New phytoplankton species in the Bay of Fundy since 1995

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A monitoring programme was initiated in 1987 to study phytoplankton populations in the Western Isles region of the Bay of Fundy, southwest New Brunswick. Samples are collected weekly from May through October, and monthly during the remaining months, to determine phytoplankton distribution and abundance at Brandy Cove, Lime Kiln Bay, Deadmans Harbour, the Wolves Islands, and mid-Passamaquoddy Bay. Since the programme was initiated, several previously absent or non-indigenous species have been found, suggesting that new species may have been introduced to the area. In order to establish a baseline for species indigenous to Bay of Fundy waters, we have taken a conservative approach and termed species reported for the first time in the Bay of Fundy system since 1995 as "new" species. New species include the following: (dinoflagellates) *Alexandrium pseudogonyaulax*, *Amphidinium carterae*, *A. sphenoides*, *Ceratium macroceros*, *Polykrikos schwartzii*, *Preperidinium meunieri*, *Protoperidinium crassipes*, and *Pyrocystis lunata*; (diatoms) *Attheya septentrionalis*, *Attheya longicornis*, *Chaetoceros radicans*, *Cylindrotheca gracilis*, *Grammatophora serpentina*, *Lithodesmium undulatum*, *Mediopyxis helysia*, *Membraneis challengeri*, *Neodenticula seminae*, *Odontella sinensis*, *Proboscia eumorpha*, *Pseudo-nitzschia subpacifica*, *Pseudo-nitzschia fraudulenta*, and *Thalassiosira punctigera*.

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Introduction

A phytoplankton monitoring programme was initiated in the Western Isles region of the Bay of Fundy, eastern Canada (Figure 1) in 1987 when the salmonid aquaculture industry was expanding rapidly amid growing concerns and claims that incidents involving harmful algal blooms (HABs) seemed to be increasing in intensity, frequency, and geographic distribution in other regions of the world (Anderson, 1989; Smayda, 1990; Hallegraeff, 1993, 1995). The objectives of the phytoplankton study when it was initiated were to (i) establish baseline data on phytoplankton populations in the lower Bay of Fundy, because little detailed work had been published since studies of Davidson (1934) and Gran and Braarud (1935); (ii) identify harmful algal species that could harm the salmon aquaculture industry; (iii) determine patterns and trends in phytoplankton populations; and (iv) provide an early warning to aquaculture and wild fisheries and regulatory agencies, such as the Canadian Food Inspection Agency (CFIA), on species that produce toxins and which can result in shellfish toxicities and closures of shellfish beds to harvesting. A long-term goal of the study was to determine whether there were changing trends in phytoplankton populations or new species introduced as a result of the increased use of inshore waters in recent years for salmon farming, tourism, fishing, and shipping, or as a result of other natural environmental parameters such as climate change, storm events, or ocean currents.

Phytoplankton transport and relocation to other regions of the world are a major concern, especially for HABs and their establishment and effects on the new environment. A number of vectors have been implicated through which phytoplankton can be transported, including changes in hydrography, currents, and winds, hull fouling, movement of live shellfish, and shipping (Minchin and Gollasch, 2002). Ship ballast tanks have been suggested as a major vector for the transport and introduction of phytoplankton species either through the water column or in sediments from the tanks (Medcof, 1975; MacDonald, 1995; Hallegraeff, 1998). Additionally, phytoplankton cells have been shown to remain viable in tanks and sediments for several days to months (Yoshida *et al*., 1996), and dinoflagellates can be germinated from ballast tank sediments (Pertola *et al*., 2006). Most vessels that come to the two major ports in the New Brunswick portion of the Bay of Fundy are bulk, ore, and container ships that arrive in ballast and leave with cargo. Routes can vary between years, with most traffic originating from the northeast, northwest, and western central Atlantic (JLM, unpublished data).

Material and methods

Sampling in the southwestern New Brunswick portion of the Bay of Fundy was initiated in 1987 in Lime Kiln Bay (where a number of salmon aquaculture sites are located) and in 1987/1988 at Brandy Cove (a brackish site influenced by the St Croix River estuary), Deadmans Harbour (an open bay with offshore influence), and the Wolves Islands (an offshore indicator site; Figure 1). An extra sampling site was added in mid-Passamaquoddy Bay in 1999, following the observation that Brandy Cove was not a good indicator site for cell densities of algal blooms within Passamaquoddy Bay.

Sampling was conducted onboard the research vessel, CCGC "Pandulus III". Weekly samples were collected from 1987 to the present between April or May and the end of September or

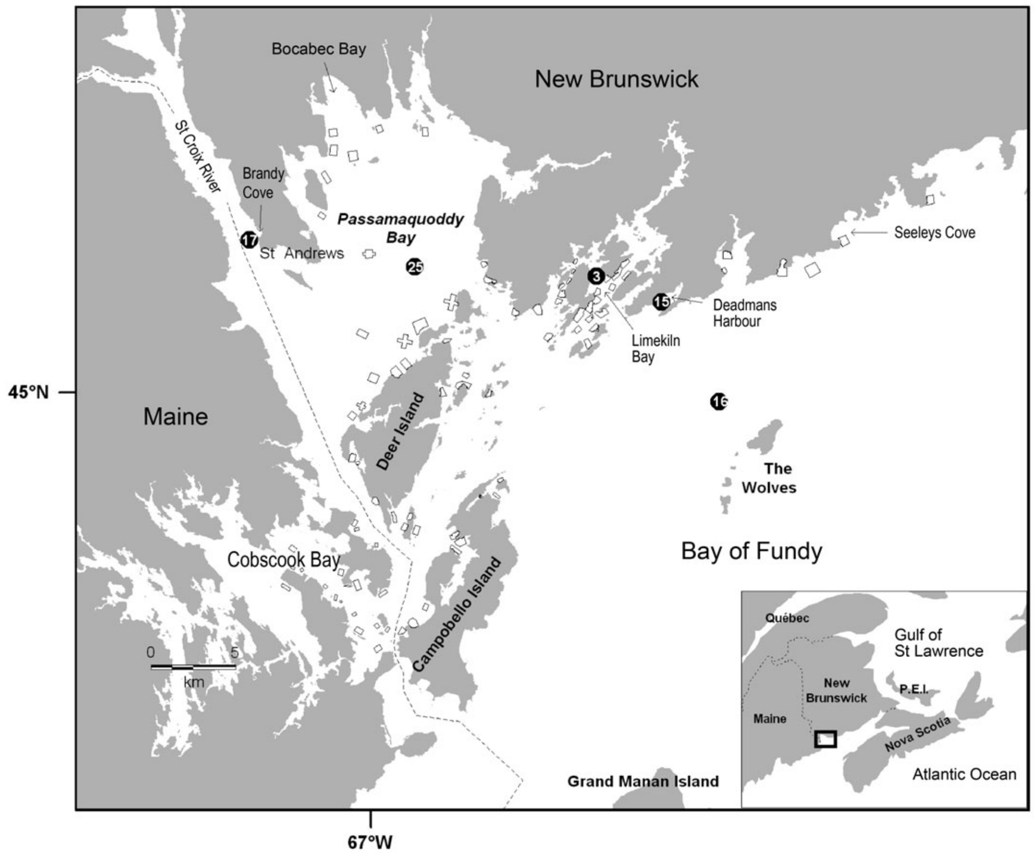

Figure 1. Map of sampling stations. Boxes indicate locations of salmon aquaculture sites.

October, depending on the initiation and decline of the spring and autumn phytoplankton blooms. Biweekly sampling was conducted in shoulder months, such as April and October (when phytoplankton cell densities had begun to increase or decrease), and monthly during all other cooler months.

Phytoplankton samples were collected at the surface by bucket from all five stations and at depths of 10, 25, and 50 m with a Niskin bottle at the Wolves Islands. Water samples (250 ml) were immediately preserved with a 5 ml formaldehyde:acetic acid solution. Later, 50 ml subsamples were settled in counting chambers for 16 h. All phytoplankton $>5\ \mu m$ were identified and enumerated (as cells l^{-1} or chains of cells l^{-1}) with the Utermöhl technique using a Nikon inverted microscope (Sournia, 1978). Because we have used the same enumeration technique since sampling was initiated, the element of error described by Wiltshire and Dürselen (2004) associated with changes in personnel, and the identification of phytoplankton in long-term datasets has been reduced. Further identification was made with either a JEOL JSM–5600 scanning electron microscope (SEM) or a Hitachi S–2400 SEM.

Data from phytoplankton analyses of the total community for the years 1987–2000 have been published already (Wildish *et al.*, 1988, 1990; Martin *et al.*, 1995, 1999, 2001, 2006), but the data from 2001 to 2006 have not yet been published.

Dinoflagellate and diatom species were defined as "new" if they had not been observed in samples prior to 1995 and now appear to be permanently established as members of the local ecosystem (Boalch and Harbour, 1977; Dahl and Tangen, 1993; Hallegraeff and Gollasch, 2006). We also noted additional taxa that included cyanobacteria, flagellates, and protists, which may have been present before 1995, but were either small or similar in structure to existing species and so may have been previously grouped with other species.

Results

Eight new dinoflagellate species and 14 new diatom species have been observed in Bay of Fundy samples since 1995, along with five additional species including flagellates, small zooplankton, cyanobacteria, and haptophytes (Table 1). Most species new to the area are cold-temperate species that exist in other regions of the world with ecosystems similar to the Bay of Fundy. They appear to have established populations in the Bay of Fundy, because they have been observed during more than one year or annually since the time of first observation. Of the 27 new species observed in the area, most were observed in 2000 (9 species) and 2001 (14 species); just one new species was detected in each of the years 1997, 2002, 2004, and 2005.

Many of the new species detected are in the larger size range of their phytoplankton group, and have distinct morphological features that would not easily have been missed or overlooked in samplings prior to 1995. One such species, *Mediopyxis helysia*, was recently described as a species of diatom new to science (Kühn *et al.*, 2006). It was isolated from the Wadden Sea in 2003 and the Gulf of Maine in 1996, and first observed in the Bay of Fundy in 2002. *Odontella sinensis*, another of the larger species, was first detected in our study waters in 2000. Although one

Table 1. New phytoplankton species in the southwest New Brunswick portion of the Bay of Fundy since 1995, and the year in which each was first observed.

Phytoplankton species	Year first observed
Dinoflagellates	
Alexandrium pseudogonyaulax	2001
Amphidinium carterae	2000
A. sphenoides	2000
Ceratium macroceros	2001
Polykrikos schwartzii	2001
Preperidinium meuneri	2001
Protoperidinium crassipes	2001
Pyrocystis lunata	2001
Diatoms	
Attheya longicornis	2004
Attheya septentrionalis	2001
Chaetoceros radicans	2000
Cylindrotheca gracilis	2001
Grammatophora serpentine	2001
Lithodesmium undulatum	1997
Mediopyxis helysia	2002
Membraneis challengeri	2001
Neodenticula seminae	2005
Odontella sinensis	2000
Proboscia eumorpha	2001
Pseudo-nitzschia subpacifica	2000
P. fraudulenta	2000
Thalassiosira punctigera	2001
Other taxa	
Apedinella sp.	2001
Chrysochromulina parkeae	2002
Commation cryoporinum	2000
Microcystis sp.	2001
Phaeocystis pouchetii	2000

possible mode of transport could have been through natural dispersal via currents from the Gulf of Maine, the species alternatively could have arrived via human transport, because our observation of the species coincided with an observation of *O. sinensis* from a ballast water sample taken from a ship at the port of Bayside (Figure 2), whose last port of call was Charleston (USA). In the same year too, *Lithodesmium undulatum* was observed in ballast water samples and water samples from our sampling programme. *Odontella sinensis* has become well established since 2000, because it has been observed in samples during January, May, and August through December of every year. In contrast, *L. undulatum* does not appear to be as successful to date, and has been seen only rarely. *Amphidinium carterae*, *A. sphenoides*, *Polykrikos schwartzii*, *Commation cryoporinum*, and *Thalassiosira punctigera* have also been observed in water samples on a regular basis since they were first observed, but they have not been seen yet in ballast water samples.

Discussion

In all, 55 species of dinoflagellate, 168 of diatom, and 30 other species, including flagellates, smaller zooplankton, and ciliates, have been recorded in our sampling programme (Wildish *et al*., 1988, 1990; Martin *et al*., 1995, 1999, 2001, 2006). Of these 253 taxa recorded since 1995, 8 dinoflagellate, 14 diatom, and 5 other taxa (including flagellates, smaller zooplankton, cyanobacteria, and haptophytes) have been documented for the first time and appear to be new to the area. It is not evident that any species have disappeared or are no longer present in the Bay of Fundy. Although a few species have been observed only occasionally or rarely since 1995 (*Ceratium minutum*, *Detonula confervacea*, *D. pumila*, *Fragilaria crotonensis*, *Licmophora flabellata*, *O. obtusa*, *O. regia*, and *Synedra* sp.), the dataset does not extend over a sufficiently long period to demonstrate the complete disappearance of these species from the Bay.

A decision to use the year 1995 to identify species as new to the region was made in order to eliminate a number of factors that might influence results. These factors include: (i) the possibility that species were overlooked or "hidden" (because so few cells were present previously through the initial years of a sampling programme); (ii) more detailed species information has become available; (iii) allowing for random errors associated with the initiation of a counting person and the personal element in microalgal species recognition; (iv) interannual and decadal variability; (v) allowing for years during which a particular species may not have been present; and (vi) the fact that expertise in identification tends to decrease with cell size. Using 1995 as the start year for determining new species to the area, we have been able to show that some indigenous species, such as *Eucampia zodiacus*, occurred at high cell densities during some years, at very low densities during others, or were not observed at all during some years (Martin *et al*., 2007). Other indigenous species, such as *Gyrodinium auroleum*, which were very abundant during the early years of the monitoring programme, seem to have decreased in occurrence more recently. Others, such as *Alexandrium fundyense*, have clearly cyclical abundance patterns, years with greater cell densities alternating with periods of low cell density (Page *et al*., 2006).

Currents and water movements may be one means through which some phytoplankton cells have been transported from the Gulf of Maine into the Bay of Fundy. Inflow from the Gulf of Maine eddy (located south of the Bay of Fundy to the east of Maine and southwest of Nova Scotia) takes place along the southern entrance to the Bay, close to the coast of Nova Scotia (Bumpus, 1960; Bumpus and Lauzier, 1965). An alternative explanation for the presence of these phytoplankton species would be through human-mediated vectors, such as ship traffic (Hallegraeff, 1998; Hallegraeff and Gollasch, 2006). Figure 2 shows shipping routes and ports for the Bay of Fundy. Most ships begin discharging ballast at the entrance to the Bay of Fundy, then continue discharging until they reach port, allowing the discharge to be dispersed throughout the Bay. Water in the Bay of Fundy has an estimated flushing time of 76 d (Trites and Garrett, 1983), indicating the potential for extended residence and a potential for cell establishment and growth. Many vessels entering the Bay of Fundy from the northeast, northwest, and western central Atlantic use water ballast, so pose quite a threat in terms of introducing non-indigenous material into the Bay. Previous studies and samples collected from ballast tanks from ships entering the Bay of Fundy in 2000 show that phytoplankton is transported into the Bay and remains viable (JLM, unpublished data). Water samples collected in the Bay of Fundy near ships discharging ballast have contained species that were also found in the

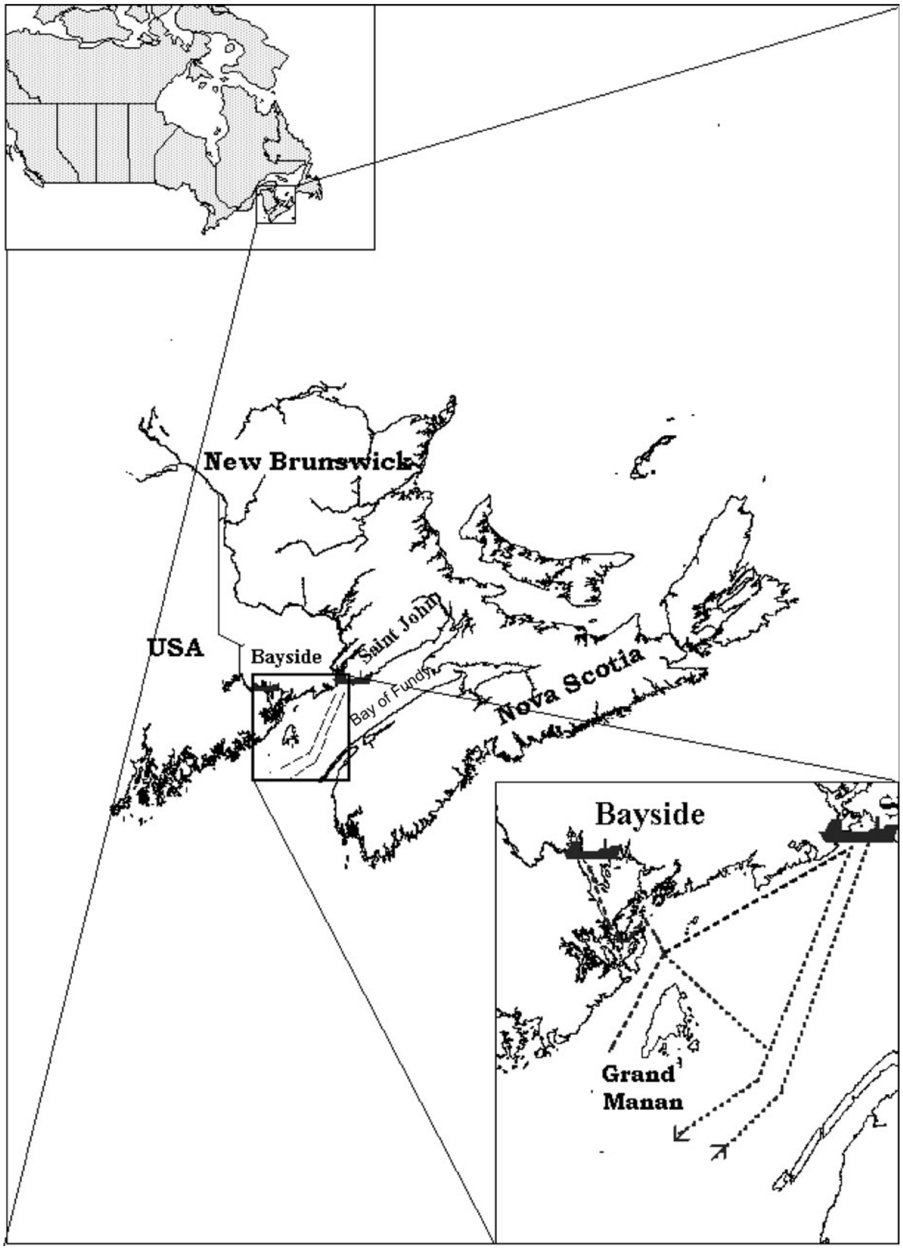

Figure 2. Map showing the shipping routes and the major ports of Bayside and Saint John in the Bay of Fundy.

ballast tanks. Similarly, authors such as Hallegraeff and Gollasch (2006) and Bolch and Salas (2007) presented strong evidence that ballast water has acted as a major vector for introductions of microalgae into ecosystems in Australia and elsewhere in the world.

Here, we have documented for the first time the presence of some phytoplankton species new to the Bay of Fundy. Although most of the new species found do not appear yet to have caused harm or problems to fisheries either in the Bay of Fundy or elsewhere in the world, one needs to treat the introduction of all non-indigenous microalgae as potentially harmful to the environment. There is always the concern that these species may become dominant and could alter community structure or cause harm to existing fisheries and fauna, for instance as has been shown in Derwent Estuary (Tasmania, Australia) by Hallegraeff and Summer (1986), the Mediterranean Sea by Vila *et al.* (2000), and in Norway by Hopkins (2002). It is also possible that, through continued monitoring, more new species will be found in the Bay of Fundy, including other taxonomic groups.

With intensified concerns and awareness throughout the world about the global spread of non-indigenous species, the documentation of baseline data for a region is considered to be important. Moreover, we have shown the necessity for this monitoring data to be long term. Such monitoring data are becoming more detailed, through advancements in the availability of and access to new taxonomy references, the description of new species, the identification through detailed documentation of species currently listed as unknown in datasets, increased monitoring and collaboration between researchers, improved access to taxonomic expertise, and the evolution of molecular tools.

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