bay, New Brunswick. In the Bay of Fundy *Alexandrium fundyense* concentrations reached 5.0×10^5 cells L^{-1} and were responsible for shellfish area closures when PSP levels increased to 640 µg STXeq /100g in soft-shell clams, *Mya arenaria*. Along the Nova Scotia shores of the Bay of Fundy toxin levels of 212 µg STXeq /100g were measured.

In southern Nova Scotia, PSP toxins were detected in shellfish and reached levels of 354 µg STXeq /100g. In the St Lawrence Estuary, there were closures and shellfish toxicities <200 µg STXeq /100g.

Newfoundland did not experience closures sue to marine toxins in 2007.

8.1.2 Finland National Report

Emil Vahtera (with contribution from Vivi Fleming-Lehtinen, Seija Hällfors, Seppo Kaitala) Finnish Institute of Marine Research

The main HAB –species in Finnish territorial waters in 2007 were the usual diazotrophic filamentous cyanobacteria *Aphanizomenon* sp. (currently considered non-toxic in the Baltic), *Nodularia spumigena* (hepatotoxic), and *Anabaena* spp. (neurotoxic). Besides theses species, haptophyceans of the genus *Chrysochromulina* (ichtyotoxic) were abundant and toxin producing dinoflagellates (*Dinophysis acuminata, D. norvegica, D. rotundata, Protoceratium reticulatum, Alexandrium ostenfeldii, Protoperidinium reticulatum, Gonyaulax spinifera*) were observed throughout the growth season generally in small numbers.

The cyanobacteria blooms were on average short and intense, and concentrated in the eastern parts of the Gulf of Finland. Surface accumulations occurred briefly in the Northern Baltic Proper and Bothnian Sea. The cyanobacteria surface accumulations were dominated by the non-toxic *Aphanizomenon* sp..

No toxic events were reported in 2007 of either humans or animals. Occasional beach closures due to accumulation of cyanobacteria surface-scums were reported.

The annual cyanobacteria bloom prognosis forecast considerable blooms in the Gulf of Finland and southern Baltic Proper. The prognosis is based on runs with three-dimensional coupled physical-biological models. The amount of available phosphate and weather condition largely govern the development of the blooms.

In 2007 phosphate concentrations were at normally high levels. However, cool and windy weather conditions prevailed during summer. These conditions are generally unfavourable for the development of surface scums and exceptionally large biomasses.

The first slight cyanobacteria blooms were observed in mid-June in the eastern Gulf of Finland, but they faded away within a few days time. At this stage *Aphanizomenon* sp. was found in samples from the western Gulf of Finland and became more common toward the end of June. Throughout July *Aphanizomenon* sp. formed frequent surface accumulations in the eastern and western Gulf of Finland. *Anabaena* spp. and *Nodularia spumigena* increased in July and remained common throughout August. An unexpected late bloom, consisting of *Aphanizomenon* sp. and *Microcystis flos-aquae*, occurred in the eastern Gulf of Finland in late October. In general, blooms were more for short periods more intense than the 10 year average in June and August, but as a whole the bloom period was shorter.

8.1.3 The Netherlands National Report

Marnix Poelman, IMARES, the Netherlands

The Netherlands experienced a quite HAB year for the fifth year in a row. No toxic events could be reported during 2007.

Phaeocystis sp. and other off shore data are only included for the first semester of 2007, since validation of the second semester have not taken place at this stage. *Phaeocystis sp.* was detected at Northwest, and Western situated off shore locations (135, 70 en 10 km from the Shore) during the period of January through June. The location 135 Northwest from the Shore showed a maximum of 1 mln cells/litre in April, where cell counts of 100.000 cells/litre were reported in May. The locations West (10 and 70 km off shore) showed maxima of 10 mln cells/litre in April and May, where levels dropped below 1 mln cells / litre in June. A maximum well below 100 mln cells/litre was detected at the 70 km off shore location.

At near shore locations cell counts for *Phaeocystis* were reported with maxima between 10–100 mln cells/litre in April and May, this occurred in the regions both North and South of the river Rhine. At near shore locations north of the Wadden Sea cell counts of 80–90 cells/litre were reported in May. Episodes of (shell)fish moralities or social effects were not reported.

Pseudo-nitzschia sp. has not resulted in any toxic episodes in 2007. *Pseudo-nitzschia* sp was reported at both off shore as near shore locations; however the threshold level of 100.000 cells/litre was only exceeded in May.

Dinophysis acuminata was detected in Lake Grevelingen in June 2007 at levels around 1.400–1.700 cells per litre; however no toxins were detected using the rat bioassays. LC-MS analyses revealed toxin levels ranging from 40–54 µg OA/kg were detected in the Western part of the lake (close to the connection to the North Sea).

D. acuminata was present in the Wadden Sea during the period September through October with ranges from 100–500 cells/litre. However, no toxins could be detected with use of rat bioassays. LC-MS analyses revealed presence of OA starting from Mid September onwards. Maximum levels of 70 µg OA/kg could be reported Mid October.

Yessotoxins were detected in mussels from the conditioning areas plots of the Wadden Sea (level ~70 µg/kg). The source of the mussels could not be traced, however it was suspected that the mussels are imported from other EU member states.

Spirolides (13-desmethyl spirolide C) was detected by LC-MS in a large amount of the shellfish in trace levels.

8.1.4 USA National Report

Don Anderson, Woods Hole Oceanographic Institute, USA

2007 was basically a "normal" HAB year for most regions of the U. S., with several noteworthy or exceptional events.

PSP. Similar to previous years, Maine, Massachusetts, California, and Washington all recorded PSP toxicity during 2007. For the first time since 2004, Oregon experienced PSP toxicity. In Maine, four humans were treated for PSP due to harvesting from a floating barrel covered in mussels. Three were hospitalized due to life-threatening exposures. It is suspected that the barrel originated in the Canadian Maritimes. Mussels on the barrel measured >18 000 μg STX/100 g. Washington State had a relatively "quiet" PSP year, but the massive bloom and high toxicity levels of 2006 carried over into 2007 resulting in a record 28 commercial geoduck tract closures in Central Puget Sound. California experienced unusual elevated PSP toxicity in winter – in both Santa Cruz and San Luis Obispo counties. As in 2006, there were no PSP events in Alaska in 2007.

A noteworthy finding in 2007 was the discovery of a widespread *A. fundyense* bloom in the offshore waters of Georges Bank in the Gulf of Maine. Cruises in May documented this bloom over most of the Bank with cell concentrations reaching 7500 cells/L. A subsequent cruise in June showed the bloom was still present although it had shifted to the south and west, with higher cell concentrations (12600 cells/L). Another cruise in October showed that some cells were still present, although over a smaller area and at lower concentrations (maximum – 2000 cells/L). Georges Bank is the site of significant hard surf clam, ocean quahog and sea scallop populations, all of which showed levels of PSP toxins. This area has been closed to the harvesting of surf clams, ocean quahogs and wholes scallops since 1989 so this toxicity had no direct impact on these fisheries. There are efforts underway to reopen these fisheries using a dockside testing protocol, so the potential for toxicity has major implications.

This was the first major *Alexandrium* survey of Georges Bank. In the past, opportunistic samples were collected that also revealed *Alexandrium* cells in these waters. Given the sporadic nature of these measurements, however, it is not clear whether a bloom of *Alexandrium* occurs every year on Georges Bank or whether 2007 was an unusual event. This area will thus be the focus of field efforts of the GOMTOX programme in 2008.

Since 2002, Florida placed a ban on puffer fish due to the saxitoxin poisoning events that happened that year. In 2007, for the second time, there were closures of shellfish beds for potential PFP (puffer fish poisoning). There were no human cases of PFP in 2007.

ASP. ASP toxins above quarantine were recorded in California and Washington. In California, domoic acid reached a maximum concentration of 610 ppm – a new record for that state for DA levels in mussels. Oregon experienced no new toxic *Pseudo-nitzschia* blooms in 2007, but saw a carry-over of toxicity in some shellfish from 2006 – although below quarantine levels. Washington's domoic acid concentrations were at relatively low levels.

NSP. *Karenia brevis* blooms occurred in three areas of Florida: the southwest, northwest and east coast. 42 manatee mortalities were associated with the southwest bloom and 13 'extra' sea turtle strandings were reported compared to the previous ten-year average. In addition, 29 sick or dead double-crested cormorants were admitted to local bird sanctuaries exhibiting neurologic symptoms of brevetoxicosis. The northwest bloom was associated with six 'extra' sea turtle strandings. The east coast of Florida had not experienced a bloom since 2002. Sixty-four 'extra' sea turtle strandings were reported and 9 manatee mortalities associated with that bloom.

CFP. Several outbreaks of ciguatera fish poisoning have been confirmed in consumers who ate fish harvested in the northern Gulf of Mexico, an area where this poisoning has not been reliably documented in the past. The fish linked to the illnesses were harvested near the Flower Garden Banks National Marine Sanctuary, a protected area of 56 square miles in the northwestern Gulf. An advisory has been issued to warn fishers and consumers of the CFP risk from this area (http://www.fda.gov/bbs/topics/NEWS/2008/NEW01790.html).

Brown tide. The south shore of Long Island, NY (South Shore Estuary) experienced a brown tide bloom for the first time since 2002. It was also the largest brown tide bloom in that area in over a decade, beginning in May and continuing through November. Peak cell densities exceeded millions per ml. The bloom was geographically expansive, covering 20 miles of Long Island's south shore bays from the Shinnecock Bay inlet to the Moriches Bay inlet. There were reports of eelgrass die offs and fish kills during the blooms.

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

Karlodinium. Chesapeake Bay experienced a *Karlodinium veneficum* bloom in 2007. It is suspected that this bloom caused a juvenile fish kill, but that is unconfirmed.

Other Events

A bloom of *Akashiwo sanguinea* occurred along Bolivar Peninsula, and Mustang Island, Texas resulting in a large fish kill (over 1 million gulf menhaden, etc.)

An extensive bloom of *Cochlodinium polykrikoides* occurred in the lower Chesapeake Bay sub-estuaries from the James River complex and also in the York River, Virginia. no confirmed mortalities or other negative impacts directly associated with this bloom were recorded.

8.1.5 French national report

Patrick Gentien, IFREMER, France

The French monitoring programme for phytoplankton and phycotoxins (REPHY) works according several levels: phytoplankton monitoring is performed on three levels corresponding to different objectives. The first level concerns 33 stations which are sampled all the year, once a fortnight: all phytoplankton species are identified and counted. The second level concerns 88 stations which are sampled all the year, once a week to once a month: only blooming and toxic species are counted. Supplementary measures are made on all stations of these two levels: temperature, salinity, turbidity, chlorophyll-a, oxygen, and nutrients. The third level concerns more than 100 stations, which are sampled only during toxic events, once a week: only toxic species are counted. The monitoring for toxins is performed on several hundreds of shellfish sampling stations, which concerns many different shellfish and many different growing methods.

The monitoring strategy is different according to the area status. In coastal production areas and natural banks, for PSP and ASP, the detection in water of toxic species (*Alexandrium* and *Pseudo-nitzschia*) above the alert threshold leads to the research of toxins in shellfish; but for lipophilic toxins (including DSP), systematic toxin research is performed in risk areas during risk periods once a week, and outside of these risk periods if presence of *Dinophysis*. On offshore natural banks, systematic toxin research is performed once a week during fishing period for the three families of toxins (lipophilic + PSP + ASP).

For lipophilic toxins, many toxic events occurred in several regions (mainly in Brittany and in Mediterranean lagoons), concerning several species of shellfish, such as mussels, oysters, donax, cockles, different sorts of clams, and -for one event- scallops. In most cases, these events were linked to the presence of *Dinophysis (*mainly *D. sacculus* or *acuminata)*, and LC/MS analysis showed the presence of OA + DTXs. No AZAs, no GYMs and no SPXs (or very few) were observed in 2007. But YTXs were observed in mussels in a Mediterranean lagoon with a maximum of 115 µg/kg, and PTXs were observed in mussels of another Mediterranean lagoon, with a maximum of 363 µg/kg.

There were only two PSP toxic events in 2007, in mussels of two Mediterranean lagoons. The maximum toxicity was 127 µg/100 g of equ. STX, and it was linked to the presence of *Alexandrium*: *A. minutum* in the first case, *A. tamarense* and/or *catenella* in the second case.

ASP toxic events were observed in many areas of Western and Southern Brittany, concerning mainly scallops *Pecten maximus* (only one event in *Donax*). The maximum toxicity observed was 183 µg/g of DA. The link of these toxic events with the presence of *Pseudo-nitzschia* could not be proved at each time, given the fact that phytoplankton monitoring is not always made on offshore areas.

A few cases of breathing difficulties had been reported in 2006 near Marseille,and had been linked to the presence of *Ostreopsis* cf. *ovata.* A reinforced monitoring of benthic species was then implemented in 2007 along two sites of the East Mediterranean French coast (Marseille and Toulon), with *Ostreopsis* counts on water samples, but also on macroalgae samples. In 2006, the maximum concentration in water had been 4500 cells per litre. In 2007, the maximum in water was 5300 cells/L in Marseille (August), 6200 in Toulon (September), and the maximum on macroalgae was 106 000 cells/g of macroalgae in Marseille (July), 85 400 in Toulon (September). No cases were reported in 2007.

8.1.6 Germany National Report

Allan Cembella AWI, Germany

During the period 2007-early 2008 few harmful algal events were reported from the North Sea or Wadden Sea coast of Germany, including waters adjacent to Helgoland, Bremen State, Lower Saxony and Schleswig-Holstein. The only major HAB-associated event was the mass mortality of herring along the Wadden Sea coast adjacent to the island Sylt near the Danish border. A dense nearshore bloom of mucilaginous algae belonging to the prymnesiophyte *Phaeocystis globosa* (colonial form) was observed in the first week of June 2007. The bloom was virtually monospecific with a west-east diameter of 2 km and a north-south extent of 5 km. Locals on Sylt reported a severe water discoloration ("brown soup") and the smell of decaying algae. Sea conditions during the peak of the bloom were characterized by high water temperature and little wind, accompanied by a slow current-driven northwards drift. A strong east wind the carried the bloom to the beaches of Sylt along with millions of dead herring at the beginning of the second week of June 2008. The *Phaeocystis* patch eventually dispersed during the following week. At the aggregative centre of the bloom there was evidence of dying cells at this time. Maximal chlorophyll a levels reached 20 µg/L in late May.

The putative cause of the mass mortality of herring (estimated at millions of fish) was low oxygen in the centre of the bloom: 17% oxygen saturation in patch nucleus vs. 100 – 110% surrounding the bloom and >120% away from the patch. Herring typically avoid such *Phaeocysis* patches but juvenile and young adult herring are preyed upon by mackerel. The arrival of this predator led to the hypothesis by fisheries ecologists this may have been a panic avoidance response by herring to predation by entering bloom as a refuge.

For Lower Saxony no measurable phycotoxins were found in blue mussels (*Mytilus edulis*). Maximal abundances of potentially toxic species were found during the monitoring period July to October. These taxa include: *Dinophysis* spp. (400 cells/L), *Chrysochromulina* spp. (5000 cells/L) and *Pseudo-nitzschia* spp. (68 200 cells/L).

No major harmful events were associated with the typical appearance of cyanobacterial blooms on the German Baltic coast including transitional waters. Transient beach-fouling, an inconvenience to bathers and beach-goers, did occur along the margins of Mecklenburg-Western Pomerania in 2007.

8.1.7 Ireland National Report

Joe Silke / Dave Clarke, Marine Institute Ireland

Amnesic Shellfish Poisoning

No particularly high or dense blooms of *Pseudo-nitzschia sp.* were observed nationally for 2007, where the highest peak of *Pseudo-nitzschia delicatissma* group were observed in July at <700 000 cells/litre for a 1 week period. For *Pseudo-nitzschia seriata* group, a one off peak in March of 2000 000 cells/litre were observed, typically levels observed throughout the year were <100 000 cells/litre.

Typically, low levels of Domoic and Epi-Domoic acid were observed in 460 scallop (*P. maximus*) tissues analysed (mainly adductor and gonad tissues), where only a small percentage of Gonad tissues analysed were found to be above the regulatory limit of 20µg/g. In 114 samples of other shellfish species (mussels, oysters, clams, and razor clams) analysed for ASP, all results were <LOQ.

Paralytic Shellfish Poisoning

Low levels of the causative PSP toxin producer, *Alexandrium spp.* were observed in 2007, where the highest level observed was in July 07 at 800 cells/litre. These low levels were correspondingly observed in 170 shellfish samples (clams, cockles, mussels and oysters) analysed by the AOAC method for PSP Bioassay, where all samples were well below the regulatory limit. The highest PSP concentration observed was 39µg/STXdiHCL100g^{-1}.

Diarrhetic Shellfish Poisoning

Low levels of the DSP causative organisms, *Dinophysis acuta* and *Dinophysis acuminata* were observed in 2007, where the highest levels observed was 880 cells/litre of *Dinophysis acuminata* in July 07. This resulted in very low levels of DSP toxins

(Okadaic Acid (OA) equivalents) in shellfish samples analysed and were all below the regulatory limits. The highest level observed was 0.08µg/g^{-1} OA equivalents in mussels (*M. edulis*).

Azaspiracid Shellfish Poisoning

Numerous closures were enforced due to the AZA contamination of mussels above the regulatory limit during 2007. Due to Azaspiracid contamination of mussels (mainly in the SW Ireland) toward the end of 2006 above the regulatory limit, these levels were slow to detoxify from the affected mussels, hence there was carry-over of AZA levels into early 2007 above the regulatory limit (highest AZA level observed was 0.72µg/g^{-1} AZA equivalents) in Southwest Ireland in January 07. These levels were observed to decrease to levels below the regulatory limit by April 2007.

AZA levels remained low until June and July where in SW Ireland, AZA levels were above the regulatory limit in a small number of samples (highest AZA level observed was 0.4µg/g^{-1} AZA equivalents). These levels decreased during August, but were observed to increase in SW Ireland in September and October 2007 (highest AZA level observed was 1.4µg/g^{-1} AZA equivalents).

Overall the number of site closures due to toxin presence above the regulatory limit was much lower than observed in 2006 and 2007, and was only confined to the presence of AZA's.

8.1.8 United Kingdom National Report

Prepared by Eileen Bresnan FRS Scotland and co-authors from UK

8.1.8.1 Scotland

In contrast to sites along the south coast of the UK, the spring bloom in Scotland was very strong during 2007. Blooms of *Pseudo-nitzschia* spp. were observed in early April 2007, Dense *Pseudo-nitzschia* concentrations were mostly absent in southwest Scotland and south Argyll and Bute. *Pseudo-nitzschia* blooms were widespread during July and the largest bloom recorded in 2007 was from southwest Shetland in early July (2.5 million cells.l^{-1}). Blooms of *Pseudo-nitzschia spp.* associated with ASP toxins in shellfish were observed in July 2007. A large bloom of *Pseudo-nitzschia* (365 000 cells.l^{-1}) in the Western Isles in late August resulted in ASP toxicity above permitted levels in shellfish. By October most *Pseudo-nitzschia* blooms had disappeared, persisting only around Orkney and on the east coast.

Dinophysis spp.

Dinophysis spp. was present above threshold levels (100 cells.l^{-1}) from late April 2007 onwards, initially appearing most frequently in Argyll and Bute, northwest Scotland and Orkney. Blooms appeared in the Shetland Islands and on the east coast in June, with the largest blooms (up to 5300 cells.l^{-1}) occurring around Shetland in July. *Dinophysis acuminate* was routinely observed while *D. acuta* was absent. These were associated with extended periods of DSP toxicity in Shetland shellfish recorded between June and August. Most other 2007 DSP toxic events occurred in Argyll and Bute in May and June. The largest *Dinophysis* bloom recorded in 2007 was from Elie, Fife in July (58 000 cells.l^{-1}).

Alexandrium spp.

Alexandrium spp. was observed much more frequently during April 2007 compared with the same time the previous year, with a large bloom present around Orkney from mid April to mid May (up to 6060 cells.l^{-1}). It was generally less widespread at all phytoplankton monitoring sites by September 2007, compared with September 2006. The largest bloom recorded in 2007 was from southwest Shetland in late July (9180 cells.l^{-1}). Although PSP toxicity was detected below permitted levels at most monitoring sites during 2007, there was some toxicity associated with the presence of *Alexandrium* on the west coast during April, May and June. *Alexandrium* occurring around Shetland in April, July, August and September was also associated with toxic events.

Prorocentrum minimum

Prorocentrum minimum was frequently observed at Scottish monitoring sites during 2007, with blooms occurring from mid April to early September. Similar to 2006, the largest blooms occurred around Shetland, with a maximum recorded level of >2.4 million cells.l^{-1} observed in late May. Blooms were also noted around Orkney and Sutherland (north and east coast) at this time, but it was only recorded in small numbers at other monitoring sites. The toxicity of this species in Scottish waters in unknown.

Other species

Protoceratium reticulatum, Lingulodinium polyedrum and Protoperidinium crassipes were infrequently observed in Scottish waters during 2007 and did not cause problems. *Prorocentrum lima* was observed in relatively small numbers during 2007, and frequently occurred at the same sites. Maximum cell density reached 560 cells.l^{-1} in Shetland in mid June.

Karenia mikimotoi

Compared with 2006, large blooms of *Karenia mikimotoi* were not observed during 2007 and recorded cell densities were mostly below 10 000 cells.l^{-1}. Cell densities were at there greatest in July, August and September, with a maximum concentration of 103 156 cells.l^{-1} recorded on the west coast of the Isle of Mull in mid August.

8.1.8.2 England and Wales

Alexandrium spp. were much less widespread than in 2006, being recorded from just 24 of the 59 sampled areas. They occurred in 141 of the 1125 samples collected. Highest concentrations were found in the River Yealm (Devon) at concentrations of 2 million cells/litre in mid July, where it persisted from March to the end of October. *Alexandrium spp*. were found much less frequently than normal in samples collected from four sites in the Fal Estuary, and were not found at all in samples collected from the site at Penryn. Samples from Weymouth inner harbour also contained *Alexandrium spp* much less frequently and at lower concentrations than previously recorded, with a maximum concentration of 86 000 cells/litre occurring in August. PSP toxins were found on only three occasions in 2007, all at Holy Island, Northumberland, in mussel flesh. *Alexandrium spp* did not coincided with PSP toxins being found on any occasion this year.

Dinophysis spp. (DSP) were found in 9 sampling areas, but often at low concentrations. Highest concentrations (800 cells/litre) were found at Holy Island at the end of June. Unusually, only low concentrations were found regularly in samples collected offshore at Blyth, Northumberland from April to August. Elsewhere, *Dinophysis spp.* only occurred infrequently and always at low concentrations. *Prorocentrum lima* (DSP) were found on thirteen occasions. Peak concentrations of 240 cells/litre were found in water samples collected from the Fleet, Weymouth, Dorset at the end of July and in early October. DSP toxins were recorded on nine occasions in samples of mussels from the Fal Estuary (1) and Holy Island (4), oysters from Holy Island (1), Salcombe (1) and the Hamble Estuary (1), and once from a sample of cockles collected from Three Rivers, South Wales.

Pseudonitzchia spp. (ASP) were found in most of the sampled areas in 2007 but were much less widespread and persistent than in 2006. They also occurred at much lower concentrations than had been seen in 2006. They breached the 'investigative' level (50 000 cells/litre) on 20 occasions and the action level (150 000 cells/litre) 4 times during summer of 2006. ASP toxins were found in shellfish flesh samples from nine harvesting areas in 2007 but always well below action level (20.0μg/g). The highest concentration (12.0μg/g) was found in a sample of cockles taken from Three Rivers at the end of June.

8.1.8.3 Northern Ireland

In 2007, thirty five sites were sampled routinely on a fortnightly basis from N. Ireland sea loughs.

Alexandrium spp. were recorded in 1.6% of samples reaching a maximum of 120 cells l^{-1} in a sample taken from Belfast Lough in June. No PSP toxins were detected.

Dinophysis spp. were present in water samples from April to October reaching a maximum abundance of 200 cells l^{-1} in a Belfast Lough sample in July. The most abundant species was *D. acuminata* with only small numbers of *D. acuta*, *D. rotundata* and *D. fortii* counted. Cells of *Prorocentrum lima* were recorded in 2.1% of all samples with a maximum abundance of 140 cells l^{-1}. One site in Carlingford Lough recorded *P. lima* cells in 18.1% of samples. Diarrhetic shellfish toxins were detected on six occasions during 2007 all in oyster samples. These positives were not linked with the presence of any known microalgal species in the water samples.

Pseudo-nitzschia spp. were present in 64.9% of samples reaching a maximum concentration of 198 528 cells l^{-1} in a Carlingford Lough sample. Toxicity, however, was confined to samples of scallops (*Pecten maximus*) from Strangford Lough. Domoic acid levels reached a peak of 30.95 μg/g whole flesh.

No major phytoplankton blooms of harmful or other microalgal species were recorded during the year.

8.1.9 Norway National Report

Submitted by Einar Dahl, Norway

This report summarizes information from weekly monitoring of toxic algae and algal toxins in mussels at about 50 stations from the Swedish to the Russian border.

<u>Yessotoxins</u>

At one monitoring station in west-Norway yessotoxins (YTX) above action level (1000 ug/kg) were detected in the end of the year.

Diarrhetic Shellfish Poisoning (DSP)

In total there were fewer problems than average. There were most problems in northern Norway, as we have seen since 2003.

Along the Skagerrak coast DSP-toxins accumulated slightly above action level (160 ug/kg) in July and August, but declined again, and remained below the action level the rest of the 2006, which was rather unusual. Along the west-coast DSP-toxins were most common in the inner part of the large fjords and less common along the outer coast. In northern Norway DSP-toxins were present already in March, when the monitoring started. This was most probably due to toxin-remnants from the autumn-situation. After a period with no DSP-toxins from about middle of April to August, DSP-toxin again accumulated in mussels above action levels in northern Norway and remained high throughout the year.

Paralytic Shellfish Poisoning (PSP)

PSP-toxins were recorded all along the coast, but not at all stations. The levels were, however, often low just above the action level (400 ug/kg) and only slightly above the quarantine level (800ug/kg), and the problems lasted generally 1–2 months, seldom more. However, at some few stations both in south, along the west-coast and in the north levels well above quarantine levels were recorded.

Other toxins
ASP, AZA and PTX were not detected above action levels in Norway in 2007.

8.1.10 Sweden National Report

The Skagerrak and the Kattegat

No major harmful algal blooms occurred in the area in 2007. In spring *Verrucophora farcimen*, a species harmful to fish, was observed after the diatom spring bloom and also in autumn. No harmful effects were noted in Swedish waters. *Dinophysis* spp., a dinoflagellate genus with representatives producing Diarrhetic Shellfish Toxins (DST) was observed during most of the year but in abundances lower than usual. DST above the regulatory limit of 160 mg per 100 g of mussel meat occurred in blue mussels at a few locations along the Swedish West coast in January-February and in July-August. *Alexandrium* spp, occurred in several locations during the year. A closure of shellfish harvesting in one area due to an indication of Paralytic Shellfish Toxin occurred in early September. New phytoplankton species were observed in the area. The diatom *Chaetoceros concavicornis*, known from the West coast of North America, was abundant from September to the end of the year. Its spines have negative effects on the gills of fish but no harmful effects were noted in Sweden. The dinoflagellate *Peridinium quenquecorne* was common in the area in early summer. It is suspected that a case of swimmers rash near Gothenburg in June was connected to high abundance of the organism.

The Baltic

No major harmful algal blooms occurred in the area in 2007. This was a year with only minor surface accumulations of cyanobacteria. This is probably due to meteorological conditions during summer. Clouds made observations from satellite less frequent than usual and strong winds probably mixed the bloom deeper in the water column. The toxin producing species *Nodularia spumigena* was less common than usual. Another cyanobacterium, *Aphanizomenon* sp. was common. Surface accumulations were observed in June and ended in August. However, accumulations of cyanobacteria were also noted in autumn in the Archipelago of Stockholm and also along the coast of the Bothnian Bay. The potentially harmful flagellate *Chrysochromulina* cf. *polylepis* was abundant at several off shore locations in the Baltic proper during autumn. No harmful effects were observed. A ctenophore, *Mnemiopsis leidyi*, was observed in the whole Baltic South of the Quark. This is an introduced gelatinous zooplankton species and there is concern about effects on the ecosystem in the Baltic. The same species was introduced to the Black Sea about twenty years ago and it is suspected to have effects on e.g. the fish populations in the Black Sea.

8.1.11 Spain National Report

ANDALUCIA[1] (ATLANTIC COAST)

DSP. Different species of *Dinophysis* led to shellfish harvesting closures in the area. *Dinophysis* cf *acuminata* bloomed (max. 5.560 cell L^{-1}) in early February off eastern Huelva. Intermittent peaks (more intense in the eastern part of the Gulf) of this species are common between April and August (max. 3.280 cell L^{-1}). *Dinophysis caudata* peaked between May and July (max. 2.760 cell L^{-1}). *Dinophysis acuta* was found from early June until the end of August. Cell maxima (7.360 cell L^{-1}) were reached in July.

ASP. Events caused by *Pseudo-nitzschia* cf *australis* (max. 172272 cell $L^{-}1$) in April.

ANDALUCIA[1] (MEDITERRANEAN COAST)

ASP. The main bloom of *Pseudo-nitzschia* spp (mainly *P. australis*) off Cádiz lasted from late March to early April and contaminated the open sea resources in this province (max. 394.944 cell L^{-1} in Algeciras Bay). Blooms of lower intensity (150000 cell L^{-1}), and a new toxin outbreak in June were reported off Málaga.

PSP. *Gymnodinium catenatum* cells observed in January were remains of the previous year event. During 2006, this species proliferated in November-December. Maximum concentrations were detected in La Línea (3.360 cell L^{-1}) (Cadiz) and later (January 2008) in Fuengirola (Málaga) (5200 cell L^{-1}). The autumn outbreak affected all shellfish production areas in the coasts of Málaga.

[1] Data provided by LCCRRPP (Fisheries and Agriculture Department, Junta de Andalucía).

BASQUE COUNTRY[2]

Analysis of ASP, DSP and PSP toxins have been carried out in wild mussels and oysters from three estuaries (Butroi, Oka and Bidasoa), twice a yea (summer and autumn). Phytoplankton was analysed from 42 stations (coastal waters and estuaries) in samples collected during four cruises between February and November. Despite the occurrence of potentially toxic species, toxins were below detection in wild shellfish.

Potentially toxic genera/species detected included:

Alexandrium spp., found in estuarine locations (max. $16 \cdot 10^3$ cell L^{-1} that represented 16% of the population); *Dinophysis acuminata* and *D. caudata* in coastal waters (<10% of the samples) with a maximum abundance of $2 \cdot 10^3$ cell L^{-1}; *Gymnodium* spp., in coastal and estuarine waters (35 and 12% of the samples, respectively, max. abundance $16 \cdot 10^3$ cell L^{-1}); *Prorocentrum minimum*, in 13% of the coastal samples and 2% of the estuarine samples, with a maximum abundance of $2 \cdot 10^4$ cell L^{-1}; cf. *Pfiesteria,* detected in the Oiartzun estuary with a maximum concentration of $3 \cdot 10^4$ cell L^{-1}.

Potentially toxic diatoms showed high concentrations in summer. *Pseudo-nitzschia* spp. was very frequent in coastal waters (72% of the samples), where it reached up to $1.5 \cdot 10^6$ cell L^{-1}. *Pseudo-nitzschia* spp. was also present in 27% of the estuarine samples, with a maximum concentration of $2.4 \cdot 10^5$ cell L^{-1} (Nervión estuary). *Pseudo-nitzschia galaxiae* was detected in a few coastal and estuarine stations; its highest abundance was $2 \cdot 10^4$ cell L^{-1} (Oka estuary).

Potential fish-killer were also detected: (i) *Chrysochromulina* spp., in 87% of the coastal samples and 37% of the estuarine samples, with a maximum abundance of $1.3 \cdot 10^5$ cell L^{-1}.

[2] Data provided by AZTI-SIO and University of the Basque Country, funded by the Basque Government.

CATALONIA[3]

PSP. *Alexandrium minutum* bloomed (1.4 10^5 cells/L) in April in open waters off Vilanova, and toxins (up to 205 µg eq STX/100g) found in clams from nearby harvesting areas. High concentrations of the same species (up to $2.3 \cdot 10^6$ cell L^{-1}) in Arenys and Estartit harbours were not associated with toxicity in shellfish nearby. *A. catenella* bloomed (3 10^4 cell L^{-1}) off Tarragona in July, and low toxin levels (83 µg eq STX/100g) were found in mussels. *A. minutum* and *A., andersoni* were present among other species in Alfacs Bay in March (max. 1.2 10^4 cell L^{-1}).

ASP. *Pseudo-nitzschia* spp. were present in high densities (2 10^6 cell L^{-1}) in Alfacs and Fangar bays, but presence of domoic acid in shellfish was not reported.

DSP. In Alfacs Bay, *Dinophysis sacculus* (max. 1.1 10^3 cell L^{-1}) was associated with the detection of lipophilic toxins in mussels (mouse bioassay). *Protoceratium reticulatum* was present in low densities (2 10^3 cell L^{-1}) in April, and yessotoxins identified (HPLC and LC/MS-MS) below regulatory levels.

Fish-killers. *Karlodinium* spp., occurred in high densities ($1.5 \cdot 10^6$ cell L^{-1}) in Alfacs Bay in June. This bloom was associated with mortalities of razor clams (*Solen marginatus*), bottom fish and mussel seed.

Skin irritations *in sunbathers at beaches in L'Escala (northern coast, Costa Brava) coincided with high concentrations of Ostreopsis spp. (up to $8 \cdot 10^6$ cell L^{-1}).*

[3] Data provided by IRTA (Agriculture Department) and ICM-CSIC (Environmental Department), both with funds from the Generalitat de Cataluña.

GALICIA[4]

ASP. During 2007, there were no harvesting closures of raft-mussels caused by *Pseudo-nitzschia* spp. Eventually, infaunal resources were affected, during the first half of September, in some of the Rías Altas (Corme-Laxe and Camariñas)

Scallops (Pecten maximus) contained DA above regulatory levels all year-round. Restricted harvesting after evisceration (according to Directive 2002/226/EC) proceeded only in the large Ría de Arousa at the end of the year.

DSP. Closures due to *Dinophysis acuminata* (max. concentration of 6640 cell L^{-1} in Portonovo, Ria de Pontevedra) affected the southern rías of Pontevedra, Muros, outer parts of Vigo and Arousa and the Estuary of Baiona. Closures lasted longer in the northern rías, where closures lasted until the end of September in Corme and from mid-August to mid-September in Ares-Betanzos. An additional autumn peak (2 weeks harvesting closures) occurred in October in Ría de Pontevedra (max. 600 cell L^{-1}).

PSP. *Alexandrium minutum* and infaunal shellfish quarantines from late June to early July in Ría de Camariñas (Galician Finisterre), and in both, infaunal and raft-mussels during September in Ría de Ares-Betanzos (max. 383130 cell L^{-1}).

Gymnodinium catenatum occurred in very moderate concentration (max. 360 cell L^{-1}) that led to one week shellfish closures in the outer parts of Ría de Vigo.

[4] Data provided by INTECMAR (Fisheries Department of the Xunta de Galicia)

MURCIA[5]

This Autonomous Community, that experienced several outbreaks of toxic aerosol and skin irritations during summer 2006, established a new programme of control for this new health risk. During 2007, there were no reports of sunbathers and others affected by this syndrome.

[5] Data provided by the Technical University of Cartagena (UPCT), funded by the Generalitat Valenciana.

8.1.12 Denmark National Report

Shellfish

Algal toxins (DSP, PSP and ASP) were not observed above the regulatory limits in shellfish during 2007 in Danish waters. However, DSP toxins were detected below regulatory limits.

Exceptional blooms of *Dinophysis acuminata* and *D. acuta* were observed in the Limfjord, figure 1. The increased concentration levels of the rather slow growing *Dinophysis* species might be the result of a major decrease in the population of mussels (*Mytilus edulis*) in the Limfjord due to major incidents of hypoxia during the previous years. The rather small populations of mussels will result in a decrease in the grazing pressure from the mussels on the *Dinophysis* species, allowing *Dinophysis* species to grow to higher concentrations than observed in the previous years !

As a response to the situation, single cell toxicity in combination with toxin content in mussel flesh were implemented into the ongoing risk analysis. This allowed the shell fishers/shellfish growers to harvest in situations where the regulatory limits of the algal concentrations were exceeded, but where the toxicity of the cells were low and the toxin content in the mussel flesh were less than 50% of the regulatory level, Jørgensen and Andersen 2007.

Marine fish cultureA bloom of the flagellate, previously/formerly known as *Chattonella* cf. *verruculosa* during a four week period in March-April in the southern part of the Kattegat in fish kills in marine fish farms, figure 1. Both overwintering fish and newly released fish were killed. At one site approx. 50% of the fish (overwintering fish) died within the first days of the bloom. The fish surviving the first kill, also survived the following period with bloom concentrations which lasted a couple of weeks! At another site 100% mortality was observed including a combination of overwintering and newly released fish.

A total loss of approx. 70 tons of fish were observed during the bloom. Economical losses as a result of the *Chattonella* bloom also included loss of production due to delayed release of fish in the marine fish farms.

HAB moitoring was not initiated untill the bloom was detected in the form of increase fish mortality/dead fish. From the available data, it is suggested that the fish kills occurred at concentrations of "*Chattonella*" of approx. 05–1 mill. cells/l. Observations from a similar bloom during spring of 2008, indicate that the fish will react to concentrations of approx. 04–05 mill. cells/l but that now increase in mortality is observed at this concentration level. The max. concentration observed was 10 mill. cells/l.

Recreative use of coastal waters

No algal problems in relation to the recreational use of Danish coastal waters were observed during 2007. No *Nodularia* or other cyanobacteria were detected in bloom concentrations, propably as a response to the windy and cold danish summer !

Reference

Jørgensen, K., and Andersen P. 2007. Relation between the concentration of *Dinophysis acuminata* and diarrhoeic shellfish poisoning toxins in blue mussels (*Mytilus edlis*) during a toxic episode in the Limfjord (Denmark), 2006. J. Shellfish Res. Vol. 26, No. 4, 1081–1087.

9 Term of Reference g)

9.1 Review the progress on intersessional updating and inputting data in the IOC-ICES-PICES HAEDAT database and developments made towards developing an integrated system.

HAEDAT

The on-line version of HAEDAT was ready for on-line input in October 2007, however around December, reappearances of bugs were reported by users. Early March 08 the programmer was given two weeks to solve the existed problems. A new programmer is now in place, since the previous attempt was not successful. Due to the development of the on-line database and the development problems, data input has not happened since 2004. There is quite a challenge for everyone now to include the data from 2004 and onwards. The reports can now be submitted in the old format, but it is preferable that submission is done after the online registration is complete.

It is estimated that the system is up and running by Easter. HAEDAT is an extremely valuable dataset with great potential to becoming extensively utilized. There are developments on the technical end that allow users to publish their data and query it through the Internet. This system was demonstrated to WGHABD in 2006 in a near complete version. Unfortunately it has taken a long time to complete and end user testing has revealed a number of bugs. It is requested that the finished version be presented in 2009.

HAIS

Harmful Algae Information System (HAIS) is a novel system which will be developed. The information will include current taxonomy names of Harmful Algae, biogeography of Harmful species, occurrence of HAB events (HAEDAT), etc. HAIS has potential to become a unique product. HAIS is developed within the joint framework of the IOC International Ocean Data Exchange Programme (IODE) and IPHAB. A meeting was held in Oostende in January 2008 about developing HAIS. Here it was discussion who the collaborating partners are, and what the related responsibilities in the development will be. At this moment a funding search is being performed, however the initial activities (workshop and HAEDAT, HABMAP, MONDAT linkage) are funded by Flanders and US NOAA, and are planned for 2008.

The existing data sources which are proposed to be incorporated in HAIS are:

- Harmful Algal events, IOC ICES PICES (HAEDAT)
- Biogeography with ISSHA (HABMAP) and OBIS
- Taxonomy with WoRMS and EoL (Tax. Reference list)
- References with AFSA and OceanDoc
- Expert Directory with IODE (OceanExpert)
- Monitoring and management design with ICES (MONDAT)

The system was agreed by WGHABD to be an ambitious development, but really functional. Proper back up structures will be guaranteed, and easy access will be guaranteed. Data should be validated on a yearly basis in order to guarantee their quality. HAIS should be disseminated as the leading HAB information system, since the involvement, trust and achievements in the component databases, so far have been great.

The monitoring plans etc. will be incorporated in the MONDAT databases. These are however not updated since 2001. However, the adaptation of the European Regulations, have resulted in changes since then. Therefore it is important to have access to the different methodologies used, the implications of results, and function of phytoplankton monitoring. At this moment not everyone has a proper view of the legislation and there is potential for misunderstanding, this may even lead to difference in interpretation of test results. The development of a well maintained and updated information system was endorsed by the working group as a useful resource to inform and prevent such difficulties arising.

10 Term of Reference h)

10.1 Review the structure and composition of the decadal HAE maps for the ICES region with special reference to the linkage between the decadal maps and the HAEDAT database and the need for new maps for specific algal species

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. IFREMER have facilitated this activity in cooperation with IOC and their ongoing support this work was appreciated and further discussions on this ongoing development were held. As reported in the previous ToR the building of the HAEDAT database necessary to deliver the information to the maps in an automated fashion and this link is still under development. The linkage of the maps on the IFREMER website (http://www.ifremer.fr/envlit/documentation/dossiers/ciem/aindex.htm) to the HAEDAT is dependant on the completion of the HAEDAT database and population of this database to bring it up to date.

However, since there is still some work necessary on HAE-DAT because is not yet established enough to stand alone. A critical step forward is to make HAE-DAT operational with completion of all input from regions/countries including data from 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.

The data shown on the now out of date IFREMER maps used to be updated by the delegates at (WGHABD), however the initiative to update electronically has resulted in a gap since 2004 in their refreshing and this was identified by the group as an unfortunate lapse.

Once this has been rectified the data provided by representatives of the different member countries of the WGHABD, and through coordination and cartographic services are ensured by the French Institute for Marine Research and Exploitation (IFREMER) the maps will be updated annually and a dynamic web access will provide new means of customizing output.

The information available differs greatly depending on the event or country concerned. As monitoring intensity, the number of monitoring stations, and the number of samplings also differ considerably, there is no direct relation between recorded and actual harmful algal events (e.g. of toxicity) in a given region. Moreover, areas with many recorded events may benefit from an efficient monitoring and management programme and have very few problems and a low risk of intoxication, whereas infrequent events in other areas may cause severe problems and represent significant health risks.

11 Term of Reference i)

11.1 Review intersessional work to generate a website to electronically archive past reports of the WGHABD and to facilitate intersessional work carried out by the group

The IOC Harmful Algal Bloom website (http://www.ioc-unesco.org/hab/index.php?option=com_contentandtask=viewandid=11andItemid=0) was demonstrated. Some of the tasks identified in this term of reference are already in place on this site. On this website are working group reports of WGHABD dating back to 1999, and links to ICES and IFREMER. In addition there are background HABs information and up to date news and activities.

The IOC Project Coordinator at the IOC-Science and Communication Centre on Harmful Algae (Henrik Enevoldsen) suggested that this website could be a suitable location to service the intersessional activities of WGHABD and facilitate the access and publication of material between members of the group. The further development of this website will be discussed between the WGHABD Chair and the IOC Project Coordinator intersessionally.

12 Term of Reference j)

12.1 To investigate and discuss the possibility of comparative studies that will use mesocosms to explore the dynamics of HABs subject to eutrophic pressures

12.1.1 Using Mesocosms to Research HABs in Comparative Studies- Can They Mimic "real life"?

A presentation on Mesocosms and their applicability to mimicking "real life" by Edna Graneli Dept. Marine Sciences, Kalmar University, Sweden and Bengt Karlson of the Swedish Meteorological and Hydrological Institute Research and Development, Oceanography, Sweden, discussed the various facilities including the Mesocosms at Rhode Island, Bergen, Netherlands and in Sweden. Experiments carried out in The Swedish Kalmar centre has been designed for large-scale experimental work on primary and secondary production and consists in enclosed water masses with volumes between 2 and 30 m^3 . Some experiments carried out here included effect of nutrient concentrations and ratios on summer HABs from Baltic Sea. In these experiments the highest Nodularia concentrations were seen in the P deficient treatments (Figure 9).

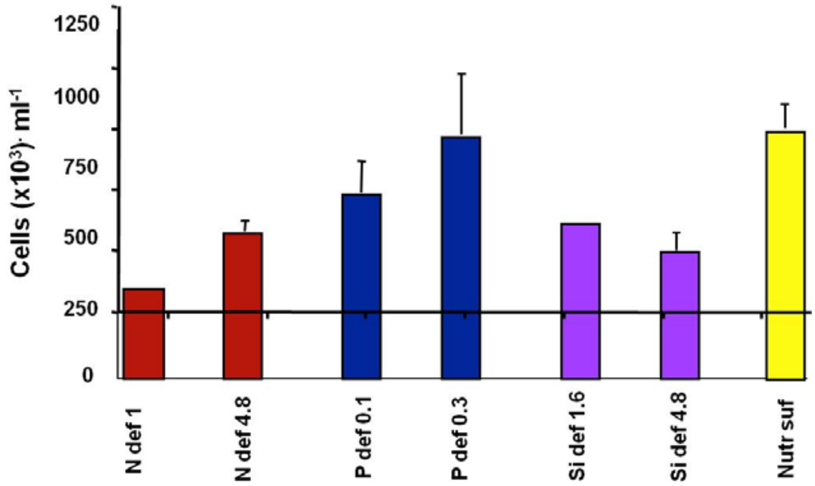

Figure 9. Highest *N. Spumigena* densities in the P- deficient treatments.

And also toxicity per cell was highest in the P-deficient treatment (not strange as nodularin contains nitrogen). But this also show that the algae under N-deficiency, although able to use N from air, will not use this N in increasing their toxicity. But more N input to the Baltic may potentially increase toxicity in the blooms. Further experiments in the mesocosms showed the N addition alone increase algal biomass (NO3), N and iron further increase, DOM (humic acids from rivers), even more but mostly increases were cyanobacteria and flagellates, adding iron and DOM made *Anabaena* increase in such amounts that the flocs could be seen in the water. Other experiments showed that zooplanktivorous fish (*Sprattus spratus*: herring, which has increased several folds in the Baltic due to overfishing of cod) was able to keep the zooplankton at normal densities and also decreased copepods numbers added in 6 times their normal densities to the mesocosm cylinders. Copeds in high densities were able to keep the diatoms down but not flagellates and dinoflagellates in the NP enriched cylinders. *Karenia mikimotoi* could not be kept under control under any copepod density. In top down experiments adding jelly fish could decrease the grazing pressure on the dominating diatom by heavy predation on the copepods. For *Karenia* it did not matter with or without jelly fish this species was not suffering from heavy grazing in any of the copepod densities.

12.1.2 Mesocosm Discussion

The WGHABD group recognized that it does not have the critical mass of expertise to consider the utility or applicability of mesocosms in great detail. They group questioned why the mesocosm concept is being addressed solely by the GEOHAB Core Project on HABs and Eutrophication and considered that the white papers and analyses being proposed below could be prepared for GEOHAB as a whole, not just one of its core projects.

The WGHABD agrees with the GEOHAB SSC on the need for a thorough assessment of the capabilities and limitations of mesocosms and recommends to IOC that

GEOHAB prepare a white paper on the capabilities and limitations of mesocosms, and in particular, emphasize the criteria and logistical issues that need to be addressed to generative data of value to those planning comparative studies. The assessment should also include possible strategies for the incorporation of mesocosms into HAB research. Some of the issues of interest to WGHABD include the following:

1) Allelochemical interactions
2) Small-scale microbial interactions
3) Life history transitions
4) Selective grazing
5) Succession through grazing
6) Trophic transfer of toxins
7) Turbulence effects on HABs
8) Nutrient effects on HAB dynamics and community composition

Which of these can be addressed through mesocosms? Which cannot? What types of mesocosm strategies might be useful in this regard. Note that this list of topics ranges beyond the focus of the Core Research Project on Eutrophication, yet their expertise can provide useful guidance in this broader context.

Concerns raised by WGHABD about mesocosms:

1) Need to use natural communities, difficulties in capturing natural blooms, or in adding cultures to mesocosms (permit issues);
2) Need to define questions that cannot be addressed in small bottle experiments.
3) What hasn't been done before?
4) The effects of time, walls, turbulence, light, etc.

The WGHAB and its members welcome cooperation with the GEOHAB SSC in the organization of the assessment and white paper either by correspondence or through a joint workshop if feasible.

13 Draft Resolutions

The ICES IOC Working Group on Harmful Algal Bloom Dynamics [WGHABD] (Chair J. Silke, Ireland) will meet in Huelva, Andalucia, Spain from 30 March–3 April 2009. The draft Terms of Reference for the next meeting is presented in Annex 3.

Annex 1: List of participants

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Annex 2: Agenda

11:00 – 12:00 **Opening, Agenda, Announcements (Chair Joe Silke)**

- Welcome
- Introductions
- Housekeeping
- Agenda – Additional Items
- Report of 2006 Meeting
- Nominations for New Chair (Selection Thursday)
- ICES Science Plan Feedback
- AOB

12:00 – 12:30 **WGPEP Update (Joe Silke)**

- Discussion regarding developments at WGPEP
- Developments towards a special theme session at ASC 2008

14:00 – 15:30 **Changes in the distributions of individual species.**

(Lead: Bengt Karlson Rapporteur Allan Cembella)

- Review developments in knowledge pertaining to the development of Verrucophora and Chattonella blooms (ToR d)
- Aerosolized Toxins and methodologies used to monitor them (ToR a)

15:45 - 17:15 **ICES Cooperative Research Report**

(Lead Richard Gowen, Rapporteur Eileen Bresnan)

- Discuss the requirements for and, if agreed, plan the preparation of a draft outline an ICES Cooperative research report on new findings and developments relating to the distribution of phycotoxins and metabolites and recent findings on the distribution of HABs and toxin producing phytoplankton species in the ICES area. (ToR c)
- Brainstorming: Contents / Headings
- Editorial considerations

Tuesday 11 March

09:30 – 10:30 **HAB Modelling**

(Lead Patrick Gentien Rapporteur Jennifer Martin)

- Discuss: Interactions with WGPBI for consideration (ToR b)
- Modelling the 2006 *K. mikimotoi* bloom in Scotland
- Discuss: holding a joint meeting of both groups in 2009

- Key questions for HAB models.. generate a questionnaire?
- Programme of Work / Assign tasks

10:50 – 12:00 **New Findings Session 1 (Lead Bengt Karlson, Rapporteur Pat Tester)**

- 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 (ToR e)

B. Luckas A. ostenfeldii comparison north sea/ Baltic
B Karlson Peridinium quinquecorne
L Turrell Alexandrium sp Scottish waters
S. Kavanagh PHYTOTEST
R Raine Habit cruise 2007
GEOHAB Meetings and Planned activities

12:00 – 13:15 **National Reports Session 1 (Lead Eileen Bresnan, Rapporteur Joe Silke)**

- Collate and Assess National Reports by Country Reps (ToR f)

Wednesday 12 March

09:15 – 10:45 **Database GIS System and Website (Lead Catherine Belin, Rapporteur Marnix Poelman)**

- review the progress on intersessional updating and inputting data in the IOC-ICES-PICES HAEDAT database and developments made towards developing an integrated system (ToR g)
- review the structure and composition of the decadal HAE maps for the ICES region with special reference to the linkage between the decadal maps and the HAEDAT database and the need for new maps for specific algal species (ToR h)
- review intersessional work to generate a website to electronically archive past reports of the WGHABD and to facilitate intersessional work carried out by the group (ToR i)
- IOC workshop in Belgium to develop a 'Harmful Algal Information System'

11:00 – 12:30 **New Findings Session 2 (Lead Allan Cembella, Rapporteur Keith Davidson)**

- 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 (ToR e)

Elie Fux SPATT Technology and new techniques
B Reguera Dinophysis SPATT

	L Turrell SPATT Resins deployment P Tester DA Test Kit A Cembella Poseidon Cruise
13:00 – 13:30	**Laboratory Tour**
13:30 – 14:00	**Continuation of Cooperative research report Discussions**
14:00 – 15:00	**National Reports Session 2 (Lead Eileen Bresnan, Rapporteur Joe Silke)** • Collate and Assess National Reports by Country Reps (ToR f)
15:00 – 15:45	**Mesocosm Studies (Lead Don Anderson, Rapporteur Henrik Enevolsen)** • Investigate and discuss the possibility of comparative studies that will use mesocosms to explore the dynamics of HABs subject to eutrophic pressures (ToR j)
16:00 – 16:30	**Reports from Related Meetings:** IBIROOS Patrick gentien Report from IPHAB (Henrik Enevolsen) IOC Activities (Henrik Enevolsen)
16:30 – 17:15	**Meeting Summary / TORs 2009**

Thursday 10 March

09:30 – 11:00	Draft ToRs for 2009 Meeting
11:00 – 11:15	**Morning Break**
11: 15 – 12: 15	Draft national Reports and WG report
12: 15 – 12:30	Decide on Location for 2009 Meeting Vote for New working Group Chair Conclusion of Meeting

Annex 3: WGHABD Proposed Terms of Reference 2009

The **ICES IOC Working Group on Harmful Algal Bloom Dynamics** [WGHABD] (Chair J. Silke, Ireland) will meet in El Rompido (Huelva) Spain from 31 March to 2 April 2008 to:

a) Discuss the compilation of national practices across ICES areas for Harmful Algae and Phycotoxins monitoring and prioritize updating of the IOC-MONDAT Data Base.

b) With reference to modelling in the ICES region, review the state of knowledge of initiation, maintenance and senescence of cyanobacteria blooms, including transfer of toxins and effects on the foodweb.

c) Discuss and formulate the description and justification for a thematic session on HABs in the Baltic Sea for the 2010 ASC.

d) Identify the requirements for observing specific TPA and HAB species in near real time using automated techniques and produce forecasts of Harmful Algal Events using observations and models.

e) WGHABD wish to pursue by interdisciplinary work with WGPBI the development of joint TORs and a joint WG sessions in 2009

f) 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.

g) Collate and assess National reports (Country Reps) and collect data for HAEDAT

h) Review the UK's DEFRA funded literature and data analysis on HABS and nutrient enrichment. Identify follow up activities.

i) Review the publications in Journal of Sea Research from the ICES Workshop on Time-series Data Relevant to Eutrophication Ecological Quality Objectives (WKEUT)

WGHABD will report by DATE to the attention of the Oceanography Committee.

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)** A wide variety of monitoring practices at the national or regional level hampers time-series comparisons across ICES region. In the first instance it is important to document these monitoring activities. While it should be noted that in parallel the group of National Reference Laboratories in Europe producing a compilation of the management practices for Toxic Algae Events, WGHABD working with IOC can assist in the completion of a similar exercise through the ICES and IOC member states for both HABs and Toxin producing algae and their impacts.. **Term of Reference b)** The cyanobacteria HABs in the Baltic Sea are thoroughly studied and a wealth of information is already gathered that could be more efficiently used in modelling, specifically in the development of species of interest models for the main HAB forming cyanobacteria. WGHABD facilitates the cooperation of modellers and

	oceanographers/biologists therefore rendering this a suitable forum for the compilation of existing knowledge of critical aspects of cyanobacteria HABs with reference to modelling. **Term of Reference c)** Harmful Algal Blooms influences the whole ecosystem in the Baltic. New knowledge pertaining e.g. to oxygen deficiency and phosphate dynamics as well as the introduction of new zooplankton species (e.g. *Mnemiopsis leidyi*) should be of interest to the ICES community. Also results from several HAB-related projects in the EU-BONUS research programme should be ready for presentation at the 2010 ICES-ASC. **Term of Reference d)** Use information from the inventory together with knowledge of bloom dynamics and existing modelling capabilities to identify (i) regional locations where the first HAB observation and forecasting systems should be implemented and (ii) the observational and infrastructural components required to achieve these capabilities. **Term of Reference e)** HAB physical-biological processes is limited. Improved knowledge of the validation of these models and the status of coupled physical-biological process knowledge is essential to improve models for HAB dynamics. **Term of Reference f)** 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 g)** National reports on the previous years highlights of each country's HAB events is a useful means of comparing the regional patterns, and synthesizing the occurrence of HAB events in the ICES area. These reports can also incorporate information necessary for HAEDAT. **Term of Reference h)** Review the 2008 UK assessment of HABs and nutrient enrichment and consider what further work might be undertaken and whether the report should be used as the basis for an ICES cooperative research report. **Term of Reference i)** The WKEUT workshop discussed 17 long-term phytoplankton datasets from European and North American coastlines in order to allow comparative analyses of phytoplankton dynamics in response to nitrification and weather driven changes. Among questions addressed to the datasets were those relating to HABs such as "Have the seasonal bloom patterns and/or bloom species, including HABs, changed over time; is it possible to identify indicator species"? This term of reference will review the publications stemming from this workshop.
Resource Requirements	The research programmes which provide the main input to this group are already 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 Facilities	None
Financial	No financial implications
Linkages to Advisory Committees	There are no obvious direct linkages with the advisory committees
Linkages to other committees or groups	WGHABD interacts with WGZE, WGPE, WGPBI.
Linkages to other organizations	The work of this group is undertaken in close collaboration with the IOC HAB Programme. IOC should be consulted regarding ToR or discontinuation of the WG prior to the ASC. There is a linkage to SCOR through the interactions of the IOC-SCOR GEOHAB Programme.

Annex 4: Recommendations

Recommendation	For follow up by:
1. As this was the third year in the current Chair's tenure, a request for potential candidates for the chair of the working group was made. The existing Chair (J. Silke Ireland) was proposed by Eileen Bresnan (Scotland) and seconded by Pat Tester (USA). There were no other delegates proposed and Mr. Silke was unanimously re-elected to serve a second term.	• Generally addressed to ICES;
2. WGHABD wish to propose a Special Theme Session on Harmful Algal Blooms in the Baltic Sea for the 2010 Annual Science Conference. The Working Group on GEOHAB Implementation in the Baltic (WGGIB) have completed their work in the preparation of the GEOHAB BALTIC plan. A wealth of information on Cyanobacterial HABs is already gathered and WGHABD wish to foster further collaborative studies in the Baltic area, the theme session is proposed to report on studies to date and as a focus to promote future potential activities.	• Generally addressed to ICES; • OCC
3.	
4.	
5.	
6.	