International Council for the
Exploration of the Sea

C.M. 1993/L:7
Ref. C

REPORT OF THE ICES-IOC STUDY GROUP ON THE DYNAMICS OF ALGAL BLOOMS (SGDAB) AND THE JOINT MEETING OF SGDAB AND THE ICES WORKING GROUP ON SHELF SEAS OCEANOGRAPHY (SSOWG)

Charleston, USA, 8-11 February 1993

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REPORT OF THE ICES-IOC STUDY GROUP ON "THE DYNAMICS OF ALGAL BLOOMS"

Charleston, 8-9 February 1993

1. OPENING OF THE MEETING

1.1 Welcome to the participants

The meeting was convened at 0930 on 8th February 1993 with preliminaries and welcoming remarks from Dr. Robert Kifer, Director of the Southeast Fisheries Science Center and the Study Group Chairperson, Ms. Beatriz Reguera of the Instituto Español de Oceanografía (Centro Oceanográfico de Vigo).

1.2 Approval of the Agenda

The approved agenda for the meeting of the Study Group on the Dynamics of Algal Blooms in Coastal Oceans (SGDAB) (**Annex I**), and a list of participants (**Annex II**) are appended to this report.

1.3 Election of Rapporteur

Dr. John Smith (Canada) agreed to act as a rapporteur.

2. TERMS OF REFERENCE

At the LXXX Statutory meeting of ICES (Warnemunde, Germany, September 1992) the Biological Oceanography and the Hydrography Committees recommended that the Study Group on the Dynamics of Algal Blooms:

a) continue and, if possible, finalize discussions on a programme for investigating the dynamics of harmful algal blooms in the ICES area; methods to be used should be discussed and the timetables for the development of pilot studies outlined;

b) describe algal population dynamics in relation to hydrodynamic processes;

c) assess the utility of mesocosm experiments for the understanding of algal population dynamics;

d) examine the cyst phase in the life histories of relevant, potentially harmful algae;

e) coordinate work on algal blooms with the activities of the Programme of Harmful Algal Blooms adopted by the Joint *ad hoc* IOC-FAO Intergovernmental Panel on Harmful Algal Blooms.

A joint session of this Study Group and the Working Group on Shelf Seas Oceanography was held from 10-11 February 1993.

This meeting was co-sponsored by IOC represented by Drs. Henrik Enevoldsen, Programme Coordinator, IOC, and Bernt Dybern, Chairman of the *ad hoc* IOC-FAO Intergovernmental Panel on Harmful Algal Blooms (IPHAB).

3. THE TITLE OF THE GROUP: ALGAL BLOOMS/HARMFUL ALGAL BLOOMS

The Chairperson noted the change of name of the Study Group since the last meeting when the term **Harmful** was included in the title. The present Study Group was established **to look at the dynamics of harmful algal blooms and to study fundamental phenomena where true progress in understanding these events can be made**; the group should continue to focus on dynamics. It was suggested that the separation of bloom dynamics from the management of their effects is somehow artificial. But hydrographers are likely to have little interest in toxin chemistry and other such aspects of harmful algal blooms and to attract their interest to the problem, it is necessary to maintain a focus on dynamics. From the practical point of view **it is not possible to include every aspect of the harmful algal bloom problem in one Working Group**. Following further discussion during agenda items 3.1 and 3.2, these matters became agenda item 6 and 7 of the joint meeting with the Working Group on Shelf Seas Oceanography.

4. THE UTILITY OF MESOCOSM EXPERIMENTS FOR THE UNDERSTANDING OF ALGAL POPULATION DYNAMICS

This session was led by T. Smayda with the help of E. Granéli and F. Colijn. These three scientists gave short talks about the state of the art of mesoscosms experiments, applied to research on noxious algal species, in their countries. A document concerning mesocosms as enclosed experimental ecosystems was presented and appended to this report as **Annex III.**

4.1 Mesocosms experiments in The Netherlands

Mesocosms are employed in the Netherlands (Colijn) to study how reduced P and N loads resulting from sanitation programmes on the Rhine River, which have decreased nutrient inputs into the North Sea by 50%, are affecting eutrophication in the Southern Bight of the North Sea. Their advantages include their use as a substitute for scarce ship time, ease of manipulation and the possibility of towing to alternate sites, and ease of controlling nutrients and mixing to achieve standard conditions.

Mesocosms are considered to be simplified ecosystems and lend themselves readily to mathematical modelling. They are used in conjunction with laboratory studies of *Phaeocystis*

and new techniques of immunolabelling of harmful species and flow cytometry for counts etc. Preliminary work with these mesocosms includes studies of light climate, mixing, reproducibility and design. Sanitation programmes on the Rhine as elsewhere have succeeded in removing relatively more P than N since N has much more diffuse sources; the effects of a relative N surplus are being studied first, with P surplus studies to follow. Zoobenthos grazing and energy transfer to the benthos can be studied in conjunction with these mesocosms. The mesocosms are constructed of black plastic with diffusers on top; irradiance falls rapidly to 1%, giving a steep light gradient, but with some radial effect in light intensity measurements. When mixed, these mesocosms provide reproducible growth measurements, species composition and succession patterns. Diel cycles in pH, oxygen and fluorescence are observable.

Mesocosms currently in use on the Elbe River were illustrated and recommended as suitable for the proposed pilot studies. There are no fouling problems with this design. Dinoflagellates have not yet been tested in these mesocosms.

4.2 Mesocosms experiments in Sweden

Mesocosms were first used in freshwater in Sweden (Granéli), originally to avoid advection effects. These mesocosm studies are large undertakings and require a great deal of teamwork. The Lund group is now studying factors underlying harmful algal blooms by means of such mesocosms. The setup consists of an above ground swimming pool which contains a number of large tubes or mesocosms. The possible rôle in harmful algal blooms of eutrophication, acid precipitation, higher trophic level changes and humic substances are being investigated. With eutrophication there is the question of what is the limiting nutrient for growth. In Sweden, 90% of P has already been removed from sewage and there is a programme to reduce N by denitrification and decreasing agricultural N runoff. The Lund group is now looking at the effects of various N/P/Si ratios since Si levels have remained fairly stable in the face of anthropogenic changes in N and P.

In terms of top down control of phytoplankton species assemblages, it is known that an increase in the predators of copepods yields decreased grazing pressure on the diatom size class. Thus, for spring and early summer phytoplankton assemblages, mesocosms containing jellyfish which prey on copepods yield large diatom populations, the same result obtained by removing copepods from the mesocosms, while adding extra copepods yields populations of very small phytoplankton. For autumn phytoplankton populations, copepods graze dinoflagellates very poorly; thus controlling the copepod population yields mixed diatoms and dinoflagellates while increasing the copepods leads to dinoflagellates only.

It has been observed that humic substances are increasing in freshwater discharge in Sweden. One hypothesis is that acid rain results in no fertilization of the forest and the increased humics represent increased levels of decomposition. Another view is that the acid rain is washing out residual humics from deep in the soil. It is not known whether there is direct use of humics by phytoplankton or whether UV radiation decomposes humics into smaller molecules which are then attacked by bacteria giving ammonia, amino acids, etc. which are then utilized by the phytoplankton. Microcosm experiments with added humic resulted in *Gyrodinium* population increases. Replication found using these microcosms is good. A new large mesocosm is being developed. Mixing in these mesocosms greatly decreases the dinoflagellate component. Stratification of these units is possible.

4.3 Mesocosms experiments in U.R.I. (U.S.A.)

University of Rhode Island mesocosm experiments (Smayda) were described, and it was concluded that it is necessary to scale the work to biological factors. The URI 13 m^3 mesocosm systems provide for mixing, stratification (by differential heating) and sediments; these systems exchange 4% of their volume per day with the bay. N nutrients are rapidly consumed in control tanks. The effect of sediments is very important, ammonia utilization can be forced, and successions can occur without changes in nutrients. In continuous cultures, nutrient ratios affect the outcome of competition. Species composition is governed by nutrients and grazers and probably by benthic processes. These mesocosms are probably suitable for nutrient enrichment studies with dinoflagellates.

4.4 Discussions and conclusions

Following these presentations on mesocosms, it was suggested that reproducibility and similarity to the natural environment should be criteria for ecological extrapolation from mesocosm experiments. Other participants wished to refocus on the problem of offshore blooms entrained in coastal currents as a means of simplifying the problem; it was stressed that what we need to know is how large the total population of interest is, and by applying a known growth rate, to estimate how long it will take the population to grow to a given size. Some participants doubted whether experiments work for species other than diatoms. Others felt that mesocosms are useful; since they can provide good vertical migration and growth rate data, provided the records are of short duration.

Long-term mesocosm studies were thought to be less valuable bacause of artifacts which develop with time. It may be necessary to capture an early stage bloom and to fill new bags at intervals throughout the bloom period. It is also necessary to simultaneously observe developments in the field. It was pointed out that mesocosms have been used successfully to study the dinoflagellate *Gyrodinium.*

It was generally agreed that growth rate measurements are important and should be one of our priorities, and that mesocosms provide a suitable tool for these measurements.

It was agreed that the tasks of the Study Group needed to continue in a more permanent forum. Possible terms of reference for a new Working Group to replace the Study Group were considered. It was decided that an *ad hoc* group of participants would prepare draft terms of reference for a new Working Group.

5. THE CYST PHASE IN THE LIFE HISTORIES OF RELEVANT, POTENTIALLY HARMFUL ALGAE

The aim of Agenda Item 7 was to examine the cyst phase in the life histories of relevant, potentially harmful algae to determine whether this stage is important in controlling the magnitude and timing of toxic blooms. This session was led by D.M. Anderson.

The need to confirm the existence of cyst or spore stages for harmful algal bloom species was noted. Harmful algal species known to have cyst stages were listed. For PSP producers,

this includes various species of *Alexandrium* and *Gymnodinium catenatum* but for *A. ostenfeldii* and other species of *Gymnodinium/Gyrodinium*, cysts have not yet been identified. The life history of *Phaeocystis* is unknown but temporary cysts now appear to have been identified for this species. No species of *Dinophysis* has a cyst stage proven to date, but growing evidence of sexual reproduction and encystment processes within the genus are being found. *Heterosigma* has a resting stage and *Chattonella* has had a cyst stage identified.

It is known that nutrient depletion can result in the induction of sexuality in *Alexandrium* in the laboratory but it is not known whether this occurs in the field. Future work on induction needs to include studies on endogenous (i.e., an internal clock) versus exogenous control. In some species, there is a mandatory period of dormancy once a cyst is formed before it can germinate; for *Gymnodinium catenatum* this period is less than 2 weeks, while for *Alexandrium excavatum* this is about 4 months; this period needs to be determined for other species.

Cyst survivability during gut passage needs to be determined; this process is sometimes mandatory for germination in non dinoflagellates. The control of quiesence following dormancy also requires further study to determine whether cues for germination are internal or external. Other relevant work in the case of *Alexandrium* is to determine how field distributions of cysts correlate with bathymetry, sediment type, water movement and bloom incidence. For population dynamics studies, we need to know germination rates and total cyst population sizes. Germinated cysts do not always appear to grow under ideal temperature conditions, etc. We also need to determine encystment rates, the toxicity of the cysts themselves (are they more or less toxic that the vegetative cells?), and the effects of resuspension. Induction of sexuality can be studied best in laboratory cultures. Mandatory dormancy can be studied using cysts of known age from cultures, sediment traps or tows. Survival following ingestion can be studied by obtaining cysts from natural faeces and laboratory experiments. Control of quiesence would be mainly laboratory work.

A detailed document discussing the different aspects of the cyst phase and the experimental to be carried out is appended to this report as **Annex IV**.

6. PILOT PROGRAMMES OUTLINE

6.1 Goals of the meeting

It was noted that the various pilot projects to be discussed at present have only regional representatives (e.g., no North American representatives in the Iberian Peninsula programme) and the question was raised as to internationalize the various programmes. The need was also expressed that the methods that will be used to measure variables such as growth rate etc. need to be clarified.

Next, the goals of the meeting were commented on. It was stated that the IOC requires results which will bring project implementation closer, and that means need to be found to encourage international cooperation in field programmes. An additional joint project was suggested, by a mixed international group; IOC could contribute to such a project particularly

if it were to be located in a developing country. The group was canvassed for suggestions for such projects.

6.2 Suggestions for an international intercomparison exercise

A joint intercalibration study of *in situ* growth rate was suggested, where investigators would come to a single laboratory or site and try out their various methods; this would also have the virtue of generating less paperwork than full scale meetings. Some participants preferred the terms *intercomparison* or *parallel analysis* to *intercalibration.*

Bahía Fosforescente in Puerto Rico was proposed as an ideal site for such an exercise because of the regular occurrence of blooms there, and the fact that it is nearly enclosed, making it rather similar to a large mesocosm. **It was emphasized that it is necessary to have a population dynamics model in mind in order to interpret the data and carry out the study; that is, we need to show the ability to produce results, not just to run experiments**. It was agreed by some participants that Bahía Fosforescente should allow good estimates of population dynamics parameters, and pointed out that a good amount of data probably already exists or can be estimated for this location. The proposed study might facilitate quantification of detailed structure and processes, but we probably cannot model the details yet. Red tides in B. Fosforescente are thought to be the result of physical accumulation of non-grazed, slow growing species, and the hypothesis that regular bloom mechanisms involve the outgrowing of predators could be tested.

All the proposed pilot studies include a mesocosm component but regular studies need to be carried out as well, and for this regular occurrence of blooms is needed in order that they be suitable for group study. A "strategic" approach was also suggested, rather than methodological and biological studies; the workshop methods utilized by other organizations was noted. The group next considered where and how to have a joint international project to study particular aspects of the problem such as ***in situ* growth rate measurements.** A discussion ensued in which logistical characteristics of various sites relative to their possible scientific merits were considered. Some participants argued that the first intercomparison exercise should be carried out in one of the pilot study regions with their targeted species. Ría de Aveiro (Northern Portugal) was then suggested by the Portuguese participants as an ideal site since there is a permanent monitoring programme in place, harmful blooms occur with predictable regularity, the basin is enclosed so that advective exchanges with the adjacent coastal waters are easily measured, and a model of the circulation is being developed by the local University.

7. DISCUSSIONS ON GROWTH MEASUREMENTS AND INNOVATIVE DEVICES

A discussion took place of how to measure growth of phytoplankton assuming a system whose limits are defined. Mitotic index methods were suggested, but many species are not amenable to this kind of analysis. It is necessary to find organisms on which many methods such as mitotic index techniques can be practised and a short list of species of interest, such as *Dinophysis* and *Pyrodinium* was noted. It should be noted that in the first meeting of this Study Group in April 1992, the question of novel methods of measuring growth was discussed extensively.

The group then turned to the questions of how to sample populations which are horizontally patchy, vertically stratified or vertically migrating. A short discussion ensued including suggestions of vertical and oblique net tows and other standard methods. The question of selective grazing of daughter cells was raised and the need for a comparative study of methods reiterated. The role of grazing was then pursued in the discussion. It was suggested the effects of pathogens, including virus infections, needed to be included in loss terms and, for appropriate size classes of harmful algae, the effects of microzooplankton grazing as well. There was then a short discussion of different methodologies for quantifying grazing, and it was pointed out that **we need to determine for the systems of interest whether the cells of harmful algae are grazed at all.** The Study Group may need to include experts on grazing if this should appear to be a significant loss term.

The properties of an instrument called the ***in situ* grain size analyzer** were described (Gentien). This instrument was developed in his laboratory and works on the principle of diffraction. It can classify particles in the size range 0.7 to 400 um in diameter, works to depths of 300 m and in suspended particulate concentrations of up to 450 mg/l. Particles are discriminated into 32 size categories and the comparison to Coulter Counter analysis of the same sample is good. The *in situ* grain analyzer can also measure particle numbers (concentration) and could be used synoptically to estimate the total number of particles in a system. It was demonstrated how this instrument was able to discriminate a very narrow layer of *Dinophysis* which could have easily been missed by bottle cast sampling techniques. It was noted that the addition of a fluorometer to the package would allow the confirmation of the presence of *Dinophysis* by its phycoerythrin signature.

Following this presentation, there was some comment on methods apparently under development including an underwater microscope and a *in situ* flow cytometry system. The subject of the use of antibodies and DNA probes was introduced to aid in counting and identification of the cells of interest. Sorting flow cytometers can provide samples of the cells of interest from a mixed population and these samples could then be used for physiological measurements. It was suggested that the Study Group exchange new methods each year at the meeting. The opinion was expressed that many of these new methods would prove quite unsuitable for operational monitoring purposes because of the high cost and the high level of sophistication of the instruments.

Some discussions of the terms "growth rate" and "population dynamics" followed to clear up possible confusion about the meanings of the expressions. It was pointed out that all the sites proposed for joint international study were intended to be physically uncomplicated in order to simplify the measurement of constants like growth rate. After discussions on three different sites (Bahía Fosforescente, Ría de Aveiro and the Gullmarfjord) Ria de Aveiro in Portugal was selected for a joint study in July 1994 with M.A. de Sampayo in charge of the local arrangements.

The circulation of a document prepared by the mesocosm group followed. Although there is some skepticism in the scientific community concerning these methods, the short-term use of mesocosms for rate constant determinations had already been agreed upon. It was pointed out that some harmful species will respond to nutrient loadings while others do not, and that not all certain harmful algal species can be studied in mesocosms.

8. PILOT PROGRAMME PROPOSALS DISCUSSIONS IN SUBGROUPS

The group split in three subgroups to have further discussions on the pilot studies and list questions to be forwarded to the hydrographers in the Joint Session.

9. RECOMENDATIONS FROM THE FIRST TWO DAYS

9.1 **Harmful Algal Blooms** presents problems quite distinct from those of normal blooms (due to their potentially serious economic and social impacts) and they therefore require special attention. The existing Study Group on the Dynamics of Algal Blooms should be transformed into an **ICES Working Group on HARMFUL ALGAL BLOOMS.**

9.2 Since mesocosms experiments can contribute to the understanding of at least some factors controlling algal populations there was agreement that **some boundary conditions might be placed on potential algal growth dynamics by experiments conducted with these devices.** The group encourages these and related mesocosm/microcosm studies, with particular emphasis on the utility of such experimental approaches to resolve representative HAB issues.

9.3 The potentially important rôle which cysts can play in population dynamics was accepted, since it is obvious that **excystment and encystment rates can influence parameters values in the population growth equations.**

9.4 The group agreed that the most important parameter to estimate was the **algal division rate (μ)**, and that a place should be chosen where the flux rates due to physical processes should be amenable to easy measurement. It was also agreed that a site should be chosen where the probability of encountering a toxic species is undeniable. It was proposed that an **ICES Workshop for Intercomparison of Methods for Measuring Dinoflagellates Division Rates *in situ*** should be carried out in Ría de Aveiro (Portugal) in the second half of July 1994.

JOINT MEETING OF THE ICES-IOC STUDY GROUP ON THE DYNAMICS OF ALGAL BLOOMS IN COASTAL WATERS (SGDAB) AND THE ICES WORKING GROUP ON SHELF SEAS OCEANOGRAPHY (SSOWG)

Charleston, 10-11 February 1993

1. OPENING OF THE MEETING

The ICES-IOC Study Group on the Dynamics of Algal Blooms (SGDAB) (Chairperson: B. Reguera, Spain) and the ICES Working Group on Shelf Seas Oceanography (WGSSO) (Chairperson: H. Dahlin, Sweden) met in the Charleston Laboratory of the Southeast Fisheries Service Center (SC, USA) from 10-11 February 1993. Hans Dahlin and Beatriz Reguera co-chaired the joint session. The list of participants is given in Annex VII. Following approval of the agenda (Annex VI), J.C. Smith (Canadá) was appointed rapporteur.

2. TERMS OF REFERENCE

The terms of reference for the SSOWG were:

a) Develop a programme for investigating the dynamics of algal blooms;

b) Plan and propose field experiments and modelling to increase understanding of the physical/chemical factors that influence the development of algal blooms.

For the SGDAB the terms were:

a) Continue and, if possible, finalize discussions on a programme for investigating the dynamics of harmful algal blooms in the ICES area;

b) Describe algal population dynamics in relation to hydrodynamic processes;

e) Coordinate work on algal blooms within the activities of the Programme of Harmful Algal Blooms suggested by the Joint *ad hoc* IOC-FAO Intergovernmental Panel on Harmful Algal Blooms.

The overall objective is to report on how to study harmful algal blooms, taking a long-term approach. It is stressed that the need is to define the important aspects of the programme since ICES needs a strong programme on harmful algal blooms to help governments understand the problems and devise appropriate means to deal with them (e.g., lowering nutrient concentrations is expensive and the need to do it must be clearly shown).

The need to focus on ICES requirements (studies of bloom dynamics and modelling) rather than on experimental details was emphasized. During discussion, attention was

directed to the diversity of bloom phenomena and the rôles of stratification and nutrients, as well as to the features associated with coastal currents which appear to be important in bloom formation. Attempts should be made to quantify the number and magnitude of such blooms to see whether they are increasing globally, and to determine the influence of anthropogenic modification of coastal waters in this process.

The use of the equations of population dynamics and collaboration with physicists would, it was felt, force useful confrontations with real systems. It was noted that special attention should be given to harmful algal blooms since that is in the ICES terms of reference.

3. INTRODUCTION

3.1 Current situation of the IOC/HAB Programme.

Participants were brought up to date on the IOC/HAB programme, originally developed in response to requests by several member states for help with harmful algal bloom problems. The current programme has a number of components, not all of which involve IOC participation. One component is the information network. Under this component the ***Harmful Algal News***, an IOC newsletter on harmful algae and harmful algal blooms, has been established and is publishing regularly. An editorial team composed of regional coeditors has been established. A ***Directory of Experts*** is to be published by the IOC in cooperation with NOAA (USA), with publication planned for the end of 1993. A ***Manual on Harmful Marine Phytoplankton***, in the series ***Monographs on Oceanographic Methodology***, is planned for publication in the first half of 1994. The IOC will also co-sponsor the ***VI International Conference on Toxic Marine Phytoplankton***, Nantes, France in October 1993. A proposal to establish a ***Scientific and Technical Cooperation Network on Harmful Marine Phytoplankton*** was submitted to the Commission of the European Communities, ***Human Capital and Mobility Programme***. Lastly, the IOC will seek the establishment of a ***Scientific Project and Communication Center*** on harmful algal blooms. Other developments include:

i) the ***IOC-Danida Training Course on the Taxonomy of Harmful Marine Phytoplankton***, 16-28 August 1993, at the University of Copenhagen, cosponsored by the Danish International Development Agency (Danida).

ii) an ***IOC/WHO/BMTC Training Course on Identification and Quantification of Algal Toxins***, to be held in Europe in the first half of 1994, cosponsored by WHO and the State of Bremen.

iii) an ***IOC Training Course on the Taxonomy of Harmful Marine Phytoplankton***, to be held in WESTPAC in 1993.

iv) Development of a training programme in relation to harmful algal blooms is also underway. A proposal for the development of modules and a feasibility study has been submitted to potential donors. Cosponsors of these courses are being sought.

v) The ***SCOR-IOC Working Group 97 on the Physiological Ecology of Harmful Algal Blooms*** will hold its first meeting in La Rochelle (France) 23-24 October, just after the VI International Conference on Toxic Marine Phytoplankton (Nantes, 18-22 October).

vi) A new staff member has joined the *Harmful Algal Bloom* team at the IOC Secretariat, and the upcoming second meeting of the ***IOC-FAO Intergovernmental ad hoc Panel on Harmful Algal Blooms (IPHAB)*** is already scheduled.

3.2 Results of the First Session of the IOC-FAO Intergovernmental Panel on Harmful Algal Blooms (IPHAB)

Attention was directed to a list of amendments to the programme in the report of the First Session of the IOC-FAO Intergovernmental Panel on Harmful Algal Blooms held in Paris, 23-25 June 1992. Following decisions of the IOC Assembly which took place 25 February-6 March 1993, the IPHAB will act as an advisory body to IOC and FAO.

A ***Programme Office*** will be set up within IOC with expert guidance as a ***Planning Group***. Tasks to be carried out are:

i) International programming of activities relative to harmful algal blooms.

ii) Establishment of an international network of scientists, managers and other relevant people.

iii) Initiation of training activities.

iv) Arrangement of relevant information to be distributed, e.g.

a) Newsletter ("Harmful Algal News")
b) Directory of Scientists
c) Advice on management problems.

To accomplish these tasks it is necessary to establish close contacts with other organizations working with harmful algal blooms such as the FAO and IOC regional bodies, other organizations within the UN system (UNDP, WHO etc.), EC, ASEAN, ICSU/SCOR and ICES. Within ICES there is a considerable expertise on harmful algal blooms, most of which is concentrated in SGDAB. However, that group has definite tasks to carry out in the short Study Group lifetime. The SGDAB participants agreed that it would be advantageous for both IOC and ICES if a closer cooperation on harmful algal blooms could be continued and expanded between the two organizations. This demands clearer organization of these questions within ICES. Until now, problems related to harmful algal blooms have been dealt with in several groups. Harmful algal blooms are, however, special, and in many parts of the world (including the ICES area) of immense importance, causing damage to ecosystems, to fisheries, aquaculture and human health.

It was suggested that, in view of the importance of harmful algal blooms to science and management, and to facilitate contacts with other organizations, there should be a special body within ICES to take care of harmful algal bloom affairs. Similar bodies for special purposes already exist, e.g., the Working Group on Introductions and Transfers of Marine

Organisms, so a special group on harmful algal blooms would not be unusual. In accordance with this suggestion it was decided to **recommend to ICES that the SG on Dynamics of Algal Blooms be transformed into a *Working Group on Harmful Algal Blooms.***

The meeting was reminded that IOC has, in fact, been cosponsoring the Study Group, and it was felt appropriate to recommend ICES to invite IOC to also cosponsor the new Working Group. This would very much facilitate future cooperation. The meeting suggested that the new Working Group on Harmful Algal Blooms should report to the Biological Oceanography Committee (and possibly also to the Hydrography Committee). The Working Group would be recommended to cooperate with several bodies (within ICES, e.g., any group dealing with plankton questions and with the Working Group on Shelf Seas Oceanography). The possible reporting to the Hydrography Committee and the suggested intimate cooperation with the Working Group on Shelf Seas Oceanography derives from the fact that hydrographical affairs are an integral part of harmful algal bloom work. The possibility of the new Working Group reporting to the Advisory Committee on the Marine Environment (ACME) was also mentioned.

The meeting also discussed other possible steps which could be taken by ICES to enhance activities related to harmful algal blooms. The extensive related activities in EC were discussed. The ties between ICES and EC should be strengthened (an initiative for cooperation between IOC and EC has already been taken); the links between ICSU/SCOR and ICES can likewise be strengthened.

3.3 A physicist's reaction on day one and two

A presentation by W. Fennel discussed the differences in approach between marine biologists and physical oceanographers. Marine biology is a complex field and adding physical aspects will even enhance this complexity. Marine biology depends on physical processes such as e.g., advection, diffusion, upwelling etc. The physical oceanography is practically independent of biological processes. Physical oceanographic processes are more readily quantifiable than those of marine biology. One advantage of physical oceanography is that the dynamical relations are governed by laws of conservation, e.g. energy, momentum mass, which can be expressed by differential equations.

Theory and modelling are required in physical oceanographic research to understand observation and to address new questions. In biology it is often necessary to describe rate processes. **For biologists and physicists to interact and to harmonize their efforts, it is necessary to introduce theory and modelling at an early stage in the programme to help determine what processes can be ignored and what needs to be studied or measured.** It is at the point of theory that physicists and biologists can meet and it is important to isolate the most crucial relationships which determine the processes to be considered.

4. DESIGN OF ALGAL POPULATION DYNAMICS STUDIES IN RELATION TO HYDRODYNAMICS AND THE DEVELOPMENT OF PILOT STUDIES

Agenda Item 4 was introduced by noting the need to know the boundaries of the population being studied and relevant inputs and outputs across these boundaries.

One of the objectives of this session was to present the physical/biological interactions aspects of the pilot studies to the hydrographers. More details about the **GULF OF MAINE**, **SKAGERRAK-KATTEGAT**, and **IBERIA** pilot projects are given in the report of the first meeting of the **ICES-IOC Study Group on the Dynamics of Harmful Algal Blooms in Coastal Waters** (C.M. 1992/L:4 Ref.C).

4.1 Presentations of Pilot Programmes designed by the SGDAB and questions forwarded to the Joint Session

4.1.1 The **GULF OF MAINE Pilot Study** is proposed as an investigation of the population dynamics of PSP producing *Alexandrium* in a buoyant coastal current. The coastal current is relevant to the distribution of toxicity in many areas. The system involves features such as river plumes, coastal fronts and wind driven up and downwelling. It is desired to model what goes on inside a defined portion of this coastal current. South winds force the coastal current offshore and cell movements complicate the growth rates measurements of this harmful species necessary for the population dynamics studies.

The observed cell concentrations within the coastal current may be affected by behaviour and physics as well as being the consequence of growth, and a means of measuring growth rates *in situ* will be required. There was a description of the ongoing programme and the progress already made, but the biology remains somewhat descriptive. An extant model of the distribution of salinity in the area was described and how particles could be introduced into the model to see how changes (e.g. in growth terms) affect the population within the moving box. The need for oceanographers in the contiguous areas of the Bay of Fundy and the Nova Scotian Shelf to provide input was identified. The occurrence of years when there is a flat thin freshwater layer were also discusssed. The large interannual variance in PSP phenomena and the ocurrence of exceptional years were also noted.

4.1.2 Many toxic species are involved in the **SKAGERRAK-KATTEGAT Pilot Study**. Three types of blooms are recognized; i) surface blooms associated with coastal currents, ii) subsurface blooms associated with the pycnocline at about 20 m depth, and 3) offshore blooms at about 40-50 m. Coastal current (surface) blooms are associated with low nutrients while those near the pycnocline may have high productivity due to cross pycnocline transport. It is also possible to find blooms near the pycnocline where there is no nutrient rich deep water and these have low productivity. The advection of *Gyrodinium aureolum* from the Skagerrak to Norwegian salmon farms was described. Subjects which require the input of hydrographers were listed:

- the need to describe processes leading to bloom accumulation from low cell concentrations next to a low nutrient pycnocline; i.e. how can physical accumulation be separated from biological growth processes?

- the significance of nutrient entrainment to the chlorophyll maximum layer at the pycnocline.

- how can one make entrainment measurements in real time so as to know where to make the right biological measurements?

- describe the microhydrodynamics of the pycnocline in relation to the needs of pycnocline populations.

- are pycnocline intrusions a mechanism for the production of subsurface blooms?

- we need to understand the surface and sub-surface origin of nutrients.

4.1.3 The **IBERIA Pilot Study** is being planned as an investigation on the population dynamics of DSP agents (*Dinophysis* spp) and PSP agents (*Gymnodinium catenatum*).

DSP, or both DSP and PSP, occur annually in the Iberian pilot study area. Both kinds of outbreak can occur:

- During the upwelling season (summer conditions, stratification), when moderate upwelling pulses and strong insolation allows the establishment of subsurface thermoclines or pycnoclines;

- at the relaxation of the upwelling (autumn conditions, mixed waters), when a warmer, nutrient-depleted shelf water is advected to the coast and to the rías.

Different mechanisms are believed to enhance growth of the same species in the two preceeding cases (predominance of either *in situ* growth rate or of physical mechanisms of concentration in coastal waters).

The toxic outbreaks can be :

- With bloom development, the high concentration of cells causing water discoloration;

- without bloom development, when the toxic cells represent a small percentage (1-5%) of the total phytoplankton population.

And in relation with the spatial distribution:

- restricted to inner areas of the rías and lagoons;

- apparently originated in the shelf area and physically transported inside the rías and lagoons.

The pilot study will concentrate on those blooms apparently resulting from exchange between the open coastal waters and the inner waters of the rías and lagoons. These outbreaks affect in a similar way the Spanish Rías Bajas and the Portuguese coastal lagoons and must be the result of a mesoscale process.

Outbreaks caused by low numbers of the causative agent, especially DSP outbreaks, are the most frequent. The importance of upwelling was emphazised as the main driving force behind the changes in phytoplankton succesion and hydrographic features.

- It is necessary to measure the growth rates of the populations established mainly in the pycnocline during summer condidions. What are the reasons for the maxima to

congregate there? Nutritional strategies, or choosing a layer where the losses due to offshore flushing rates are minimal?

- To contribution of *in situ* growth and advectives processes needs to be quantified, especially during the sudden autumn outbreaks.

- It is known that at the relaxation of upwelling and inshore advection of shelf water, a population of large dinoflagellates, associated with PSP or DSP or both, is established. What are the physical/chemical conditions of the water which determine the dominance of one species or the other?

4.2 Presentation of other typical cases of harmful algal blooms

Some Baltic studies illustrated phytoplankton population dynamics in relation to physical forcing. These included the joint, multinational Baltic Sea Patchiness Experiment, PEX 86', which concentrated on the dynamics of the vernal phytoplankton bloom. Other examples described the hydrodynamical control by frontal processes of toxic cyanobacterial blooms in the Gulf of Finland.

Physical processes in the lower estuary of the St. Lawrence River and the northern Gulf of St. Lawrence were described, with emphasis on the effect of runoff on the development of fronts and the distribution of the plume and its entrained cells. The objective of further study of this system will be to determine the relative importance of physical factors versus *in situ* growth in determining the magnitude and distribution of PSP blooms in the area.

4.3 A systematic discussion of the three pilot projects

Physical mechanisms relative to the accumulation of dinoflagellates at convergences and shear fronts and how to distinguish these were discussed by R. Geyer, as well as the use of acoustic Doppler techniques to measure current velocities and help validate numerical models of the types of convergence at fronts. It was pointed out that numerical models are validated by intense sampling, and that tidally dominated systems require intense effort for understanding their dynamics. Other approaches to measuring motion at fronts were also considered, such as drifters and dyes. Drifters require the use of global positioning systems for accuracy, and dyes cannot be used with the required frequency. By coupling numerical models and these other approaches it is possible to use models to reasonably fill gaps in the empirical picture. Other techniques mentioned included ocean surface current radar, HF radar and normal current meters.

The meeting then divided into groups to finalize the Pilot Studies discussions, each group to put together an administrative plan and an operating plan, bearing in mind the need to integrate physics into the biological plans. Each Pilot Study must be designed to produce general results and they must show how to do this. The plans should explicity show where physical/biological cooperation is required and what is of interest to physicists, i.e., what is in the plan besides service hydrography?

4.3.1 Notes from the submeeting on Physicists and Biologists about the SKAGERRAK-KATTEGAT Pilot Study

The dinoflagellate *Gyrodinium aureolum* first emerges during summer in the western part of the Skagerrak, and generally as a subsurface population in low numbers. It is not clear if the North Sea is the seeding area or if the *G. aureolum* remains during winter in the Skagerrak area. In the next stage of a bloom development, *G. aureolum* will normally be found along the northwest coast of Denmark, still as a subsurface population. When large blooms are formed in the Skagerrak area, the largest concentrations are found in the coastal current area or close to it. The big blooms of this species may appear as both surface and as subsurface blooms. During years when *G. aureolum* blooms in the Skagerrak area, the southwest coast of Norway is the most likely area to find the largest concentrations and also harmful effects like fish-kills.

A number of biological/physical interactions seem to be involved in bloom forming processes of *G. aureolum*:

- The large scale circulation bringing *G. aureolum* from the North Sea or western part of Skagerrak, to the central part and coastal current zone.

- The bloom formation process which includes the biological growth of the algae and the increase of the density of the population through concentration processes of a scattered population. The latter process is probably of great importance when the off-shore surface water containing the algae is advected towards the coast and flows below the coastal current due to the higher density of the downwelling off-shore water.

- The latter process involves a number of biological and physical interactions like the speed of vertical water movements controlled by Ekman transport and Ekman driven up- and down-welling in relation to the swimming speed of the algae, the swimming to compensate for sinking, diurnal migration, self shading effects, etc.

In the Skagerrak-Kattegat area the occurrence of stratification is a well known feature. Subsurface chlorophyll maxima also occur regularly in the area. Thus, the second topic discussed was the biological and physical micro-dynamic processes and their possible interactions in, or close to, the pycnocline and the significance of chlorophyll maxima for the formation of bloom events. The following points were made:

- Entrainment of deep-water across the pycnocline is an important process in bringing nutrients to the surface waters. The entrainment process is associated with vertical water movements in the water column and turbulence in the mixed layer close to the pycnocline. However, the turbulence level in the upper layer is usually much higher owing to wave stirring and thermal convection. Thus, phytoplankton cells who want to stay in a pycnocline population have to counteract the dispersion caused by the advection-diffusion processes. The physical-biological interactions which relate to the entrainment processes are not clear.

- Vertical movements of the pycnocline caused by changes in the circulation pattern due to wind stress, internal waves, etc., will move the subsurface chlorophyll maxima as well.

The biological response to such vertical displacements could be several, like concentrating a population, separating species, bringing a population to an unfavourable depth level, etc.

- Further, mass balance studies and nutrient budgets of subsurface chlorophyll maxima, taking the physical-biological interactions into special consideration, are needed if the bloom forming processes are to be understood in the Kattegat-Skagerrak area.

The subgroup concluded that there was a considerable amount of data and knowledge available on the two topics which, as a first step, could be worked up by the physicist and the biologist together. **Thus, the subgroup suggested that a four days workshop on the physical-biological interactions of the *G. aureolum* blooms and the subsurface chlorophyll maxima in the Kattegat-Skagerrak area would be the next step. The results of the workshop could then be used to design what experiments should be carried out in the proposed pilot project.**

4.3.2 Notes from the submeeting about the IBERIA Pilot Study

A change in wind regime at the end of the upwelling season always results in an increase of temperature in the whole water column and an increase in the populations of large dinoflagellates. It is desirable to be able to predict what kind of toxicity (PSP, DSP) is going to be, and the levels that will result.

When upwelling relaxes, where does the warmer water comes from? From the poleward surface slope current? Warmer offshore water not affected by upwelling? Is there any significant influence of river plumes?

There is evidence of long-shelf spreading of PSP toxicity during autumn blooms of *Gymnodinium catenatum*. Is this a transport of cells, or a transport of conditions (higher temperature, different "f" ratio) that enhance cyst germination/cell growth ?

During summer outbreaks, some differences between the open Portuguese coast and the Spanish rías need to be made more explicit. On the Portuguese coast, there is evidence of a lower salinity lens limited by the shelf break. Its cross-shelf position is likely to be modified by wind forcing. It is thus necessary to establish an adequate meteorological data base to enable estimation of Ekman transports. On the other hand, it is essential to carry out direct current measurements in the wind-driven surface layer.

The subgroup noted that there are divergences in the interpretation of Spanish and Portuguese observations that need to be reconciled after a deeper analysis of the respective data, and regional meetings and workshops are planned in 1993, and 1994.

4.3.3 Notes from the submeeting about the GULF OF MAINE pilot study

Nothing further was added to previous statements on the Gulf of Maine Pilot Study.

5. A BIOLOGIST'S REACTION ON DAY THREE AND FOUR

General problems of harmful algal blooms were next addressed by F.J.R. Taylor. Theories of blooms are well developed and models have been produced but do not work very well at present for toxic blooms. The problem of prediction of toxic blooms is like the problem of predicting weather and climate. There is the problem of predicting blooms in the short term of days and weeks and on longer time scales of between years and greater. Long term changes in major nutrients can be linked to climate change and we wish to know what effects this will have on the species composition of the phytoplankton population and on the frequency and intensity of toxic blooms.

There are at least three fundamental types of toxic algae:

i) benthic types both macro and microalgae (e.g., those responsible for Ciguatera toxins);

ii) types from the mixed pelagic environment (e.g., diatoms);

iii) those from the stratified pelagic environment (flagellates).

Buoyant forms such as *Trichodesmium* and *Noctiluca* also are important in certain regions. In fresh water, neurotoxin and hepatotoxin producing cyanobacteria are important; certain areas of China have high incidences of liver cancer as a result of mycrocystin producing organisms.

One can also recognize at least four fundamental ecosystem types.

i) The pilot studies are all similar in that they are associated with the **open coastal type of ecosystem.**

ii) There is also the **semi-enclosed type of ecosystem** which may be eutrophic; in this type it is also possible to distinguish deep (fjords) and shallow types.

iii) Another type is **major estuarine fronts.**

iv) The fourth is the **open ocean** where types such as *Gymnodinium breve*, *G. catenatum*, *Gyrodinium aureolum* and *Trichodesmium* (now thought to be a neurotoxin producer) occur.

Within systems one can distinguish i) **low** and ii) **high nutrient types**, with high nutrients occurring either **naturally** (e.g., by upwelling) or **anthropogenically** (by eutrophication). Blooms have a characteristic **seasonality**, often occurring within short "windows" of time and often involving cysts or spores. Toxic blooms are ultimately **site specific**. Sometimes indices can be used for their prediction, e.g., temperature can be used to predict *Chattonella* blooms in the Sea of Japan. Some of the complications in dealing with toxic blooms are the occurrences of both toxic and non-toxic forms in the same species, behavioural complications such as swimming, allelopathy ('chemical warfare') and its effect on grazing and the food web and selective grazing, say, of non-toxic forms.

In the Strait of Georgia (British Columbia, Canada), that is a good example of a large, semienclosed, deep estuarine ecosystem (case ii), regularly produced blooms of *Heterosigma* cause serious damage to the salmon cage culture industry. The blooms do not appear until the temperature reaches 15°C but the salinity must also decline to 15 ppt. Duration of the bloom depends on strong stratification (this lasted 4 months in 1989) and termination of the bloom depends on stratification breakdown. These events are preceded by a typical spring bloom followed by an interval and then the big freshwater runoff from the Fraser River following which stratification occurs and *Heterosigma* appears. This organism is allelopathic, it blooms during a time of low surface major nutrients and migrates vertically for nutrients and light if the stratification is less than 10 m (it is usually 5-8 m). Fall wind events lead to the breakdown of this system. The *Heterosigma* populations are horizontally advected; the growing population remains about the same at the mouth of the Fraser River while large quantities are exported down the Strait of Georgia where they encounter fish farms. Years of low snowfall like the winter of 1991-92, result in high salinity and low micronutrient levels (as in 1992) and result in no bloom. It was recommended that these other types of blooms and ecosystems be included in the ICES programme.

Long-term records for the occurrence of *Heterosigma* in the area are qualitative. In Japan, the situation is similar but there are also anoxic bottom waters. The *Heterosigma* situation in Naragansett Bay (Rhode Island, USA) is different again. **An intercomparison study of *Heterosigma* blooms in the three localities (Strait of Georgia, Sea of Japan, Narragansett Bay) was proposed.**

6. DISCUSSION OF PROPOSALS FROM SUBGROUPS AND OF THE RECOMMENDATIONS ON THE FUTURE NAME AND TERMS OF REFERENCE OF THE STUDY GROUP

Discussion then began on the future of the SGDAB. It was emphasized that the proposal for a new Working Group should point out to ICES the need to continue and enhance work on harmful algal blooms. This might even result in an upgrade to an special working group. The question of which committee to report to was again mentioned and it was suggested that we look for more organizations to cooperate with, particularly the EC which has great resources, and SCOR.

It was emphasized that the term *blooms* should be retained for political and historical reasons since by long usage its meaning is readily understood by non-specialists. The creation and maintenance of a data base was discussed, but so far there has been no attempt to identify resources for such an undertaking. Some participants felt that the present Study Group (SGDAB) should accept all harmful algal bloom responsibilities, including the country reports, while the other **Working Group on Phytoplankton and the Management of their Effects** should deal with phytoplankton productivity questions. A suggestion to enlarge the harmful algal bloom database to include a worldwide data base might gradually occur with IOC participation. The name of the new upgraded group should include the word **'dynamics'** which encompasses a multidisciplinary (hydrography and ecology) aspect which the participants considered to be very important in defining the uniqueness of the proposed Working Group.

Some participants suggested that the proposed terms of reference should give more emphasis to the collaboration with physical oceanography. Much of this could be done in the terms of reference to actual meetings, or the problem could be solved by the cosponsorship of the Hydrography Committee. Plans for the next meeting were discussed in the evening following this meeting and during the following meeting of the WGSSO. The idea of combining the proposed methods workshop with the next meeting was considered. It was proposed and accepted that B. Reguera should continue as chairperson for at least one more year, and suggested that joint planning groups should be appointed to propose the agenda for the next meeting.

The ad hoc group that formulated the proposed terms of reference also proposed some titles. The group favoured the title: **WORKING GROUP ON HARMFUL ALGAL BLOOM DYNAMICS.**

The ad hoc subgroup which proposed the terms of reference and name for the new Working Group presented these proposals in the form of a document which also included a section headed 'What is the ultimate objective of the proposed Working Group? Anticipated outcomes'. This was followed by the statement of its objective:

'Develop the ability to explain and eventually predict harmful algal bloom occurrences based on a fundamental understanding of the underlying mechanisms and forcing functions, including hydrology and organism behaviour and ecophysiology'.

7. RECOMMENDATIONS

The Study Group on the Dynamics of Algal Blooms and the Working Group on Shelf Seas Oceanography (Joint Session) recommends that:

The ICES-IOC Study Group on the Dynamics of Algal Blooms should be re-established as the **Working Group on Harmful Algal Bloom Dynamics** (Chairman: Ms B. Reguera, Spain) and will meet in Vigo, Spain from 11-14 May 1994 to:

a) continue the development of an understanding of the dynamics of harmful algal blooms, including experimental aspects of HAB Dynamics;

b) review progress in the implementation and/or execution of physical-biological interaction investigations in the pilot study areas (Gulf of Maine, Skagerrak-Kattegat, Iberia);

c) review the workshop on Modelling the Population Dynamics of Harmful Algal Blooms, and propose further steps to improve the dialogue between physicists and biologists;

d) finalize planning of the Workshop on "Intercomparison of *in situ* Growth Rate Measurements";

e) consider the integration of ongoing research activities on harmful algae phenomena in the ICES area into the existing global international programme on harmful algal blooms (IOC-FAO/OSLR/HAB);

f) evaluate strategies useful in investigating HABs, and in mitigating their detrimental effects on marine ecosystems, e.g., the efficacy of regional HAB monitoring systems;

g) consider the development of a HAB database;

h) collate and discuss national reports on harmful algal blooms (HABs);

A Joint session with the Working Group on Shelf Seas Oceanography will be held on 11-12 May 1994 to consider agenda items a, b and c.

IOC should be invited to co-sponsor this Working Group.

Justification:

Because harmful algal blooms present problems quite distinct from those of normal blooms due to their potentially serious economic and social impacts, they require special attention. It is an important international problem, and ICES needs to play a major role because of its importance to member countries and ICES' unique interdisciplinary capabilities. IOC-FAO are developing an international programme that features ICES participation as a major regional player for the North Atlantic area, as well as involvement in a worldwide educational programme.

The problem requires focused effort on harmful algal bloom dynamics (including the effect of the population dynamics of harmful algal bloom events, and the rôle of physical-biological interactions), and should not be submerged into a general study of plankton ecology. The focused effort will make it easier to attract the required hydrographers and chemists to the problem. To facilitate the growth of this important multi-disciplinary aspect to the problem, joint sessions with the Working Group on Shelf Seas Oceanography are proposed.

The Study Group on the Dynamics of Algal Blooms recommends that:

A Workshop on "Intercomparison on *in situ* Growth Rate Measurements" should be held at Ría de Aveiro (Portugal) from 25-29 July 1994 under the chairmanship of Ms M.A. Sampayo (Portugal) to undertake an intercomparison study of *in situ* growth rates of dinoflagellates in support of the study of harmful algal blooms.

Justification:

Good estimates of population dynamics parameters, 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. The chosen site has a model available, is physically

uncomplicated which will facilitate intercomparison of techniques, and there is a certainty of finding relevant target species.

The Working Group on Shelf Seas Oceanography recommends that:

A Workshop on "Modelling the Population Dynamics of Harmful Algal Bloom (HABs)" should be held at Vigo (Spain) from 4-7 May 1994 under the co-chairmenship of Dr P. Tett (UK) and Dr W. Fennel (Germany) to:

a) investigate the use of numerical models in improving understanding of the dynamics of HABs;

b) use the above models to assist in the design of sampling strategies, interpretation, and the forecasting of HABs;

c) develop a dialogue between physical and biological oceanographers with respect to HABs, including the role of physical inputs and temporal and spatial scales.

Justification:

Many biologists working on HAB problems are insufficiently aware of the role which modelling can play in advancing their work and achieving results, (in the design of sampling strategies, interpretation of data, and forecasting. It was suggested that this situation could be improved by creating a dialogue between students of harmful algal, and existing groups of biological and physical oceanographers. This workshop will provide a suitable occasion for such a dialogue, with emphasis on population dynamics including the importance of physical inputs, and temporal and spatial scales.

ANNEX I

SECOND MEETING OF THE ICES STUDY GROUP ON THE DYNAMICS OF ALGAL BLOOMS IN COASTAL OCEANS

8-11 February 1993

Southeast Fisheries Science Center. Charleston, S C, U.S.A.

1. **Registration.**

2. **Welcome to the participants.**

3. **Opening of the meeting.**
   - 3.1 The title of the group: Algal Blooms/Harmful Algal Blooms.
   - 3.2 Terms of reference.

4. **Introduction of participants.**

5. **Approval of the agenda**
   - 5.1 Designation of a rapporteur

6. **The utility of mesocosm experiments for the understanding of algal population dynamics.**
   - 6.1 Brief introductions about their experience on the subject (10-15 mn each) by Prof T. Smayda, Dr E. Granéli, and Dr F. Colijn.
   - 6.2 Recent results found in the literature
   - 6.3 What mesocosm experiments should be carried out and how
   - 6.4 Discussions and conclusions.

7. **Examine the cyst phase in the life histories of relevant, potentially harmful algae**
   - 7.1 Review of recent relevant work
   - 7.2 What experiments to carry out and how
   - 7.3 Discussions and conclusions

8. **Pilot Programmes outline**
   - 8.1 Pilot programme proposals discussions in subgroups.
   - 8.2 List of questions/problems to be forwarded to the members of the Shelf Seas Oceanography WG during the joint session on days 10-11 February.
   - 8.3 Designation of one person from each pilot programme to present their regional case to the hydrographers

9. **Discussions on innovative devices already applied in other international programmes (i.e. the Adriatic programme) that could used in the ICES programme.**
   - 9.1 Presentation by P. Gentien of the "*in situ* grain size analyzer"
   - 9.2 Other suggestions

10. **Propose agenda for joint discussions between participants in the SGDAB and the SSOWG.**

ANNEX II

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ANNEX III

ENCLOSED EXPERIMENTAL MARINE ECOSYSTEMS

There is growing worldwide experience and expertise with mesocosms/microcosms. Results of mesocosm/microcosm experiments have been published in a few summary books (Grice & Reeve 1982, Lalli 1990, Anonymous 1991).

Mesocosms/microcosms are the two basic enclosure experimental ecosystems. Which type is used and its "physical" design characteristics depend on the objectives of the research. Mesocosms may be transportable for *in situ* deployment or fixed enclosures on land, with flow-through seawater capability and of variable volume. Mesocosms are defined as enclosures of at least 1 m^3, while microcosms are usually < 1 m^3. Both enclosure types can be operated in batch, semi-continuous or continuous culture mode, and the experiments replicated.

Depending on the questions being asked and the region, including depth, being evaluated, mesocosms/microcosms should be run with or without bottom sediments, with or without grazers, in batch, continuous or semicontinuous culture mode, with natural phytoplankton communities or inoculated species, and with or without stratification and physical fine structure. The duration of the experiment should not exceed a few weeks.

Historically, experimentally manipulated mesocosms/microcosms have used total phytoplankton community biomass as the response parameter, and have focused on the effects of nutrient additions on population growth. To date, there has been very little use of mesocosms/microcosms in studies on harmful algal blooms. For such research to be relevant to harmful algal bloom events, however, appropriate physical and biological scaling must be achieved beyond that normally used in mesocosm/microcosm experimentation. For example, an evaluation of the role of physical conditions such as turbulence/stratification and physical habitat fine structure is required, as well as measurements of species-specific responses, rather than the less useful bulk measurements of response, such as biomass. Since species-specific responses are coupled to physical habitat conditions, with the role of the latter significantly more difficult to establish, there is need for improvement of mesocosm methodology for use in harmful algal bloom research. There is great need for appropriate scaling of biological processes to the physical habitat, and difficulty in achieving this experimentally. It is therefore particularly important to establish the relationships between mesocosm size and the range of physical scaling factors (i.e., turbulence, pycnocline structure, small-scale motion, etc.) achievable in these systems in order to establish whether the proposed experimentation, and which types, are ecologically relevant. Such efforts are recommended. A drawback might be that some species will be difficult to keep in such systems.

Mesocosms/microcosms are best suited to derive rate constants rather than to simulate the complex ecological conditions under which harmful algal blooms develop and are regulated. Mesocosms/microcosms can be used to study harmful algal population dynamics, including certain rate processes, in response to various factors such as nutrient loads, nutrient ratios, grazers and pathogens in the absence of interference from complex physical conditions such as diffusion, advection, entrainment, etc. which occur under natural conditions. Such

results would help to provide physiological constants needed in the development of mathematical models of harmful blooms. These experiments would also put into better perspective the collective role of physical conditions in regulating bloom dynamics relative to biotic processes. Mesocosm/microcosm experiments can also provide insights into the ranges in the rates of various processes. Such information would be helpful in the design and in carrying out field studies, *in situ* measurements, and in the design of ecological experiments.

The mobility of some of these systems makes them easy to use in different situations. The cost of installation of a full system may range from 100 US $ for a simple system to 100,000 US $ for a very sophisticated system.

Some examples of contemporary problems of harmful algal ecophysiological dynamics which would appear to be appropriate for study in mesocosms/microcosms are:

1. What is the role of nutrient concentrations on specific growth rates of species of interest, given that:

   a. Macro or micro nutrients may differentially affect growth of particular species.

   b. Nutrient ratios may differentially affect species.

   c. Organic nutrients, including exudates from cultured shellfish, may differentially affect the growth of particular species.

2. What are the growth properties and characteristics of the bloom initiation, exponential growth and decline phases under simulated conditions, and how do they compare with *in situ* behavior?

3. What is the role of grazers in the selection of harmful species, including their specific grazing affects, inhibition of grazing on species of interest, and in determining food web structure?

4. What is the role of species behavior, such as vertical migration, reaction to turbulence, and discontinuous growth such as cyst formation in determining population growth dynamics?

As with all experimental approaches, mesocosm/microcosm experimentation has both limitations and advantages. Such experimentation is therefore not recommended as a general panacea for resolution of harmful algal bloom issues requiring experimental approaches. Rather, judicious selection of which scientific issues can be attacked using mesocosms/microcosms and incorporating suitable biological and physical scaling, and yielding results applicable to *in situ* processes, is required. The utility of this approach may be applicable to certain population dynamics' issues, but not to others; suitable for experimentation on some species, but not others; appropriate to certain regional bloom event issues, but not to others, etc. What is needed is continuous, ongoing methodological improvement to increase the range of tractable problems amenable to mesocosm/microcosm experimentation. Investigators are therefore encouraged to resolve

the utility of mesocosm/microcosm approaches to harmful algal bloom issues requiring experimental manipulation, including those listed above.

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ANNEX IV

THE IMPORTANCE OF RESTING CYSTS IN THE BLOOM DYNAMICS OF HARMFUL ALGAE

Background

Many phytoplankton species, including some harmful ones, have dormant stages in their life histories. The alternation between dormant and benthic stages and a motile, vegetative existence is a complex process that must be considered to understand and model harmful bloom dynamics. Cyst germination provides an inoculum at the initiation phase of many blooms, and cyst formation subsequently removes substantial numbers of cells in later stages. However, our knowledge of resting stage physiology and ecology is restricted to a few species, and even for those, is limited in scope. The Study Group reviewed present knowledge, and identified a number of areas where additional research is needed. Methods needed to address these issues were discussed.

Present Understanding

Resting stages have been documented for harmful algal species belonging to several different phytoplankton classes, though for one important group, the diatoms, none are yet known for harmful forms. The following harmful species in the ICES region are known to produce a resting stage: *Alexandrium tamarense, A. catenella, A. fundyense, A. lusitanicum, A. minutum, A. monilatum, Gymnodinium catenatum, Prorocentrum lima, Heterosigma akashiwo,* and *Nodularia spumigena*. Since all but two of these are dinoflagellates, the focus here is on their cysts or hypnozygotes. Temporary, asexual cysts, short-lived resting cells that result when the theca is shed in response to adverse conditions, are not considered since they are probably not a major factor in bloom dynamics. Hypnozygotes or cysts are highly-resistant, dormant cells produced as a result of sexuality (Pfiester and Anderson, 1987). Their formation (encystment) and germination (excystment) dynamics are thought to be important in many harmful blooms. All subsequent discussion of cysts in this text refers to hypnozygotes .

Most toxic or harmful dinoflagellate species reproduce asexually, but under certain conditions sexuality occurs (reviewed in Pfiester and Anderson, 1987). In laboratory cultures, nutrient depletion induces sexuality (e.g. Pfiester, 1975; Turpin et al, 1978; Walker and Steidinger, 1979; Anderson et al, 1984). Nutrient limitation has also been implicated in sexual induction in natural populations, but the environmental cues that stimulate encystment in natural populations are not well-defined, and recent studies indicate that factors other than macronutrient availability (e.g. iron stress) may be involved (Doucette and Harrison, 1989). Furthermore, given the discovery of endogenous control of cyst germination for *A. tamarense* (discussed below), the possibility of endogenous or "clock"-regulated sexuality must be considered.

Dinoflagellate hypnozygotes undergo a variable period of dormancy before they excyst. The length of this mandatory interval varies considerably among species (12 hrs. to 6 months; Pfiester, 1977; Anderson, 1980), and for a single species, can vary with the storage conditions. Thus cysts of *A. tamarense* stored at 4 °C mature in 6 months, whereas storage

at warmer temperatures shortens the mandatory interval to 3 months or less. In *Gymnodinium catenatum*, the mandatory interval is less than 2 weeks (Blackburn et al., 1989) and is not affected by the temperature of storage (Bravo and Anderson, submitted). Cysts of *Gonyaulax polyedra* lose the germination capacity after one year of storage in oxygenated conditions, but germination proceeds after one year storage in anoxic conditions (Blanco, 1990). The duration of the mandatory interval can have a significant effect on the timing of recurrent blooms, as species with a long maturation requirement may only seed one or two blooms in a year, whereas those that can germinate in less time may cycle repeatedly between the plankton and the benthos and contribute to multiple blooms in a single season (Binder and Anderson, 1987).

After the mandatory dormancy interval is complete, cysts can remain in a resting state (quiescence) due to non-optimal environmental conditions. The primary stimulus for excystment of temperate species is generally accepted to be a shift in temperature to favorable levels, as occurs in seasonal warming or cooling (Huber and Nipkow, 1922, 1923; Anderson and Wall, 1978; Anderson and Morel, 1979). Spontaneous germination of cysts without a change in temperature has also been noted (von Stosch, 1973; Pfiester, 1975 and 1977; Binder and Anderson, 1987). In addition, large numbers of cysts can also remain in the sediments even though ambient temperatures are suitable for excystment and cell division (Anderson et al., 1983; Blackburn et al., 1989). This is attributed to the burial of many cysts below the oxygenated surface layer of sediments and an absolute requirement for oxygen during germination (all species tested thus far; Anderson et al., 1987; Blanco 1990). Some species must be exposed to light for either brief (Binder and Anderson, 1986) or prolonged intervals (Anderson et al., 1987) before excystment is possible. Cysts that are buried deep in the sediment can thus remain quiescent for years, their fate either being eventual death if anoxia persists, or germination should they be transported to the sediment surface or overlying water. Cyst longevity, which probably depends most critically on temperature and oxygen, varies considerably between species, but can be 5-10 years in natural marine sediments (Keafer et al., 1992). Freshwater species are known to have survived for 17 years in lake sediments (Huber and Nipkow, 1922, 1923).

Internal or endogenous control of cyst germination has recently been demonstrated in *A. tamarense* (Anderson and Keafer, 1987) and in *Scrippsiella trochoidea* (Costas et al., 1990). Mature, quiescent cysts did not germinate in a consistent manner when exposed to optimal growth conditions throughout the year, but instead showed a variable response depending on the season. An endogenous annual clock was implicated, which might explain the germination of cysts deposited in deep waters where seasonal environmental cues such as temperature or daylength are small or nonexistent.

The geographic distributions of the cysts of several species have been mapped using both qualitative and quantitative approaches. For example, the simple presence or absence of *A. tamarense* cysts was determined along the coast of southern New England, highlighting regions with the potential for PSP, including several that had no prior history of the problem (Anderson et al., 1982). More comprehensive, quantitative surveys have been conducted for the same species in the Gulf of Maine (Lewis et al, 1979; Thayer et al., 1983; Anderson and Keafer, 1985), the Bay of Fundy (White and Lewis, 1982), and the lower St. Lawrence estuary (Cembella et al., 1988; Turgeon et al., 1990). The geographic distribution of *Alexandrium excavatum* on the Patagonian shelf has been studied by Carreto and Orozco (1989) who suggested that the high concentration of cysts in the surface sediment represents

a great potential danger of toxic episodes. Cysts of *A. minutum* have been mapped as well, revealing a pattern whereby seedbeds are transported into the upper reaches of estuaries by bottom currents during one season, and flushed back to their origins by strong runoff at other times (Erard le Denn, unpublished). The distribution and abundance of cysts have been shown to correlate with bathymetric features (e.g. basins), with the fine clay and silt sediment fractions, and with the transport pathways of major current and frontal systems (Dale, 1976; Harland et al., 1980; Tyler et al., 1982; Anderson and Keafer, 1985; Cembella et al., 1988).

Finally, cysts of toxic species have been shown to be toxic, though the level of this toxicity is somewhat unclear. Dale et al.(1978) suggest that cysts can be 10 times as toxic as vegetative cells, a conclusion supported by Oshima et al., (1992). Both of these studies relied on comparisons between cysts and motile cells from natural populations that might well represent different genetic stocks, and no study has yet examined this issue in the laboratory using cysts and motile cells of the same clone.

Future Work

Many studies relating to resting stages have been made, but only some are relevant to the Study Group's focus on the dynamics of blooms. Following are listed some areas where future efforts could be directed.

Life History Studies. For several harmful species it is not known whether resting stages exist, e.g.: *A. ostenfeldii, Gyrodinium aureolum, Chrysochromulina polylepis, Prymnesium parvum,* and *Phaeocystis pouchetii.* No species of *Dinophysis* has a cyst stage proven to date, but growing evidences of sexual reproduction and excystment processes within the genus are being found (Reguera et al., 1990; MacKenzie, 1992; Moita and Sampayo, 1993; McLachlan, 1993; Hansen, 1993; Reguera et al., submitted). The lack of a resting stage cannot be assumed simply because such cells have not been observed in laboratory cultures, as sexuality is often inhibited by the artificial laboratory environment. Furthermore, some important harmful species such as *Dinophysis* are not yet in culture. However, when a species is in culture, it is best to try to induce cyst formation by mixing different clones and manipulating nutrient levels so that a particular nutrient (N or P) becomes limiting (Walker and Steidinger, 1979; Anderson et al., 1984). Sometimes, careful attention must be paid to the "cleanliness" of the glassware and the medium, since contaminants introduced during autoclaving or from the glass itself can be inhibitory (Anderson et al., 1984). An alternative to culture manipulations is to concentrate and incubate natural populations, as was done for *Gymnodinium catenatum* (Bravo, 1986; Anderson et al., 1988). The procedure involves collection of a mixed plankton assemblage using a plankton net, and the resuspension of that material in filtered seawater from the study site, to which has been added a low level of nutrients and trace metals. The assemblage is incubated in the laboratory at the temperature of the ambient water at an appropriate daylength. Within a few days, cysts will be produced by many of the species in the sample. These are of a known age, and can readily be isolated by micropipette to study morphology or germination characteristics.

Induction of Sexuality. Our knowledge of the underlying mechanisms for sexual induction is inadequate and conflicting. Laboratory experiments implicate nutrient limitation as a triggering factor, but field studies have not provided conclusive proof that this type of external control is actually operative in blooms. The possibility of endogenous, "clock"

control of encystment must be evaluated. Quantitative studies of sexual induction require manipulations of laboratory cultures, preferably in continuous or semi-continuous cultures for those species that tolerate mixing, as batch cultures create a highly artificial nutrient environment that can obscure certain life cycle transformations (Anderson and Lindquist, 1985). Measurements of nutrient concentrations and the abundance of different life cycle stages during blooms can provide useful information as well, though it is difficult to account for the nutritional history of the population in sufficient detail to identify thresholds for induction. Internal nutrient pools may be more important in induction than the ambient nutrient concentration in the water surrounding a cell, yet pool sizes are extremely hard to measure for a single species in a mixed plankton assemblage. This is one of several instances where new methods are needed that permit the separation of the cells of a harmful species from co-occurring organisms and detritus for analysis.

Mandatory Dormancy. Several methods can be used to determine mandatory dormancy intervals for newly formed cysts or spores. If cysts can be formed in culture, they can be collected and stored under different temperature and light conditions, and periodically exposed to optimal growth conditions to assess germination success (Anderson, 1980; Blackburn et al., 1989). Cysts produced in an incubated plankton tow (described above) or collected in a sediment trap (deployed underneath a bloom but sufficiently far above the bottom to avoid resuspension of "old" cysts) also have a clearly-defined age, and can be isolated by micropipette for periodic germination trials. The least attractive option, but one which will work if a recent bloom has produced abundant new cysts, is to collect and work with surface sediments directly. Cysts can be isolated individually for germination trials, or an alternative procedure can be attempted that uses unprocessed sediment. This involves the preparation of numerous aliquots of a sediment sample, counting the cysts of interest in many of these aliquots to obtain a statistically sound estimate of initial concentrations, and then incubation of the subsamples. The difference between the initial counts and the number of cysts remaining after incubation provides a good measure of germination success (Anderson et al., 1987).

Control of Quiescence. Mature cysts can remain quiescent for months or years due to both exogenous and endogenous control. This information is not known for numerous cyst-forming harmful species, but may determine the timing and size of the excystment inoculum in the initiation phase of blooms. Determination of the temperature "window" for germination of a species or the light requirements of that species both require laboratory manipulations of cysts. As described above, these can be produced in laboratory cultures, or obtained from incubated plankton tows, sediment traps, or surface sediments. Laboratory methods for obtaining this information are given in Anderson and Morel (1979), Anderson et al. (1987), and Binder and Anderson (1987).

Geographic Distribution of Cysts. The distribution and abundance of cysts within a region have been investigated for only a few harmful species within the ICES region. Although cyst mapping can be extremely labor intensive and time-consuming, it does provide an important perspective on possible sites for bloom initiation and a general indication of the magnitude of the seedbeds. Quantitative surveys are not likely to be worth the large expenditure of effort on a repeated basis, but should be conducted at least once in a region under study. If resources are limited, a qualitative survey can still be used to demonstrate the presence or absence of the cysts of a target species. Samples for such a study are simple to collect using small corers, pumps, and even plankton nets dragged along the sediment surface. The utility

of this approach and the procedures followed are given in Anderson et al. (1982). Quantitative cyst mapping must account for the cells that are buried several cm below the sediment surface (Anderson et al., 1987). The fate of cysts buried below the oxygenated surface layer is not known, but their potential importance should be considered since resuspension or benthic animal activity can bring them into contact with favorable conditions for germination. Mapping studies have thus enumerated the total cysts in the top two, and sometimes top six, centimeters of sediment (Anderson and Keafer, 1985; Tyler et al., 1982; White and Lewis, 1982; Cembella et al., 1988; Turgeon et al., 1990). Methods for obtaining the vertical profile of cysts in a core sample are given in Anderson et al. (1987). This is one area where immunological probes or other techniques to "tag" cysts of interest are needed to expedite the tedious process of scanning sediment samples for cysts, which are often in low abundance compared to sand grains, detritus, and other organisms or particles in a sediment sample.

Encystment and Excystment Rates. Within the context of the dynamics of algal blooms, major uncertainties are the rates at which cysts germinate and at which motile cells form cysts. Equally important is the degree to which a population is synchronized during these transformations. It is not sufficient to know what fraction of a cyst population will germinate at a certain temperature; the distribution of germination events through time must be known as well. In other words, if a certain number of cysts germinate from a seedbed, it matters a great deal whether they all germinate at once, or whether their excystment is prolonged in time. These rate and synchrony measurements are difficult to obtain. Encystment rates in natural sediments can be estimated in some cases by examining the chlorophyll fluorescence of individual cysts, as some species fluoresce red under blue excitation because they are synthesizing chlorophyll immediately prior to germination (Anderson and Keafer, 1985). Alternatively, excystment rates can be obtained using laboratory cysts incubated at different temperatures, or through frequent quantitative cyst counts in cores to determine the change in cyst abundance in natural sediments. In this latter case, care must be taken to process numerous replicates, as small scale patchiness introduces a degree of variability in cyst abundance that can easily obscure a low level of germination (Anderson and Keafer, 1985; Blanco, 1989).

Encystment rates can be approximated by tabulating planozygotes as a percentage of the total motile population of a species in a bloom (Anderson et al., 1983; Coats et al., 1984). For some species such as *Alexandrium tamarense*, planozygotes are so different morphologically that they can be identified readily under the microscope. For other species, complex cytological techniques are needed to distinguish planozygotes from vegetative cells (Coats et al., 1984). One method which could be applied to obtain rate estimates from bloom populations would utilize sediment traps deployed just below the thermocline, or drifting with populations. Frequent collection of material from the traps, coupled with frequent cell counts of the bloom population could provide an estimate of the fraction of the population that forms cysts through time. A problem arises in efforts to determine encystment rates using laboratory cultures, as nutrients are exhausted so quickly in batch cultures that some cells begin the process of cyst formation, but do not complete it due to unfavorable conditions (Anderson and Lindquist, 1985). The low yield of cysts (10-20%) reported in such studies is deceptive, since a large percentage of the motile population can be planozygotes that presumably would have formed cysts had conditions been suitable.

Longevity. Two methods have been used to estimate the longevity of marine dinoflagellate cysts. One involves the storage of a sediment sample in the laboratory, with periodic cyst isolations and germination attempts. This is a long process, as most cysts will survive for years under these conditions. An alternative is to couple cyst profiling in sediment cores with isotope measurements, such as ^{210}Pb (Keafer et al., 1992). These data can be used in a simple model to determine the half-life of cysts in natural sediments.

Resuspension dynamics. Given the potential importance of cysts as an inoculum for blooms, the manner in which germination occurs should be known. If cysts simply germinate from the sediment surface, as would be expected in deeper waters (Anderson and Keafer, 1985), the problem in estimating inoculum size is much simpler than if resuspension by storms or currents moves cysts away from the sediment surface and into the water column. Intensive vertical profiling of the water column would be necessary to determine the extent to which cyst resuspension occurs in an area.

Cyst Toxicity. It is now well established that cysts can be toxic (Dale et al., 1978; Oshima et al., 1992), though the level of cyst toxicity relative to that in vegetative cells is uncertain. Studies are needed that determine the extent to which ingestion of cysts by scallops and other shellfish contributes to the PSP toxicity in an area, especially at times when vegetative cells are absent from the water column. The absolute toxicity of cysts can be obtained using laboratory cultures, though it is difficult to completely separate the cysts from vegetative cells even with density gradient procedures. Processed sediment samples can be analyzed for toxicity directly, though results are always more uncertain than laboratory cultures due to the heterogeneous nature of sediment samples. Documenting the link between cysts and shellfish toxicity is difficult, but useful information can be obtained by examining the guts of shellfish for intact or partially digested cysts. Dissected shellfish tissues can be processed with an ultrasonic probe and sieves using the same procedures employed for sediment samples.

Discussion.

For a number of harmful algal species, it is clear that cysts can be important in the dynamics of blooms (Anderson, 1984), yet unfortunately the list of areas where our knowledge is inadequate is long and challenging. Within the context of bloom dynamics, however, not all topics deserve equal priority, as some have more academic than practical importance. For example, high priority should be assigned to projects that attempt to demonstrate the existence of a resting stage in the life history of a particular species, and that strive to determine the physiological characteristics of that resting stage in the laboratory (mandatory dormancy interval, temperature "window" for germination, light requirements, etc.). Likewise, estimates of the rates of encystment and excystment and the exogenous or endogenous factors triggering sexuality are critical unknowns that must be addressed in our efforts to model bloom dynamics. Cyst mapping is of lower priority, however, and should be conducted on a one-time basis in a region, and infrequently thereafter, unless the cysts are thought to be a direct and significant source of toxicity. A low priority could also be assigned to studies on the longevity of buried cysts. In general, cyst studies should not dominate a field program within a region, but are necessary as a complement to studies of plankton bloom dynamics.

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ANNEX V

AGENDA

JOINT MEETING OF THE ICES STUDY GROUP ON THE DYNAMICS OF ALGAL BLOOMS AND THE WORKING GROUP ON SHELF SEAS OCEANOGRAPHY

10 - 11 February 1993

Southeast Fisheries Science Centre, Charleston, SC, U.S.A.

1. **Opening of the meeting**
   - 1.1 Presentation of participants
   - 1.2 Approval of agenda
   - 1.3 Appointment of a rapporteur

2. **Terms of reference**
   - 2.1. Terms of reference and what we are expected to achieve
   - 2.2. Short discussion on what we want to achieve

3. **Introduction**
   - 3.1. Current situation of the IOC/HAB programme
   - 3.2. Results of the first session of the IOC-FAO Intergovernmental Panel on Harmful Algal Blooms
   - 3.3. A physicist's reaction on day one and two

4. **Design of algal population dynamics studies in relation to hydrodynamics and the development of pilot studies**
   - 4.1 Presentation of pilot programmes designed by the SG on the Dynamics of Algal Blooms
   - 4.2. Presentation of questions/problems forwarded to the joint session by the study group
   - 4.3. Presentation of some typical cases of harmful algal blooms
   - 4.4. A systematic discussion of the three pilot projects
   - 4.5. Evaluate the possibilities to include modelling
   - 4.6. Outline a plan for achieving an increased understanding of the dynamics of harmful algal blooms

5. **A biologist's reaction on day three and four**

6. **Discuss proposal from subgroup and decide on the recommendation on the future name and terms of reference of the study group**

ANNEX VI

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