Mapping chlorophyll-*a* fluorescence in a blue mussel (*Mytilus edulis*) seed collection farm

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The primary objective of this study was to assess the phytoplankton biomass distribution throughout a blue mussel (*Mytilus edulis*) seed collection farm using active fluorescence mapping technology. Within the farm we measured a fluorescence depletion of up to 30% compared to outside the farm. The depletion generally extended 10 m past the farm, although during ebb tide it appeared to follow currents and was traceable further (500 m) downstream past the farm. The scale and magnitude of this "shadow effect" was attenuated during flood tide. Within the farm, there were generally increases in fluorescence between sets of longlines. Unexpected areas with high phytoplankton biomass were also observed inside the farm during the ebb tide.

RÉSUMÉ

Sonier, R., Comeau, L.A. and Lanteigne, L. 2010. Mapping chlorophyll-*a* fluorescence in a blue mussel (*Mytilus edulis*) seed collection farm. Can. Tech. Rep. Fish. Aquat. Sci. 2870 : v + 17 p.

L'objectif primaire de cette étude fut d'évaluer la distribution de la biomasse phytoplanctonique à l'intérieur et autour d'une ferme de collecte de naissains de moules bleues (*Mytilus edulis*). La fluorescence active (*in-situ*) fut utilisée afin de déterminer l'étendue et la persistance des gradients de fluorescence à l'intérieur de la ferme. À la profondeur des collecteurs de naissains, une diminution de fluorescence atteignant jusqu'à 30 % fut notée comparativement à l'extérieur de la ferme pour la même profondeur. Cette diminution s'étendait généralement jusqu'à 10 m à l'extérieur du bail, toutefois durant la marée perdante cette diminution semble suivre les courants dominants et est détectable même jusqu'à 500 m à l'extérieure de la ferme. Cependant, la fluorescence augmente rapidement entre les blocs de longues lignes (« corridors »), et s'accroit considérablement environ 10 mètres de distance à l'extérieur de la ferme. Certains endroits localisés démontrent des valeurs élevées en fluorescence au centre de la ferme surtout durant la marée descendante.

INTRODUCTION

Primary production rates in estuaries are among the highest on earth, and the transfer of this productivity through the food web often yields prolific fisheries (Gobler *et al.* 2005). The yields from culturing marine shellfish in estuarine environments have steadily increased over the last two decades. In 1996, bivalve production accounted for 25% of the world's aquaculture production in terms of value (Gibbs, 2004). In Atlantic Canada, shellfish farming has grown progressively over the past eighteen years, during which the annual production increased from 1,463 t in 1986 to 28,039 t in 2004 (Comeau *et al.* 2006). For instance, blue mussel (*Mytilus edulis*) production on Prince Edward Island (P.E.I.) greatly increased during this time and is now responsible for 80% of Canadian mussel production (Drapeau *et al.* 2007). Likewise, aquacultural production of the American oyster (*Crassostrea virginica*) was approximately 4 million oysters in 2002, and production is estimated to reach 54 million oysters in 2010 (Sonier, 2006).

Presently, the Department of Fisheries and Oceans in Atlantic Canada grants few sites (leases) for new development or expansion of shellfish aquaculture. Faced with this dilemma, the growers aim to optimize crop distribution at their leases and/or bay-scale to improve the productivity of suspension-cultured shellfish. Therefore, some shellfish growers are reconsidering their growing layout according to the uneven crop growth within their active leases, which can cover areas of up to 300 acres. Recently, a cost benefits study of P.E.I. blue mussel production analyzed sock spacing and socking densities. The results of the study suggested that there was a positive effect of high sock spacing on shell growth and abundance for small, densely packed seeds (Comeau *et al.* 2008).

Mussels feed on phytoplankton, and strong depletions in chlorophyll are commonly observed above mussel beds (Maar *et al.* 2007). At low algal concentrations, mussels can close their valves and temporarily stop feeding to reduce their oxygen uptake and respiration and save energy (Dolmer, 2000; Riisgård *et al.*2003). The growth of filter-feeding bivalves is largely controlled by food availability. In turn, food availability is affected by phytoplankton dynamics, the concentration of suspended particulate matter (seston), community composition (i.e. quality and species) and transport rates (currents and waves) (Strohmeier *et al.* 2008). One of the shellfish industry's concerns is that

seeds and juveniles are deployed with inadequate consideration to localized inequalities in growing conditions, such as phytoplankton concentrations and current velocities. For instance, several areas have a high phytoplankton abundance with a low crop biomass, or vice-versa. Shellfish growers may gain some control over the management of these characteristics by taking into account localized patterns and trends in phytoplankton availability from punctual (seasonal blooms) or non-punctual (runoffs, anthropogenic enrichment) sources.

All phytoplankton species contain chlorophyll, the primary molecule used for photosynthesis, which allows chlorophyll *a* (chl-*a*) to be used as a general indicator of phytoplankton biomass worldwide (Trotter *et al.* 2008). The well-known optical characteristics of chlorophyll molecules allow for easy detection and quantification of the phytoplankton biomass using optical fluorometric techniques (Röttgers, 2007). Fluorescence readings with field fluorometers have many advantages, such as specificity (no two molecules excite and emit at the same wavelength), simplicity (no treatment is required for many applications and filters can be smudged without affecting accuracy) and speed (readings can be taken on-site in nearly 2 s). Recent technologies use optical readings rather than chemical analysis to measure chlorophyll concentrations in water. Optical fluorometric readings are much more practical, and the scientific community recognizes the readings as a reliable data collection technique (Fujiki *et al.* 2007; Pinto *et al.* 2001; Maxwell and Johnson, 2000) Therefore, fluorescence mapping can provide shellfish growers with a "snap shot" of their leases in terms of its primary production

The primary objective of this study was to assess the phytoplankton biomass distribution throughout a shellfish farm using active fluorescence mapping technology. As a first step towards this goal, our laboratory has begun assessing both the magnitude and persistence of localized features within culture estuaries. We used field fluorometers to describe spatial and temporal patterns in phytoplankton biomass by measuring chl-*a* levels in the farm. Our preliminary results highlight the importance of frequent samplings with field fluorometers to identify persistent features in highly dynamic estuarine environments. The combination of fluorometric data with Geographic Information System (GIS) information can also be used to describe spatial and temporal patterns in phytoplankton biomass.

MATERIALS AND METHODS

STUDY SITE

The mussel farm chosen for this study was a seed collection site located in Lamèque Bay, which is situated in northeastern New Brunswick, Canada. The farm is in proximity to the Shippagan Harbour (Figure 1). In our study, mussel spat was collected on ropes that were arranged in a grid layout to maximize the surface area available for larvae settlement (Figure 2).

CHLOROPHYLL MEASUREMENTS

In-situ active fluorescence measurements

A field fluorometer (model 10-AU) manufactured by the Turner Design© company was used, which has the capability to give real-time fluorescence readings while water is pumped continuously through the instrument. Using the fluorometer, transects were carried out through an aquaculture lease and between longlines to acquire relative fluorescence data and measure the phytoplankton biomass. The results were used to determine whether there was an increase or decrease in fluorescence measured in Relative Fluorescence Units (RFU). Changes in RFU were measured at a chosen water depth (approximately 1.5m) as we evaluated the interior and immediate exterior surroundings of the shellfish farm under study.

Transects and GIS

Sampling transects were repeated several times by boat between the lines and outside the lease on September 13th, 2006 through the course of a tidal cycle (ebb tide and flood tide). Each transect continued for approximately one hundred feet (30 m) outside the lease to be certain to measure any continuous or residual depletion that occurred inside the lease that did not have time to recuperate or stabilize. Our boat navigated across the farm along pre-determined transects. Seawater was continuously pumped from the side of the vessel through the field fluorometer, which was connected to a portable computer (Itronix® Toughbook laptop). Thousands of fluorescence data were collected and related to the geographic latitudinal and longitudinal coordinates. Those coordinates were simultaneously logged using a handheld global positioning system (GPS; Garmin

60CX). Geographic tracks were imputed from the GPS data logger into geomatic software (MapInfo® version 6.5). This allowed us to develop a GIS protocol to map the chlorophyll at the farm. From this data, using MapInfo software, a series of chlorophyll biomass maps were produced, which represent the relative fluorescence levels in color.

RESULTS

Our observations at the mussel seed collector lease demonstrated a fluorescence reduction (Figure 3 and 4) compared to outside of the lease at the same depth. Transects made alongside the longlines of the mussel seed collector lease showed that there was a 0-30% reduction in fluorescence (Figure 5) compared to a significant increase in biomass approximately 30 feet outside the lease. Moreover, the chlorophyll-*a* biomass increased in corridors between the mussel collectors longlines as well. During ebb tides, the depletion appeared to follow the currents and was traceable after passing through the `farm as a "shadow effect." However, we observed that this shadow effect showed irregularities during flood tide. In nearly every transect, especially those conducted during the flood tide, the relative fluorescence values declined alongside the mussel collector longlines, and there were increases in fluorescence in corridors and outside of the lease (Figure 3 and 5).

Following the direction of the flow at ebb tide, the shadow effect was monitored south of the mussel collection lease. In contrast, on the north side of the map, pockets of high phytoplankton biomass were observed, which were brought by the river to the north (Figure 3, *blue and green colored areas*). As shown in Figure 4, during flood tide, a pocket of highly chlorophyll concentrated water is visible at the south of the map that arrives from the outside of the bay, which is close to the ocean (*blue and green colored areas*).

Phytoplankton biomass appeared higher in transects that were made close to the shore than in those made near the channel waterway (Figure 6). When the data was collected during the ebb tide, the phytoplankton concentrations were high towards the inner bay, and they decreased by approximately 22% downstream towards to mouth of the bay, after passing though the farm (Figure 7). However, unexpected areas of high phytoplankton biomass were observed in the middle of the farm, as shown in Figure 3.

The fluorescence transect made closest to the channel showed that there was an increase of phytoplankton biomass in the area, whereas there were no mussel lines in the middle of the lease. On either side of the empty area where the mussel seeds were collected, the fluorescence values decreased (Figure 8).

DISCUSSION

Mussels are well-known phytoplankton grazers, and strong depletions in chlorophyll have been reported above mussel beds (Maar *et al.* 2007). Prior to our study, the majority of studies on the relationship between bivalves and primary production have been undertaken on bivalves in natural benthic environments. Such studies have shown that bivalves, like blue mussels, can reduce phytoplankton biomass (Ogilvie *et al*. 2000). It is important to extend this research to shellfish aquaculture sites to study the interactions between pelagic (or mid-water) growing structure and the environment. Also, comparisons between the reduction of phytoplankton biomass occurring over a natural mussel bed and aquaculture leases would be relevant future research. Additionally, it is important to examine the interactions of floating structures, such as OysterGro© cage or Vexar® bags, which are used for oyster suspension aquaculture. The "shadow effect" that we observed during low tide is an interesting result for shellfish growers. It suggests that if a new lease is to be added at the end of the one surveyed, there would be a potential deficit of food downstream of the first lease.

Areas with high concentrations of phytoplankton that may have been generated by nutrient-rich water running off from the river were also observed. Moreover, studies have suggested that bivalves can promote primary production by converting particulate nitrogen into dissolved inorganic nitrogen, and hence making it available for phytoplankton utilization (Ogilvie *et al.* 2003). Trottet *et al.* (2008) showed that phytoplankton productivity in a mussel farm was enhanced because the mussels helped supplement the concentrations of nutrients locally present. This study indicated that the net production rates of phytoplankton were significantly greater inside the mussel farm than outside the farm (Trottet *et al.* 2008). The ammonia excreted by the mussel seeds may compensate for the phytoplankton uptake by promoting localized blooms around the farm and between blocks. This relationship may explain the augmentation of biomass in

the corridors. Dense populations of suspension feeders play a key role in coastal ecosystems where their large grazing potential strongly influences the plankton community by consuming the phytoplankton and detritus above the seabed. These events drive phytoplankton succession towards smaller, faster-growing species. At the same time, bivalve populations fertilize the phytoplankton community by excreting nutrients (Nielsen and Maar, 2007).

Although our results showed a slight reduction of the phytoplankton biomass at the shellfish farm studied, some publications have suggested that shellfish aquaculture is important for controlling the risk of eutrophication in bays and estuaries. Grazing by bivalves may effectively control phytoplankton biomass (Prins *et al.* 1994). In New Zealand, mussels were shown to have a stabilizing influence on phytoplankton biomass by reducing the high ambient levels during the winter and by slightly increasing the low levels during the summer (Ogilvie *et al.* 2003). Therefore, shellfish aquaculture plays a significant role in controlling eutrophication throughout coastal areas, such as those in South East Asia (Ferreira *et al.* 2007). Shellfish can also control phytoplankton blooms produced by anthropogenic sources of nitrogen. For example, oyster reef restoration has been used in the Chesapeake Bay to control eutrophication (Pomeroy *et al.* 2006). Bivalves accelerate nutrient recycling and provide the water column with dissolved nitrogen, which is the limiting factor for primary production in most aquatic environments (Sarà, 2007; Mazouni, 2004). Oysters contribute to this recycling activity through their excretions, which support the regeneration of primary production (Chapelle *et al.* 2000).

In terms of mapping techniques, remote sensing has been used extensively to provide quantitative information on the distribution of phytoplankton in inland waters. Remote sensing relies on surrogate chl-*a* mapping; however, since chl-*a* is common to nearly all phytoplankton species, it cannot provide any information on the taxonomic composition of the phytoplankton communities (Hunter *et al.* 2008). Fluorescence monitoring can be used to assess live phytoplankton cells, which are able to perform photosynthesis. In theory, fluorescence is used to quantify the total food available to shellfish in a system, but bivalves do not exclusively feed on active phytoplankton. Bivalves also feed on inactive cells, bacteria and protists, which do not fluoresce. Thus,

it would be informative to characterize the organic vs. inorganic matter and classify species in water samples from locations with extremely low or high fluorescence values.

Our preliminary results demonstrate the importance of frequent samplings to identify persistent trends in highly dynamic estuarine environments. Many factors, such as changes in currents and temperature, can cause stress and lead to reductions in food uptake. In future studies, many additional parameters that influence fluorescence and phytoplankton biomass should be examined, such as light intensity, currents, total suspended matter, mussel sizes, filtrations rates, as well as nutrient inputs (enrichment) from river systems and runoffs. Additionally, *in situ* fluorescence readings obtained from mapping surveys should be converted into absolute chlorophyll concentrations (µg/L). Such a conversion requires the collection of water samples and extraction of chlorophyll.

ACKNOWLEDGEMENTS

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FIGURES

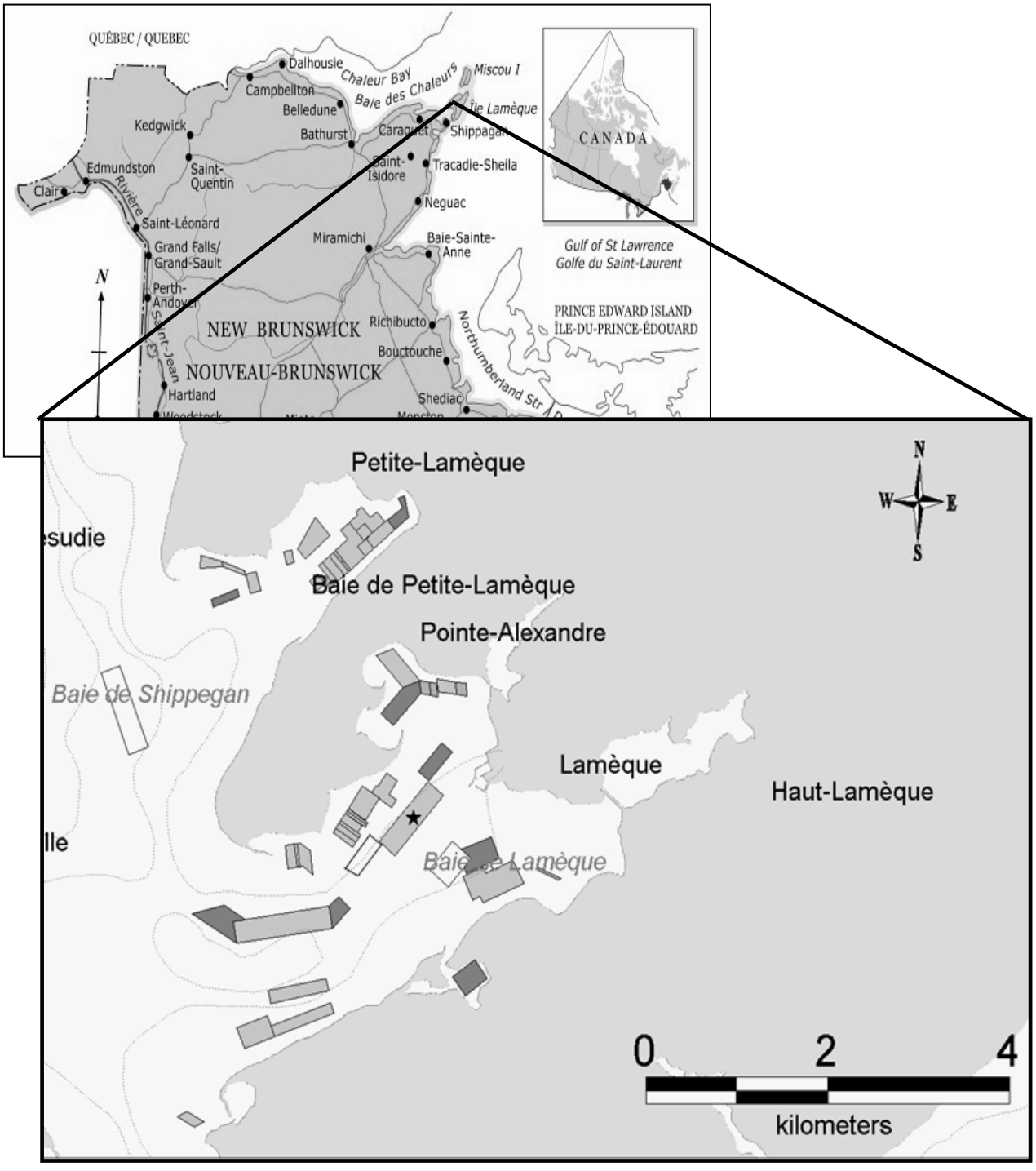


Figure 1 - Mussel collection lease (★) in Lamèque Bay in northern New Brunswick.

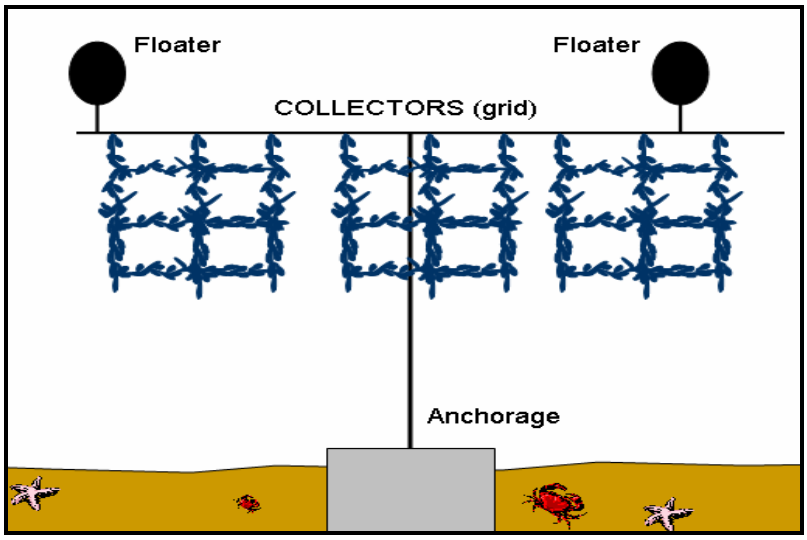


Figure 2 - Mussel seed collectors in a grid layout in Lamèque Bay.

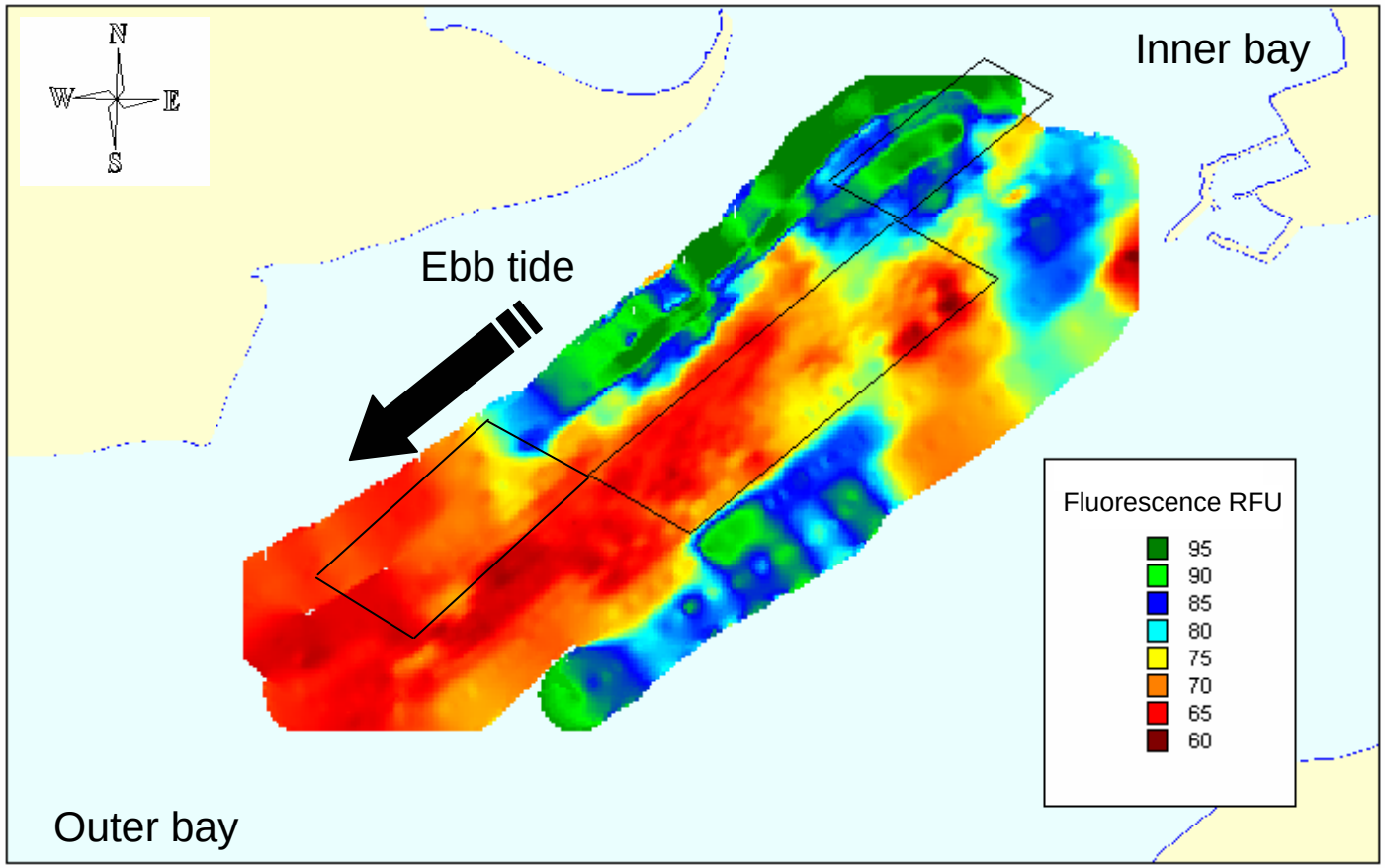


Figure 3 - Fluorescence (RFU) mapping of a mussel seed lease in Lamèque Bay, N.B. on September 13th, 2006 during ebb tide. Rectangles outline the mussel leases layout.

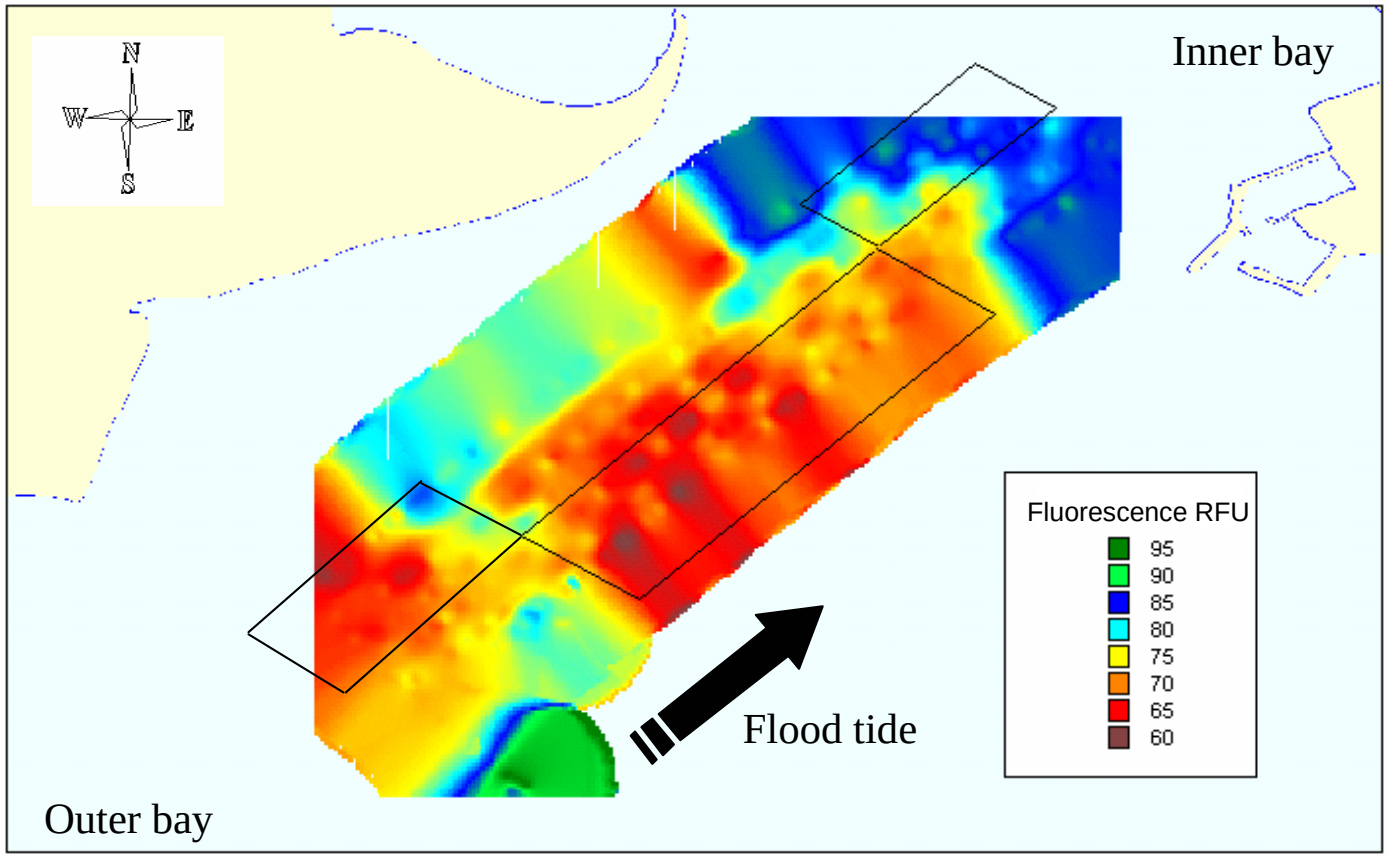
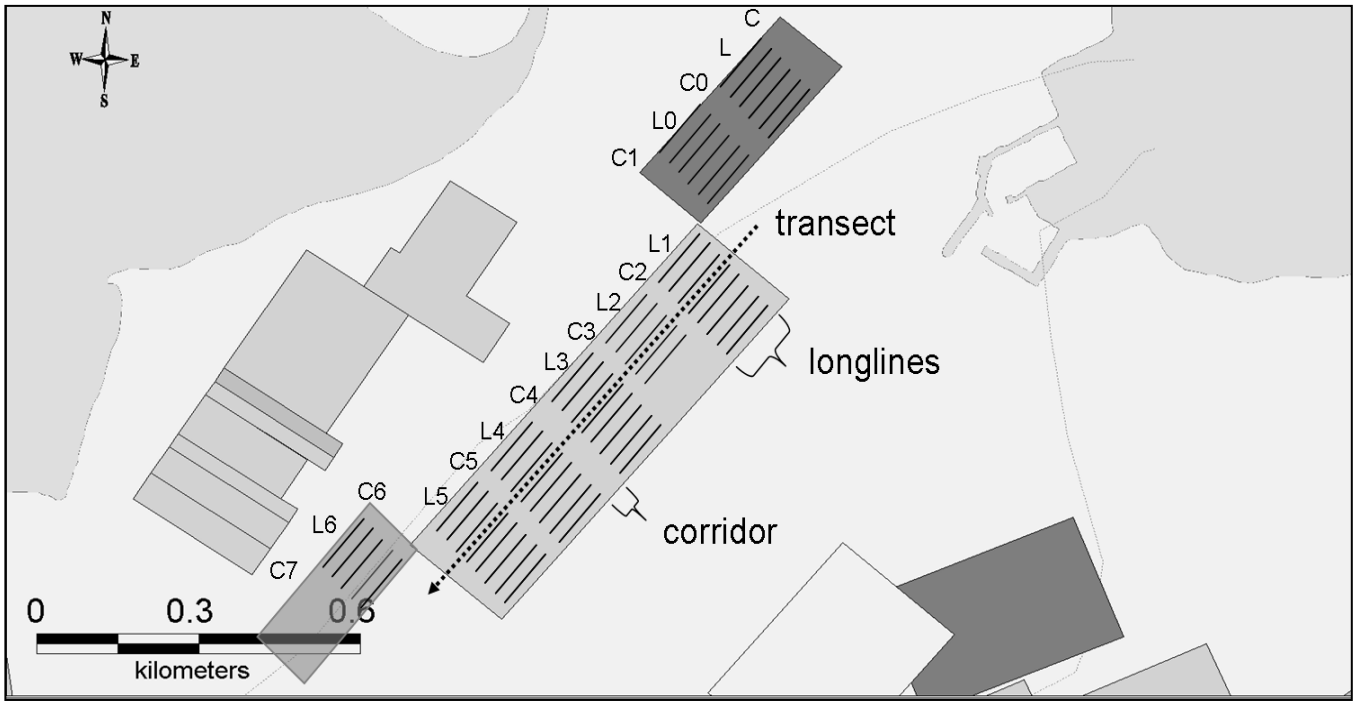


Figure 4 - Fluorescence (RFU) mapping of a mussel seed lease in Lamèque Bay, N.B. on September 13th, 2006 during flood tide. Rectangles outline the mussel leases layout



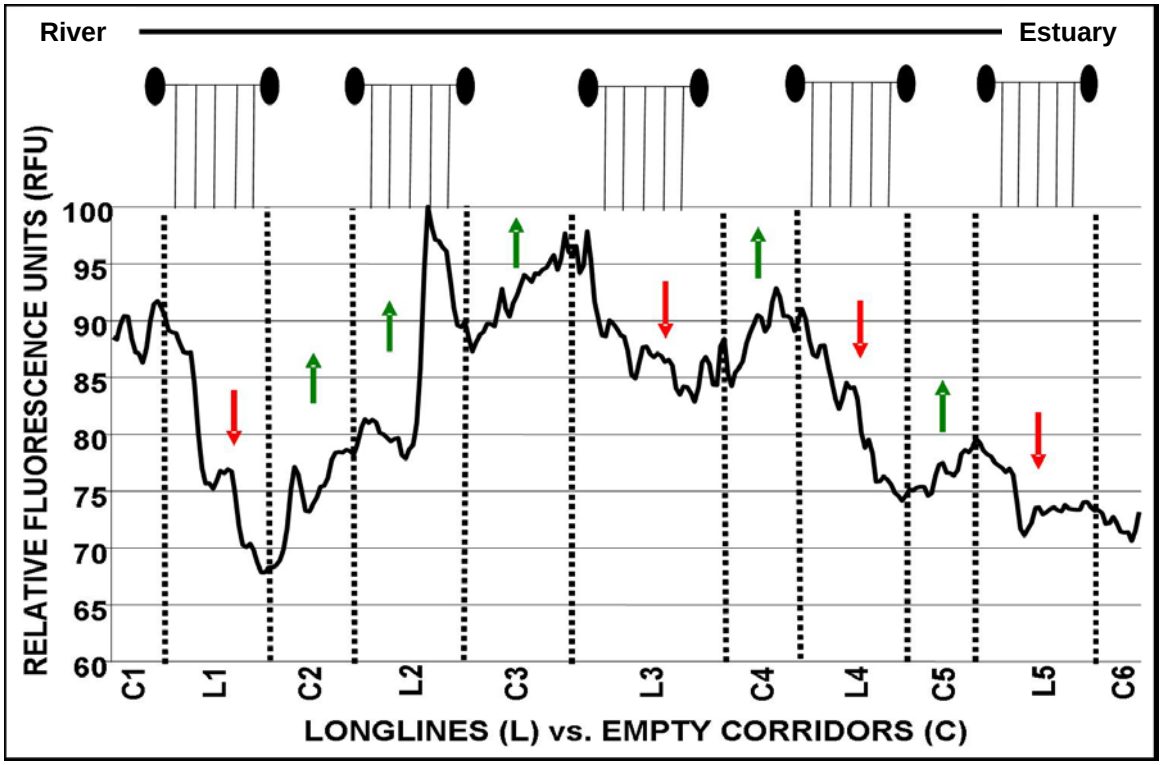


Figure 5 - Longitudinal view of fluorescence (RFU) variations along a transect of a mussel seed lease in Lamèque Bay, N.B.

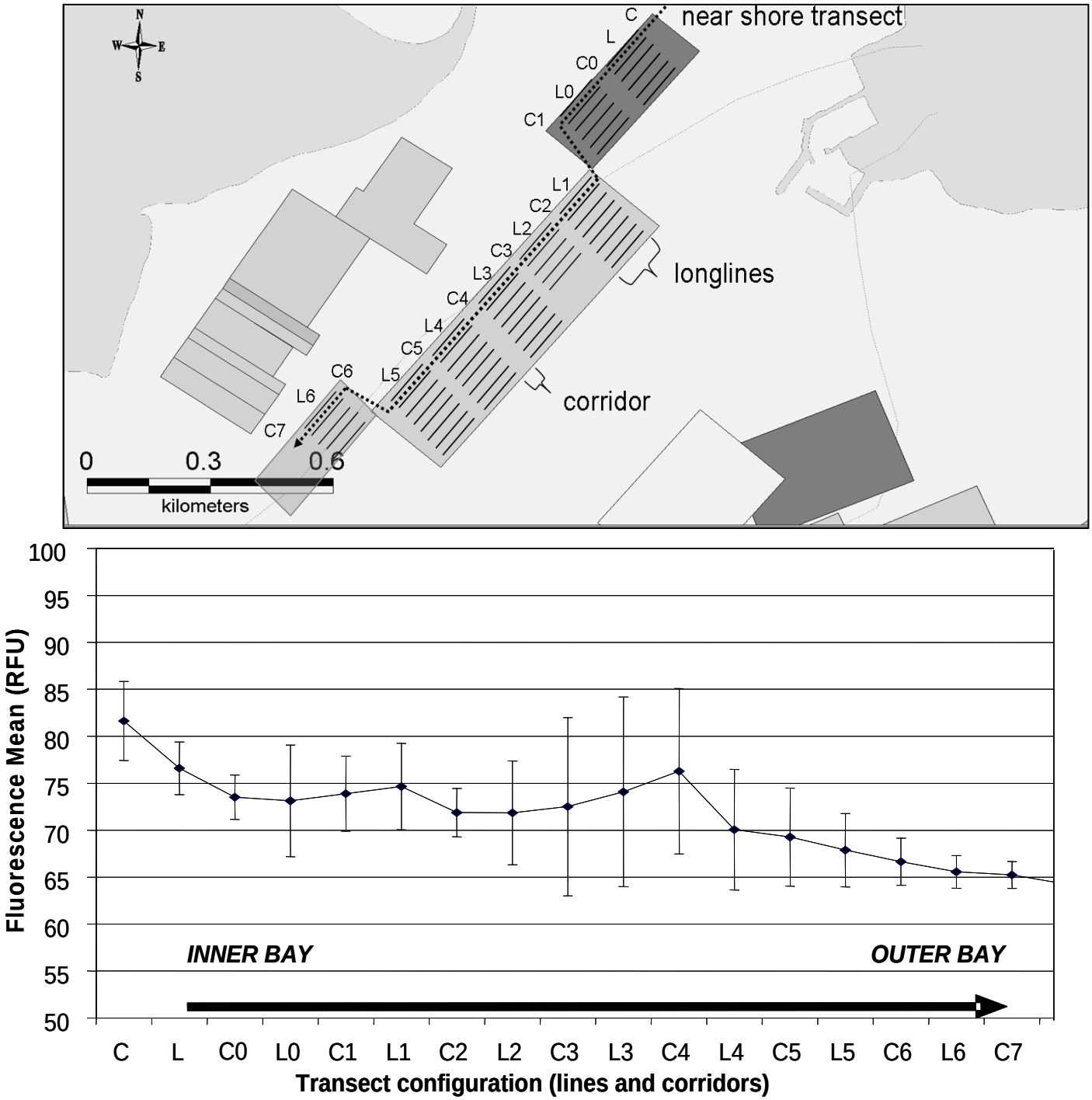


Figure 7 - Fluorescence (RFU) measurements along a transect line close to the shore in Lamèque Bay, N.B. during ebb tide.

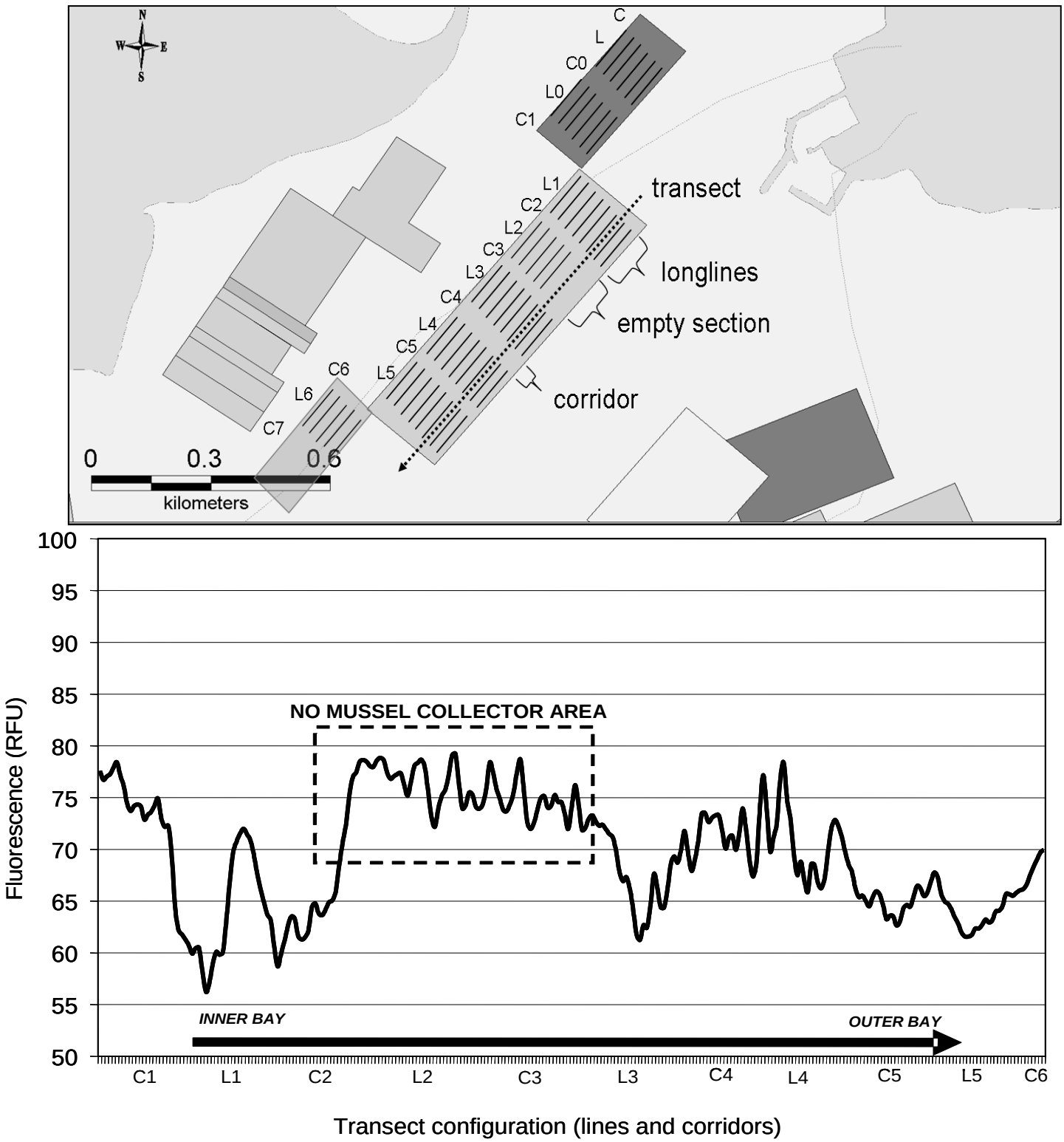


Figure 8 - Fluorescence (RFU) measurements along a transect line close to the navigation channel in Lamèque Bay, N.B. during ebb tide. The empty section is labeled with the boxed area.