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Developments for a Canadian GEOHAB (Global Ecology and Oceanography of Harmful Algal Blooms) Program: 2001 Workshop Report

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ABSTRACT

Martin, J.L. (Editor). 2002. Developments for a Canadian GEOHAB (Global Ecology and Oceanography of Harmful Algal Blooms) Program: 2001 Workshop Report. Can. Tech. Rep. Fish. Aquat. Sci. 2400: vii + 44 p.

This report summarizes oral presentations, discussions and group recommendations arising from a two-day Workshop held in Montréal, Quebec on October 19-20, 2001. The aim of the workshop was to determine the level of interest and to bring researchers from throughout Canada together for the first time to discuss collaborative research and establish research goals for Harmful Algal Bloom (HAB) research. Participants from government, university and industry assembled to discuss developing a Canadian component of the international Global Ecology and Oceanography of Harmful Algal Blooms (GEOHAB) program. The mission of GEOHAB is to "foster international co-operative research on Harmful Algal Blooms (HABs) in ecosystem types sharing common features, comparing the key species involved and the oceanographic processes that influence their population dynamics". The scientific goal of GEOHAB is to "improve prediction of HABs by determining the ecological and oceanographic mechanisms underlying their population dynamics, integrating biological, chemical, and physical studies supported by enhanced observation and modelling systems".

Thirty participants attended the Workshop and represented the following: Fisheries and Oceans Canada, Health Canada, Canadian Food Inspection Agency, National Research Council, University of British Columbia, Université du Québec, Université Laval, McGill University, Dalhousie University, University of Western Ontario, St. Croix Estuary Project, and North Coast Water Quality Biotoxin Program. Participants each gave a brief presentation on their research interests and how they could contribute towards a Canadian program. A title for the Canadian GEOHAB program was proposed: "Population Dynamics of Canadian HABs (*Heterosigma, Alexandrium* and *Pseudo-nitzschia*)". Three themes for work were outlined: 1) population dynamics of key HABs in comparable ecosystems; 2) biologically mediated growth and loss processes; and 3) systems and data analyses towards improved prediction.

Plans were made to proceed with a national Canadian GEOHAB program, including submitting a letter of intent (LOI) to the Natural Sciences and Engineering Research Council of Canada (NSERC) International Opportunity Fund (IOF) for funding to organize a workshop in June, 2002 to develop research proposals for NSERC, and Fisheries and Oceans Canada's (DFO) Science Strategic Fund (SSF) and Environmental Science Strategic Research Fund (ESSRF).

RÉSUMÉ

Martin, J.L. (Editor). 2002. Developments for a Canadian GEOHAB (Global Ecology and Oceanography of Harmful Algal Blooms) Program: 2001 Workshop Report. Can. Tech. Rep. Fish. Aquat. Sci. 2400: vii + 44 p.

Ce rapport résume les présentations orales, les discussions et les recommandations de groupe qui dérivent d'un atelier qui a eu lieu le 19-20 octobre, 2001 à Montréal, Québec. Le but de cette rencontre était de regrouper pour la première fois des scientifiques du Canada afin de discuter de recherche collaborative et d'établir des buts scientifiques pour les floraisons d'algues nuisibles (FAN). Des participants du gouvernement, des universités et de l' industrie se sont rassemblés pour discuter de la possibilité d'initier un volet canadien au programme d'écologie et océanographie globale des floraisons d'algues nuisibles (ÉOGFAN). La mission d'ÉOGFAN est "d'encourager la recherche coopérative internationale des floraisons d'algues nuisibles dans les types d'écosystèmes ayant des caractéristiques semblables, en comparant les espèces clés impliquées et les processus océanographiques qui influencent la dynamique des populations". Le but scientifique d'ÉOGFAN est "d'améliorer la prédiction des FAN en déterminant les mécanismes écologiques et océanographiques qui sont à la base de leur dynamique des populations, unifiant les études biologiques, chimiques et physiques qui sont soutenues par les observations intensives et par les systèmes de modélisation".

Les trente participants à cet atelier représentaient les groupes suivants: Pêches et Océans Canada, Santé Canada, l'Agence Canadienne d'Inspection des Aliments, l'Université de British Columbia, l'Université du Québec, l'Université Laval, l'Université McGill, l'Université Dalhousie, l'Université de Western Ontario, le Conseil national de recherche, le projet de l'estuaire Ste-Croix et le programme de biotoxine de la qualité des eaux de la côte nord. Tous les participants ont donné une courte présentation de leur recherche ainsi que leur contribution potentielle au programme canadien proposé. Le titre soumis pour le programme canadien d'ÉOGFAN est le suivant: "Dynamique des populations des FAN canadiens (*Heterosigma, Alexandrium* et *Pseudo-nitzschia*)". Les grandes lignes des trois thèmes choisis sont: 1) dynamique des populations des espèces clés des FAN dans des écosystèmes comparables; 2) liens biologiques entre les processus de croissance et de perte; et 3) observation originale et analyses de données en vue d'une prédiction améliorée.

La décision a été prise de procéder avec le programme canadien d'ÉOGFAN en soumettant une lettre d'intérêt aux fonds d'initiative internationale (FII) du conseil de recherche en sciences naturelles et en génie du Canada (CRSNG) afin de couvrir les frais d'un atelier en juin 2002 qui aura pour but de développer des propositions scientifiques pour le CRSNG ainsi que pour les fonds stratégiques en sciences (FSS) du ministère des Pêches et des Océans (MPO) et des fonds de recherche stratégique en sciences environnementales (FRSSE).

Workshop on a Canadian Program on Global Ecology and Oceanography of Harmful Algal Blooms (GEOHAB)

INTRODUCTION

Several international organizations and agencies have established programs or working groups focused on specific aspects of harmful algal blooms (HABs) and their deleterious impacts on ecosystems, fisheries, recreational resources, and human health. Nevertheless, until recently there has been no international science agenda on research directed towards studying the biological, chemical, ecological and physical factors that regulate HAB dynamics and a prominent gap has existed in coordinating international efforts. The Scientific Committee on Oceanic Research (SCOR) and the Intergovernmental Oceanographic Commission (IOC) initiated a coordinated international science effort with an international workshop sponsored by the two organizations in Havreholm, Denmark on October 13-17, 1998. Twenty countries were represented and John Cullen (Dalhousie University, Canada) chaired the workshop. The report can be found on the SCOR website at http://www.jhu.edu/~scor. A Scientific Steering Committee was subsequently formed to write a Science Plan on GEOHAB (http://www.jhu.edu/~scor/GEOHAB_2001.pdf). The focus of GEOHAB is to foster a coordinated international science plan on the ecology and oceanography of HABs in the context of large-scale comparative ecosystems involving multidisciplinary studies on processes and mechanisms. This approach, ultimately leading to improved monitoring and prediction, will require the co-operation of researchers in many countries affected by HAB events.

The mission of GEOHAB is to "foster international co-operative research on HABs in ecosystem types sharing common features, comparing the key species involved and the oceanographic processes that influence population dynamics". The science goal is to "improve prediction of HABs by determining the ecological and oceanographic mechanisms underlying their population dynamics, integrating biological, chemical, and physical studies supported by enhanced observation and modelling systems". The science plan describes five program elements: 1) biodiversity and biogeography, 2) nutrients and eutrophication, 3) adaptive strategies, 4) comparative ecosystems, and 5) observation, modelling and prediction.

Canada is both affected by HABs and has been very active in HAB research in the past. Unfortunately, in recent years there has been a gradual decline in funding for HAB research in Canada. DFO provided seed money to initiate a workshop for the participation of oceanographers and marine ecologists to help develop a Canadian GEOHAB program. One strategy for Canadian researchers to continue to play an important role in the implementation of international programs is the formation of a national counterpart to the international GEOHAB program.

A preliminary meeting to discuss Canadian involvement in the international GEOHAB program was held May 26, 2001 in Nanaimo, British Columbia, immediately following the 7th Canadian Workshop on Harmful Marine Algae. A decision was made to hold a Workshop in Montréal on October 19 and 20, 2001, with the following *Ad hoc* Steering Committee, to organize the October Open Science meeting: Allan Cembella, Maurice Levasseur, Jennifer Martin, Suzanne Roy, Curtis Suttle, Max Taylor, and Ian Whyte.

The Canadian Workshop, held October 19 and 20 in Montréal, was chaired by Bjorn Sundby (Université du Québec, McGill University) and organized with the intent of coordinating scientific research and cooperation in order to develop international capabilities for assessment, prediction and mitigation of HABs through a better understanding of the ecology and oceanography affecting these algae. The Workshop Agenda is in Appendix 1. The Workshop was an Open Science meeting with a representative from the International Program (Allan Cembella), and participation from universities, government departments and agencies, industry and whoever else was interested in being involved. The list of participants is in Appendix 2. Participants were not necessarily required to be currently involved in HAB research but were to be able to link their knowledge – for example, as oceanographers, modellers, etc. Although 30 participants attended the Workshop, additional researchers indicated interest in a Canadian GEOHAB initiative and asked to be kept informed about the progress of the program. The Workshop provided a starting point so that participants (and those interested researchers unable to attend) could collaborate in future Canadian GEOHAB activities, including writing a joint Science Plan for submission to NSERC and other funding agencies.

Workshop participants

PRESENTATION SUMMARIES

Participants gave the following summaries of their research, as well as suggestions for how they would be able to contribute to a Canadian GEOHAB program. Some participants sent summaries and for those who did not, the editor summarized their presentation from notes. These summaries are listed below in the order that they were presented at the Workshop.

TOPICS OF RESEARCH FOR CONSIDERATION AND DISCUSSION IN A GEOHAB PROGRAM

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These proposed themes focus on harmful algal species with significant socio-economic impact to the west coast of Canada.

1) Effects of eutrophication due to salmon farms on the harmful phytoplankton community in Clayoquot Sound

The public perception that salmon farms are polluting and fostering formation of HABs along the 27,000 km coastline of British Columbia (BC) is encouraged by media coverage of salmon kills caused by harmful algae. The media stated that the recent kill of 80,000 farmed salmon in Clayoquot Sound by *Heterosigma akashiwo* was caused by increased eutrophication from salmon farm sites in the Sound. However, research to date on this topic has suggested no evidence of increased formation of harmful algal blooms (HABs) caused by salmon farming. Nevertheless, no detailed scientific research has been undertaken in BC to qualify and quantify interactions between oceanographic and environmental influences on harmful phytoplankton communities within Clayoquot Sound. Conducting research on phytoplankton communities in Tofino Inlet, adjacent to and distant from farm sites, would provide knowledge of bloom dynamics related to ecological and oceanographic features of the Inlet. In addition, this study would afford unequivocal scientific proof of any influence of salmon aquaculture on the formation, distribution, and periodicity of HABs.

2) Comparison of bloom dynamics of *H. akashiwo* in three distinct ecological regions around Vancouver Island - the Strait of Georgia, the Broughton Archipelago and Clayoquot Sound

Heterosigma akashiwo has been responsible for over $35 million loss in aquacultured salmon in British Columbia, with major mortalities occurring in July to October 1997. Bloom densities of 350,000 cells mL^{-1} were recorded in north Clayoquot

Sound, with the alga recorded to 18 m depth and over 80% salmon mortality. In Simoom Sound in the Broughton Archipelago, densities over 50,000 cells mL^{-1} were recorded with only minor salmon mortalities. In the Strait of Georgia, the alga was found to a depth of 25 m and at Departure Bay and the Gulf Islands, densities recorded were 90,000 and 8,000 cells mL^{-1}, causing over 78% mortality to farmed salmon. A comparison of temperature and salinity at lighthouses in the Strait of Georgia from June through October 1996 and 1997 clearly demonstrated similarities in annual temperature but substantially lower salinity during these months in 1997. Similarly, temperature and dissolved oxygen in locations in Clayoquot Sound during 1996 and 1997 were similar, but a significant decline in salinity was observed during 1997. These preliminary observations suggested a direct correlation between lowered salinity with increased stratification of the water column and the formation of *Heterosigma* blooms. More detailed studies on the ecological and oceanographic conditions that prompt formation and senescence at physically different oceanographic environments is required to provide bloom predictive capabilities.

3) Comparison of the bloom dynamics of *Alexandrium* spp. in Grappler Inlet in Barkley Sound and with Echo Bay in the Broughton Archipelago

From seasonal records of paralytic shellfish poisoning (PSP) in the sentinel species *Mytilus californianus*, taken from inlets around Vancouver Island from 1989 to 1994, a clear distinction was evident in the timing of *Alexandrium* spp. blooms in the Broughton Archipelago relative to other locations. Peak levels of PSP toxins in Echo Bay, Gilford Island in the Broughton Archipelago occurred in the early part of the year whereas at all other sites around Vancouver Island the peak occurrence was in summer and late fall. This difference provides an opportunity to clearly understand the ecological and oceanographic mechanisms causing bloom formations of *Alexandrium* spp. in different ecosystems within BC and also in other countries.

4) Comparison of bloom formation of *Pseudo-nitzschia* spp. in Holberg Inlet and Long Beach on the west coast of Vancouver Island

The razor clam is known to strongly retain domoic acid and is therefore an excellent long-term indicator of *Pseudo-nitzschia* spp. blooms, particularly in very remote areas. A 2 yr study was undertaken on the toxicity in clams in McIntyre Bay in the Queen Charlotte Islands in northern BC, and those taken from Cox Bay, near Long Beach on the west coast of Vancouver Island. Over the period of the study the northern clam contained less than 1.2 $\mu g\ g^{-1}$ of domoic acid in tissue, whereas those from Vancouver Island had 5-37 $\mu g\ g^{-1}$. The occurrence and infrequency of *Pseudo-nitzschia* spp. at these two locations was reflective of the different ecosystems. Lack of detailed ecological and oceanographic data reflected the logistics and lack of resources in adequately monitoring these sites, particularly those in the north. Further study is required to determine site-dependent occurrence of *Pseudo-nitzschia* spp. in different ecosystems such as the fjord-protected Holberg Inlet and open-ocean Long Beach. Expansion of this study to Oregon, Washington and Alaska would provide invaluable scientific information on *Pseudo-nitzschia* spp. on the west coast of North America.

5) Distribution of *Cochlodinium polykrikoides* in inlets on the west coast of Vancouver Island

In 1999, for the first time, blooms of *Cochlodinium polykrikoides* were responsible for salmon mortalities in Holberg Inlet and Quatsino and Kyuquot Sounds. Data collected in that year indicated that *Cochlodinium* was capable of out-competing *H. akashiwo* in all of the locations where they co-existed. *Cochlodinium* was also observed in bloom densities in Quatsino, Kyuquot, Nootka and Clayoquot Sounds on the west coast of Vancouver Island in 1999, suggesting a wider distribution than had been acknowledged previously. Although *Cochlodinium* cells were observed in the water column of these inlets in the following 2 yr, few blooms or farmed fish mortalities were recorded. Clearly, interannual variance in environmental and oceanographic conditions dictate the excystment and subsequent growth of these species. Certain shallow areas in these Vancouver Island inlets are considered probable cyst areas for *Cochlodinium*. Since *Cochlodinium* is a relatively recently discovered harmful alga on the west coast of Canada, any further study of its temporal and spatial distribution around Vancouver Island is deemed worthy of investigation.

Most of these themes concerning regional differences on the Pacific could be applied to comparisons with fjord, embayment and open-ocean ecosystems on the Atlantic coast.

THE BAY OF FUNDY PHYCOTOXIN PROGRAM AND FUTURE INVOLVEMENT WITH A CANADIAN GEOHAB PROGRAM

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HAB and marine toxin research in the Bay of Fundy has a long history at the St. Andrews Biological Station. For example, we began our work in the late 1970s with the earlier work on population dynamics of *Alexandrium fundyense* and establishing the linkage between PSP toxins, the food web and herring mortalities.

Work during the past 14 yr has included collecting data for a phytoplankton database documenting abundance and distribution (all species, including HAB species), salinity, temperature, nutrients (nitrate and nitrite, silicate, phosphate and ammonia), and fluorescence. Research has focused on environmental change and biodiversity as well as bloom initiation, dispersal and transport. The ultimate goal is towards prediction (or more accurately hind-casting) and early warning. Other research has included toxin uptake and depuration studies.

Our contribution to a Canadian GEOHAB program would fit into all program elements: 1) biodiversity and biogeography; 2) nutrients and eutrophication; 3) adaptive strategies; 4) comparative ecosystems; and 5) observation, modelling and prediction.

INTEREST AND CONTRIBUTION TO THE GEOHAB PROGRAM

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My work, led by Dr. Maurice Levasseur, focuses essentially on the role of climate variability on HABs. Our contribution to the GEOHAB program will be in the form of 1) elucidating the relationship between climate variability and HABs by hind-casting from historical data of *Alexandrium tamarense* blooms in the St. Lawrence (Program Element 1); and 2) improving our prediction ability of HAB events by developing indices and models for the St. Lawrence (Program Element 5). The approaches developed within our studies will be applicable to other Canadian coastal areas.

CLIMATE VARIABILITY AND TOXIC *ALEXANDRIUM TAMARENSE* BLOOMS IN THE ST. LAWRENCE: THE ROLE OF RAINFALL, RIVER RUN-OFF AND WIND

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Blooms of the toxic dinoflagellate *Alexandrium tamarense*, responsible for PSP, are annually recurrent events in the Estuary and Gulf of St. Lawrence. The analysis of abundance data for this algal species between 1989 and 1998 at Sept-Îles, a presumed initiation site in the northwestern Gulf of St. Lawrence, revealed yearly fluctuations in the onset, duration and magnitude of *A. tamarense* blooms. Hydrological and meteorological data for the region indicate that rainfall, local river run-off and wind are highly related with the pattern of bloom development each year. Results from the 10-yr dataset reveal that high river run-off from a prolonged spring freshet or from heavy rainfall events in the summer and fall were found to initiate *A. tamarense* blooms, and prolonged periods of high river run-off combined with weak winds favored the continued development of *A. tamarense* blooms. A strong correlation between surface salinity and the presence of *A. tamarense* cells was found at this station and could be used as a predictive tool for the presence of cells in this system.

The hypothesis that *Alexandrium* blooms are associated with brackish water is not new but it is the first time that we have the tool (a coupled ocean-ice model for the St. Lawrence) and the data (more than 12 yr of monitoring data from 11 coastal stations and 3 yr of cruise data) to test this hypothesis and to explore the possibility of using this climate-bloom relationship to develop a predictive tool.

INTERACTIONS BETWEEN HARMFUL ALGAE AND BENTHIC ORGANISMS: BENTHIC-PELAGIC COUPLING

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Grazing by suspension-feeding benthos, such as bivalve molluscs, is known to play a significant role in controlling phytoplankton biomass in shallow estuaries worldwide. Benthic organisms may thus replace zooplankton as the dominant grazer in nearshore waters. It is, therefore, important to determine the contribution of benthic organisms to the removal of harmful algal cells from the water column, and their accumulation in bottom sediments via the production of biodeposits (faeces and pseudofaeces). These processes have not been previously quantified for harmful algae, but it is well established that they can provide a key element in understanding phytoplankton dynamics in coastal waters. Such biological removal processes (top-down control) will need to be incorporated in the development of models describing the population dynamics of HABs.

The grazing pressure exerted by suspension-feeding benthos will depend on the bivalve species composition, the cell density and cell toxicity of toxic phytoplankton, and the relative composition of toxic and non-toxic species in a mixed phytoplankton assemblage. Bivalve species vary greatly in their ability to feed on toxic algae, and therefore in their capacity for toxin accumulation, as best documented for paralytic shellfish poisoning (PSP) toxins. Resistant species experience negligible feeding rate inhibition even in the presence of bloom densities of highly toxic *Alexandrium* spp., whereas sensitive species can experience varying degrees of feeding inhibition above a threshold level of cell toxicity. Recent work suggests that bivalves occurring at historical peak abundances in shallow Long Island, NY estuaries, could potentially control the initiation of brown tides of *Aureococcus anophagefferens* via grazing. Work in my laboratory (ECOHAB research) has also demonstrated that pronounced differences in grazing rates on toxic PSP-producing dinoflagellates may occur among bivalve populations of the same species depending on their long-term history of exposure to toxins and potentially their genetic makeup.

Benthic organisms can also affect the availability and water column resuspension of cysts and motile cells trapped in biodeposits via bioturbation of sediments and grazing activities. Thecate dinoflagellate cells of some toxic species are known to remain viable following ingestion by bivalve molluscs, and cysts of toxic algae have been identified in the gut contents of several suspension-feeding species, including offshore sea scallops. Benthic biodeposition and bioturbation may thus play a role in the transport and initiation of HABs but these processes have not been examined to date.

Sensitive benthic species can also be used as effective bio-indicators or early-warning systems for the presence of toxic and harmful algae, and thus act as an effective proxy or complement to phytoplankton monitoring. Development of rapid, inexpensive and sensitive toxicity bioassays is especially important in cases where phytoplankton toxins have not yet been chemically characterized.

Our main, proposed contribution to the Canadian GEOHAB program would be to quantify species-specific grazing and biodeposition rates of harmful algal species by key benthic species commonly occurring in Atlantic waters affected by HABs. Tracking the fate (viability, capacity for resuspension) of vegetative cells and cysts of harmful algae following ingestion by benthic organisms could also be incorporated in these studies. Collaboration with DFO and university researchers involved in modelling of phytoplankton dynamics in two major Atlantic estuaries (Bay of Fundy and the Gulf of St. Lawrence Estuary) would allow integration of biological loss processes of HABs in these models. HABs frequently co-occur with non-toxic species, especially during bloom initiation and termination phases, and past work has focused primarily on grazing in unialgal suspensions. Therefore, the emphasis of this work will be on the quantification of grazing rates in mixed assemblages of toxic and non-toxic species, in order to provide estimates of grazing pressure under more ecologically relevant conditions. The proposed research fulfills objective #3 of the International GEOHAB Science Plan: "to describe and quantify chemical and biological processes affecting species interactions", for example, by assessing "the importance of toxin production in selective grazing" and characterization of "modes of action of bioactive compounds on other organisms". It also fulfills the goal of understanding biologically mediated growth and loss processes, one of the three research goals identified at the 2001 Canadian GEOHAB workshop.

Additionally, our laboratory can contribute towards the development of methods for mass production of toxic algal species using bioreactor technology developed at the Institute for Marine Biosciences (IMB). These studies would allow production of harmful species (e.g. *Alexandrium* and *Pseudo-nitzschia* species, two of the targeted organisms for the Canadian GEOHAB program) under controlled conditions. These could provide a reliable inoculum for mesocosm studies of algal ecophysiology and behavior undertaken in other national and international laboratories. We have already successfully tested this technology using a high-toxicity *A. tamarense* strain from the Gulf of St. Lawrence Estuary, and could extend and test new methods for other HABs, including domoic-acid-producing *Pseudo-nitzschia* spp.

Finally, we can contribute to the program by developing novel, rapid bioassays for harmful algae involving behavioral, physiological, biochemical and/or cellular responses of early life history stages of bivalve molluscs. We have developed two such bioassays to date, based on the burrowing inhibition on infaunal bivalves caused by PSP toxins, and the feeding inhibition of suspension-feeding bivalves by brown tide (*A. anophagefferens*) cells.

NORTH COAST WATER QUALITY AND BIOTOXIN PROGRAM

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The North Coast Water Quality and Biotoxin Program coordinates testing services for shellfish harvesting and growing interests on the North Coast mainland and Queen Charlotte Islands in British Columbia.

In most other regions of Canada, the federal government provides these services. However, in 1964, a decision was made at the federal level to suspend these services to the North Coast. This resulted in one-third of BC's 27,500-km coastline being permanently closed for most shellfish activity for almost 40 yr. This closure impacted both First Nation traditional harvesting and recreational harvesting, and it has precluded the development of a shellfish farming industry on the North Coast.

The North Coast Water Quality and Biotoxin Program started out as a community-driven pilot project which was initiated in 1998 and expanded in 1999. The goal was to re-establish an official biotoxin sampling and testing program for the North Coast which would meet both the needs of the region and federal regulatory testing standards. Participants include two lease sites and the First Nations communities of Metlakatla, Kitkatla, Kitimaat, Lax Kw'alaams, Hartley Bay, and Haida Gwaii.

Biotoxin and phytoplankton sampling occur weekly from May 1-October 31 and every two weeks from November 1-April 30. Currently there are 13 biotoxin monitoring stations; six of these are also phytoplankton monitoring sites. Up to 15 more biotoxin monitoring stations may be established in the near future. Three biotoxins are monitored: Paralytic Shellfish Poisoning (PSP), Amnesic Shellfish Poisoning (ASP) and Diarrhetic Shellfish Poisoning (DSP).

Northern Laboratories Ltd., based in Prince Rupert, is the only laboratory in Canada which has been approved to both: 1) prepare PSP and ASP extracts for the Canadian Food Inspection Agency (CFIA), and 2) process fecal coliform tests for Environment Canada. Biotoxin samples must reach the laboratory within 24 h of collection, and water samples must reach the laboratory within 6 h of collection. Given the remote location of most of the North Coast communities participating in the North Coast Water Quality and Biotoxin Program, this local laboratory capacity is vital.

Currently, Dr. B. Shaw (Ocean Ecology) analyzes regular phytoplankton samples and provides reports on the prevalence of *Alexandrium*, *Pseudo-nitzschia* and *Dinophysis* spp. at each of the six monitoring stations. The *Alexandrium* and *Pseudo-nitzschia* results are correlated to CFIA's mouse bioassay results for PSP and HPLC results for ASP, respectively. In June 2001, Environment Canada conducted shoreline surveys and fecal coliform testing in Hartley Bay, Kitimaat, Kitkatla, Humpback Bay, Oona River, Metlakatla and Lax Kw'alaams. On October 10, 2001, the Pacific Shellfish Classification Committee classified these growing areas as either *Approved* or *Closed*, based on the survey results.

The North Coast Water Quality and Biotoxin Program has successfully met both Environment Canada and the CFIA's testing requirements, and areas of the North Coast are ready to be opened, pending approval by Fisheries and Oceans Canada.

Since the inception of this program, several alternative technologies have been investigated in a series of studies. These technologies include the MIST QUANTI™ test for PSP, the MIST™ ALERT test for PSP, phytoplankton monitoring and the use of satellite data as a predictive tool for toxic algal blooms. The North Coast Water Quality and Biotoxin Program is interested in participating in a Canadian GEOHAB program as a potential partner for scientific research on biotoxins and alternative monitoring/testing technologies.

This program is supported by: Community Futures Development Corp., The Community Economic Adjustment Initiative, Prince Rupert Economic Development Commission, Fisheries Renewal BC, The Skeena Queen Charlotte Regional District, Northern Laboratories Ltd., The Haisla Fisheries Commission, and The Canadian Food Inspection Agency.

SURVEY OF HARMFUL CYSTS IN THE SEDIMENTARY COLUMN

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As part of some HAB life cycles, a resistant stage called a cyst occurs in the sediments. Cysts fall to the sediment surfaces following reproduction of the toxic dinoflagellate vegetative cells in the water column. Cysts can be buried directly as a result of hydro-dynamic processes in the deposit zone or through sedimentary mixing (bioturbation) by benthic organisms. Various invertebrates, as suspension feeders or deposit feeders (molluscs and polychaetes), can also consume cysts. We plan on evaluating 1) cyst burial by sedimentary transport or by bioturbation processes, and 2) cyst predation by invertebrates in the sedimentary column.

1) Cyst quantification in the sedimentary column in their natural habitats

We have developed a new method for quantification of biogenic structures in the sedimentary column using computed axial tomography or CAT Scan. This method enables us to quantify and determine the position of cysts in the sedimentary column. We sampled 10 sediment cores from a location where cysts were previously observed. Sediments were collected from a sandy biotype under strong hydrodynamic conditions and a muddy sand environment in low hydrodynamic conditions. Each sediment core was sealed with a paraffin plug to preserve biogenic structures and cyst position in the sedimentary column. All scanner analyses were done using the INRS-Géoressources CAT Scan in Québec (100 μm resolution). The following protocol was used for each sediment core: one longitudinal cut, and transverse cuts of 5 mm thickness between 0 and 200 mm. The sediment space occupied by the biogenic structures was calculated from five values of tomography intensity (IT) chosen in three biogenic structures, (15 IT values) by cut layer and in sediment matrix. These structures are chosen so that the IT value is nearest to the IT value of the sedimentary matrix, in order to get an IT value limit for biogenic structures. For cysts, we try to delimit their values (IT) in order to quantify them in each cut. These values are calculated for every cut and results are presented under a bioturbation in profile in relation to the number of cysts and quantified according to depth.

2) Cysts buried in the sedimentary column by bioturbation processes

In order to measure the effect of sedimentary mixing (bioturbation) on the physico-chemical properties of sediments and cysts burying in sediments, we have developed the following experimental approach. The experimental setting is made up of several modules, each containing four reconstituted sediment cores. Sediment cores are 20 cm in length and 10 cm in diameter. A tube system positioned at different levels allows water or sediment samples to be taken. A water sample is taken from the sediment surface (1 cm). In the sedimentary layer, samples are extracted at depths of 1 cm, 5 cm and 14 cm. Each sediment core is filled with defaunated sediment up to 15 cm and the rest of the column filled with circulating seawater. One week after the stabilization of sediments, we add a natural density of benthic organisms for each of the studied habitats (one or many species). During the experiment (14 d), the temperature is maintained at 13°C, the salinity at 27 psu and the photoperiod at 12 h light/12 h dark. After settlement of benthic organisms, cysts are added to the sediment surface in densities equivalent to the natural habitat in order to evaluate burial by benthic organisms during the experiment.

Every 2 d, water is sampled at the four depths to evaluate dissolved oxygen, chemical oxygen demand (COD), nitrate, nitrite and ammonium ions. Variations of these parameters in sediment water will give us an evaluation of the modifications of the global system by bioturbation. Dissolved oxygen is measured directly with a voltametric microprobe (Luther et al. 1998). Measures of COD are done using the UVS method developed by Deflandre and Gagné (2001). Nitrate, nitrite and ammonium are measured by colorimetric methods (Alpkem autoanalyzer). Bacterial counts are done on sediments sampled with a syringe during the experiments. Bacteria are counted with an epifluorescence microscope after coloration (DAPI methods, Porter and Feig 1980). Following the experiment, all sediment cores 0-5 cm deep are cut in slices of 0.5 cm thickness and 1 cm thickness from depths of 5-15 cm. Cyst counting for each slice is done microscopically in order to get a profile of cyst densities according to depth in the sedimentary column.

3) Cyst predation by benthic organisms

We want to evaluate cyst predation by dominant benthic organisms. A known quantity of cysts will be put on the sediment core surface filled with natural densities of characteristic species from muddy sand or sandy biotopes. Every day, for the duration of the experiment (10 d), we will collect faeces at the surface sediment to evaluate the number of cysts that passed through the organisms' guts. Following the experiment, cores will be disassembled and the sediment sieved to evaluate the ratio of migrated cysts to absorbed cysts.

Collaborators on these projects are B. Long (INRS-Géoressources) and B. Sundby (ISMER).

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BIOLOGICAL FACTORS AFFECTING GROWTH AND MORTALITY OF HABS

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Nuisance blooms of phytoplankton organisms are sporadic but recurrent events in Canadian marine waters. While physical conditions set the stage for HABs, physiological responses to local conditions (such as nutrients) and biological interactions determine the amplitude and length of blooms. Like other phytoplankton species, HABs are sensitive to zooplankton grazing and pathogen infection (viruses and bacteria). However, while meso- and microzooplankton control of phytoplankton is generally well studied, little is known about their impacts on HABs. How morphological features, poor nutritional quality, intracellular or excreted toxins as well as other allelopathic compounds in HABs work as feeding deterrents are poorly known. A variable susceptibility of toxic algae to grazers is considered to reflect varying toxin levels depending on algal growth conditions. Although there are only a limited number of studies, it has been shown that certain phytoplankton species release compounds to retard grazing under nutrient-limited conditions. On the other hand, pathogens are now considered to be extremely important for the termination of phytoplankton blooms. The most common pathogenic agents in the sea are viruses (typically numbering 10 billion per litre) and there is indication that HABs may be important hosts for viruses. For example, recent observations in the St. Lawrence Estuary suggest that viruses could play an important role in *Pseudo-nitzschia* spp. bloom dynamics. The relative contribution of grazing and viral lysis on mortality of HABs remains nevertheless essentially unknown.

In the context of the Canadian GEOHAB program and in collaboration with Drs Maurice Levasseur (IML), Suzanne Roy (ISMER), Stephen Bates (Moncton), Curtis Suttle (UBC), Allen Cembella (NRC), and Michel Gosselin (ISMER) we propose:

1) To determine the susceptibility of *P. seriata* and *A. tamarense* to grazing by meso- and microzooplankton and viral infection when grown under different environmental (nutrient, light, UV) conditions;

2) To investigate the capability of *P. seriata* and *A. tamarense* to produce allelopathic compounds in order to avoid competition, grazing and infection, and to determine conditions under which such compounds are produced and released;

3) To improve our capability to monitor and study *P. seriata* and *A. tamarense* blooms in the St. Lawrence Estuary by using appropriate molecular probes and flow-cytometry;

4) To isolate and maintain in culture pathogens specific to *P. seriata* and *A. tamarense* in order to conduct laboratory studies on their influence on bloom

dynamics of these toxic species and to explore the possibility of using them for bio-control.

LOSS PROCESSES IN REGULATING HABS

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The big issue for the Canadian GEOHAB program to address is the role of loss processes, especially mortality, in regulating phytoplankton blooms. Even small increases in mortality can shift production from net negative to net positive. Within the broad perspective of loss processes, viral mortality is potentially very important but much less well understood. Viruses are now recognized as major mortality agents for microbial communities in marine systems (Suttle 1994; Fuhrman 1999; Wommack and Colwell 2000). Virus-like particles have been described in more than 40 taxa of algae (Van Etten et al. 1991), and there is good evidence of termination of phytoplankton blooms by viral lysis (Bratbak et al. 1993). Among the toxic bloom-forming phytoplankton in Canadian waters, *Heterosigma akashiwo* is known to be infected by viruses. This alga is of international concern and has caused major fish kills in Canada, Japan, United States and Australia. Japanese investigators have shown that virus-infected cells are more abundant near the termination of blooms (Nagasaki et al. 1994, 1996). As well, in British Columbia coastal waters, a number of different viruses infecting *H. akashiwo* have been isolated, including double-stranded DNA and RNA viruses that bear little resemblance to known viruses (Lawrence et al. 2001; Tai et al. in press). Given these viruses are widespread in coastal waters of BC, it implies that they are agents of mortality. Their involvement in preventing bloom initiation or causing bloom termination is unknown. Not only are viruses infecting *H. akashiwo* widespread in surface waters, but they also occur at depths of at least 40 cm in coastal sediments. This reservoir of infectious viruses may also be important in bloom dynamics.

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STUDY OF *ALEXANDRIUM TAMARENSE* BLOOM DYNAMICS IN THE ST. LAWRENCE ESTUARY (CANADA): A MODELLING APPROACH

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Toxic dinoflagellate blooms are greatly influenced by their physical environment, and hence climate forcing (e.g. precipitation, wind conditions). Biological factors, such as the spatial distribution and germination rates of resting cysts, the growth rate and vertical migrations of vegetative cells, also play a significant role in the development, maintenance and dissipation of these blooms. Exactly how these different physical and biological factors affect the dynamics of blooms of the PSP producer *Alexandrium tamarense* in the St. Lawrence Estuary (SLE) is still not well understood. The recent development of climatically driven circulation models for coastal areas and their coupling with appropriate biological models offer the opportunity to test hypotheses linking bloom dynamics with particular climatic, hydrodynamic and biological conditions. To explore the causes of an *A. tamarense* red tide observed in 1998 in the SLE, we used a Regional Ocean Model that reproduces the three-dimensional water circulation under atmospheric, hydrological and oceanic forcings. In the two scenarios tested, the initial spatial distribution of *A. tamarense* vegetative cells was based on the known spatial distribution of resting cysts in sediments of the SLE and a 20% germination rate. First, vegetative cells were followed as passive Eulerian tracers in order to determine if the distribution of cysts, the timing of germination, and advection in the SLE could explain the general distribution of *A. tamarense* cells during the red tide. Second, a growth rate of 0.5 division per day was set for vegetative cells to verify if cellular growth can account for the *A. tamarense* abundance observed during the red tide. Our preliminary results highlight the potential of this approach to understand and eventually predict toxic blooms in coastal waters.

We expect to pursue this modelling effort in the context of the Canadian GEOHAB program and more particularly to develop a coupled bio-physical model for the toxic diatom *Pseudo-nitzschia seriata* which is now responsible for ASP outbreaks in the St. Lawrence Estuary and Gulf. We would also like to run a comparative study between the St. Lawrence, the Strait of Georgia, the Bay of Fundy and the Gulf of Maine in order to highlight differences and similarities in term of impacts of meteorological and hydrodynamic forcing on HABs. This would imply collaborations with J. Martin, I. White and D. Anderson.

DISTRIBUTION OF DOMOIC ACID CONCENTRATIONS IN SHELLFISH AND OF *PSEUDO-NITZSCHIA* SPP. IN THE ST. LAWRENCE FROM 1999 TO 2000

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The presence of domoic acid (the toxin responsible for ASP) in shellfish has been monitored in the Estuary and the northern Gulf of St. Lawrence since 1997 by the Canadian Food Inspection Agency (CFIA). The results indicate a spatial and temporal evolution of domoic acid contamination between 1998 and 2000. Trace amounts of this toxin were first detected in the gonads of sea scallops from fishing areas offshore of the Îles-de-la-Madeleine in the summer of 1998. In 1999, the concentration of domoic acid in the digestive glands of sea scallops from the same area reached 585 $\mu g\ g^{-1}$, whereas the adductor muscles were not contaminated. At the same time, concentrations of domoic acid close to 25 $\mu g\ g^{-1}$ digestive gland were measured in sea scallops from the Havre-aux-Maisons Lagoon while trace amounts were measured for the first time in soft-shell clams collected on the Lower North Shore of the Gulf of St. Lawrence. In 2000, the digestive glands of sea scallops from the Îles-de-la-Madeleine remained toxic and trace amounts of domoic acid were measured in shellfish all along the North Shore, from Tadoussac to Havre-Saint-Pierre. In addition to the CFIA data, the Harmful Algae Monitoring Program from Institut Maurice Lamontagne (Fisheries and Oceans Canada) observed the presence of two potentially domoic acid-producing diatoms in the St. Lawrence: *Pseudo-nitzschia seriata* and *Pseudo-nitzschia delicatissima*. Analysis of data showed a link between domoic acid in some shellfish from the Îles-de-la-Madeleine and North Shore and the presence of *P. seriata*. Dense blooms of *P. delicatissima* (with no *P. seriata*) did not cause toxicity. Laboratory analyses performed on a *P. seriata* strain isolated from the St. Lawrence Estuary during a toxic event showed the ability of *P. seriata* to produce domoic acid whereas all attempts made with *P. delicatissima* from other regions of eastern Canada have so far been negative. These new results show that *P. seriata* blooms in the St. Lawrence and the resulting shellfish toxicity due to domoic acid represent a potential risk that needs to be addressed in the future.

In the context of the Canadian GEOHAB program and in collaboration with Drs. Michel Starr (IML), François Saucier (IML), Stephen Bates (Moncton), Suzanne Roy (ISMER) and Michel Gosselin (ISMER), we are proposing to conduct an in-depth analysis of the data collected during the IML monitoring program in order to determine, as much as the data permit, the key factors controlling these toxic diatom blooms in the

St. Lawrence. We are also planning to conduct laboratory studies on the physiology and ecology of these species (*P. seriata* and *P. delicatissima*) and to develop a coupled bio-physical model based on the one currently developed for *Alexandrium tamarense*.

NUMERICAL MODELLING OF THE CIRCULATION AND PARTICLE TRANSPORT IN COASTAL STRATIFIED WATERS

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My research expertise is in the field of coastal oceanography, with emphasis on the dynamics of physical oceanographic processes and their influence on dissolved and particulate matter transport in stratified coastal waters. My studies are carried out in lagoons, bays and small estuaries in the Gulf of St. Lawrence and elsewhere in the world, and the scientific approach is through field time series measurements and numerical 3D modelling. More recently, I became involved in the study of shellfish and finfish mariculture site selection and the environmental impacts of these sites on the surrounding nearshore ecosystem. For example, on-going field and numerical modelling studies are carried out in the baie de Gaspé, in the Magdalen Islands lagoons and in the Richibucto estuary, within the Gulf of St. Lawrence.

My contribution to the Canadian GEOHAB program can be within a multi-disciplinary study of the environmental factors affecting the onset of a HAB in coastal stratified waters, the dispersion of the bloom and its decay within coastal waters, bays and fjords in eastern and western Canada.

Field equipment such as current meters, tide gauges, CTDs and others, as well as ISMER's 50 m research vessel, can be made available for the GEOHAB study. The numerical models to be used are the MIKE3 integrated suite of three-dimensional models (http://www.dhi.dk) including a hydrodynamic module, a Lagrangian particle transport module, a suspended sediment module with a three layer parameterization of sediment, a water quality module including the cycling of oxygen, nutrients and BOD and a eutrophication module including the cycling of nutrients, detritus, bacteria, phytoplankton and zooplankton. All the MIKE3 modules are coupled within the same Windows environment.

REPORT ON PARTICIPATION IN THE GEOHAB WORKSHOP

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I am a doctoral candidate in the interuniversity oceanographic program (Laval, McGill, UQAR, expected date of completion April 2002). My thesis research has been in the North Water Polynya, eastern Canadian Arctic, exploring interactions between phytoplankton and other protists and bacteria. In particular I am interested in what promotes species-specific blooms of phytoplankton in general. In 1997, I worked on the HAB species *Gymnodinium catenatum* with Dr. Hallegraeff at the University of Tasmania in Australia. The initial aim of my study was to investigate the effect of CDOM (chromatophoric, dissolved organic matter, or humic substances) on *G. catenatum* and associated bacteria. During this research, I isolated a bacterium than causes rapid lyses and death in several HABs (*G. catenatum, Chattonella marina* and *Heterosigma akashiwo*). Subsequently, more algicidal bacteria have been isolated. The implications of this phenomenon include the suggestion that HAB incidents may occur when natural loss processes in the water column or sediments fail to operate and conditions become favourable for mass recruitment of HABs. Algicidal bacteria may also contribute to the rapid decline of existing blooms.

I am currently applying for post-doctoral positions in Canada and overseas. However, since my primary interest is in the ecological and oceanographic conditions that promote species-specific blooms, I hope to be able to eventually work on HAB projects in Canada. I appreciate the opportunity to contribute ideas, based on my experience, in planning future directions.

BACKGROUND AND INTEREST IN THE CANADIAN GEOHAB PROGRAM

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I have worked mainly on laboratory-based projects that study the physiology of phytoplankton. My involvement with HAB research began soon after the 1987 discovery that the diatom *Pseudo-nitzschia multiseries* was the source of domoic acid in mussels in PEI. I have since been studying factors that control the growth of and toxin production by diatoms of this genus (e.g., irradiance, temperature, nutrients, iron, bacteria, and organic compounds). Comparisons have been made with non-toxic *Pseudo-nitzschia* species. Findings from these laboratory experiments now need to be verified in field populations of this diatom genus. For example, laboratory cultures of *P. multiseries* show that toxin production does not begin until the cells slow, then stop their cell division during stationary phase, due to silicate or phosphate limitation. Domoic acid has nevertheless been found during blooms of *P. multiseries*. It is possible that the toxin may be produced during brief periods of nutrient depletion and cessation of cell division, in between pulses of nutrients that flow out of rivers following rainfall events. Any field approach requires the application of existing and novel methods to assess growth and cell physiology in natural populations. Other laboratory work includes the development of cell surface antibodies that enable the distinction between *P. multiseries* and *P. pungens*, and the testing of rRNA probes that successfully distinguish these species in Canadian waters. The process of sexual reproduction in *P. multiseries*, *P. pungens*, and *P. pseudodelicatissima* has also been elucidated for the first time. Possible differences in toxicity between new cells resulting from sexual reproduction, and those older cells which have decreased in apical cell length, may also have implications for bloom toxicity and periodicity. Thus, a study of sexual stages of this diatom genus would be important in any field study.

The use of laboratory cultures, which is an accepted component of the GEOHAB Science Plan, thus has several advantages. These include the ability to: determine growth and toxin production control factors under controlled conditions; develop molecular probes; study gene expression (e.g., I am on the PhD thesis of Katie Rose Cellineri – MIT-MIT Joint Program, who is studying the up- and down-regulation of gene expression in *P. multiseries*, using the DNA microarray approach); and compare clonal and strain differences and variability.

Of particular interest has been the study of the interaction between bacteria and *P. multiseries* in culture. When bacteria are eliminated from the diatom cultures by antibiotic treatment, toxin production by these axenic cultures decreases dramatically to very low levels. Reintroduction of any of a number of bacteria to the previously axenic cultures results in a significant enhancement of toxin production, sometimes exceeding the original level. We have recently discovered, using scanning electron microscopy (in collaboration with Dr. Irena Kaczmarska and Jim Ehrman – Mount Allison University), several types of bacteria attached to the surface of *P. multiseries* cells. Most of the bacteria are attached between the girdle bands and along the raphe, where organic material can be seen to be extruding from the cell. The significance of these attached bacteria for domoic acid production remains to be determined, as does the mechanism of enhancement of toxin production by free-living bacteria. The bacteria also require characterization. These experiments demonstrate the need to consider bacteria when studying the bloom dynamics and toxicity of field populations.

Since the discovery of domoic acid as a phycotoxin in 1987, other countries around the world have become concerned about the possible presence of toxigenic species of *Pseudo-nitzschia* in their own waters. As a result of such concerns, and because of certain seabird and marine mammal mortalities, researchers around the world have now documented nine species of toxic *Pseudo-nitzschia*. This has led to international collaborations with researchers in Scotland, Ireland, Bermuda, South Africa and Chile. As well, collaborative work is underway with scientists studying populations of toxic *Pseudo-nitzschia* spp. along the Atlantic coast of NS, in the upper Gulf of St. Lawrence, and in the Bay of Fundy.

Current work involves a component of the ESSRF-funded program "Integrated Ecosystem Studies for Modelling Mussel Aquaculture-Environment Interactions". Samples are being collected for the identification and enumeration of phytoplankton species, including *Pseudo-nitzschia* and other toxic species, in selected embayments of PEI during the late summer and autumn seasons. The goal is to document the species composition and dynamics of toxic and benign phytoplankton in relation to ambient nutrient concentration and the level of mussel aquaculture within the embayments. This work is being carried out in collaboration with Jennifer Martin, Murielle LeGresley and Fred Page (St. Andrews Biological Station).

My interest in GEOHAB is thus 1) to maintain in culture different clones of several *Pseudo-nitzschia* species for directed laboratory experiments, and 2) to study the bloom dynamics of *Pseudo-nitzschia* and other toxic species, in relation to benign phytoplankton species and nutrient conditions, in contrasting ecosystems in Canadian waters and internationally.

MULTIVARIATE COMMUNITY STRUCTURE AND TIME SERIES ANALYSES OF PHYTOPLANKTON FROM THE BAY OF FUNDY: SOME WORK IN PROGRESS

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As described earlier in this workshop proceedings by J. Martin et al., phytoplankton in the Bay of Fundy has been monitored at several locations for over 14 yr. At each location a single water bottle sample has been obtained from each of several depths, at monthly to weekly intervals throughout the year (the sampling interval is monthly in the winter and weekly during the spring, summer and fall periods). The samples have been manually sorted under a microscope to determine species composition and the abundance of each species.

In an effort to describe and explore the temporal and spatial variability in the community structure of the phytoplankton community in the Bay of Fundy, we have begun to use a series of univariate and multivariate statistical analyses techniques. The techniques include traditional indices of diversity as well as cluster and ordination techniques based on the Bray-Curtis and Jaccard indices of similarity. The primary software tool used to implement these techniques has been PRIMER, an analysis tool developed by Clarke and Warwick that has become widely used within the biodiversity community. We have also used descriptive statistical approaches such as cumulative abundance curves.

As an example of these approaches, the cumulative abundance curves of the total dinoflagellate abundance from one of the Bay of Fundy monitoring station for the years 1987-99 is shown in Fig. 1. The curves indicate that the timing of the dinoflagellate annual cycle is relatively constant from year to year. Figure 2 shows the two dimensional representation of the multi-dimensional scaling (MDS) ordination analyses of the monthly mean species specific abundance matrix for one of the monitoring stations. Each number on the figure represents the MDS co-ordinates of one of the monthly means. The bolded numbers represent the averages of the MDS co-ordinates for each monthly mean. The line joining successive points highlights the sequential nature of the ordination and indicates the seasonal cycle in the community structure. These types of analyses are being used to explore the temporal and spatial structure in the phytoplankton community within the Bay of Fundy and to identify the species or taxonomic levels that are defining the patterns. They are also being used in conjunction with environmental data to suggest potential environmental patterns associated with the biological patterns.

Abundance estimates of organisms, such as phytoplankton, are often highly variable. If repeated samples are taken at a location the variance in the estimates of abundance can be quite high, in some cases the magnitude of the variance equals the mean. This is well known and attributed to small-scale plankton patchiness. As a result, time series of the abundance of organisms, such as phytoplankton, that are based on point samples are often characterized by frequent spikes. This variability needs to be taken into consideration when trying to interpret the time series of abundance. For example, are there interannual differences in the phasing and amplitude of the annual cycle, or are the apparent differences consistent with the expected level of sample variability?

Statistical time series methods appear to hold promise for analyzing this type of data. We have begun to explore the utility of a state space approach for examining interannual variability in phytoplankton abundance. This analysis is based on a stochastic cycle model embedded in the framework of a Kalman filter, with the associated fixed interval smoother, and using maximum likelihood parameter estimation. To date, the approach has been applied to a time series of the total abundance of dinoflagellates per sample collected from a single sample site, the Wolves. The time series extended from the year 1987 through 2000, and consisted of over 300 samples or abundance estimates. The method indicates that the annual cycle in log-transformed dinoflagellate abundance can be adequately described by a simple sine curve with fixed annual period, but randomly drifting amplitude and phase. The analysis suggests that there has been significant (at 90%) interannual variability in the amplitude and phase of the dinoflagellate abundance cycle within the Bay of Fundy for only 2 of the 12 yr examined.

In the context of GEOHAB, there are several lines of investigation we are interested in pursuing. We have interests in statistical analyses of abundance and environmental datasets, utilizing new observational technologies and in process modelling. With respect to statistics, we would like to continue exploring descriptive and predictive statistical methodologies. These would likely include analyses addressing questions such as how the harmful algae species fit into the larger phytoplankton community structure, whether the time series of phytoplankton abundance is associated with environmental variability and whether time series methods offer a useful tool for analyzing and forecasting the abundance of single species of harmful algae. With respect to new environmental technologies, we are interested in using multi-spectral optical techniques and optical imaging analyses approaches to obtain species-specific datasets on finer temporal and spatial scales than traditional bottle sampling and manual sorting approaches have allowed. We are also interested in utilizing circulation models as tools for enhancing understanding of phytoplankton dynamics. Hopefully, some of these interests can be pursued in collaboration with interested colleagues with data and models from other geographic areas.

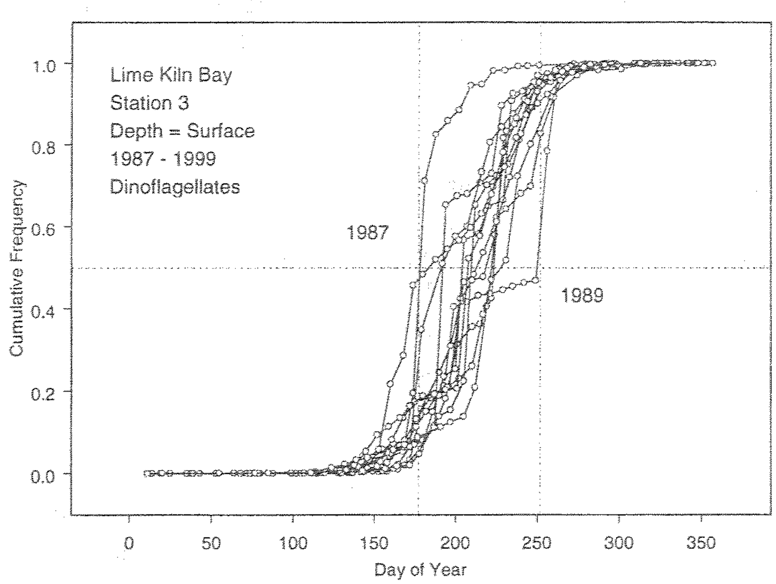

Fig. 1. Cumulative abundance curves of all dinoflagellates collected from a monitoring station within the Bay of Fundy during the yr 1987-99.

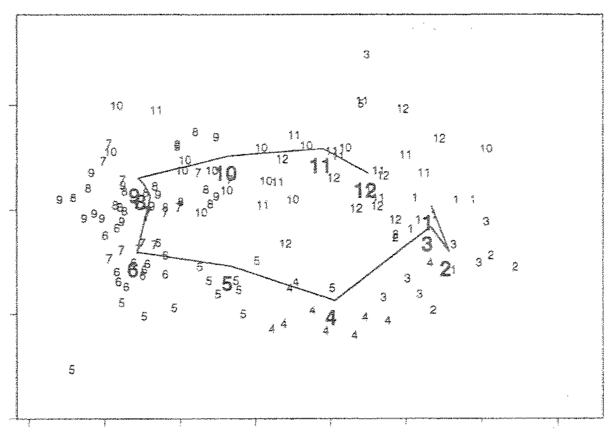

Fig. 2. A multi-dimensional scaling ordination plot based on the matrix of species-specific monthly mean abundances of phytoplankton and sample date (month and year) for a Lime Kiln Bay monitoring station within the Bay of Fundy during the years 1987-2000. Each abundance value was group averaged (month-year) and fourth root transformed and organisms were aggregated to the species level. 2D stress was 0.19, cyclicity was 0.066 (P=0.001) and seriation was 0.116 (P=0.001). Numbers indicate the sampling month and the large numbers indicate the centroid of the MDS co-ordinates for each month.

HYPOXIA IN THE DEEP WATERS OF THE LAURENTIAN TROUGH, LOWER ST. LAWRENCE ESTUARY

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Hypoxia has become a growing concern in coastal regions throughout the world (Rosenberg 1985; Nixon 1995; Rabelais and Turner 2001). A watermass is hypoxic when the concentration of dissolved oxygen is so low as to cause stress for aquatic organisms. In general, this happens when the oxygen concentration is 2 mg L^{-1} (65 μM) and less. At this level, many fish species cannot survive, and the benthic community structure undergoes significant modifications (e.g. Diaz and Rosenberg 1995; Sagasti et al. 2000; Karlsen et al. 2000). Notable examples of hypoxia can be found in the Baltic, the Kattegat, and on the shelf region of the northern Gulf of Mexico that receives the outflow of the Mississippi River. The extent of the Gulf of Mexico hypoxia has more than doubled during the last 15 yr, which has prompted our neighbours to the south to initiate an extensive research program (Goolsby 2000).

During a recent cruise to the Lower St. Lawrence Estuary, sporadic measurements of dissolved oxygen revealed concentrations of 65 μM and less in the bottom 50 m of the water column (Gobeil, C., unpublished). This is below published values of the oxygen concentration in this region (90 μM) (Dunbar et al. 1980; Yeats 1988), suggesting we may be experiencing a trend towards hypoxia and perhaps even anoxia. A rough estimate suggests that the area bathed in water of such low O_2 concentrations may cover more than 1000 km^2 (Gobeil, C., unpublished). This observation is cause for concern because of the effects low oxygen will have on benthic and epibenthic fauna, and on nutrient release by sediments and subsequent effects on primary production. We may also be observing a trend of decreasing oxygen concentrations that could ultimately lead to complete anoxia in the deep water, which would be nothing less than an environmental disaster.

Bottom water hypoxia in the Laurentian Trough is not a seasonal phenomenon because the water column is permanently stratified and the bottom water is isolated from the atmosphere. This distinguishes the St. Lawrence from shallow environments such as the Gulf of Mexico shelf, where hypoxia is seasonal. In these areas, hypoxia develops in the bottom water during spring and summer, but the conditions return to oxic when vertical mixing of the water column by strong winds in fall and winter replenishes the deep water oxygen. In the St. Lawrence, oxygen in the deep water cannot be replenished by mixing. The oxygen concentration at a given location is determined uniquely by the oxygen concentration in the water that flows from the Atlantic Ocean along the bottom towards the head of the Laurentian Trough and by the local rate of oxygen consumption. There are no local sources of oxygen. Thus, whereas the bottom water oxygen concentration in the Cabot Strait region is about 60% of saturation, it is now down to 15% of saturation in the Estuary.

HYPOTHESES AND QUESTIONS

We hypothesize that, as in the case of the Gulf of Mexico, the low oxygen concentrations are at least in part a response to an increased loading of nitrogen nutrients (Nixon 1995). Unlike freshwater environments, where phosphorus usually is the limiting nutrient, ammonia and nitrate can limit primary production in coastal marine areas. Alternatively, it could be a response to other factors including changes in fresh water run-off, increased terrestrial organic matter input, change in the supply and/or properties of bottom waters supplied from the Atlantic Ocean. Therefore, the existence of low oxygen concentrations in the bottom waters raises a number of questions, including:

- Is this a recent phenomenon or is it normal for this region?
- What is the extent of the area that is now underlying low oxygen concentrations and is it changing?
- Has nutrient supply to the estuary, biological production, and fluxes of organic matter to the deep water been changing?
- What is the rate of oxygen consumption in bottom water and underlying sediment? How does this compare to the rate of oxygen supply?
- What are the present controls on oxygen supply and oxygen consumption?
- Is the North Atlantic oscillation an important factor in determining the flow of deep water and oxygen supply in the Gulf of St. Lawrence?
- Would changes in fresh water run-off affect the residence time and the oxygen balance in the deep water?
- Would changes in fresh water runoff and associated changes in stratification and vertical mixing affect the primary production in the St. Lawrence Estuary?
- What would be the effects of hypoxia on the ecology of the St. Lawrence Estuary?

RESEARCH NEEDS

1) Document the extent of the hypoxic region in terms of oxygen and key nutrients;
2) Consolidate historical data and evaluate the evolution of the hypoxic region;
3) Refine the present understanding of the water flow in the near-bottom layer in the Gulf and the oxygen supply to the deep water in the lower Estuary;
4) Determine the rates of oxygen consumption in water and sediment in the hypoxic region;
5) Develop a mass balance for oxygen supply and consumption for the deep waters of the Estuary and Gulf, including the hypoxic region. Establish the relative role of water column and benthic respiration on the apparent oxygen utilization in the bottom waters;
6) Develop a conceptual model of oxygen supply and use along the flow path of the bottom waters entering the Estuary;

7) Describe the benthic community characteristics (diversity, abundance and assemblages) in relation with oxygen concentration and differentiate between the decrease in diversity associate with the estuarine gradient and the decrease in oxygen concentration.

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SUMMARIES BY THE EDITOR

CONTRIBUTIONS/COLLABORATIONS WITH HEALTH CANADA

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Health Canada's research group has become more involved in toxic syndromes since 1987, when domoic acid was detected in shellfish. The involvement of Health Canada is primarily clinical, with cardiovascular abnormalities being investigated and receptor sites on peripheral organs. In addition, collaborative research is being carried out with NRC in the study of spirolide toxicity. Presently, the knowledge is limited to the effects of toxins on humans.

RESEARCH AT THE UNIVERSITY OF WESTERN ONTARIO

Lisa Pickell and Charles Trick

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The primary HAB research focus at the University of Western Ontario is studying physiology and productivity of *Heterosigma akashiwo* blooms, and mechanisms for toxicity and effects of iron. Currently, a project is ongoing tracking the historical profiles of bloom occurrances in British Columbia through settling cores. This technology can be applied to a Canadian GEOHAB program.

SUPPORT FROM DFO – BEDFORD INSTITUTE OF OCEANOGRAPHY

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BIO has a satellite receiving system covering the east coast of Canada. The SeaWiFS footprint at present is 1 mi^2. Expertise in a wide variety of disciplines is available at BIO and a number of researchers (working in the fields of modelling, oceanography, radionucleides, remote sensing, etc.) have expressed interest in keeping current in Canadian GEOHAB progress.

MONITORING ALGAL BLOOMS WITH *IN SITU* OPTICAL SENSORS

John Cullen

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In situ optical sensing technology and rapid diagnostic assays can provide new opportunities for autonomous environmental monitoring and assessment of phytoplankton blooms and seston quality. Moored optical sensors can provide continuous measurements of water properties for documented long-term trends in water quality, in addition to transient plankton events, including HABs. Optical sensors are tools that can be incorporated into an existing phytoplankton monitoring program to provide 'real-time' measurements.

FUTURE OF THE CANADIAN GEOHAB

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When identifying priorities for the Canadian GEOHAB program, the role of viruses needs to be addressed. As does the majority of the east coast, British Columbia has a long history of HAB-related events. For example, historical PSP data from the west coast shows a 6-8 yr cycle, with shellfish toxicities worse in 1993 and 2001. *Alexandrium* spp. tends to be seeded in inlet systems where there is no evidence of eutrophication. Phytoplankton data, in varying formats, exists for monitoring stations in southern BC. One must also realize that organisms are unique and have different triggering mechanisms for bloom formation.

Although it is difficult to take information from one body of water and from a given species and transfer it to another region, it would be beneficial to attempt adapting or applying the east coast models for the west coast. Models tend to be location specific with the challenge of looking at coastal regions and trying to predict what is going to happen.

CANADIAN GEOHAB GOALS

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A goal of the international GEOHAB program is to develop international research on HABs. In the past, much work on HABs has been descriptive. Biological, physical and statistical models have improved in recent years and need to be linked to HABs.

Monitoring technology has been improving so that real-time measurements can be obtained; taxonomic and toxin-specific probes are being developed; rapid diagnostic methods are more promising; and optical sensing equipment is improving. A conserted effort is necessary to address the problem of prediction.

CANADIAN GEOHAB

Michel Gosselin

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The main species of concern in the Gulf of St. Lawrence are *Alexandrium tamarense* and *Pseudo-nitzschia seriata*. Program Elements 2 (Nutrients and Eutrophication) and 3 (Adaptive Strategies) of the GEOHAB Science Plan can be addressed through laboratory and mesocosm work in response to nutrient changes.

PAST WORK IN THE ST. LAWRENCE AND FUTURE INVOLVEMENT IN A CANADIAN GEOHAB PROGRAM

Suzanne Roy

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Research has focused on the resuspension of resting cysts, with particular emphasis in 1996 on the resuspension of 10 cm sediment in the Saguenay Fjord region. CAT Scan equipment has been used to detect cysts in sediments. Various strains of *Alexandrium* spp. from the St. Lawrence are being characterized. In addition, a remote sensing effort has been undertaken using SeaWiFS.

SHELLFISH TOXICITY AND DATABASES

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The Canadian Food Inspection Agency (CFIA) has long datasets of shellfish data and can provide a supporting role in a Canadian GEOHAB initiative.

GENERAL DISCUSSION

After the presentations on Day 1, the following common themes were suggested with the hopes of narrowing the topics to 3 on Day 2: (they are not listed in order of priority)

- viral and bacterial – loss mechanisms
- modelling
- molecular probes
- retrospective analyses
- adaptive strategies
- non-indigenous species
- predictive and loss strategies
- ecology
- comparative ecosystems
- international interactions
- monitoring using technology such as optical sensors

A general consensus was reached to include both field and laboratory studies, with a focus in the field on areas where natural blooms are commonly known to occur. Where models are being applied, retrospective analyses and data collection are important as well as determination of growth rates for organisms, grazing rates and observations during 'non-bloom' years.

Although all HABs are a health concern, a suggestion was made to study three organisms: *Alexandrium* spp., *Pseudo-nitzschia* spp. and *Heterosigma akashiwo*. As these species are found in other regions of the world, it would enable international collaborative and comparative studies. In addition, Canada has three types of ecosystems: coastal embayments, large-scale estuaries, and fjords. Although prediction is often listed as an end target, it was noted that this is often a very difficult task and researchers are often unable to deliver, so the group should be cautious with such statements. It might be more realistic to make a prediction and follow-up with the degree of deviation from the prediction.

Day 2:

OVERVIEW OF THE INTERNATIONAL GEOHAB PROGRAM AND LINKAGES WITH A CANADIAN PROGRAM

Allan Cembella (member, International GEOHAB Science Steering Committee) described the International GEOHAB Program. Its focus is to foster a coordinated international science plan on the ecology and oceanography of HABs in the context of large-scale comparative ecosystems involving multidisciplinary studies on processes and mechanisms. A role of the international scientific committee is to help countries link with other countries in collaborative research. Although they will not provide peer review of a proposal, they will establish whether a proposal submitted by a particular country meets the criteria for a GEOHAB proposal and, where applicable, will write a letter of endorsement. They are not able to provide financial assistance. In order for a program to be approved under the GEOHAB umbrella, it is essential to provide linkages to international research efforts.

Studies currently supported by GEOHAB include the Baltic program (with a focus on cyanobacteria and *Dinophysis*) and a China project. A draft application form for submission to the international steering committee was circulated.

POSSIBILITIES FOR FUNDING

Suzanne Roy contacted NSERC and highlighted the following funding grants available under NSERC:

- Research Network Grant (RNG)
- Strategic Project Grants (SPG)
- Networks of Centres of Excellence (NCE)
- Collaborative Research Opportunity Grants (CRO)
- International Opportunity Fund (IOF)

Discussion followed to determine the main objective and goals for a Canadian GEOHAB program. The objective is to coordinate and focus the efforts of the Canadian scientific research community towards research on HABs of relevance to Canada. The scientific goal is to improve prediction of HABs by determining the ecological and oceanographic mechanisms underlying their population dynamics, and by integrating biological, chemical, and physical studies supported by enhanced observation and modelling systems. The program will be based on determining the biologically mediated processes affecting bloom dynamics and will apply novel information systems to study HABs in Canadian waters. In order to meet the international GEOHAB criteria, the Canadian multidisciplinary approach will be coordinated with research from other countries.

Work will focus on the following algae: the dinoflagellate *Alexandrium* spp., the raphidophyte *Heterosigma akashiwo*, and the diatom *Pseudo-nitzschia* spp. These genera were selected for focused research based on the following criteria: their distribution in Canadian waters, available Canadian expertise on the particular species, and the fact that these particular species have been observed in many non-tropical regions of the world.

It was stressed that the sediment environment be included in studies and that models incorporate all life stages of organisms as well as information such as vertical migration, fate of temporary cysts, cyst germination, bloom initiation and dissipation, excystment (including percentage of excystment), and deep-water cysts. Regions of

Canada that do not have HABs need to be studied to determine why they do not occur in those particualar locations.

RESEARCH THEMES

Participants expressed interest in all five elements of the international GEOHAB Science Plan: 1) biodiversity and biogeography, 2) nutrients and eutrophication, 3) adaptive strategies, 4) comparative ecosystems and 5) observation, modelling and prediction. However, it was agreed that a Canadian GEOHAB program would focus on key HAB species (*Alexandrium*, *Heterosigma* and *Pseudo-nitzschia)* and the following three themes:

1. population dynamics of key HAB species in comparative ecosystems
2. biologically mediated growth and loss processes
3. novel observation systems and data analyses towards improved prediction

Theme 1. **Population dynamics of key HAB species in comparative ecosystems.**

Bloom dynamics of the three above genera will be compared using common methodologies and innovative approaches in several ecosystems - including coastal Atlantic embayments, the Bay of Fundy, Gulf of St. Lawrence, British Columbia coastal embayments and the Georgian Straight. Commonalities and differences between sites will be compared with the goal of determining factors associated with bloom occurrences. The large geographic extent and varying coastal environments where blooms of the same organism occur provide an ideal natural laboratory in Canada to examine bloom dynamics. Measurements will include detailed physical and chemical oceanographic descriptions, growth rates of HAB organisms, nutrient uptake kinetics, vertical migration, cyst-related processes and loss processes. Hypotheses will be tested using retrospective databases, analyzing new data, observations and data testing.

An example will be a comparison between the Bay of Fundy, Ship Harbour, Barclay Sound and the Gulf of St. Lawrence where blooms of *Alexandrium* and *Pseudo-nitzschia* occur. This multidisciplinary comparison has never been attempted in the past. Population dynamics, growth rates, and nutrients could be studied with the same approach to determine whether results are consistent. This will result in a common approach with common instrumentation and the ability to test hypotheses in all environments.

As species of *Alexandrium*, *Heterosigma* and *Pseudo-nitzschia* are common elsewhere in the world, information derived may be extended and compared to results from other regions of the world.

Theme 2. **Biologically mediated growth and loss processes.**

This theme targets the need for laboratory and field studies dealing with biologically mediated processes influencing population dynamics of HABs. Although knowledge in this field is deficit, Canada can capitalize on existing expertise. Biological processes controlling growth and mortality of the selected genera will include:

- grazing-related losses (from pelagic and benthic grazers)
- allelochemical interactions
- the influence of fungi, bacteria, and viral pathogens
- sedimentation and biodeposition, and sediment chemistry as it relates to vegetative cells and cysts

Theme 3. **<u>Novel observation systems and data analyses towards improved prediction</u>**

This theme involves using innovative and existing techniques and approaches to observe HAB organisms *in situ* and to detect their physiological and toxicological status in the field. Antibody and nucleotide probes are being developed with future use in semi-automated bulk or single sample analyzers such as flow cytometers (FlowCam). Optical measurements from moorings can provide continuous monitoring and detection of many HABs. In addition, bio-optical methods are being refined to extract information on species composition from remote sensors or long-term monitoring for ocean colour. These data will be analyzed using circulation and biological models. Predictive models will also be explored using novel statistical methods to analyze existing data and data assimilation methods incorporating near real-time observations into physical/biological models. Research activities will include:

- continued development of novel detection methods (including single cell methods or species identification; development of physiological probes)
- development of novel observation methods (for both HABs and associated communities)
- enhanced capability for long-term and synoptic observation
- novel data analyses using retrospective and historical databases
- development of data assimilation forecasting models for application in coastal observation systems to improve prediction

TITLE/ MISSION

The group chose "Population Dynamics of Canadian HABs (*Heterosigma, Alexandrium* and *Pseudo-nitzchia*)" as the title for its Canadian GEOHAB program. The mission of the Canadian program is to coordinate and focus the efforts of the Canadian scientific research community into harmful algae research of relevance to Canada.

As discussed earlier, the Canadian approach will be to feature three research themes focusing on three key organisms in comparative ecosystems using innovative approaches. It will be a coordinated, multidisciplinary and international approach. Although Canadian waters have a number of HABs, the group decided to focus on three genera (*Alexandrium*, *Heterosigma* and *Pseudo-nitzschia*) where it will be possible to build on existing knowledge and expertise within Canada. Canadian waters provide a unique opportunity to study HABs as most HAB species (except those restricted to the tropics) have been observed.

SUMMARY AND CLOSING

This Workshop was the first time that Canadian researchers attempted to form a coordinated national research team to work on HABs. It provided an excellent opportunity to discuss a bi-coastal research plan.

Future plans and action items for a Canadian GEOHAB program:

- complete a technical report summarizing the Montréal Workshop – Fisheries and Oceans Canada Phycotoxin Working Group (PWG)
- write letter to the GEOHAB steering committee
- establish a writing committee (S. Roy, J. Martin, J. Cullen, M. Levasseur, C. Suttle) to write a document for distribution to interested parties, funding agencies, and national and international colleagues (with the help of S. Bates and M. Taylor)
- write a letter of intent (LOI) for the NSERC International Opportunity Fund (IOF) (S. Roy)
- host a meeting in Montréal in June 2002, led by university personnel, to develop an NSERC Research Network (RN) proposal. DFO personnel will also develop DFO proposals for submission to the Strategic Science Fund (SSF) and Environmental Science Strategic Research Fund (ESSRF)

Those interested individuals who were not able to attend the Workshop, but who are willing to share their expertise for the program will be kept informed.

Currently, other countries are increasing their HAB research efforts. Canada will likewise hopefully increase its funding for HAB research in the next few years. This workshop demonstrated the enthusiasm and willingness of Canadian researchers to collaborate amongst themselves and with researchers around the world to better understand HABs. Enough interest was shown to warrant continuing the development of a Canadian GEOHAB program.

ACKNOWLEDGEMENTS

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Appendix 1

Canadian GEOHAB
Hôtel de l'Institut
Montréal, QC

Meeting Agenda

Friday – October 19, 2001 (Léonard-Gagnon Room)

8:30 Welcome, Introduction (Bjorn Sundby)
8:40 International GEOHAB Science Overview (Allan Cembella)
9:15 Discussion
9:50 Overview (SOLAS) Maurice Levasseur
10:00 Health Break
10:20 Informal 5 min **(or less)** presentations by participants indicating their background and interest in Canadian GEOHAB program
12:00 "Working Lunch" provided in the Workshop room

13:00 Continue 5 min presentations
14:30 Select topics for break-out session
15:00 Health Break
15:20 Break-out into discussion groups by topics selected in plenary
17:00 Meet in plenary to summarize discussions/themes

Saturday – October 20, 2001 (Paul Émile-Lévesque Room)

8:30 Discussion of potential international links (Allan Cembella)
9:00 Funding Agencies (Suzanne Roy)
10:00 Health Break
10:20 Continuation of sub-groups; Research discussion
12:00 "Working Lunch" provided in the Workshop room

13:00 "Pulling everything together"; discussion
15:00 Health Break
15:20 Election of "writing committee"
16:30 Preliminary drafting of Letters of Intent (LOIs)
17:00 Closing remarks

Included in discussions: future workshops, Canadian GEOHAB committee, etc.

Appendix 2

CANADIAN GEOHAB WORKSHOP
Montréal, Québec
19-20 October, 2001

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