Final Technical Report

Physical Processes Between the Kennebec Estuary, Casco Bay and the Inner Shelf: Potential Implications for Contaminant Transport and Red Tides.

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Alexandrium spp. in Eastern Casco Bay and the Kennebec River Estuary.

INTRODUCTION

The freshwater discharge of the Kennebec River (> 4000 m^3/s in spring) can profoundly influence the circulation and hydrography of the estuary and adjoining continental shelf waters in the Gulf of Maine. It has been suggested (Franks and Anderson, 1992) that eastern Casco Bay, just west of the mouth of the Kennebec River, is the source region for red tide blooms occurring in the western gulf during spring and summer. Shellfish consume the motile (swimming) dino-flagellates and accumulate saxitoxins. In turn, the shellfish are consumed by humans causing a potentially fatal neurological disorder called paralytic shellfish poisoning (PSP).

The most significant red tide organism in the gulf is a toxic dinoflagellate, *Alexandrium spp.* A critical feature of the life cycle of this dinoflagellate is the formation of a dormant cyst stage at the end of a bloom. The cysts sink and over winter in the sediment creating a seed population that germinates the following spring to fuel a new bloom. Numerous hypotheses exist as to the importance of inshore versus offshore populations as potential seed for toxic blooms as well as the conditions for bloom formation and advection throughout the gulf. Considerable interannual variability has been observed in terms of the occurrence and extent of toxic blooms in the Gulf of Maine such that an extensive monitoring program is operated by the State of Maine for the protection of human health. The objectives of this study were to address the following:

1) Does a seed population exist in the Casco Bay/Kennebec River region?
2) Is there a mechanism for generation of blooms?
3) Does the Kennebec River act as a repository of cysts?
4) Does the two layered circulation of the Kennebec River contribute to the movement or dispersal of cysts through the formation of an estuarine turbidity maximum?

HYDROGRAPHY OF THE REGION

The physical oceanography component of this project was conducted by Dr. Neal Pettigrew and Dory Kistner of the University of Maine. Earlier findings have demonstrated that the Kennebec River exhibits a strong two-layered estuarine circulation and flood-ebb asymmetry, which contribute to the formation of an estuarine turbidity maximum (ETM) near the upstream limit of salinity intrusion. The ETM was documented in the upper part of the estuary under a Sea Grant funded comparative study of three Maine estuaries in 1993–1995 (Wong and Townsend, 1999; Mayer et al., 1996), and its position was observed to oscillate with the tide. The current pattern associated with an ETM and the strength of the bottom landward flowing currents could be relevant to the issue being discussed here. If the ETM forms after the spring freshet as hypothesized and moves upstream throughout the summer, it may serve to trap not only sediment in the estuary but also benthic cysts of *Alexandrium spp.* These cysts may then be stored in the upper estuary until the next spring when they are flushed out onto the adjacent shelf to fuel that season's red tide. However, the timing of the formation of the ETM at the mouth and its current structure is not known and there are no details on its movement upstream. In addition, the sediments in the upper Kennebec River near Merrymeeting Bay have never been examined for cysts although the substrate is fine grained and suitable for their storage.

The results of a previous EPA funded study (Pettigrew, pers. comm.) suggest that the Kennebec plume exerts a major influence on the oceanography of Casco Bay and the inner shelf even under low outflow conditions. During high runoff conditions, the plume can completely dominate the local residual circulation and hydrographic fields. Thus, information on the density structure of the water column and the strength and direction of the residual and tidal currents in Casco Bay are critical to understanding the mechanism responsible for initiating a bloom.

METHODS

Sediment samples were collected at each station using a Shipek sampler to determine the presence and abundance of *Alexandriums spp.* cysts. Sub-samples of fine-grained sediment were removed from the top 3 cm of the grab sample using small diameter corers and refrigerated in the dark. Prior to counting, each sample was re-suspended in a standard volume of filtered seawater and a sub-sample of this suspension was sonicated for 30 seconds (2 second pulses at 45 watts). The sample was then sieved to collect particles in the 20-75μm size range, rinsed into a centrifuge tube, fixed with paraformaldehyde (final concentration 1%) for 30 minutes and centrifuged at 700G for 10 minutes. The pellet was re-suspended in methanol and refrigerated for two days to permeabilize the cysts. After two days, the methanol was replaced with deionized water and the cysts stained with Primuline (0.2 mg/ml final concentration) for 30 minutes. After re-centrifuging, the pellet was resuspended in freshly deionized water and a subsample was counted using epi-flouresence microscopy (Yamaguchi, et al., 1995). Concentrations of cysts/cc for samples collected in 1997 were calculated using the entire core volume. Concentrations of cysts/cc for samples collected in 1998 were separated into 0-1cm and 1-3cm depth sub-samples which provided additional information on distributions within the sediment.

At each of the stations, a vertical net tow (20μm) was collected to the depth of the 1% light level determined by Secchi disk depth (Z_s * 3) or to just below the sub-surface chlorophyll maximum indicated by an *in situ* fluorometer profile. Net samples were fixed with Lugol's solution and counted directly using a light microscope. Discrete chlorophyll samples were also collected using Niskin bottles and analyzed fluorometrically by the method of Holm-Hansen, et al. (1965).

RESULTS

In the Kennebec River, no cysts or motile cells were observed in 1997 (Figure 1). In April, 3 sediment and net tow samples were obtained below the region known as Fiddler's Reach off Doubling Point in Arrowsic, ME. The river bottom is scoured in this region and sediment samples were difficult to obtain. In June, 3 sediment and net tow samples were collected above Bath, ME, to Merrymeeting Bay where bottom sediments are more available. A study that sampled the Kennebec River for phytoplankton populations in September, 1993, and May, June, July and September, 1994 (Wong and Townsend, 1999) reported dinoflagellate abundance was low except in June and July with no direct reference to *Alexandrium spp.* Sampling was not repeated in the Kennebec River in 1998 with the exception of several net tows above Bath, ME, in October where no motile cells were observed (Figure 2b). The Kennebec River appears to be an unlikely repository of red tide seed populations.

In the New Meadows River, no cysts and negligible motile cells (0.1/L at a station northwest of Bear Island) were observed in April, 1997 (Figure 2). The following April (1998) cyst concentrations of 19.3 to 46.5 cysts/cc of sediment were observed with motile cell concentrations less than 1 per liter.

Sampling was reduced to a single station in October, 1998, where 23.7 cysts/cc of sediment were found and no motile cells were present. While the sampling matrix for both rivers in both years was not ideal for statistical comparison, these data suggest that the New Meadows River is more important as a potential source of cysts to seed a spring bloom of *Alexandrium spp.* in some, but not all, years.

Offshore sediment and water samples indicated a similar pattern when comparing the Kennebec and New Meadows regions of the continental shelf to 100m. No cysts were observed off the mouth of the Kennebec in April of either year (Figures 1a and 2a). In June, 1997, cyst concentrations ranged between 0 and 19.2 cysts/cc of sediment. In October, 1998, concentrations of 13.1 to 243.5 cysts/cc were observed for the same stations. When present, motile cells were observed in concentrations to 1.5 cells/L in April and to 2.4 cells/L in June, 1997 (Figure 1b). Concentrations of motile cells off the Kennebec River in April and October, 1998, were always <1 cell/L (Figure 2b). Off the New Meadows River, cysts were always present seasonally in the sediments where concentrations ranged between 0.0 and 33.3 cysts/cc of sediment in April, 1997, and 0.0 to 41.4 cysts/cc in April, 1998. Similar values were observed in June, 1997, with 3.3 to 49.0 cysts/cc and in October, 1998, with 4.8 to 106.3 cysts/cc. Patterns in the distribution of *Alexandrium spp.* motile cells off the New Meadows River were similar to the offshore waters of the Kennebec; ranging to 3.5 cells/L in June, 1997, and <1 cell/L in all other sampling months.

DISCUSSION

Our results demonstrate the interannual variability that occurs in this region for the distribution of *Alexandrium spp.* cysts in the sediments and motile cells in the water column. Comparisons can be drawn between the Kennebec and New Meadows Rivers, and between the rivers and the offshore region of Casco Bay. Not much is known concerning the relative importance of 'offshore' versus 'inshore' populations, but higher temperatures and light intensities at inshore beds may promote cyst germination. Thus, although inshore cyst beds are smaller, they may be more active and more significant to bloom initiation (Keller, 1999). The New Meadows River and Casco Bay were shown to be more significant repositories of cysts in sediment than the Kennebec River.

The only region where significant concentrations of cysts were present in both sampling months and both years was offshore of the New Meadows River in Casco Bay. Cysts were present in the upper reaches of the New Meadows River in April of 1998, but not in April of 1997. Cysts were also found off the mouth of the Kennebec River in the summer of 1997 and the fall of 1998, but not in April of either year, with no cysts found within the Kennebec River. The present study was undertaken in conjunction with investigations on the currents and hydrography of the region. River discharge values measured from USGS flow gauges on the Kennebec and Androscoggin Rivers show maximum flow occurred before and during the April samplings in both years. The high river discharge would have flushed the channel into the bay. Kistner and Pettigrew (pers. comm.) did not find a strong ETM in April, 1997. It is known that high river flow can be fatal to the mechanisms sustaining the ETM by moving the region of estuarine mixing out of the channel and onto the adjacent shelf. It was observed that during the summers of 1997 and 1998, at high flow conditions, the 2 psu isohaline was pushed almost entirely out of the channel – as far as 27 km from mouth of the Kennebec River in October, 1998. The high cyst concentrations in October, 1998, may have been the result of a bloom that had already occurred in summer and generated cysts that sank to the sediments.

Motile cell concentrations measured in this study were very low compared to values observed during blooms (> 100 cells/L) or reported periods of toxicity when cell concentrations ranging from

<100 to 1000 cells/L have been observed (Anderson et al., 2000). Net tows may be considered more qualitative than quantitative, however, we feel that the concentrations of motile cells per liter that we calculated may be used quantitatively. April and October of any year may not be times when high numbers of motile *Alexandrium spp.* cells should be expected, whereas the month of June should fall within the growth period which leads to the development of a bloom if one occurs. Our highest motile cell concentrations were observed in June of 1997, a year when significant toxicity was measured along the coast in this region by DMR (L. Bean, pers. comm.).

These results confirm the wide distribution of *Alexandrium spp.* in Casco Bay and the inner continental shelf. In this highly dynamic region, it is not possible to determine whether there is an offshore, inshore or multiple sources for *Alexandrium spp.* blooms based solely on cyst or motile cell concentrations. Factors such as light, temperature and nutrients which might trigger blooms must be considered. Model simulations driven by environmental and hydrographic conditions in conjunction with a large-scale cyst map of the region presently being undertaken by McGillicuddy et al. (2000) could provide important insights

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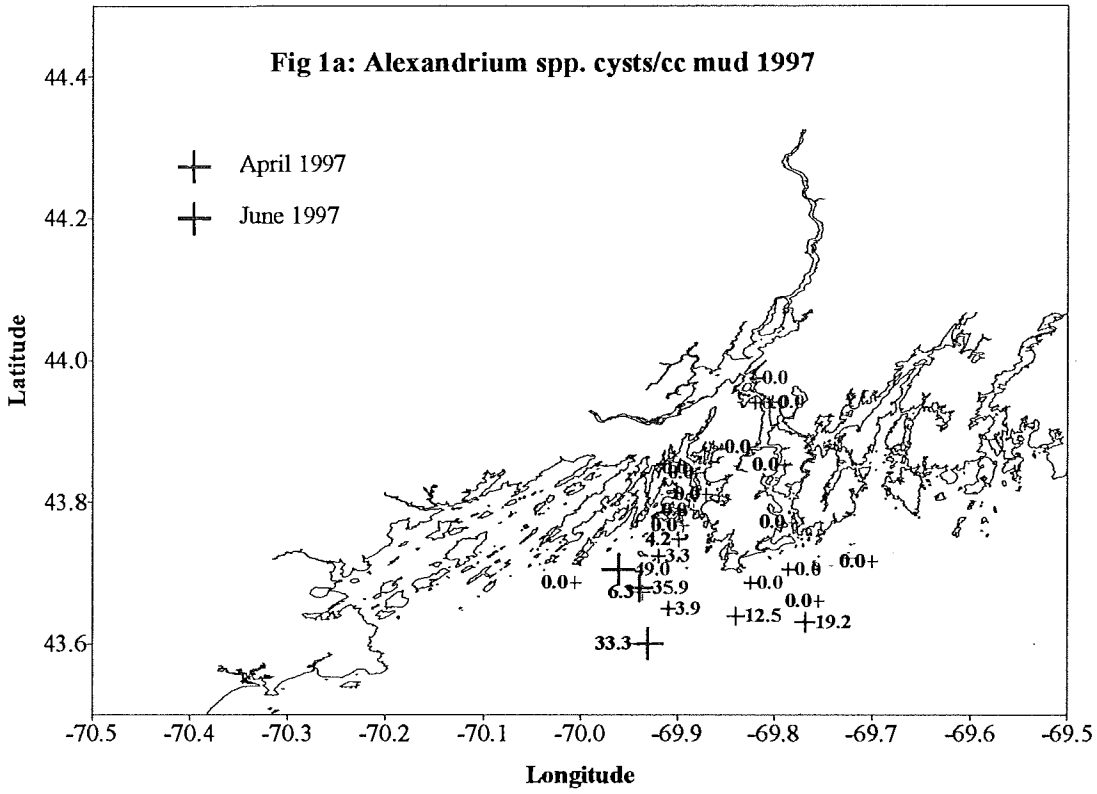
Fig 1a: Alexandrium spp. cysts/cc mud 1997
April 1997
June 1997
Latitude
Longitude
44.4
44.2
44.0
43.8
43.6
-70.5
-70.4
-70.3
-70.2
-70.1
-70.0
-69.9
-69.8
-69.7
-69.6
-69.5
0.0
4.2
3.3
49.0
35.9
6.3
3.9
12.5
19.2
33.3

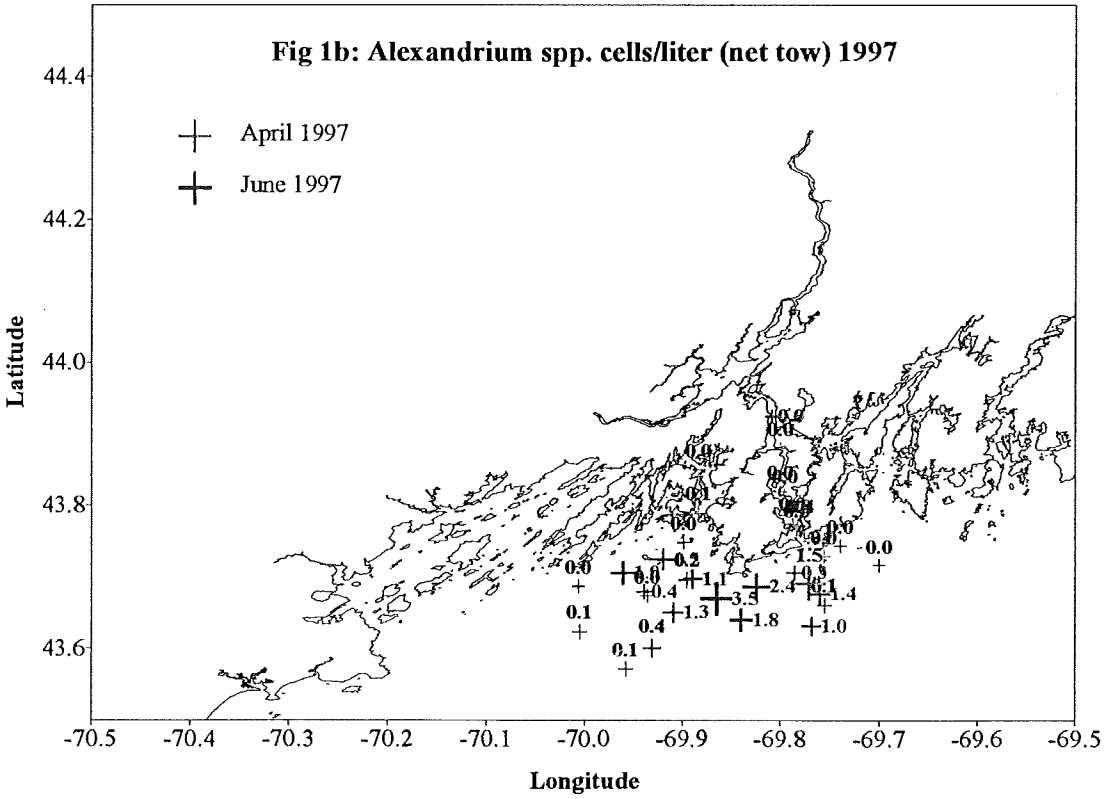
Fig 1b: Alexandrium spp. cells/liter (net tow) 1997
April 1997
June 1997
Latitude
Longitude
44.4
44.2
44.0
43.8
43.6
-70.5
-70.4
-70.3
-70.2
-70.1
-70.0
-69.9
-69.8
-69.7
-69.6
-69.5
0.0
0.2
1.1
0.4
1.3
3.5
2.4
1.8
1.0
1.4
1.5
0.1
0.4
0.1

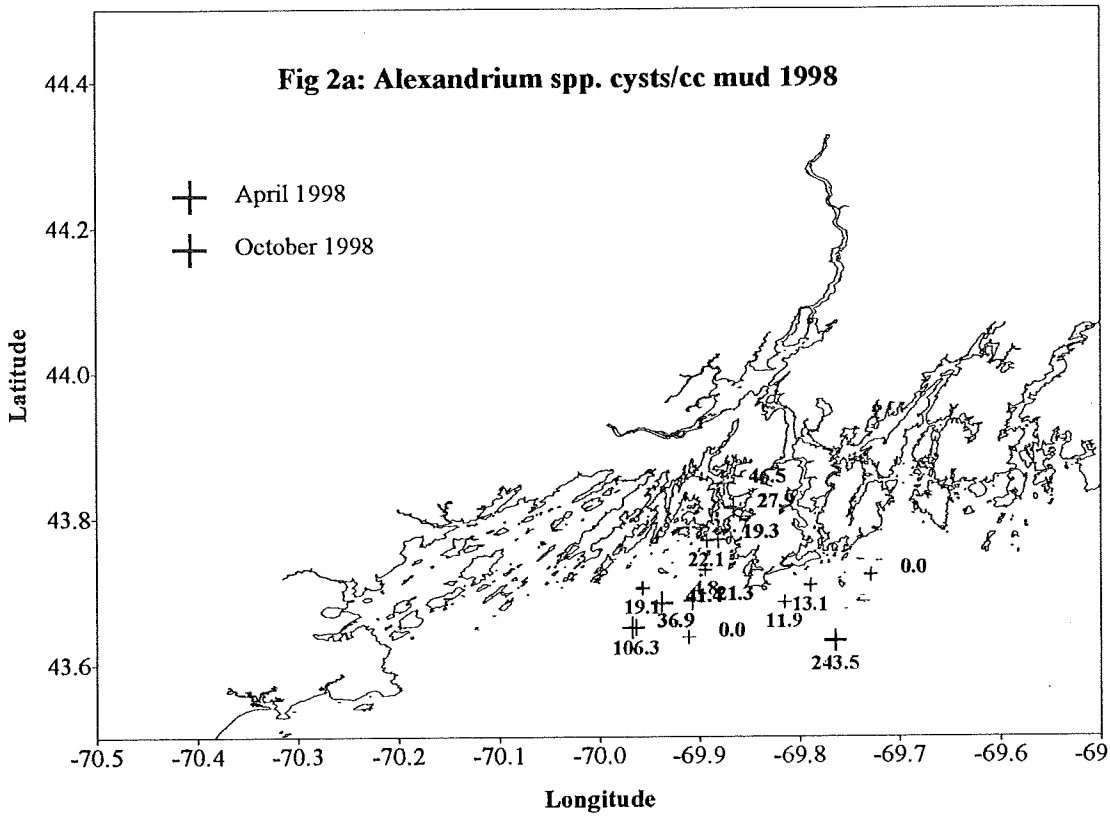
Fig 2a: Alexandrium spp. cysts/cc mud 1998
April 1998
October 1998
Latitude
Longitude
44.4
44.2
44.0
43.8
43.6
-70.5
-70.4
-70.3
-70.2
-70.1
-70.0
-69.9
-69.8
-69.7
-69.6
-69
46.5
27.6
19.3
22.1
0.0
19.1
41.2
21.3
13.1
36.9
0.0
11.9
106.3
243.5

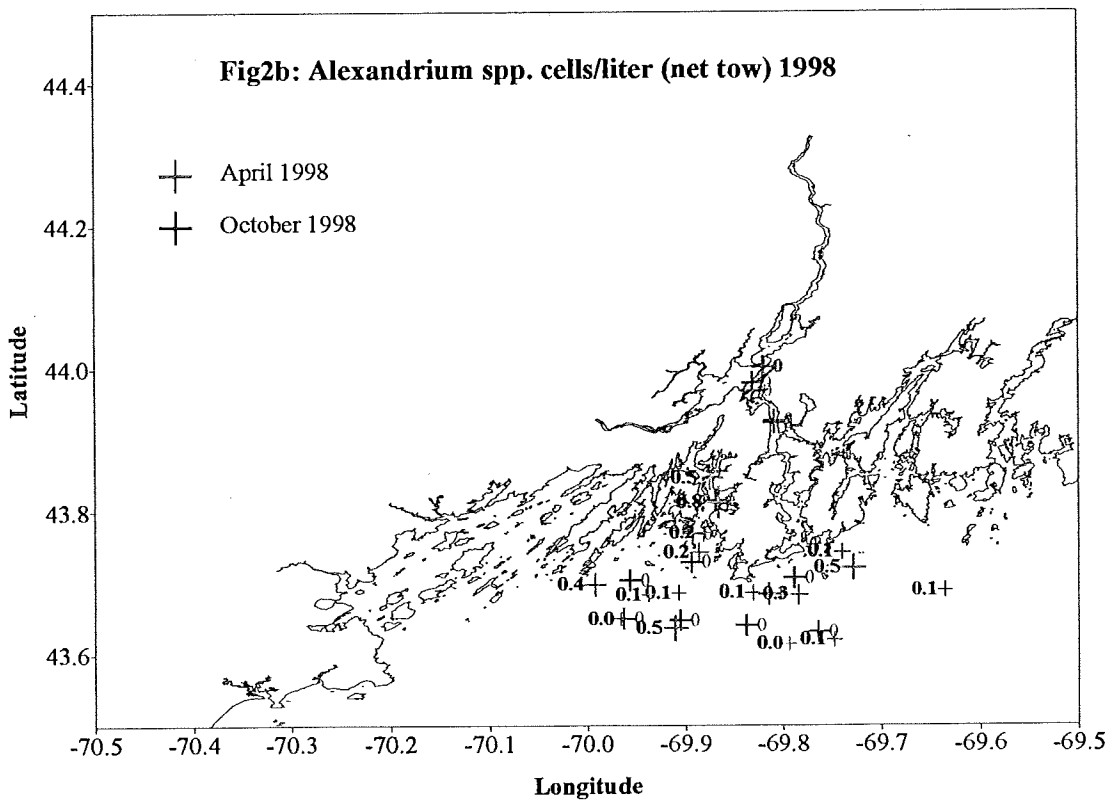
Fig2b: Alexandrium spp. cells/liter (net tow) 1998
April 1998
October 1998
Latitude
Longitude
44.4
44.2
44.0
43.8
43.6
-70.5
-70.4
-70.3
-70.2
-70.1
-70.0
-69.9
-69.8
-69.7
-69.6
-69.5
0.4
0.1
0.1
0.1
0.3
0.1
0.0
0.5
0.0
0.1
0.5
0.1