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REPORT OF THE

ICES/IOC WORKING GROUP ON HARMFUL ALGAL BLOOM DYNAMICS

La Roche Canillac, France 22–26 April 1997

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1 WELCOME AND OPENING OF THE MEETING

The ICES-IOC Working Group on Harmful Algal Bloom Dynamics (WGHABD) was convened at ACRO (Association Canillacoise pour la promotion de la Recherche en Océanographie) in St. Pardoux- La Croisille (22-26 April 1997). The meeting was organized by Ian Jenkinson and was chaired by Patrick Gentien (France). 24 scientists from 12 countries took part: they are listed in Annex I.

Tom Osborn was appointed as a rapporteur for the whole session. In plenary session of the WGHABD, individual participants introduced themselves and their institute and gave a concise description of their major field of research.

Since the NATO/ASI meeting on « The Physiological Ecology of Harmful Algal Species », held in Bermuda, (27 May - 6 June, 1996) gathered a great number of scientists emcompassing a large range of fields of expertise, it was felt of interest to present a summary of the recommandations formulated during this meeting. Don Anderson provided informations on the contents of the book which will be available before end 1997 and presented the recommendations which are presented in Annex VI.

2 TERMS OF REFERENCE

At the 84th ICES Annual Science Conference in Reykjavik (Iceland), the council resolved (C. Res. 1996/2:52) that:

The ICES-IOC Working Group on Harmful Algal Blooms Dynamics (Chairman: P. Gentien, France) will meet in La Roche Canillac (22-26 April 1997) to :

\$1 - examine the results of the Workshop on Development of *In Situ* Growth Rate measurements for dinoflagellates and consider future publication;

§2 - collate the National Reports in the usual form;

§3 - review the mapping exercise and propose a format suitable for publishing the work;

§4 - establish recommendations concerning the limitation of transfer of harmful phytoplankters through ballast water discharges;

§5 - define a methodology for estimating the impact of grazers on a given HAB and impact of HAB on recruitment of grazer populations and assess the experimental biases inherent in each method;

§6 - evaluate the role of micro-organic nutrient dynamics and heterotrophic interactions in the initiation and maintenance of HAB;

§7 - evaluate and assess the use of remote and *in situ* optical sensing technology in HAB dynamics studies.

§8 - review the status of development of taxonomic coding systems with a view to recommending the adoption of a single coding system for use in ICES;

§9 - prepare plans for a joint meeting with the Working Group on Phytoplankton Ecology in 1998;

\$10 - consider the design of an experiment to elucidate the role of physical-biological interactions in harmful blooms in ICES area.

3 SUMMARY OF THE CONCLUSIONS

<u>**Term of reference 1**</u>: examine the results of the Workshop on Development of In Situ Growth Rate measurements for dinoflagellates and consider future publication

Since all the sub-reports were not yet available, it was agreed that during 1997 an exchange of data between the participants will be organized in order to discuss each sub-report in a holistic framework. The final report should be finished in February 1998 and revieved by the WG on HABD in 1998.

<u>**Term of reference 2 :**</u> collate the national reports in the usual form

The compiled national reports are appended in Annex II. Country members (1 report is missing) presented in plenary session a summary of their respective national reports for 1996. It is noticeable that in all ICES countries, HAB toxic events have been quite mild. No correlation with meteorology could be found. Some members hypothesized a long term cycle like White's 19-year cycle.

In order to have easier electronic access to the information and data accumulated in the annual National Reports, it was proposed by the IOC Science and Communication Centers that an information data-base be established (see preliminary outline in Annex V). Taking into consideration (i) the usefulness of the ICES national reports on HAB's as the only easy accessible overview of HAB occurrences in ICES area, (ii) the well received results of the HAB mapping exercise (see ToR 3), and (iii) the results of the ICES-IOC survey on HAB monitoring practices (1995-96), the WGHABD supports the establishment of an information data-base compiling the information contained in the National Reports, the results of the mapping exercise, and the results of the ICES-IOC survey. If feasible, the information data-base should be compatible with a GIS system.

In a first phase the results of the mapping exercise could be available as image files only. The information data-base should include national focal points for monitoring data. The WGHABD encourages the IOC Science and Communication Centers to prepare the information data-base prior to the 1998 meeting and make it available at this occasion for demonstration.

Term of Reference 3: review the mapping exercise and propose a format suitable for publishing the work.

Maps of toxin presence for the whole ICES area have been produced by IFREMER (C. Belin) according to the proposals made by the WGHABD in 1996. They are presented in annex III. Countries which have not submitted their data are shaded. Maps are produced under ArcView and could be made available under any of the compatible formats. The yearly update of these maps and their publication should be discussed by ICES.

<u>**Term of Reference 4 :**</u> establish recommendations concerning the limitation of transfer of harmful phytoplankters through ballast water discharges

Critical points concerning HAB species have been identified which are:

* the focus on HAB species is broadened to include other algal taxa, for example toxic diatoms and flagellates, and that the transfer of motile cells in addition to resting stages is investigated

* Studies on the transport of HAB dinoflagellate cysts in ballast tanks sediments continue, and that the relative importance of water column versus sediment origin of cysts be addressed in relation to dinoflagellate life cycles.

Since the transfer of harmful phytoplankters is included in a larger set of problems, it is recommended that:

* Liason with the Study Group on Ballast Water and Sediments continues in order to update knowledge on HAB species transfer in ballast water and sediments, and relevant treatment options.

* Liason with the Working Group on Introduction and Transfer of Marine Organisms be established to address the issue of accidental transfer of HAB organisms via movement of shellfish stocks.

<u>Term of Reference 5</u>: define a methodology for estimating the impact of grazers on a given HAB and impact of HAB on recruitment of grazer populations and assess the experimental biases inherent in each method

A bloom of an algal species – accumulation of biomass above normal levels – implies some relaxation or thwarting of grazing pressure. Even in cases where physical processes concentrate a diffuse population, one would expect dense patches of a benign algal population to attract and concentrate swimming grazers, which would then consume the nascent bloom.

Therefore, studies of grazer (pelagic and benthic) population grazing impact should be incorporated into attempts to understand HAB dynamics. More attention should be paid to benthic grazing as a phytoplankton production loss-term.

Detailed technical and scientific recommendations are listed in the discussion of this term of reference.

<u>**Term of Reference 6**</u>: evaluate the role of micro-organic nutrient dynamics and heterotrophic interactions in the initiation and maintenance of HAB

It is now clear that many HAB species rely on autotrophic and heterotrophic pathways which provide them with competitive advantage over strict autotrophs. Bloom dynamics, effects of eutrophication on species composition of phytoplankton assemblages, life cycle of key HAB species cannot be understood without a thorough examination of mixotrophy switches and potentials.

Most of the studies conducted on mixotrophy have not led to a quantification of the processes, which is urgently needed if realistic models of nutrient transfer are to be achieved.

Detailed technical and scientific recommendations towards this goal are listed in the discussion of this term of reference.

<u>**Term of Reference 7**</u>: evaluate and assess the use of remote and in situ optical sensing technology in HAB dynamics studies

This term of reference was not addressed in the absence of competent scientists from Russia, U.S.A. and Italy. It is, however, recognized that this set of techniques would be of great help in understanding the meso-scale in which these toxic blooms develop.

<u>Term of Reference 8:</u> review the status of development of taxonomic coding systems with a view to recommending the adoption of a single coding system for use in ICES

There is at present a number of taxonomic coding systems which include phytoplankton (NCC, NODC). The ICES Working Group on Marine Data Management is currently reviewing existing taxonomic coding systems with a view to recommending the adoption of a single system. The Working Group on Phytoplankton Ecology is also recommending a practical check list of phytoplankton species in the ICES area. It is therefore recommended that any decisions on adopting an existing system or developing a new one be carried out in association by these groups. It is recommended that the deliberations of the workshop on taxonomic nomenclature at the 8th International Conference on Harmful Algae, Vigo, Spain, June 1997, are taken into consideration.

Term of Reference 9: prepare plans for a joint meeting with the Working Group on Phytoplankton Ecology in 1998

It was felt by the WGHABD members that a joint meeting with WGPE would be necessary in 1998, in order to review the report on the Kristineberg workshop on growth rate measurements and review the status of taxonomic coding in relation to the establishment of a phytoplankton database. Moreover, it would be of interest for the WGHABD members to be informed of the latest developments in phytoplankton ecology. A regular joint meeting of one day every two or three years would be profitable to both groups.

<u>**Term of Reference 10:**</u> consider the design of an experiment to elucidate the role of physical-biological interactions in harmful blooms in ICES area

Considering the history of the WG, the sub-group recommended, in order to facilitate progress in this area, compiling the available scenarios for the various harmful events in the ICES area in order to facilitate the communication between the disciplines and identification of information gaps, sampling problems,... This compliation will be made on the basis of a questionnaire identifying the critical steps in the development of a toxic event. A joint description of the basic systems will form a common base for discussion and modeling.

Understanding how these thin layers of particles develop, what is their relation to the physical fields of temperature, salinity, and density, and what physical processes are involved in forming these biological and chemical layers is necessary for quantitative explanation of physical-biological interactions. The theme session at the ASC in Baltimore on this subject provides some basis for the establishment of a joint investigation between some members of the group.

4 DETAILED DISCUSSION OF THE TERMS OF REFERENCE

Term of Reference 1 :

The ICES/IOC Workshop on Intercomparison on *in situ* growth rate measurements (Dinoflagellates) was held in Kristineberg, Sweden, 9 - 15 September 1996. The list of participants to the workshop is appended in Annex V.

1. Opening of the Workshop

The workshop was opened by the chairman Dr. Odd Lindahl. He also welcomed participants and gave a short presentation of Kristineberg Marine Research Station with history back to 1877.

1.1 Approval of the agenda and rapporteur

The tentative agenda was approved by the workshop. Mr. Einar Dahl was appointed as rapporteur.

1.2 The purpose of the workshop

The purpose of the workshop was to intercompare a number of methods, traditional as well as newly developed, in order to measure the *in situ* growth rates of dinoflagellates. Estimates of population dynamics, such as growth rates, are essential to providing the means to quantify the detailed structure and processes which lead to a capability to model algal populations and bloom development.

2. Kristineberg and the Gullmar fjord

Kristineberg is located at the mouth of the Gullmar fjord on the west coast of Sweden. The hydrographical conditions in the surface layer of the area are dynamic with different waterbodies flushing through, mainly driven by the wind. That changes may occur was demonstrated by a large raise in salinity at 1m depth from 3 to 8 September due to northeasterly winds blowing the surface water out of the fjord just before the workshop. The most typical hydrographical situation is a rather strong stratification due to the influence of Baltic water. Due to the different waterbodies flushing the area the plankton diversity over time was variable.

3. Logistics

Kristineberg Marine Research Station has new and well equipped laboratories, partly with accreditation certificate, and members of the skilled staff was allocated to support the workshop with technical assistance and basic data as nutrients and chlorophyll. Arrangements for the mesocosm were made at or on the quay and the unialgae cultures were kept in culture rooms with climate control.

4. Presentation of participants and methods

The following participants took part in the workshop and used the techniques/methods listed below. The methods were applied on unialgal cultures and natural communities enclosed in mesocosms.

Participants	Country	Technique/method
Dahl, Einar Sampayo, Maria Antonio Catherine Legrand	Norway Portugal Sweden	Cell counts
	Spain	Mitotic index
Reguera, Beatriz	Span	Whote much
Gentien, Patrick	France	Viability of cells
Lindahl, Odd	Sweden	Plankton cages, ¹⁴ C-methods
Davidsson, Lennart	Sweden	
Hernroth, Bodil	Sweden	
Edler, Lars	Sweden	
Fredrik Norén	Sweden	

Participants	Country	Technique/method
Anderson, Don Allan Cembella Granèli, Edna Carlsson, Per Lars-Åke Gisselsson	USA Canada Sweden Sweden Sweden	Flow cytometry, DNA/RNA/cell
Peperzak, Louis Sandee, Ben	Netherlands Netherlands	Flow cytometry, DNA
Micheli, Carla Barbini, Roberto Fantoni, Roberta Palucci, Antonio Ribeho, Sergio	Italy Italy Italy Italy Italy	LIDAR Fluorosensor System Laser fluorometer
Colijn, Franciscus Hartig, Peter	Germany Germany	PAM technique

5. Coordination of the experiments

The participants agreed to put their efforts on algae samples from mesocosms and cultures, with field sampling as an option. It was decided to set up two mesocosms, one without (control) and one with nutrients. Nutrient data from the Gullmar fjord just prior to the workshop showed concentrations at or below detection level both for inorganic phosphorous and nitrogen.

It was decided to focus on the cultures of *Prorocentrum micans* and *Alexandrium fundyense*. The need for careful coordination to obtain comparable results was expressed. One important point in that respect was the schedule of sampling the mesocosms as well as the cultures. In order to fulfill this point sampling, followed by subsampling for the different methods, were carried out every second hour. The measuring frequency could be different for the different methods applied.

6. Experimental set-up and testing

Strong winds and heavy waves damaged the *in situ*, floating mesocosm first established at the dockside and delayed the start up of the experiment half a day. As an alternative twelve 300 L polyethylene barrels were placed in the shade on the quay and filled with water from 3 m depth by a low volume diaphragm pump. Six barrels represented the control and six the nutrient enriched mesocosm. One barrel with nutrients was placed on a float in an orientation accessible to the LIDAR equipment. Phosphate and ammonium were added to achieve a starting concentration of about 0.5 and 5 μ M respectively in the enriched mesocosm tanks.

Cultures of *P. micans* (CCMP 1589) ans *A. fundyense* (CA28) which arrived at Kristineberg three days before the experiment start up, were kept at 20 °C in a 12:12 light:dark cycle at 19 W m^{-2} (measured with a Zemoko dosemeter).

Parallell to the experimental set-up, the participants tested their equipment and methods by using material from the field or cultures.

7. Sampling during the experiment

The sampling started 06.00 a.m. on 11 September, about twelve hours after the barrels were filled, and continued with a frequency of every second hour until 06.00 p.m. on 12 September.

Before each sampling, the barrels were gently, but carefully, mixed by a plastic disc mounted on a shaft. Then a sample of about 7 L from each barrel was immediately collected and mixed with the samples coming from the other barrels belonging to the same type of mesocosm. From these composite, subsamples were distributed for the different measurements.

8. Measurements and analyses

Some measurements and analyses could be performed during the experiments. Results of the following parameters were presented to the participants during the experiment: temperature, salinity, nutrients, chlorophyll, cell counts, ¹⁴C-uptake and the viability of cells. The measurements of particulate organic carbon (POC) and dissolved organic matter (DOM) were sent to laboratories in Gothenburg for analyses. However, most of the measurements and analyses had to be fulfilled by the participants at their laboratories. It was decided to send sub-reports to the chairman within two months.

9. Present status of the workshop

Looking in retrospect, it was far too optimistic to believe that the sub-reports should be ready within two months. Only some few of the participants were able to manage that. At the ICES WG-meeting in Saint-Pardoux-La-Croisille, a collection of the available sub-reports were distributed. However, some subreports were still missing but were in progress.

At the WG-meeting, some preliminary results from the experiment were presented and the need for inter-comparisons and harmonization of the data was strongly expressed. Thus, it was agreed that during 1997 an exchange of data between the participants will be organized in order to discuss each sub-report in a holistic framework. The final report should be finished in February 1998 and revieved by the WG on HABD in 1998.

Term of Reference 2 : collate the National Reports in the usual form

The compiled national reports are appended in Annex III. Country members presented in plenary session a summary of their respective national reports for 1996. It is noticeable that in all ICES countries, HAB toxic events have been quite mild. No correlation with meteorology could be found. Some members hypothesized a long term cycle.

In order to have easier electronic access to the information and data accumulated in the annual National Reports, it was proposed by the IOC Science and Communication Centers that an information data-base be established (see preliminary outline in Annex). Taking into consideration (i) the usefulness of the ICES national reports on HAB's as the only easy accessible overview of HAB occurrences in ICES area, (ii) the well received results of the HAB mapping exercise (see ToR c), and (iii) the results of the ICES-IOC survey on HAB monitoring practices (1995-96), the WGHABD supports the establishment of an information data-base compiling the information contained in the National Reports, the results of the mapping exercise, and the results of the ICES-IOC survey. If feasible, the information data-base should be compatible with a GIS system.

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Canada

In Canadian coastal waters, PSP toxicity continues to be the predominant phycotoxin problem of concern to regulatory authorities. During the past year, PSP toxicity in the Bay of Fundy, along both the New Brunswick and Nova Scotian coasts, traditionally areas of high summer toxicity in mussels, *Mytilus edulis* and soft shell clams *Mya arenaria*, was unusually low. In general, peak domoic acid levels in shellfish have receded dramatically in eastern Canada since the major incidents in eastern Prince Edward Island in the late 1980s. Regular monitoring for domoic acid in shellfish is maintained for key stations in British Columbia and Atlantic Canada. Toxicity associated with DSP has been identified at only a few locations in Atlantic Canada, in Nova Scotia and Newfoundland, and toxicity levels have remained <1 μ g okadaic acid equivalents per g shellfish tissue, therefore toxicity monitoring is confined to a few key aquaculture sites. Some unusual observations deserve special mention here:

- a) Alexandrium ostenfeldii was recently found in phytoplankton populations from the lower St. Lawrence estuary and in coastal embayments in southeastern Nova Scotia. This species is toxic but has an unusual toxin profile; it is readily distinguishable from A. tamarense with which it co-occurs in Nova Scotian waters.
- b) Studies on the dynamics of benthic *Alexandrium* cysts at mussel aquaculture sites in Newfoundland have recently confirmed the hypothesis that winter toxicity in mussels in this area is due to resuspension and ingestion of benthic cysts and not to cryptic populations of vegetative cells or prolonged toxin retention times.

- c) The source of episodic DSP toxicity in mussels and sea scallops from Nova Scotia has not yet been definitively identified, but it does not appear to be linked to Dinophysis spp. blooms, which tend to occur in early summer (June/July) and early fall (September). Extensive analysis by sensitive LC-MS methods of field phytoplankon populations rich in *Dinophysis* spp. (*D. norvegica*, *D. acuminata* and *D. acuta*) morphotypes similar or identical to toxigenic European forms, failed repeatedly to show the presence of DSP toxins. Attention is now focused on the epiphytic dinoflagellate *Prorocentrum lima* which is found attached to substrates at mussel aquaculture sites; isolates produce DSP toxins (primarily DTX4, okadaic acid and diol-esters) in culture, whereas in mussels DTX1 tends to predominate.
- d) A novel class of lipophilic bioactive compounds named spirolides has been found in digestive tissues of bivalve shellfish and in specific size fractions of plankton net tows in Nova Scotian waters. Spirolides are of unknown human health significance but they provoke a dramatic neurotoxic response in mice upon i.p. injection in the conventional DSP mouse bioassay. These compounds are circumstantially linked to the presence of relatively featureless spheroidal cells of mean diameter 42 μm (dubbed GB-42), now believed to have affinities with gonyaulacoid dinoflagellates. Spirolides in the plankton are always found in association with low to moderate levels of PSP toxins.

Denmark

Relatively low phytoplankton biomasses and concentrations were registred in the summerperiod in Danish waters. The biomasses were dominated by diatoms (e.g. *Skeletonema costatum* and *Rhizosolenia fragilissima*) and dinoflagellates (e.g. *Prorocentrum minimum*). DSP was registred locally at the east coast of Jutland in the beginning of the year (jan-feb) in a situation with low concentrations of *Dinophysis* spp. Neither PSP, ASP or fish kills due to HAB's were registred i 1996. An exceptional local bloom of *Dinophysis acuminata* (max. conc. 11 x 10^3 cells/L) was registred in october in Præstø Fjord. No harmful effect were registred in connexion with the bloom.

Finland

Early summer 1996 was exceptionally cold in the Northern Europe and consequently, the late-summer cyanobacterial bloom in the Gulf of Finland and the Northern Baltic proper was delayed with ca. 2 weeks from its normal timing, being most intense in late August. The bloom was dominated by *Nodularia spumigena* and *Aphanizomenon flos-aquae*. No toxic effects were reported on the Finnish coast.

In July 1996, an intense bloom of *Heterocapsa triquetra* was observed in the western Gulf of Finland. The bloom biomass advected to coastal zones colouring the water reddish brown on several areas along the Finnish SW coast. This was the first observation of a red tide by *Heterocapsa* in the Finnish waters since mid 1970s. Okadaic acid was, for the first time analysed in the Gulf of Finland in mussels in 1993 and in flounder in1996. Traces of OA were found in the soft tissues of Mytilus edulis. In flounders, maximum concentration of 200 ng OA g-1 was observed in the liver tissue (Pimia et al. 1997).

France

DSP toxicity (*Dinophysis* spp) affected 18 sites in 1996. All sites, except one in Mediterranean, were situated on the Atlantic coast, along the southern coast of Brittany and around Loire estuary.

PSP toxicity (*Alexandrium minutum*) was recorded in the same site than the last seven years, i.e. Morlaix bay in Western Brittany. But it was also recorded for the first time in a site of northern Brittany. A study on *Alexandrium minutum* cysts along the whole coast of Brittany, also revealed a geographical extension of these cysts in the sediment of Brittany areas.

Germany

(see also report "MURSYS" distributed by Bundesamt für Seeschiffart und Hydrographie. Hamburg) North-Sea :

After a cold winter, the le large diatoms *Coscinodiscus wailesii* and *C. concinnus* developed dense populations. During collapse of these populations large amounts of lipid acids were set free into the water column and dispersed about several 100s of km² north of the East-Frisian Islands, easily detected by satellite remote sensing. First it was supposed, that a tanker had washed its tanks with palm-oil. Analysis of the fatty-acid composition showed that the fatty acids were different from any commercial product but were similar to those of *Coscinodiscus*. Many Eeiderducks and other

seabirds were oiled and washed ashore. A similar event had been reported earlier by Grøntved (1951) from the open North-Sea.

Dinophysis acuminata developed high population densities, up to 70 000 cells -l, in the open North Sea off Helgoland but also in a brackish water reservoir on the coast of Lower Saxonia. As no mussels were harvested at these sites, no adverse effects have been reported.

In the waters off Eiderstedt the raphidiophyte *Fibrocapsa japonica* was reported again at cell concentrations of 30 000 cells-l. Also *Heterosigma akashiwo* was recorded several times but no adverse effect was reported. For the first time, the raphidiophyte *Chattonella* was found in samples. This species was not found in plankton samples but was restricted to sediment samples taken off List/Sylt. It may be an undescribed species.

The Prymnesiophyte *Prymnesium patellifera* formed dense blooms up to 6 million cells-l, in backwaters near Büsum, Schleswig-Holstein in a place were earlier fish-kills occurred.

The large colony forming prymnesiophyte *Phaeocystis* and the large heterotrophic dinoflagellate *Noctiluca scintillans* did not form large blooms in 1996, in contrast to nearly every year.

The health authorities analysed fresh mussels and imported mussel products. Only few of the fresh mussels contained low concentrations of PSP-toxins but 7 samples of the canned mussel products contained PSP- toxins; in some cases the PSP toxin concentrations were just below the allowed levels ($800 \mu g/kg$). DSP-toxins were not recorded in any sample. So far, ASP is not analysed.

Baltic Sea :

Cyanobacteria such as Aphanizomenon flos-aqua, Anabaena sp.and Nodularia sp. reached cell numbers of 15 million cells-l in Flensburg Fjord, Kiel Bight, and the open Baltic Sea as well as in the Boddenwaters of Rügen. Prorocentrum minimum produced a bloom of a maximum of 225 million cells-l in the Arcona Sea, Dinophysis acuminata was registered in low numbers.

Ireland

The 1996 results were notable for the very low numbers $(40 - 200 \text{ cells/l}^{-1})$ of *Dinophysis* sp. compared with previous years. A bloom of *Alexandrium tamerense* was recorded in Cork Harbour.

Closure (2 weeks) of markets due to D.S.P. contrasts markedly to the closure period enforced in the southwest in the period of 1994/95 which lasted for up to 10 months. Harvesting closure due to P.S.P. lasted 2 weeks: it was the first time P.S.P. was detected in Irish waters.

Unexplained toxicity was detected in mussels from Killary Harbour in the absence of known toxic phytoplankton cells and lasted 8 months.

Norway

Problems due to harmful algae were small in Norway in 1996. Only two events of local shellfish toxicity due paralytic toxins were recorded. However, in one case as much as 16 000 MU per 100 g mussel meat measured.

Portugal

An unusual event was the *Lingulodinium polyedrum* red tide. This species has been responsible for red tides in the forties and after became rare in the phytoplankton population. A possible explanation can be that the heavy rains we had this year disturbed coastal sediments, bringing cysts to the upper layer and favoured excystment. For HAB, in general it was a very mild year.

Scotland

PSP was detected on the east and west coasts of Scotland and also in the Orkney and Shetland Islands. Voluntary Closure Agreements (VCA) were used when required and in one location, a closure order was placed on offshore scallops under FEPA 1985 legislation. DSP in mussels on part of the north east coast required a VCA. Domoic acid was detected for the first time in Scottish waters during 1996, and ASP will now be included in the routine monitoring during

1997. Regular phytoplankton monitoring was carried out for the first time, and 21 sites were sampled weekly from April to September and monthly thereafter. The programme demonstrated the appearance of potentially toxic cells in Scottish waters in conjunction with toxicity in shellfish in certain areas.

Spain

1996 was an unusally quiet year in the Galician coast, where a very intensive monitoring takes place to ensure a safe marketing of the large mussel production and other bivalves in the area. There were only a few isolated and mild DSP episodes in the Galician Rías Bajas associated with low levels (less than 1000 cells/l) of *Dinophysis spp.* Scallops still showed remains of ASP toxins associated with the 1995 blooms of *Pseudo-nitzschia* species.

In Catalonia (Mediterranean coast), small scale episodes known from previous years, took place in different embayments. These included development of non toxic blooms affecting tourism (*Alexandrium taylori*) or wild/cultured fish populations (*Gyrodinium corsicum*) and toxic blooms of *Alexandrium minutum* (PSP) rendering the wild mussels toxic or causing water discolorations.

Sweden

In the Skagerrak and Kattegat area there were only a few exceptional phytoplankton blooms in 1996.

In the beginning of April a bloom of a non identified dinoflagellate, resemblig *Gyrodinium aureolum*, was observed in Byfjorden. No adverse effects were reported. In the end of June and beginning of July there was a large bloom of *Emiliana huxleyi* in a large part of eastern Skagerrak. Cell densities were high enough to color the water greenish. There were no reports of adverse effects caused by this bloom. In the beginning of July, *Alexandrium minutum* was for the first time observed along the Swedish Skagerrak coast. Cell densities were less than 5 000 cells/L and no toxic events were reported.

Between 15 and 25 of September *Prorocentrum minimum* developed a bloom in Laholm Bay in the southeast Kattegat. There were no reports of adverse effects caused by this bloom.

The concentration of Okadaic acid in mussel meat ranged from 10 to 70 mg/kg mussel meat between January and April. From May through August the concentrations were less than 10 mg/kg mussel meat. In September they started to increase and in November the highest concentration measured was 430 mg/kg mussel meat. In December values up to 730 mg/kg mussel meat were measured. The food administration in Sweden has a limit for consumtion of 400 mg/kg mussel meat, whereas EU-countries have agreed on a limit of 80-160 mg/kg mussel meat.

In large parts of the Baltic Sea blooms of bluegreen algae with considerable surface accumulation were common from the end of July till the beginning of September. Toxicity was registered in a algal sample obtained east of Gotland.

U.K.

The year 1996 was unusual in that no Alexandrium blooms (or PSP toxins in flesh samples) were detected off the North East coast of England, although they have previously been a regular occurrence. Blooms in other areas were present for about half the time period, compared with 1995.

U. S. A.

1996 was an unusual year for HAB events in the United States for several reasons. First, there was virtually no PSP in New England, an area which has had recurrent outbreaks in 22 of the past 24 years. It is of note that the same was generally true for the Bay of Fundy region of Canada, where 1996 PSP levels were below detection levels at most stations and times, whereas PSP is typically quite high and regular in those waters. Thus we can conclude that some unknown factor or factors operative on a regional level was responsible for the lack of *Alexandrium* blooms. A workshop with participants from Canada and the New England states explored possible explanations for the regional lack of toxicity, but did not identify any probable explanations.

The second unusual feature of 1996 was the bloom of *Gymnodinium pulchellum* in the Indian River Lagoon, Florida, in September and October. The water was discolored by the blooms, and fish kills were observed. There were also human respiratory problems (stinging sensations in mouth and lungs, and sinus irritation) and eye irritation. This bloom represents the first record of *G. pulchellum* in the western North Atlantic and Americas. An isolate is being cultured to confirm ichthyotoxicity. *Gymnodinium pulchellum* Larsen 1994 is also known as *Gymnodinium* sp. 'type 84-K' and *Gymnodinium* sp. (Japan). The Japanese *Gymnodinium* sp. tested by Endo e al (1992) contained oxidized brevetoxins.

The third unusual event was the first record of NSP in the northern Gulf of Mexico waters of Louisiana, Mississippi, and Alabama in November and December. Economic damages were severe due to the quarantine of oyster harvesting for several months during the peak holiday season. Loss etimates are \$300,000 per day. This bloom of *G. breve* is thought to have originated in offshore waters, with delivery of the cells by onshore water mass movement. One other noteworthy feature of this bloom is that cels were persisting (and perhaps growing) at 15.5 psu, even though the prevailing view is that *G. breve* cells lyse at this salinity, and do not grow well at salinities lower than 24 psu. It may well be that a different genetic strain of *G. breve* was responsible for the northern Gulf bloom, compared to that responsible for outbreaks on the west Florida shelf. The coast of Texas was also hit by a destructive G. breve bloom in September and October, 1996. The water was discolord, and approximaely 5 million fish died. Salinity ranges were 25-42 psu. This bloom was probably advected from offshore waters with subsequent *in situ* growth.

Another unusual event was the 18 month Florida red tide of *Gymnodinium breve* which ended in mid-1996. This was a very large, long-duration bloom along the west Florida shelf that caused extensive damage due to aerosolized toxin, NSP, and fish and animal mortalities. The latter included the mortality of 150 manatees, an endangered species.

Other HAB events are more "normal" compared to other years. PSP was detected in California, Washington and Alaska, at generally low levels. The New York brown tide occurred, but with cell densities of *Aureococcus anophagefferens* about an order of magnitude lower than those from major bloom years. The Texas Brown Tide continued for yet another years, representing the longest documented marine algal bloom. This problem first occurred in December 1989 in the Laguna Madre section of Texas, and has persisted ever since.

No ASP or DSP was reported in the U.S. in 1996.

Term of Reference 3 : review the mapping exercise and propose a format suitable for publishing the work.

Maps of toxin presence for the whole ICES area have been produced by IFREMER (C. Belin) according to the proposals made by the WGHABD in 1996. They received the agreement of the whole group. Two cases of ASP have been noted in Europe (Danmark and Spain). They are presented in annex III. Countries which have not submitted their data are shaded. Maps are produced under ArcView and could be made available under any of the compatible formats. The yearly update of these maps and their publication should be discussed by ICES.

<u>**Term of Reference 4**</u>: establish recommendations concerning the limitation of transfer of harmful phytoplankters through ballast water discharges

A newly formed ICES Study Group on Ballast Water and Sediments (SGBWS) met in La Tremblade, France on April 21st 1997 to discuss the role of ballast water and sediments in the transfer of aquatic organisms between different geographic areas. The Study Group addressed in some detail a wide range of organisms transported in ballast and reviewed ongoing studies in ICES countries. The WGHABD also includes ballast water in it's Terms of Reference in connection with the ballast tank transport of HAB species.

Most of the existing information on HAB species in ballast is concerned with the transport of dinoflagellate cysts in tank sediments. Cysts are frequently found in sediments which have accumulated at the bottom of dedicated ballast tanks and cargo holds used to carry ballast water, and are occasionally found in ballast water. In addition to dinoflagellates, resting spores of diatoms are frequently present in ballast tank sediments and germination and culturing studies have shown that many ballast tank phytoplankton are viable. There is to date little information on other HAB organisms in ballast e.g. toxic flagellates, diatoms or cyanobacteria. Whilst ballast tank transport of viable dinoflagellate resting cysts has been well documented in recent years, proving conclusively that ballast transport alone is responsible for the appearance of HAB species in new areas is often problematic. However, in the context of HAB, this is perhaps less important than demonstrating that ballast transport does occur, and that cysts of potentially harmful species can be discharged to areas previously free from the harmful effects associated with particular HAB species. Dinoflagellate cysts are an important component of ballast water and sediment biota which can contain taxa ranging from prokaryotic organisms to fish, and whilst it is recognised that complete elimination of risks from all taxa is unrealistic, efforts can be made to limit the risks from organisms recognised as harmful in other areas. It is difficult to accurately predict which ballast-borne organisms may become nuisance species in the discharge environment, but known HAB species should always be regarded with caution.

When addressing how to limit the transfer of non-motile cysts in ballast tanks, it is important to consider their origin. For example, there is some evidence to suggest that cysts in ballast sediments may originate from motile cells in the water column taken up during ballast loading which encysted once inside the ballast tanks, but it is unlikely that motile cells could encyst unless gamete production and fusion had already occurred. Non-motile resting stages may also originate from sediments in the area where ballast is loaded if the water is shallow and the seabed is disturbed during ballast loading - this can occur when vessel operations or other external factors cause resuspension of sediments which release benthic cysts to the water column. If cysts originate from motile cells in the water column, it may be possible to make better assessments of the risks involved in ballast loading at particular times of year in certain locations, and to avoid ballasting in those areas during periods of greatest risk.

The shipping industry is subject to substantial economic pressures and is unlikely to readily adopt prevention or treatment measures which will significantly increase their operating costs. In addition, the safety of each vessel and it's crew is of prime importance, and any proposed treatment options for ballast water must not compromise vessel stability or safety. Recommendations outlined in the International Maritime Organisation (IMO) Resolution A.774 (18) provide guidance for countries advising shipping operators on safe ballast water practices which may reduce the risk of introducing non-native or harmful species to new areas:

<u>Non-release of ballast water</u> - whilst undoubtably an effective method in preventing transfer of harmful organisms, it is in very many cases impractical, particularly when vessels are involved in bulk cargo (e.g. oil, gas, ore) transport and only carry cargo in one direction. The ship must be fully ballasted when "light ship" and in order to make cost-effective use of it's capacity, cannot afford to retain ballast on board.

Ballast water exchange and sediment removal at sea or in designated areas - this method is regarded as, in the absence of more scientifically based means of control, a potentially effective way of limiting the probability of transporting freshwater or coastal species to other freshwater or coastal areas. This option, which would probably involve continuously flushing ballast tanks rather than total reballasting, is likely to be favoured by some, as it is relatively inexpensive and experimental trials during oceanic voyages have shown that when correctly carried out, it can greatly reduce planktonic flora in ballast tanks. However, problems associated with this method include how safely the practise can be carried out by all vessels in all sea conditions, ensuring that vessels have complied with the regulations, and of particular importance in the ICES area, mid-water exchange in regional seas such as those around continental Europe may not reduce the diversity and abundance of phytoplankton in ballast tanks, but in fact exacerbate the problem. In addition, many shipping routes in the ICES area are relatively short, and survival of motile phytoplankton may be greater than during trans-continental voyages.

<u>Preventing or minimising uptake of contaminated water or sediment during ballasting</u> - in the context of HAB, port and harbour authorities should be vigilant regarding red tide events or shellfish toxicity in waters under their jurisdiction, and prevent ballast loading at these times. The major problem with this option is how port authorities would be kept aware of such events - unless red tides or HABs were clearly evident (e.g. water discolouration) or regular monitoring was carried out (e.g. monitoring shellfish toxicity as part of a national programme), operators would be unaware of problems.

Discharge to shore-based facilities for treatment or controlled disposal - this option is possibly the most costly, as shore based reception facilities for ballast water and sediments would require major investment by port operators, and would probably be resisted by the industry. Particular care should be taken when disposing of ballast tank sediment. Vessels which carry ballast only in dedicated ballast tanks tend not to routinely dispose of sediment, but as these tanks are rarely inspected, it is not known how sediment accumulations may be resuspended during ballast loading and discharge, or if rough weather conditions cause cysts to be transferred from tank sediments to ballast water which can then be readily discharged in the normal way. The crews of vessels which carry ballast in flooded cargo holds are often instructed to remove sediment from the tanks when empty and dispose of into port waters. This practise carries very high risks of discharging sediment-borne organisms to the receiving environment and should be avoided.

In addition, IMO encourages research into new ballast treatment strategies including physical and chemical control options. Chemical methods for preventing the germination of some dinoflagellate cysts have been investigated in laboratory studies, but when scaled up to field conditions, would prove costly and result in problems with chemical disposal. Some physical methods have been trialled, with heat treatment showing potential as an effective method for preventing germination of Gymnodinium catenatum cysts. Other physical methods (e.g. microwave, UV, electric shock, filtration and centrifugation) have been proposed for similar investigations.

The ICES SGBWS is to review options for the control of dissemination of organisms by ballast, and WGHABD should monitor their progress in relation to HAB species transfers. The WGHABD should also consider the movement of commercial shellfish stocks in the accidental transfer of motile and resting stages of phytoplankton, and whilst this item may be addressed by the Working Group on the Introduction and Transfer of Marine Organisms (WGITMO), it is an issue which is highly relevant to HAB events, particularly in Europe where restrictions on movements of shellfish stocks are less rigorous than for example in the USA and Canada. The WGHABD recognises that blanket precautionary measures may not be the most suitable for reducing transport of HAB species in ships' ballast, and that regional

differences in phytoplankton ecology, HAB history, shellfish exploitation and shipping activity should be taken into account when developing mitigative strategies. The WGHABD is also aware that legislative powers over ballast water management are complex and that any regulatory developments are likely to be established through the framework of IMO.

To conclude, the WGHABD recommends that :

- 1. The focus on HAB species in ships' ballast is broadened to include other algal taxa, for example toxic diatoms and flagellates, and that the transfer of motile cells in addition to resting stages is investigated.
- 2. Studies on the transport of HAB dinoflagellate cysts in ballast tank sediments continue, and that the relative importance of water column versus sediment origin of cysts be addressed in relation to dinoflagellate life cycles.
- 3. Liason with SGBWS continues in order to update knowledge on HAB species transfer in ballast water and sediments, and relevant treatment options.

4. Liason with WGITMO be established to address the issue of accidental transfer of HAB organisms via movement of shellfish stocks.

<u>Term of Reference 5</u>: define a methodology for estimating the impact of grazers on a given HAB and impact of HAB on recruitment of grazer populations and assess the experimental biases inherent in each method

A bloom of an algal species – accumulation of biomass above normal levels – implies some relaxation or thwarting of grazing pressure. Even in cases where physical processes concentrate a diffuse population, one would expect dense patches of a benign algal population to attract and concentrate swimming grazers, which would then consume the nascent bloom. Therefore, interactions between HAB phytoplankters and grazers are extremely important in HAB bloom dynamics.

Grazers of phytoplankton occupy two aquatic habitats: pelagic and benthic. Pelagic grazers include protistan microzooplankton, crustacean mesozooplankton (chiefly copepods), and pelagic larvae of many benthic animals, including crustaceans and bivalve mollusks. Benthic grazers include many invertebrate groups, including commercially-important bivalve mollusks.

There are several ways in which grazers can interact with harmful algal blooms. Grazing impact can prevent or terminate blooms, and grazers are the initial animal entry-point for phytoplankton toxins to be transported through marine food webs, possibly causing vectorial intoxication of upper-trophic-level consumers such as fish, shellfish, and marine mammals. In order to quantify and understand these processes, proper measurement of grazing rates is required.

Field and laboratory experimental methods to address questions of HAB interactions with pelagic grazers have been adapted from protocols designed to elucidate feeding processes in unperturbed systems. Interference with expected feeding or other behaviors implies toxic or harmful effects. Histological examination of affected animals, compared with normal or control animals, can offer insights into the mode of action of a harmful phytoplankter as well. Although copepods often are responsible for the largest portion of pelagic grazing, an analysis of who is eating whom in bloom and in non-bloom situations could reveal trophic shifts associated with bloom initiation. An example of this would be the cascade effect of increasing predation on copepods by higher trophic levels, such as finfish and ctenophores, leading to reduced grazing pressure on phytoplankton.

To assess the impact of grazer communities on harmful blooms, one must measure grazing rates of individual animals, quantify the abundance and composition of the grazer community, and toxic and non-toxic phytoplankters in the natural assemblage. If this is done, individual-animal grazing rates can be multiplied by abundances of those grazer taxa, and the numbers of target phytoplankton cells removed by this population grazing impact can be estimated as a proportion of the total abundance of the population of the target phytoplankters.

Grazing rates of individual grazers must be determined experimentally. This is done by adding known numbers of grazers to natural assemblages in experimental aliquots, and comparing numbers of various phytoplankton taxa remaining in experimental aliquots with samples preserved at the initial onset of the experiment, and with ungrazed controls which are allowed to incubate with experimental aliquots under the same conditions for the same period.

The major limitation to such experiments is the precision and time consumed in quantifying phytoplankton assemblages by microscopic counting. Specifically, if the experimental error in the phytoplankton counting exceeds the amount of phytoplankton removed by grazing, then grazing will go unrecorded. This becomes particularly troublesome at high phytoplankton concentrations, such as those typically found in cultures, and occasionally during extraordinary natural blooms. Specifically, at typical phytoplankton counting precision levels of + or -10%, at high phytoplankton levels of 10-5 or 10-6 target cells per liter, 10% of the total phytoplankton may exceed the amounts of cells removed by grazers during a typical grazing experiment. In such cases, one might wish to use alternative methods such as radioisotope-labelling techniques (Watras et al. 1985), later applied to blooms of cyanobacteria in the Baltic (Sellner et al. 1986).

Quantification of phytoplankton by microscopic techniques, though laborious, allows measurement of grazing upon target toxic, as well as co-occurring non-toxic phytoplankters. The feeding upon non-toxic taxa will likely affect rates of feeding upon taxa (Turner and Anderson, 1983). Quantification of the phytoplankton composition and abundance in the initial sample provides a baseline against which grazing impact can be compared, allowing estimation of the percentage of the target population removed by grazing.

Once rates of grazing for individual grazers are obtained, these can be multiplied by field abundances of those grazers to obtain estimates of grazing by the entire population of each measured grazer taxon; this requires quantification of components of the natural grazer assemblage. For grazers such as copepods and other metazoans, abundances can be obtained using nets with appropriate mesh, equipped with flowmeters. For abundances of protists and other micrograzers, samples should be taken with water bottles, and protists should be concentrated either gravimetrically or by screening through a fine mesh, as most appropriate for the grazers of interest. After collection, zooplankton or other grazer assemblages should be preserved and subsequently counted and identified microscopically.

To estimate zooplankton community grazing impact, animals to be used in experimental studies should be collected from the same waters where blooms occur. This ensures that grazers are pre-conditioned to feeding in the bloom assemblage. If one uses the most abundant grazers as experimental animals, then extrapolation of the population grazing for these taxa can estimate grazing impact by much of the total grazer community. Experimental animals should be sorted alive under a dissecting microscope, and quantitatively added to experimental suspensions. After applying the formulae of Frost (1972) to counts of phytoplankton in initial, control and experimental containers, grazing rates can be estimated, accounting for growth of phytoplankton during incubations, in control containers.

If one seeks to understand possible effects of toxic phytoplankters upon fecundity of, and recruitment to grazer populations, one should precondition grazers to experimental food suspensions prior to beginning each experiment. This will ensure that the experimental food suspension is that reflected in any resulting egg production or larval recruitment (Tester and Turner, 1990).

If using metazoan grazers such as copepods, one should test hatching of eggs into larvae (nauplii) rather than assuming that all eggs produced actually hatch into viable larvae. The reason is that studies by Ianora and Poulet and colleagues have revealed that there are considerable variations in hatching success of eggs produced, and that these can relate to the composition of ingested food.

Information on interactions between HAB's and pelagic larvae of bivalve mollusks is very rare. Recruitment failure of bivalves has been seen when blooms are coincident with the larval stage. Examples include failure of bay scallop sets during the North Carolina red tide of 1987 and during the recurring brown tides in Long Island, NY, bays. Recruitment of bivalves is, however, inherently variable, therefore, it is difficult to attribute recruitment failure directly to the HAB. Laboratory methods, in which bivalve larvae, spawned in captivity, are exposed to cultured algal isolates, have revealed lethal and sub-lethal effects of HAB species. These experiments follow well-established aquaculture protocols which may introduce experimental biases when applied to interpretation of field observations. Some possible biases include : 1) concentration of both algae and larvae at densities far above those encountered in nature, 2) feeding a unialgal or simple mixed algal diet that does not represent a natural assemblage very well, and 3) use of cultured algae that may differ physiologically or genetically from field populations. Some of these biases can be addressed with thoughtful experimental design. The strength of this laboratory approach is, however, that a carefully-controlled experiment can provide unequivocal data that often cannot be obtained in field studies. Currently, work is needed urgently on questions of how much and how long the exposure to an HAB species must be before bivalve recruitment is affected or grazing pressure is affected significantly.

If benthic consumption of phytoplankton does not represent a significant loss term for the phytoplankton populations in a given environment, then bivalves and other benthic filter-feeders can be removed from HAB dynamics models. Information from existing carrying-capacity, food flux, and pelagic-benthic coupling models can contribute to making this determination. It should be noted that suspension culture of bivalves (rope or long-line) presents a special case wherein a portion of the benthic community is introduced into the pelagic environment. In environments where molluscan consumption is substantial, a heirarchy of questions need to be addressed to determine the possible contribution of changes in molluscan feeding to bloom initiation, maintenance, and decline. There are three possible feeding behaviors that may occur when bivalves are presented with a potential HAB species. 1) normal feeding, in which case, there is no change in grazing pressure from bivalves, 2) the pseudofeces response, in which cells are filtered from suspension, but not ingested, and finally released as pseudofeces. This response can have several consequences, including partitioning of phytoplankton biomass from suspension to the benthic boundary layer, possibly leading to hypoxia, partitioning of propagules (cysts, spores, or resting cells), change in the sediment-benthic boundary layer dynamic with respect to partitioning of inorganic and organic nutrients. 3) Elimination of feeding and filtering, from either behavioral changes or mortality of the bivalves. In this last case, benthic grazing pressure can be relaxed temporarily or permanently, and this may be a critical step in bloom initiation. All of these responses have been demonstrated in laboratory feeding studies similar to those described above for bivalve larvae. Essentially all of the experimental biases described for larval feeding studies apply to post-set bivalves as well. However, field observations are again difficult to attribute unequivocally to a particular portion of the phytoplankton counting methods similar to those described for community. Selective feeding can be investigated in laboratory or field manipulation type experiments, with phytoplankton counting methods similar to those described for copepods. Flow cytometric evaluation of grazed and ungrazed phytoplankton assemblages offers some promise of automating the tedious counting process, particularly as species-specific fluorescent tags are developed.

Design of field studies must take into account changes in feeding behavior that accompany any change in algal diet, as well as the possibility that harmful effects may be delayed until some threshold is reached. Molluscan aquaculture provides an opportunity to obtain observations and both water and tissue samples because shellfish farmers regularly check their stocks and generally have expectations of survival and growth rates. One additional approach, identification of a distinctive histological syndrome associated with dinoflagellate feeding, is in development for oysters. Such a biomarker would be useful in both field and laboratory investigations.

Recommendations

Studies of zooplankton population grazing impact should be incorporated into attempts to understand HAB dynamics.

The effects of variations in phytoplankton toxicity upon grazers and their consumers should receive more attention.

Phytoplankton and zooplankton ecologists and phycotoxin chemists should collaborate more closely and extensively to further elucidate the interplay of HAB's and pelagic and benthic food webs.

Evaluate benthic grazing as a phytoplankton production loss term in bloom-prone areas, with a goal of identifying areas with substantial benthic grazing and with little benthic grazing for comparitive field studies.

Continue to explore mechanisms (traditional toxins or other physiological or physical interactions) by which some phytoplankton species, especially dinoflagellates, cause harm in benthic consumers, so that expectations of effects can be generalized rather than needing full evaluation on a species-by-species basis.

Conduct controlled experiments to determine thresholds of cell density or percentage of the phytoplankton assemb lage at which harmful effects occur in both larval and post-set bivalves.

Develop field protocols for bivalve mollusks to test laboratory findings in real-world HAB events.

<u>**Term of Reference 6**</u>: evaluate the role of micro-organic nutrient dynamics and heterotrophic interactions in the initiation and maintenance of HAB

Mixotrophy is the ability of an organism to be both phototrophic and heterotrophic, in the latter case utilizing either organic particles (phagotrophy) or dissolved organic substances (osmotrophy).

The significance of mixotrophy in general phytoplankton ecology is still largely unknown, and even less is known for harmful algal species. In this report, phagotrophy and osmotrophy are considered separately.

Phagotrophy by phytoplankton:

Phagotrophy can be seen as an important factor regulating phytoplankton dynamics. Thus we may ask, for example: What part of grazing on bacteria is due to phagotrophy by phytoplankton? How can we detect phagotrophy? Among which members of the different phytoplankton taxonomic groups is phagotrophy most common? What environmental conditions trigger phagotrophic behaviour? Why do some primarily photosynthetic organisms exhibit phagotrophy, i.e. is there a nutritional advantage? Can phagotrophs outcompete autotrophs and/or heterotrophs? Is phagotrophy an alternative to photosynthesis and uptake of dissolved inorganic or organic substances, to acquire supplementary carbon, macronutrients (N, P, Si), trace elements or special compounds, e.g. vitamins, that the algae cannot synthesise ? Or is phagotrophy only a remnant of an ancient, formerly important behaviour among planktonic organisms, that has little ecological significance at present? These questions remain largely unanswered.

Phagotrophy among algae is presumably regulated through external (and to a certain extent by internal) abiotic or biotic factors that influence cell physiological state. If light is too low to allow for sufficient CO₂ fixation to meet metabolic demands of the cell, phagotrophy can supplement or even substitute for photosynthesis as a source of organic carbon A similar argument may be used to explain phagotrophy with respect to nutrients, i.e., phagotrophy may supply the organism with N or P or some micronutrient if dissolved sources have been exhausted For phagotrophy to be effective, there has to be a sufficient supply of suitable organic particles (prey organisms). Phagotrophy might thus be induced in environments where the organism encounters a high concentration of prey, e.g. bacteria or microflagellates. The three most important triggering/regulating factors for phagotrophy are assumed to be light, nutrient availability, and prey abundance.

It is reasonable to assume that algae use phagotrophy to obtain macro- (N. P) and micro-nutrients (e.g. vitamins), when dissolved inorganic or organic nutrients are growth-limiting. Mixotrophy may be a primitive trait, a notion that is supported by the fact that groups with many phagotrophs are evolutionarily old. Some phagotrophic algae have evolved from primitive heterotrophs whereas others are "secondary" phagotrophs, where the character has evolved from strict phototrophy.

Mixotrophy should not be viewed as a single strategy developed by planktonic organisms, placed between the two extremes of nutrition: autotrophy and heterotrophy. There is probably a continuous gradient between truly autotrophic and heterotrophic organisms. Some organisms will initiate phagotrophy only in the presence of high quantities of prey, whereas for others phagotrophy appears to be more dependent on abiotic factors, such as light. Some algae may be efficient phagotrophs but poor phototrophs, whereas others may be obligate photoautotrophs, still capable of phagotrophy. From an ecological point of view, it is expected that there is a trade-off between phagotrophy and photosynthesis. Both modes of nutrition have certain costs, and the simultaneous ability to perform both has high costs enables the organism to outcompete heterotrophic or strictly photosynthetic competitors under certain environmental conditions.

Phagotrophy in relation to environmental conditions

Light, nutrient availability, and prey concentration may interact in enhancing or suppressing phagotrophy in more or less complicated ways. It is also likely that there is some genetic(i.e. selective) adaptation involved when switching from one mode of nutrition to another. If this is true, then the mode of nutrition may not immediately track changes in environmental conditions, as is the case for physiological changes taking place when a phytoplankter is exposed to different light regimes.

Nitrogen and phosphorus, the most important inorganic nutrients for algal growth, are often in low concentrations in marine waters. Some phytoplankton species that are not good competitors for the limiting nutrient at low concentrations may have retained or developed the capacity to prey on other plankton organisms in order to obtain the required nutrients. The ability to use an alternative nutrient source may enable these species to coexist with, or outcompete, species that rely strictly on dissolved nutrients (Rothhaupt 1996b).

According to Keller *et al.* (1994), phagotrophy by the phytoflagellate *Ochromonas sp.* is used to supplement its nutrition when light or inorganic nutrients are limiting. However, there was an apparent threshold of response, as phagotrophy decreased or even ceased after periods of prolonged darkness. Bird and Kalff (1989) estimated that 50% of the carbon demand by the chrysophyte *Dinobryon sertularia* was met by phagotrophy. Assuming an assimilation efficiency of 60% for carbon (Calow 1977) and close to 100% for nitrogen, Bird and Kalff (1989) estimated that nearly 100% of the N demand by *Dinobryon* could be met by phagotrophy. For the marine red-tide dinoflagellate *Gymnodinium sanguineum*, Bockstahler and Coats (1993 b) calculated that natural populations of this algae in the Chesapeake Bay, were able to balance N requirement by ingesting nanociliates (< 20 μ m).

Little is known about the connection between mixotrophy and toxin production. Okadaic acid (OA) concentrations in cells of *Prorocentrum lima* were higher (11.2-14.2 pg OA cell⁻¹) when organic P (as glycerol-PO₄) was supplied instead of inorganic P (7.5-8.9 pg OA cell⁻¹) (Tomas and Baden 1993).

The ecological significance of phagotrophy

Until recently, phytoplankton were regarded as strictly phototrophic, with a well defined position at the base of the food webs of lakes and oceans. Recently we have learned that the nutritional demands of a growing number of phytoplankton species have the potential to be met at least partly, or under certain environmental conditions, through heterotrophic nutrition (Rothhaupt 1996b). This finding has direct implications for our view of algal survival strategies, and also for the ability of phagotrophic species to out-compete other algae under less favourable growth conditions, e.g. in waters poor in inorganic nutrients or under low light. It also affects the traditional view of the "microbial loop" (Azam *et al.* 1983), where DOC is thought to be channelled from algal photosynthesis to bacteria and then up the food chain through heterotrophic flagellates, ciliates and mesozooplankton. Are phagotrophic phytoplankton that feed on bacteria "taking back" some of the lost (excreted) photosynthetic DOC? How can we estimate the fluxes of carbon and nutrients between different trophic levels in the plankton food web when phagotrophic algae are involved?

Summary- Phagotrophy

Phagotrophy is widespread in certain groups of photosynthetic organisms, including several important toxic or harmful species, such as C. polylepis, P. piscicida, H. akashiwo, A. tamarense, G. galateanum and H. triquetra . Phagotrophy most probably occurs in the Dinophysis species responsible for diarrhetic shellfish poisoning (DSP). Phagotrophic algae have been shown to prey on bacteria, other algae, and even microzooplankton. Phagotrophy may substitute for photosynthesis, and thus may be an alternative way of acquiring reduced carbon. Phagotrophy may also enable the organism to obtain macronutrients (P, N), micronutrients, vitamins or other organic substances that the organism cannot synthesise itself. Both low light and nutrient deficiency have been shown to promote phagotrophy, with grazing rate generally dependent on prey concentration. However, some phagotrophic algae graze independent of light conditions. There is a wide range in degree of mixotrophy, from species that can only supplement their nutrition with phagotrophy to species that are able to grow phagotrophically in complete darkness. Mixotrophic organisms may have a competitive advantage over strict heterotrophs or strict photoautotrophs under specific environmental conditions. However, there is most likely a cost attached to being mixotrophic that makes phagotrophic algae photosynthetically less efficient than obligate autotrophs and less efficient grazers than heterotrophic unicellular organisms. Under certain conditions phagotrophic algae can be the primary bacterivores of the microbial food web. Phagotrophy among algae was "rediscovered" only about 10 years ago, and the ecological significance of this mode of nutrition for the organisms themselves, as well as for the plankton ecosystem, is still poorly known.

OSMOTROPHY or Utilization of Dissolved Organic Matter (DOM) by Phytoplankton,

The ecological importance of phytoplankton utilizing dissolved organic matter as a significant part of their nutrition is still an open question. That some phytoplankton can grow heterotrophically on dissolved organic carbon compounds (DOC) in the dark have been known for some time. Soil extract is known to increase the growth of phytoplankton cultures (e. g. Provasoli et al. 1957). Prakash and Rashid (1968) and Prakash et al. (1973) found that dissolved organic matter in the form of humic substances increased both yield and growth rates of marine dinoflagellates and diatoms. Granéli et al. (1985) suggested that the dinoflagellate *Prorocentrum minimum* was able to use N in humic substances since the biomass yield increased considerably when humic substances and phosphate were added to the medium. Cell N- content of *P. minimum* grown with additions of humic substances were also comparable to cells grown with inorganic nitrogen (Granéli et al. 1985). Organically bound P can be utilized by phytoplankton by the action of alkaline phosphatases (phosphomonoesterases e.g. Berman 1970), but the utilization of organically bound nitrogen other than urea or amino acids is less well known.

Phytoplankton production in most coastal waters is considered to be N-limited. Thus DOM represents a potentially large N pool for phytoplankton in these areas. Moreover, riverine inputs of DON are also a large potential N- source for phytoplankton in coastal waters.

Importance of DOM as a nutrient source

The pool of DOM is composed of a large number of known and unknown components. Only about 50% of the DON has been characterized (Sharp 1983), being dominated by urea and amino acids (free and combined). DON is the dominating component of the flux of N from land to sea, generally comprising 60-90% of the total dissolved N-export (Meybeck 1982). Nitrogen content of riverine humic substances is usually 1-3 % (by weight) and the phosphorus content

is about 0.2% (Hedges 1987). The DON-concentrations in marine waters are generally 3-5 μ mol l⁻¹ (Sharp 1983). Suzuki et al. (1985), however, reported values of 20-40 μ M in surface oceanic waters using a new high-temperature catalytic oxidation method. Since then, Hansell (1993) has measured DON concentrations in nearshore and open-ocean waters consistently lower than 10 μ M also using high-temperature catalytic oxidation. Regardless of the exact amount of DON present in marine waters, the DON pool is usually substantially larger than the pool of inorganic nitrogen.

DON is usually not considered as available for phytoplankton when management of eutrophied coastal areas is discussed. However, several experiments have shown that DON stimulates phytoplankton production. Phytoplankton in a stratified system increased more in particulate N than could be explained by the uptake of inorganic N and urea (Price et al. 1985), presumably by utilization of organic nitrogen compounds. Carlsson et al. (1993) found a stimulation of the microbial food web and increased regeneration of inorganic N which in turn increased phytoplankton production when riverine humic substances was added to a natural plankton community.

Direct utilization of DOM by phytoplankton

High molecular weight polymeric compounds are too large to pass the cytoplasmic membrane by active transport or facilitated diffusion(Payne 1980). Therefore, only the part of the DOM that occurs in low molecular form (e g amino acids) can be taken up directly through the cell membranes with the aid of specific transport enzymes (permeases), without enzymatic degradation (Raven 1980). A mechanism to take up high molecular weight compounds is by pinocytosis, a process only studied to a minor extent in phytoplankton (Klut et al 1989).

Utilization of amino acids not necessarily directly linked to growth have been reported. The red-tide dinoflagellate Gymnodinium breve incorporated several amino acids directly into proteins, during logarithmic growth using inorganic nitrogen as the main N-source (Baden and Mende 1979)

Results by Carlsson et al. (in prep.) show that the toxic dinoflagellate Alexandrium catenella is able to grow well on nitrogen bound to humic substances isolated from river water. *Alexandrium catenella* showed the same growth yield without bacteria or with a natural bacterial community present. Aminopepeptidase activity was negligible in *A. catenella* cultures grown with humic substances without bacteria, but high in the treatment with bacteria. Thus, in the treatments with bacteria, A. catenella might have used amino acids as a N source, cleaved extracellularly by the bacterial aminopeptidases, but the high growth of *A. catenella* in the axenic treatment cannot be explained this way. Perhaps *A. catenella* was able to use the HMW DOM directly, as indicated by the presence of FITC-labelled dextran molecules of 2000 kDa size found in vacuoles of *A. catenella* (Legrand and Carlsson, in prep.).

Three phytoplankton species, among them the toxic prymnesiophyte Prymnesium parvum, have been shown to posses another enzyme: cell-surface L-amino acid oxidases that oxidize amino acids and primary amines to release ammonium that is taken up by the cell (Palenik and Morel 1990).

Indirect utilization of DOM by phytoplankton - the role of bacteria and regeneration of inorganic nutrients

The grazing activity of heterotrophic flagellates on bacteria, regenerates inorganic nutrients. The regenerated inorganic N and P are then available both for bacteria and phytoplankton uptake. A transfer of nutrients from organic to inorganic form occurs in this way.

Bacteria are considered to be the main organisms using DOM as substrate. Since the bacterial C:N ratio is lower than for phytoplankton (about 3-7, Nagata 1986), they need more N per unit biomass than phytoplankton. DOM in marine waters has a high C:N ratio, about 15 (by weight) (Benner et al. 1992), while riverine DOM that enters coastal waters has an even higher C:N ratio (about 50) (Malcolm 1985). When bacteria are grazed by heterotrophs such as heterotrophic nanoflagellates and ciliates, inorganic N is released. The amount of N that is regenerated has been estimated to be between 10-50% of the bacterial N ingested (Andersson et al. 1985) and it will be ultimately available for phytoplankton. The rest of the N is retained in the heterothrophic flagellate biomass and is thus available for higher trophic levels (Azam et al. 1983).

Possible connections between terrestrial supply of DOM and occurrence of red tides

Prakash and Rashid (1968) suggested that most, if not all, dinoflagellate blooms in coastal waters are connected to humic or other nutritional factors entering the waters after heavy rainfalls or land drainage. Baden and Mende (1979) found that Gymnodinium breve utilized amino acids even when inorganic N sources were present and speculated that even if amino acid concentrations in marine environments usually are low, the amounts of free amino acids can become substantial in areas of low water turnover and high productivity, areas which coincidentally are sites for red tide outbreaks. Granéli and Moreira (1990) have shown that rivers draining from agricultural soil and thereby rich in

inorganic N and P, stimulate growth of diatoms, whereas river water from forest areas, rich in humic substances, increases growth of dinoflagellates. Coastal waters can be heavily influenced by river runoff, containing both inorganic and organic nutrients. Especially in coastal areas influenced by rivers draining forested land, the contribution of organically bound N can be very high. A large increase of the discharge of humic substances by rivers into Swedish coastal waters the last 20 years (Andersson et al. 1991, Forsberg 1992) coincides with increased number and intensity of dinoflagellate blooms in the same area (Granéli et al. 1989).

Conclusions -DOM utilization

Substantial experimental evidence shows that phytoplankton do not use DOM as a carbon source in natural environments, except perhaps during extreme conditions. It is, however, evident that many phytoplankton species benefit from organic substances present in seawater or reaching coastal zones with rivers. The growth stimulating effect is caused either by trace metal complexation by the organic molecules, but also by direct utilization of organic bound N occurring in small molecules such as urea and amino acids. Indirect utilization of organically bound N via remineralization by heterotrophic grazers cropping bacteria that previously used the DOM as a substrate is also a mechanism for phytoplankton utilization of N in DOM. Direct ingestion of high-molecular weight organic molecules might also be a mechanism so far overlooked.

DOM in river water entering coastal waters are subjected to two major breakdown processes: bacterial degradation (Carlsson et al. 1993) and photochemical transformations that increase their availability to bacteria (Kieber et al. 1989). In coastal waters influenced by river runoff, increased growth of phytoplankton, including blooms of harmful algal species might therefore occur as an effect of indirect utilization of N previously bound to DOM.

1. Mixotrophy by HAB species

Justification: It is now clear that many HAB species have the potential for mixotrophic nutrition. The ability to utilize both photosynthetic and heterotrophic pathways may give species of mixotrophic HAB a competitive advantage over strictly autotrophic members of the phytoplankton. Thus, we cannot understand bloom dynamics, the effects of eutrophication on plankton communities, or life cycle dynamics of key HAB taxa without thoroughly understanding the forces driving mixotrophy and the extent to which autotrophic processes are supplemented by heterotrophy.

Recommendations:

- Develop reliable methods to detect and quantify mixotrophic nutrition
- Determine the mixotrophic potential of key HAB species
- Identify factors that induce or enhance phagotrophy in photosynthetic organisms
- Investigate the effects of natural and anthropogenic eutrophication on HAB mixotrophy
- Evaluate the effects of mixotrophic nutrition on toxicity of HAB species
- Assess the significance of mixotrophic HAB on the structure and function of marine food webs
- There is an urgent need to know the impact of mixotrophy, if realistic mathematical models of nutrient transfer within the microbial loop are to be achieved.

<u>**Term of Reference 8**</u>: review the status of development of taxonomic coding systems with a view to recommending the adoption of a single coding system for use in ICES

The WGHABD recognizes the need to develop a single taxonomic coding system for phytoplankton. Nevertheless, the taxonomic expertise in the WGHABD is not broad enough for the Group to address the question on its own. In order to draw on a broader expertise, it is therefore recommended that any decisions on adopting an existing system or developing a new one be carried out in association with the ICES Working Group on Marine Data Management, the Working Group on Phytoplankton Ecology and the WGHABD. It is recommended that the deliberations of the workshop on taxonomic nomenclature at the 8th International Conference on Harmful Algae, Vigo, Spain, June 1997, be taken into consideration.

Term of Reference 9: prepare plans for a joint meeting with the Working Group on Phytoplankton Ecology in 1998

WG members, in general, felt that joint meetings with WGPE should not be held on a regular basis. However, it was felt that a joint meeting with WGPE would be necessary in 1998, in order to review the report on the Kristineberg workshop on growth rate measurements and review the status of taxonomic coding in relation to the establishment of a phytoplankton database. Moreover, it would be of interest for the WGHABD members to be informed of the latest developments in phytoplankton ecology. A joint meeting of one day when specific tasks would be benefit from a joint expertise, would be enable to produce detailed and pertinent answers.

<u>Term of Reference 10:</u> consider the design of an experiment to elucidate the role of physical-biological interactions in harmful blooms in ICES area

Several specific cases of harmful events were discussed to develop consensus on how to approach the problem. Harmful Algae are found in the complicated coastal ocean environment where physical processes have both short time and space scales. The large concentrations found in many blooms suggest that both net population growth and motion of the particles relative to the local water (due to behavioral characteristics and/or density differences) are fundamental issues. To identify and understand the interactions between the physical field and the different biological species, it is necessary to focus on specific locations and species. This presents an impediment to progress because the detailed information about the physical and biological fields is often very difficult to obtain with sufficient temporal and spatial resolution. To facilitate progress in this area the sub-group recommended compiling the scenarios for the various harmful events in the ICES area in order to facilitate the communication between the disciplines and identification of information gaps, sampling problems,... These scenarios should identify the salient features of the population history to enable modeling of bloom initiation, the effects of growth, grazing, behavior, advection as well as environmental fluctuations on seasonal cycles as well as random events. These informations will be compiled by P. Gentien and T. Osborn during the intersessional period from contributions from identified experts. Functional grouping of species may be possible on the basis of these scenarios.

Development of the instrumentation capable of high resolution and detailed measurement of physical parameters led to the development of the fields of oceanic finestructure (>1 m) and microstructure (< 1 m), the latter being the study of small-scale turbulence in the ocean. Similar techniques for detailed sampling of the biological and chemical fields have been much more difficult and slower in coming. Reports on the development of a particle size and concentration instrument show significant detail in the particle fields. When combined with taxonomic information one gets the strong impression that the particle dynamics play a significant role in their population dynamics for such concentrating of biological populations enhance their growth and survival. The historic methods of sampling give the wrong picture of the distributions. Rates and effects are greater than mean values due to concentration into layers. Understanding how these thin layers of particles develop, what is their relation to the physical fields of temperature, salinity, and density, and what physical processes are involved in forming these biological and chemical layers is necessary for quantitative explanation of physical-biological interactions.

5 PROPOSED TERMS OF REFERENCE FOR THE 1998 WGHABD

§1. collate and assess National Reports and update the mapping of HABs;

§2. Prepare a review document on the population scenarios for the different Harmful Algae species in the ICES area

§3. examine the population dynamics and assess the role of harmful benthic microalgae in benthic and pelagic food webs;

§4. review strategies that could be used to control harmful algae, identify the systems where bloom control may be possible and highlight promising methods which require further research in order to reduce the extent and effects of HABs.

Justification for §2.

The objective is to produce descriptions of the population life histories for each specific region and species of interest in their oceanographic context. This information will be used as the basis of communication between physical oceanographers and the physiological ecologists for modeling work. While physicists are trained to simplify problems by neglecting details in order to make models that explain the features of the system, biologists must examine and

identify the details that separate species. A joint description of the basic systems will form a common base for discussion and modeling.

These case descriptions will include details of algal life histories, physical processes, and interactions with other organisms. These scenarios should identify the salient features of the population history to enable modeling of bloom initiation, the effects of growth, grazing, behavior, advection as well as environmental fluctuations on seasonal cycles as well as random events. To be compiled by P. Gentien and T. Osborn during the intersessional period from contributions of identified experts

Justification for § 3.

Benthic harmful microalgae are an important source of phycotoxins transfered through benthic and pelagic food webs. The WGHABD has not previously addressed this problem, since studies on HAB dynamics usually focus on events and processes in the pelagic domain, where stratification can contribute to bloom aggregation. Biomass and growth rate estimates for toxigenic benthic species (e.g. *Prorocentrum* spp., responsible for some D.S.P. outbreaks) are often considered difficult to ascertain because growth is spatially heterogeneous (in « patches »). Nevertheless, the fact that these populations are relatively stationary may yield advantages to studying growth rates, nutrient dynamics, susceptibility to grazing, allelopathic interactions and microscale processes. Thus, whilst benthic harmful microalgae warrant special attention, the results may prove highly relevant for interpreting similar mechanisms for pelagic blooms.

Justification for §4.

Social pressures are increasing for scientists to do more than study the fundamental ecology and oceanography of HABs. The statement is often made that we are studying the problems, not the solutions. It is indeed true that an increase in understanding underlying mechanisms is necessary if strategies, such as prediction or nutrient loading reduction are to be attempted, but direct manipulation of bloom populations should also be considered. There are many possible control strategies that can be evaluated, ranging from biological (introduction of parasites, viruses, pathogens) to mechanical (e.g. clay flocculation). We recognize that many HABs occur on such large scales and in such dynamic hydrographic systems that control is not feasible. There are, however, localized HABs or blooms that have initiation zones where preventative measures might reduce subsequent bloom density and extent. Thus, there is value in identifying ecosystems where bloom control may be possible. Care must be taken to emphasize that we are not promising success, but only reccomending practical lines of applied research that have thus far been ignored in pursuit of fundamental scientific understanding of bloom dynamics.

6 PROPOSED TERMS OF REFERENCE FOR THE 1998 JOINT MEETING OF WGHABD AND WGPE

§1. Review the results of the Workshop on development of *in situ* Growth Rate Measurements of dinoflagellates held in Kristineberg, 1996

§2. Review the status of taxonomic coding systems with a view to recommend the adoption of a single coding system for use in ICES.

Justification for §1.

The Kristineberg workshop which was designed and developed by the Working Group, was held in late 1996 in Kristineberg, Sweden. the final report will be produced in early spring of 1998 and evaluated together with the WGPE.

Justification for §2.

The WGHABD recognizes the need to develop a single taxonomic coding system for phytoplankton. Nevertheless, the taxonomic expertise in the WGHABD is not broad enough for the Group to address the question on its own. In order to draw on a broader expertise, it is therefore recommended that any decisions on adopting an existing system or developing a new one be carried out in association with the ICES Working Group on Marine Data Management, the Working Group on Phytoplankton Ecology and the WGHABD. It is recommended that the deliberations of the workshop on taxonomic nomenclature at the 8th International Conference on Harmful Algae, Vigo, Spain, June 1997, be taken into consideration.

ANNEX I

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ANNEX II

NATIONAL REPORTS

Status of HAB's for Danish waters in 1996

Compiled by Dr. Per Andersen. Bio/consult as, Johs. Ewaldsvej 42-44, 8230 Aabyhøj, Denmark. phone. 45 86 25 18 11, fax. 45 86 25 81 73, E-mail: bioconjp@inet.uni-c.dk.

The phytoplankton in Danish coastal waters and fjords in 1996 was characterized by relatively low concentrations and biomasses in the spring and most of the summer period. During summer the biomasses were dominated by diatoms e.g. *Skeletonema costatum, Rhizosolenia fragilissima* and the dinoflagellate *Prorocentrum minimum* which bloomed on several locations. The low biomasses during the summer period were primarely the result of low input of inorganic nutrients from land, caused by low run-off during spring and summer.

The following toxic and potentially toxic algae were registred in high concentrations:

Dinoflagellates

Dinophysis acuminata, Prorocentrum minimum, Prorocentrum micans

where as the following toxic and potentially toxic algae were registred in low concentrations:

Dinoflagellates

Alexandrium ostenfeldii, Alexandrium tamarense, Dinophysis norvegica, Dinophysis acuta, Dinophysis rotundata, Prorocentrum lima, Gyrodinium aureolum, Gymnodinium sanguineum, Noctiluca scintillans

Diatoms

Pseudonitzschia delicatissima-group, Pseudonitzschia seriata-group

Others

Dictyocha speculum (-"Si-skeleton"), Phaeocystis pouchetii, Chrysochromulina spp., Nodularia spumigena, Aphanizomenon flos-aquae.

No fishkills caused by HAB's were registred in 1996.

Intensified monitoring and/or closing of shellfishery due to high concentrations of *Dinophysis acuminata*, *Dinophysis norvegica*, *Alexandrium* species and *Pseudo-nitzschia*-species were imposed at several occasions in areas at the east coast of Jutland, in the Limfjorden as well as in the Wadden Sea. DSP-toxins were registred in shellfish at the east coast of Jutland in january/february in a situation with very low concentrations of *Dinophysis* spp. (<100 cells/L). Mussel fisheries were stopped in the area. There is no reports of human intoxications caused by consumption of danish shellfish during 1996.

PSP and ASP were not registred in 1996.

An exceptional local bloom of *Dinophysis acuminata* (max. conc. 11×10^3 cells/L) was recorded in October in Præstø Fjord. It occurred together with the bluegreens *Nodularia spumigena* and *Aphanizomenon flos-aquae*. No harmful effect were registred in connexion with the bloom.

Harmful algal blooms in FINLAND in 1996

TOXIC CYANOBACTERIA

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Location	Northern Baltic Proper and the Gulf of Finland
Dates Augus	t
Effects No de	tectable effects have been reported
Management	Warning and information in mass media
Causative speciesNodul	aria spumigena and Aphanizomenon flos-aquae
Environment	mainly open sea, temp over 16 oC
Advected population	??
Previous occurrence	a yearly phenomenon
Additional comments	the bloom was 2-3 weeks later than usually, due to the exceptionally cold weather
	in early summer
Individual to contact	Eija Rantajärvi
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00931	Helsinki
Finlar	d
tel +3	58 9 613941
email	rantajarvi @ fimr. fi
HETEROCAPSA TRIQUET	RA RED TIDE
Location	western Gulf of Finland

Docution		
Dates	July	
Effects	-	
Management		-
Causative specie	esHetero	capsa triquetra
Environment		open and coastal sea, dense populations colouring the water reddish brown
Advected popul	ation	-
Previous occurre	ence	mid 1970s
Individual to contact		Eija Rantajärvi
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	email :	rantajarvi@ fimr. fi

PRESENCE OF OA

Loacation		Western Gulf of Finland
Dates	August	
Effects	??	
Management		-
Causative specie	sDinoph	ysis acuminata (?), D. norvegica (?)
Environment		the upper 0-15 m, typically forming maxima occurring in the deeper part
Advected popula	ation	
Previous occurre	ence	Dinophysis spp. occur regularly in the Baltic Sea. This is the first observation of
		OA in the Baltic Sea - not studied before
Individual to con	ntact	Harri Kankaanpää
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	tel +358	3 9 613941
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National Report for Ireland, 1996.

Phytoplankton

The 1996 results were notable for the very low numbers of *Dinophysis acuminata* and *Dinophysis acuta* present in the samples compared with previous years. Both species were typically detected at levels in the range 40 - 200 cells/litre. The maximum cell count of *Dinophysis acuminata* was 600 cells/litre in Roaring Water Bay on 3 September and the maximum cell count of *Dinophysis acuta* was 1160 cells/litre in Bantry Bay on 28 August.

Alexandrium tamerense, with a maximum cell count of 875,000 cells/litre was recorded at in Cork Harbour in July.

In May an extensive bloom of *Phaeocystis spp*, tentatively identified as *Phaeocystis globosa*, was recorded along the east coast from Dublin to Clogher Head.

Shellfish Toxicity

DSP

In the south and southwest positive bioassays results were only obtained on one occasion in early September in mussel samples from inner Bantry Bay, Kenmare Bay and Cork Harbour. Harvesting of shellfish from these locations was prohibited for a period of 2 weeks. No positive results were obtained from the other shellfish growing areas. This contrasts markedly with typical closure periods of shellfish production areas of 8 - 10 weeks in other years and is in marked contrast to the closure period enforced in the southwest in the of 1994/95 which lasted for up to 10 months.

PSP

In July, a bloom of *Alexandrium tamerense* was observed during routine analysis of water samples from Cork Harbour. Toxins were detected, at a concentration above the threshold for human consumption, in mussel samples and harvesting of all shellfish from the area was prohibited for a period of 2 weeks. Chemical analysis of samples showed that the main toxins present were gonyautoxin2 (GTX2) and gonyautoxin3 (GTX3). This was the first time since 1987 that PSP toxins were detected in shellfish from Ireland.

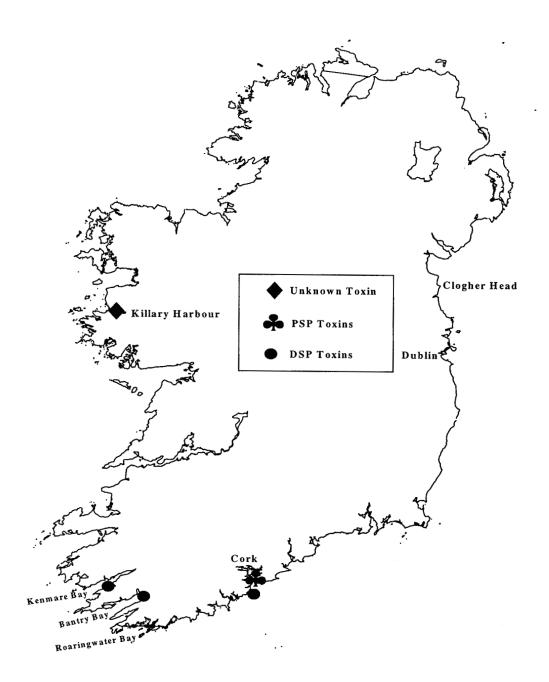
"Unexplained toxicity"

As reported last year, unexplained toxicity was detected in mussels from Killary Harbour. Toxicity, as measured by both DSP rat and mouse bioassays, persisted from November 1995 through to June 1996. Throughout this period no known toxic phytoplankton species were observed in water samples. Harvesting of all shellfish from Killary Harbour was prohibited from November 1995 through to June 1996. A previously unknown toxin has been isolated by Prof. Yasumoto in Japan. and work is ongoing to characterise the toxin. As yet the origin of the toxin is unknown.

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HARMFUL ALGAL BLOOMS IN NORWAY 1996

Paralytic Shellfish Toxins

In 1992 a regular monitoring of algae, in 1996 at 24 stations, and control of shellfish toxicity by mouse bioassay along the Norwegian coast were established. The results from this monitoring programme concerning Paralytic Shellfish Toxins in 1996 are summarised.

	Alexandrium spp. occurred almost all along the coast, but most numerous along the ast. Toxins were only recorded at single stations along the west coast up to y. Most frequently the levels were low, 200-400 ME.
DATES station	April - September 1996. 16000 ME/100g were recorded at on the north-west coast in May, and 2100 MU on a in northern Norway in July.
EFFECTS	Toxins recorded above the action level (400 ME/100g) according to mouse bioassay.
MANAGEMENT DECISIONS	Harvesting was locally banned. The public was warned against picking toxic mussels.
CAUSATIVE SPECIES	Alexandrium spp.
ENVIRONMENT	No information
ADVECTED POPULATION	Mainly due to in situ growth ?
PREVIOUS OCCURRENCES year to	A few historical records, and more or less regular occurrences along the west coast the recent years, however, the spatial and temporal extent may vary significantly from one another.
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Prymnesium spp.

LOCATION	Ryfylke, Sandsfjord system, the westcoast of Norway.
DATES	July-August 1996
EFFECTS	No
MANAGEMENT DECISIONS	Monitoring.
CAUSATIVE	Prymnesium parvum/patteliferum
ENVIRONMENT	Brackish water.
ADVECTED POPULATION	Mainly in situ growth.
PREVIOUS OCCURRENCE	Yearly blooms since 1989
ADDITIONAL COMMENTS	

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Diarrhoeic Shellfish Toxins

In 1992 a regular monitoring of algae, in 1996 at 24 stations, and control of shellfish toxicity by mouse bioassay along the Norwegian coast, were established. The 1996 results from this programme concerning Diarrhetic Shellfish Toxins are summarised.

LOCATION	Dinophysis spp. were recorded all along the Norwegian coast but most numerous along the south coast and in the innermost part of the Sognefjord at the west coast.
DATES	Toxins in mussels were recorded only at one station in mid-Norway in July.
EFFECTS	Toxins recorded above the action level according to mouse bioassay.
MANAGEMENT DECISIONS	Harvesting was locally banned. The public was warned against picking toxic mussels.
CAUSATIVE SPECIES	Most probably Dinophysis spp., with D. acuminata and D. acuta as the most potent species.
ENVIRONMENT	The problem occur over a wide range of temperatures and salinities.
ADVECTED POPULATION	Along the southern coast there are some evidence that the algae and toxin problems are spread by advection. But along the west coast the "hot spots" seems to be rather patchy which indicate local concentration of the algae and/or in situ growth.
PREVIOUS OCCURRENCES	A few more dubious historical records. A yearly, more or less large scale and long lasting phenomenon since 1984 according to mouse bioassay. The phenomenon has never been so extensively monitored as since 1992.
INDIVIDUAL TO CONTACT	Einar Dahl, Institute of Marine Research, Research Station Fl¿devigen, N-4817 His, NORWAY tel. +47 370 59000, fax. +47 370 59001 E.mail: einar.dahl@imr.no

DSP

DSP toxins were detected at Aveiro, Minho, Lima, Mondego estuaries and Óbidos lagoon; positive bioassays at Sagres were negative with HPLC, nevertheless mussel harvest was closed.

1. and 2. Location and data of occurrences

- Minho estuary: January 1 March 11; May 21 June 17
- Lima estuary: January 1 March 11; May 21 July 22
- Aveiro Lagoon: June 19 September 23; October7 21
- Mondego estuary: July 1 September 23
- Óbidos Lagoon: May 20 September16
- Ericeira: February 26 March 19; June 24 July 22
- Albufeira Lagoon: February 26 March 11; June 25 July 1
- Sagres: July 2 September 18; September 23 October 28 and December 6 -31.

3. Effects:

Mostly mussels (*Mytilus edulis*) from these regions presented DSP toxins, in the case of the mussels from Sagres we could not find DSP toxins by HPLC analysis, but the mice died during the first 24 hours after injection, so the harvest was closed in the region.

DSP toxins were determined both by the mouse bioassay and through HPLC.

4. Management decisions:

Harvest of affected species closed during toxication.

5. Causative species:

Dinophysis cf. acuminata

The highest detected concentrations (cells/l) were:

- Minho estuary and litoral North: D. cf. acuminata 750 (June 19)
- Aveiro Lagoon: D. cf. acuminata 4207 (June 24)
- Mondego estuary: D. cf. acuminata 500 (June 24)
- Obidos Lagoon D. cf. acuminata 833 (june 19)

6. Environment: Temperature range: 16° - 19°C Salinity range: 24 - 36⁰/₀₀

7. Advected population or in situ growth: Most probably a combination of both.

8. Previous occurrences:

Since 1987, the first year of confirmed occurrence, the problem has occurred every year, with a break in 1993. This year the most affected areas were Minho estuary, Aveiro Lagoon, Mondego estuary, Óbidos Lagoon and Sagres.

9. Individual to contact:

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PORTUGAL 1996

PSP

Only Mondêgo Estuary, Algarve coast and Formosa Lagoon were affected with values over 80 µg/100g.

1. and 2.- Location and areas of occurrence:

- Mondêgo Estuary (Figueira da Foz): January 9.
- Algarve coast (Sagres): September 25 October 15..
- Algarve coast (Faro/Vila Moura): August 29; September 17 24.
- Formosa Lagoon (Tavira): September 30 October 3.

3. Effects:

Only some mollusc bivalve species from the affected areas presented PSP toxins over $80 \mu g/100g$ (highest detected values):

- Mondêgo Estuary (Figueira da Foz): Scrobicularia plana 128.6 µg/100g (January 9)
- Algarve coast (Sagres): Mytilus edulis 178 µg/100g (October 2).
- Algarve coast (Faro/Vila Moura /Olhão): Venus gallina 234 μg/100g (August 29) and Donax spp 431 μg/100g (September 17)
- Formosa Lagoon (Tavira): Ruditapes decussatus 90.8 µg/100g (September 30)

4. Management decisions:

Bivalve species with PSP values over $80 \,\mu g/100g$ closed to harvest.

5. Causative species:

The causative species was Gymnodinium catenatum.

The highest detected concentrations (cells/l) were:

- Mondêgo Estuary: 416 Empty cysts and 100 cysts (January 9)
- Algarve coast (Sagres/Lagos): 400 (October 1)
- Algarve coast (Faro/Vila Moura /Olhão): 2083 (September 12)
- Formosa Lagoon: 1053 (September 12)

6. Environment: Temperature range: 17° - 20 °C Salinity range: 34 - 37⁰/₀₀

7. Advected population or in situ growth: A combination of both

8. Previous occurrences:

Since 1986, with a break in 1991, *G. catenatum* has been the responsible species for PSP at the Portuguese coastal zone. In 1993 and 1994 all the coast has been affected beginning in the South and spreading to the North. In 1995 the main affected area was Algarve coast, in an extensive way, covering all litoral, sea Lagoons and Estuaries. This year only very restricted areas and only some bivalve species at Mondêgo Estuary, Algarve coast and Formosa Lagoon were affected for a short time.

9. Individual to contact:

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PORTUGAL 1996

ASP

Domoic acid was detected in very small amounts < $20 \ \mu g/g$ in almost every bivalve species all around the portuguese coast for short periods scattered in time and coincident with the occurrence of *Pseudo-Nitzschia* spp mainly *P. australis* in concentrations below 100 000 cell/l.

7. Advected population or in situ growth:

Most probably a combination of both.

8. Previous occurrences:

The first detected occurrence of Domoic acid in bivalves over 20 μ g/g was in smooth callista (*Callista chione*) in 1995 as reported.

9. Individual to contact:

Maria Antónia de M. Sampayo and Maria da Graça Vilarinho IPIMAR Av. Brasília 1400 Lisboa PORTUGAL Phone: 351 1 3017361 Fax: 351 1 3015948

PORTUGAL 1996

Red Tides Without Harmful Effect

1. and 2. Location and data of occurrences:

- Litoral Figueira da Foz. March 17
- Obidos Lagoon: April 16; May 29 and July 30
- Formosa Lagoon (Algarve coast): June 14 20
- Peniche Bay: May 21, June 25; July 2 and 24
- Litoral Setúbal Sines: Sept. 26 Oct. 19

3. Effects: Water discoloration

4. Management decisions:

5. Causative species (highest detected concentrations in cells/l) were:

- Litoral Figueira da Foz, March 17: Mesodinium rubrum (0.8×10⁶)

- Obidos Lagoon, April 16; May 29 and July 30: Skeletnema costatum and Thalassiosira lavenderii (2.3×10⁶)

- Formosa Lagoon, June 14 20: S. costatum, T. lavenderii and Glenodinium foliaceum (3.53×10⁶)
- Peniche Bay, May 21, June 25; July 2 and 24: T. lavenderii and Chaetoceros socialis (5.97×10⁶)
- Litoral Setúbal Sines, Sept. 26 Oct. 19: Lingulodinium polyedricum (0.72×10⁶); Prorocentrum micans (0.87×10⁶)

6. Environment:

- Litoral Figueira da Foz. March 17: Temperature range: 15 - 16°C Salinity range: 36%0

- Obidos Lagoon, April 16; May 29 and July 30: Temperature range: 15 - 22°C

Salinity range: 34 - 37%o

- Formosa Lagoon, June 14 - 20: Temperature range: 24 - 26 °C Salinity range 36.5 - 37.6 % o

- Peniche Bay, May 21, June 25; July 2 and 24: Temperature range: 16 - 18.5°C Salinity range: 36 %o

- Litoral Setúbal - Sines, Sept. 26 - Oct. 19: Temperature range: 17 - 18°C Salinity range: 36 %o

7. Advected population or in situ growth:

Most probably a combination of both at Figueira da Foz litoral, Peniche Bay and Setúbal/Sines litoral, *in situ* growth at Óbidos and Formosa Lagoons.

9. Individual to contact:

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PORTUGAL 1996

Fish and /or fauna Mortalities

1. and 2.- Location and areas of occurrence:

- Albufeira Lagoon: Feb. 19 - May 27; August 5 - 20 and December 11

- Fish ponds at Fuzeta (Formosa Lagoon) Algarve coast: August 5 - 13

- Lagos - Salgada Lagoon : August 19

3. Effects: Mussels and/or fish mortalities

4. Management decisions:

In the case of Albufeira Lagoon to intensify water flush by dredging the channel to the sea. In the case of fish ponds to change the water in the affected ponds as fast as possible

5. Causative species (highest detected concentrations in cells/l) were:

- Albufeira Lagoon: small flagellates, Sekeletonema costatum, Thalassiosira pseudonana, Chaetoceros calcitrans,

Gymnodinium sp (73.9×10^6)

- Fuzeta fish ponds: total phytoplankton: 17.5×10^6 (Heterosigma inlandica 3.3×10^6)

- Espiche: total phytoplankton 86×10^6 (small flagellates and *Chaetoceros calcitrans*)

6. Environment:

- Albufeira Lagoon, Feb. 19 - May 27; August 5 - 20 and December 11: Temperature range: 14 - 19°C Salinity range: 17 - 34.5 %o sub surface oxygen depletion - Fuzeta fish ponds, 5 - 13 August: Temperature range: 22 - 28°C Salinity range: 37.8 - 40 %o Oxygen: anoxia at critical time (05 - 08hours) - Lagos - Salgada Lagoon , August 19: Temperature: 23- 25°C

Salinity: 28 %o

7. Advected population or in situ growth:

- in situ growth at Albufeira and Salgada Lagoons

- advected seed population with high in situ growth in the case of Fuzeta fish ponds.

8. Previous occurrences:

- at Albufeira Lagoon some years when the opening to the sea closes there are eutrophization.

- some years ago also in aquaculture ponds at Algarve we could relate fauna mortalities with the occurrence of Heterosigma inlandica.

9. Individual to contact:

Maria Antónia de M. Sampayo and Maria da Graça Vilarinho **IPIMAR** Av. Brasília 1400 Lisboa PORTUGAL Phone: 351 1 3017361 Fax: 351 1 3015948

1. Locations: East coast of Scotland a) south east coast b) north east coast

2. Dates of Occurrences: a) late April/early May 1996b) late April to early July 1996

3. Effects: a) PSP in mussels at one site (192 g/100g) and one scallop sample from offshore (23 g/100g).

b) PSP in mussels in Dornoch Firth, maximum level 976 g/100g on May 13. Declined in June and not detected by early July. DSP detected in same area during July. PSP toxins in offshore scallops to maximal level of 232 g/100g from April to July.

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4. Management Decisions:

b) Voluntary Closure Agreements (VCA) of mussel beds for PSP from May to early July and during July for DSP in Dornoch Firth.

5. Causative Species: Dornoch Firth - Alexandrium spp. detected from May to July up to 1100 cells/litre. Dinophysis spp. from May to September, max. concentration 2500

6. Environment:

7. Advected Population or in-situ growth: not known

8. Previous Occurences: PSP has recurred on the Scottish east coast since 1990, although in different locations. DSP has not previously been detected in this region.

9. Additional Comments:

 Individual to Contact: Godfrey Howard/Elspeth Macdonald SOAEFD Marine Laboratory PO Box 101, Victoria Road Aberdeen AB11 9DB, UK

1. Locations: Orkney Islands

2. Dates of Occurrences: 23 April to 23 September

3. Effects: PSP in mussels (max. 555 g/100g) mostly in Scapa Flow, Kirkwall Bay and Wide Firth. Other species affected included cockles, scallops, queen scallops and razor fish. Low levels (up to 27 μg/100g) in velvet crabs. Scallops from east of Orkney contained up to 640 μg/100g.

4. Management Decisions: FEPA 1985 closure order imposed on 17/07/96 and revoked on 23/09/96 for offshore scallop gonads.

5. Causative Species: Alexandrium spp. observed at two sites in Orkney until July with highest levels found in May (2000 cells/litre) at Scapa Flow.

6. Environment:

7. Advected Population or in-situ growth: Not known

8. Previous Occurences: PSP has regularly occurred in Orkney since 1990.

9. Additional Comments: Low levels of PSP detected in scallop gonads during winter 1995-96.

 Individual to Contact: Godfrey Howard/Elspeth Macdonald SOAEFD Marine Laboratory PO Box 101, Victoria Road Aberdeen AB11 9DB, UK

- 1. Locations: Shetland Islands
- 2. Dates of Occurrences: Early June to early September
- 3. Effects: PSP first detected on 05/06/96, peaked on 15/07/96 (1108 μg/100g). Toxins still detectable in SW Shetland until early September.
 ASP Traces of domoic acid detected by HPLC during June.
- 4. Management Decisions:
- 5. Causative Species: Alexandrium spp. observed from April to August, up to 3600 cells/litre during June. Pseudonitzschia spp. exceeded 3 x 106 cells/litre in July.
- 6. Environment:
- 7. Advected Population or in-situ growth: Not known
- 8. Previous Occurences: PSP previously detected in Shetland, but always below the action level. First record of domoic acid in Scottish waters.
- 9. Additional Comments: Cyst survey during winter found relatively high numbers of Alexandrium tamarense cysts in Shetland sediments.
- 10. Individual to Contact: Godfrey Howard/Elspeth Macdonald SOAEFD Marine Laboratory PO Box 101, Victoria Road Aberdeen AB11 9DB, UK

- 1. Locations: West Coast of Scotland
- 2. Dates of Occurrences: Late April to late July.
- 3. Effects: PSP detected sporadically in mussels from NW coast, peaked in late July (771 μg/100g), but mostly < 200 μg/100g. On central west coast, levels reached 118 μg/100g on 07/07/96.
 DSP short-lived events in two areas no action required.
- 4. Management Decisions: Voluntary Closure Agreement (VCA) agreed for NW coast and central west coast as required in collaboration with shellfish farmers and Environmental Health Officers.
- Causative Species: Alexandrium found sporadically over the west coast from March to September up to 1600 cells/litre, but mostly < 500 cells/litre. Dinophysis commonly found from April to October.
- 6. Environment:
- 7. Advected Population or in-situ growth: Not known
- 8. Previous Occurences: PSP has regularly occurred on the west coast since 1990.
- 9. Additional Comments:
- 10. Individual to Contact: Godfrey Howard/Elspeth Macdonald SOAEFD Marine Laboratory PO Box 101, Victoria Road Aberdeen AB11 9DB, UK

- 1. Locations: West coast and Orkney Islands
- 2. Dates of Occurrences: August
- 3. Effects: Widespread kills of littoral organisms, particularly cockles, razor fish and annelid worms. Some farmed salmonid mortalities attributed to anoxia following bloom decline.
- 4. Management Decisions:
- 5. Causative Species: Extensive bloom of Gymnodinium cf. mikimotoi, up to 3 x 106 cells/litre.
- 6. Environment: Widespread region affected, so not known.
- 7. Advected Population or in-situ growth: Thought to be advected population.

8. Previous Occurences:

9. Additional Comments:

10. Individual to Contact: Marie Kelly/Elspeth Macdonald SOAEFD Marine Laboratory PO Box 101, Victoria Road Aberdeen AB11 9DB, UK

HARMFUL ALGAL BLOOMS IN GALICIA IN 1996

1.- <u>Location</u>: Punctual areas in the Galician Rias.

2.- Date of Occurrence:

Sporadic, very localized and short lasting (maximum, 12 days) closures througout the year.

3.- Effects: DSP toxicity in the bivalves.

4.- Management Decision: Harvesting is banned when DSP toxin is present.

5.- <u>Causative Species</u>: *Dinophysis spp.* in concentrations below 1000 cells $.1^{-1}$.

6.- Environment: Very variable.

7.- Advected Population or In situ Growth: Probably "in situ" growth.

8.- Previous Ocurrences: Very low intensity and short lasting DSP episodes compared with

previous years.

(Pecten maximum)

9.- <u>Additional Comments</u>: As a consequence of the 1995 *Pseudo-nitzschia* episodes, scallops remained toxic with low levels of ASP toxins.

10.- Individual to Contact:

J. Mariño; J. Maneiro; Y. Pazos; C. Salgado; M. Bermúdez; F. Arevalo. Centro de Control da Calidade do Medio Mariño. Peirao de Vilaxoán s/n 36611 Vilagarcía de Arousa Pontevedra. España.

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HARMFUL ALGAL BLOOMS IN CATALUÑA IN 1996

1.- Location: Harbour of Arenys de Mar

2.- Date of Occurrence: May and June 1996.

3.- Effects: Red water discoloration in the harbor, max. dinoflagellate concentration 2×10^7 cells/l

4.- Management Decision: Sampling twice a week the harbour and adjacent waters.

5.- Causative Species: Alexandrium minutum Halim.

6.- Environment: Calm weather, temperature of 16-20 °C, salinity 26-38 psu.

7.- Advected Population or In situ Growth: In situ growth.

8.- <u>Previous Ocurrences</u>: Present every year in the Catalan coast.
9.- <u>Additional Comments</u>:
10.- <u>Individual to Contact</u>:

Maximino Delgado Instituto de Ciencias del Mar Ps. Joan de Borbó s/n 08039 Barcelona

HARMFUL ALGAL BLOOMS IN CATALUÑA IN 1996

1.- Location: Beach of La Fosca (Costa Brava, Catalonia)

- 2.- Date of Occurrence: July August 1996
- 3.- Effects: Green patches in the beach, negative effect on tourism.
- 4.- Management Decision:
- 5.- Causative Species: Alexandrium taylori Balech.
- 6.- Environment: Calm weather, temperature of water:20-26 °C, salinity 37-38 psu.

7.- Advected Population or In situ Growth: In situ growth.

8.- Previous Ocurrences: summers 1994 and 1995.

9.- Additional Comments: Non-toxic water discoloration. Recurrent every year.

10.- Individual to Contact:

Maximino Delgado Instituto de Ciencias del Mar Ps. Joan de Borbó s/n 08039 Barcelona

HARMFUL ALGAL BLOOMS IN CATALUÑA IN 1996

1.- Location: Alfacs Bay (Ebro Delta, Catalonia)

2.- Date of Occurrence: From December 96 to March 97.

3.- Effects: Mortality of wild fauna.

4.- <u>Management Decision</u>: Monitoring the dinoflagellate concentration before pumping of water to ponds.

5.- <u>Causative Species</u>: Gyrodinium corsicum Paulmier.

6.- Environment: Salinity 35-37 psu, temperature 11-17°C.

7.- Advected Population or In situ Growth: In situ growth.

8.- <u>Previous Ocurrences</u>: previous two years on the same period.
9.- <u>Additional Comments</u>: This species was associated with fish mortalities in aquaculture ponds in 1994. But in 1996, also mortalities of wild fish have been reported.
10.- <u>Individual to Contact</u>:

Maximino Delgado Instituto de Ciencias del Mar Ps. Joan de Borbó s/n 08039 Barcelona

HARMFUL ALGAL BLOOMS IN CATALUÑA IN 1996

1.- Location: Harbor of Vilanova i la Geltrú.

2.- Date of Occurrence: January and February 1996.

3.- Effects: Dinoflagellate bloom (max. concentration 694000 cells/l) associated with PSP detection in wild mussels from the harbour.

4.- Management Decision: Sampling twice a week the waters of the harbor and external waters.

5.- Causative Species: Alexandrium minutum Halim.

6.- Environment: Salinity 35-38 psu, temperature 11-14°C. Calm weather.

7.- Advected Population or In situ Growth: In situ growth.

8.- <u>Previous Ocurrences</u>: Yearly occurrence in the Catalonian coast.
9.- <u>Additional Comments</u>:
10.- <u>Individual to Contact</u>:

Maximino Delgado Instituto de Ciencias del Mar Ps. Joan de Borbó s/n 08039 Barcelona

HARMFUL ALGAL BLOOMS IN BALEARIC ISLANDS IN 1996

1.- Location: Harbour de Palma de Mallorca

2.- Date of Occurrence: April 1996

3.- Effects: Waters abnormally reddish brown

4.- <u>Management Decision</u>: once PSP toxicity was confirmed, the local government forbade fishing in the harbour of Palma de Mallorca

5.- Causative Species: Alexandrium minutum Halim

6.- <u>Environment</u>: Fine weather, appropriate temperature and high degree of enclosure

7.- Advected Population or In situ Growth: in situ growth

8.- Previous Occurrences: during April and May 1995

9.- Additional Comments:

10.- Individual to Contact:

Vincenç Forteza Unvisersitat de les Islas Balears Dep. Biologia Ambiental Campus Universitari, 0707 - Palma, SPAIN Tel: +34 71 173348 Fax: +34 71 173184

HARMFUL ALGAL BLOOMS IN BALEARIC ISLANDS IN 1996

1.- Location: Beaches of Peguera (South of Mallorca Island)

2.- Date of Occurrence: July-September 1996

3.- Effects: greenish-brown coloration near the coast, affecting negatively tourism

4.- Management Decision:

5.- Causative Species: Alexandrium taylori Balech

6.- Environment: Stability of the water column, increase in water temperature

7.- Advected Population or In situ Growth: in situ growth

8.- Previous Occurrences: summer months since 1985

9.- Additional Comments:

10.- Individual to Contact:

Vincenç Forteza Unvisersitat de les Islas Balears Dep. Biologia Ambiental Campus Universitari, 0707 - Palma, SPAIN Tel: +34 71 173348 Fax: +34 71 173184

- 1. Location: Fal estuary
- 2. Date of occurence: 11th June early August
- 3. Peak cell concentration: 780,000 cell per litre
- 4. Effects: PSP in bivalve flesh samples (292 units)
- 5. Causative species: Alexandrium tamarense
- 6. Environment: no data.
- 7. Advected population or in situ growth: no data.
- 8. Previous occurrence: Yes
- 9. Individual to contact: I.Laing

1. Location: River Avon

2. Date of occurence: 18th June - 29th July

3. Peak cell concentration: 380,000 cell per litre

4. Effects: PSP in bivalve flesh samples

5. Causative species: Alexandrium tamarense

6. Environment: no data.

7. Advected population or in situ growth: no data.

8. Previous occurrence: Yes, but first time that toxins detected

9. Individual to contact: I.Laing

- 1. Location: Weymouth Harbour
- 2. Date of occurence: July
- 3. Peak cell concentration: 970,000 cell per litre
- 4. Effects: non toxic
- 5. Causative species: Alexandrium tamarense
- 6. Environment: no data.
- 7. Advected population or in situ growth: no data.
- 8. Previous occurrence: Yes, peak cell concentrations lower than in previous years (5 million cells per litre)
- 9. Individual to contact: I.Laing

1. Location: Milford Haven

- 2. Date of occurence: 10th July 19th July
- 3. Peak cell concentration: 70,000 cell per litre
- 4. Effects: PSP in bivalve flesh samples (1307 units)
- 5. Causative species: Alexandrium tamarense
- 6. Environment: no data.
- 7. Advected population or in situ growth: no data.
- 8. Previous occurrence: Yes
- 9. Individual to contact: I.Laing

HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1996

I. Location: Pleasant Pt., Eastport to Canadian border (Area 1)

2. Date of Occurrence: *Mya arenaria* closure promulgated June 25, 1996 Repealed Sept. 27 (left in effect until this time only as a precautionary measure)

3. Effects: PSP in *Mytilus* and *Mya*

4. Management Action: Affected area closed to the harvest of *Mya arenaria* (*Mytilus edulis* permanently closed in this area)

- 5. Causative Species: Alexandrium tamarense
- 6. Environment:
- 7. Advected Population or In Situ Growth:
- 8. Previous Occurrences: Generally a yearly occurrence, since 1958, when monitoring began.

9. Additional Comments:

10. Individual to Contact: John W., Hurst, Jr.
 Laurie L. Bean
 Department of Marine Resources
 West Boothbay Harbor, ME 04575

Algal Bloom Reports - United States - 1996

1. <u>Locations</u>: Moriches and Shinnecock Bays, New York. The bloom was present mainly in eastern Moriches Bay, Quantuck Bay, and western Shinnecock Bay.

2. <u>Dates of Occurrence</u>: From May through October with peak concentrations ranging from 7.6×10^4 to 4.7×10^5 cells/ml occurring from late June through July.

3. <u>Effects</u>: Impacts on various shellfish species (scallops, hard clams, & mussels) and on submerged aquatic vegetation (eelgrass) have previously been reported. Other effects are aesthetic - water discoloration and reduced transparency.

4. Management Decisions: Continue weekly monitoring program.

- 5. <u>Causative Species</u>: Aureococcus anophagefferens
- 6. <u>Environment</u>: Temperature: 14.5 22.0 deg.C Salinity: 25-30 ppt Dissolved oxygen: 6.0 - 9.1 mg/l Water column stability: mixed
- 7. Advected population or in-situ growth: in-situ growth
- 8. <u>Previous Occurrences</u>: 1989: $<1.3 \times 10^5$ cells/ml in Moriches Bay $<2.3 \times 10^4$ cells/ml in Shinnecock Bay 1990: $<10^3$ to 9.6×10^5 cells/ml 1991: $<10^3$ to $>10^6$ cells/ml 1992: $>10^6$ cells/ml 1993: up to 2×10^5 cells/ml 1994: up to 3.8×10^4 cells/ml 1995: $>1.5 \times 10^6$ cells/ml

9. Additional comments:

10. Individual to Contact:

Dr. Robert Nuzzi Bureau of Marine Resources Suffolk County Dep't. of Health Services Riverhead, New York 11901 516-852-2082

TOTAL P.03

Algal Bloom Reports - United States - 1996

1. **Locations:** Great South Bay, New York. The bloom was present throughout the bay with highest concentrations found in its central portion from Patchogue Bay to the Robert Moses Causeway bridge.

2. <u>Dates of Occurrence</u>: From May through December, with peak concentrations ranging from $1.3x10^4$ to $1.8x10^5$ cells/ml occurring in mid June, and a secondary peak of from $2.9x10^4$ to $1.1x10^5$ cells/ml occurring in November and December.

3. <u>Effects</u>: Impacts on various shellfish species (scallops, hard clams, & mussels) and on submerged aquatic vegetation (eelgrass) have previously been reported. Other effects are aesthetic - water discoloration and reduced transparency.

4. Management Decisions: Continue weekly monitoring program.

5. <u>Causative Species</u>: Aureococcus anophagefferens

- 6. <u>Environment</u>: Temperature: 5.5-27.0 deg.C Salinity: 20-28 ppt Dissolved oxygen: 6.0-10.5 mg/l Water column stability: mixed
- 7. Advected population or in-situ growth: in-situ growth
- 8. <u>Previous Occurrences</u>: 1985, 1986: >10⁶ cells/ml 1988: $10^3 - 5 \ge 10^5$ cells/ml (June-Aug) 1989: <2.5 \u2265 10⁴ cells/ml (April-Sept) 1990: <1 \u2265 10⁴ cells/ml (May-Dec) 1991: <10⁴ cells/ml (Jan-June) 1992: $10^3 - 10^6$ cells/ml (Jan-Dec) 1993: <10³ - 2.6 \u2265 10⁵ cells/ml (Jan-Mar, Aug-Nov) 1994: up to 10^6 cells/ml (June-July) up to 1.2 \u2265 10⁴ cells/ml (Aug-Oct) 1995: 2.8 \u0365 to >10⁶ cells/ml in July

9. Additional comments:

10. Individual to Contact:

Dr. Robert Nuzzi Bureau of Marine Resources Suffolk County Dep't. of Health Services Riverhead, New York 11901 516-852-2082 04/18/1997 13:33

12708015 P.02

HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1996

- 1. Location: Indian River Lagoon, Florida, Area 15
- 2. Date of Occurrence: September and October, 1996, possibly August
- 3. **Effects:** Caused surface water discoloration and was associated with fish kills in the same area at the same time. Associated with human respiratory irritation (stinging sensation in mouth and lungs and sinus irritation) and eye irritation.
- 4. **Management Action:** Local shellfish beds were closed because of potential unknown toxicity until mouse bioassays for lipid-soluble toxins were run. Shellfish beds were reopened because mouse bioassays revealed no signs of toxicity.
- 5. **Causative Species:** Gymnodinium pulchellum Larsen 1994 also known as Gymnodinium sp. 'type 84-K' and Gymnodinium sp. (Japan). The Japanese Gymnodinium sp. tested by Endo et al. 1992 contained oxidized brevetoxins.Counts as high as 19.7 x 10⁶ cells liter⁻¹ were recorded during this bloom.
- 6. **Environment:** Shallow lagoon. At time of bloom, salinity ranged from 26 to 37 ppt. Temperature ranged from 26 to 28°C.
- 7. Advected Population or In Situ Growth: In situ growth.
- 8. **Previous Occurrences:** No bloom under the name of *G. pulchellum*, but there have been previous blooms in this system caused by a *Gymnodinium* sp. of the same size ($< 25\mu$) and similar morphology. A specimen of this species was previously identified once from Florida Bay, FL but no blooms have been recorded.
- 9. Additional Comments: This bloom represents the first record of *G. pulchellum* in the western North Atlantic and Americas (Steidinger, Landsberg, Truby and Roberts, in prep.). An isolate is being cultured to confirm ichthyotoxicity. Samples were collected specifically to look for *Pfiesteria*-like species, none were found.
- Individual to Contact: Dr. Karen A. Steidinger
 Florida Department of Environmental Protection
 Florida Marine Research Institute
 100 Eighth Avenue S. E.
 St. Petersburg, FL 33701-5095
 Tele: 813-896-8626
 Fax: 813-823-0166
 email:steidinger k@sellers.dep.state.fl.us

HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1996

- Location: Florida 1. Florida Bay, Area 16
- Date of Occurrence: January 1996 January 1997 2.
- Effects: Yellow-green to pea green discolored sea water with 3. decreased water clarity. In a previous year, sponge mortality conicidental with bloom areas.
- Management Action: Restoration of the bay is the long term 4. goal of an interagency plan, part of the restoration involves freshwater delivery.
- Species: Blue green alga or cyanobacterium, 5. Causative <u>Synechococcus</u> elongatus, up to 10^6 cells mI¹. Can co-occur with small (< 10 μ m) centric diatoms and other cyanobacteria, particularly coccoid forms. Chlorophyll a levels up to > 20 μ g liter⁻¹.
- Environment: Shallow subtropical lagoon with salinites from 6. essentially freshwater to marine (up to 38.3 o/oo) and temperatures from 20.99 to 33.5 C. In previous years, the bay has been hypersaline. Resuspension events from winds and tidal action common. Resuspended sediments can change surface discoloration to browns and tans.
- Advected Population or In Situ Growth: In situ growth within 7. sub-basins of bay. High residency time within sub-basins, but sub-basins flushed by rain and storm events through narrow channels.
- Previous Occurrences: Bloom has been on-going since 1991 but 8. the intensity and geographic coverage varies seasonally.
- Additional Comments: 9.

10.	Individual to Contact:	Dr. Karen A. Steidinger
		Florida Department of Environment
		Protecti
		Florida Marine Research Institute
		100 Eighth Avenue S. E.
		St. Petersburg, Fl 33701-5095
		Tele: 813-896-8626
		Fax: 813-823-0166

email: steidinger@sellers.dep.state.fl.us

Environmental Protection HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1996

- 1. Location: Florida Pensacola to the Florida Keys - Area 16
- Date of Occurrence: January June, October and November 1996 (inshore, offshore), September - December 1996(offshore central west coast) and continued into 1997 as an offshore bloom.
- 3. Effects:Human respiratory irritation from the Florida Panhandle to SW Florida. Three cases of NSP in June, two involved children. Dead fish - inshore and offshore. Dead birds, e.g.,cormorants. Dead turtles suspect. About 150 manatees died from red tide exposure in March and April in SW Florida inshore waters where red tide rarely establishes in winter.
- 4. Management Decision: Shellfish harvest bans due to <u>Gymnodinium</u> <u>breve</u> red tide in most major shellfish harvesting areas along the west and north coasts of Florida at some point in year.
- 5. Causative Species: <u>Gymnodinium breve</u> Inshore and coastal water samples up to 45 miles offshore had cell concentrations ranging from negative to 10⁷ cells Liter^{-1.}
- 6. Environment: Occurred in nearshore and shelf waters with wide salinity range (15.5 to 36 o/oo) and temperatures of 12 to 32 C.
- 7. Advected Population or In Situ Growth: Advected population from offshore waters between Tampa Bay and Charlotte Harbor. In February, warm water intrusion documented off southwest Florida which preceeded establishment of red tide in Pine Island Sound. In November, Loop Current water documented within 30-50 miles off shore off North Florida and northern Gulf. North Florida blooms occurred in May-July as well as October-November. In September offshore bloom off Tampa Bay to Charlotte Harbor, mainly between 10 and 35 miles that remained offshore until it started to dissipate in February/March 1997. Still monitoring.
- 8. Previous Occurrences: Usually patchy along Florida coast, not necessarily coast wide.Jan-Dec 1995, Sept-Dec 1994; Jan 1993 (cont. of Dec 1992)Sept-Dec 1992; Jan-Feb 1991; Feb-Mar, Oct-Nov, 1990; March-May 1989; Oct-Dec, 1988; Jan/Feb, May-July, Sept/Oct, 1987; Sept-Dec 1986, Sept-Dec 1985, Jan-March, May-Aug,1984, Jan/Feb, Oct-Dec, 1983; Jan-April, July-Oct,1982; Sept/Oct, 1981; Jan/Feb, June-Nov, 1980; and before.
- 9. Additional Comments: The 1996 red tide year had severe consequences in loss of living aquatic resources such as

manatees, fishes, birds, and probably sea turtles. Humans were affected with the respiratory irritant known as mred tide aerosol, and local economies suffered losses due to reduced revenues from tourism and recreational activities.One Bay system was continually closed because of long-term retention of brevetoxins in a small edible clam and rainfall events. In addition, all Gulf States had G. breve blooms in 1996 which is the first time this bloom distribution has been documented, although G.breve has been documented throughout the Gulf in low concentrations. Florida strains of G. breve typically lyse at 15-17 ppt and blooms do not do well below 24 ppt, so Florida bays are usually protected by a salinity barrier, but some other Gulf States have recorded G. breve populations below 15 ppt and one Florida sample at a north Florida pass was recorded as 15.5 ppt. This needs to be investigated with north Gulf G. breve isolates.

10. Individual to Contact:Dr. Karen A. Steidinger Florida Department of Environmental Protection Florida Marine Research Institute 100 Eighth Avenue S. E. St. Petersburg, Fl 33701-5095 Tele: 813-896-8626 Fax: 813-823-0166 steidinger_k@sellers.dep.state.fl.us

HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1996

MISSISSIPPI

- 1. Location: Hancock, Harrison, and Jackson Counties; Area 17.
- 2. Date of Occurrence: 26 October 1996 through 11 December 1996.
- 3. Effects: Water discoloration, due in part to co-occurring blooms of non-toxic species; moderate fish mortalities both inshore and on barrier islands; mortalities of waterfowl and marine mammals; respiratory irritation reported by DMR and Wildlife, Fisheries, & Parks personnel; not uniformly distributed.
- 4. Management Action: Shellfish harvest bans due to *Gymnodinium breve* red tide in Middle Bay and Mississippi Sound (Area 8) of Jackson County from 2 November 1996 to 25 February 1997; western Mississippi Sound in Hancock County (Area 2) closed from 7 November 1996 to 26 February 1997 for Pass Marianne reefs; Long Beach tonging reefs in nearshore western Mississippi Sound closed 7 November 1996 to 10 December 1996.
- 5. Causative Species: Gymnodinium breve. Inshore waters in the immediate vicinity of routinely harvested oyster reefs had cell concentrations ranging from zero to greater than 6.3 x 10^5 cells per liter; water samples from the vicinity of the barrier islands had cell counts ranging from negative to in excess of 13.6 x 10^6 cells per liter.
- 6. Environment: Occurred in vicinity of barrier islands and in nearshore waters with salinity ranges of 5 30 ppt, and water temperature range of 12 27.1°C.
- 7. Advected Population or In Situ Growth: Advected population from Gulf of Mexico waters in vicinity of Mississippi's barrier islands and the Chandeleur Island chain in eastern Louisiana. Some *in situ* growth may have occurred in Middle Bay (east Jackson County) and in the vicinity of Cat Island (Area 4) in MS Sound.
- 8. **Previous Occurrences:** None.

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- 9. Additional Comments: Presence of brevetoxin confirmed in 6 specimens of lesser scaup sent in for necropsy. Bloom appeared to be followed by *Prorocentrum* spp. bloom with okadaic acid production; data to support or refute pending. Bloom timing coincided with the beginning of roe mullet season; roe of *Mugil cephalus* tested negative for brevetoxin.
 - Individual to Contact: Dr. Cynthia A. Moncreiff USM Institute of Marine Sciences Gulf Coast Research Laboratory 703 East Beach Drive, P.O. Box 7000 Ocean Springs, MS 39566-7000 Phone: (601) 872-4260; FAX: (601) 872-4204 E-mail: cmoncrei@whale.st.usm.edu

TYPE OF TOXICITY (PSP, DSP, ASP, NSP, ETC.): NSP; likely followed by DSP

DURATION OF TOXIC EPISODES

Maximmum toxicity (ug/100g) 3,060 μg/100g (84 MU) Dec × Nov × Oct Sep Aug Jul Jun May Apr Mar Feb × Jan × code 17 17 MS MS area YEAR 1996 1997

This table should be used to indicate the duration of the toxic episodes and the maximum level of measured toxicity.

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HUMAN INTOXICATIONS

YEAR	MONTH	AREA (CODE)	COMMENTS
1996	Oct	17	No human intoxications reported.
1996	Nov	17	No human intoxications reported.
1996	Dec	17	No human intoxications reported.
1997	Jan	17	No human intoxications reported.
1997	Feb	17	No human intoxications reported.

MORTALITY OF FISH AND OTHER MARINE ORGANISMS

YEAR	MONTH	AREA (CODE)	COMMENTS
1996	Oct	17	Anchoa mitchilli, Mugil curema; Archosargus probatocephalus
1996	Nov	17	Tursiops truncatus (n=20); >107 waterfowl; fish (moderate)
1996	Dec	17	Tursiops truncatus (n=10); Sciaenops ocellatus (moderate)
1997	Jan	17	No major mortalities reported.
1997	Feb	17	No major mortalities reported.
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HARMFUL ALGAL BLOOMS IN THE UNITED STATES --- 1996

- 1. Location: Coastal Alabama, area 17
- 2. Date of Occurrences: 23 October 1996 through 6 December 1996
- 3. Effects: Water discoloration brownish-red in some areas. Respiratory irritation along Gulf Shores area. No significant fish kills.
- 4. Management Action: Shellfish harvest closure 10 November 1996 to 6 December 96 due to *Gymnodinium breve* red tide in Alabama coastal waters. Public Health notice issued 29 October 1996 for Gulf Shores Area.
- 5. Causative Species: *Gymnodinium breve*. Inshore and bay water samples had cell concentrations ranging from 0 to 400,000 cells/liter.
- 6. Environment: Occurred in near shore beach areas of Gulf Shores and Fort Morgan with salinity ranging from 24ppt to 27ppt and temperature ranging 20°C to 25°C and Mobile Bay and Mississippi Sound areas with salinity ranging 7ppt to 27ppt (High salinity was recorded near Mississippi state line in Grand Bay. This reading was greatly above normal with strong WNW wind and rising tide.) and temperature ranging 12°C to 26°C.
- Advected Population or In Situ Growth: Advected population from West Florida waters (Perido Bay area). The population moved westward along Alabama coastal waters and was carried into Mobile Bay and Mississippi Sound waters due to unusual wind and current conditions.
- 8. Previous Occurrences: None.
- 9. Additional Comments: Special thanks to Dr. Karen Steidinger and staff, and to Dr. Bob Dickey and staff.
- 10: Individual to Contact: Dr. Lewis A. Byrd

Director of Seafood Quality Assurance Alabama Department of Public Health Mobile, Alabama 36608

334-344-6656 phone 334-343-9061 fax

HARMFUL ALGAL BLOOMS IN THE UNITED STATES—1996

LOUISIANA

- 1. Location: Louisiana coastal waters from the Louisiana/Mississippi border to 90°40'W
- 2. Date of Occurrence: 11/12/96 to 12/17/96, but toxicity on-going (as of Mar. 13,1997)
- 3. Effects: Some fish, duck, and porpoise deaths may have been due to *Gymnodinium breve*, but not confirmed.
- 1. Management Action: Shellfish harvest bans due to *Gymnodinium breve* from Nov. 13, 1996 to Feb. 28, 1997, Mississippi River Gulf Outlet to the Louisiana/Mississippi River border. Some areas opened Feb. 28, 1997 after 2 consecutive negative toxin tests (mouse bioassay); some areas still closed (as of Mar. 13, 1997).
- Causative Species: Gymnodinium breve. Inshore waters east of Mississippi River: Exceeded 5,000 cells/liter from Nov. 12, 1996 (start of sampling) to Dec. 4, 1996 with maximum abundance of 6.20 x 10⁵ cells/liter on 11/20/96. Inshore and shelf waters west of Mississippi River and east of 90°40'W. Observed at low abundances (less than or equal to 5,000 cells/liter) from 11/13/96 -12/4/96. Also unusually high numbers of *Dinophysis caudata* and *Prorocentrum* spp. during the same period.
- 1. Environment: Occurred in nearshore and shelf waters between 5 and 34.5 ppt and 14.5 and 21°C. Abundances above regulatory limits (>5,000 cells/liter) occurred in Louisiana waters between 18 and 28 ppt, but data from Mississippi suggests a lower salinity (15 ppt).
- 1. Advected Population or In situ Growth: There is no direct data at present to assess this question, but reported dates when *Gymnodinium breve* was present in Florida, Alabama, and Mississippi waters suggest advection from east was a factor. Growth must also have occurred to maintain such high numbers for such a long time.
- 1. Previous Occurrences: None documented for Louisiana waters.
- 1. Additional Comments: Besides lower temperature and salinity than usually associated with *Gymnodinium breve* blooms, the blooms occurred in shallow water when winds were strong, light levels were low, and water turbidity was high. In addition, oysters in areas which are still closed have remained toxic (by mouse bioassay) much longer than expected.

10. Individual to Contact:	Dr. Quay Dortch
	Louisiana Universities Marine Consortium
	8124 Highway 56
	Chauvin, LA 70344
	Phone: 504/851-2800
	FAX: 504/851-2874
	E-mail: qdortch@lumcon.edu

HARMFUL ALGAL BLOOMS IN THE UNITED STATES-1996

LOUISIANA

Location: Louisiana coastal waters Grand Isle to 90°40'W up to 20 mi offshore and Terrebonne Bay

1. Date of Occurrence: September, 1996

2. Effects: Reports of bioluminescence widely distributed.

Management Action: None

Causative Species: Alexandrium monilatum

Environment: Occurred at salinities from 15 to 30 ppt.

Advected Population or In situ Growth: Unknown

Previous Occurrences: Blooms frequent in fall in same area.

Additional Comments: Extent and intensity of surface bioluminescence much more widespread and prolonged than usual, but not reports of discolored water. Higher salinities in nearshore and estuarine waters than usual. Measured abundances never exceeded 100,000 cells/liter, but samples for counts were not necessarily from areas where intense bioluminescence was reported.

10. Individual to Contact: Dr. Quay Dortch Louisiana Universities Marine Consortium Highway 56 Chauvin, LA 70344 Phone: 504/851-2800 FAX: 504/851-2874 E-mail: qdortch@lumcon.edu

HARMFUL ALGAL BLOOMS IN THE UNITED STATES -- 1996

- 1. Location: Red Tide: Coastal Texas Matagorda Bay to Port Isabel (this includes) San Antonio, Aransas, Matagorda and Corpus Christi Bays)...Code 18. Brown Tide: Upper Laguna Madre, Baffin Bay and lower Laguna Madre, Texas. Nueces, Kleberg, Kenedy, Willacy and Cameron Counties.
- **2. Date of Occurrence:** Red Tide: 11 September 1996 through 20 October 1996. Brown Tide: Persistent throughout the entire year.
- **3. Effects:** Red Tide: Dead fish (approximately 5 million) inshore and offshore within coastal lagoons. Water discoloration brownish-red. Respiratory irritation throughout the entire area. Noted oyster die-offs in areas of high concentrations. Brown Tide: Water discoloration continues, same as last year.
- **4. Management Action:** Red Tide: Shellfish harvest bans due to *Gymnodinium breve* red tide Matagorda Bay south to Texas-Mexico border. Bays closed from September 1996 February 1997. Brown Tide: Same as last year.
- Causative Species: Red Tide: Gymnodinium breve, with cell concentrations reaching 3 X 10⁸ cells/liter. Brown Tide: Aureoumbra lagunensis, with cell concentrations reaching 4 X 10⁹ cells/liter.
- 6. Environment: Red Tide: Occurred in nearshore and shelf waters and in coastal embayments and lagoons. Salinity ranges 25-42 0/00; Temperature ranges 21-27°C. Brown Tide: Confined to Baffin Bay and Laguna Madre. Environmental conditions same as last year.
- 7. Advected Population or In Situ Growth: Red Tide: Initial population advected into nearshore waters, with in situ growth occurring in coastal embayments. Brown Tide: In situ growth.
- 8. Previous Occurrences: Red Tide: Previously occurred in 1986. Brown Tide: Bloom started in December 1989 and has been present continuously in Baffin Bay and Laguna Madre to the present time.

9. Additional Comments:

10: Individual to Contact:	Dr. Dean A. Stockwell
	Marine Science Institute
	The University of Texas at Austin
	750 Channelview Drive
	Port Aransas, TX 78373
	Phone: 512-749-6705 Fax: 512-749-6777
	Email: dean@utmsi.zo.utexas.edu

DURATION OF TOXIC EPISODES

TYPE OF TOXICITY (PSP, DSP, ASP, NSP, ETC.): NSP

YEAR	arca		code	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Maximum toxicity (ug/100g
1996		18	NSP									x	x	x	x	200-260 mouse units
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This table should be used to indicate the duration of the toxic episodes and the maximum level of measured toxicity.

MORTALITY OF FISH AND OTHER MARINE ORGANISMS

YEAR	MONTH	AREA (CODE)	COMMENTS
1996	Sept/Oct	18	Approximately 5 million fish killed along Texas coast and
		•	including Matagorda, San Antonio, Aransas, and Corpus
			Christi Bays. Approximately 12,000 Bull Reds (Red Drum)
			were killed during this event. Age structure information
[is available through Texas Parks and Wildlife Department
			(Contact: Larry McEachren 512-729-2328).
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HUMAN INTOXICATIONS

COMMENTS	None reported.									
AREA (CODE)	18									
YEAR MONTH	1									
YEAR	1.996									

HARMFUL ALGAL BLOOMS IN THE UNITED STATES: 1996

- 1. Location: Area 21, Drakes Estero, Marin County (50 miles north of San Francisco). This is the most productive commercial shellfish growing area in California at present. Only oysters are grown and harvested for human consumption. Mussels are used as a monitoring tool. Affected Species: bay mussels (*Mytilus edulis*).
- 2. Date: May 22-24; July 9-11.
- 3. Effects: Relatively minor episodes, each short lived. May event resulted in a maximum measured toxicity of 160 ug/100 g tissue. July maximum was 130 ug.

4. Management Action: Outer Estero closed to harvesting. Inner Estero placed on batch release.

5. Causative Species: Alexandrium catenella. Confirmed by routine phytoplankton monitoring.

- 6. Environment: Each event occurred during a slight warming trend which broke down quickly. The July warming trend appeared to be better developed. Imagery poor.
- 7. Advected/In Situ: May: Unknown. Warming trend did not last beyond a couple days. Imagery shows no clear sign of advection. Toxicity due to in situ population that experiences a small increase due to warming conditions? July: advection more plausible due to appearance of small amount of warm water advected nearsbore.

8. Previous Occurrences: Alert levels of PSP toxicity routinely occur in this region each year.

9. Additional Comments: This was a year of exceptionally low PSP activity (see enclosed graph).

10. Contact: Gregg Langlois California Department of Health Services Marine Biotoxin Program 2151 Berkeley Way, Room 118 Berkeley, CA 94704 510-540-3423 510-540-2716 (Fax) glangloi@ix.netcom.com DURATION OF TOXIC EPISODES

YQY TYPE OF TOXICITY (PSP, DSP, ASP, NSP, ETC.): 28 COUNTY

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		AMAMAN	244	21															
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This table should be used to indicate the duration of the toxic episodes and the maximum fevel of measured toxicity.

Figure 1. PSP toxicity in Californnia during the ten year interval from 1986 to 1996. **PSP TOXICITY: 1986-1996** TIME (Years) PSP TOXICITY (ug/100 g tissue) Ó 

### HARMFUL ALGAL BLOOMS IN THE UNITED STATES -- 1996

1. Location: East and West sounds, Orcas Island; Quartermaster Harbor, Maury Island; Mystery Bay, Marrowstone Island; Discovery Bay; Sequim Bay, Washington

2. Date of Occurrence: Various times from 1 July to 22 October 1996

3. Effects: Closure of blue mussel (*Mytilus edulis*), and Pacific oyster (Crassostrea gigas) harvest

4. Management action: There was some product recall.

5. Causative species: Suspected to be Alexandrium catenella.

6. Environment: Not available

7. Advected Population or In Situ Growth: Probably in situ growth

8. **Previous Occurrences:** PSP commonly occurs in mussels and other bivalve molluscs throughout western Washington inland waters (Puget Sound, Admiralty Inlet, San Juan Islands, etc.) during the summer.

### 9. Additional Comments:

10. Individual to Contact: Mr. Frank Cox Washington State Department of Health Shellfish Programs Airdustrial Center, Bldg. 4 P.O. Box 47824 Olympia, WA 98504-7824

### HARMFUL ALGAL BLOOMS IN THE UNITED STATES -- 1996

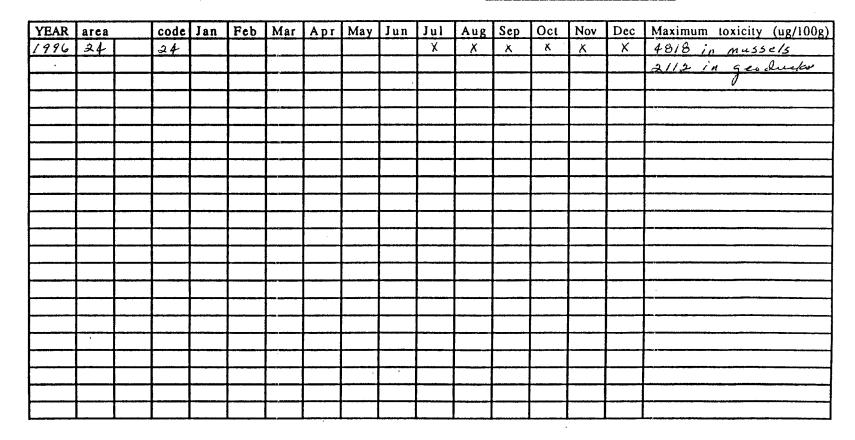
- 1. Location: Port Gamble, Murden Cove, Agate Pass, Skiff Point, Washington
- 2. Date of Occurrence: Various times from 9 September to 23 December 1996
- 3. Effects: Closure of geoduck (Panope generosa) harvest.
- 4. Management action: There was some product recall.
- 5. Causative species: Suspected to be cysts of Alexandrium catenella.
- 6. Environment: Water depth varied from about 10 to 35 m.
- 7. Advected Population or In Situ Growth: Probably cysts
- 8. Previous Occurrences:

**9.** Additional Comments: Cysts were probably stirred up into the water column when the geoducks were harvested by divers using a suction-type device. Toxic and non-toxic periods alternated at individual sites. This is a relatively new commercial fishery potentially worth about \$5 million.

10. Individual to Contact: Mr. Frank Cox Washington State Department of Health Shellfish Programs Airdustrial Center, Bldg. 4 P.O. Box 47824 Olympia, WA 98504-7824

# **DURATION OF TOXIC EPISODES**

TYPE OF TOXICITY (PSP, DSP, ASP, NSP, ETC.):  $\rho \leq \rho$ 



This table should be used to indicate the duration of the toxic episodes and the maximum level of measured toxicity.

# HARMFUL ALGAL BLOOMS IN THE UNITED STATES — 1995

## ALASKA

- 1. Location: Kodiak Island, Alaska at Thumps Up Cove (mile 34.5 Chiniak Highway).
- 2. Date of Occurrence: 24 March 1996
- 3. Effects: Four individuals ate Butter Clams (Saxidomus giganteus). One individual vomitted 1 1/2 hours after eating. PSP level of steamed butter clams 68 ug/100 grams. Vomitus sampled were negative for PSP.
- 4. Management Action: NA
- 5. Causative Species: Unknown
  - 6. Environment:
- 7. Advected Population or In Situ Growth:
- 8. Previous Occurrences: Area has been implicated with PSP illnesses in the past
- 9. Additional Comments:
- Individual to Contact: Michael J. Ostasz
   Shellfish Coordinator, Seafood Program
   State of Alaska
   Department of Environmental Conservation
   555 Cordova Street
   Anchorage, AKASKA 99515
   Tel: (907) 269-7638
   Fax: (907) 269-7510
   E-Mail: mostasz@envircon.state.ak.us

# HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1995

## ALASKA

- 1. Location: Pillar Creek area on Kodiak Island, Alaska (58°25'30"N, 152°48'55"W)
- 2. Date of Occurrence: 10 May 1996
- 3. Effects: One individual seen at Hospital Emergency Room (tingling and numbness by mouth)

PSP Tests:

- a) Mussel soup (mussel cooked in wine sauce) -> negative for PSP.
- b) Vomitus -> negative for PSP
- c) Mussels from area (6 days later) 456 and 435 micrograms/100 grams
- 4. Management Action:
- 5. Causative Species: Suspect Alexandrium
  - 6. Environment:
- 7. Advected Population or In Situ Growth:
- 8. **Previous Occurrences:** General area has had PSP problems in the past from unapproved (classified) area of harvest
- 9. Additional Comments:
- **10.** Individual to Contact:

Michael J. Ostasz Shellfish Coordinator, Seafood Program State of Alaska Department of Environmental Conservation 555 Cordova Street Anchorage, AKASKA 99515 Tel: (907) 269-7638 Fax: (907) 269-7510 E-Mail: mostasz@envircon.state.ak.us

## HARMFUL ALGAL BLOOMS IN THE UNITED STATES - 1995

## ALASKA

- **1.** Location: Hoonah, Alaska
- 2. Date of Occurrence: 6 March 1996
- 3. Effects: One individual ill 3 hours after consuming cockle clams with symptoms of shortness of breath, nausea, vomiting, tingling of lips/tongue and diarrhea. Clams harvested on 6 February 1996. Clams eaten steamed with seal oil and alcohol. Person hospitalized and treated for fluids replacement.

Samples negative for PSP (32-33 ug/100 grams)

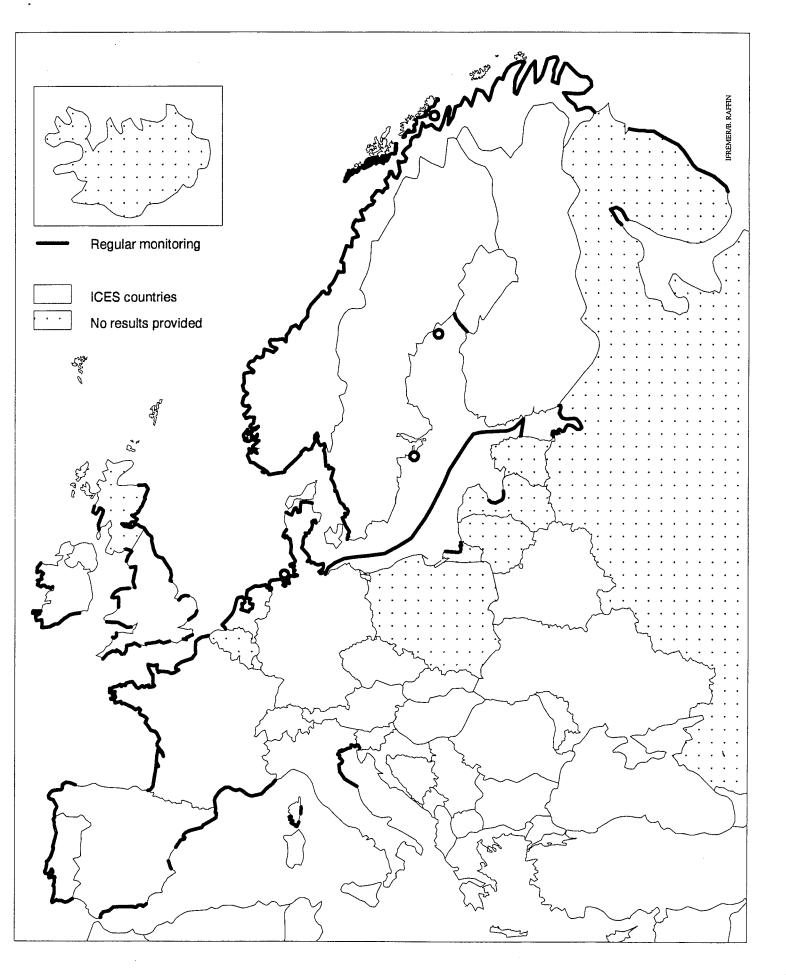
- 4. Management Action:
- 5. Causative Species: Suspect Alexandrium
  - 6. Environment: Unknown
- 7. Advected Population or In Situ Growth:
- 8. Previous Occurrences:
- 9. Additional Comments:
- **10.** Individual to Contact: M

Michael J. Ostasz Shellfish Coordinator, Seafood Program State of Alaska Department of Environmental Conservation 555 Cordova Street Anchorage, AKASKA 99515 Tel: (907) 269-7638 Fax: (907) 269-7510 E-Mail: mostasz@envircon.state.ak.us

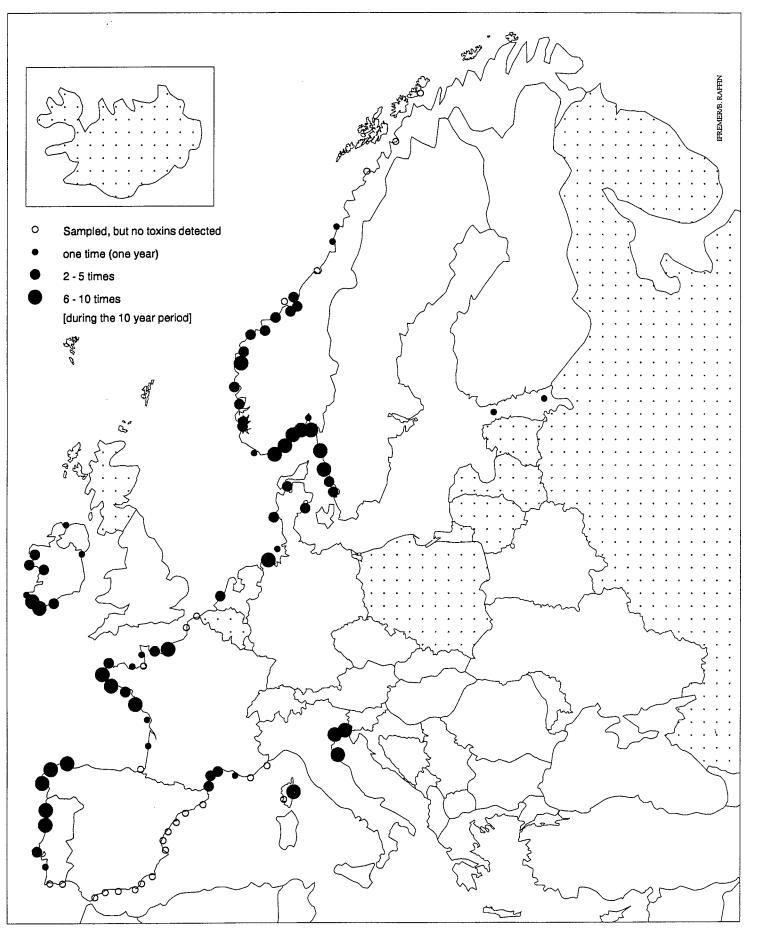
### ANNEX III

### DECADIAL MAPS OF PHYTOPLANKTON TOXINS IN THE ICES AREA

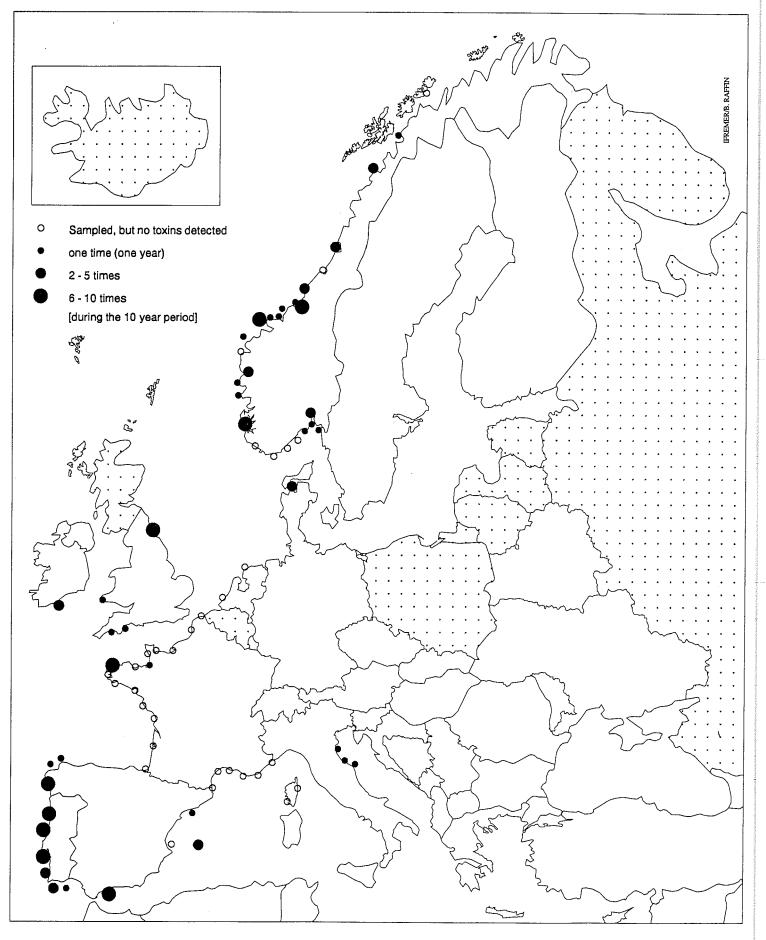
Period 1987-1996



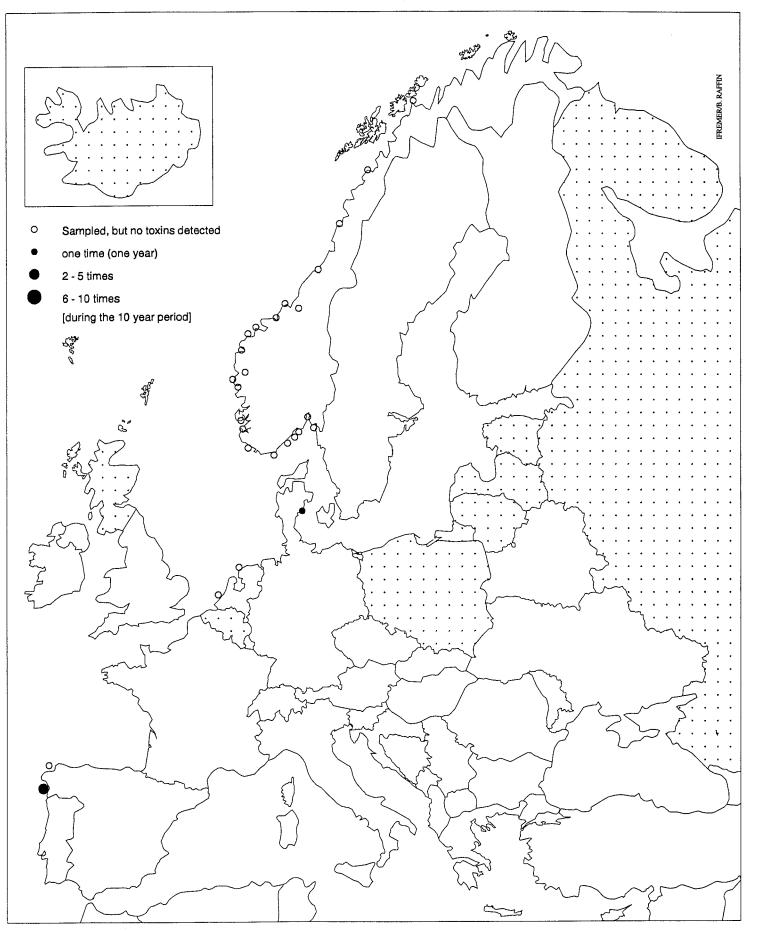
# Presence of DSP toxins



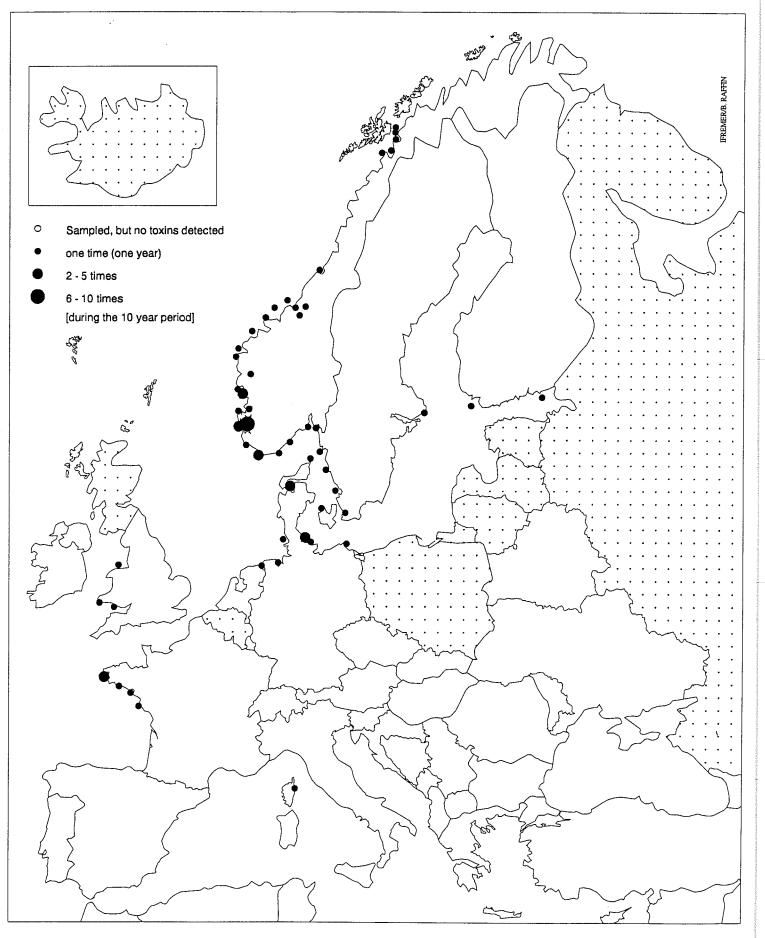
# Presence of PSP toxins

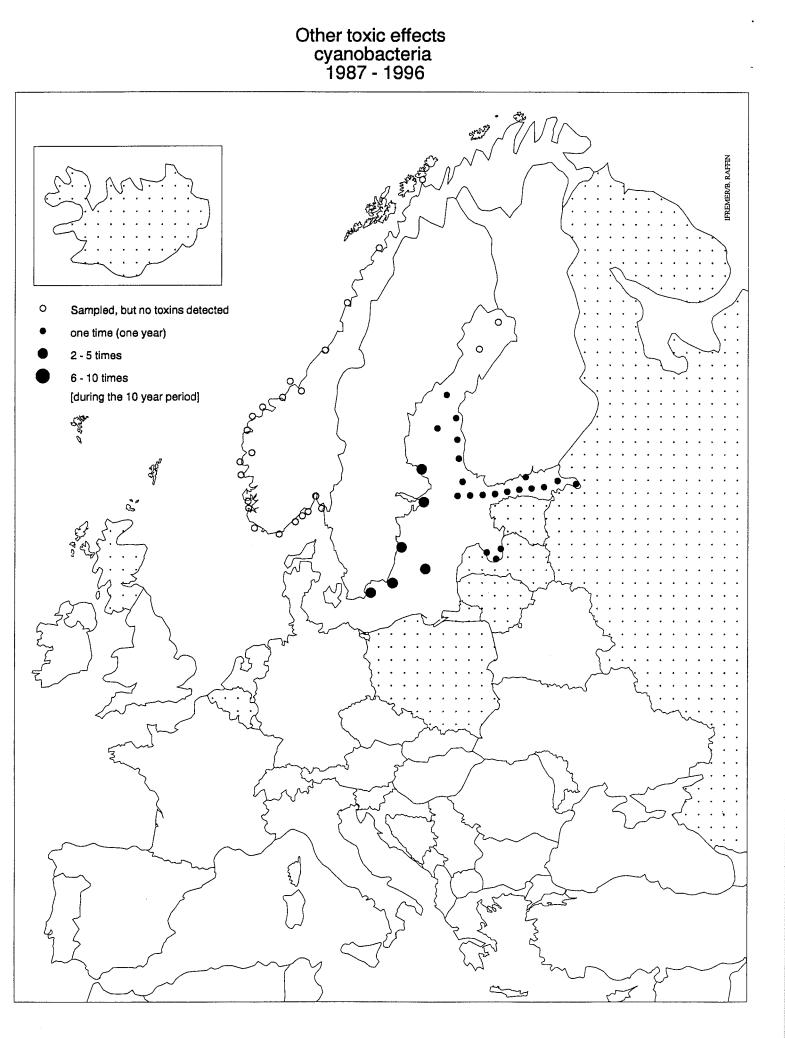


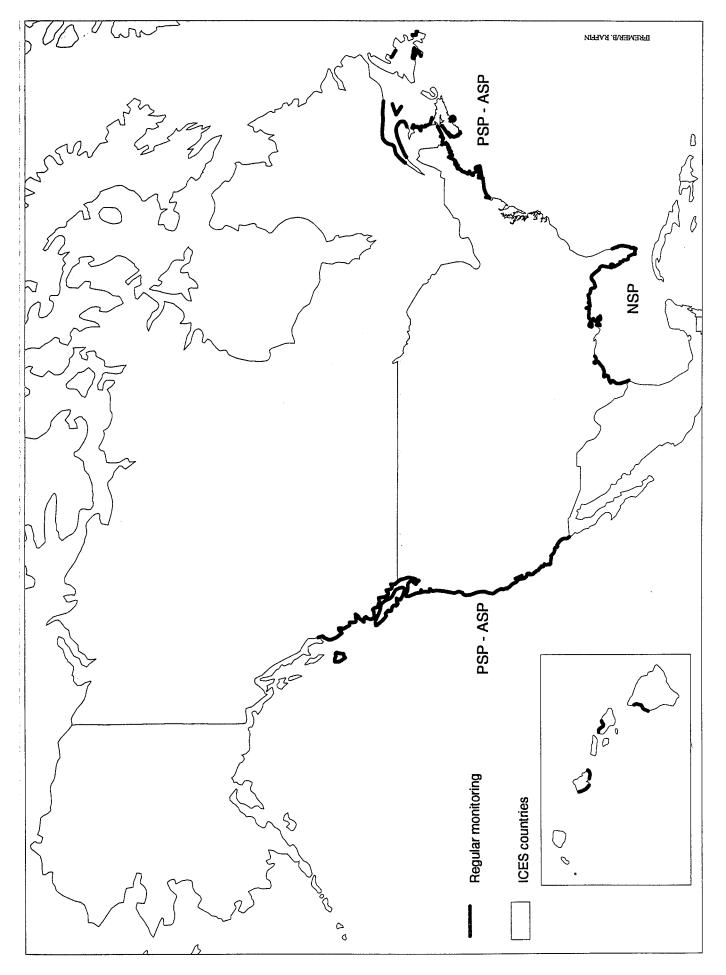
# Presence of ASP toxins

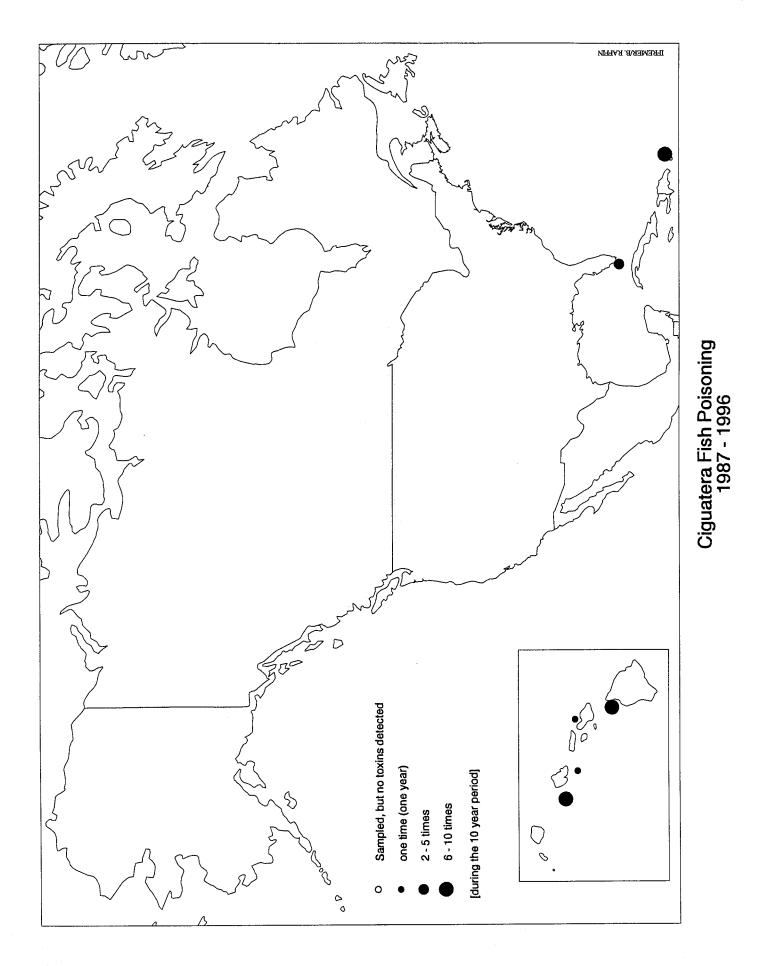


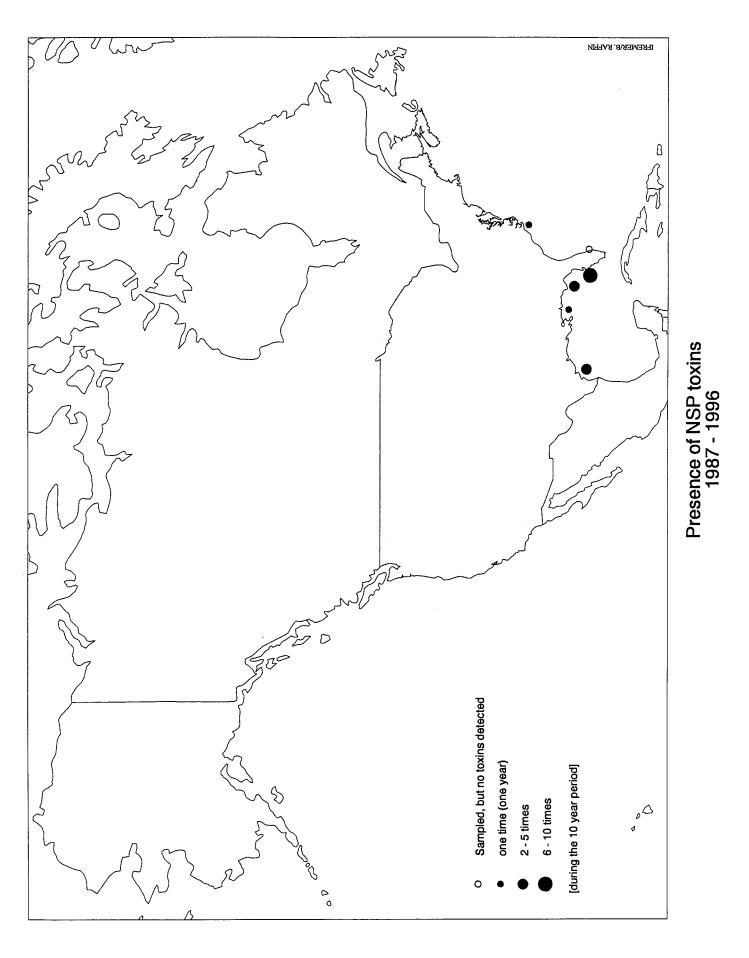
# Animal and plant mortalities

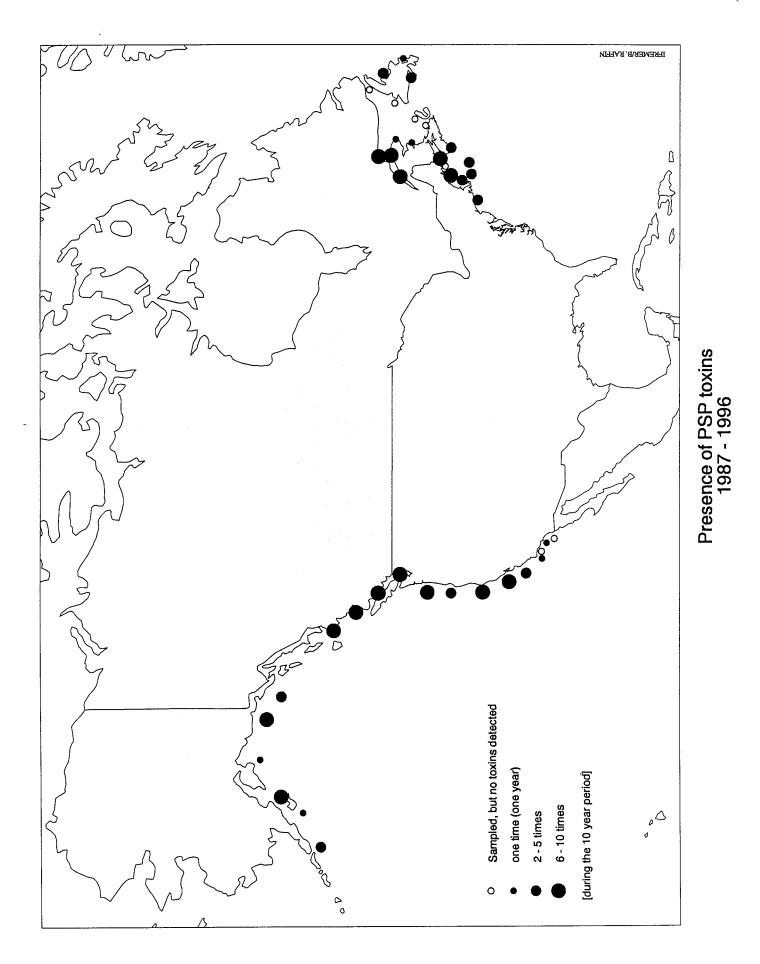


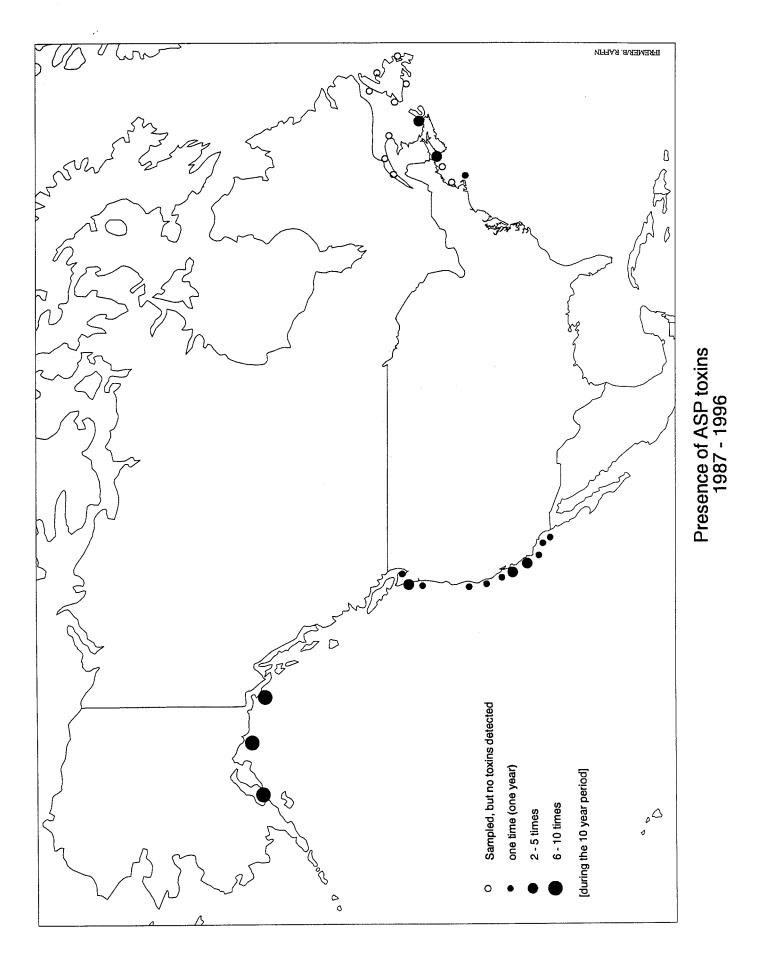


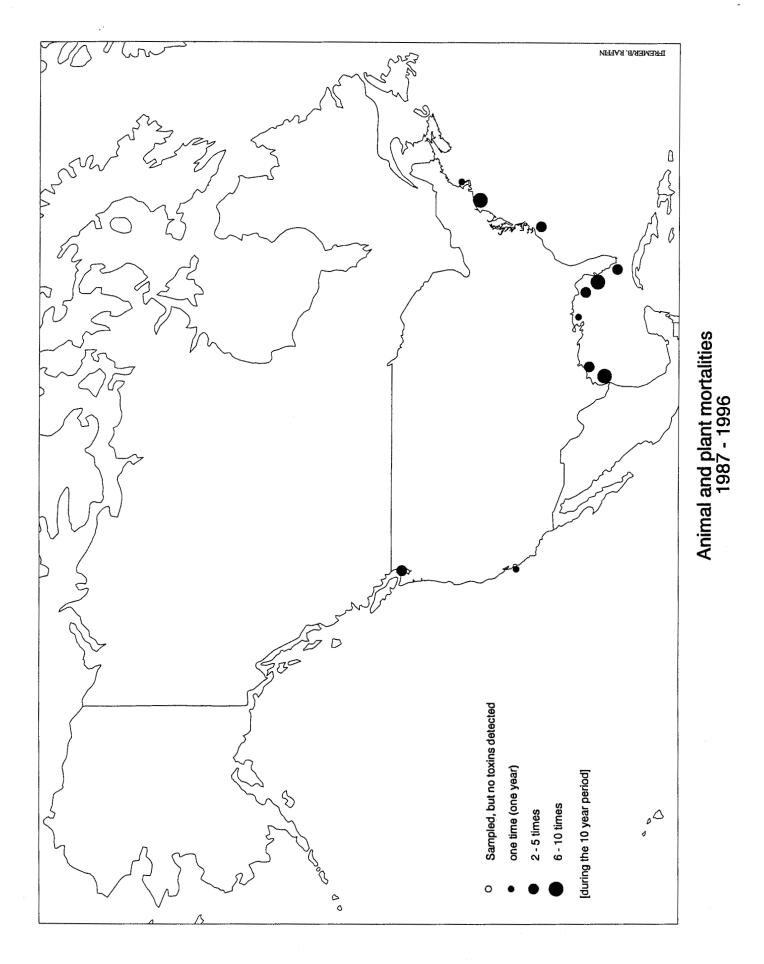












#### ANNEX IV

### **References to term of reference 6**

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### ANNEX V

### DEVELOPMENT OF A COMPUTER INFORMATION DATA-BASE ON HARMFUL ALGAL OCCURRENCES WORLDWIDE

### 1) Objectives:

To gather existing data on harmful algae occurrences and make it available in a computer format in order to facilitate the search of information. To improve future gathering of data on harmful algal occurrences.

### 2) Justification:

It responds to the attempts periodically proposed within ICES since 1982 and to the priorities included within the Educational Programme Element of the IOC HAB Programme.

### 3) Expected outputs of the project:

Compilation and spreading of information on harmful algal occurrences worldwide. Accessability in a format that allows easy search of data. In the long term improvement of the basis for studies of long term trends in harmful algal occurrences.

### 4) Feasability:

The advisory group could be the ICES-IOC WG on the Dynamics of Harmful Algal Blooms, which already compiles the ICES National HAB Reports. The IOC Science and Communication Centres on Harmful Algae in Copenhagen and Vigo will be in operation until at least the year 2000 and 2001 respectively, and is offering its staff and facilities. Scientific supervision of data is offered by Dr. Jorge Diogène from the IOC-IEO Scientific and Communication Centre on Harmful Algae in Vigo. The mapping exercise carried out by C. Belin IFREMER is a natural component of the information database. Collaboration with other scientists/institutions are welcome. The results of the ICES-IOC Survey (1995-96) on HAB monitoring practices worldwide will be included in the information database.

### 5) Proposed steps:

5-a) To consult the ICES/IOC WGHAB in order to define the proposal in relation to:

- potential users of the service
- potential countries involved (ICES, extension to non-ICES countries)
- input requirements and format (which data/information) (Algal blooms, toxic events,...)
- mechanism for data gathering
- information output format (data, text, tables, graphs, summaries, ...)
- appropriate software to use
- other...

5-b) To gather data on harmful algal episodes in marine systems. This would include the annual reports compiled in the past by ICES, and would subsequently expand to include data from outside the ICES area.

5-c) To make periodical updates of the data base and to facilitate the transfer of information either by internet or by "snail-mail".

6) The data base:

Name:

The name of the data base will be: IOC-ICES Harmful Algal Bloom Data Base (HABDAT)

### Potential users of the service:

Any professional or institution working in relation to HAB. Scientists studying global trends on HAB may especially be interested.

<u>Potential countries involved:</u> ICES and IOC member states. The starting will be the ICES National Reports on HAB. The aim is to have a data base with global coverage.

### Input requirements and format:

The information introduced in the data base will be the one presented in the ICES National Reports Cards. The information from previous national reports will be included. The base will include both algal blooms and reports on toxic events.

### Mechanism for data gathering:

Information data will for the ICES area be collated through the WGHABD. A mechanisk to up-dta more than once a year will be explored. For IOC member states which are not ICES members the IUCES format will be used and the Intergovernmental Panel on Harmful algal Blooms will be used as the appropriate mechanism.

### Information output format (data, text, tables, graphs, summaries, ...):

The information will be accessible by internet from an appropriate web-site of IOC and ICES.

### Software:

The IOC has access to the UNESCO data-base ISIS that is freely available, also for WWW use.

# ANNEX VI

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## PARTICIPANTS TO THE KRISTINEBERG WORKSHOP

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### ANNEX VII

### NATO Advanced Study Institute on the Physiological Ecology of Harmful Algal Blooms (Co-sponsored by IOC and SCOR)- Don Anderson

This Advanced Study Institute (ASI) was planned and implemented by SCOR/IOC Working Group # 97 on The Physiological Ecology of Harmful Algal Blooms. The WG was asked to assemble and evaluate information on the ecology, physiology, and bloom dynamics of harmful phytoplankton and to assess the present state of knowledge and identify issues that need priority attention. This is one step towards a better understanding of the apparent worldwide increase in the frequency and geographical extent of harmful algal blooms.

The Terms of Reference for SCOR/IOC WG # 97 were:

- 1. To review and analyze data on the physiological ecology and biochemical aspects of harmful algal blooms, especially those resulting in toxic episodes, paying particular attention to nutritional, environmental and physiological factors.
- 2. To assemble the WG's findings and submit for publication a report, summarizing the state of knowledge and identifying the areas of future research.

At the first meeting of the WG in October 1993 at L'Houmeau, France, the decision was made that the best means for meeting these objectives was to apply for funds to convene a NATO Advanced Study Institute. An ASI is a "post-doctoral course of approximately 10 days duration, with an attendance of 12 to 15 lecturers and 60 to 80 participants. It is a high-level teaching activity with a carefully defined subject systematically presented, and treated in depth by lecturers of international standing". One advantage of an ASI for this WG was that it could fulfill the Terms of Reference by providing support for a major workshop, involving many more than just WG members, as well as covering the costs for the publication of proceedings in the NATO ASI Series. The venue for the ASI was the Bermuda Biological Station for Research (BBSR), a facility with adequate lecture hall space, fully equipped teaching laboratories, easy access to the ocean, and housing and dining facilities on-site for all participants. Following the first WG meeting, the chairman wrote a proposal to NATO requesting funds for the meeting. The proposal was accepted and approximately \$90,000 awarded. SCOR and IOC both contributed \$15,000 each to the budget.

The ASI was held at the BBSR from May 27 - June 6, 1996. There were 87 lecturers and students from 28 countries, distributed according to the requirements of NATO. The 34 invited oral presentations were divided into two Themes: "Autecology" and "Ecophysiological Processes and Mechanisms".

**General Recommendations:** In addition to the specific research recommendations listed below, ASI participants agreed upon several recommendations to SCOR and the IOC. These are: **Patrick - I will supply these later when I get back to my office computer. They include recommendations for one or two new SCOR Working groups. ****

### **Research Recommendations**

During the ASI, participants gathered into working groups in order to carry out the task of identifying key areas of research. The topic areas for these groups were agreed upon in Plenary session discussions. In carrying out this task, groups were asked to write statements indicating: 1) our present state of knowledge of a particular research area, with a justification as to why that area is important, and 2) our view of the future direction of the research, with a bulleted list of suggested approaches for achieving the goals. These recommendations are listed below (in random order).

### 1. Mixotrophy by HAB species

**Justification:** It is now clear that many HAB species have the potential for mixotrophic nutrition. The ability to utilize both photosynthetic and heterotrophic pathways may give species of mixotrophic HAB a competitive advantage over strictly autotrophic members of the phytoplankton. Thus, we cannot understand bloom dynamics, the effects of eutrophication on plankton communities, or life cycle dynamics of key HAB taxa without thoroughly understanding the forces driving mixotrophy and the extent to which autotrophic processes are supplemented by heterotrophy.

### **Recommendations:**

- [°] Develop reliable methods to detect and quantify mixotrophic nutrition
- [°] Determine the mixotrophic potential of key HAB species

- [°] Identify factors that induce or enhance phagotrophy in photosynthetic organisms
- ^o Investigate the effects of natural and anthropogenic eutrophication on HAB mixotrophy
- ° Evaluate the effects of mixotrophic nutrition on toxicity of HAB species
- [°] Assess the significance of mixotrophic HAB on the structure and function of marine food webs

### 2. Thin layers and microphysics of the pycnocline

**Justification:** Different strategies lead some species, amongst which a large proportion of the harmful species, to accumulate into thin layers (some tens of centimeters) associated with the pycnocline for long periods of time. Spatial scales in these features have not yet been investigated but there is evidence that they can extend, at least, to the 10 km scale.

For different species, greater knowledge of behavioral responses responsible for such accumulations or confinement would allow better understanding of HAB dynamics. Detailed studies of a few target species might lay the basis for typifying functional groups of species. Such detailed studies would thus also provide insights into biomodification of the physico-chemical environment.

Although all the detailed processes leading to and maintaining these thin layers are not yet understood or even described, the displacement and maintenance of these layers are of paramount importance to the prediction of harmful events. The behavior of these boundary layers has not yet been sufficiently investigated by physicists (for example in cross-shelf and along-shore transport).

### **Recommendations:**

- [°] Characterize the microphysics and chemistry at the viscous and intermediate sub-ranges in order to quantify the various processes influencing the development of HABs.
- ° Characterize the processes leading to the formation and the erosion of thin-layers.
- ^o Develop an understanding and modeling of mesoscale advection in thin layers in relation to cyclic forcing, weather events and biologically mediated reduction of turbulent viscosity (through turbulence suppression, e.g. by differential solar heating of phytoplankton layers, and/or by viscoelastic polymer secretion).
- [°] In order to understand 3D-movements of these features, development of appropriate instruments is necessary (density-adjusting floats, fine-scale profilers, particle analyzers, video systems, in situ rheometers).

### 3. Macronutrients

**Justification:** There has been an increase in anthropogenic nutrient inputs to many coastal areas. Correlative evidence indicates that HABs have increased in response to elevated nutrient loading in localized regions (e.g., North Sea, Baltic Sea, Seto Inland Sea, Tolo Harbor). Some HABs have also been related to natural nutrient dynamics (e.g., upwellings) in coastal areas. The relationship between macronutrients and mechanisms of HAB formation/maintenance remain to be determined. In particular, it is unknown whether HAB species have unique nutrient utilization capabilities and strategies which allow them to out compete other species, or whether, in the presence of high nutrient availability, the occurrence of HABs is controlled by grazing pressure. Other unresolved issues include the role of nutrient supply ratios in selecting for HAB species, and the importance of DOM either directly, as a nutrient source, or as an indirect promoter of HABs.

### **Recommendations:**

- ^o Determine the nutrient utilization capabilities and strategies of selected HAB species in comparison with cooccurring phytoplankton. Interactive field and laboratory studies are essential. Field studies may require the development of new technologies for focusing on individual species in conjunction with more traditional approaches.
- ^o Determine how nutrient concentration, composition, ratios, and cycling influence the occurrence of HABs. Retrospective analysis of long-term data available in some regions or evidence preserved in cores can provide information about the past occurrence of HAB in areas where nutrient inputs have increased. Process-oriented

field studies of current HABs, especially recurrent, predictable HABs where all phases of the bloom dynamics can be studied, are essential.

- ^o Evaluate the role of DOM as either a direct source of nutrients which HAB species can utilize or as an indirect stimulant of HABs through interaction with bacteria and the microbial loop.
- ^o Investigate the relative importance of top-down control in regulating species composition of HABs.

### 4. Population genetics/biogeography

**Justification:** Morphological features and life histories are the primary criteria used to distinguish phytoplankton species. However, a single group of organisms defined in this way a species may include multiple genetic variants or strains. Definition of species and their associated infraspecific variants is critical for understanding the basis of biodiversity, toxin production, physiological optima and tolerances and origins of HABs on local and global scales. The presence of genetic variation even within populations of apparently unspecific blooms creates serious problems for studies based on clonal cultures because it is unclear how representative these cultures are of natural populations.

### **Recommendations:**

^o There is an urgent need to expand the study of genetic variation in HAB species with particular reference to the roles of biogeography, genetic isolation and long-term changes in genetic diversity. Molecular tools are particularly useful for these studies. Currently available techniques are limited and progress is slow because of the small number of investigators in this area. More technology needs to be developed, particularly those techniques that bridge the critical gap between lab investigations and natural populations. More training opportunities need to be provided for phytoplankton ecologists to learn these tools.

^oThe requirement of morphotaxonomy to provide a platform for species identification and genetic comparisons necessitates a renewed effort in this area. The impending loss of expertise in morphotaxonomy creates a need for training new people in traditional as well as advanced methods of recognition of morphospecies.

^oSupport for culture collections is declining. However, a primary requirement for the study of genetic variation among HAB organisms is the availability of multiple isolates of the same species from different geographic areas. Recognition of the importance of culture collections is required and more financial support is needed to maintain and expand them.

^o Physiological studies should recognize the potential for variability for such parameters as environmental optima and tolerance ranges, quantity of toxins and toxin profiles because the patterns of variation may yield useful insights into the relative importance of adaptive factors controlling the distribution of organisms. More technology, such as automated counting methods, need to be developed that facilitates the examination of larger sample sizes.

^oMultidisciplinary approaches to the study of genetic variation can yield critical insights. Support for multidisciplinary investigations that examine HAB species from both morphological and subcellular perspectives should be encouraged.

^o There is provocative evidence that anthropogenic dispersal of HAB species has occurred. The introduction of harmful organisms may be accompanied by the introduction of harmful genes that may be incorporated into native populations. Further assessment of the extent of role of human-assisted dispersals and their occurrence in the past needs to be conducted.

### 5. Freshwater/stratification

**Justification:** Some degree of vertical stability is essential for the development of phytoplankton blooms. Such stability can be provided by density gradients caused by heat or by low salinity inputs. Examples of the latter include estuaries and buoyant coastal currents. However, freshwater input also brings with it a supply of poorly characterized, land-derived nutrients: macronutrients, such as nitrogen and phosphorous from agricultural and industrial/domestic sources; micronutrients including metals and vitamins; and, dissolved and particulate organic materials. The latter may serve as growth promoters (e.g. chelators) or directly as nutrients.

The linkage between HABs and freshwater input has been established in many areas (e.g. *Phaeocystis* blooms associated with Rhine River discharge, *Heterosigma* blooms in the Strait of Georgia, *Alexandrium* blooms in the southwestern Gulf

of Maine coastal current, and *Pyrodinium* blooms along river plume fronts). Research is needed to understand the mechanisms underlying this linkage and to elucidate the relative importance of the many factors which may be operating.

### **Recommendations:**

- ^o Determine in situ growth rates and physiological status of algal species within and outside areas of freshwater influence as they relate to the chemical constituents of the water;
  - ° Determine the relative importance of physical effects and algal behavior vs. direct growth stimulation;

[°] Elucidate the mechanisms underlying blooms occurring within low salinity water masses vs. those occurring at the boundaries of water masses;

° Develop an understanding of the influence of the timing of freshwater inputs and stratification to bloom dynamics

### 6. Small scale physics, behavior, and photosynthesis

**Justification:** We have a conceptual model, often termed the Margalef mandala, that provides a framework for describing how physiology, behavior and hydrography interact to promote or maintain algal blooms. We also have good descriptions of algal blooms in several well-studied regions. However, we are not yet able to couple sufficiently detailed information on algal physiology and behavior into realistic hydrodynamic models to describe single species bloom events in natural water columns. Consequently, our present numerical models are not robust in the real world.

### **Recommendations:**

- [°] Incorporate physics into all HAB initiatives to provide information on the environmental conditions that support bloom development, maintenance and decline.
- ^o Develop effective methods to characterize how organisms behave under the influence of variable environmental factors.
- [°] Develop appropriate methods to characterize photosynthesis and nutrition under relevant, natural hydrodynamic variability. Conventional culture techniques are not sufficient.
- [°] Improve coordination among physiological/behavioral characterizations, biological-physical modeling, and quantitative descriptions of hydrography and natural communities.

### 7. Ecophysiology of toxin production in HAB species

**Justification:** Toxic secondary metabolites associated with harmful blooms in aquatic ecosystems are produced by several groups of photosynthetic eukaryotic and prokaryotic microorganisms, including non-photosynthetic bacteria. To date, research on these compounds has focused on structural elucidation, detection methods and mechanisms of action. The next major research efforts required for understanding the physiological function of toxin production are rigorous investigations of their biosynthetic pathways and genetic regulation. Some data are available on these topics but we have only begun to understand these processes.

The effect of environmental factors on production of marine and freshwater biotoxins (known as phycotoxins when derived from algal sources) has not been investigated in sufficient detail for any of the toxin groups, but some information is available for a limited number of strains of toxigenic species. For example, production of toxins associated with PSP (paralytic shellfish poisoning) has been studied in *Alexandrium* spp., *Pyrodinium bahamense*, and *Gymnodinium catenatum*, and there have been several investigations on the production of NSP (neurotoxic shellfish poisoning) toxins by *G. breve*, DSP (diarrhetic shellfish poisoning) toxins in *Prorocentrum* spp., anatoxin in *Anabaena* spp., and domoic acid, the toxin which causes ASP (amnesic shellfish poisoning), by *Pseudo-nitzschia* spp.

Biosynthetic schemes for certain PSP toxins (saxitoxin and neosaxitoxin) have been proposed and the structures of many analogues have been elucidated, yet the details of the synthetic pathway(s) and genetic regulation among the different toxin-producing organisms remain largely unknown or hypothetical. In contrast, the structure of ASP toxin (domoic acid) is known and its biosynthesis is well described - we can now ask probing questions concerning its regulation and physiological importance. A total understanding of the function of these toxins will only be achieved when we determine

their structure, biosynthesis, genetic regulation and ecophysiology. Emphasis should be placed on the functional roles of toxins in the primary source organisms, e.g. do these toxins serve an intrinsic or an extrinsic function? Understanding the genetic basis for toxin production will also provide valuable insights into their ecophysiological role in bloom dynamics.

### **Recommendations:**

- Determine the kinetics of growth, toxin biosynthesis, and interconversions.
- Identify the cellular location of the toxins and the timing of their
- production in the cell cycle.
- Characterize the components of the toxin biosynthetic pathways.
- Determine if toxins are directly involved in predator-prey interactions.
- Determine if toxins serve an allelopathic function against other algae, bacteria or fungi, including the role of other compounds in stimulating or inhibiting the effect of the toxins.
- Determine the critical interactions between extrinsic environmental factors and the genome in regulating toxin biosynthesis, catabolism and sequestration (phenotypic vs. genotypic variation in HAB species).
- Model the production of toxins for extrapolation to natural populations. Determine the evolutionary origin of the toxin biosynthetic genes.
- Develop and maintain international sources for toxin standards and reference materials.

### 8. Bacterial-algal interactions in HAB population dynamics, cellular growth, and toxicity.

**Justification:** Co-existing with algal communities containing HAB species are microbial assemblages which, like the algae, undergo changes in species composition, exhibit nutrient competition, and produce bio-active compounds, including toxins. These and other microbial processes are influenced by the algal community and, in turn, can potentially influence the population dynamics, cellular growth, and toxin characteristics of the algae. It is now clear that bacteria can produce both intracellular and extracellular compounds with algicidal as well as growth inhibiting or stimulating effects. Moreover, available evidence demonstrates that bacteria not only can modify the toxicity of algal cells, but can themselves synthesize certain `algal' toxins.

### **Recommendations:**

We must characterize natural bacterial assemblages associated with HABs, and establish whether production of growth/toxicity modifying substances and/or toxins occurs on temporal and spatial scales permitting them to influence algal bloom dynamics, cellular growth, and toxicity. It is also essential to determine whether bacteria constitute an important source of 'algal' toxins in the context of trophic transfer and seafood safety issues. Suggested approaches to achieve these objectives include:

- Apply classical and molecular approaches to characterize the temporal/spatial association of bacterial assemblages with HABs, and define the systematic as well as functional relationships between bacteria and these algae.
- Isolate and characterize bacterial and algal bio-active metabolites, and determine their respective effects on bacterial and algal growth and/or toxicity, including elucidation of the mechanisms involved and the patterns of synthesis in natural communities.
- Evaluate the role of toxigenic bacteria as a point of entry for the transfer of 'algal' toxins between trophic compartments in aquatic food webs, and assess the contribution of these microbes to the toxification of fisheries resources.
- Determine the distribution of toxin genes among bacteria and extrachromosomal elements (e.g., plasmids, viruses) as a means of explaining the phylogenetic diversity of organisms capable of synthesizing certain 'algal' toxins, and identifying potential mechanisms for the lateral transfer of these genes.

### **10. Trace Metals and Chelator Interactions**

**Justification:** Trace metals including iron, manganese, cobalt and selenium have been hypothesized to limit the production of harmful algal blooms. Examples include limitation of *Alexandrium tamarense* (Gulf of Maine), *Aureococcus anophagefferens* (Peconic Bay), *Heterosigma sp.* (Osaka Bay) and Chattonella antiqua (Seto Inland Sea) by iron, the limitation of *Chrysochromulina sp.* (Kattegat) by cobalt and the inhibition of *A. tamarense*, and *C. antiqua* by copper. In most cases, the importance of a single trace metal was determined when the addition of that particular metal stimulated the growth of a HAB organism in culture.

In recent years, the importance of chemical speciation and organic complexation in the open ocean has revolutionized our view of how trace metal chemistry affects phytoplankton growth dynamics. Current data on trace metal concentrations in coastal waters, along with information on

chemical speciation and biological interactions indicate that it is plausible that trace metals influence the growth and species composition of coastal phytoplankton communities. This new information on trace metal availability now needs to be applied to the development, density and frequency of harmful algal blooms.

### **Recommendations:**

^oObtain more detailed information on trace metal concentrations and speciation in coastal waters.

• Determine techniques to estimate their bioavailability to harmful algal bloom species.

°Examine the mechanisms of uptake.

°Examine the effects of multiple nutrient limitation.

°Determine the minimum cell quotas for trace metals.

°Examine the effects of trace metal limitation on cellular biochemistry including toxin biosynthesis.

°Examine the importance of bacteria trace metal interactions.

#### 11. Life Cycles

**Justification:** Knowledge of life cycles and cell cycles is fundamental to the understanding of formation, maintenance, and decline of harmful algal blooms. The water column or benthic origins of bloom inocula, cell growth, bloom termination and long-term species are mediated by poorly understood or largely unknown life cycle event. Most HAB species have benthic phases, which are believed to be important in species survival and persistence, but control on interactions between planktonic and benthic phases remain to be elucidated.

### **Recommendations:**

- ^o Elucidate life histories for all HAB species (including asexual and sexual reproduction).
- ° Develop rapid and reliable molecular/immunological probes for detection of life cycle stages, leading to improved and/or automated recognition.
- ° Describe cell cycle morphologies (for example, division stages) facilitating calculation of in situ growth rates.
- ^o Determine endogenous cell cycles, for example, the gating of cell division on a diurnal basis and annual cycle of excystment.
- ^o Identify the specific nutritional and environmental triggers as well as genetic controls involved in stage transformation, and the conditions required for successful transformation.

° Determine the survival and metabolic activity of benthic stages.

### **12. Emerging techniques and technology**

**Justification:** An understanding of HAB phenomena requires the detection of causative species and the toxins they may produce. Analogous problems exist in biomedical diagnostics, and many techniques and technologies used for those purposes are applicable to HAB research. For example flow cytometry, antibody and DNA probes, phycotoxin-specific receptor binding assays have been transferred and used successfully in the laboratory and, to varying degrees, in field studies. The interfaces between traditional disciplines have often yielded scientific breakthroughs, thus interdisciplinary collaborations should be fostered to solve the difficult issues relevant to understanding HAB phenomena. Programs should be developed that foster cross-training and collaborations between biomedical and HAB researchers.

### **Recommendations:**

Inexpensive, simple, efficient, and accessible methods for identification and detection of all HAB species and toxins must be developed. Methodologies and tools should be made widely available to the field as a whole as they are developed. Confirmation of the efficacy and utility of new techniques in the field requires multi-disciplinary investigations that could include: taxonomy, physical oceanography, ecology, and biochemistry.

Develop probes and assay methods for HAB species identification. DNA and antibody probes have been developed and tested in the laboratory for a limited number of HAB species. These probes must now been tested in the field and new probes developed for other species.

^o Develop new methods for toxin detection. Receptor assays for toxins in HAB species have been demonstrated in the laboratory. These assays now need to be tested for utility in natural populations.

° Develop methods for the measurement of in-situ growth rates. Methods for determining in situ growth rates are needed in order to understand the mechanisms driving HABs. Flow and imaging cytometric methods have been demonstrated to be useful in the field and show promise for HAB species.

[°] Develop probes for physiological status. Many fluorescent probes for measuring intracellular conditions have been developed for mammalian systems. Application of these tools to algal physiology may yield new understanding of the regulation of HAB population dynamics.

Since the choice of reference taxa is critical due to potential genetic variability, culture repositories should be supported and expanded internationally.