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PACIFIC COAST RESEARCH ON TOXIC MARINE ALGAE

J.R. Forbes (ed.)

Institute of Ocean Sciences
Department of Fisheries and Oceans
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Proceedings of a workshop held at the Institute of Ocean Sciences, Sidney, B.C., 30 April 1991.

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ABSTRACT

Forbes, J.R. (ed.). 1991. Pacific coast research on toxic marine algae. Can. Tech. Rep. Hydrogr. Ocean Sci. 135: 76 p.

This report comprises the proceedings of the workshop on Pacific coast research on toxic marine algae, which was held at the Institute of Ocean Sciences in Sidney, British Columbia, on 30 April 1991. Representatives from the federal and provincial governments, universities and industry participated in presentations of current research and management activities on the west coast of Canada. Research results on the effects of toxic phytoplankton on farmed fish, physical factors affecting the development of blooms, remote sensing techniques, and analytical procedures were presented. Summaries of inspection and monitoring activities on the west and east coasts of Canada, as well as research activities in Washington State and Atlantic Canada provided a basis for comparison in discussion sessions.

Key words: phytoplankton, toxins, phycotoxins, red tide, aquaculture, blooms, remote sensing, management, monitoring

RESUME

Forbes, J.R. (ed.). 1991. Pacific coast research on toxic marine algae. Can. Tech. Rep. Hydrogr. Ocean Sci. 135: 76 p.

Ce rapport comprend les actes de latelier sur les recherches effectuées sur la côte du Pacifique portant sur les algues marines toxiques, qui a eu lieu à l'Institut des sciences océaniques à Sidney, Colombie-Britannique, le 30 avril 1991. Les représentants des gouvernments fédéraux et provinciaux, des universités et de l'industrie ont participé à des présentations portant sur la recherche actuelle et les activités de gestion sur la côte ouest du Canada. On a présenté les resultats de recherches portant sur les effets d'espèces toxiques de phytoplancton sur les poissons d'aquiculture, sur les facteurs physiques influant sur le développement de prolifération, sur de techniques de télédétection, ainsi que sur les méthodes d'analyse. Des sommaires des activités d'inspection et de surveillance sur les côtes est et ouest du Canada, ainsi que sur les activités de recherche dans l'Etat de Washington et dans les provinces maritimes, permettaient de faire des comparaisons lors des débats.

Mots clés: phytoplancton, toxines, phycotoxines, marée rouge, aquiculture, prolifération, télédétection, gestion, surveillance

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I am pleased to thank the following people for their excellent assistance, contributing to the success of the workshop. Gwen Groberman, Linda White and Art Walters provided help with the organization and conduct of the workshop. Ian Whyte and Sharon Thomson carefully reviewed the manuscript of the report. Terry Tebb volunteered to chair the discussion session at the end of the meeting, and the members of the Phycotoxins Working Group provided useful advice.

INTRODUCTION

Blooms of toxic marine algae have resulted in substantial losses in the developing salmon aquaculture industry in B.C. over the last several years (Black 1991). They also have been responsible for significant restrictions in areas available for recreational and commercial harvest and growing of shellfish (Chiang 1991), as protection from the clear risk as a human health hazard.

A workshop on research on toxic marine algae on the west coast of Canada was held at the Institute of Ocean Sciences in Sidney, B.C., on 30 April 1991. Sixty participants from federal and provincial agencies, universities and industry attended. The impetus for the workshop came from a variety of sources, including regional Department of Fisheries and Oceans (DFO) management, the national DFO Phycotoxins Working Group, and provincial and industry sources who were interested in sharpening the focus of DFO in this field.

The major objectives were to improve cooperation between the various agencies and individuals engaged in research in this field, as well as provide a framework to assess the types of projects that the federal government should take on to augment research and management activities by others in the region. As such, the workshop built on and had a rather different focus from a workshop on the impact on fisheries and aquaculture of exceptional algal blooms, held at the Bamfield Marine Station in Bamfield, B.C., in 1988 (Clayton 1988). This workshop was also intended to provide some balance to two recent workshops (Bates and Worms 1989, Gordon 1991), intended to be national in scope, but which have had virtually no input from the west coast.

During morning sessions, results of various research projects on distribution of toxic species, modes of toxicity, remote sensing applications and analytical procedures were presented. In the afternoon, requirements of the aquaculture and wild harvest industries were discussed. A review of Department of Fisheries and Oceans research activities on the east coast was given, and presentations were made on inspection programs in the Pacific and Atlantic regions. Information on research and monitoring activities related to salmon aquaculture in Washington State was also presented. The workshop concluded with a discussion session at which a considerable number of areas in which our knowledge is limited were identified.

Participants who presented oral or poster papers were asked to submit a summary of their presentation for inclusion in these proceedings. These submissions comprise the bulk of the report. There was substantial variation in the length and organization of the summaries, but it was decided to include them essentially as submitted, with only minor stylistic editing. Taxonomic nomenclature has been retained in the form submitted by the individual authors. It should be noted that discrepancies do occur with some taxa. John Spence, Director, B.C. Aquaculture Research and Development Council, was not able to attend, but did provide material on concerns of the aquaculture industry regarding toxic algae. This material is included in the report, together with the conclusions and recommendations of a joint B.C.A.R.D.C. and B.C. Oyster Board review of requirements regarding algal toxins to meet product quality control and purity objectives. Material presented by Larry Albright on the distribution of harmful *Chaetoceros* spp. and their lethal mode of action is being prepared for publication elsewhere and does not appear here. The report concludes with a review of the discussion session and some comments on areas that deserve particular attention.

The results of the workshop give an indication of the present level of effort and understanding of the source, nature and consequences of toxic algal blooms on the west coast of Canada.

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THE EFFECTS OF *HETEROSIGMA AKASHIWO* ON JUVENILE CHINOOK SALMON

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The increasing incidence and extent of toxic algal blooms throughout the world suggests that algal prey organisms, ensuing food chain organisms, and cage-reared fish are becoming more vulnerable to poisons derived from algae (White 1982, Anderson *et al.* 1985, White and White 1985, Hallegraeff *et al.* 1988). In temperate coastal waters members of the *Alexandrium* genus (formerly *Gonyaulax* and later *Protogonyaulax*) have been implicated in juvenile and adult fish kills (White 1981a, 1982, Mortensen 1985, Potts and Edwards 1987, White *et al.* 1989).

The neurotoxins in the gonyaulacoid genus *Alexandrium* are a family of tetrapurine derivatives, commonly called Paralytic Shellfish Poisons (PSP), which can accumulate in invertebrates, such as molluscs (Shumway and Cucci 1987) and planktonic crustaceans (White 1981b, Ives 1985, Watras *et al.* 1985), but without lethal effects. However, mortality of finfish larvae from PSP is known to occur either by direct ingestion of the toxic algae or by ingestion of toxic prey zooplankton (White *et al.* 1989). Concern is now being expressed that increased frequency of PSP blooms will seriously affect the year class recruitment of certain fish stocks (Robineau *et al.* 1991).

The Chloromonad *Heterosigma akashiwo* (Hada) Hada, is gaining increasing notoriety as a fish killer worldwide (Gaines and Taylor 1986, Gowen 1987, Fraga 1988, Murphy 1988, Boustead *et al.* 1989, Stockner 1989). Major fish kills in the salmon farming industry in the northeast Pacific since 1986 have accounted for losses in excess of \$15 million (Rensel *et al.* 1989, Black 1990). As the cause of fish mortality in *H. akashiwo* blooms is unknown, any information on the mechanisms causing death would expedite mitigation of losses.

Possible mechanisms causing death of fish from algae are, 1) physical damage to gills from spines of algae such as *Chaetoceros* spp. (Bell 1961, Gaines and Taylor 1986), 2) mechanical obstruction of gills from algal mucus causing asphyxiation (Anon. 1988), 3) asphyxiation caused by oxygen demand of senescent algal cells (Evelyn 1972, Okaichi 1983, Holmes and Lam 1985), 4) gas-bubble trauma caused by extreme oxygen saturation of seawater resulting from algal photosynthesis (Renfro 1963), and 5) ichthyotoxins such as the brevetoxin polyketides produced by *Gymnodinium breve*, or the hemolysins produced by *Gyrodinium aureolum* and *Chrysochromulina polylepis* (Yasumoto 1990), or the tetrapurine neurotoxins from species of *Alexandrium* mentioned previously.

In the summer of 1990, *H. akashiwo* blooms in excess of several km^2 at San Mateo bay on the west coast of Vancouver Island, and at Blind Bay in the Strait of Georgia provided an opportunity to perform bioassays and histopathological examination on freshwater and saltwater acclimated juvenile Chinook salmon exposed to these blooms (Black *et al.* 1991). No notable concentrations of other known or suspected ichthyotoxic algae were evident in the algal blooms at either location.

At San Mateo Bay in Barkley Sound the bloom was 95% *H. akashiwo* at 200000 ± 8000 cells.mL^{-1}, in surface water of 16° C and 24 salinity. In the bioassay juvenile Chinook salmon were placed in seawater from the Pacific Biological Station (PBS), as a control, placed in the bloom and in the bloom sparged with oxygen. Freshwater Chinook exposed to the bloom and to the oxygenated bloom exhibited mean times to death, which were not significantly different, at 118 ± 30 and 130 ± 34 min, respectively. Similarly, saltwater acclimated Chinook showed a mean time to death of 81 ± 45 and 110 ± 40 min in the bloom and in the oxygenated bloom, respectively. No mortalities occurred in the control seawater, which showed oxygen saturation levels about 50% higher than the corresponding levels in the bloom and oxygenated bloom seawater.

At Blind Bay the bloom was in excess of 95% *H. akashiwo* at 793000 ± 257000 cells.mL^{-1} in the surface water at 24° C and 24 salinity, and a Secchi disc was invisible 25cm under the surface. Freshwater Chinook exposed to the bloom and to the oxygenated bloom exhibited mean times to death of 35 ± 12 and 30 ± 11 min, respectively, which were not significantly different. No mortalities occurred in the control seawater, which contained a 40% higher oxygen saturation level than the corresponding levels in the bloom and oxygenated bloom water.

The rates of fish mortalities in the blooms were independent of saltwater acclimation. Survival times of fish in the bloom at Blind Bay were reduced by two thirds, relative to survival times at San Mateo Bay, most probably from the increased cell concentration, although increased susceptibility from temperature stress could not be dismissed.

Increased oxygen demand associated with osmoregulation of freshwater fish when placed in seawater appeared to have no impact on survival of the fish exposed to algal blooms. Mortalities were not associated with low ambient oxygen levels as oxygen concentrations were well above saturation concentrations. Oxygen supersaturation resulting from algal blooms has been implicated in gas-bubble trauma leading to mortalities of fish in Galveston Bay (Renso 1963). However, the higher oxygen concentrations were not a factor in these fish mortalities.

Both freshwater and saltwater acclimated fish when exposed to the bloom behaved as if anesthetized. During the bioassay moribund fish were placed immediately into Davidson's solution as were the pithed control fish at the end of the bioassay. Histological comparison of the moribund fish with the control fish indicated no pathological change to gills and only a normal quantity of mucus. The latter observation would support the observed lack of coughing response in the test fish, although this behaviour has been reported as a function of exposure to *H. akashiwo* (Campbell 1988). The considerably higher density of algal cells in the bloom relative to the few cells observed in the fish gills further corroborated the suggestion that asphyxiation from clogged or damaged gills was not the cause of death.

Histological damage to fish gills from algae such as *Gyrodinium aureolum* and *Gonyaulax excavata* has included necrosis and sloughing of the epithelia of secondary lamellae, accompanied by pyknosis of the primary lamellar epithelium, and congestion of branchial vessels (Jones *et al.* 1982, Roberts *et al.* 1983, Mortensen 1985). Also, the disappearance of the mucus coat on gills of fish exposed to *Chattonella antiqua* and *Gymnodinium* sp. has been considered to lead to locally impaired osmoregulation (Shimada *et al.* 1982, 1983). These features were not evident in any of the fish gills examined from San Mateo Bay or Blind Bay. The expansion of the intercellular spaces in the primary and secondary lamellae of the gill has also been associated with algal exposure (Shimada *et al.* 1983); however, this characteristic can also result from delayed fixation (Speare and Ferguson 1989). The absence of any abnormalities in the gills suggested that physical damage to the gills was not the cause of the observed mortalities.

Histological examination by light microscopy showed no pathological abnormalities of gill, kidney, liver, pancreas, spleen or intestine in any of the 97 fish examined from San Mateo Bay or in any of the 41 fish examined from Blind Bay. Identical histological features were observed for internal organs of fish exposed to 'Flagellate X', an alga considered to be responsible for fish deaths in Scotland, although numerous foci of haemorrhage were apparent in atrical heart tissue, indicative of cardiac arrest (Gowen *et al.* 1982). Heart tissue was not examined in detail in the present bioassay because this organ was so small in the juvenile fish.

Juvenile Chinook in the *H. akashiwo* blooms did not die from low ambient oxygen levels, oxygen supersaturation, asphyxiation from clogged gills, the disappearance of normal gill mucus or excess mucus formation in the gill, physical gill damage, or damage to other internal organs. Data are consistent with death from an ichthyotoxic agent. The physiological effects suggested rapid assimilation of a toxic agent to produce neurological damage to the fish, or rapid loss of biomembrane integrity in the gills which would not have been evident at the light microscope level of examination.

Preliminary growth studies on *H. akashiwo* have demonstrated a doubling in the cell density using continuous light relative to a light:dark (2:1) cycle. Stationary phase *H. akashiwo* cultured under continuous light could attain densities of 1.2 - 1.3 x 10^6 cells.mL^{-1}. When juvenile Chinook were treated with cultured *H. akashiwo*, time to death was extended over ten times that observed from natural blooms mentioned previously, indicating only minor production of toxic metabolites. Nevertheless, variations in the levels of toxins in algae are known to be dependent on environmental conditions of growth (Roszell *et al.* 1990).

Marine teleosts obtain much of their freshwater requirement for osmoregulation by intestinal absorption of ingested seawater (Shehadeh and Gordon 1969). Toxins or inorganic elements ingested with this water would also be absorbed quickly. Elements in the cultured *H. akashiwo* were compared with specimens from San Mateo Bay and Blind Bay to assess possible metal toxicity. In all algal samples major elements (with concentrations in excess of 1mg.g^{-1} dry weight) were potassium, magnesium, phosphorus, calcium and bromine. Minor elements (from 10μg.g^{-1} to 1mg.g^{-1} dry weight) were iron, zinc, titanium, aluminum, silicon, boron, iodine, manganese and strontium. Trace elements (at levels below 10μg.g^{-1}) were lithium, copper, chromium, selenium, arsenic, cadmium, tin, molybdenum, lead, mercury, nickel, vanadium, cobalt and scandium. No toxic levels of elements were detected in *H. akashiwo*, whether cultured or from wild blooms, as all levels were within the range of concentrations determined recently in the spot prawn *Pandalus platyceros* (Whyte and Boutillier 1991).

A digalactosyl glycerolipid, considered a hemolysin, was the principal toxic component of *Chrysochromulina polylepis*, which caused extensive fish kills in Scandinavian waters (Skjoldal and Dundas 1991). An unusual fatty acid profile, with 17% 14:0, 62% 16:0, and 21% 16:1n7t acids was reported for *H. akashiwo* by Fernandez-Reirez *et al.* (1989) and prompted closer examination of the lipid fraction of this alga. The total fatty acid profile of *H. akashiwo* cultured at PBS exhibited 5.1% 14:0, 12.4% 16:0, 6.2% 16:1n7, 8.6% 18:3n3, 23.5% 18:4n3, 17.2% 20:5n3 and 2.8% 22:6n3 acids respectively, which agreed favourably with former values reported by Nichols *et al.* (1987) for *Chatonella antiqua* and *H. akashiwo*. The possibility that specific fatty acids are somehow involved in toxicity is considered remote, however, the nature of the toxic agent remains unknown.

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HARMFUL EFFECTS OF THE DIATOM *CHAETOCEROS CONCAVICORNIS* ON ATLANTIC SALMON

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Blooms of harmful phytoplankton in Puget Sound, Washington have proved to be a major impediment for the marine culture of salmonids by private fish growers, resource management agencies and tribes. The diatoms *Chaetoceros concavicornis* and *C. convolutus* have been a problem since the beginning of net-pen salmon culture efforts in the early 1970's. I conducted laboratory studies on the effects of *Chaetoceros concavicornis* on seawater-acclimated Atlantic salmon (*Salmo salar*) averaging about 250g in weight. Arterial blood-gas, histopathology, and behavioral measures were collected during 6 to 96 hour static bioassays. Although exposure bioassays have been performed by others using *Chaetoceros* spp. collected in the field or grown in laboratory cultures, no blood-gas monitoring results have previously been reported.

Atlantic salmon responded to the addition of 10^5, $2x10^5$ and $4x10^5$ cells.L^{-1} of *C. concavicornis* by displaying a cough reaction of regular, but slowly diminishing frequency, suggesting a sort of acclimation. The 10^5 concentration reflects the maximum concentration of *C. concavicornis* documented in Puget Sound to date, most other records are nearer 10^4 cells.L^{-1}. At the lower, more commonly occurring concentration of 10^4 cells.L^{-1}, no change in initial cough frequency compared to control fish was noted. Fish coughing, and in some cases acclimation, is common by salmonids exposed to small doses of many environmental irritants or chemicals.

Mucus production on the gills appears to increase from exposure to the *C. concavicornis*, macroscopically showing a mixture of mucus, diatom cells and setae; no bleeding was grossly evident. Histopathology investigations have not been completed. Blood-gas monitoring with microelectrodes immediately upon death of the salmon or at termination of an exposure bioassay has demonstrated a significant reduction of blood-oxygen partial pressure for all concentrations of the diatom used; carbon dioxide partial pressure was significantly greater for blood from all fish except those exposed to 10^4 concentrations. The low oxygen partial pressure of blood samples collected from fish immediately upon death (<20mm Hg from dorsal aorta) supports the argument that *C. concavicornis* interferes with normal gas exchange mechanisms of the fish.

Interpretation of these preliminary results suggests that oxygenation (not aeration) within net-pens surrounded by perimeter skirts may be an effective method to offset the negative effects of *C. concavicornis* on salmon. Due to the wide depth distribution of *C. concavicornis* and the impracticality of conducting visual surveys by boat or airplane, methods such as airlift pumping or towing net-pens that have been useful during blooms of *Heterosigma akashiwo* may be less effective.

THE AUGUST/SEPTEMBER 1990 RED TIDE EVENT ON THE WEST COAST OF CANADA: SATELLITE AND AIRCRAFT OBSERVATIONS

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Introduction

In early September 1990 a denser and more extensive red tide than any observed previously in the area, was reported off the west coast of Vancouver Island. The bloom was primarily due to the dinoflagellate *Gonyaulax spinifera*, which, although non-toxic, caused substantial shellfish mortality and was of great concern to local aquaculture operations. Record high surface water temperatures were observed at this time, and probably contributed to the event. The effects of increasing population in the watershed of the Strait of Georgia and Puget Sound may also have contributed.

In the few surface observations that are available, surface cell concentrations as high as 10^7 cells.L^{-1} were encountered, equivalent to chlorophyll concentrations of several hundred mg.m^{-3}. It was evident from visual observations that the colour changes in surface waters were very strong, and should be detectable by available satellite sensors, even though the only sensor designed for water colour observations (the NASA CZCS) is no longer operating. Both the AVHRR and the TM can be used for studies of near-shore blooms. In the present case the AVHRR has the advantage of providing daily repeats capable of following the expected evolution of such a bloom with spatial resolution adequate for the large areas of discoloured water observed offshore.

An airborne imager suitable for mapping water colour changes is available on the west coast of Canada, operated commercially by Borstad Associates Ltd. This is a Compact Airborne Spectral Imager (CASI), (Babey and Anger, 1989). Although spatial coverage from such an airborne instrument is very limited, it can also be deployed under cloud which would block satellite observations, and has sufficient spatial resolution to monitor blooms in small coastal bays and inlets. Images show typical patterns formed near the coast and the detailed spectra are useful for interpreting the satellite images.

AVHRR imagery

The AVHRR is flown on each of the US NOAA weather satellites (NOAA 9, 10, 11...) and is the major source of satellite thermal and visible data used in oceanography. AVHRR imagery has neither the high sensitivity nor the special spectral bands needed for mapping the subtle water colour changes associated with normal concentrations of phytoplankton. Also its spatial resolution (1.2 km on the satellite track, increasing to 2.5 by 7km at the edges of the swath) is not high enough to examine the many red tide events which form in local areas of sheltered inlets.

In the present case, however, the bloom, at least in its later stages, covered a very large area and the wide coverage and daily repeat of the AVHRR could be used to advantage. Higher resolution would have been valuable, especially for mapping the early stages of the bloom and the filaments in late August. TM imagery will therefore also be acquired for this study.

The spectral bands of the AVHRR imager are listed in Table 1. The instrument lacks the sensitivity and the short-wave bands of the CZCS, which was designed specifically for mapping of near surface biomass, but the band 1 radiance will be increased by the presence of seston in near-surface water, whereas the corresponding increase in band 2 should be much less, due to the higher absorption by water at these longer wavelengths. Over land, the high near-infrared reflectance of chlorophyll pigments gives higher signals in band 2 than in band 1. The remaining three bands provide thermal data.

TABLE 1
The 5 spectral bands of the AVHRR imager

1	580 - 680	nm	(visible)
2	725 - 1100	nm	(near infrared),
3	3000 - 3500	nm	(short thermal infrared),
4	10500 - 11300	nm	(thermal infrared, split window)
5	11500 - 12500	nm	(thermal infrared, split window)

Images were acquired in digital form from the western Canadian receiving stations at the University of British Columbia (UBC) in Vancouver, and the Atmospheric Environment Service (AES) of the Federal Government in Edmonton, for the period July, August and September. The AES facility records data from whole passes of the satellite every day for weather forecasting purposes, but re-uses the tapes after 10 days. UBC stores limited segments for the Canadian west coast in a long-term archive, but operates less regularly.

Images used are listed in Table 2. These were rectified to a common geographic grid centered at 49° N, 126° W with a scale of 1000m/pixel using software written at IOS to run under the commercial EASI/PACE package on a Sun 3/260 system. The rectification assumes a known altitude, orbital inclination and scanner geometry, and makes use of one or more ground control points. Results are accurate to about one pixel (rms) when compared to the World Data Base II coastline.

Observations of the bloom with AVHRR imagery

Table 2 lists areas off the west coast of Vancouver Island in which the satellite images detected measurable colour change (radiance increase in band 1 or band 2) compared to that expected for clear water. Thick clouds block the measurements, but on the days listed, large clear areas covering from 25,000 up to 95,000km^2 were present. The band 1 (red) images should show the greater signal increase if reddened water is present. At the wavelengths of band 2 (near infrared) water is much more strongly absorbing, and the expected signal increase is lower by about the ratio of the mean water absorption coefficient in the two bands (about a factor of 15). For vegetation on land, where the spectral properties of chlorophyll reflectance are more important than those of water absorption, the situation is reversed, and the radiance increase in band 2 would be higher.

Clouds are a major problem in interpreting satellite images. Thin clouds also cause a radiance increase over water, but the increase has a different spectral form from that for suspended matter in water. For the two AVHRR bands, thin cloud gives roughly the same signal increase in bands 1 and 2. If a difference image is formed by subtracting band 2 from band 1, then expected radiance increases due to seston in water will remain, but the effect of clouds will be greatly reduced.

Table 2 lists the areas in km^2 for which the signal increase in the difference images exceeded 3, 5 and 7 digital steps on each of the 10 days for which relatively cloud-free AVHRR data was available.

The strongest signal increase due to the bloom in these difference images was observed on September 3, and corresponded to about 10 digital steps in the 10-bit AVHRR data. However, patches of discoloured water were observed in late August having higher radiance increases in band 2 than in band 1. This is an extremely unusual observation, and implies very high, near-surface cell densities sufficient to completely mask the absorption effects of water. It will also cause the image differences calculated in Table 2 to go negative in some areas, causing errors in the area estimates. Table 2 also lists these areas of enhanced radiance in band 2.

TABLE 2
The areas in km^2 of enhanced signal observed on AVHRR band 1 - 2 difference images, and band 2 images. Dates and sources of images used are shown. All were imaged by the satellite during ascending passes shortly after local noon.

Date 1990		**Source**	**Area clear (1000 km^2)** >3	**Enhanced signal band 1 - band 2** >5	>7	>3	**band 2 image** >9
Jul 27	UBC	65	50	0	0	0	0
Aug 24	UBC	25	250	0	0	75	0
Aug 25	UBC	25	80	0	0	150	5
Aug 26	UBC	65	450	0	0	260	8
Aug 27	UBC	50	1500	15	0	250	0
Sep 3	AES	85	1200	600	100	0	0
Sep 4	AES	95	1600	500	100	20	0
Sep 13	AES	95	300	0	0	5	0
Sep 20	AES	55	200	0	0	0	0
Sep 22	AES	50	100	0	0	0	0

The area of slight but measurable radiance increase apparent at the end of July appears due to the normal growth pattern of phytoplankton, as imaged previously with the CZCS in this area (Pan *et al.*, 1988). A long period of cloud prevented collection of satellite imagery in early August. Occurrences of the bloom were reported among the U.S. Gulf Islands in this time period. The available images give no indications of bloom in these areas, but the limited spatial resolution of the AVHRR would make it hard to observe this.

The first indications of the bloom in satellite images appear in the images collected in late August (Fig. 1). These show dense patches and filaments forming in an area about 100km across, centered about 100km off the central coast of Vancouver Island. The four day sequence shows that filaments within 20km of the shore were advecting to the north-west in the Vancouver Island coastal current. Further off shore the motion was mostly southwards.

For September 3 1990, the band 1 image shows a significant radiance increase in a patch measuring about 25km by 80km (long axis along the coast) centered about 50km off the mouth of Juan de Fuca Strait (Fig. 2a). Ship observations showed 10^4 to 10^5 cells.L^{-1} in this area. The corresponding band 2 image (Fig. 2b) showed only very small radiance increases, as expected for suspended material in water. Since cloud gives roughly the same signal in bands 1 and 2, their effect will be greatly reduced in a difference image (Fig. 2c) formed by subtracting band 2 from band 1. This image clearly shows the extent of the bloom at this time. The only other areas of water showing significant signal increases in Figure 2c are the Fraser River plume and coastal inlets containing glacial meltwater or other suspended sediment.

Figure 2d shows the surface temperature pattern on September 3, as imaged by the thermal band 4 of the AVHRR. The bloom was located in water of a temperature intermediate between the cold, mixed water of the Juan de Fuca Strait (10-12° C) and the warmer water off-shore (16-18° C). The thermal patterns for September 3 and 4 suggest that surface water associated with the bloom was being collected in the centre of a counter-clockwise eddy.

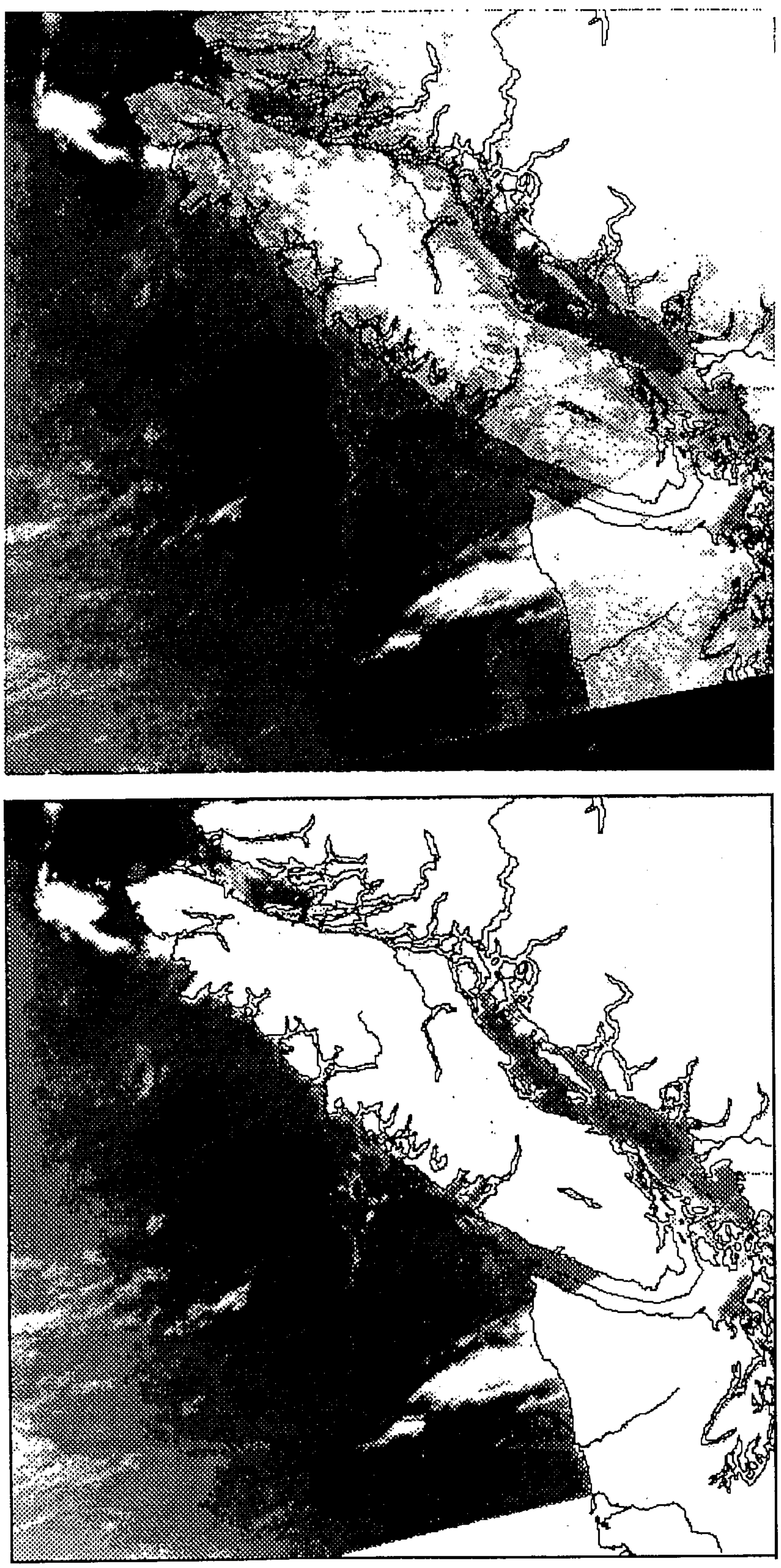


Fig. 1

Geometrically corrected AVHRR images (band 1 (top) and band 2 (bottom)) of the coastal waters off Vancouver Island showing radiance increases due to patches of an intense plankton bloom on 26 August 1990.

Satellite images for September 3 and 4 show significant fading of the bloom band 1 increase between the two days. By September 13 the bloom had faded to levels that were only just detectable.

The ratio of signals in bands 1 and 2 is illustrated in Figure 3. The figure compares cuts through images in these bands for August 26 and September 3. On September 3 the band 2 radiance increase in the extended patch was very small, being about 0.07 of the increase in band 1, as expected for suspended material in water. On August 26, however, the band 2 radiance increased in the filaments by as much as 5.8 times the increase in band 1. This is comparable to ratios for green vegetation on land. The band 2 radiance increase for the patches shown in Figure 3 is only about 25% that for land, implying that about this fraction of a 1.2 X 1.2km field of view is covered by high densities of algae.

These high cell densities would be expected to increase the sea surface temperatures by causing solar irradiance to be absorbed in a shallower surface layer than would be the case for clear water. The thermal images show that this is occurring. On August 26, for the peak ratio of 5.8, the thermal band 4 image shows a corresponding temperature increase of 4.6° C, sufficient to raise significantly the plankton growth rate.

The numbers in Table 2 give some indication of the time scale for the bloom. During the sequence of four images in late August the area covered by the dense filaments increased rapidly, as indicated by the band 2 radiances. Areas showing a significant increase in this band increased in the ratio 1:2:3.5:3.5 over the four days. If the value of the increase is proportional to suspended biomass in the surface layer, then the surface biomass increased in the ratio 1:1:4.5:7 over the four days. The satellite observations therefore suggest a growth of biomass comparable to the maximum expected rate of a daily doubling time. It should be noted, however, that flotation into the surface layer will also cause such an apparent biomass increase for satellite observations.

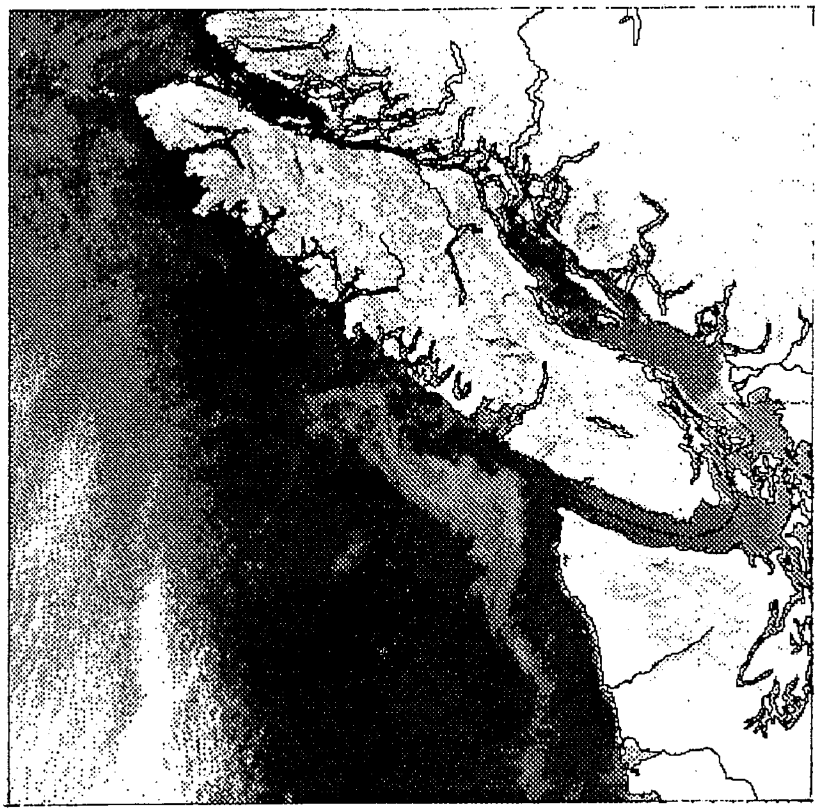
The band 2 radiance increase in the extended patch observed in early September is very small, but suggests some reduction in the total amount of biomass from that observed at the end of the August sequence. Between September 3 and 4 the amount remained roughly the same. By September 13 it had diminished by at least a factor of 10.

Airborne imaging spectrometer observations

Airborne observations using the CASI imaging spectrometer were made for smaller areas near shore. Figure 4 shows spectra measured with the CASI in different concentrations of the red tide. The strong peak centered at 705 - 710nm appears associated with the chlorophyll reflectance variation which gives the "red edge" in vegetation spectra on land.

Spectral coverage of AVHRR bands 1 and 2 are plotted on the Figure. It seems unfortunate that the small gap between bands 1 and 2 should coincide almost exactly with the strong spectral peak. Band 2 to band 1 ratios for the 3 spectra can be calculated (for the spectral range covered by the CASI) as 0.15, 0.48 and 1.08 respectively. Spectra for the extended patch on September 3 for which the ratio was <0.07 would therefore be expected to lie below those shown in Figure 4. Spectra for the filaments observed on August 26 for which the ratios were in the range 3 - 5.8 would show considerably higher radiances in the near infrared, approximating those for leaf spectra on land, and implying extremely dense surface mats of red tide algae.

Figure 5 shows a single band (701 - 714nm) image of the red tide near islands in Barkley Sound on September 4 1990. The area shown is about 1.2 by 3km and is imaged with a resolution of about 3m. Concentration of the bloom into filaments as seen further offshore in AVHRR imagery is evident.



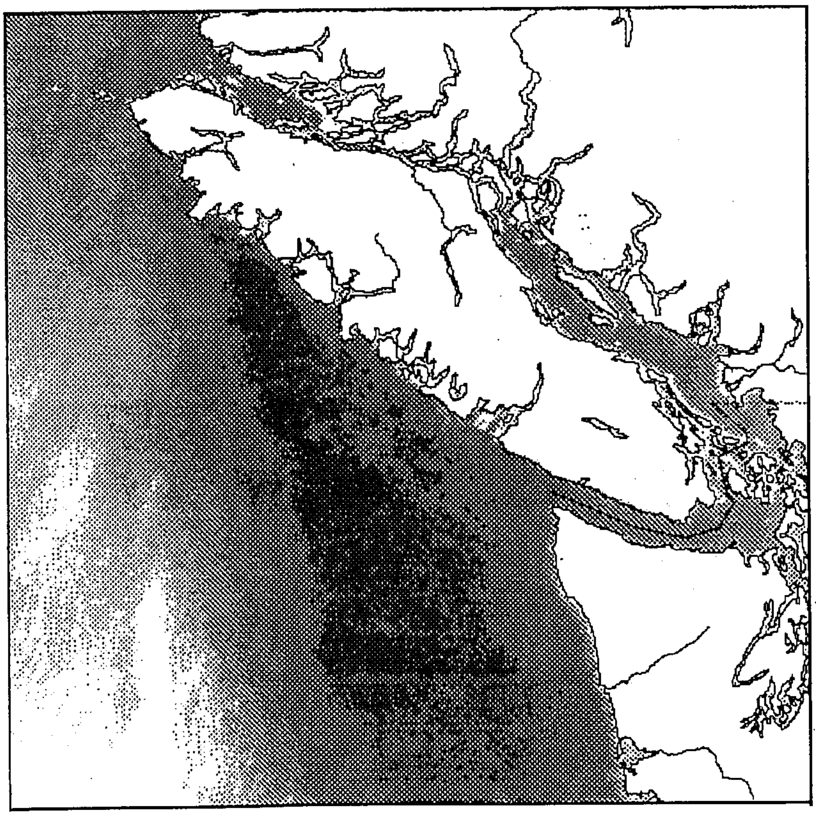


Fig. 2 a. and b.

Geometrically corrected AVHRR images of the coastal waters off Vancouver Island showing radiance increases due to the bloom on 3 September 1990, eight days after the images shown in Fig. 1 (band 1 (a) top, band 2 (b) bottom).

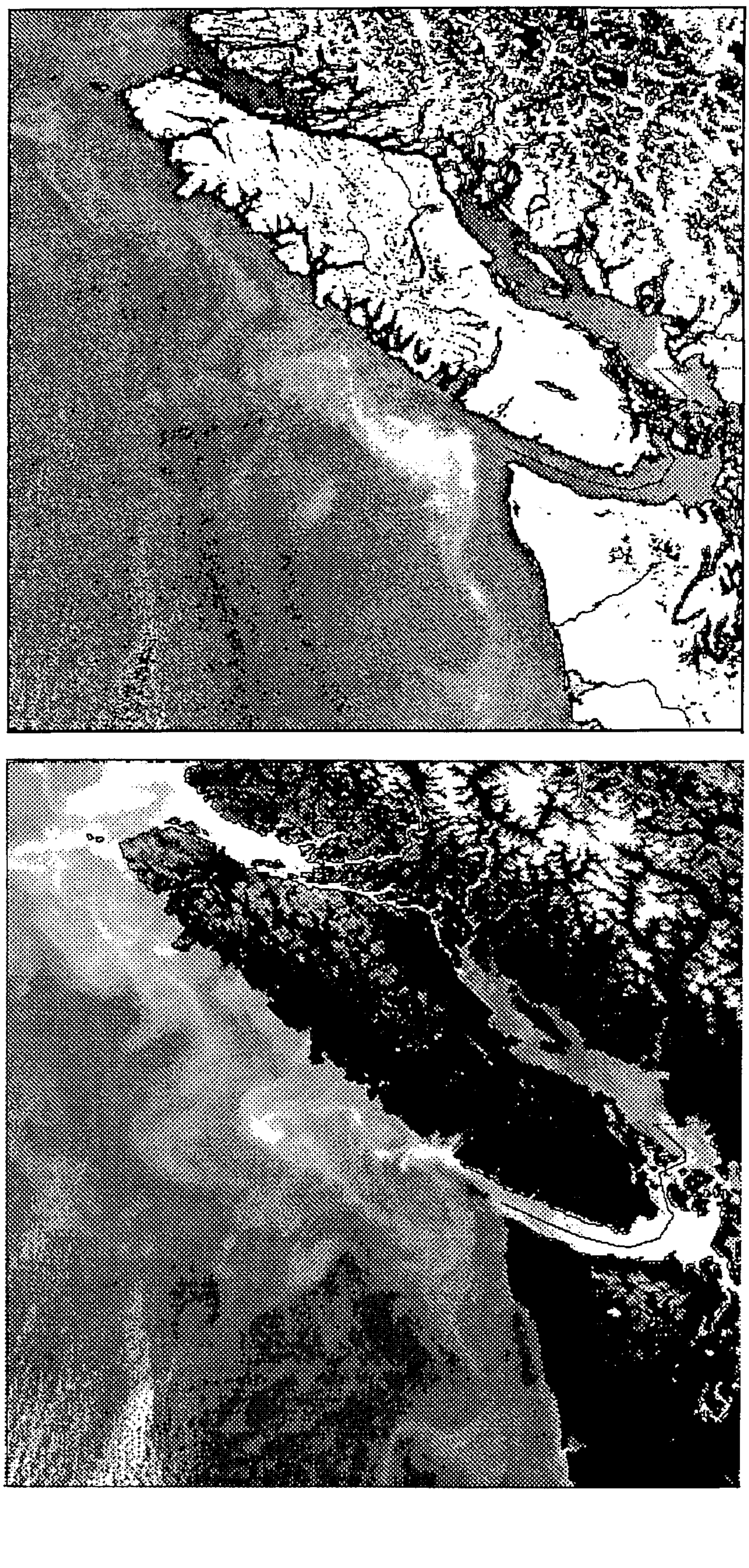


Fig. 2 c. and d.

As for (a) and (b), but (c) top: difference image (band 1 - band 2) suppressing radiance due to thin cloud, and (d), bottom: thermal image (band 4) showing the associated temperature patterns. Lower temperatures are shown as lighter shades.

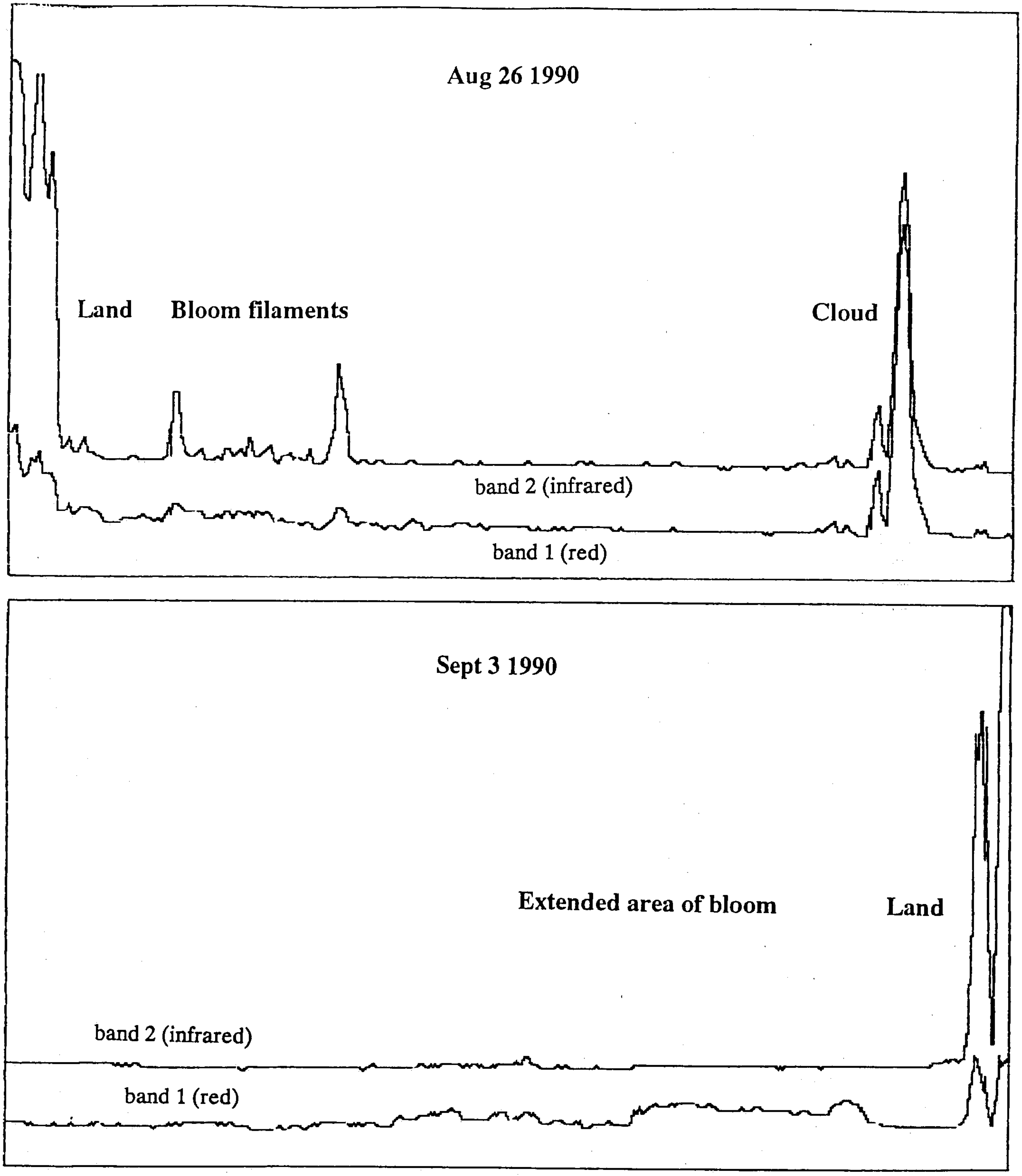


Fig. 3 Cuts through images collected on 26 August (top) and 3 September (bottom), crossing the regions of greatest radiance increase. The relative increases in band 1 (lower curves) and band 2 (upper curves) are very different on the two days.

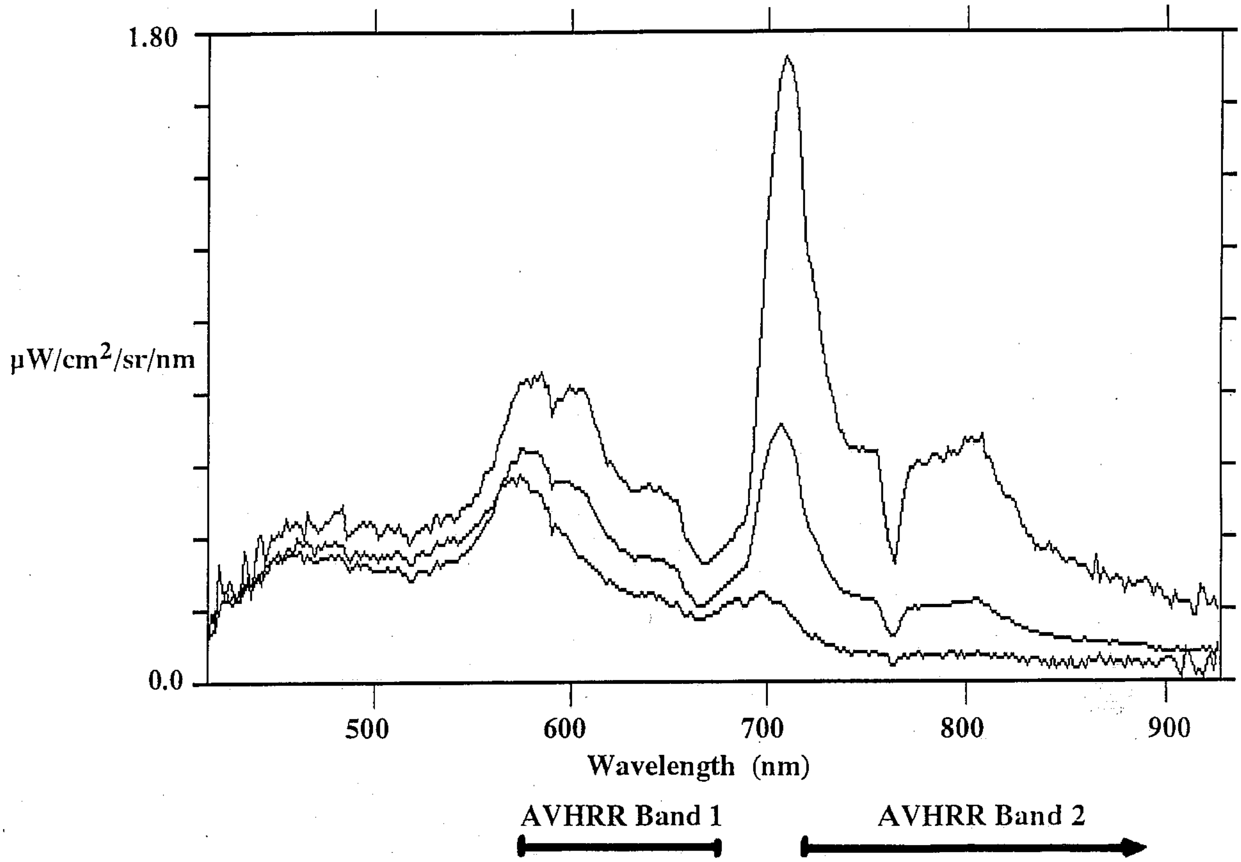
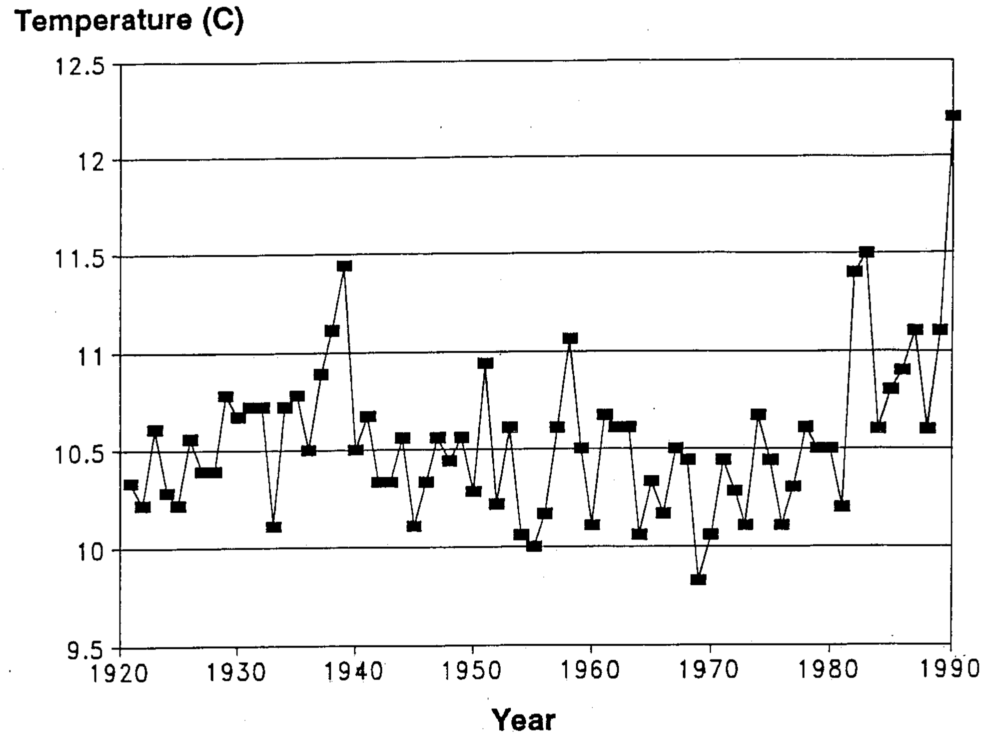


Fig. 4 Three calibrated spectra of different concentrations of *Gonyaulax spinifera* observed with CASI in coastal inlets of Vancouver Island on 4 September 1990, showing (below) the spectral coverage of AVHRR bands 1 and 2.

Fig. 5: see Appendix 1: Colour plates (p. 69)

a



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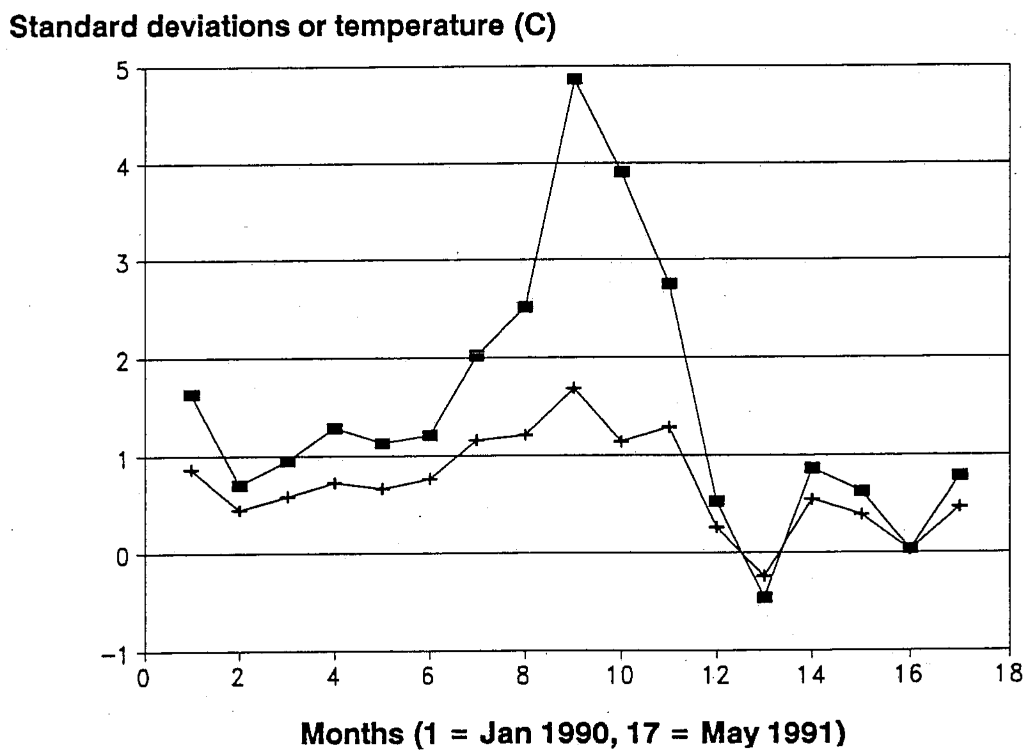

Fig. 6 a. Mean monthly temperatures for September from 1921 to 1990, showing the record warm temperature at the time of the event discussed here.

b. Anomalies in the monthly mean temperatures for 1990 and 1991 (up to May), expressed in ° C (crosses) and in standard deviations of the monthly means (squares). September and October have the lowest standard deviations (0.35° C and 0.29° C respectively). Other months have standard deviations in the range of 0.47° C to 0.63° C.

Conclusions

This event was the most intense and extensive red tide ever observed on the west coast of Canada. The cause of the bloom is unknown, but may be related to either warmer coastal water temperatures or increased nutrients, or a combination of both. Near surface coastal water monitoring stations on the west coast of Vancouver Island (Freeland, 1990) show a warm temperature anomaly at this time, but only of a magnitude that is observed about once every 10-20 years. However, the Race Rocks station which measures the well-mixed waters of Juan de Fuca Strait, recorded the highest September mean temperature in the 70 years of observation (Fig. 6a), at almost 5 standard deviations from the long-term mean. The anomaly was not appreciable in August, but remained high through October (Fig. 6b). This appears to be the clearest indication that unusually high temperatures were occurring.

We show that AVHRR visible and thermal observations can be extremely useful in mapping and studying such plankton blooms. Calculations based on the known cell diameter show that densities of a few times 10^3 cells.L^{-1} should be detectable with the AVHRR, and that peak densities in the dense filaments must have approached 10^8 cells.L^{-1}. These extreme cell densities have important implications for the evolution of the present event, and for planning the mapping of future blooms with satellite and airborne sensors.

The airborne CASI instrument is useful for imaging areas near shore where high spatial resolution is required. It also gives spectra which are useful for estimating cell concentrations and for interpreting the satellite imagery. Airborne instruments can also be used under cloud when satellite observations are impossible.

A more sensitive satellite instrument with coverage similar to the AVHRR is badly needed for monitoring future red tide events. Frequency and severity of such events may be expected to increase in future under the combined influence of any global warming and of increased eutrophication of coastal waters.

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THE SECHELT HARMFUL ALGAL RESEARCH PROJECT (SHARP): AN ENVIRONMENTAL STUDY OF HARMFUL BLOOMS IN A BRITISH COLUMBIA FJORD

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Introduction

We have recently concluded the main phase of a 2 ½ year (three summer) study of the ecology of phytoplankton blooms in Sechelt Inlet, B.C. This inlet was chosen because of accessibility, a previous oceanographic study (Lazier 1963), well defined inputs and outputs, and considerable aquaculture activity. At the start of the study there were 15 fish farm sites and 12 oyster leases (British Columbia Ministry of Agriculture, Fisheries and Food data). In 1986 a devastating fish-killing bloom of the flagellate *Heterosigma* occurred in the inlet and adjacent waters (Gaines and Taylor 1986), but no prior planktological studies had been done in this region and consequently the event was inexplicable. Paralytic shellfish poison (PSP) has also occurred periodically within the inlet.

Our study focussed on species known to be harmful (fish killers, shellfish poison producers: see Table 1), but complete community analyses were performed in order to allow interspecies interactions to be examined. Methodology was essentially 'low-tech' since a small boat was the only way to carry out sampling at sufficient frequency (every two weeks) to describe blooms. Here we will summarize some of the main features of the study in the context of relevance to aquaculture. Complete details are contained in a report (Taylor *et al.* 1991) and a more complete description and analysis of the results will be published in the primary literature.

The inlet

Sechelt Inlet is a fjord located 43km northwest of Vancouver. It consists of a main axis, 29km long, with two side extensions (Fig. 1): the larger Salmon Inlet, which is the head of estuarine circulation, and the smaller Narrows Inlet, which has a secondary sill separating an inner basin from the rest of the system. The main axis is 250 - 300m deep, opening to Agamemnon Channel by Skookumchuk Narrows, a shallow sill area through which strong tidal currents flow (maximum flood $7.5 m.s^{-1}$). Anoxia occurs in the deepest water of the 80m deep inner basin of Narrows Inlet (Smethie 1987), with low levels also occurring in the deeper waters of the main axis. The town of Sechelt at the southern end of the main axis is the only location of substantial human input.

Methods

Closing bottles were not used to obtain a profile of the upper water column since these can miss concentrated layers formed by flagellates under stratified conditions. As standard electrical power was not available it was not possible to perform pump profiling. Instead, we modified the dividable hose method of Odd Lindahl (unpubl. ms.) which uses quick-connect sections of garden hose to obtain segments representing depth intervals. To reduce smearing we used 4cm rigid PVC piping with smooth bore valves at the lower end of each segment. In such a system, resolution is determined by the length of the segments. In ours there were two 1.5m upper segments, followed by six 3m segments for the deeper samples. Carefully lowering the connected segments, we were able to obtain an excellent integrated profile of the upper water column (see Sutherland *et al.* 1991 for the design and testing). Samples from each segment were taken for quantitative cell counts and standard phytoplankton environmental parameters.

In winter the sampling frequency was monthly, but in the main phytoplankton growth period (March to October) it was increased to biweekly to avoid missing short-lived blooms. Closer spacing was logistically impossible.

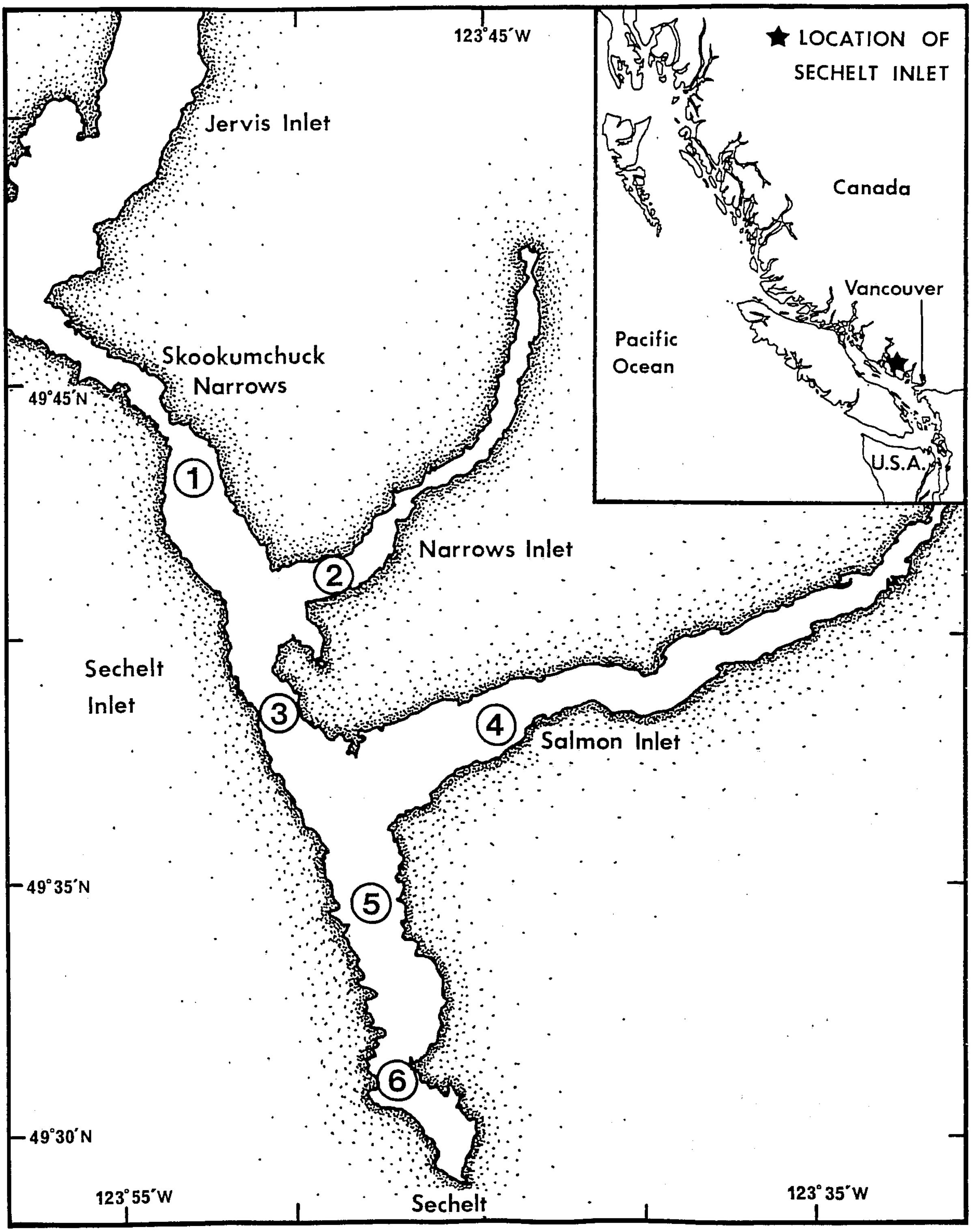

Fig. 1 Map of Sechelt Inlet, showing the primary stations from which data were collected. Inset: location on the B.C. coast.

TABLE 1

Actual (A) or potentially (P) harmful phytoplankton species in British Columbia waters (for details see Gaines and Taylor 1986). PSP: paralytic shellfish poisoning. DSP: diarrheic shellfish poison. ASP: amnesic shellfish poison.

Species	Cause of	A / P
Protogonyaulax[1] *catanella*	PSP	A
Protogonyaulax tamarensis	PSP	A
Protogonyaulax acatanella	PSP	A
Dinophysis acuminata	DSP	P
Nitzschia pungens	ASP	P
Nitzschia pseudodelicatissima	ASP	P
Chaetoceros concavicorne	Fish kill	A
Chaetoceros convolutum[2]	Fish kill?	A?
Heterosigma akashiwo	Fish kill	A
Undescribed chloromonad[3]	Fish kill	A
Dictyocha speculum[4]	Fish kill	P
Chrysochromulina polylepis	Fish kill	P

1 The genus name *Alexandrium* is used by some authors for all species of *Protogonyaulax*.
2 May be a form of *C. concavicorne*; less harmful to fish.
3 A bloom killed fish in 1990. Until brought into culture its group attribution remains speculative.
4 Only the askeletal form is known to be harmful to date.

Results

General inlet features

The inlet is a classic fjord system, with a shallow entrance sill and estuarine circulation and periodic overturn of the deep water. Looking at the inlet as a whole the following observations can be made. All the potentially harmful species of phytoplankton known from B.C. coastal waters (Table 1) are present in the system to varying degrees, although the dominant phytoplankter overall was the common chain-forming diatom *Skeletonema costatum* (usually two-blooms per year; see below). There was clear evidence of nitrogen limitation over long periods in the summer, with much of the water column above 5m containing less than 1 to 2μM.L^{-1} nitrate, and ammonium much less. At these periods, levels of 4μM^{-1} were only reached below 10m. Over the whole main axis N:P ratios of close to 8 were the norm.

The seasonal cycle was typical for temperate coastal inlets, with a spring diatom bloom starting in late February or early March (earlier than in less sheltered waters) and lasting to May. This was followed by a summer predominated by flagellates, particularly nanoflagellates and a lesser, but more diverse, fall diatom bloom in late August or September.

Inter-annual variability

Considerable differences could be seen from year to year in the development of blooms of particular species. A massive summer bloom of *Heterosigma* in 1986 was not repeated in any of the following four years, although minor blooms were present in each year of our study. *Protogonyaulax* (*Alexandrium*) *catenella* had a single outburst in September 1989 (during which a B.C. record level of 32000μg PSP toxin per 100g shellfish was recorded; Fisheries Inspection Branch, Department of Fisheries and Oceans). Some of the variation could be reasonably linked to weather changes. Unusually poor weather (cloudy, windy) in May 1989 led to brief recapitulation of the spring bloom in the summer and an unusual appearance of the fish killer *Chaetoceros concavicorne*. Other organisms, such as *Dinophysis* spp. seemed to be much more regular in their summer occurrence.

Within-inlet variability

The six stations formed a transect along the main axis of the inlet, with two (2, 4) being taken as representative of the side inlets. Examination of profiles of distribution showed considerable differences within the system and allowed inferences to be drawn as to the origins of blooms. Highest phytoplankton abundance, reflected both in chlorophyll *a* and cell counts, occurred at the mouth and at the southern end of the main axis. Tidal mixing was probably responsible for continued supply of nutrients to surface waters in both regions.

The profiles showed clear evidence for blooms entering from the outside through Skookumchuk Narrows (*Heterosigma*, May 1990), favouring flagellates within Narrows Inlet (blooms of several species of dinoflagellates and the silicoflagellate *Dictyocha*: see Sutherland and Taylor 1990), stimulation of blooms in the main axis due to outflow from Narrows Inlet (*Heterosigma*, August 1990), introductions from Salmon Inlet (*Chaetoceros concavicorne, Protogonyaulax catenella*) and blooms originating from the southern end (*Protogonyaulax*). *Dinophysis fortii* and *Nitzschia pungens* consistently developed blooms in the fall at the southern end.

Conclusions

Based on our experience the presence of all the harmful species within the system is unremarkable: we have yet to find a location within the usual salinity range that is free of any of them. The presence and temporal succession of the other species was equally unremarkable, not only for B.C. waters, but for other temperate coastlines of both northern and southern hemispheres.

The study showed that this type of investigation can yield extremely important information for aquaculture, revealing areas of consistently high versus low phytoplankton abundance, as well as probable source locations of harmful blooms. Characterization of the seasonal succession allows an aquaculturist to "follow along" and have a reasonable expectation of events to come, allowing for anomalous weather patterns. The strong interannual variability of some harmful blooms within the system, e.g. *Heterosigma* and *Protogonyaulax*, means that a single year study would be inadequate in determining risk in a body of water.

Our interpretation of the within-inlet pattern is that the two side inlets have very different influences on the system: generally speaking Narrows Inlet favours flagellates, whereas Salmon Inlet is more general, being a source for fish-killing diatoms. Several studies have shown that water over shallow to medium depth anoxic basins favours flagellates, particularly dinoflagellates (see e.g. Iizuka 1972). Shallow areas are also likely to act as seed beds for organisms with temperature-activated benthic cysts or spores. In the Sechelt system only the southern end of the main axis has any extensive shallow areas, the rest being deep and fjordic and thus unlikely to lead to autochthonous blooms.

One early concern with the development of aquaculture facilities was the possibility of eutrophication from animal excretion leading to increased harmful blooms. The expected impact of fish farms of various sizes has been calculated and the area of expected influence predicted (Weston 1986). In terms of phytoplankton in a reasonably well-circulated inlet, such activities can be expected to be negligible. We found no sign of increased nitrogen loading in the system over the period of the study. Indeed, the absolute levels of nitrate and ammonium, plus the low nitrogen:phosphorus ratios, indicate that the upper 10m of the system is nitrogen limited for most of the summer.

This type of study would seem to be an essential prerequisite for the development of a body of water for intensive aquaculture purposes. Unfortunately this has almost never been the case in the past, siting of farms being carried out with highly inadequate information. This has been largely due to the lack of such information for most coastal regions. There is a drastic shortage in trained phytoplankton analysts and the inverted microscope method (Hasle 1978) is time-consuming. The present study involved the microscopic analysis of over 2300 samples with a minimum analysis time of one to two hours per sample! Simple early warning systems, based on information from the farms themselves, can lead to steps to ameliorate the impact of blooms, if such are known, but cannot lead to sufficient understanding of the systems to predict future blooms. We hope that studies such as ours can lead to the latter, given adequate information.

What then is the bottom line? Is this inlet a good or poor location for aquaculture on the British Columbia coast? The 1986 *Heterosigma* bloom which led to approximately $2.5 million in losses, occurred both within and outside the inlet. After that episode, some farms relocated to the well-mixed open passages outside the inlet (Agamemnon Channel). Ironically, our evidence suggests that this flagellate is introduced from outside (Fig. 2) and a 1989 bloom killed $4 million in Agamemnon Channel and was negligible within the inlet. Our data indicate that, once introduced, blooms of *Heterosigma* tend to be concentrated at the entrance to Narrows Inlet and are much less elsewhere and so this indicates that, if this region was avoided, risk from *Heterosigma* would be lessened. Harmful *Chaetoceros* seem to exit from Salmon Inlet and are also more prevalent in the south end in September.

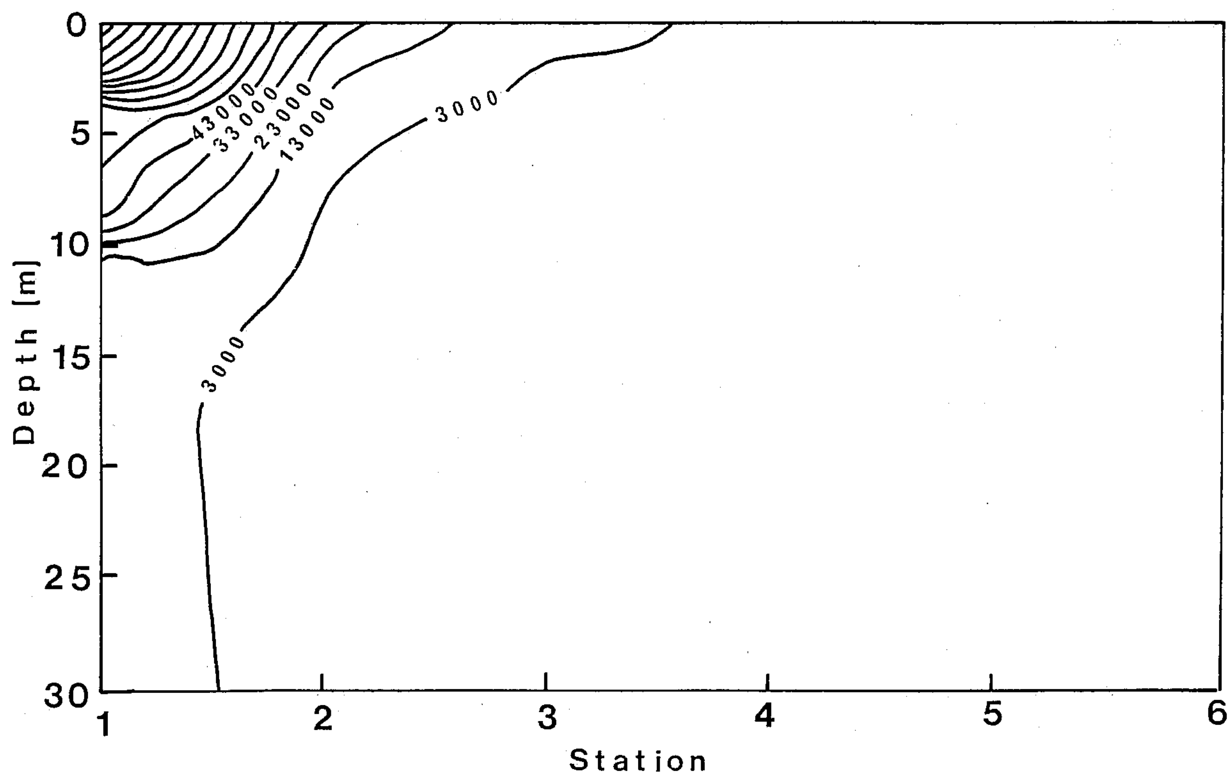

Fig. 2 Example of a longitudinal profile of the upper 30m. North is to the left. Cell numbers are per litre. This distribution of *Heterosigma* in September, 1989, illustrates an introduction through Skookumchuk Narrows. Subsequent development was centred on station 2 (Narrows Inlet entrance), but eventually spread in low numbers throughout the system.

As far as shellfish are concerned, high phytoplankton abundance is favourable. This was consistently found at both ends of the main axis. However, highest *Dinophysis fortii* occurs in the south end and highest *Protogonyaulax* near Salmon Inlet. At present, oysters are the principal product grown. These do not appear to be as subject to shellfish poison accumulation as other species of bivalves. However, if others are grown, more extensive testing for PSP, DSP and ASP within the inlet will be necessary.

If due consideration is taken for within-inlet differences, the inlet does not appear to be unfavourable for aquaculture. The inlet is large enough, generally steep sided, with sufficient circulation that eutrophication effects appear to be very localized, i.e. beneath the cages.

There is an old controversy: in fish farming are enclosed bodies of water to be preferred or are open locations better? Flagellate blooms are usually favoured by stratification and this often occurs earlier in sheltered waters. The answer seems to rest both on scale and on whether blooms originate within it or come from outside it: if yes, no and if no, yes.

Acknowledgements

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B.C. MINISTRY OF AGRICULTURE, FISHERIES AND FOOD STUDIES ON THE MANAGEMENT OF SALMON FARMING ALGAE PROBLEMS

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Since the earliest attempts to culture salmon in marine net cages on the B.C. coast algal problems have been recognized as one of many challenges the farmer had to overcome on his way to financial success (Kennedy 1978). The scope of this problem did not become fully apparent until the sudden growth of this industry in the mid 1980's. While the problem is far from unique to B.C.'s industry (Black and Gowen 1988), the distribution of industry and the resources available to overcome this problem has required the evolution of a unique local situation. Some lessons on how to manage the effects of algal blooms have been learned locally (Black 1988) and internationally (Shirota 1989), and a program based on industry participation in plankton monitoring has been developed to assist local industry in developing ways of managing the plankton problem. The monitoring program has four critical components: collection of a historical data base; a mechanism for communicating developing bloom situations; development of knowledge on ways of minimizing the impacts blooms have on stocks; and training farm staff (Black 1990a, b, c).

As can be seen from Table 1, losses to algal blooms in the early portion of this program were very high. In 1986 38% of all the stock in marine cages on this coast were lost to algal blooms. Four years later this percentage has undergone a continuous reduction to a level closer to 5% per annum. It is hoped that this represents a level of loss which is maximal as a result of lessons learned from the plankton monitoring program and studies initiated to support the industry. One of the principal areas of development was the increased knowledge of blooms of the alga *Heterosigma akashiwo*. This is one of the two dominant problem algal species (Table 1) in British Columbia.

TABLE 1
Historical losses of farmed salmon to phytoplankton blooms in the B.C. aquaculture industry.

Year	% of Industry Stock Lost	Value of Lost Production	Principal Algal Species Involved
1986	38 %	$ 2.5 M	*Heterosigma*
1987	19 %	$ 3.9 M	*Chaetoceros*
1988	< 1 %	$ 0.5 M	*Heterosigma*
1989	5 %	$ 4.2 M	*Heterosigma*
1990	4 %	$ 4.0 M	*Heterosigma*

Some of the principal findings of these studies are:

1. Blooms can develop in areas distant from salmon farms; however, they may be maintained in water bodies containing fish farms long after that source ceased to supply algae (Black *et al.* 1990).

2. A network of farms monitoring algae can give invaluable lead time in preparing for the occurrence of a bloom on the site (Black *et al.* 1990).

3. Blooms of this species are associated with warm (>16° C) stratified waters (Black *et al.* 1990).

4. Atlantic salmon and trout are more susceptible to loss due to this algal than Chinook or Coho (Black 1990d).

5. Large fish are more susceptible than small fish (Black *et al.* 1991).

6. The mechanism of death does not appear to involve demand for or supply of oxygen to the fishes tissue (Black *et al.* 1991).

7. A labile, fast acting, low molecular weight toxin appears to be responsible for the fishes death (Black *et al.* 1991).

8. Lack of clinical symptoms in humans who have consumed fish which have died of exposure to this alga suggests the toxin may not be a hazard to seafood consumers.

9. Field observations suggest that fish given the opportunity to avoid the bloom through the use of deep nets survive better than fish maintained in shallow nets (Black *et al.* 1990).

10. The choice of farm site in a location with turbulent vertical mixing of the water column (such as at a shallow sill in a fjord or inlet) may result in very high mortalities (Black *et al.* 1990).

11. The cessation of the bloom appears to be associated with the loss of water column stratification in an area (Black *et al.* 1990).

12. The end of the bloom does not result in a cessation of mortalities induced by the bloom. Outbreaks of the disease Vibriosis are common in stocks which have survived a bloom, suggesting that the toxin induced stress or the cessation of feeding during a bloom may lower the cultured fishes' resistance to this ubiquitous pathogen.

Other aspects of algae also constitute a threat to salmon farming on this coast. Most notable of these is algae of the *Chaetoceros* genus. However, our understanding of the risk these species pose to salmon may require more taxonomic work on the differentiation of the forms presently identified as *C. convolutum* and *C. concavicornis* or a better understanding of environmental factors involved in the development of secondary spicules on the setae of these species.

Further, two more algal species have been associated with a bloom in 1991 which bear investigation. One is an undescribed chloromonad (F.J.R. Taylor, U.B.C., pers. commun.). The other is a species that Rosemary Waters (Guildford, U.K., pers. commun.), supported by Y. Hara (Japan), has good evidence is a cold water species of *Chattonella*. This genus has been associated with mortalities of fish in Japan. Overall, it would appear that the knowledge gained towards reducing losses to algal blooms appears to have assisted industry in reducing its losses to algal blooms. Most of the serious blooms and most of the knowledge development has, however, centered around the species *Heterosigma akashiwo*. We may be and likely are inadequately knowledgeable to control losses to blooms of other species such as *Chaetoceros* spp. or the species detected in 1991. More work is required on the taxonomy and morphological response of both *H. akashiwo* and these other species to changing environmental conditions.

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A NEW APPROACH FOR IDENTIFYING PSP TOXIC SHELLFISH

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Introduction

Paralytic shellfish poisoning (PSP) occurs in individuals that have consumed shellfish previously exposed to toxic dinoflagellates, marine organisms that are the source of a variety of neurotoxins, collectively termed Paralytic Shellfish Toxins (PST). Bivalve shellfish, such as clams and mussels that are contaminated with PST do not normally exhibit characteristic signs of PSP, and therefore can not be easily identified as potential health hazards. As a result, it is imperative that bivalve molluscs are tested for the presence of PSP prior to making shellfish available to the consumer. At the present time, the mouse lethality bioassay, initially developed by Sommer and Meyer (1937), remains the accepted procedure for PSP detection. Associated with the mouse bioassay, however, are the disadvantages of requiring a large number of mice of specified age and strain, and the concern regarding inter-laboratory accuracy of detecting low PSP toxic shellfish. During the last 15 years there have been numerous attempts at replacing the mouse bioassay with alternative bioassay, chemical and serological tests for PSP. In our laboratory, we discovered that the intertidal crab, *Hemigrapsus oregonesis*, exhibits varying signs of resistance to the toxicity of saxitoxin, a principal PST, when exposed to a "Red Tide Bloom". The seasonal resistance of this small shore crab to saxitoxin was further demonstrated to be associated with the presence of a high molecular weight protein, termed Saxitoxin Induced Protein (SIP). Preliminary studies using the SIP protein complex as a serological probe for identifying PSP contaminated shellfish were initiated and showed promise.

The purpose of the present study was to perform a field study with PSP contaminated butterclams and to determine the usefulness of our serological test in detecting PSP toxic clams.

Materials and methods

Bivalve mollusc samples:

PSP toxic butterclams were obtained from a variety of locations in the Prince Rupert area (Canoe, Gasboat, Prescott and Dudevoir Passages, Kitkatla and Hunts Inlets, and Larsen Harbour, to name a few), courtesy of the Department of Fisheries and Oceans, Inspection and Special Services Branch. Samples underwent an acid extraction and were analyzed for total PSP toxicity according to the standard mouse lethality bioassay. Samples used for the serological SIP assay were dissected to remove the foot tissue and this material was homogenized, centrifuged and prepared for an ammonium sulphate extraction (75% NH_2SO_4 cut). Samples were centrifuged and resuspended in phosphate buffered saline prior to determining the protein content and final dilutions for the SIP ELISA assay.

ELISA procedure:

Affinity chromatography procedures, utilizing protein A, purified SIP IgG, coupled to an Aminolink matrix (Immunopure AG/AB immobilization kit, Pierce, Rockford Il.) were used to purify the SIP standard for direct ELISA. Purified IgG was diluted into a blocking buffer to a concentration of $75\mu g.mL^{-1}$ and cross-absorbed to crude nontoxic butterclam extracts with known PSP toxicities. The clam homogenate sample was diluted to 1:5000, 1:10000, 1:20000 to ensure a protein range of 0.3 - $4\mu g.mL^{-1}$. Following application of the sample to the ELISA plate, the cross-absorbed IgG was added and incubated for 2 hr at room temperature. The secondary antiserum was anti-rabbit IgG alkaline phophatase conjugated antibody and the substrate was nitrophenyl phosphate. The absorbency readings were measured at 405nm.

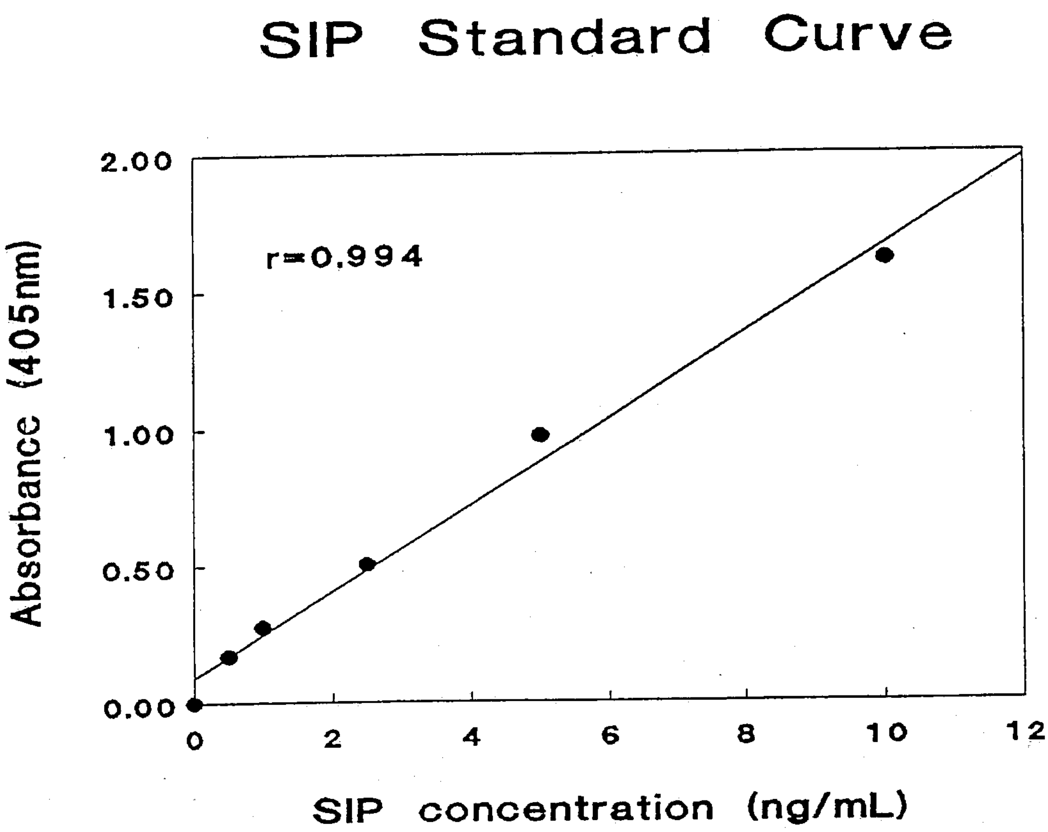

Fig. 1 Standard curve for SIP absorbance in the clam serological assay.

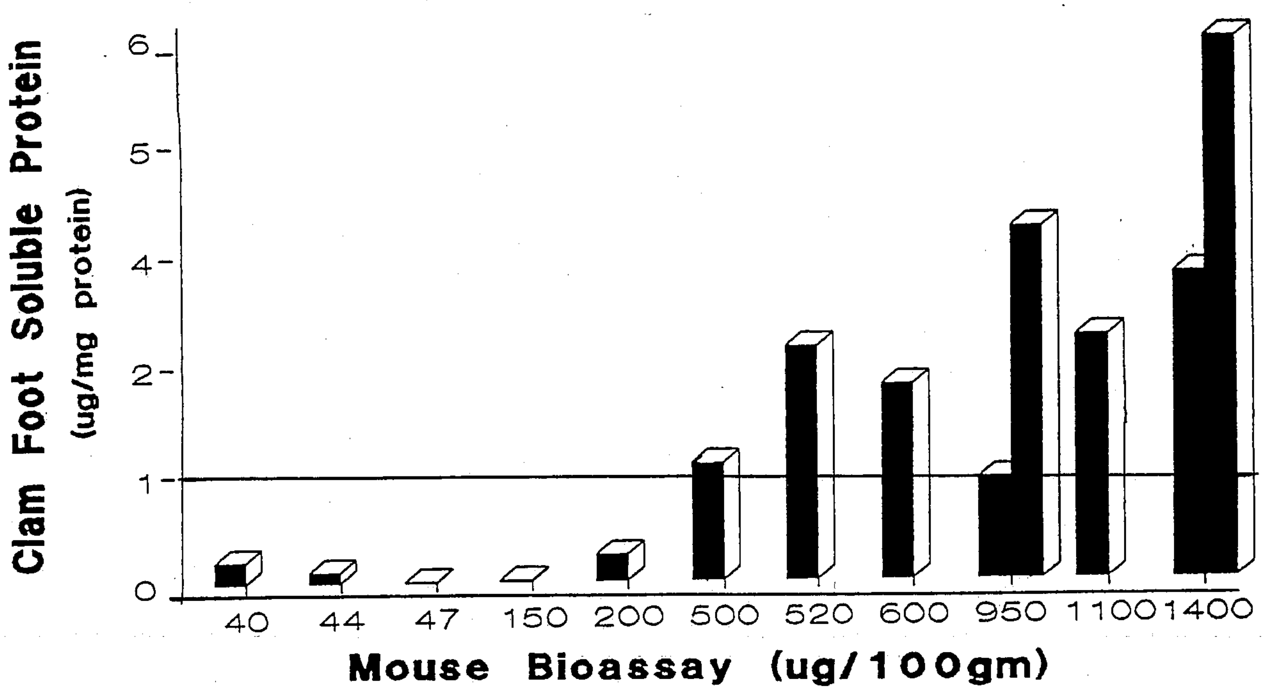

Fig. 2 Comparative results of the relationship between the clam ELISA assay and the mouse bioassay.

Data analysis:

Affinity purified crab SIP was used at the concentration range of 0.5 - $10 ng.mL^{-1}$. The clam SIP equivalent was determined from this standard following correction for dilution. The final value was expressed as a percentage of total protein in the sample. The standard curve was analyzed by a linear regression. The prediction of correct estimates concerning whether samples should be re-tested for PSP content by the mouse lethality bioassay, following the results from our serological test, was made from a random binomial probability estimate.

Results and discussion

Results of preliminary studies concluded that the clam foot was the best source of tissue for detecting SIP cross-reactive antigens that corresponded to actual PSP toxicity values. Although we were able to detect cross-reactive antigens in the clam siphon and adductor muscle as well, the degree of cross-reactivity in these organs precluded determining any meaningful association between clam PSP content and SIP cross-reactivity.

The standard curve for SIP in the clam serological assay is presented in Figure 1. An excellent correlation ($r = 0.994$) between SIP concentration and absorbency readings at 405nm was obtained. It is important to note that the source of the SIP is the crab, and therefore we are effectively measuring cross-reactive antigens in the clam sample and possibly not actual SIP. As a result, we have referred to the SIP equivalents measured in the clam samples as 'clam soluble protein complex' until a better characterization of the cross-reactive clam proteins is made.

Figure 2 presents the results of our serological clam assay plotted against the results of the standard mouse bioassay on a limited number of samples. Standardizing the serological assay result for the amount of protein in the assay was shown to improve the correlation ($r = 0.775$; $P < 0.05$; $n = 10$) between the two assays. We have since expanded our field testing to include over 52 different analyses with varying PSP

TABLE 1
Random probability estimates of correctly identifying PSP toxic shellfish on basis of soluble protein measurements in the butter clam[1]

Sample size (n)	PSP Toxicity range[2]	Correct predictions (%)	Probability of correct answers by random guess (%)	Psig
10	ND-46	100	0.01	0.001
10	47-57	90	1.07	0.001
10	57-89	100	0.10	0.001
10	90-300	100	0.01	0.001
12	300-2000	100	<0.10	<0.001

[1] Prediction criterion = 0.1% clam soluble protein complex (SCPC). e.g. PSP toxicity < 90μg, given %SCPC≤ 0.1% soluble clam protein. PSP toxicity > 90μg, given %SCPC≥ 0.1% soluble clam protein.

[2] estimated by mouse bioassay ($\mu g.100g^{-1}$ shellfish)

toxicities and have confirmed the significant correlation ($r = 0.826$; $P < 0.01$; $n=52$) found between the two assay procedures. On the basis of this result, and the fact that we relied on a specific cross-reaction to identify clam samples which contained unacceptable PSP toxin levels, we established a clam soluble protein cut off value of $1\mu g.mg^{-1}$ protein (or 0.1%) clam protein for the prediction of shellfish containing PSP. It is easily seen that samples in the higher toxicity range (> $200\mu g.100g^{-1}$ sample) contained more immunoreactive protein. To test this further, the results of our complete study (52 samples; PSP toxicity ranges of 40 - $2000\mu g.100g^{-1}$ shellfish) were randomized and a blind study was performed to determine the probability of correctly predicting clam samples that should undergo further PSP testing by the standard mouse bioassay (Table 1).

The results in Table 1 show the random probability of correctly identifying PSP toxic shellfish on the basis of our soluble clam protein measurement. The data was analyzed using a binomial probability estimate of obtaining a correct answer by a random guess as well as the level of significance in obtaining the number of correct answers in groups of PSP toxic shellfish subdivided into specific levels of PSP toxicity. The analysis indicates that the probability of obtaining our answer by random guess work is very small in all ranges of PSP toxicity. We conclude therefore that the serological method, reported herein, could be a very useful screening method for identifying PSP containing clams.

The approach that we recommend, with the assay results in mind, is that samples yielding an absorbency value of greater than $1\mu g.mg^{-1}$ soluble clam protein would not require further mouse bioassay testing, since the probability has been shown that they contain PSP toxins above the acceptable limit. This approach would save the necessary mouse bioassay testing for samples that are below the $1\mu g.mg^{-1}$ soluble protein cutoff, and which normally would require supportive ancillary testing by the mouse bioassay for confirmation of safety assurance. This conservative approach, while retaining the requirement for mouse testing for final confirmation of absolute toxicity, could be considered useful in increasing monitoring frequencies of those shellfish regions with historic sporadic outbreaks of PSP. Moreover, increasing the amount of testing during actual coastal closures following sufficient evidence by the present method, could be viewed as recommendations for earlier re-testing using the mouse bioassay. In conclusion, this screening method in practice may contribute to a reduction in the number of mice that are used for PSP testing and thus the subsequent cost of performing the mouse lethality bioassay.

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ADVANCE WARNING AND SCALED RESPONSE PLANNING FOR TOXIC BLOOMS

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Introduction

During the second half of August 1990 an exceptional red tide developed off the west coast of Vancouver Island (Forbes *et al.* 1990, Gower and Borstad (In press)). This was the largest such event recorded along the B.C. coast and, while caused by a dinoflagellate not known to be toxic, *Gonyaulax spinifera*, it created considerable public concern. Substantial shellfish mortality was associated with this event, primarily in Barkley Sound, where *G. spinifera* intermixed with or was replaced by *Prorocentrum micans*, but also to a lesser degree along the outer coast in the area of Long Beach.

The Department of Fisheries and Oceans provides inspection services to ensure that harvested wild and farmed stocks of shellfish are free from algal toxins. The program is expensive to operate, but has important implications for human health. Although the area covered by regular inspection has increased over the last several years to cover the south coast and Vancouver Island, logistics and cost factors limit expansion to the central and north coasts. The aquaculture and wild shellfish harvest industries in B.C. have experienced major losses over the last several years due to algal blooms. Closures of harvesting areas lead to lost sales and reduction of market share for shellfish harvesters. *Chaetoceros* and *Heterosigma* blooms lead to direct loss of fish on fish farms, as well as costs incurred in attempting to avoid exposure by moving pens and reduction in growth due to suspension of feeding.

The *G. spinifera* bloom and continuing industry problems with toxic algae indicate the need to be able to provide advance warning of algal blooms developing offshore and respond in a timely manner to provide information to the public, inspection services and industry on the blooms as they develop. This year the DFO is initiating a project to investigate the feasibility of developing a practical and cost-effective warning system and reviewing appropriate response approaches.

Long-term goals and approaches

Advance warning of toxic blooms would let shellfish growers and fish farms take precautionary measures such as anticipatory harvest, moving of stock, or cessation of feeding. It would also permit DFO Inspection Services to deploy field effort more effectively by indicating where sampling should be concentrated. Three major approaches have been considered: remote sensing, either airborne or by satellite (Yentsch 1987), direct water monitoring (Stockner 1991) and predictive modelling (Menesguen *et al.* 1990, Thomas and Gibson 1990a, 1990b). Each has advantages and disadvantages, some of which are indicated in Table 1.

Current project plans

The initial stage of the project will be to obtain comprehensive documentation of blooms occurring in offshore and northern B.C. coastal waters, in particular the causative organisms, magnitude, frequency and seasonality of blooms. This will form the basis for effective programs to provide advance warning of blooms. The reasons for concentrating on the offshore and northern areas are that there is concern that major blooms may develop offshore and then be advected onshore, and that records for northern B.C. waters are virtually non-existent.

The approach will be two-fold. First, a long-term data base (10 years) of phytoplankton identity and abundance from the continental shelf of B.C. will be analyzed. This will be used to identify and quantify problem species, assess the temporal and spatial variability of phytoplankton community composition on the coast, and identify physical and chemical factors influencing the occurrence of particular species or species groups. Second, a field

TABLE 1
Advantages and disadvantage to advance warning approaches

REMOTE SENSING SATELLITE

Advantages: Large spatial coverage
Moderate cost
Straightforward logistics

Disadvantages: Current sensors are not very sensitive to algal blooms
Algal community composition information unavailable
Weather-dependent
Samples surface only

AIRBORNE

Advantages: Moderate spatial coverage
Rapid deployment coastwide to problem areas
Moderately sensitive to community composition
Logistics moderately straightforward

Disadvantages: High cost
Samples surface only
Requires moderately good weather
Limited availability (one unit currently in B.C.)

DIRECT WATER MONITORING

Advantages: Immediate identification of causative organism
High precision quantification
Can sample at depth
Associated physical and chemical measurements can be made

Disadvantages: Restricted spatial coverage
Logistics of sample and data distribution are difficult
High cost
If shore based - will miss blooms that develop offshore
If ship based - logistics problems are compounded
Requires skilled taxonomists

PREDICTIVE MODELLING

Advantages: Very low cost once models are in place
Building block approach can allow incremental spatial coverage from restricted to coastwide
Can direct water sampling specifically to areas at risk

Disadvantages: Requires long-term research effort to implement, with associated expense
What if you are wrong?

program similar to that in place in the 1960's (Quayle 1969) will be implemented. This will involve placing sampling gear on board DFO patrol and science vessels to sample blooms that they observe or that are reported to them in the vicinity of their areas of operation. These vessels are deployed coast-wide over the period when problem blooms may be expected to develop. Observers on fisheries branch aerial patrols will be involved in reporting blooms to the project coordinator and directly to vessels. Regular samples will also be obtained from selected light stations along the coast. We recognize that this system will not capture all blooms, as demonstrated by Quayle (1969), but it will provide us with substantially more information than presently available. We will cooperate with DFO Inspection Services, industry monitoring programs and university researchers to document blooms coastwide as comprehensively as possible.

Scaled response planning will involve considering guidelines for the level of response based on the identity of the organism in a bloom and evidence of the concentrations and magnitude that the bloom will achieve, representing the levels of risk to the industry. Levels of response that could be considered include:

1. Report of occurrence to people calling in to a phytoplankton watch program
2. Advisory distributed to industry and Inspection Services
3. Extended sampling to establish distribution and abundance
4. Remote sensing mapping of the developing bloom area
5. Track prediction for bloom advection

Note that the first two levels are currently in place for a restricted area of the south coast, under the direction of the Phytoplankton Watch program (Stockner 1991).

This program will be considered a pilot project only at this stage. The conundrum that bedevils this type of program is that you can't respond to all blooms and by the time you recognize that a bloom may cause problems, you should already have responded to it!

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AQUACULTURE INDUSTRY CONCERNS REGARDING TOXIC ALGAE

J. Spence, B.C. Aquaculture Research and Development Council

Salmon farming

The most pressing concern was that the Plankton Watch program of E. Stockner would be funded in 1991. This funding has now been confirmed by B.C. Ministry of Agriculture Fisheries and Food. In addition, BCARDC will assist in an NRC-IRAP application to cover the costs of an assistant.

I hope that Department of Fisheries and Oceans (DFO) programs will complement this valuable work in Plankton Watch. A close working liaison should be established between DFO and Plankton Watch. BCARDC will be willing to assist in this process if necessary.

Mechanisms of algal toxicity

Additional research is needed in B.C. on the mechanisms of algal toxicity. We anticipate that Dr. Max Taylor and his students will be developing further projects in this area.

Analytical Techniques

The shellfish industry has a strong interest in forecasting and responding to the threat of algal toxins in shellfish - PSP and DSP toxins, domoic acid, etc.

In particular, the industry wish is to investigate the development of reliable, rapid and quantitative techniques for measuring the levels of toxins in seafood and in phytoplankton. They are discussing the possibility of testing HPLC techniques as an alternative or supplement to mouse or rat tests for algal toxins.

The need for a more comprehensive approach to toxic algal monitoring

In the past, programs have been ad hoc or regulatory in nature. We need to move to a more comprehensive approach to these problems addressing all aspects of the source - fundamental research, toxicology, new quantitative assays, etc. - that serve all the needs of our seafood industry, including the wild harvest, shellfish cultivation and salmon farming.

Appended are the Major Findings and Recommendations of Phase II of the Industry Science and Technology (ISTC) Fisheries Sector Campaign - Shellfish Quality Control and Product Purity Projects:

Project II Algal Toxins - Conclusions, recommendations and further research

Prepared by Monty Little, Syndel Ltd. for B.C. Aquaculture Research and Development Council and B.C. Oyster Board, March 1991.

Conclusions and Recommendations

There is a need for a rapid, inexpensive, site-specific system to detect the onset, presence and clearance of toxic algal contamination at shellfish operations in the province.

The principal criteria for this system are suggested:

1. Provision for input from the farm site
2. Compatible and complementary to DFO mussel watch program
3. Provide early warning of impending blooms
4. Offer mitigating procedures

It is felt that such a site-specific system would help to overcome prolonged closures (normally resulting in loss of sales and markets to farmers), but still provide the safety needed for human health.

A farm monitoring system is suggested with adherence to a standardized method paralleling the Phytoplankton Watch Program of the B.C. salmon farming industry. It is thought that the latter program could be expanded to include shellfish industry sites. An early warning system makes sense and has proved reliable and beneficial for finfish farmers. A co-operative approach with the B.C. salmon farmers is suggested.

Further Research

Quick, qualitative assays are needed to identify the presence of algal toxins in B.C. shellfish growing waters. Quick validation of farm-site pathogen identifications is difficult. In conjunction, tests for toxins must be reliable and reproducible. More research is needed to develop these types of qualitative methods.

An extraction protocol is required if PSP is to be monitored directly from phytoplankton as is being done for ASP.

Use of a standardized physicochemical method must be developed and adopted, as bioassays are no longer in favour (HPLC and electrophoretic methods most likely choices for PSP). A standard procedure for the extraction and analysis of all the toxins still remains to be adopted and, until such time, private laboratories will not be able to offer back-up services to the industry.

The transfer of PSP identification technology from government to private laboratories should be considered to accelerate the development of appropriate protocols.

Canada should strive to become the internationally recognized source of PSP, ASP and DSP toxin standards.

SELECTED CONCERNS OF THE WILD HARVEST INDUSTRY

K. Vautier, United Fishermen and Allied Workers Union

The major concern of many in the wild harvest industry relates to loss of market caused by long closures. These are due primarily to limited inspection capabilities and sanitary shellfish closures, with fewer due to algal toxins.

The industry has been able to take advantage of the increased level of inspection, which has removed the blanket closure on the west coast of Vancouver Island. A further expansion would be desirable, particularly into Areas 7, 8, 9 and 10 on the central coast.

Algal toxin problems are relatively low on the list of priorities compared to other problems. However, the industry would like to see simple, fast, specific and cost-effective new analytical procedures that could lead to site-specific or producer exemption from the blanket closures.

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PARALYTIC SHELLFISH MANAGEMENT PROGRAM IN BRITISH COLUMBIA

R. Chiang, Department of Fisheries and Oceans, Fish Inspection Branch, Burnaby, B.C.

Introduction

Poisoning from the consumption of toxic bivalve molluscs has long been recognized by native communities along the Pacific coast. The earliest recorded victims were crew members from Captain Vancouver's voyage in 1793. The federal Department of Fisheries and Oceans has compiled paralytic shellfish toxicity records in British Columbia since 1942 (Quayle and Bernard 1966, Quayle 1969). While the frequency and intensity of toxic dinoflagellate blooms may vary from year to year, the probability of having a toxic bloom somewhere along the British Columbia coast approaches certainty in any given year. The challenge for a monitoring program is to detect when and where toxic blooms may occur.

Program objectives and components

The paralytic shellfish (PSP) monitoring program is a major component of the Canadian Sanitary Shellfish and Toxicity Management Program. The objectives of the Canadian program, in the context of shellfish toxicity monitoring, are as follows:

1. Public safety: to provide reasonable assurance that shellfish harvested from open areas are not toxic.

2. Resource utilization: to promote the best use of shellfish resources and optimize the long term economic return to Canada.

By providing a safe shellfish product to the consumer, a sound shellfish toxicity management program achieves not only the first objective, but also reinforces the second objective by enhancing consumer demand. The integrity of bivalve fisheries can only be preserved through a diligently managed shellfish program that encompasses both public safety and resource management concerns.

Goal paths

To achieve the stated objectives, a PSP management program must be able to **detect** and **respond** to toxic blooms in a timely manner. Open areas must be monitored.

Risk assessment

The risk to public health is a function of the extent of a monitoring program and the response time required to implement closure actions (Chiang 1988). The risk decreases with better monitoring or faster (i.e. reduced) response time. If the response time increases, the risk will become higher unless the delay could be compensated for by more intense monitoring. In remote areas where the logistics of sampling, transportation and the regulatory agency's ability to respond to sudden outbreaks preclude reasonable response time, it may be prudent to keep the areas closed.

PSP toxicity monitoring

In recent years the major component of the toxicity monitoring program consists of a series of sampling locations where mussels are collected on a weekly or biweekly basis. Local residents are paid a fee for sample collection. In 1990, there were 62 sites in the south coast of British Columbia where 2039 samples were collected. The locations of these sites are illustrated in Figure 1.

To supplement the mussel monitoring sites, field samples are also collected at other locations by fishery officers and patrol crews from time to time. Commercial geoduck fishermen, scallop fishermen and oyster growers have also provided samples from other locations on request. There were 488 samples from these sources in 1990.

To provide for safety verification, commercial bivalves are sampled periodically at federally registered processing plants. In 1990, a total of 439 commercial samples were taken. It should be noted that commercial samples can serve as compliance verification measures only. A toxicity monitoring program must not rely solely on commercial samples.

Closure decisions

It is internationally accepted that when the toxicity level exceeds 80μg of toxin per 100g of edible tissue, the area of harvest shall be closed and the product rejected. The decision to close an area is never easy. The spontaneous nature of toxic dinoflagellate blooms seldom afford the decision makers the luxury of delineating closure boundaries that encompass the toxic area exactly. By the time sample results are known, the data could be outdated. A toxic bloom may be extensive or sporadic, thus affecting massive areas or only spotty locations. The intensity of a bloom may also vary, resulting in significant differences in toxicity levels among bivalves of the same or different species. The decision to close an area should therefore involve not only the 80μg criterion, but also the dynamics of the bloom. A good monitoring program may provide insights to the intensity and the extensiveness of a bloom. It may also indicate whether the bloom is active or subsiding. In the absence of perfect information, it is prudent, however, to include a sizable safety zone in the closure of an affected area.

Controllability

The essence of a shellfish program is controllability. It is known that toxicity levels can vary significantly among individual clams dug from the same beach at the same time. Quayle (1969) has reported that the toxicity levels of 41 individual butter clams so harvested had a range of 50 - 1568μg. The mean was 669μg with a standard deviation of 340μg. Given the high variability of toxicity levels among individual clams and the sporadic nature of PSP blooms, a PSP toxicity management program must focus on the **ability** to detect toxic blooms or bloom conditions in a body of water. This requires that on-site monitoring samples be taken at regular intervals. In the absence of such data, lot by lot analyses of commercial products will not provide the same degree of safety confidence, because product homogeneity cannot be assumed. Areas open to bivalve harvesting must therefore be monitored.

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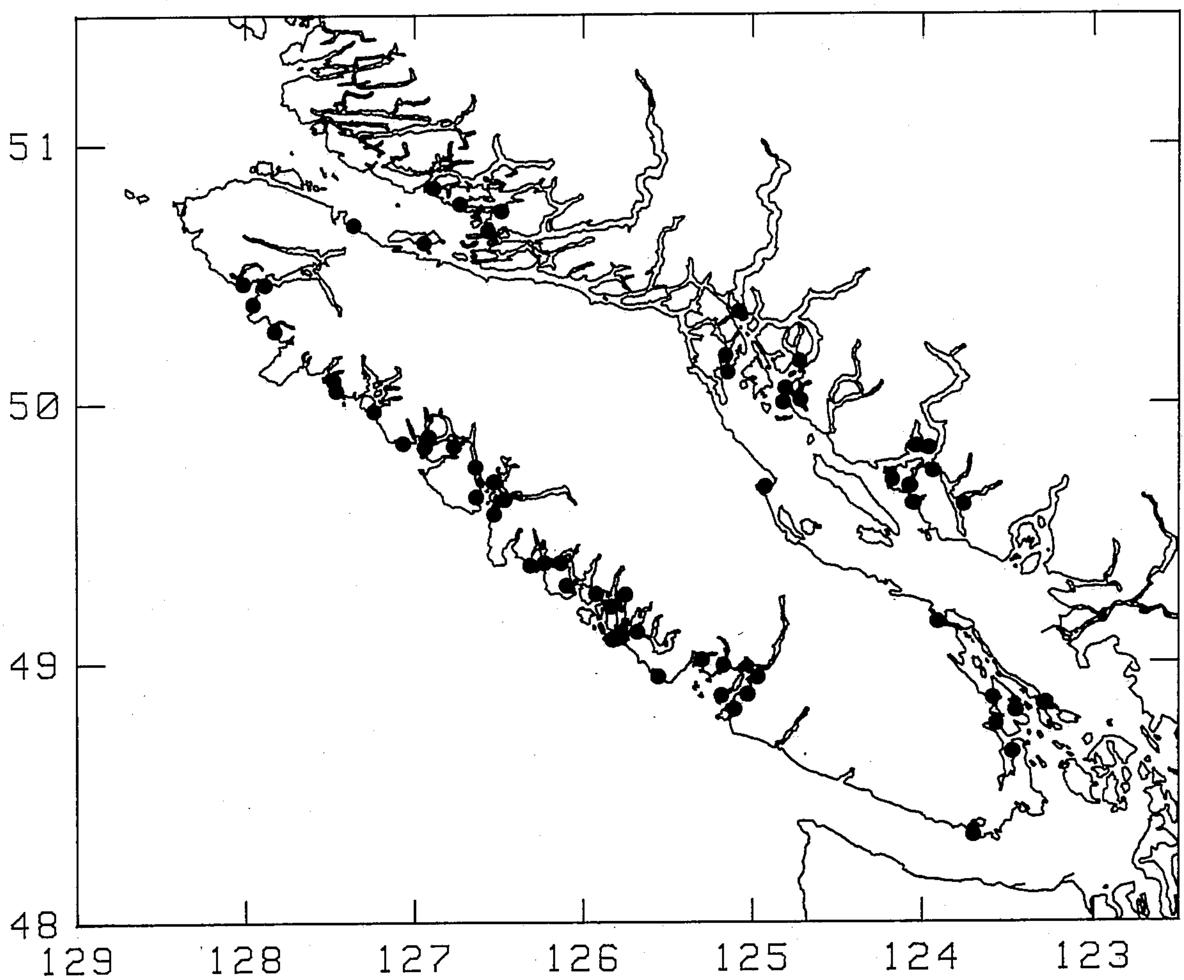

Fig. 1 Locations of the 62 sampling sites in 1990.

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DEPARTMENT OF FISHERIES AND OCEANS INSPECTION - EASTERN COAST PROGRAMS

S. Stephen, Department of Fisheries and Oceans, Inspection, Regulations and Enforcement Branch, Ottawa

Canada has been monitoring molluscan shellfish for the presence of paralytic shellfish poison (PSP) toxins since 1943. The Association of Official Analytical Chemists (AOAC) mouse bioassay has been the only approved detection method used in Canada since that time.

Originally, shellfish extracts were prepared by Department of Fisheries and Oceans (DFO) Inspection labs in the regions and shipped to the Department of Health and Welfare (DHW) in Ottawa, where the bioassays were done. Turnaround time on results would often be a week or longer. This arrangement made management of shellfish harvest areas difficult.

Areas like the Bay of Fundy portion of Scotia-Fundy Region are prone *Alexandrium* spp. blooms and most, if not all, of southwest New Brunswick harvesting areas are closed each summer due to PSP. Pressure from industry and the need for more rapid results led to the first bioassay lab on the east coast being established at Blacks Harbour, New Brunswick, in the mid 1980's. With most of the PSP problems focussed in the Bay of Fundy, the Blacks Harbour laboratory did as many as 5000 bioassays in some years. The majority of samples came from southwest New Brunswick and southwest Nova Scotia.

In November 1987 things changed with the mussel crisis and domoic acid, or Amnesic Shellfish Poison (ASP). DFO received additional funds and Inspection began its enhanced shellfish monitoring program.

As a result, today there are mouse bioassay labs in St. John's, Newfoundland, Moncton and Blacks Harbour, New Brunswick, and Sept Iles, Québec. Two regional labs, Halifax and Moncton, and the National Inspection lab in Ottawa have the capability to test for ASP using the approved DHW high performance liquid chromatography (HPLC) method. Nearly 31 Inspection 'person-years' are involved at least in part in the shellfish program. This includes field and lab staff involved in sampling, analysis, and shellfish plant and product inspections. Although the program is a national one it is administered by each of the four east coast regions (Newfoundland, Gulf, Scotia-Fundy and Québec) on a regional basis.

Newfoundland Region's shellfish industry is almost exclusively based on cultured blue mussels and the majority of the aquaculture sites are located in Notre Dame Bay. Historically, Newfoundland was not known to have any PSP. That was until 1982 when the first confirmed PSP illness occurred after a man consumed a quantity of mussels (White and White 1985). There have been isolated incidents of PSP in Newfoundland shellfish since that time. A monitoring program is in effect, but on a smaller scale than in other regions. Approximately 400 mouse bioassays were done last year.

The Gulf Region is perhaps the most unusual in that it has jurisdiction in portions of each of the four Atlantic provinces (Newfoundland, New Brunswick, Prince Edward Island and Nova Scotia). Its shellfish industry is a mixture of soft-shell clams, oysters, quahogs, bar clams and cultured mussels. PSP is found in the area, but not nearly to the same extent or frequency as in the Bay of Fundy. There have been no PSP closures for bivalves since 1988, but closures were made in six areas in 1989 for moonshells, whelks and winkles. There were no closures due to ASP this past winter and in fact the level of domoic acid did not reach $5 \mu g.g^{-1}$ in any sample analyzed. Gulf Region anticipates doing 2000 bioassays this fiscal year with 800 of those going for HPLC analysis for ASP.

Unlike many European countries, Canada operates closure of shellfish areas based on phycotoxin levels in shellfish, not on numbers of the causative organisms in the surrounding waters. The Gulf Region was the first Inspection region to participate actively in a phytoplankton monitoring program. Originally developed by Gulf Science Branch, the techniques used allow for routine water column sampling for phytoplankton using simple and inexpensive methods. Samples are then examined to identify and enumerate a variety of phycotoxin producing species. Detection of PSP, ASP or diarrhetic shellfish poison (DSP) producing organisms gives Inspection Services an early warning system and allows staff to increase sampling of molluscs in potential problem areas. Inspection in the Gulf Region currently has 25 water sampling sites and expect to do 1400 samples this year.

In Scotia-Fundy Region there are a variety of shellfish species harvested: soft-shell clams in the Bay of Fundy, cultured mussels on the Southern and Eastern shores, and mussels and oysters in Cape Breton. Testing is also carried out on roe-on scallops harvested from Georges Bank. Sampling is on a lot-by-lot basis, with ASP testing done by an independent laboratory in Nova Scotia. The lab is also routinely monitored by Inspection and must participate in split sampling. In southwestern New Brunswick alone there are 20 key sites and approximately 1400 harvest area samples are taken annually. All are tested for PSP with about 300 also being tested for ASP. Data on PSP levels in local shellfish in southwestern New Brunswick is probably the most detailed in Canada and dates back to the beginning of PSP monitoring in Canada. Sampling normally increases in frequency during the warmer summer months, with many locations being sampled twice a week. The majority of PSP closures predictably begin around the July first weekend. A 24-hour toll free 'Clam Line' is operated out of Blacks Harbour allowing industry and the general public to obtain information on which shellfish areas are open. Domoic acid also appeared in shellfish in southwest New Brunswick on 1 September 1988. Closures occurred in and around St. Andrews with some lasting until mid-October.

Although the southwest New Brunswick Inspection does not participate in a phytoplankton watch program, it has had strong connections with staff at the St. Andrews Biological Station of DFO. They receive information from staff at the station on increases in numbers of *Alexandrium* and *Nitzschia* spp. in the area and, in turn, they advise of any changes in toxin levels or types in shellfish. Additional cooperation is obtained through close ties with John Hurst of the Maine State Department of Marine Resources. Maine generally sees a rise in PSP levels in shellfish a few days in advance of southwest New Brunswick.

Scotia-Fundy Region has started a pilot phytoplankton monitoring project in southwest Nova Scotia. The techniques used are those developed in the Gulf Region. They are currently monitoring water samples from 11 sites. Recently, Inspection staff from Halifax together with Gulf Region DFO personnel, carried out a one day seminar for local industry on the project. The local mussel industry is actively involved in collecting the water samples in several of the sites. This participation is based, in part, on interest expressed at the Second Canadian Workshop on Harmful Marine Algae held last October in Dartmouth, N.S. Over 300 water samples are expected to be analyzed from the 11 sites. In addition, 360 shellfish samples are going to be collected from the same locations to compare phytoplankton numbers with toxic levels in the shellfish. Nearly 630 routine PSP samples are slated to be taken in southwest and eastern Nova Scotia.

Québec's industry is primarily in clams and mussels. Mussel aquaculture there is evenly distributed. There are approximately 1600 PSP samples done annually, with a small proportion of them also being analyzed for ASP. As in southwestern New Brunswick, Québec Region has had close ties to DFO Science Branch at Mont Joli. Inspection staff have been kept informed on the status of phycotoxin producing species in their routine phytoplankton sampling.

To summarize, Inspection Services continues to use the mouse bioassay for PSP and HPLC for ASP, respectively, as the main monitoring tools for the shellfish program. It is not anticipated that the pilot phytoplankton monitoring programs being carried out in the Gulf Region and, to a lesser extent, in Scotia-Fundy Region will become a permanent monitoring tool for the shellfish program at this time.

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TOXIC AND OTHERWISE HARMFUL MARINE ALGAL BLOOMS: A CANADIAN EAST COAST PERSPECTIVE[1]

J. Worms, Department of Fisheries and Oceans, Science Branch, Gulf Region, Moncton, N.B.

In describing the evolution of marine phycotoxin related research on the Canadian east coast, one has to refer to the pre- an post-1987 'mussel crisis' eras. The discovery of domoic acid, a novel phycotoxin from the diatom *Nitzschia pungens* f. *multiseries*, in December of 1987, was the signal for a dramatic increase in the amount of research dedicated to studying numerous aspects of natural toxins produced by phytoplankton.

The situation prior to December 1987

Up to December 1987, paralytic shellfish poisoning (PSP) was the only marine phycotoxin documented and recognized in Atlantic Canadian coastal waters. The problem has long been identified in the St. Lawrence River estuary and in the Bay of Fundy, and programs to monitor shellfish toxicity have been in place since 1943 to prevent consumption of tainted molluscs. Research programs were targeted mainly at the dynamics of *Alexandrium tamarense* and *A. fundyense* populations, including cyst population surveys, in order to improve predictability of dinoflagellate blooms. Efforts had also gone into improving analytical techniques for the detection and quantification of PSP toxins. The relative geographic stability of the PSP pattern of occurrence and the effectiveness of the shellfish monitoring program in protecting consumers' health did not provide the necessary incentive for increased research efforts.

The December 1987 'mussel crisis'

In mid-November 1987, several consumer complaints related to food poisoning were traced to cultured mussels from eastern Prince Edward Island. The food poisoning episode subsequently reached alarming proportions and was shown to be caused by domoic acid. This unfortunate episode showed the limitations of the existing shellfish monitoring program. It was clear that, although no one could have predicted or prevented the occurrence of a novel phycotoxin, limiting the shellfish monitoring program to those areas with a documented PSP history was not sufficient. Another clear message was that the problem posed by marine phycotoxins was far more complex than previously thought, especially in view of the increasing use of estuaries and embayments by the rapidly-developing shellfish aquaculture industry. The crisis, because of its far-reaching human health implications and resulting public exposure, eventually helped sensitize higher levels of Department of Fisheries and Oceans (DFO) Science and Inspection management and prompted the establishment of long-term, coordinated research and monitoring programs.

The post-crisis situation

During the crisis, a multi-agency Analytical Working Group (AWG) was assembled to coordinate the work of the many research laboratories involved in the search for the toxin. Beyond this immediate pressing target, the need for a long-term expanded research program rapidly became obvious. New resources were therefore requested from Treasury Board to boost capabilities in Québec, Gulf and Scotia-Fundy Regions, those most impacted by the crisis. Based on a preliminary list of research priorities established by AWG in early 1988, new projects were started. This working group was disbanded in the aftermath of the crisis, but the concept of a coordinating body was retained by DFO Science, resulting in the creation of the DFO Phycotoxins Working Group (PWG) in early 1988. Since then, PWG's main mandate has been to ensure that all research needs were adequately covered

[1] Summary of a presentation made at the Workshop on Pacific Coast Research on Toxic Marine Algae. Copies of the full text, including 36 references, are available from the author.

with no undue duplication of effort. Research projects of DFO and, to a large extent those of other institutions, are organized around several major themes as follows:

1. **Chemical methodology**: Development and implementation of bioassays and analytical techniques to detect and quantify marine phycotoxins.

2. **Phytoplankton population dynamics**: Large-scale monitoring of phytoplankton populations with special emphasis on harmful blooms and their controlling factors. These projects rely heavily on physical and chemical oceanographic support.

3. **Toxin uptake and depuration by shellfish**: Field and laboratory studies of toxin uptake, storage and release by commercially exploited wild and cultured molluscan shellfish.

4. **Biological and biochemical aspects of toxin production**: Mechanisms of toxin production, including biosynthesis pathways, environmental and physiological control factors, and the role of bacteria in toxin production.

5. **Aquatic toxicology**: Transfer routes in the food web and impact of toxins on marine animals, including zooplankton, ichthyoplankton, and juvenile and adult fish.

6. **Fate of phycotoxins**: Role of bacteria in the biodegradation of toxins released into the water column.

During the first two years of operation of the new program, much emphasis was placed on domoic acid (amnesic shellfish poisoning or ASP) and *Nitzschia pungens*. Research was directed towards analytical, ecological (field) and physiological (laboratory) aspects, partly in support of regulatory activities. Coordination between Science and Inspection Services is especially critical to ensure optimum efficiency and cost-effectiveness of toxicity monitoring programs. While Inspection Services have a mandate of safeguarding the health of consumers against phycotoxins and other types of intoxications, it is the role of Science Branch to provide Inspection Services with tools that will make its control programs more effective.

In September 1989, the first Canadian Workshop on Harmful Marine Algae was held in Moncton, N.B. This was the first in a series of annual workshops sponsored by DFO to provide scientists a forum to present recent findings and exchange ideas. A second workshop took place at the Bedford Institute of Oceanography in Dartmouth, N.S., in October 1990. From the presentations and discussions held during these workshops, PWG drafts a series of recommendations reflecting important issues and concerns identified by participants and requiring more attention. These recommendations are used as guidelines during the regional planning process to ensure that no critical research issue is left unattended. Most of the first workshop was dedicated to ASP while the second workshop featured a mix of presentations on ASP, DSP and PSP. Although held on the east coast, this series of workshops and PWG activities are designed to serve all Canadian regions facing phycotoxin problems and could benefit west coast as well as east coast scientists.

Future directions

Over the last few years, the concern has been repeatedly raised that occurrences of toxic and otherwise harmful phytoplankton blooms have increased globally for reasons which are still debated. It is clear, however, that more such outbreaks are reported from more geographic areas, involving more species from more classes and that there is an ever increasing list of toxins. Whatever the reasons for this perception, the problem of toxin-producing and otherwise harmful algae has become a world-wide phenomenon, reaching a scale in Canada which is now of major concern to both cultured and wild fisheries, and to human health. There is a need to consider the occurrences more comprehensively than at present, in the context of a large-scale environmental/biological/fisheries issue. After the preceding few years of intensive, highly targeted research, the Canadian marine phycotoxins research program needs to broaden its scope to account for the magnitude of the problem.

Based on information from Canadian and American sources, and comparable records from western European countries, the following comments are offered:

1. The phenomenon of plankton blooms having harmful effects is no longer confined to a handful of dinoflagellate species, but spans over at least 6 classes: Haptophyceae (*Chrysochromulina*, *Prymnesium*, *Phaeocystis*), Cyanophyceae (*Lyngbya, Aphanizomenon, Nodularia*), Bacillariophyceae (*Nitzschia*, *Amphora*), Dinophyceae (*Alexandrium*, *Gyrodinium*, *Dinophysis*, *Prorocentrum*), Raphidophyceae (*Heterosigma*) and Chrysophyceae (*Aureococcus*), which includes some very ubiquitous species.

2. In addition to the toxicological effects on humans, it is now clear that fish and shellfish populations can also be negatively affected, through either lethal toxicity, or physiological or mechanical damage.

3. Animal species which have not previously been reported to accumulate toxins, either in their whole body or in specific edible parts, have been shown to accumulate amounts of toxin(s) significant enough to create a human health concern.

4. The occurrence of toxic and otherwise harmful planktonic, benthic and epiphytic algae and possibly bacteria is not confined to inshore areas but also extends well into the oceanic environment.

It is therefore evident that the problems posed by toxin-producing and otherwise harmful algae are no longer minor or occasional. The problems encountered require a much broader approach than that of simply developing or improving detection methods to manage crises, or of adjusting monitoring systems to ensure consumer safety. The time has come to take a broader and more in-depth look at this issue to determine whether we are, in fact, seeing the early manifestations of a larger environmental problem. Are changes occurring as a consequence of human activities: e.g. via modifications of environmental variables and nutrient availability, modification of competition/exclusion schemes due to pollution, introduction of new species/strains through transport and release of ballast water, or new and/or increased use of coastal areas? Or are these changes the result of large-scale hydrographic variation, or climate change? Are the combined effects of several or all of the above creating conditions favourable to the spreading of known harmful algae and the emergence of new ones, resulting in a hazard to inshore marine populations and human consumers and prompting for a serious reevaluation of the uses of the coastal environment? These and other related questions are the challenge we have to face in the years to come. A background document for discussion has recently been circulated among DFO Science Branch top managers explaining and documenting the increased concern over harmful blooms and outlining a proposed course of action for the immediate future.

This revised approach in no way negates previous and ongoing research on more targeted subjects. This work is essential to elucidate specific aspects of the dynamics, physiology, biochemistry and toxicology of a given toxin-producing species in a given environment. Results from such research, merged with information from other disciplines, will help to draw a more global picture of the phenomenon of harmful blooms, and, it is hoped, better our capacity to predict, mitigate and eventually prevent the occurrence of such events. Strengthening marine phycotoxin research on the west coast of Canada and merging the DFO component with its east coast counterpart makes this program truly pan-Canadian as originally intended, and constitutes a first, logical step towards better integration of research programs at a global scale.

AIRBORNE REMOTE SENSING OF RED TIDES

G. Borstad, G.A. Borstad Associates Ltd., Sidney, B.C.

In September 1990 G.A. Borstad Associates Ltd. of Sidney, B.C., carried out an aerial survey of the *Gonyaulax spinifera* bloom off Vancouver Island on contract for the Department of Fisheries and Oceans, using its Compact Airborne Spectrographic Imager (CASI). This work was described in a workshop talk by Jim Gower of I.O.S., and presented in a poster by the company.

Borstad Associates has been developing water colour surveillance methods since 1978, and their CASI represents the international state-of-the-art. The small instrument can be mounted in a single engine float aircraft and operated by one person. A real-time display, flexibility of instrument operating parameters and the company's custom designed data processing software allow rapid quantitative image measurements of phytoplankton concentration over large areas.

Fig. 1 and 2 show examples of the spectral and image output products of Borstad's CASI system. Spectra are used primarily for research and determining optimal configuration for the instrument's imaging mode. The image shown here illustrates surface phytoplankton patterns in the vicinity of an aquaculture site in Vernon Bay, Barkley Sound, on September 4, 1990.

Fig. 1: see Appendix 1: Colour plates (p. 69)

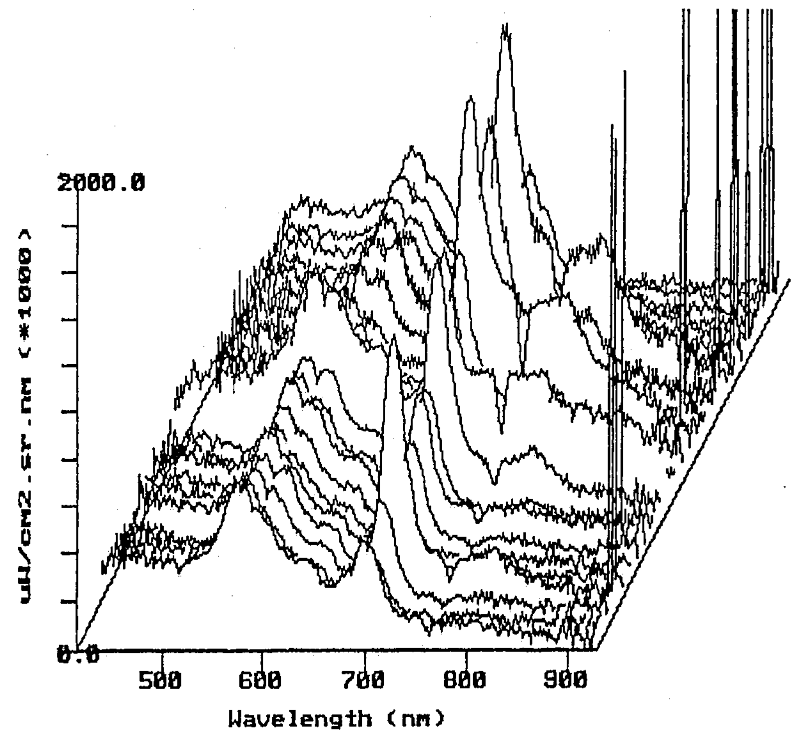

Fig. 2 Example of a sequence of individual spectra acquired by CASI from a transect heading north along the west coast of Vancouver Island. The z axis is the along-track time (or distance) axis. The sharp spikes at 925nm are spurious effects caused by the present calibration software where very low signals are encountered.

SEA SURFACE TEMPERATURE OBSERVATIONS OFF VANCOUVER ISLAND

H. Freeland, Department of Fisheries and Oceans, Institute of Ocean Sciences, Sidney, B.C.

1990 Observations at Amphitrite Point

Sea surface temperatures at Amphitrite Point are reported daily to the Institute of Ocean Sciences. Figure 1 shows a plot of daily observations from 1 May 1990 to 1 May 1991. The smooth line represents the annual cycle as determined by averaging about 56 years of observations. The shaded areas represent periods when the temperature observations are warmer or colder than normal. This plot clearly shows that there was a high temperature event that affected the coast of Vancouver Island during the summer of 1990, and is roughly coincident with the period of the red tide seen that summer. The temperature peak anomaly occurred around 4 September, which is roughly the peak of the red tide event. At that time temperatures were about 2° C above normal; that is a very large anomaly at Amphitrite Point. The monthly mean SSTs for August and September 1990 were both about 2 standard deviations above the mean, thus this represents a rare event.

Arrangements have been made to receive the observations from Amphitrite Point in near real time. Thus if a similar event should occur in the future it should be possible to provide some warning based on the occurrence of large temperature anomalies.

What does the future hold?

In Figure 2 we compare the SST observations at Race Rocks with averaged northern hemisphere temperatures as compiled by Jones *et al.* (Jones *et al.* 1986, Jones 1988). The Northern Hemisphere time series is much longer than our local SST time series, so these are referenced to different time periods for the computation of the mean. The latter time series shows a well established warming trend of about 0.4° C/century. The warming trend is not, however, the same for all seasons. August temperatures are rising only at the rate of 0.2° /century, whereas December temperatures are rising at 0.8C° /century. Though the time bases for estimation of the mean are different, it is clear that the local SST time series shows a large degree of coherence with the Northern Hemisphere average time series (which is derived from land observations only). Thus we might reasonably expect that the local SSTs will share in the large scale climate warming that is observed. The local time series of SST are all relatively short. (The series above from Race Rock was chosen because it is one of the longest series.) However, the local SST time series all show a systematic warming trend of about 1° C/century that is marginally significant at the 95% confidence level, see Freeland (1990). If this steady climate amelioration is associated with the increased atmospheric burden of radiatively active gases, then we can expect that amelioration to continue and the probability of seeing west coast SSTs above 14° C will increase. Similarly, the probability of a red tide might then be expected to increase in the future.

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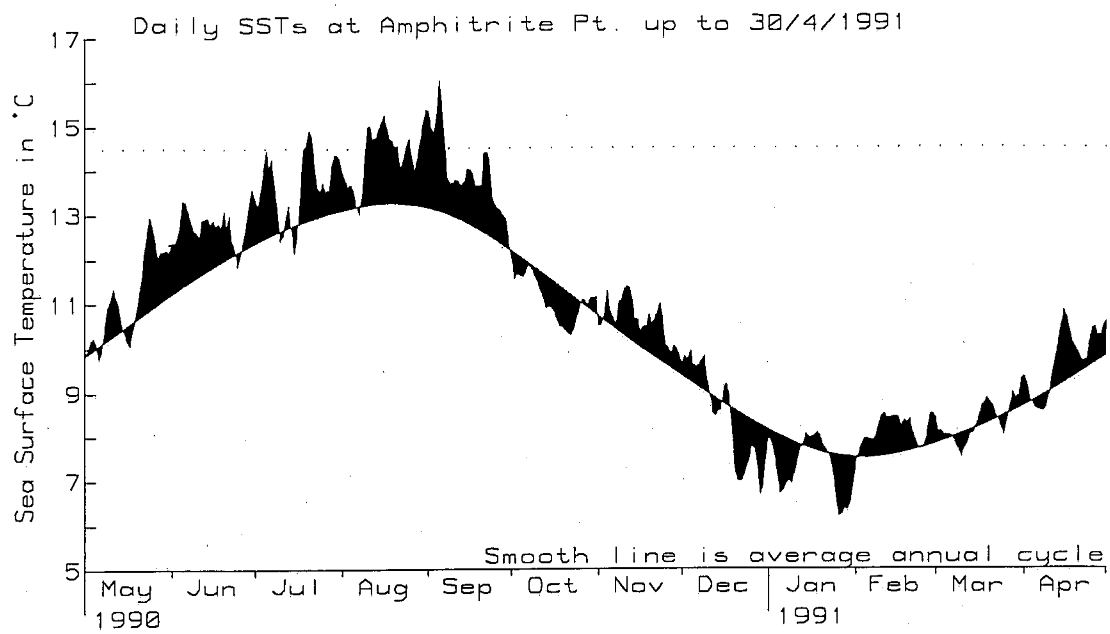

Fig. 1 Daily sea surface temperature observations at Amphitrite Point from 1 May 1990 to 1 May 1991. The smooth line represents the annual cycle (56 years observation).

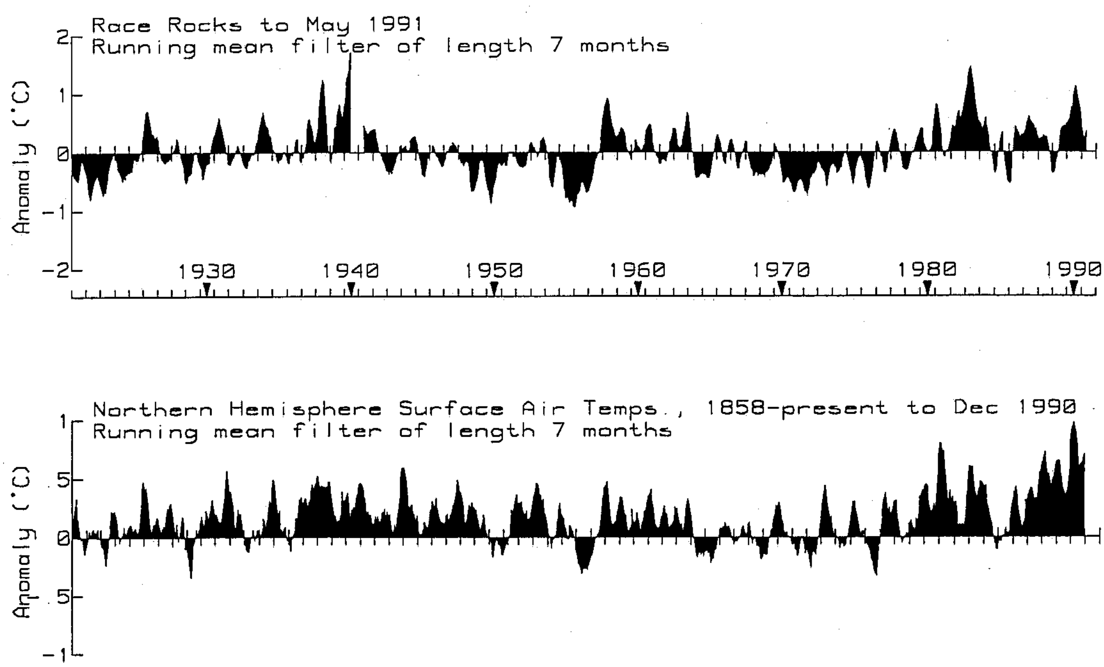

Fig. 2 Sea surface temperature anomalies at Race Rocks (upper panel) compared with Northern hemisphere air temperature anomalies (lower panel)

NOXIOUS PHYTOPLANKTON BLOOMS AND MARINE SALMON CULTURE IN PUGET SOUND, WASHINGTON

R.A. Horner and J.R. Postel, School of Oceanography, and
J.E. Rensel, School of Fisheries, University of Washington

Introduction

Worldwide, the occurrence of noxious and toxic phytoplankton blooms is apparently increasing (Anderson 1989; Smayda 1989), but it is not clear if this represents actual increases in toxic species or increased awareness and reporting. While blooms of toxic dinoflagellates and paralytic shellfish poisoning have long been an issue in Puget Sound waters, problems associated with algal blooms that kill pen-reared salmonids are relatively new (Rensel *et al.* 1989; Horner et al. 1990 and references therein) and a number of phytoplankton species have been implicated. Recent research on the problem in British Columbia and Alaska has been limited to laboratory bioassays with Pacific salmon, but evidence from Washington suggests that Atlantic salmon, the species of commercial importance in Washington waters, may be more susceptible to phytoplankton problems than Pacific salmon species. However, adequate, rapid histopathological and hematological examinations of damaged gill tissue, blood, and body tissues have not been accomplished and the precise cause of fish mortality, e.g., mechanical injury of gill tissue or toxic poisoning, is not known.

Local History

In Washington State, 14 net-pen farms raise Atlantic (*Salmo salar*) or Pacific (*Oncorhynchus* spp.) salmon. Current production is estimated to be more than 5 million pounds annually. In the last three years, phytoplankton blooms have been involved in the mortality of more than two million fish, with monetary losses estimated near $10 million. Fish growers consider phytoplankton-related problems to be a high research priority because they threaten current production and increase the risk and expense related to development of new sites. The problem is compounded because the seasonal, geographic, and vertical distributions of the problem species are not known.

In Puget Sound, fish distress or death is often the first indication of the high abundance of the diatom, *Chaetoceros convolutus* Castracane. However, a similar species, *C. concavicornis* Mangin, has been more common in Puget Sound recently and is the species being used in our experimental work. *Chaetoceros*, however, has not been a major problem in Puget Sound recently.

Instead, after being a serious problem for fish growers in British Columbia waters for several years, the flagellate *Heterosigma akashiwo* (Hada) Hada, appeared in bloom concentrations in western Washington waters in autumn 1989. Although it was apparently present in low numbers in mid-August (our unpublished data), *H. akashiwo* was first visually apparent as widespread and intense surface patches in early September. It rapidly moved toward the salmon pens at Cypress Island, leaving growers relatively little time to harvest their fish. Three farms (four net-pen sites) lost about 95% of their stock, resulting in about $4 million of insurance claims.

In early July, 1990, another bloom of *H. akashiwo* hit fish farms in central Puget Sound, with cell numbers $>3\text{-}18 \times 10^6$ cells.L^{-1}. One farm lost 70-80% of its fish, while two other farms, that were able to move their pens out of the bloom area, lost 10-20%. All told, about 1.27 million fish, valued at $4-5 million, died. This bloom extended to Cypress Island, Port Townsend, and Port Angeles, as well, but growers in those areas experienced minimal losses. We know of only two possible incidents of fish kills caused by this organism in Washington waters prior to 1989: at Lummi Island in 1976 and at Manchester about the same time.

Currently, the only mitigation procedures for any noxious blooms involve moving the pens away from the algae, reducing exposure to surface water containing the bloom with the use of plastic skirts and using airlift pumps for upwelling of algal-free water, or reducing fish activity and respiration by withholding food. Harvesting of fish after exposure to what may be toxic blooms is controversial because the mechanism(s) by which the fish are killed are not known.

Research Project

Our project involves 1) laboratory and field investigations on the causes of fish deaths and ecological characteristics of problem-causing phytoplankton species; 2) field studies of environmental factors accompanying blooms; and 3) field studies of the vertical distribution of problem species and the effectiveness of mitigation techniques.

Our objectives are three-fold:

1. In the laboratory, we are determining the causes of phytoplankton-induced fish mortality and characterizing ecological requirements for some of the problem-causing species.

2. In the field, we are characterizing the phytoplankton assemblage seasonally and annually at several salmon net-pen sites, focusing on vertical distribution of noxious species and hydrographic parameters, and developing rapid monitoring methods for noxious phytoplankton, as well as possible mitigation techniques.

3. We are preparing a guide for the identification of phytoplankton in Puget Sound, but also including methods for the collection of phytoplankton and environmental data and suggestions for mitigation procedures.

Our project is working on two levels. In the laboratory, a pre-doctoral student is experimenting with cultured and natural phytoplankton populations, especially *Chaetoceros concavicornis*, to try to determine if mortality is caused by physical (gill blockage) or chemical (toxin) means. He is using live *Chaetoceros* cells and cell-free filtrate water. Fish are monitored for visible changes in behavior and changes of blood gases (CO_2 and O_2). Gill tissues are being examined for gross morphology, and gills and other organs collected for more rigorous histopathological examination. The work also includes assessing the physiological requirements of *Chaetoceros* spp.

In the field, we are working at net-pen sites with several fish growers to learn more about the species present and the ecological factors that affect local phytoplankton and phytoplankton blooms. Technicians at four fish farms collect samples from several depths and identify the phytoplankton species every day during spring and summer. They are looking primarily for *Chaetoceros* and *Heterosigma*. Once each week they collect phytoplankton samples for us to analyze. Samples are also taken for nutrient and chlorophyll determinations that are analyzed at the University of Washington. Each farm area has an irradiance meter to determine incoming solar radiation. Samples from 1990 have been analyzed and we are now working to correlate the data. This information will allow us to characterize the phytoplankton communities at several farm sites during crisis and non-crisis periods, while focusing on the vertical distribution of noxious species.

In the future, we plan to direct much of our effort to learning more about the biology of *H. akashiwo* because of the recent blooms in Puget Sound and the massive fish kills. This project is sponsored by the Washington Sea Grant Program with Karl Banse and Frieda Taub as Principal Investigators.

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THE PHYTOPLANKTON WATCH PROGRAM IN BRITISH COLUMBIA

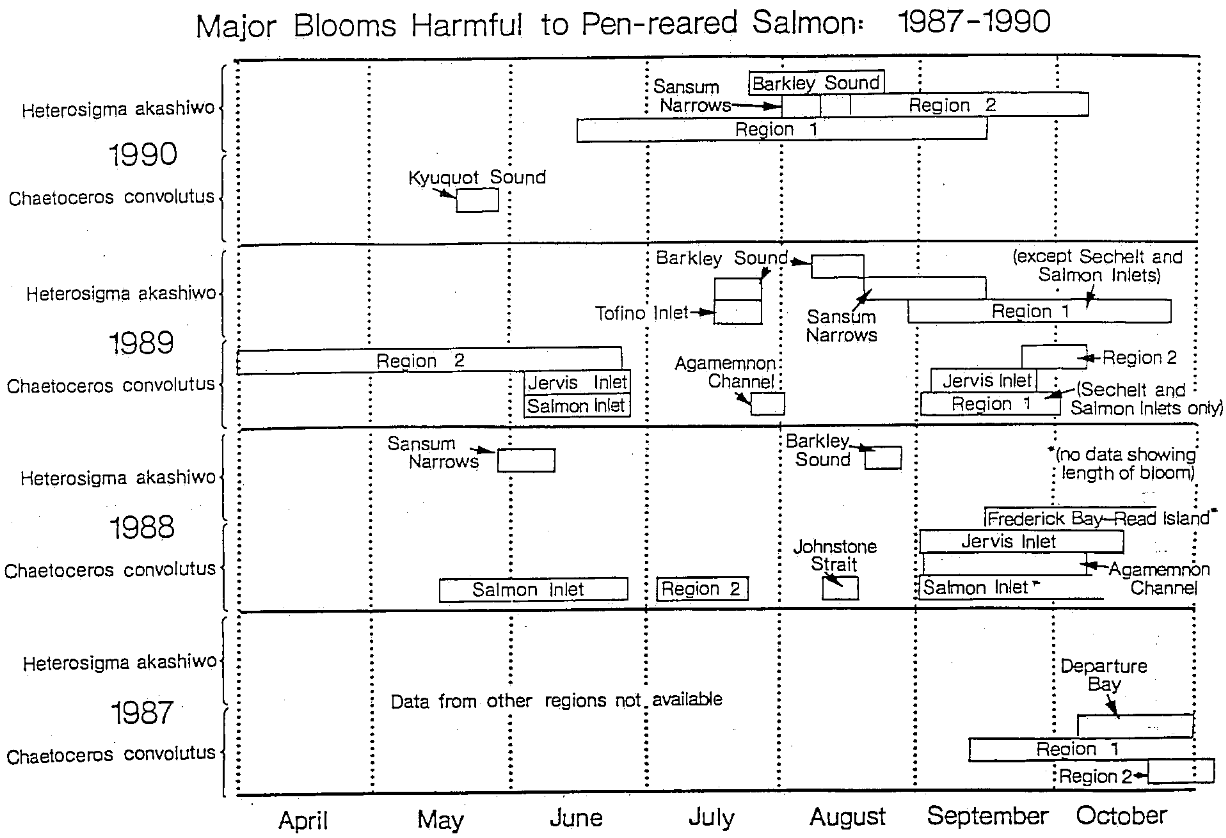
E. Stockner, Eco-Logic Ltd., West Vancouver, B.C.

In the summer of 1986 after a massive *Heterosigma akashiwo* bloom with heavy fish mortalities, the Phytoplankton Watch Program (PWP) was developed to identify and 'watch' for harmful phytoplankton species. The need for more plankton information, better sampling methods and improved communication was deemed essential for the growth and development of the aquaculture industry in British Columbia. Since the program's inception the level of awareness in the industry to the benefits of monitoring the water for harmful phytoplankton has greatly increased. The harmful species in coastal waters, e.g. *Chaetoceros convolutus*, *C. concavicornis* and *Heterosigma akashiwo* are being monitored at most all sites. Species that have caused closures in the shellfish industry, e.g. *Protogonyaulax* (= *Alexandrium*) *catenella*, *P. tamarensis* and *Dinophysis* spp., are also being watched, together with information on some important physical and chemical variables (temperature, oxygen, salinity, water transparency). The need to manage finfish and shellfish stocks through times of intense phytoplankton blooms has become synonymous with the survival of the aquaculture industry. Major blooms that have caused industry losses in British Columbia waters between 1987 and 1990 are illustrated in Fig. 1.

The program focuses on education, data gathering, communication and standards. Workshops have been an important element of the PWP with on-site instruction of sampling techniques, microscopic enumeration methods and species identifications. Lectures and workshops given by the coordinator are also an important component of the PWP's education package. Each data farm or site selected to participate in the program collects weekly samples and records physico-chemical conditions at the time of sampling and sends this to the Program Coordinator in West Vancouver, where a quantitative count is completed and results added to a computer database. Phytoplankton samples are also examined on the farms for their immediate use and for information transfer to a 1-800 telephone line[1]. Close communication is maintained with the data farms to help farm staff properly identify phytoplankton species, carry out required sampling and counting procedures and to supply information to assist them with potential problems.

One of the principal duties of the PWP coordinator is to alert farms of impending harmful bloom situations. The toll-free 800 line together with direct phone calls to each farm are the main links of communication to the industry. By placing current information obtained from the farms on the message tape of the 800 line answering machine the coordinator is able to keep the aquaculture industry up-to-date on current phytoplankton conditions. The 800 line is well used by farms, especially during bloom periods. Data farms are also asked to communicate information from other farms in their area, particularly when a harmful bloom is seen. The convenience of a toll-free line and a message that is available 24-hours a day is a vital component of the communication segment of the program. Finally, the PWP has endeavored to achieve uniform sampling and counting methods throughout the industry. The cell density of certain diatoms can seriously affect salmon, so it is necessary to enumerate cell density per unit volume. When reporting counts, results are only useful if compatible quantitative methods have been used. Reliable comparisons of phytoplankton and water quality results among farms and sites can only be accomplished by application of standard procedures.

[1] 1 800 663 2713 (April - September inclusive)



Region 1: Sunshine Coast, Region 2: Campbell River/Desolation Sound

Fig. 1 Major blooms harmful to penned-reared salmon, 1987 - 1990.

DISCUSSION AND CONCLUSIONS

T. Tebb and J.R. Forbes, Department of Fisheries and Oceans

The workshop brought together diverse participants working in the field of research on toxic marine algae in the Pacific region. We hope that it will lead to increased cooperation among various agencies. In particular, new lines of communication with individuals in universities and the Province of B.C. will place DFO in a good position to initiate research activities with a longer-term focus than the current, frequently reactive projects that it undertakes, while remaining within fairly stringent funding controls. A number of new activities may develop. These include implementing a mechanism for documentation of serious blooms (species, magnitude, location), to permit an assessment of long-term changes in frequency, size and range of blooms; and research on bloom dynamics at selected locations in diverse environments along the coast, to establish the physical, chemical and biological factors contributing to outbreaks of monospecific blooms and controlling the particular species responsible.

Participation in the workshop by industry representatives was less than anticipated, and we recognize that a better mechanism for consultation on industry requirements needs to be implemented.

Discussions at the conclusion of the workshop reflected the broad concerns of the participants and identified numerous areas where new research could address problems known to be encountered by industry. Max Taylor provided a long list of questions (Table 1) that need to be resolved, stimulating discussion on some specific topics.

Does the presence of fish farms result in increased frequency of algal blooms? The consensus was that research to date indicates that nutrient input from farms has only a minor impact on algal production. Some of the current research in the Bay of Fundy in eastern Canada is aimed at establishing how much the industry can grow before causing eutrophication problems.

Is eutrophication of coastal waters resulting in increased frequency and magnitude of monospecific algal blooms? It was reported that an International Oceanographic Commission committee examining this question has concluded that eutrophication does appear to generate increasing frequency and distribution of blooms in some cases, but not in others. This may be a result of the differing proportions of various nutrients, as well as overall nutrient loading (Smayda 1990). In this region, it was suggested that the Fraser River plume may act as a natural nutrient pump. Additional loading appears fairly low, although it is not clear how much impact overall sewage loading, particularly on the south coast, is having.

There was little discussion of the issue frequently raised by industry that new analytical methods for algal toxins need to be brought onstream (see e.g. Spence 1991). The objective would be both to boost the capacity for official inspection programs, allowing for faster turnaround, and to allow self-monitoring, resulting in increased flexibility in management and harvesting strategies.

Larry Albright suggested that one approach to dealing with fish farm losses might be to examine the potential for bio-engineering in either the fish or feed to reduce susceptibility to toxins. It was noted that study of the sources of variability in resistance exhibited by different salmon species might be profitable.

What is the best way to approach these problems? Priorities need to be set. Should the focus be on specific industry and public health issues or should it be oriented to longer-term, open-ended research? Jean Worms noted that, on the east coast, industry and DFO inspection are involved in monitoring species composition and abundance. This provides immediate feedback for industry to take ameliorative action and DFO to initiate

TABLE 1. Outstanding questions for the B.C. coast. A list prepared by Max Taylor.

Are there areas where harmful species do not occur or form blooms?

Will there be regional sorting of farm species based on differential sensitivity to prevalent blooms?

Is the frequency of red tides increasing as in other parts of the world where eutrophication has become prevalent?

Do *Protogonyaulax* blooms originate offshore as in the Atlantic?

Do *Protogonyaulax* blooms exhibit a 7-year cycle? If the blooms are cyclical what is the driving mechanism?

Are cysts of *Protogonyaulax* responsible for the winter toxicity in shellfish on the northern B.C. coast or is this solely the result of longer depuration times in colder water?

Where are the seed beds for species such as *Protogonyaulax* and *Heterosigma*, and what are the expected advection patterns?

What are the limiting micronutrients (metals, vitamins, ...) for *Heterosigma akashiwo* in B.C. waters?

Do blooms of *Heterosigma* always follow diatom blooms? Are diatom blooms somehow necessary for its blooming (e.g. by conditioning the water with exudates, by stripping the upper euphotic layer of nutrients, ...)?

Does DSP occur on this coast and, if so, how much of a threat do the *Dinophysis* species pose?

Which species of *Nitzschia* produce toxins? Do other species of pennate diatoms produce toxins?

Are *Chaetoceros concavicorne* and *C. convolutum* different species or different morphs of the same species? Is the former responsible for most of the fish deaths?

What are the environmental factors which promote the formation of large spines in *C. concavicorne*?

Can B.C. experience harmful *Chrysochromulina* blooms as in Scandinavia?

Is the askeletal form of *Dictyocha* a problem in B.C. waters? Is silicon metabolism somehow tied to toxin production?

Are there regions where phosphate is the main limiting nutrient? Would we find greater toxicity in these regions?

Are there further harmful species?

Are harmful species being introduced in ballast water?

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harvesting closures as necessary. Other organizations (DFO Science, N.R.C, universities) are involved in long-term programs to improve understanding of problems relating to the development of blooms. On the west coast, the Plankton Watch in B.C. and a similar program in Washington State operate monitoring programs, with fish farms doing basic identification and abundance estimates, and Eco-Logics Ltd. (in B.C.) and the

University of Washington providing periodic (generally weekly) quality control. These programs both operate telephone information and reporting networks. There has been relatively little ongoing research on the broader issues relating to causes of algal blooms. Max Taylor emphasized that, in his view, the only way to answer many of the problems is to undertake multi-year intensive monitoring: species-level ecology is the key to understanding. However, it was suggested by others that sampling frequency might need to sacrificed to enhance density of coverage over the coast. Taylor observed that most red tides are studied only after they have reached their peak, at which time little useful information can be gleaned. Data needs to be collected on conditions prior to the onset of blooms in order to gain an understanding of the processes leading to them. As an addendum, he also noted that there is a dearth of trained phytoplankton taxonomists, an issue that needs attention if new research is to be initiated.

As noted above, the workshop provided the opportunity to increase communication between agencies. The consensus was that this is critical to dealing with research problems associated with toxic algae. Jean Worms observed that, with various groups proposing approaches that they feel competent to deal with and addressing specific problems of concern to them, points of collaboration will naturally emerge.

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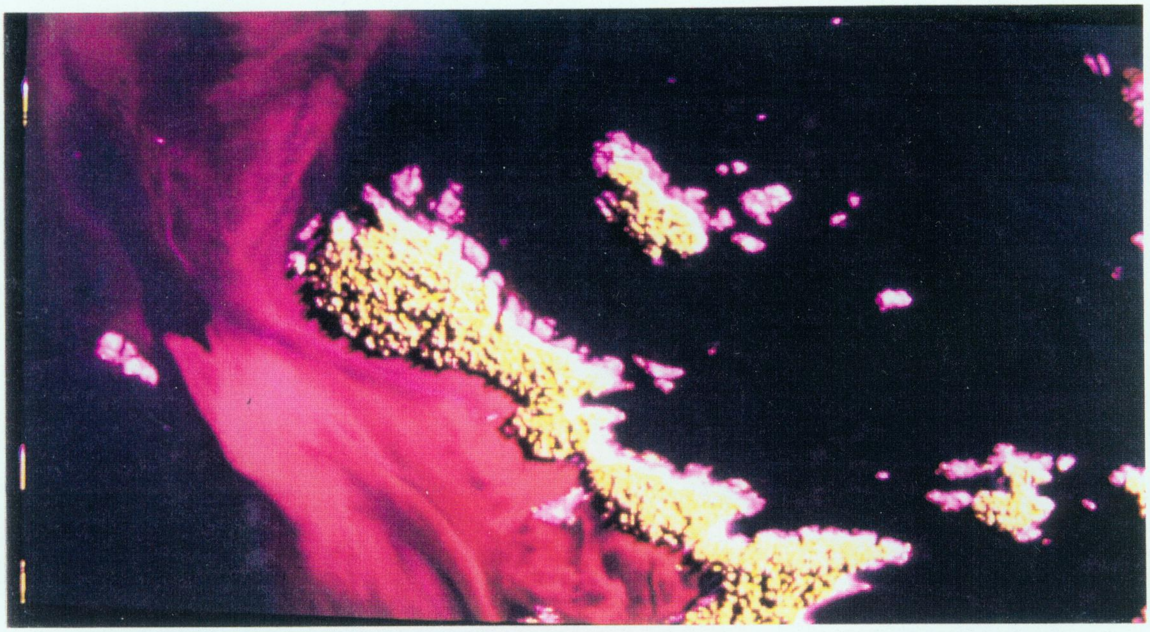
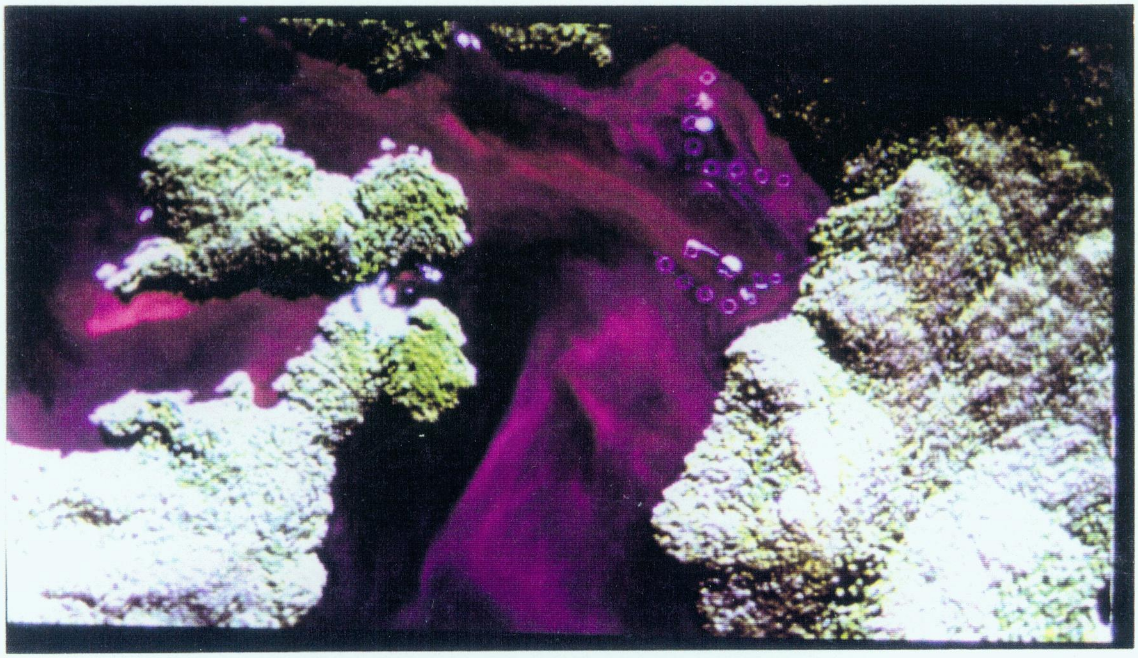
Spence, J. 1991. Aquaculture industry concerns regarding toxic algae, p. 41-42. *In* J.R. Forbes (ed.) Pacific coast research on toxic marine algae, Can. Tech. Rep. Hydrogr. Ocean Sci. 135: 76p. (This report)

Appendix 1: Colour plates



J.F.R. Gower and G. Borstad. The August/September 1990 red tide event on the west coast of Canada: satellite and aircraft observations (p. 11): Fig. 5

A single band (701 - 714nm) image collected with the CASI of a red tide near islands in Barkley Sound on 4 September 1990. The area shown is about 1.2 by 3km and is imaged with a resolution of about 3m.



G. Borstad. Airborne Remote Sensing of Red Tides (p. 55): Fig. 1

Green/blue ratio (600nm / 480 nm) image of phytoplankton distribution in Vernon Bay, inner Barkley Sound. Purple areas are highest ratio and phytoplankton concentration. Data from G.A. Borstad Associates Ltd. CASI flight, 4 September 1990.

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Appendix 2: Workshop agenda

WORKSHOP

PACIFIC COAST RESEARCH ON TOXIC MARINE ALGAE

INSTITUTE OF OCEAN SCIENCES, SIDNEY, B.C.
30 APRIL 1991

ORAL PRESENTATIONS

0900 - 0915 J.C. Davis, Regional Director, Science, Department of Fisheries and Oceans
Introductory Remarks

0915 - 0935 J.N.C. Whyte, Department of Fisheries and Oceans, Nanaimo
Effects of Heterosigma akashiwo on juvenile Chinook salmon

0935 - 0955 L.J. Albright, Simon Fraser University
Harmful Chaetoceros spp.
A. Temporal and spatial distribution along the B.C. coast.
B. Concentrations harmful to penned salmonids.
C. Observations on their lethal mode of action.

0955 - 1015 J. Rensel, School of Fisheries, University of Washington
Harmful effects of Chaetoceros spp. on Atlantic salmon

1015 - 1040 COFFEE

1040 - 1100 J.F.R. Gower, Department of Fisheries and Oceans, Sidney
Remote sensing of a massive red tide off the west coast of Vancouver Island

1100 - 1120 F.J.R. Taylor, University of British Columbia
HARP The harmful algal research project in Sechelt Inlet, 1987-1990

1120 - 1140 E.A. Black, B.C. Ministry of Agriculture, Fisheries and Food
Research on Heterosigma and other toxic algae by the B.C. Ministry of Agriculture Fisheries and Food

1140 - 1200 D.D. Kitts and D. Smith, University of British Columbia
A new approach for identifying PSP toxic shellfish

1200 - 1220 J.R. Forbes, Department of Fisheries and Oceans, Sidney
Advance warning and scaled response planning for toxic blooms

1220 - 1330 LUNCH

1330 - 1340 Submission from J. Spence, B.C. Aquaculture Research & Development Council
Research requirements: the aquaculture industry's viewpoint

1340 - 1400 K. Vautier, United Fishermen and Allied Workers
Research requirements: the harvesting industry's viewpoint

1400 - 1420 R. Chiang, Department of Fisheries and Oceans, Burnaby
Inspection programs: Pacific Region

1420 - 1440 S. Stephen, Department of Fisheries and Oceans, Ottawa
Inspection programs: Atlantic regions

1430 - 1500 COFFEE

1500 - 1520 J. Worms, Department of Fisheries and Oceans, Moncton
Research programs: Atlantic regions

1540 - 1640 **DISCUSSION SESSION: RESEARCH PRIORITIES AND AREAS OF COOPERATION**

Moderator: T.A. Tebb, Department of Fisheries and Oceans, Nanaimo

POSTERS/DISPLAYS

G.A. Borstad Associates
Remote sensing imaging of phytoplankton concentrations and associated phenomena

H. Freeland, Department of Fisheries and Oceans, Sidney
Sea surface temperature anomalies along the west coast

R. Horner, J.R. Postel and J. Rensel, University of Washington
Noxious phytoplankton blooms and marine salmon culture in Puget Sound, Washington

E. Stockner, Eco-Logics Ltd.
The Phytoplankton Watch Program in British Columbia

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