Stable Isotope Analyses of Sediment and Invertebrate Samples from Boston Harbor and Massachusetts Bay

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Introduction

Previous studies using stable isotopes of nitrogen and sulfur as tracers have demonstrated the incorporation of sewage-derived materials into the food webs of Boston Harbor and Massachusetts Bay (Tucker et al, 1999; Moore et al, 1996). These studies have also documented the recovery of Boston Harbor as sewage inputs decreased over the past decade. As a continuation and augmentation of these studies, we analyzed the following sediment and biological samples. Figures 1a and 1b presents station maps for all analyses types.

All sample analyses for ^{15}N were performed at the Marine Biological Laboratory's Stable Isotope Laboratory. Analyses for ^{34}S were performed at Coastal Laboratories, in Austin, Texas.

Sediment Samples from Nutrient Flux Stations

Boston Harbor

The δ^{15}N signatures of surface sediments and sediment profiles from two stations in Boston Harbor analyzed previously showed a change over time from light values similar to those measured in sewage sludge to heavier values more similar to those from marine sediments. The implication was that sewage-derived organic matter (OM) in the harbor sediments was being lost over time since sludge disposal ended and other treatment improvements were put in place. In this study, we added two sediment profiles to the existing data from each of these two stations, BH02 and BH03 in the central harbor, and also added profiles from two other harbor stations, BH08A in Hingham Bay, and QB01 in Quincy Bay. These profiles came from May 1998 and October 2000 surveys. They were chosen to further document "recovery" of harbor sediments, to span the most time and to give us the latest results available before sewage effluent was diverted offshore. The Hingham and Quincy bay profiles provided more spatial coverage and a contrast to the central harbor results.

The two endmember values for δ^{15}N that we used previously were a sludge value of 3.3 ‰, from an April 1990 sample, and 6‰ for marine sediments, taken from Massachusetts Bay sediment profiles and literature values. Sediment profiles from Station BH03 taken in September, 1991, before sludge disposal ceased, showed δ^{15}N values of 4‰ in subsurface sediments, suggesting a large contribution of sewage organic matter. Each successive profile from this station increased in δ^{15}N values, becoming more and more similar to "normal" marine sediments. The May 1998 profile at station BH03 nicely filled the gap between previously run samples, and fit with the clear progression towards heavier values with time that we had observed before (Fig. 2a). With the exception of a high value of 6.8‰ at the surface of the October 2000 profile, the October 1998 and October 2000 profiles were similar, both having subsurface values of 6.2‰ and decreasing to 5.2‰ at 10 cm. These results suggest that further depletion of sewage organic matter in these sediments may occur slowly, although values at depth are still relatively light, and further suggest these sediments are nearing values similar to marine sediments.

We observed similar results at Station BH02 (Fig. 2b), although the changes at this station have not progressed through time so smoothly as at BH03. The two new profiles added in this study were similar to each other, and had the heaviest values of the series. However, these values remained for the most part lower than 6‰, suggesting that sediments at this site had not yet recovered to the same extent as had those at Station BH03.

Profiles from the two southern harbor stations (Fig. 2 c and d) had higher values than the profiles from the central harbor stations, and all values were higher than 6‰. Neither station showed a trend towards lower values with depth, as was typical for Station BH03. The profiles from Station QB01 in particular trended in the opposite direction, with heavier values up to 7.5‰ at depth in the core. This pattern is typical in soil profiles where increasing depth corresponds to isotopically heavier, worked-over material (Nadelhoffer and Fry, 1988). Profiles from May 1998 and October 2000 differed little within stations, except for high surface values of 7‰ in October 2000.

Massachusetts Bay

Previous results comparing one nearfield station in Massachusetts Bay (MB01) to the Stellwagen Bay station (MB05) suggested some sewage influence present in the nearfield area of Massachusetts Bay. Cores from Station MB01 in November 1992 showed lighter values (~5.5 ‰) at middepths of the profile than at the surface or as compared to the marine value of 6 ‰ (Fig. 3a). A direct comparison of October 1994 cores from the two stations showed isotope values at MB01 were still lighter than those at MB05 (which were about 6 ‰), but heavier than they had been in 1992. These results suggested that sewage organic materials had been lost from the MB01 sediments in the intervening years, and that these sediments were approaching values similar to what we considered typical marine sediments (those at MB05). Samples run in this study from all three nearfield stations (MB01, MB02, MB03) and the Stellwagen station (MB05) from September 2000 were nearly indistinguishable. All four profiles showed heavier values of about 7‰ at the surface, and around 6.5‰ through the rest of the profile, although values from Station MB05 at 8-10 cm did deviate from the three nearfield stations slightly and were heavier.

The fact that all four stations appeared similar in September 2000, before the bay outfall became operational, suggests a fairly uniform isotopic "setting" in the nearfield and eastern Massachusetts, and a dominance of bay- or region-wide processes in the system. These results should provide a good background against which to assess results from post-diversion samples.

We also analyzed surface sediments from all four stations and all surveys in 1999 and 2000 (Fig. 3 b and c). Del ^{15}N values did not differ greatly between stations, but values from 2000 were somewhat heavier (average 6.9) than those from 1999 (average 6.6). Surface values from MB01 and MB02 were very similar to each other, whereas MB03 was more similar to MB05. MB03 exhibited a seasonal cycle, with lowest values in the spring and fall, and MB05 showed a similar but weaker pattern. When added to previously collected data, the upward trend from 1999 to2000 seemed to continue a pattern that began in late 1992, when most surface δ^{15}N values were lighter, < 6‰. This pattern is consistent with patterns observed in cores collected by USGS (see below).

Our previous assumption for coastal marine background value for sediments was 6‰ (Tucker et al., 1999). This value was assumed from our earlier core profile data and from literature values. The data from this study indicate that a slightly heavier value may be more appropriate. The profile data from all the nearfield stations as well as the Stellwagen station converge on about 6.5‰.

Sediment Traps and Sediment Cores from USGS Stations in Massachusetts Bay

Selected samples from the USGS core sites and long-term monitoring moorings were analyzed in this study to augment our sediment core data and, from the traps on the moorings, to provide information on fresh material being delivered to the benthos. Sediment core samples were from Station 2, which is a sandy site, and Station 3, which is a depositional site similar to our benthic flux stations. The time span covered by these samples was from December, 1989 to May 2002 for Station 2 and October, 1991 to May 2002 for Station 3. All of the results presented for the pre- outfall diversion period were from fall or winter collections. Results from the post-diversion period include two spring collections, May 2001 and 2002.

Sediment trap samples were from two moorings. One, the LNB mooring, is just south of the midpoint of the diffusers, and is well within the nearfield. The other buoy is located east of Scituate. Samples from the May-September, 1999, May-October 2000, and May-October 2001 collections were analyzed from both moorings. In addition, samples from June to October, 1991 from the LNB buy were also analyzed. Sediment trap samples are stored three ways: with formalin or sodium azide as preservatives, and with no poison added. Our first analysis of these samples included samples stored all three ways. The formalin and no poison samples yielded similar results, but the samples with formalin always had a lighter δ^{15}N value and a higher nitrogen content than the non-poisoned samples (see Fig. 5a and 5b). Remineralization of some of the organic matter in the traps during the four months of deployment in the non-poisoned samples would be consistent with both a lower nitrogen content and a heavier δ^{15}N value. Therefore, we think the better samples may be the formalin preserved ones. However, we did not run both types of samples for the complete data set, so we present data from both types of samples. As expected, samples treated with azide were not suitable for nitrogen stable isotope or content analyses. Samples were dominated by the azide itself, resulting in very light isotope values (around 1 ‰) and very high nitrogen content (between 5 and 10%; data not shown).

Two major events occurred over the time these samples were collected. In December, 1992, a major storm battered the area for several days (Dec. 11-16), causing resuspension of bottom sediments in Boston Harbor and the deeper waters of Massachusetts Bay. The second event was the diversion of sewage effluent from Boston Harbor to Massachusetts Bay, which occurred Sept 6, 2000.

Sediment Cores

Data on silver concentration and *Clostridium perfringens* spores from sediment cores suggest that the 1992 storm redeposited harbor sediments mixed with bay sediments in depositional areas of the bay like Station 3 (Bothner, 2001) and that less depositional areas like Station 2 were completely scoured of fine material (Bothner, pers. comm.). The nitrogen stable isotope data from the same cores support this theory completely. Results from Station 3 show a drop in δ^{15}N values from 7‰ (October 1991) to around 6.2‰ in samples collected just after the storm in February 1993 (Fig. 4a). These lighter values were accompanied by an increase in the nitrogen content of the sediments. Both of these results would be consistent with inputs of sewage-derived organic matter. At Station 2, Bothner reports that silver and *C. perfringens* inventories that had been present in the sediments were completely winnowed out by the storm. There was a very sharp drop in the $\delta^{15}N$ signal from an average of 9.3‰ in 1989 and 1991 to 5.8‰ after the storm (Fig. 4b). In addition to the depleted $\delta^{15}N$ signature, the nitrogen content, which is quite low in these sandy sediments (typically between 0.02 and 0.05%), was even lower (< 0.01%), suggesting very little organic material had reaccumulated in the sediments by the time the post-storm sample was taken. The low isotope value suggested that most of this new material was from the harbor.

From 1993 until 2000, the $\delta^{15}N$ values at station 3 continued to show an inverse relationship with nitrogen content, and gradually increased to an average value of 7.5‰ while the nitrogen content dropped. In a depositional, biologically active site like Station 3, the remineralization rate might exceed the deposition rate of fresh organic matter, which would tend to lower the N content and raise the $\delta^{15}N$ signal of sediment organic matter. In contrast, at Station 2, as small amounts of organic matter accumulated in these sandy, low activity sediments, %N and $\delta^{15}N$ followed a roughly parallel pattern.

Samples collected at Station 3 in September 2000, shortly after the bay outfall came online, showed a decrease in the $\delta^{15}N$ signature to 6.2 ‰ This signal has changed little in all subsequent samples through May 2002. These lighter values may be related to inputs of sewage particulates, but other evidence such as silver or *C. perfringens* spore concentrations are needed to support this supposition. The nitrogen content of the surface sediments at Station 3 has increased since the outfall came online, but the increase was delayed in comparison to the shift in the isotope values, possibly because more organic matter needed to accumulate to cause a response in the %N than it did to change the isotope ratio.

At Station 2, the fall and winter post-diversion samples were isotopically lighter than those collected before diversion, showing a pattern similar to that from Station 3 that may be related to sewage influence. However, the spring samples were heavier, indicating a seasonal response at this station not apparent at Station 3. It appears that in these very organic matter poor sediments, small changes in organic matter type or delivery are detectable by stable isotopes, whereas some of these subtle changes may be masked by higher organic content in the depositional sediments.

A few other sediment samples were run for general comparison to the nearfield samples, however they do not correspond directly to other samples because the time of collection was different. Samples from two Cape Cod bay stations in August, 2000, had $\delta^{15}N$ values averaging 6.8 per mil. However their nitrogen contents were quite different, with samples from CCB7 richer in nitrogen (average 0.35%) and those from CCB6 very similar to Station 3 with about 0.14%N.

Two other samples from a station east of Stellwagen Bank (USGS Station GOM8) were also analyzed with the idea of determining the $\delta^{15}N$ value of a “pure” marine sample. Results from this site were quite low in $\delta^{15}N$ (avg 5.7‰) and relatively rich in nitrogen (0.27%). These open ocean values did not seem consistent with those from the more coastal nearfield samples, and in retrospect it seemed obvious that this site was not suitable for comparison to Massachusetts Bay, as they are two separate systems responding to different physical and biological regimes. We will continue to consider our Stellwagen basin site (MB05) as an appropriate reference point for Massachusetts Bay.

Sediment Traps

Results from sediment traps from the LNB mooring, like the results from cores, showed a post-diversion decrease in δ^{15}N (Fig. 5a). The results from the non-poisoned samples, for which we have one extra datapoint, show the pre-diversion samples had values of 6.6 and 7 ‰ (October 1991 and September 1999, respectively) whereas the post-diversion samples had values of 5.1 and 5.4 ‰ (September 2000 and October 2001, respectively). The formalin-preserved samples showed the same trend, but post diversion values were even lower (4.7‰). Again, these light values may be related to sewage inputs, but supporting data on other sewage tracers are needed. Results from the Scituate mooring on the two post diversion dates were very similar to those from the LNB samples. The single pre-diversion sample, however (September 1999) was quite a bit lighter than the LNB samples from the same time, and not much heavier than the post-diversion samples. Because the effluent plume from the harbor outfall as well as the bay outfall travels along the coast in this area, it may be that a significant sewage component has long been present in the POM of this area. Again, supporting data are needed. Trap values from both moorings were lighter in general than surface sediment values most likely due to the fact that this is "fresher" material.

Mussels and other Biological Samples collected from Nearfield Hardbottom Sites

On September 20, 2000, samples were collected from four drumlin tops, two north and two south of the bay outfall diffusers. The following samples were collected:

Station T7-1 (north of diffusers, potential post-diversion reference): *M.* m*odiolus*, *Mytilis edulis*, *Agarum* (kelp), *Ptilota* (red macroalga)

Station T1-3 (north of diffusers): *Modiolus modiolus*

Station T4-4 (south of diffusers): *Modiolus*, and an encrusting corraline alga

Staton T8-1 (south of diffusers, potential post-diversion reference): *Modiolus*

All of these samples are considered "baseline" or pre-discharge. Although discharge began September 6, 2000, we feel insufficient time had passed since discharge began for the samples to be affected.

At each of these stations, several individuals of *Modiolus* were collected. *Modiolus* is a filter feeder, and stable nitrogen isotope analysis of its tissues provides an integrated signal of water column particulate organic matter (PON). The addition of the sulfur analysis provided a sensitive indicator of freshwater versus seawater inputs. Freshwater and marine endmembers are separated by nearly 20‰, with seawater sulfate having a δ^{34}S signature of about 20 ‰ (Peterson and Fry, 1987).

We analyzed 3 *M. modiolus* individuals from each site for nitrogen and sulfur isotopes, choosing individuals that spanned the size range collected at each site. These sizes, as measured by shell length, ranged from 9.7 to 12.5 cm. We also analyzed two other individuals, one fromT1-3 (11.3 cm) and one from T7-1 (13.1 cm; this was an unusually large individual) that each had a large barnacle on its shell. We also analyzed the barnacles. At Station T7-1, some of the mussels in the collection were *M. edulis*. We analyzed two of these individuals to compare to *M. modiolus* from the same site. AT station T4-4, some heavily encrusted rocks were also collected. We sampled encrusting corroaline algae from these rocks, and also took the opportunity to sample some hydrozoans, and encrusting sponges.

The $\delta^{15}N$ signature for *M. modiolus* across all four sights varied by only 1 ‰, ranging from 9.2 to 10.2 ‰. The $\delta^{34}S$ values ranged between 14.8 and 16.5 ‰. By interpreting either isotope alone, these samples would be indistinguishable, especially by using ^{15}N alone. However, a dual isotope plot clearly separates all these samples by station, albeit within a small range of isotope values. This was a surprising result, and is derived largely by the difference in the sulfur signal. The most northerly station, T7-1, had the most enriched sulfur signature, while the most southerly had the most depleted. This pattern may reflect freshwater input from the harbor area and the prevailing north to south current pattern in the bay. In terms of $\delta^{15}N$, Station T8-1 was clearly different than the other three stations, whereas Station T7-1 and T4-4 were indistinguishable from each other.

A correlation between individual size and $\delta^{15}N$ was not apparent when all the data were analyzed together; however, when analyzed site by site, there was a strong relationship at two of the four stations, T1-3 and T7-1 ($P < 0.01$, r2=0.99, n=4). At station T4-4, the linear regression also had a very high r^2 (0.94, n=3) but the relationship was not significant. At station T8-1, there was no relationship. There was also no relationship between size and $\delta^{34}S$ for all the data or site by site.

M. edulis from T7-1 had values slightly lower than *M. modiolus* at any of the sites, averaging about 9.0 ‰ (±0.1 ‰) (Fig. 7). *M. edulis* feeds from a smaller size fraction of the PON, and this may account for the difference. For comparison, samples of *M. edulis* collected in October, 1994, from navigation buoys within the nearfield, BG Buoy and Boston Buoy, had $\delta^{15}N$ signatures of 7.1 and 6.9 ‰, respectively (Tucker et al, 1999).

An interesting aside was the finding that barnacles attached to shells of *M. modiolus* were depleted in $\delta^{15}N$ by about 1.8‰ as compared to their hosts (Fig. 7). This is probably another example of differences in size selection of food items among filter feeders.

Results from the two algae samples and the sample from the encrusted rocks at T4-4 are shown in Figure 7. The kelp (Agarum) sample from T7-1 had a low $\%^{15}N$ of 4.5‰, whereas the red alga (Ptilota) had a much higher signature of 8.2 ‰. These differences are probably due to differences in fractionation of nitrogen compounds during uptake and metabolism. The red alga had some calcareous encrustations among its fronds, and although we tried to avoid them as much as possible in the sample, they may have contaminated the algal signal. The kelp, on the other hand, was clean. The encrusting corraline alga scraped from the rocks collected at T4-4 had an average $\delta^{15}N$ value of 7.7 ‰, but this sample was likely contaminated with other organic matter (S.D. for analytical reps = 1.1 ‰). Also, it was difficult to collect enough sample material from the rocks for analysis. [Further analysis of encrusting algae may need to be done using another technique, such as analyzing the ^{15}N in extracted chlorophyll.] The hydrozoans and the encrusting sponge from the same rocks had similar $\delta^{15}N$ signatures, 7.2 and 7.8 ‰, respectively.

Archived Invertebrates

Samples of two benthic amphipods, *Leptocheirus pinguis* and *Ampelisca spp*., taken in 1993 from a site in Boston Harbor previously used for sludge disposal revealed differences in their stable nitrogen isotopic signatures. *L. pinguis* had a δ^{15}N signature of 3.6‰, which was very similar to the value we had measured for sludge (3.3‰), whereas *Ampelisca* had a δ^{15}N value of 6.4‰, more similar to typical values for PON. This difference was consistent with the different feeding strategies employed by the two amphipods, and suggested that a large portion of the diet of *L. pinguis* was derived from sludge, whereas *Ampelisca* obtained most of its food from the water column. In this study we proposed to expand on these findings by analyzing preserved amphipod samples from the Benthic Monitoring Study. These samples would possibly provide a long term record of changes in food sources to the benthic community at this site in Boston Harbor (Benthic Monitoring Station T03 and Benthic Nutrient Cycling Station BH03), over the time that sewage treatment was improved. We also analyzed amphipod samples from the nearfield area of Massachusetts Bay to provide a contrast to the harbor samples, to provide a baseline for the bay before the ocean outfall became operational, and to provide a contrast with the hardbottom samples also analyzed in this study.

Boston Harbor

We analyzed August samples from harbor station T03 of *Ampelisca abdita* (each year from 1991-2000), *Leptocheirus pinguis* (each year from 1992-2000), and *Unciola irrorata* (each year from 1992-1996, 1998, 2000). Bay samples were from August collections of *U. inermis* at stations NF04 in all but three years from 1992-2000, and at NF13 in all but two years from 1994-2000. In addition, samples of *U. irrorata* from the two occasions it was collected from Massachusetts Bay stations were analyzed (from Aug. 1998 at NF04 and August 1999 at NF13). Samples were composites of 5 individuals except for three samples for which five individuals were not available [*U. inermis* samples from NF04 in Aug 1992 (1 individual) and August 1998(3 individuals), and from NF13 in August 1994(2 individuals)]. Analytical precision was less than 1%, and sample replication was at worst ± 6%, or about 0.5‰.

At the time of collection, samples were preserved in formalin with Rose Bengal as a staining agent, and later transferred to ethanol. Individuals were hand picked from the original collection vials by Isabelle Williams (ENSR), and transferred to MBL, where they were dried and ground as well as possible to homogenize. The small sample size and inherent difficulties in homogenization may have led to some sample variability. Also, preservation techniques may have caused deviations from "fresh" values (Bosley and Wainright, 1999); however, as all of these samples were treated similarly, trends in the data over time should not be affected.

Del ^{15}N values for *A. abdita* from station T3 ranged from 7.0 to 9.1 ‰, with heaviest values occurring in 1993. For *L. pinguis*, values ranged from 5.8-7.8 ‰, also with heaviest values in 1993. The range of values for *U. irrorata* fell between that of the other two amphipods, and was narrower; 7.6-8.3 ‰.

These values for archived harbor amphipods *L. pinguis* and *A. abdita* were on average about 3 and 2 ‰ heavier, respectively, than those we had previously reported (Tucker et al, 1999). Therefore, we are not able to fit our earlier results into the pattern we observed during this study. The reason for the large difference is unclear. The early samples were dried fresh, so we may be seeing a preservation effect. Also, the early samples were of a single individual, and therefore may not have been representative. The 1993 *L. pinguis* may have had a gut full of sediment. However, because the difference in early results and these results are similar for both species, it seems more likely that the difference is not one of methodology. Rather, it may be a seasonal difference. The samples from the previous study were collected in late February, 1993, whereas all of the samples in this study were from August collections. We chose August samples over the other available samples (from April) because of higher abundances and because August samples should integrate over a large part of the productive season. What we may have inadvertently found in this comparison is a large seasonal shift from a winter dependence on older, benthic material, and a summer dependence on fresh material from primary production. However, this is purely speculative, and we are left with interpreting the current results apart from the previous ones.

Del^{15}N values for *A. abdita* were on average about 1.5‰ heavier than those of *L. pinguis*. Although this difference does not represent a full trophic level difference, it does seem to indicate that the two species are using different food sources, with *A. abdita* more dependent on benthic versus water column sources. *U. irrorata* values were always higher than those for *L. pinguis*, but usually lower than for *A. abdita*, suggesting *U. irrorata* may utilize both water column and benthic food sources (see Fig. 8a).

We observed a temporal pattern in the results for *A. abdita*. Del^{15}N values increased from 1991 to 1993, and decreased after. A similar, but more variable pattern was also observed for *L. pinguis*. For *U. irrorata*, there was no discernable pattern with time. The decrease with time from 1993 to 2000 was highly significant for *A. abdita* ($P < 0.01$; $r^2 = 0.74$; Fig. 8b), and apparent but weaker for *L. pinguis* ($P = 0.06$, $r^2 = 0.47$; Fig 8c). (Regressions against body length were insignificant.) Unfortunately, without knowledge of the δ^{15}N signature of the PON or DIN through this time, we cannot speculate on what these changes mean other than that it appears there has been a shift in δ^{15}N of the food source of these animals.

Massachusetts Bay

The range of δ^{15}N values from nearfield amphipods was narrower than observed for harbor amphipods and lower values such as were measured for *L. pinguis* were absent. At station NF04, δ^{15}N for *U. inermis* ranged from 6.7-8.9‰ , with lowest values in 1992 and highest in 2000. In contrast, values for *U. inermis* at NF13 varied only between 8.0 and 8.6‰. The two samples of *U. irrorata* from nearfield stations NF04 and NF13 were similar to harbor *U. irrorata* samples; both had δ^{15}N values of 8.2‰ (Fig. 8a)

In the nearfield, results for *U. inermis* showed a temporal pattern at station NF04. The pattern was opposite the one we observed in the harbor; that is, values increased over time from 1992-2000 ($P = 0.01$, $r^2 = 0.73$). At station NF13, where samples were only available from 1994-2000, there was not a significant trend with time. Again, without knowledge of PON and DIN δ^{15}N signatures, or other ancillary data, we cannot give a specific reason for the change, nor for why the patterns at the two stations differed.

Conclusions

The recovery of Boston Harbor sediments from decades of sewage disposal was clearly documented by changes in the stable nitrogen isotope ratios in sediment organic matter. In particular, the changes with time at Station BH03 since sludge disposal ended have been dramatic. The most recent results added by this study show that the harbor continues to change towards typical offshore marine conditions, however if we use Stellwagen basin values as a target, we might expect further changes now that the effluent is being diverted offshore.

Sediments in the nearfield have also experienced notable changes in their $\delta^{15}N$ values over the past decade. The deposition of harbor sediments onto a nearfield depositional site as well as the complete winnowing of fines from a sandy site after the December, 1992 storm were clearly evident in the isotope signal, and correlated well with silver and *C. perfringens* data (Bothner, 2001). Such correspondence with other data is very important in interpreting stable isotope results, especially where distinct and well-separated endmember values are unknown or unavailable. For instance, the change towards heavier values in $\delta^{15}N$ in surface sediment from USGS station 3 corresponded inversely to changes in the nitrogen content of the sediment, leading to an interpretation about relative rates of organic matter supply versus remineralization that we would not have made in the absence of the %N data (and should in any case be considered speculation). The change in $\delta^{15}N$ in late 2000 back towards lighter values corresponds in time to the start-up of the bay outfall, but without other data on other sewage tracers, for example, we can only say that there has been a change. However, as nitrogen is so important in food web considerations, simply knowing there has been a change may be valuable information.

Sediment profiles from the nearfield have also shown change over time. However, the three nearfield stations and the Stellwagen (reference) station had nearly indistinguishable profiles in September 2000. These results should provide a very good reference point against which to assess any deviations among stations we might observe at later dates.

Stable isotope results in *M moliolus* from four hardbottom sites fell within a narrow range (within about 1‰ for nitrogen and less than 2‰ for sulfur) that should provide a good reference point for future analyses. Within this range, however, some subtle but interesting differences were observed. Dual isotope plots of δ^{15} N and $\delta^{34}S$ from *M. modiolus* collected at four hardbottom sites revealed differences among sites that would not have been evident from the nitrogen isotope alone. The difference in the sulfur isotope was most important in separating the sites, and followed a north to south gradient, with the more northern sites showing the more marine signal. Station T8-1 was different from the other three sites in both its nitrogen and sulfur isotopes. At two sites, $\delta^{15}N$ values showed a significant relationship with the size of the individual (as measured by shell length). In addition, analyses of *M. edulis* and an unidentified barnacle revealed somewhat lighter $\delta^{15}N$ values than in *M. modiolus*, suggesting differences in particle size selection among various filter feeder that could be important in assessing the entry of sewage particulates into food webs.

The results from harbor and bay amphipod samples raise new questions about changes in their food sources. In general, *L. pinguis* had the lighest $\delta^{15}N$ values of the three amphipod species analyzed from the harbor, possibly indicating a closer connection to the sediments. *A. abdita* had the heaviest values, with *U. irrorata* in between. In the harbor, *A. abdita* got heavier in the two years following the cessation of sludge disposal, and then decreased, significantly, with time thereafter. *L. pinguis* showed a similar but weaker pattern, whereas *U. irrorato* showed very little change with time. These changes were in the opposite direction of changes we observed in surface sediments. All of these samples were collected in August, and should reflect the growing season. If these results are more indicative of POC derived from water column productivity, then they present interesting questions about changes that were occurring in the phytoplankton, which could be related to dominant species or may be related to the dissolved inorganic nitrogen pool. Although harbor concentrations of nitrogen did not change significantly until outfall diversion, the isotopic signal of the DIN may have changed as treatment processes changed.

Amphipod samples from one station in Massachusetts Bay, Station NF04, showed a change toward heavier isotope values with time consistent with changes observed in depositional sediments, but with a difference in $\delta^{15}N$ of about 1‰. This difference is typical for suspended particulates and sediment organic matter, and is consistent with a water column food source for *U. inermis*. Indeed, changes in water column productivity are implicated in the sediment signals. However, the amphipods that exhibited the trend with time were from a station some distance from the sediment collection sites, whereas the amphipods from the station (NF13) in close proximity to the sediment sites did not show a significant trend with time. The lack of a trend may have been caused by the absence of early data from this site, because results from later sample collections were similar between the two sites. Because of the similarities, it seems likely that a significant divergence between these two sites, possibly related to differing positions in the prevailing north to south current regime in the bay and/or proximity to the outfall diffusers, would be detectable.

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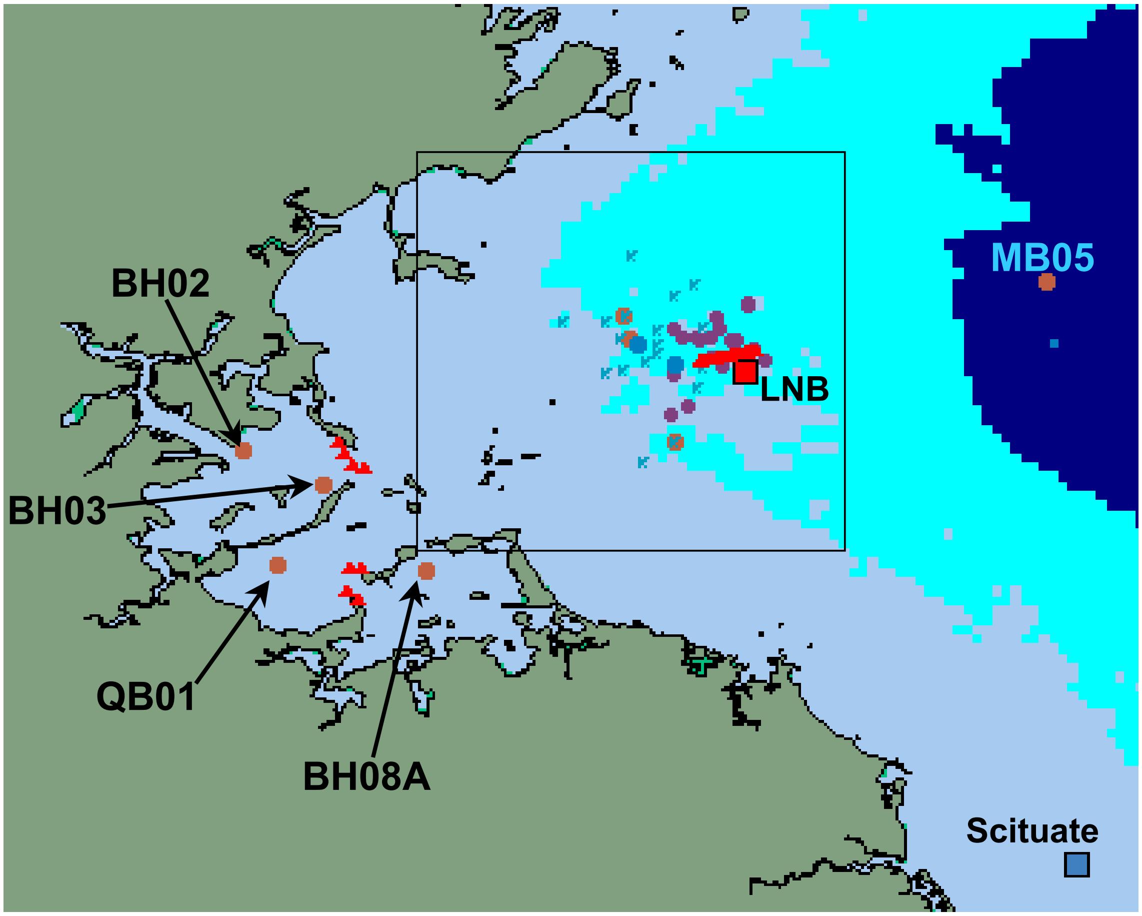


Figure 1a. Expanded station map showing harbor sediment and amphipod collection stations BH02 (T2) ,BH03 (T3), QB01, and BH08A , sediment station MB05 in Stellwagen Basin, and the location of the USGS long-term monitoring buoys in the nearfield (LNB) and near Scituate, MA. Figure 1b. on the next page shows an enlargement of the area within the box that includes sediment and hardbottom sampling stations. The red triangles designate effluent outfalls.

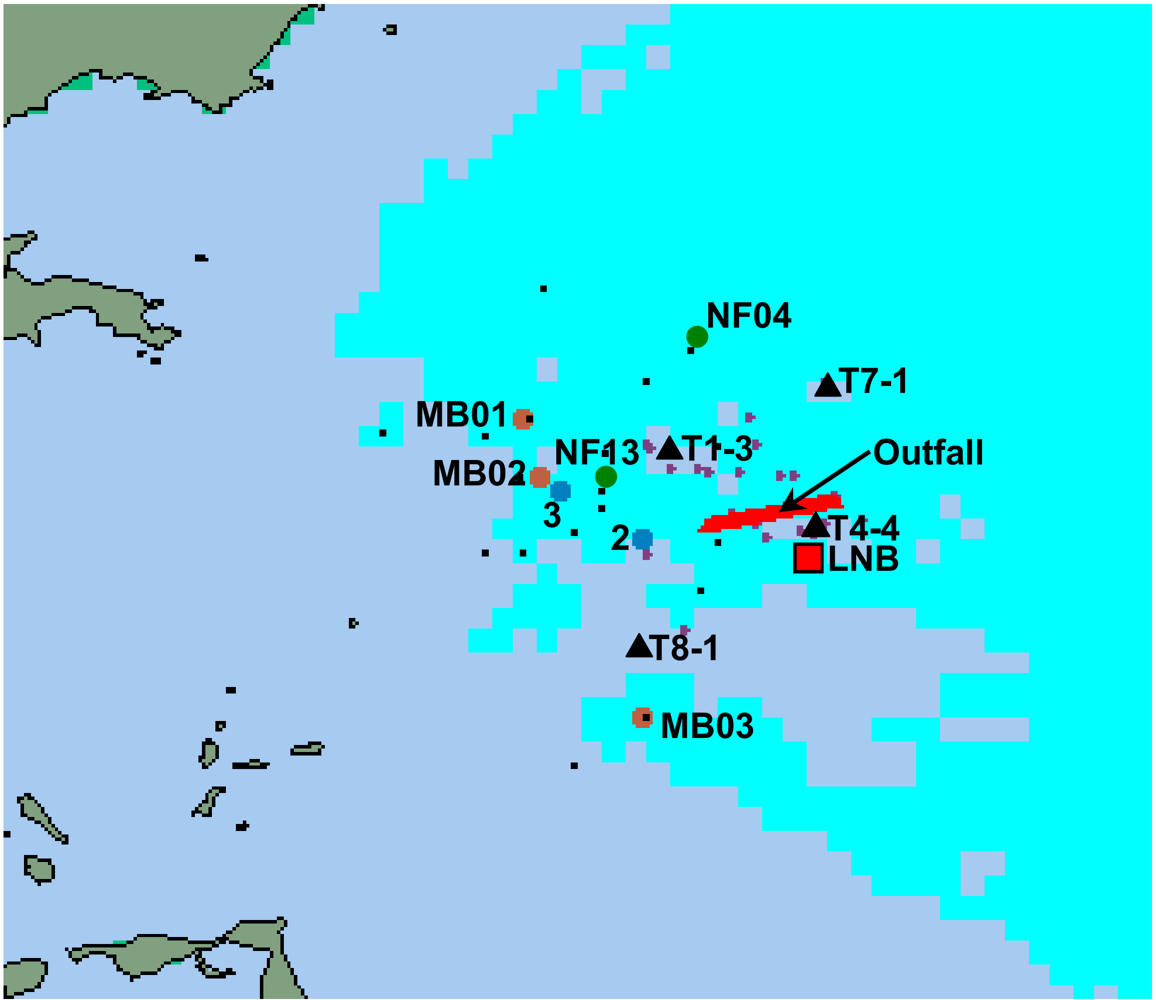


Figure 1b. Blowup of box in Fig. 1a. showing nearfield stations where sediment samples were collected: Benthic Flux stations MB01, MB02, and MB03 (orange-brown circles) and USGS long- term monitoring stations 2 and 3 (blue circles). Stations where invertebrate samples were collected were Benthic monitoring stations NF04 and NF13 (green circles) and Hardbottom stations T7-1, T1-3, T4-4, and T8-1 (black triangles). The USGS long-term monitoring buoy (LNB; red square) and the bay outfall diffusers (red triangles) provide reference to Fig. 1a.

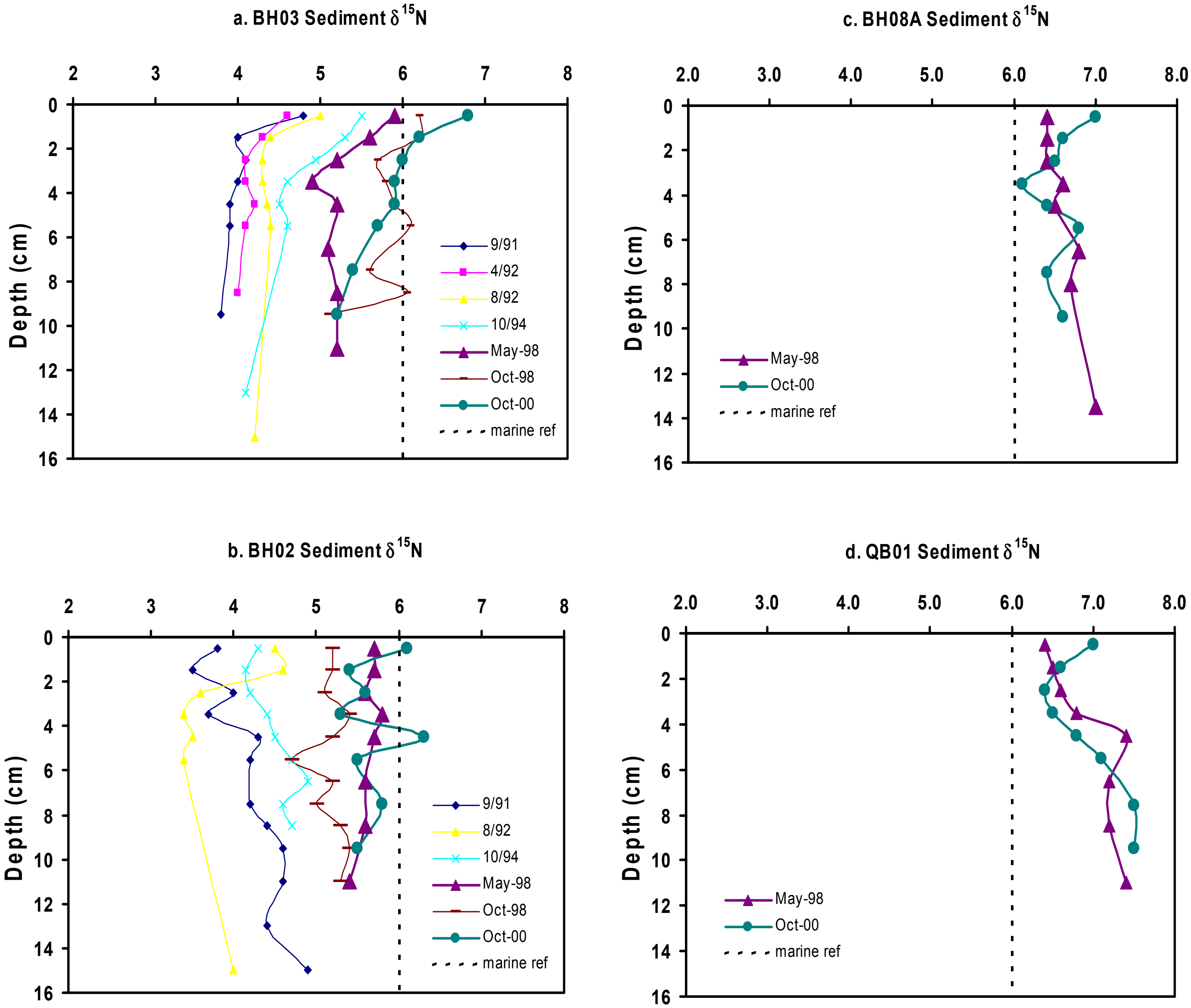
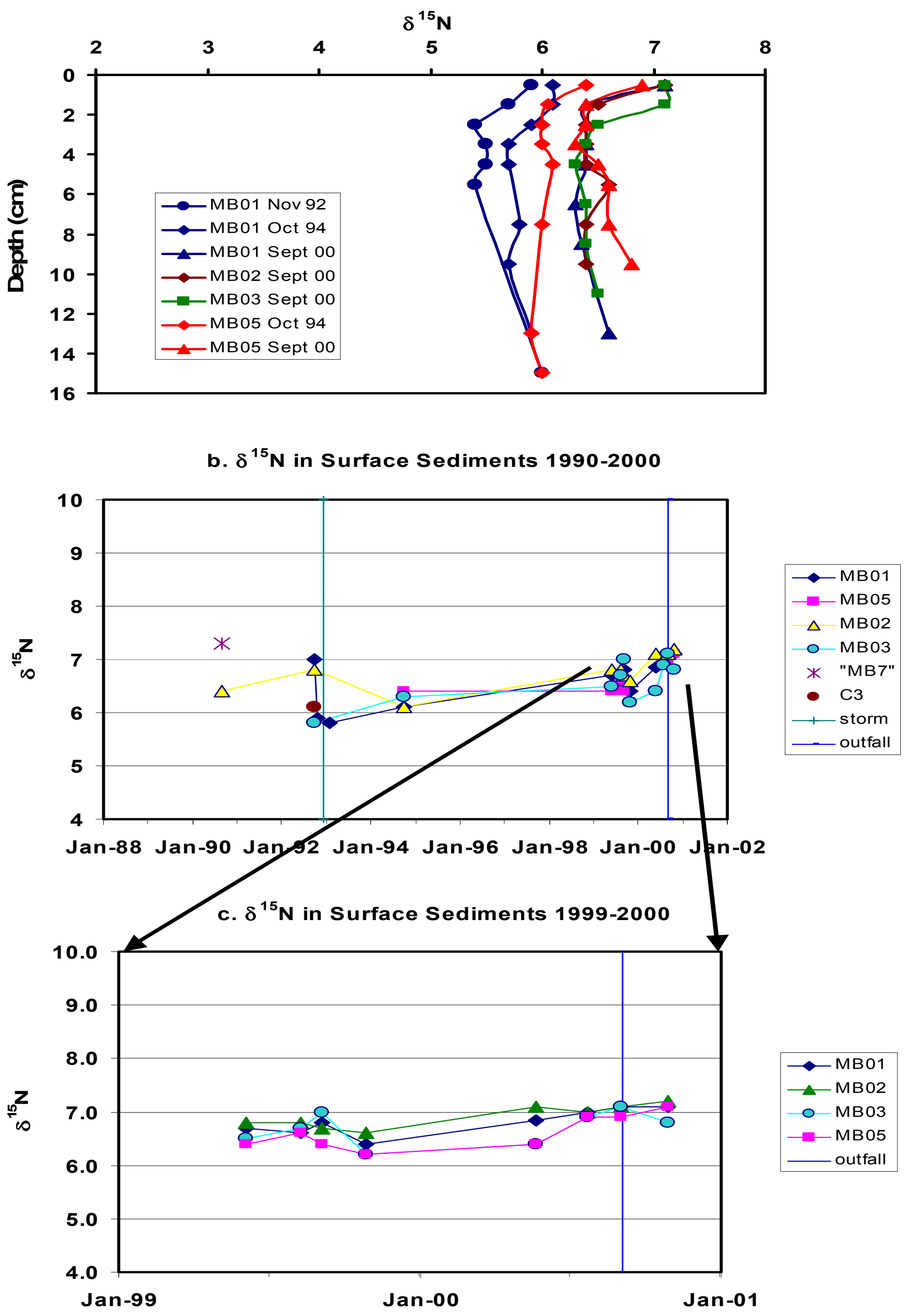


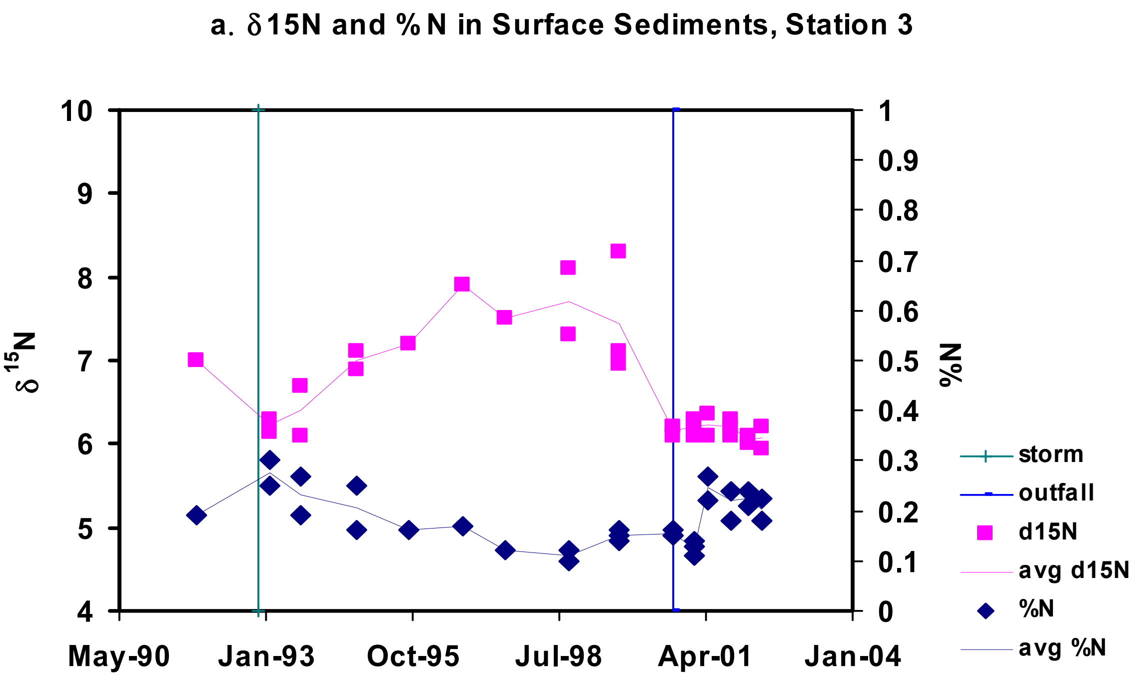
Fig. 2. δ^{15}N profiles from Boston Harbor Stations a.) BH03, b.) BH02, c.)BH08A, and d.) QB01. The May 1998 (purple triangle) and October 2000 (blue-green circles) profiles are data produced in this study (heavier lines and larger symbols). The dashed line at 6‰ represents the marine endmember value used in the previous study.


a. Mass Bay Sediment Profile δ ^{15}N

b. δ ^{15}N in Surface Sediments 1990-2000

c. δ ^{15}N in Surface Sediments 1999-2000

Fig. 3. δ ^{15}N in Massachusetts Bay sediments. a.) Sediment profiles showing similarity in September, 2000. b.) Surface sediments from Mass Bay stations from 1990 through 2000. The results from this study are from 1999-2000, shown expanded in c. In b. the vertical line at December, 1992 marks the Atlantic Coastal Storm. In b. and c. the vertical line at September 6, 2000 marks the date the outfall came on line.



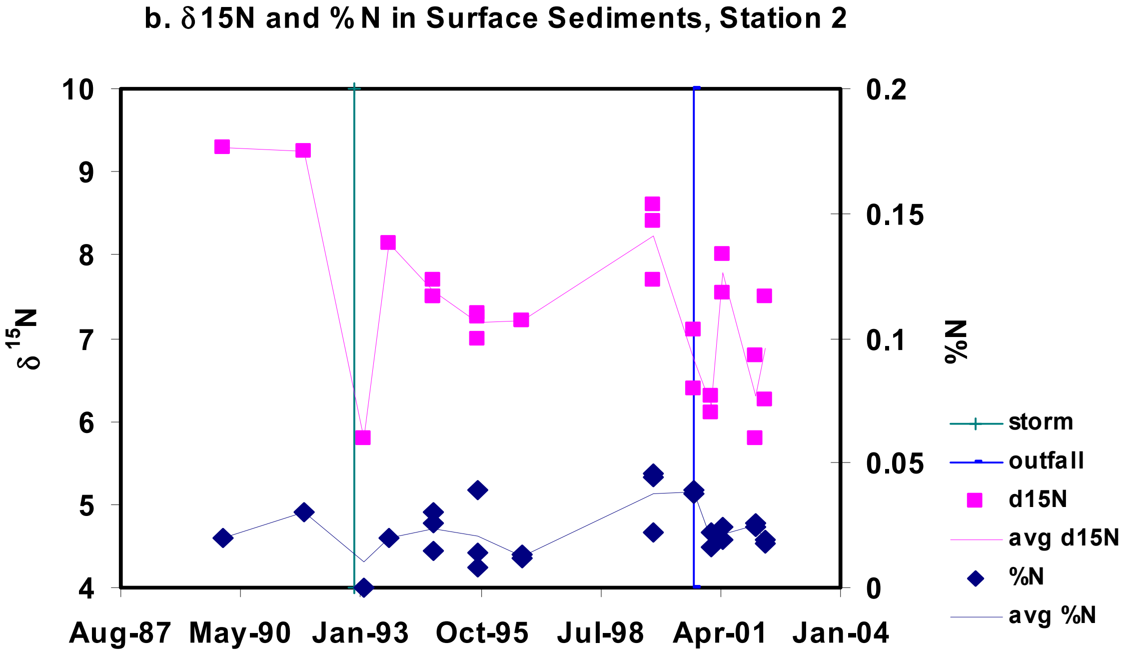
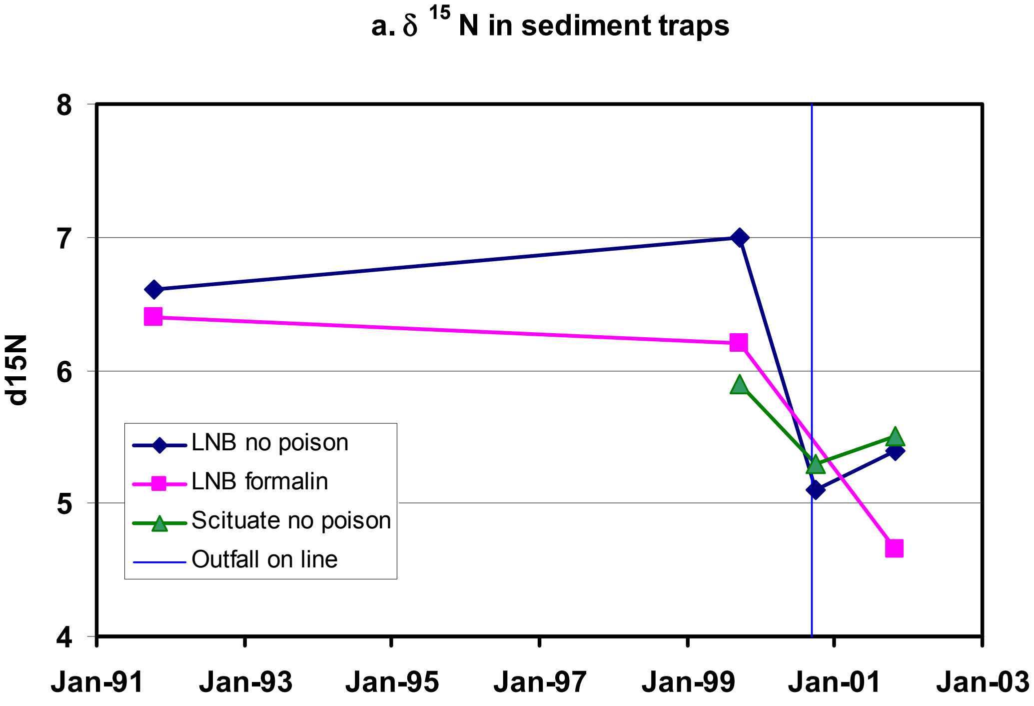


Fig. 4. δ^{15}N and % N in surface sediments at a.) USGS Station 3 and b.) USGS Station 2. the vertical line at December, 1992 marks the Atlantic Coastal Storm, and the line at September 6, 2000 marks the date the outfall came on line.



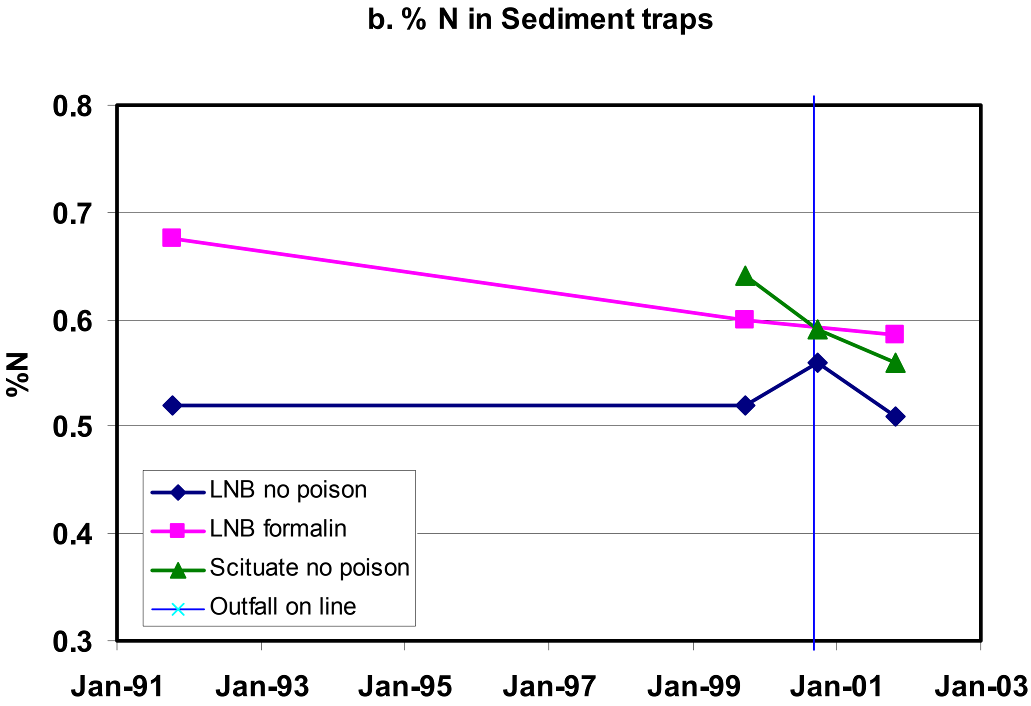
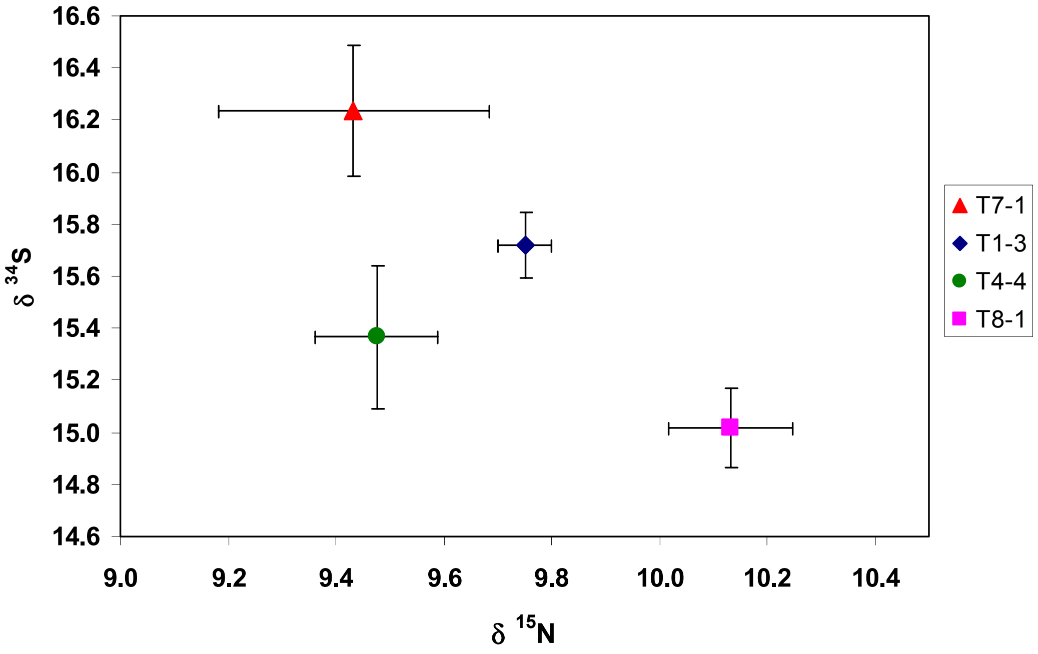


Fig. 5. Sediment trap (a) $\delta^{15}N$ and (b) %N from the LNB and Scituate moorings.

a. δ^{15}N and δ^{34} S in *M. modiolus*



b. Shell length vs δ^{15}N in *M. modiolus*

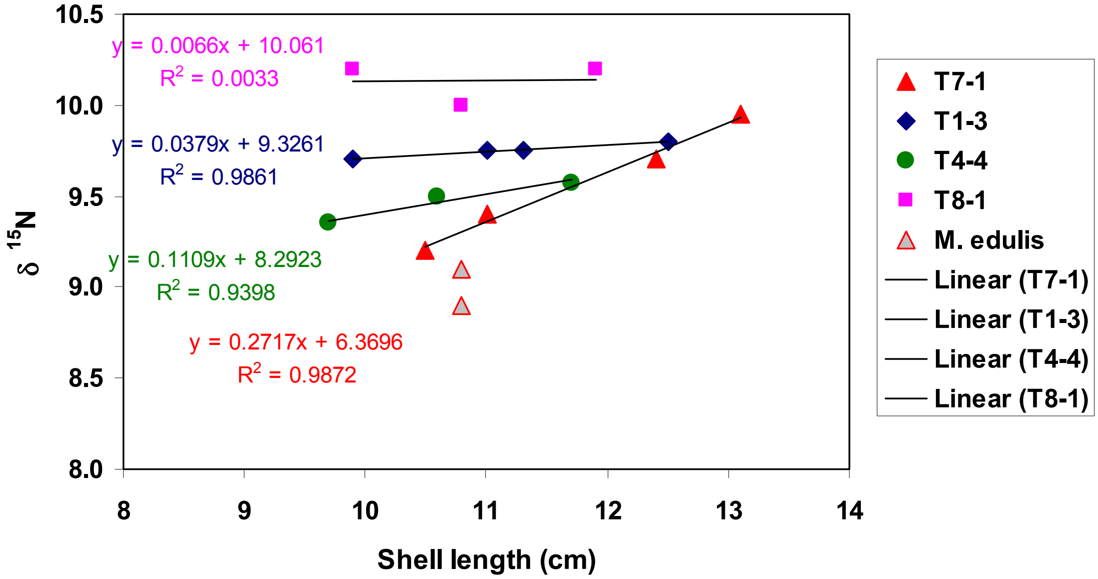


Fig. 6. a.) Dual isotope plot, δ^{15}N vs δ^{34}S, of *M.modiolus* from nearfield hardbottom sites. b.) Correlation of δ^{15}N with shell length in *M. modiolus*. Regressions were significant at T7-1 and T8-1 only; see text.

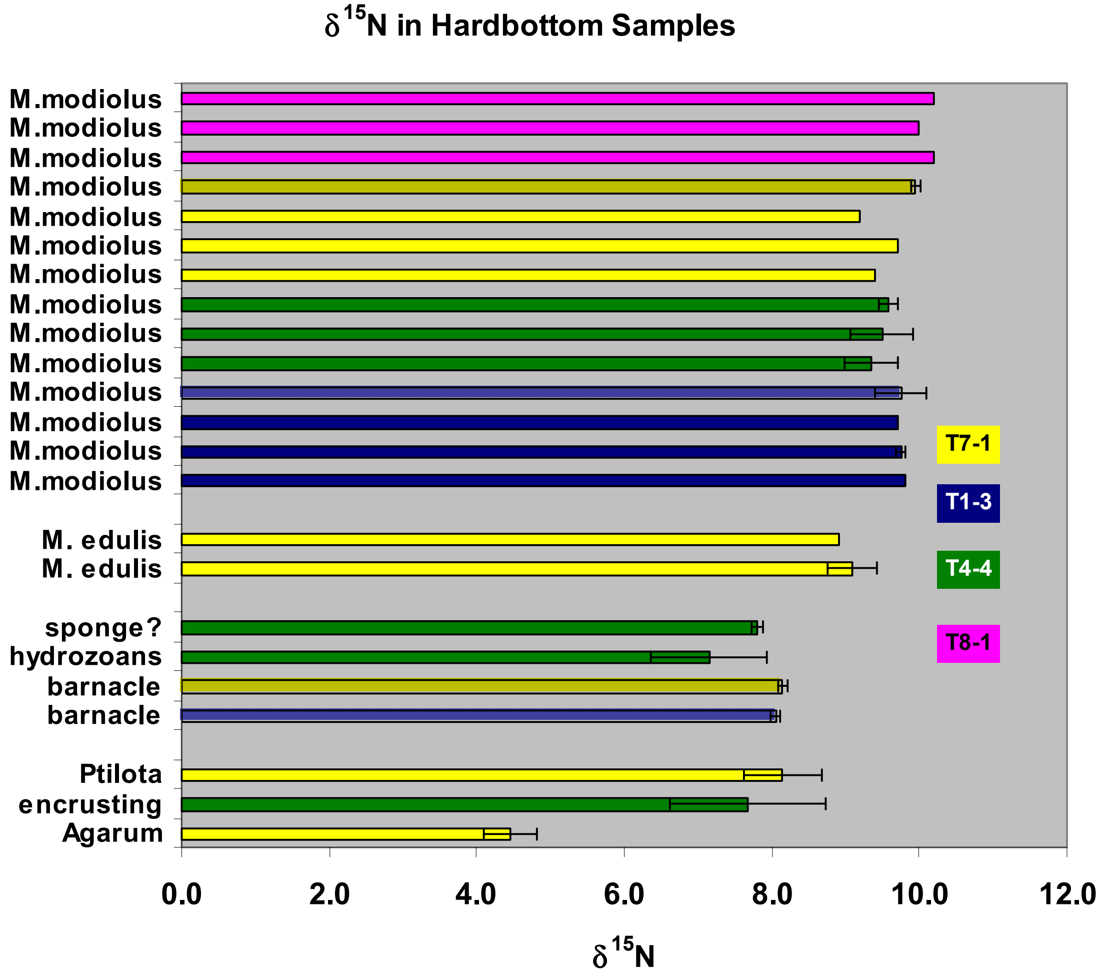
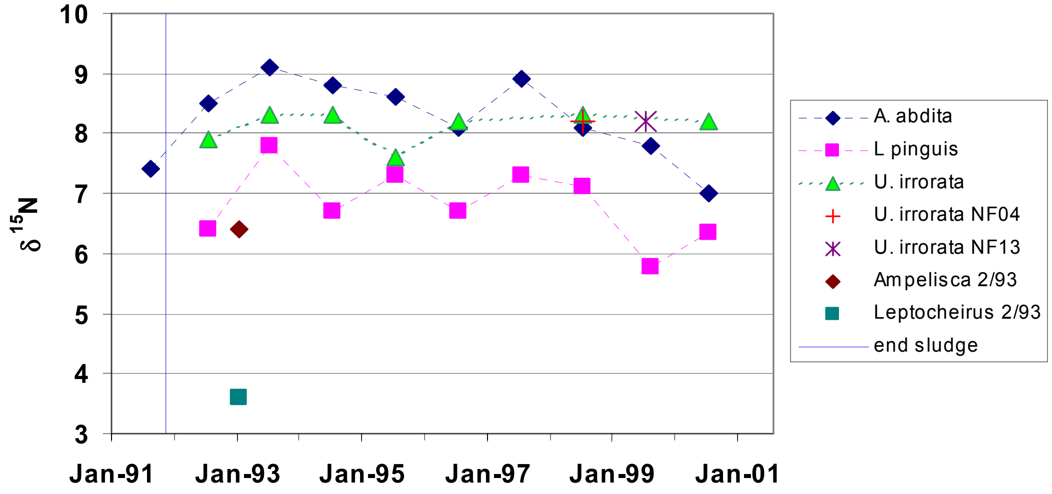
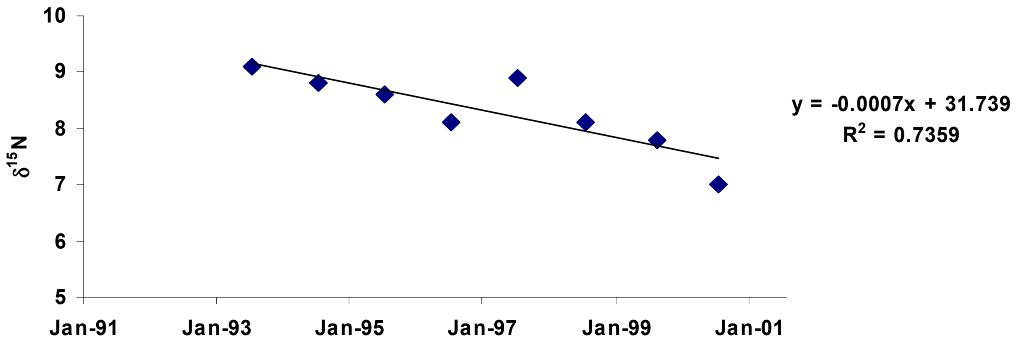


Fig. 7. δ^{15}N in samples from four hardbottom stations: T7-1(yellow bars), T1-3 (blue bars), T4-4 (green bars), and T8-1 (pink bars). Stippling within bars of a given color designates a *M. modiolus* individual and its associated barnacle.

a. δ^{15}N in amphipods at Station T3



b. T3 *A. abdita* 1993-2000



c. T3 *L. pinguis* 1993-2000

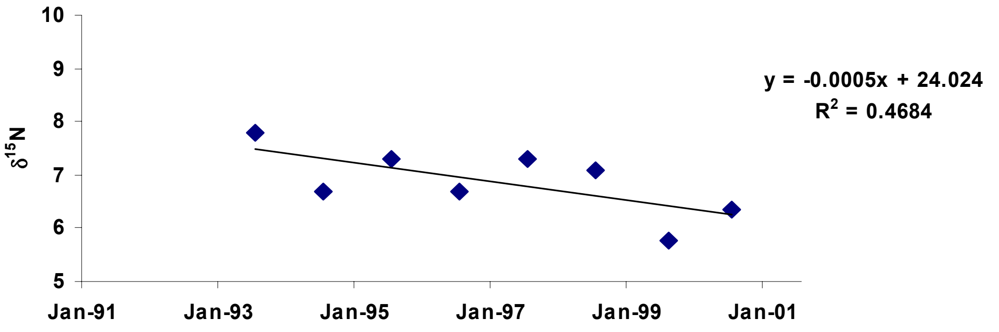
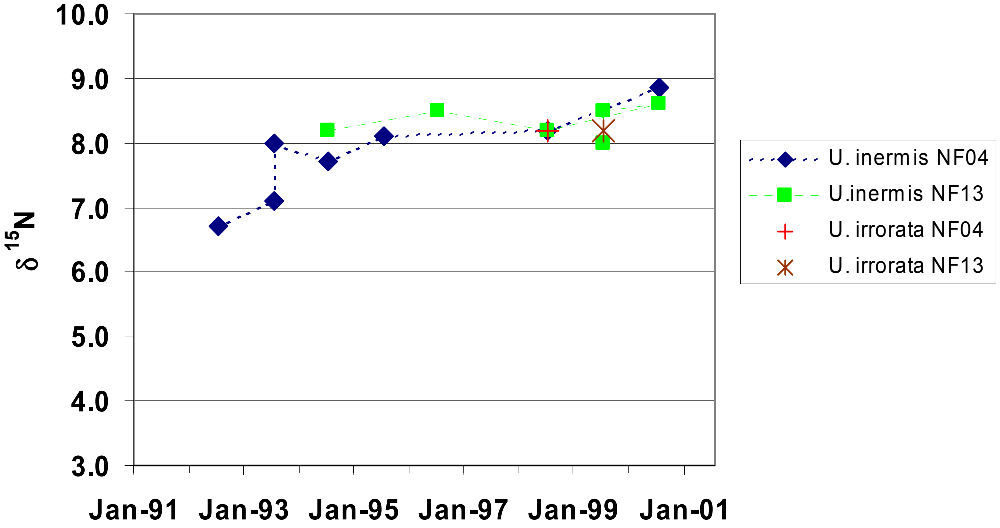


Fig. 8. a.) δ^{15}N in August samples of *A. abdita*, *L. pinguis*, and *U. irrorata* at Station T3 from 1992-2000, with results from February 1993 as well as for nearfield *U. irrorata* for comparison. b.) regression of δ^{15}N with time from1993 – 2000 for *A. abdita*; $P<0.01$ c.) regression of δ^{15}N with time from1993 – 2000 for *L. pinguis*; $P=0.06$. δ^{15}N in *U. irrorata* did not show a trend with time.

a. δ^{15}N in Amphipods at NF04 and NF13



b. NF04 *U. inermis*, 1992-2000

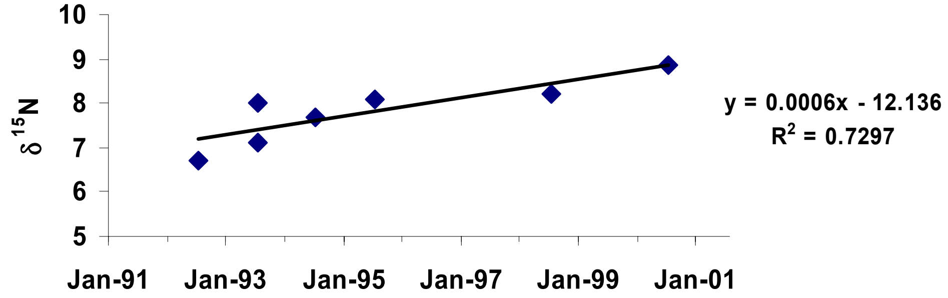


Fig. 9. a.) δ^{15}N in August samples of *U. inermis* and *U. irrorata* at nearfield stations NF04 and NF13 from 1992-2000; b.) regression of δ^{15}N with time for *U. inermis* at station NF04; *P*=0.01. δ^{15}N in *U. inermis* from NF13 did not show a trend with time.

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