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Analysis of harmful blooms of the dinoflagellate *Heterosigma* at the Pacific Biological Station Mariculture Facility in 1993 and 1997

Introduction

Blooms of the Raphidophyte Heterosigma carterae (formerly known as Heterosigma akashiwo) have caused numerous fish kills in salmon farms along the coast of British Columbia and around the world. In B.C., these blooms have caused a cumulative financial loss estimated at \$30-40 million for fish farms, with losses in 1997 alone estimated to be \$10-20 million (Whyte 1999). These blooms have raised many questions both about the organism itself as well as how to ameliorate effects on fish stocks. Farms usually have little warning of impending blooms and there is at present no way of predicting whether a bloom is toxic or not. However, information gathered during bloom events at salmon farms may lead to an increased understanding of this plankter and possibly give some predictive tools that can be used to prepare for the onset of these blooms. During the summers of 1993 and 1997, blooms of Heterosigma carterae caused heavy losses in the salmon stocks held in netpens at the Experimental Mariculture Facility of the Pacific Biological Station. In this report, we describe some observations which appear to be more systematic than simple chance events, and which may contribute to an understanding of early-warning signs of Heterosigma blooms.

The Experimental Mariculture Facility of the Pacific Biological Station was established in 1974 in Departure Bay adjacent to the Pacific Biological Station. Over its 25 years the facility has experienced periodic plankton blooms which have caused mortalities in stocks of Pacific and Atlantic salmon being held for experiments. Early experiences with lethal blooms at this site were with the diatoms Chaetoceros convolutus and C. concavicornis. However since 1989 we observed a shift to *Heterosigma carterae* as the main causative organism (although Chaetoceros continues to be present) (see Aquaculture Updates #29 and #62). Early instances of Heterosigma blooms caused only minor losses, 2.2% of the stocks in 1989, and 1.1% in 1991. But these were followed by deadly blooms in the summers of 1993 and 1997, with overall stock losses of 78% in 1993 and 75% in 1997.

June2, 1999

The annual plankton monitoring program at the Experimental Mariculture Facility begins with the arrival of the spring diatom bloom (usually at the beginning of March) and continues into the summer (when flagellates are the primary concern) and on through until the fall diatom bloom dies off. Initially the monitoring consists of weekly samples to detect the presence of potentially harmful plankton; once detected, the sampling frequency is increased as the counts rise. Since *Heterosigma* can grow quickly (cells can make from 1 to 5 divisions a day (Honjo 1992)) daily sampling is begun when cell counts reach 500,000 cells per litre (a threshold based on our experience at the site). Water samples are taken from the surface at mid-day (two o'clock) with the resulting count being used for plotting blooms. Samples are taken at mid-day due to the motility of *Heterosigma* (1.0 to 1.3 m per hour) and its diel vertical migration that allows it to reach depths of 10 meters during the night. Samples are taken at a standard location near the centre of our cage-complex using a bucket on a rope, sampling 0.5 m below the surface of the water. When counting Heterosigma a measured subsample is taken from the bucket (1 to 10 cc) and put into a settling chamber, fixed with Lugol's Fixative and allowed to settle (from 1 to 5 hours depending on the volume). The settled sample (or a percentage of the sample) is counted on a Wild M40 inverted microscope at 100X magnification and transformed to a "cells per litre" concentration. Water temperature (degrees C), salinity (parts per thousand, ‰) readings at 1 and 4 m and a Secchi disc reading (m) for turbidity are taken daily at a standard location and time.

Recent blooms

The first incidence of a major bloom of *Heterosigma carterae* occurred in the summer of 1993 with the concentration "spiking" twice to lethal levels (Fig. 1). The first spike occurred on July 25 and reached 7.1 million cells per litre, resulting in the loss of 20.3% of the salmon stocks on site. Sunny weather, no measurable winds and a coincident prolonged period of weak tidal flushing combined to provide ideal conditions for the *Heterosigma* to bloom. Tides during this day fell only 1 m from high

tide at 12:20 PM to low tide at 5:30 PM. then rose 2.1 m to a high at 12 midnight, creating 12 hours of minimal water exchange. Within 2 days the weather turned overcast and daily counts dropped below 1 million cells per litre. The *Heterosigma* persisted below this level for a month before spiking again to 7 million cells per litre on August 25. Weather conditions were again sunny with light winds and again there was the period of low tidal flushing (a high tide dropping 0.5 m to a low then rising 0.6 m to the next high, over a period of 9.4 hours). This second spike killed 70.0% of the remaining fish. Temperature readings during the two spikes were: first spike 16°C at 1 m, 14.9°C at 4 m; second spike, 18.4°C at 1 m and 17.3°C at 4 m: salinities were between 26 and 27‰ for both spikes. All year classes of Atlantic and chinook salmon stocks were effectively wiped out (96% and 98% lost, respectively). Stocks of coho on site fared substantially better, losing 25% (Table 1).

Following three summers essentially free from fish mortality due to plankton blooms, another major bloom of *Heterosigma* occurred in the summer of 1997. Blooms reached lethal levels on four separate occasions and reached concentrations ten times as high as recorded in the 1993 bloom (Fig. 2). The first spike occurred on June 20 with concentrations reaching 90 million cells per litre, resulting in the loss of 19.3% of the salmon stocks on site. Cell-counts dropped down for two months before rising again on August 24 reaching 21 million cells per litre, which killed off 30.0% of the fish. Following a brief period of lower levels, counts rose again, peaking on August 31 at 27 million cells per litre and killing 19.7% of surviving stocks. The final spike came on September 20 with counts of 15.9 million cells per litre and a resulting loss of 37.7% of the fish on hand. Coinciding with each of these spikes was a period of poor tidal

flushing. Weather conditions were calm and overcast or partially overcast for the first three spikes and sunny and calm for the last spike. Water temperatures at 1 m ranged from 15.7°C to 19.8°C over the four lethal spikes, 13.9°C to 18.9°C at 4 m depth. Salinities at these depths ranged from 24.8‰ to 29.0‰ (Fig. 4 and 5). In total, 75% of the fish on site were lost over the summer. By species, the mortalities were Atlantic salmon 75.6%, chinook 73.3% and the one stock of coho 37.7%.

Mortality

An examination of the mortality rates from the two blooms shows that Atlantic and chinook salmon were hit the hardest, while coho salmon fared much better (Table 1). Comparatively lower mortality in the coho stocks is consistent with our observations from previous blooms. In our experience, survival of chinook in plankton blooms tends to be somewhat better than that of Atlantic salmon, however in these two blooms the final totals didn't show a substantive difference. There appears to be a weak "age of fish" effect in the 1993 bloom where later year classes survived better than earlier ones, however this wasn't as apparent in the 1997 bloom. One obvious discrepancy occurs in the unusually high mortalities seen in the 2-year old chinook during the 1997 bloom (93.4%) when compared to the 3-year old chinook (45.1%). This may be due to greater sensitivity of the 2-year-old stock, which was a firstgeneration hybrid of ocean-type (Big Qualicum River) and stream-type (Yukon River) life histories. The chinook stocks on site during previous blooms were nearly all of the Big Qualicum River stock that is common in the B.C. salmon farming industry. It appears that the Yukon-Big Qualicum hybrids had a reduced tolerance to Heterosigma blooms inherited from the Yukon River stock.

Threshold effects

The *Heterosigma* cell counts reached during spikes of the two blooms did not appear to correlate directly with the level of mortality experienced; higher cell-counts did not result in proportionally greater mortality (Fig. 1, 2). In the 1993 bloom, both major spikes peaked at about 7 million cells per litre, but the first spike killed 20.3% of the fish on site, the second killed 70%. Possible reasons for this non-linearity include differences in virulence of the toxic agents accompanying *Heterosigma* blooms, an accessory effect of higher temperature during the second spike, and a cumulative or sensitising effect from the previous exposures. Measurements from the 1997 bloom do not elucidate this question, since the highest cell-count coincided with the highest temperature in the first spike but caused the lowest percent mortality in fish on site. Furthermore the loss of only 19.7% from the third spike does not support the suggestion of a cumulative effect.

Inconsistent virulence of *Heterosigma's* toxicity remains a possibility. Research at the Pacific Biological Station has demonstrated that cultures of *Heterosigma* collected from blooms showing an obvious toxic effect frequently do not remain toxic in the laboratory (Whyte 1999). The pattern of mortality in the 1993 and 1997 blooms is perplexing in that each spike appeared to afflict some fish within a given stock, but seldom killed the majority of the sensitive fish, as evidenced by the subsequent partial mortality a stock would suffer in later spikes. We have experienced *Heterosigma* blooms that caused little or no mortality despite cell-counts in excess of those observed to cause

fish mortality at our site. The nature of *Heterosigma's* toxicity may be more complex than in other lethal phytoplankton and it may also be more intimately linked to short-term variation in the growth environment of the plankter. Death of salmon from blooms of the diatom *Chaetoceros* is not due to release of a toxin but rather to physical damage to gill tissue by its spines.

We noted no husbandry factors associated with the patterns of mortality, such as cage loading densities (range 1-5 kg/m³), diets or recent stock history. There may have been a contributing role from nocturnally depressed oxygen levels (a condition we see during blooms when tidal action is weak), however no oxygen data were collected during these episodes. The inclusion of all fish stocks and the severity of the losses in both blooms suggest that the main agent was the direct toxic effect associated with the plankter itself.

Potential warning signs

(A) Low salinity

Each year Departure Bay experiences a period of low salinity water in June and July when a plume of fresh water largely from Fraser River spring runoff enters the Strait of Georgia (Fig. 3). Salinity at our site drops from 26.3‰ in May to 24.1‰ in June and 24.5‰ in July (long term monthly mean of averaged daily readings at 1 and 4 m). Water quality measurements from the two bloom years showed that our site experienced unusually low salinity water during this period (Fig. 4 and 5). In 1993 salinities were 1.5‰ lower than the longterm average in May and 3.3‰ lower in June; in 1997 the salinities were 1.8‰ lower than average in both June and July. It has been suggested that episodes of high runoff are a possible contributing factor to the observed incidence of *Heterosigma* blooms (Yamochi and Abe

1984). Therefore daily monitoring and watching for any periods of unusually low salinity may be useful as an early warning sign of *Heterosigma* blooms in the summer.

(B) Rapid increase in water clarity

When a *Heterosigma* bloom does occur it seems to be mono-specific, not as a part of a general multi-species bloom like annual spring diatom blooms. It has been suggested that this effect is due to toxins from Heterosigma killing or suppressing competing plankton (Honjo, 1992). Observations from the 1993 and 1997 bloom years showed that water clarity frequently increased dramatically (as seen in the Secchi disc readings) a day or two before the concentration of *Heterosigma* spiked to lethal levels. This effect was seen to occur in four of the six lethal episodes experienced as well as in one of the three episodes where cell counts spiked above 5 million cells per litre without causing mortalities in salmon stocks. The mean Secchi readings for the nine spikes with counts in excess of 5 million cells per litre show a 50% increase in water clarity from four days before a bloom to the two days before the bloom (Fig. 6). Therefore any sudden increase in water clarity during the summer should be noted, as this could be a sign of an imminent increase in Heterosigma concentrations.

Predisposing factors

We consider that our site location provides some natural conditions favourable to *Heterosigma*. We have found cysts (resting stages) of *Heterosigma* in bottom sediments over a 1-km radius, hence seed material for a bloom is likely present whenever conditions become favourable. Departure Bay also has weak tidal flushing (streams average 2-5 cm/sec at mid-tide) and sustained wind or storms are often necessary to mix water layers so as to dissipate a bloom. In general, periods of poor tidal flushing and calm weather should trigger careful monitoring for *Heterosigma*.

Recommendations

What can aquaculturists do to reduce the risk from Heterosigma blooms? The first recommendation would be to choose a site with good water flow and water depth, and if possible, away from any areas known to have a history of *Heterosigma* blooms. If the site is already established, then monitoring of seasonal salinity variations, tidal conditions, plankton composition and abundance and watching for sudden changes in turbidity, may allow for some warning of lethal plankton blooms. When blooms do occur it is advisable to try to minimize any stress on the stocks; discontinue feeding, mortality recovery and stock handling, to allow the fish to seek areas of lower plankton concentration. Some options exist for farms situated in deep-water sites to take advantage of the fact that Heterosigma prefers the top 10 m of the water column (Haigh and Taylor 1990). The use of deep netpens (15 m or more) may allow the fish to stay below the bloom till it dissipates. Other farms have had some success by enclosing their netpens with tarps, then upwelling or pumping water from depth. However, great care must be taken to ensure the intake is below the bloom throughout the vertical migration cycle and that the water being pumped isn't anoxic.

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Table 1. Total cumulative mortality percentages from the summers of 1993 and 1997 by species and age, with the number of netpens of each involved.

		1993 Bloom		1997 Bloom	
Species	Age (yr)	Percent	Number of	Percent	Number of
		mortality	netpens	mortality	netpens
Atlantic	1	-	-	72.2	12
	2	94.7	1	89.5	6
	3	97.2	1	84.7	1
	4	100	1	75.8	1
	Total	96.0	3	75.6	20
Chinook	1	96.4	1	-	-
	2	99.0	2	93.4	1
	3	99.9	5	45.1	1
	4	100	2	-	-
	Total	98.2	10	73.3	2
Coho	1	24.8	2	-	-
	2	25.6	1	-	-
	3	0	-	35.6	1
	Total	25.1	3	35.6	1
All Fish	Total	78.1		75.0	

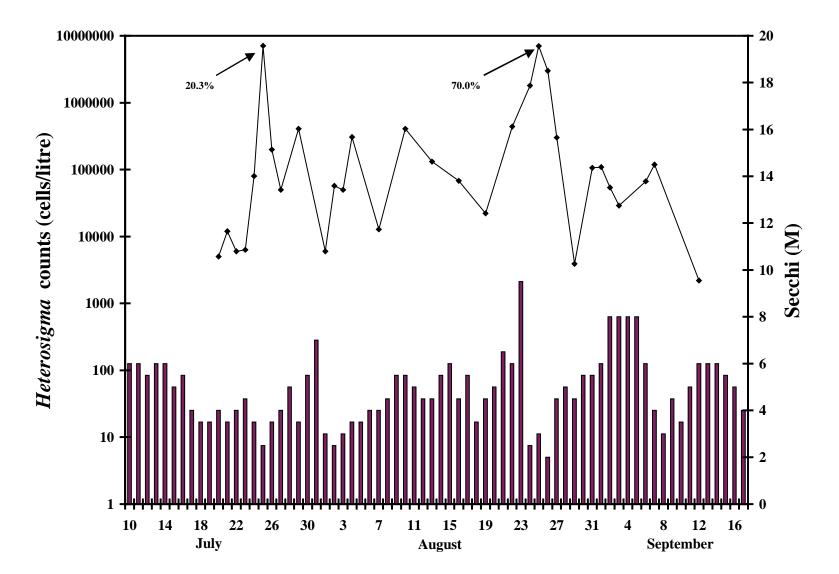


Figure 1. *Heterosigma* levels in cells per litre during the summer of 1993 (line) with daily Secchi Disc readings in metres (bar) (arrows showing percent mortality at each spike).

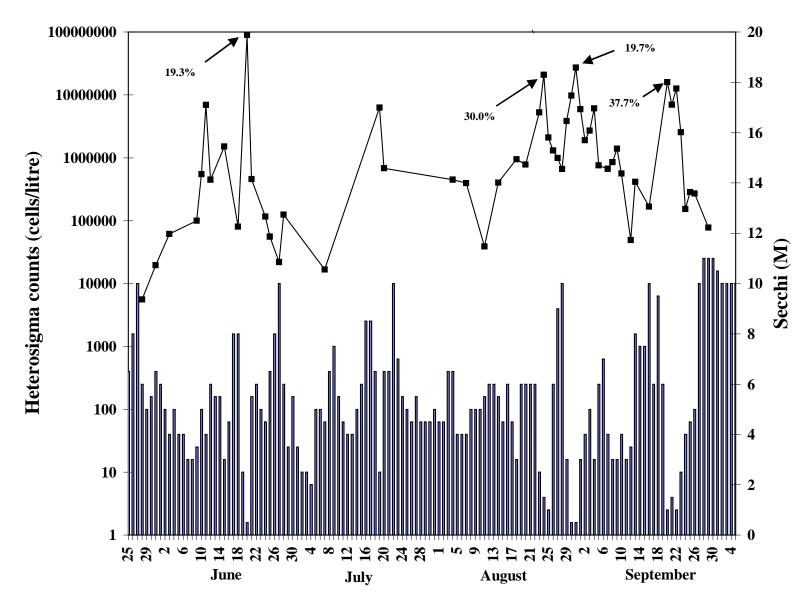


Figure 2. *Heterosigma* levels in cells per litre during the summer of 1997 (line) with daily Secchi Disc readings in metres (bar) (arrows showing percent mortality at each spike).

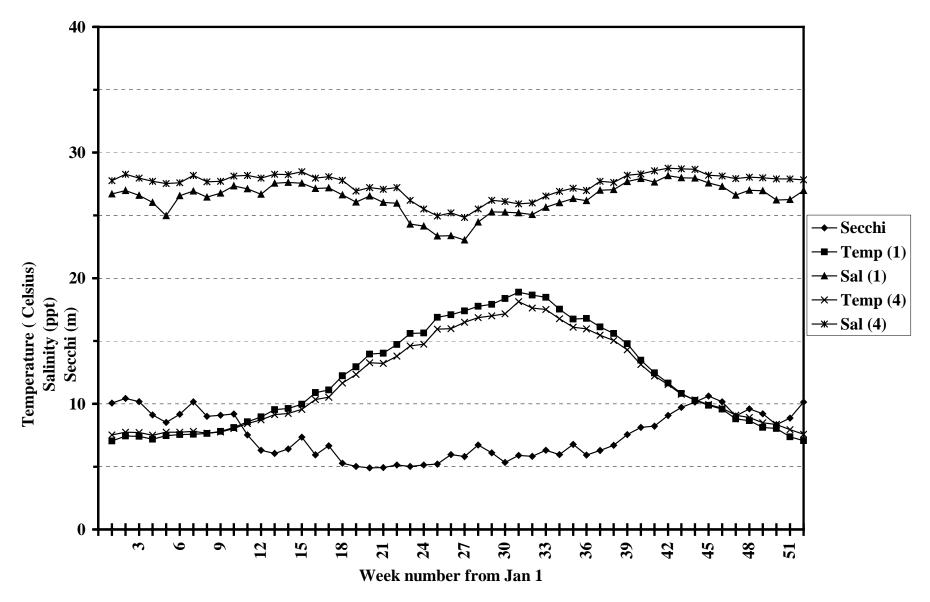


Figure 3. Average weekly water quality measurements for the period 1990 to 1998 at the Experimental Mariculture Facility of the Pacific Biological Station in Departure Bay.

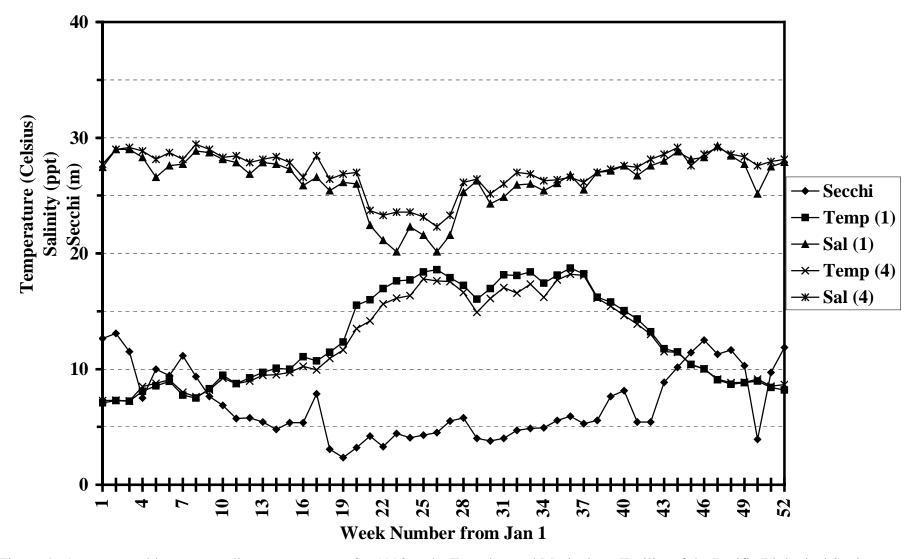


Figure 4. Average weekly water quality measurements for 1993 at the Experimental Mariculture Facility of the Pacific Biological Station in Departure Bay.

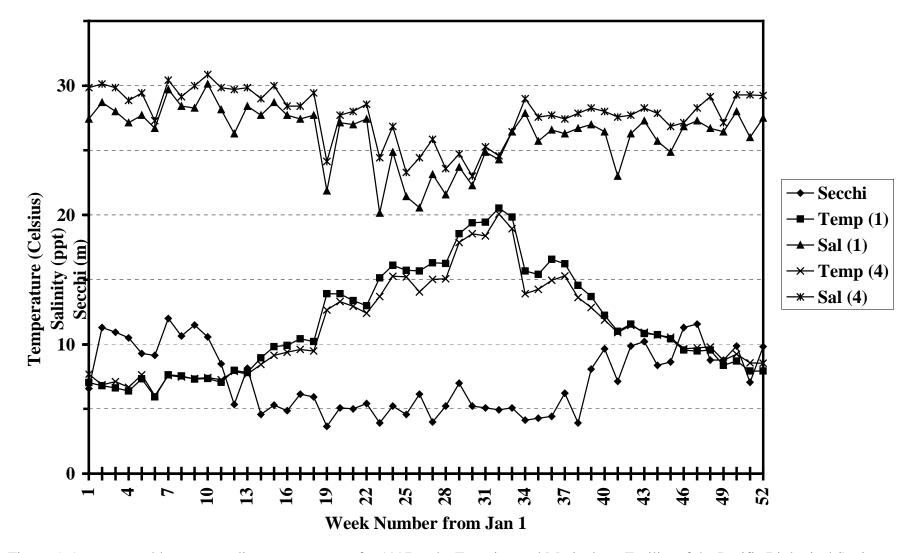


Figure 5. Average weekly water quality measurements for 1997 at the Experimental Mariculture Facility of the Pacific Biological Station in Departure Bay.

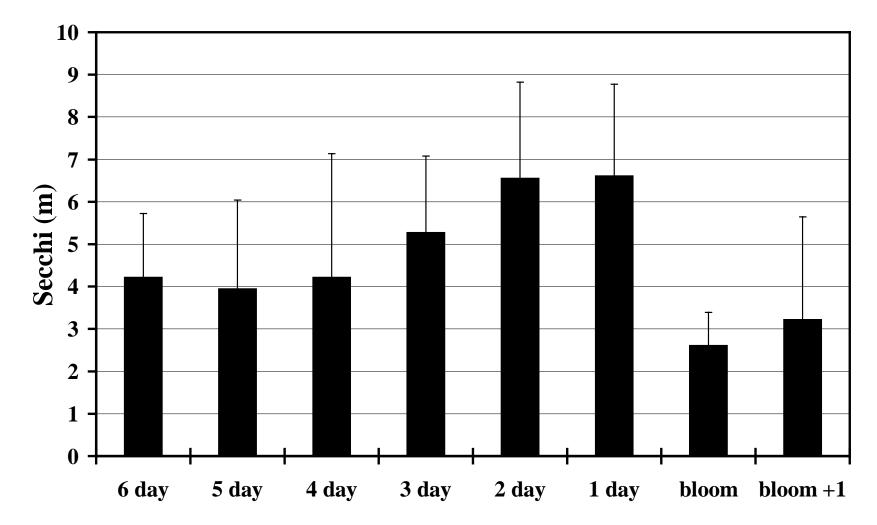


Figure 6. Average Secchi Disc reading leading up to a *Heterosigma* bloom (> 5 million cells per litre) (error bars showing standard deviation). Data from all blooms including non-lethal ones in 1993 and