

The effects of infaunal biodiversity on biogeochemistry of coastal marine sediments

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Abstract

We tested the role of benthic infaunal functional diversity in regulating the biogeochemistry of nearshore sediments using laboratory microcosms. Single and multispecies assemblages of deposit-feeding polychaetes (*Clymenella torquata*, *Spio setosa*, and *Leitoscoloplos fragilis*) were used, and fluxes of oxygen and phosphate, as well as profiles of oxygen and pH in the sediment pore water, were measured. Significant differences in flux rates were found among treatments that were unrelated to polychaete abundance or biomass alone. Multispecies assemblages had lower flux rates of both oxygen and phosphate than rates calculated from the single-species treatments. Depth-integrated oxygen and pH profiles also showed significant differences between multispecies assemblages and selected single-species treatments. These differences were most likely from species-specific feeding and burrowing behavior and species-related interactions. Coefficients of variance for both oxygen and pH were highest for microcosms with no polychaetes and lowest for the assemblages, indicating a dampening effect of multispecies assemblages on pore-water heterogeneity. When oxygen flux data was incorporated into a model of oxygen dynamics in Long Island Sound, results indicated that shifts in the benthic community composition could change sediment oxygen consumption rates sufficiently to disrupt the balance between the physical supply of oxygenated water and biological oxygen demand. The results of this study confirm the importance of benthic functional biodiversity to nearshore sedimentary processes and suggest that losses of functional diversity can have significant effects on ecosystem function.

Over the past several decades, there has been increasing interest in the health of the global environment, with specific emphasis on the importance of elemental cycling, climate change, and biodiversity loss. Worldwide species extinction rates are comparable to those of the end of the Mesozoic era and, as such, significant efforts are underway to understand how anthropogenic influences on biodiversity might alter or impair functions or services of the earth's ecosystems.

Early studies of biodiversity effects on ecosystem function focused primarily on terrestrial plant communities and revealed significant effects on physical, biogeochemical, and ecological processes. For example, Naeem et al. (1994) confirmed that diversity loss affected community respiration,

short-term organic matter decomposition, soil nutrient and water retention, primary productivity, and vegetation structure. Tilman et al. (1997), working in experimental grassland plots, demonstrated that both species and functional diversity affected productivity, nitrogen dynamics, and light penetration. Niche differentiation and the coexistence of many species having differential effects (covariance effects) on each other are often cited as the mechanisms causing differences in productivity, but some disagreement remains as to the exact mechanisms (*see* Wardle 1999; Kinzing et al. 2001 for recent reviews of empirical and theoretical work).

Studies on aquatic biodiversity have frequently examined correlations of environmental factors with species distributions, but little is known about specific relationships between biodiversity and ecosystem function. This is undoubtedly in part because of the difficulties of manipulating species and measuring ecosystem function in a dynamic, fluid environment. However, some studies suggest a causal role of diversity as moderator of ecosystem-level processes. For example, Cardinale et al. (2002) found interspecific facilitation as a mechanism by which multispecies assemblages of suspension feeders in streams can attain more food resources than single-species assemblages. Similar studies are emerging in coastal marine systems. Duarte (2000) illustrated that diversity of seagrass communities played an important role in maintaining stability and linkages to other trophic levels. Duffy et al. (2001) found grazer species composition had significant effects on seagrass growth, although species rich-

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ness did not have an effect. In the rocky intertidal, Paine (2002) found that increased consumer diversity altered rates of macroalgal production by reducing the dominance of competing producers. Studies of subtidal fouling communities showed that species richness influenced recruitment dynamics of these systems (Stachowicz et al. 2002). In spite of these examples, there is sparse experimental evidence linking diversity and function in other marine habitats, such as mangroves (Field et al. 1998), coral reefs (Ogden et al. 1994), or the open ocean (Legendre and Rivkin 2002). Similarly, linkages between microbial biodiversity and ecosystem function are underexplored (Finlay et al. 1997).

Marine sedimentary habitats provide an excellent testing ground for examining relationships between functional diversity and ecosystem function because of the extensive literature on geochemical and ecological processes and the significant effect of infauna on organic matter diagenesis and sediment seawater exchange. These effects are complex because benthic infauna have highly variable effects on sediment biogeochemistry (Marinelli 1994; Marinelli et al. 2002), in part because of the variability in activity rates and community complexity and in part because of the sedimentological environment (Huetzel et al. 1996). The existing experimental evidence for marine sedimentary environments generally supports the tenet that functional diversity influences ecosystem processes. For example, Chua and Brinkhurst (1973) found a reduction in respiration rate of oligochaetes kept in multispecies cultures rather than separately, and Weinberg and Whitlatch (1983) found increased growth of small suspension-feeding bivalves when kept in close proximity to a head-down deposit-feeding polychaete. Andersen and Kristensen (1988) examined effects of macrofauna on community respiration in comparing different taxa: an amphipod, a polychaete, and a hydrobid snail species alone and in combination in light and dark systems. Their results indicated differential effects of different taxa on sediment properties and processes. The assemblage greatly enhanced dissolved inorganic nitrogen flux and stimulated a flux of oxygen into sediments in light treatments, whereas monospecific and control treatments fluxed oxygen out of the sediment. More recently, Biles et al. (2003) found that infaunal diversity increased sediment ammonium flux out of the sediment under flow conditions, although when no flow was present, no diversity effect was found. Emmerson et al. (2001) also examined diversity effects on sediment ammonium release but found no predictable pattern in their extensive experiments. These studies affirm the suggestions of Levin et al. (2001) and Smith et al. (2000) that our understanding of benthic biodiversity effects on ecosystem processes is highly speculative and requires rigorous experimental evaluation.

In this study, we examined the effects of benthic functional diversity on sediment seawater fluxes and pore-water constituents that are important participants in elemental cycling in sedimentary systems. The questions we address are: (1) Do benthic flux rates in assemblages of three different species of deposit feeders reflect simple additive effects of each species in monocultures? (2) Are pore-water variables such as pH and oxygen different between single- and multispecies communities? (3) Do increases in functional rich-

ness have any effect on the spatial variability of pore-water constituents?

Materials and methods

Microcosms—Laboratory microcosms (~10-cm-diameter \times 30-cm-long pipes capped on the bottom) were used to examine the effect of single-species versus multispecies assemblages on sedimentary geochemical processes. The experiment consisted of five treatments, each with five replicates: a control with no macrofauna added, three single-species treatments, and a treatment containing all three species (*Clymenella torquata*, *Spio setosa*, and *Leitoscoloplos fragilis*). The top cover of each microcosm contained ports and a stirring mechanism that allowed sampling and replacement of overlying water while preventing vertical stratification when flux measurements were taken. Lids were affixed to the microcosms only when flux measurements were being made, all other times the microcosms sat in a flow-through seawater bath.

Sediment—Sediment collected from a subtidal (~4 m depth) area at the mouth of the Poquonock River, adjacent to Long Island Sound, southeastern Connecticut, was frozen for 48 h to kill the zoobenthos while retaining a significant proportion of the microbial community. A previous study at the field site (1994) found a sediment composition of 53% median sand (250–500 μm), 5% silt-clay (<63 μm), and 0.87% organic carbon (ash-free dry weight; Whitlatch unpubl. data). After thawing, the sediment was sieved through a 300- μm mesh screen, homogenized, and added to the microcosms to achieve a sediment depth of ~20 cm. The microcosms were placed in a filtered (10 μm) flow-through seawater bath and kept in the dark at 20°C. The salinity was relatively constant over the entire experiment at 32 ± 1 . The microcosms were allowed to equilibrate for 2 weeks before macrofaunal organisms were added. After organisms were added to the microcosms, small circulating pumps and air stones were added to the water bath to keep the water fully oxygenated.

Organisms—Three species of polychaete worms (*L. fragilis*, *C. torquata*, *S. setosa*) were selected on the basis of their cosmopolitan distributions, frequent co-occurrence, and functional differences. *L. fragilis* (hereafter referred to as *Leitoscoloplos*) is a motile, subsurface, conveyor belt deposit feeder (Fauchald and Jumars 1979) that feeds near the oxic–anoxic boundary (1–10 cm depth depending on the sediment properties) and moves sediment from depth to the surface (Bianchi 1988). *C. torquata* (hereafter referred to as *Clymenella*) is a tube-dwelling conveyor belt species that feeds head-down at depths up to 20 cm below the sediment–water interface. However, Dobbs and Whitlatch (1982) noted that *Clymenella* can feed on surface sediments, and others have shown that some species of malanids will move freshly deposited material to depth (Levin et al. 1997). *S. setosa* (hereafter referred to as *Spio*) is also a tube dweller that feeds at the sediment–water interface but can switch between surface deposit feeding and suspension feeding depending on the availability of suspended particles (Fauchald and Jumars

Table 1. The mean wet weights (\pm SD), number of individuals of each species added to each microcosm, and field densities of each species scaled to the surface area of the microcosm ($7.85 \times 10^{-3} \text{ m}^2$). Unpublished data (Whitlatch) was obtained from cores collected in 1994. Although *Spio* was not found in that analysis, individuals were found in abundance at the same collection site in 2001, and other spionid species were present in 1994. NA, not applicable.

Treatment	Mean wet wt (g)	Added per core (n)	Published abundances (n)	Unpublished 1994 data (n)
<i>Leitoscoloplos</i>	0.2270(\pm 0.0212)	7	3–21*	0–32
<i>Clymenella</i>	0.8321(\pm 0.1035)	5	2–5†	12–38
<i>Spio</i>	0.2571(\pm 0.0539)	6	2–8‡	0
Assemblage	0.8278(\pm 0.1175)	13 (5,3,5)	NA	NA

* Bianchi 1988.

† Dobbs and Whitlatch 1982.

‡ Dauer et al. 1981.

1979; Dauer et al. 1981). Tubes of *Spio* are generally <10 cm deep. After ingestion of food at the surface, they defecate within the tube and move their fecal material to the surface (Dauer et al. 1981).

All organisms were collected from tidal flats in the Poquonock River directly adjacent to, or in very close proximity to, Long Island Sound; maintained for 16 days in separate containers of treated sediment; then added to the microcosms. Individuals were selected randomly then grouped and weighed collectively prior to addition. The numbers added to each treatment mimicked natural densities of the species found in nearshore communities (Table 1). Abundances of organisms at the collection site were not measured at the time of collection, although previous work in the area (18 June 1994, Whitlatch unpubl. data) suggests that microcosm densities were similar to naturally occurring field densities (Table 1). Most of the organisms burrowed immediately into the sediment, and all had burrowed within several hours of addition to the microcosms.

The experiment was maintained over a 4-month period,

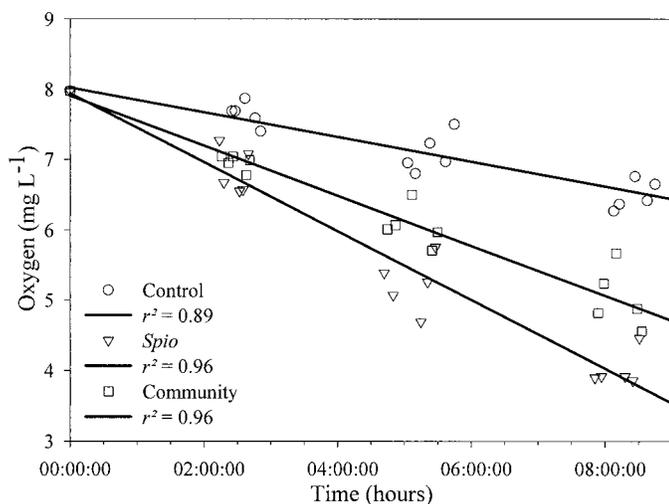


Fig. 1. Representative oxygen flux measurements for selected treatments on Day 54 showing fit of the flux data to three representative treatments.

starting with organism addition (Day 1). Organisms were fed an aquaculture food (Omega Booster, Aquafauna BioMarine) once during the experiment. An amount equivalent to a 3-week pulse of carbon to the benthos of Long Island Sound (0.75 g of food; Dam Guerrero 1989) was added to each microcosm on Day 57. The addition of a labile food source could change the relationships of macro- and microfauna to each other (Tenore et al. 1982), as well as macrofaunal foraging behavior (Taghon et al. 1980), and therefore could affect pathways and rates of remineralization. Furthermore, Aller (1994) discussed the potential for microbial priming, or elevated rates of remineralization of otherwise refractory material (beyond background levels) when labile carbon is deposited on the sediment surface; hence, a shift in metabolic processes could be explained by the input.

Flux measurement—Flux measurements of oxygen and phosphate were conducted on four dates (Days 48, 54, 70, 95) over the 4-month experiment. Fluxes were determined by fitting the microcosm lids with stirrers and allowing constituents to accumulate or deplete in the water column over 3–8 h, depending on oxygen demand. Oxygen concentrations were monitored with a microelectrode (Diamond General microelectrode and Unisense PA2000 picoammeter) every 1–2 h during flux measurements, and water samples from all microcosms were taken sequentially at times corresponding to roughly 100%, 75%, and 50% of O_2 saturation of the microcosms with the lowest oxygen levels. Approximately 20-ml water samples were taken, frozen (-20°C) for preservation, and subsequently thawed when analyzed. The microelectrodes were calibrated with the use of a two-point calibration curve, with the upper concentration consisting of ambient seawater (20°C , salinity = 31) at 100% saturation and the lower concentration consisting of seawater purged with N_2 gas. The calibration equation and flux estimates were determined by regression (see Fig. 1 for sample flux data).

Phosphate was measured on thawed samples by a modification of the U.S. Environmental Protection Agency method 365.2 for low-range determination of orthophosphate on a SmartChem autoanalyzer (Westco Scientific). An eight-point calibration curve was generated in duplicate for each

run (typically 60–129 samples) of the analyzer. All reagents and standards were prepared in acid-washed glassware, and standards were prepared in a nutrient-free artificial seawater matrix.

Sediment pore-water measurements—Oxygen concentration and pH of the sediment pore water were determined on Day 113 with the use of a Diamond General combined pH/oxygen microelectrode with a Unisense picoammeter (for oxygen) and a Microscale Measurements high-impedance ($>10^{15}$ ohm) millivolt meter (for pH). For the oxygen microelectrode calibration, a two-point calibration curve was used as described above. The pH electrode calibration was conducted by generating a three-point calibration curve using standard reference solutions (pH 4, 7, and 10; Fisher Scientific).

A digital servo-driven micromanipulator (National Aperature) permitted submillimeter-scale resolution in the vertical direction. Two pH/oxygen combination electrodes were mounted roughly 2 cm apart on the manipulator motor. Three of the five replicate microcosms of each treatment group were randomly selected for microelectrode measurements. Within each microcosm, three profiles of the double-mounted electrode were taken, resulting in six replicate profiles for each microcosm. For each profile, the double-mounted probes were positioned at least a centimeter from the edge of the microcosm and as far as possible from the previous profiles in order to obtain the most representative conditions. All three replicate profiles within a microcosm were run in ~30 min.

Data analysis—Oxygen and phosphate fluxes were calculated by regression of concentration versus time for each experimental replicate and each measurement time. Mixed-model analyses of variance were conducted on the calculated fluxes to determine whether fluxes were significantly different among the treatments through time. As such, the main effects were time, treatment, and the time \times treatment interaction. If no significant interaction was found, the analysis was rerun without the interaction term. Differences among treatments were analyzed by least squares means ($\alpha = 0.05$). All statistical analyses were performed with the SAS statistical software package.

Per-individual organism flux rates were calculated by subtracting the average control flux value from each average treatment value and then dividing by the number of organisms in each treatment (see Eq. 3). This “attributable” flux rate is a measure of the enhancement (through bioturbation activities, respiration, etc.) each individual of a given species has on the exchange of water and solutes across the sediment–water interface. The per-individual calculation might be more appropriate than a biomass calculation because sedimentary alterations are driven more by spatial considerations associated with the extent of nonlocal processes (e.g., depth of burrows, length scales associated with feeding) and less with the mass of individuals per se. A proportional deviation equation was applied to the flux data (as in Emmerston et al. 2001) to examine possible species richness effects on fluxes. This equation permits comparisons between an actual community flux and a theoretical community flux (of

the same species composition) calculated from the single-species measurements, referred to herein as yielding. Yielding was calculated as

$$D_T = \frac{O_T - E_T}{E_T} \quad (1)$$

where O_T is the observed flux in the mixed culture, or the three-species treatment, and E_T is the estimated community flux rate based on single-species treatments (Eq. 2).

$$E_T = A + \sum_i (E_i p_i) \quad (2)$$

A is the measured flux of the control, E_i (Eq. 3) is the attributable organism flux rate of species i , and p is the number of individuals of species i in the assemblage.

$$E_i = (M_i - A)/p_i \quad (3)$$

M_i is the measured value for a single-species treatment, and p_i is the number of organisms in a given treatment.

Results are easily evaluated; deviations from zero imply that species interactions affect the system in some way to either inhibit or accelerate a process—in this case, degradation of organic matter and associated solute fluxes across the sediment–water interface. If values of D_T are positive, the three-species assemblage is exceeding the expected flux; this is called overyielding. If D_T is negative, the assemblage flux is less than that expected from the single-species measurements, called underyielding.

To determine the effects of single- versus multispecies treatments on pore-water characteristics, a nested analysis of variance was conducted for depth-integrated pore-water pH and oxygen profiles. Differences among treatments were analyzed by least squares means ($\alpha = 0.05$). Replicate data from each of the two probes within one profile run of the dual-mounted microelectrodes were averaged and used as a single estimate for each profile. Replicate profiles served to estimate experimental error, and the core within-treatment effect was treated as one level of nesting. To further evaluate the role of functional diversity on sedimentary properties, the coefficient of variance was calculated for the oxygen and pH profile data at each depth for each treatment. These results were used to assess the degree of spatial heterogeneity of the biogeochemical properties of the sediment pore water. Once a coefficient of variance profile for both oxygen and pH was calculated for each treatment, it was then integrated as above. No statistical analyses were performed on these estimates; rather, coefficients of variance were used for qualitative comparisons only.

As a post hoc analysis, a Pearson correlation matrix was constructed with SAS in order to examine other possible links between infaunal biomass, abundance, and flux rates. For this calculation, the flux data were averaged across all dates.

Results

Oxygen flux—The average oxygen fluxes ($\text{mmol m}^{-2} \text{d}^{-1}$) for each treatment over the four dates were: Control, -29.76 ; *Leitoscoloplos*, -31.95 ; *Clymenella*, -34.04 ; *Spio*, -42.36 ; and three-species assemblage, -36.64 (Fig. 2).

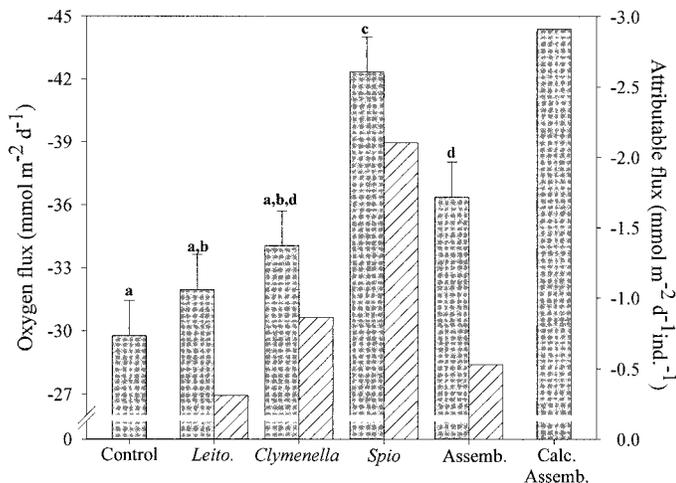


Fig. 2. Oxygen flux for each treatment averaged across all dates. Solid bars are measured fluxes for each treatment (left axis). The hatched bars represent the attributable flux rates or per-individual flux rates (right axis). The expected community flux is calculated as described in methods. Error bars are standard error, and common letters represent no significant difference. The measured assemblage flux is substantially lower than the expected assemblage flux, representing an underyielding of 17%.

Analysis of variance indicated a significant treatment ($F_{4,92} = 8.82$, $P < 0.0001$) and date ($F_{4,92} = 250.58$, $P < 0.0001$) effect on oxygen flux. The interaction term was not significant ($F_{12,80} = 0.66$, $P = 0.7835$) and was subsequently dropped from the model. Pairwise multiple comparison analyses showed that *Spio* ($t_{92} = 5.44$, $P < 0.0001$) and the three-species assemblage ($t_{92} = 2.97$, $P = 0.0038$) were significantly greater than the Control. The *Spio* treatment was also significantly greater than the *Leitoscoloplos* ($t_{92} = 4.50$, $P < 0.0001$), *Clymenella* ($t_{92} = 3.59$, $P = 0.0053$), and three-species assemblage ($t_{92} = 2.47$, $P = 0.0153$) treatments. Oxygen flux in the three-species assemblage was marginally different from the *Leitoscoloplos* treatment ($t_{92} = 2.03$, $P = 0.0456$; Fig. 2). Differences in treatment flux rates cannot be accounted for by the range of individual abundance or biomass in this experiment (Figs. 3, 4), suggesting that functionality or species identity might be the most important regulator of oxygen flux. We determined Pearson correlation coefficients, post hoc, to verify these findings and found no significant correlations between biomass and oxygen flux ($r = -0.249$, $P = 0.686$) or abundance and oxygen flux ($r = -0.463$, $P = 0.432$). Interestingly, the theoretical oxygen flux for the three-species treatment, on the basis of attributable rates for individuals, was $-44.39 \text{ mmol m}^{-2} \text{ d}^{-1}$ ($n = 5$ for *Leitoscoloplos*, $n = 3$ for *Clymenella*, and $n = 5$ for *Spio*). Comparison with the measured value of $-36.64 \text{ mmol m}^{-2} \text{ d}^{-1}$ illustrates that the coexistence of these species lowers total sedimentary oxygen flux in a way that was not predictable from the effects each species had on oxygen flux while living in a monoculture.

The application of the yielding equations to the oxygen flux data show that, on average (i.e., across all dates), the three-species assemblage underyielded 17% with respect to oxygen flux. This trend held on all dates except Day 70,

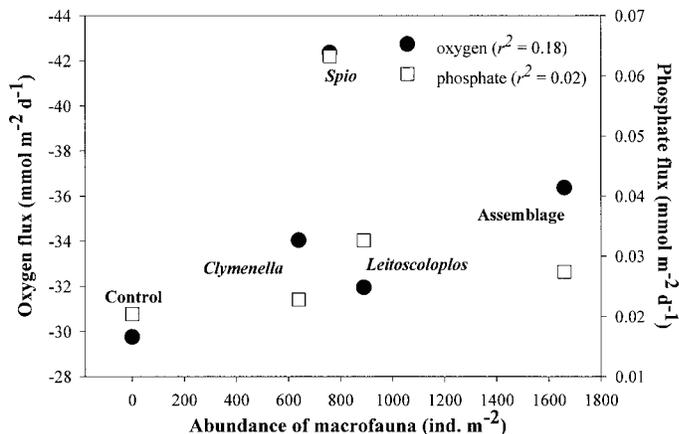


Fig. 3. Abundance of macrofauna plotted against oxygen and phosphate flux. No detectable effect was found, as noted by the r^2 values.

when the measured community and calculated fluxes were almost identical. This date was 13 d after the food addition, which might have stimulated enhanced oxygen utilization in the three-species treatment relative to the single-species treatments, perhaps because of an overwhelming effect of fresh labile material on the activities of micro- and macroorganisms. On all other sampling dates, the three-species assemblage underyielded from 15% to 43%. This result suggests that when these three species are functioning as part of an assemblage, interactions occur that influence rates of sediment oxygen uptake relative to single-species groups.

Phosphate flux—Time averaged phosphate fluxes ($\text{mmol m}^{-2} \text{ d}^{-1}$) for each treatment were: Control, 0.020; *Leitoscoloplos*, 0.033; *Clymenella*, 0.023; *Spio*, 0.063; and three-species assemblage, 0.027 (Fig. 5). Analysis of variance indicated significant treatment ($F_{4,59} = 4.70$, $P = 0.0023$), date ($F_{3,59} = 6.03$, $P = 0.0042$), and treatment \times date interaction ($F_{8,59} = 2.87$, $P = 0.0091$) effects on phosphate flux. Pairwise multiple comparison analyses showed the *Spio* treatment was significantly greater than the Control ($t_{59} = 3.78$,

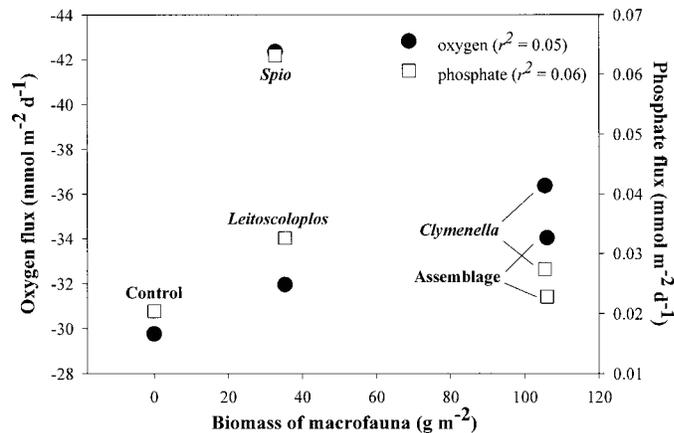


Fig. 4. Biomass of macrofauna versus oxygen and phosphate flux rates. No detectable effect of biomass on flux rates was found, as noted by the r^2 values.

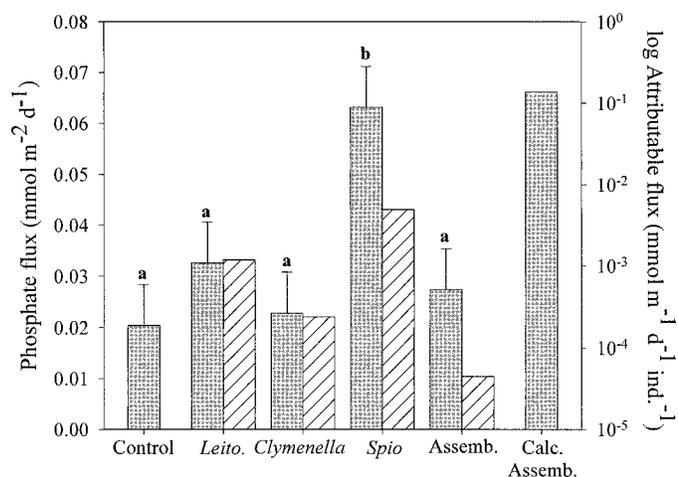


Fig. 5. Phosphate flux for all treatments averaged across date. Solid bars are measured fluxes (left axis), attributable fluxes are cross-hatched (right axis); the expected community is calculated as in methods. The attributable fluxes are graphed on a log scale. Error bars are standard error, and common letters indicate no significant difference. The difference between the measured and calculated assemblage represents an underyielding of 59%.

$P = 0.0004$), *Leitoscoloplos* ($t_{59} = 2.70$, $P = 0.0089$), *Clymenella* ($t_{59} = 3.57$, $P = 0.0007$), and three-species treatments ($t_{59} = 3.10$, $P = 0.0030$; Fig. 5). The significant interaction term suggests the treatment differences varied with date and therefore the pairwise comparisons should be viewed cautiously. Fluxes in the *Spio* treatment, however, were a factor of three larger than the other treatments. As such, the treatment \times time interaction might have been driven by changes in *Spio* treatments alone across time.

As with oxygen, phosphate fluxes do not appear to be driven strictly by organism abundance or biomass. Pearson correlation coefficients for the relationship between biomass/phosphate flux ($r = -0.234$, $P = 0.705$) and abundance/phosphate flux ($r = -0.142$, $P = 0.819$) indicate the lack of association between traditional measures of organism abundance and phosphate flux (see also Figs. 3, 4). The measured three-species treatment phosphate flux ($0.027 \text{ mmol m}^{-2} \text{ d}^{-1}$) was greatly depressed compared with the calculated value ($0.066 \text{ mmol m}^{-2} \text{ d}^{-1}$). When applied to the yielding calculations, this discrepancy resulted in a 59% decrease (range 41–75%) in phosphate flux in the three-species treatment. This again illustrates that the coexistence of these three species lowers sedimentary phosphate flux in a way not predictable from measurements made of each species living in monoculture.

pH profiles—In all treatments containing infauna, pH was higher at depth than in the Control microcosms (Fig. 6). The *Spio* treatment had the lowest pH in the upper 10 mm. Statistical analysis of the integrated profiles indicated a significant effect of treatment ($F_{4,10} = 5.99$, $P = 0.0100$). Pairwise comparisons by least square means showed that pH in the *Clymenella* and assemblage treatments were not different from one another but were significantly greater than the *Lei-*

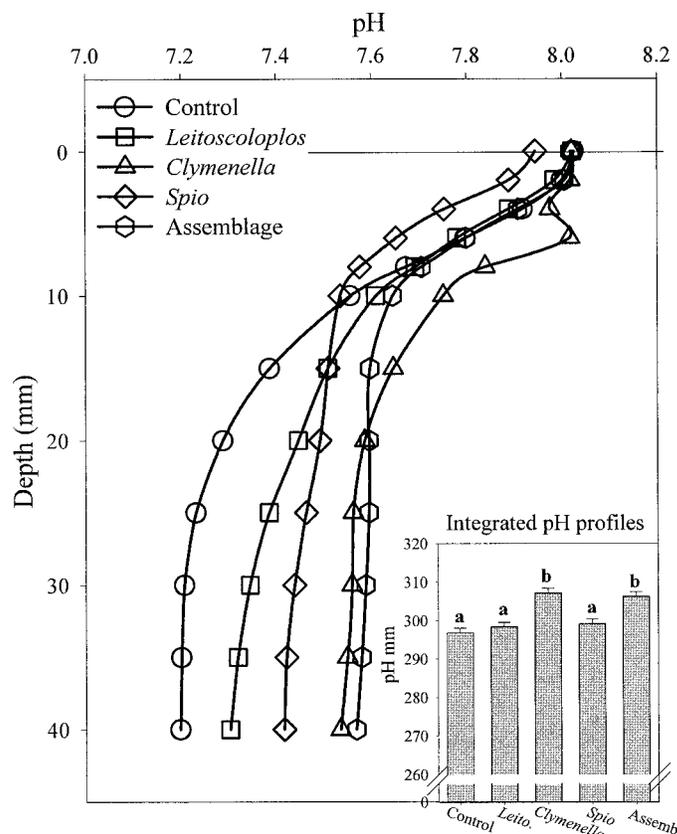


Fig. 6. Average pH profile with depth for each treatment taken on Day 113. The insert in the lower right shows the depth-integrated values for pH (note units are in pH mm), with standard error represented by the bars and common letters indicating no significant difference.

toscoloplos, *Spio*, and Control treatments (Fig. 6). Specifically, *Clymenella* was significantly greater than the Control ($t_{10} = 3.73$, $P = 0.0039$), *Leitoscoloplos* ($t_{10} = 3.17$, $P = 0.0100$), and *Spio* ($t_{10} = 2.90$, $P = 0.0159$) treatments. Similarly, the three-species assemblage was significantly greater than the Control ($t_{10} = 3.41$, $P = 0.0067$), *Leitoscoloplos* ($t_{10} = 2.85$, $P = 0.0173$), and *Spio* ($t_{10} = 2.58$, $P = 0.0275$) treatments. Because all these differences were between the three-species assemblage or *Clymenella* and the other treatments, the presence of *Clymenella* as an individual or as part of the assemblage appears to have a disproportional effect on pH.

Oxygen profiles—The oxygen profiles include the upper 15 mm of the sediment column only (Fig. 7). Below this depth, oxygen concentrations were zero for all microcosms measured. Analysis of variance found a significant effect of treatment on depth-integrated oxygen profiles ($F_{4,10} = 4.63$, $P = 0.0225$). Pairwise comparisons showed that the integrated oxygen concentration for the *Spio* treatment was significantly lower than the Control ($t_{10} = 3.10$, $P = 0.0112$), three-species assemblage ($t_{10} = 2.92$, $P = 0.0153$), and *Clymenella* ($t_{10} = 3.47$, $P = 0.0126$) treatments. *Leitoscoloplos* was also significantly lower than *Clymenella* ($t_{10} = 2.48$, P

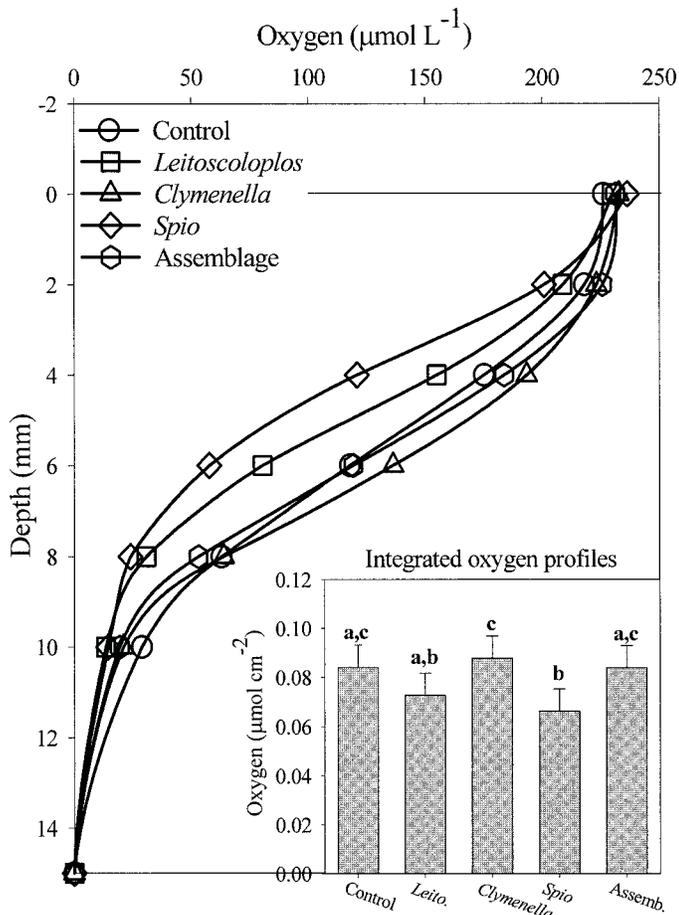


Fig. 7. Average oxygen profile with depth for the upper 15 mm of the sediment taken on Day 113. Insert shows the integrated values for the upper 15 mm, with error bars as standard error and common letters indicating no significant difference.

= 0.0323). Consistent with the high measured fluxes, the oxygen gradient in the *Spio* treatment is particularly sharp (Fig. 7).

Oxygen versus phosphate—Phosphate is several orders of magnitude more adsorptive in oxic conditions compared with anoxic conditions (Krom and Berner 1980); hence, a negative relationship between phosphate flux and measures of sediment pore-water oxygen concentration can exist. Regression analysis was conducted to assess the dependence of phosphate flux on both oxygen fluxes and depth-integrated oxygen concentration (determined from profiles). No significant relationship between oxygen flux and phosphate flux was detected. However, a negative relationship was found between phosphate flux and the depth-integrated oxygen concentration (Fig. 8), with roughly 88% of the variance in phosphate flux explained by the variance of depth-integrated oxygen within the sediments. The lack of relationship between oxygen flux and phosphate flux is not surprising. Although both are affected predominantly by organic matter decomposition, oxygen flux is influenced by oxidation of reduced species (generally significant in nearshore sediments), whereas phosphate is affected by adsorption and

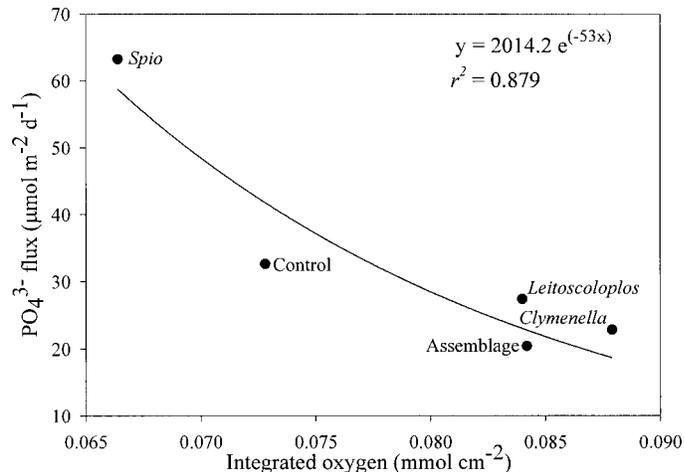


Fig. 8. Phosphate flux versus integrated oxygen profiles for each treatment. Note the total oxygen in sediment pore water can account for almost 88% of the variance in phosphate flux. This is likely because of the adsorption dynamics of phosphate in oxic and anoxic sediments, with greater adsorption occurring in oxic sediments compared with anoxic sediments (Krom and Berner 1980).

mineral formation. Depth-integrated oxygen concentration, calculated from profiles, might be the better predictor of the significance of phosphate adsorption because the profiles characterize the available oxic regions where adsorption can occur.

Variability in pore water—Spatial variability of the integrated pore-water profiles across depth, as indicated by coefficients of variance, revealed a striking pattern (Fig. 9). In general, Control microcosms had the highest coefficients of variance, whereas single-species treatments had intermediate values, and the three-species treatment had the lowest values. This pattern suggests that diversity decreases the amount of spatial heterogeneity in the pore-water constituents.

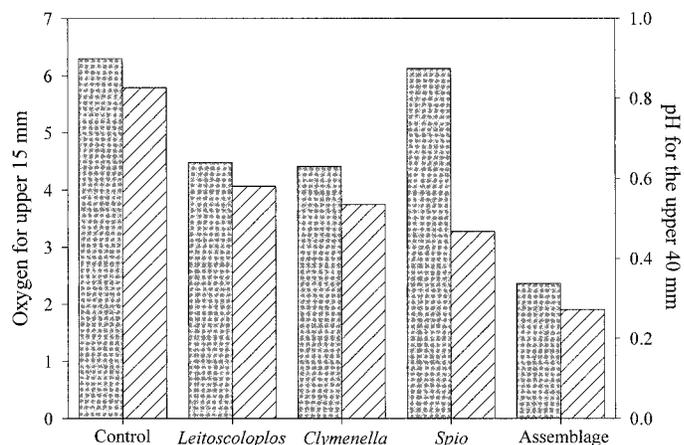


Fig. 9. The depth-integrated coefficient of variance for oxygen and pH. Solid bars represent oxygen and hatched bars pH. Note that in both cases, variance is highest in the Control, intermediate in the single-species treatments, and lowest in the community.

Discussion

Results from this microcosm study indicate that species diversity, functional diversity, or both affect the functioning of marine sediments. The yielding equation indicates a 17% and 59% reduction in oxygen and phosphate flux rates (Figs. 2, 5), respectively, in the three-species assemblage relative to the single-species treatments. In addition, the coefficient of variation for both oxygen and pH profiles was lower in the three-species assemblage relative to the single-species treatments and Control. Both of these findings show that more functionally complex assemblages operate differently from depauperate ones and that functional diversity effects are not simply the summation of individual species effects. These findings are consistent with terrestrial studies examining biodiversity and ecosystem function in plant communities (e.g., Tilman et al. 1997) and aquatic studies examining biodiversity and nutrient transport (Cardinale et al. 2002; Biles et al. 2003). The mechanisms are not clearly understood, although recent work has begun to address these processes (Cardinale et al. 2002). Thus, although many sedimentary functions are enhanced or regulated by individual infauna, diversity and community configuration are additional controlling influences.

The effects of the species assemblage on fluxes were independent of infauna abundance or biomass (Figs. 3, 4). This is consistent with the results of Marinelli and Williams (2003), who found nonlinear relationships between density of individuals and ammonium fluxes, as well as silicate: ammonium flux ratios; the results of Marinelli (1994), who found nonlinear relationships between organism density and irrigational flux; and the laboratory simulations of Aller and Aller (1998), who found enhanced remineralization processes when diffusion distances, a proxy for density, were decreased. These results suggest that spatial configuration and infaunal activity are important attributes affecting transport and reaction rates in sediments and further suggest that simple models of bioturbation must be expanded to account for such nonlinearities. Possibly in diverse communities, different ecological strategies allow more organisms to coexist (facilitation) in closer proximity to one another relative to a monospecific community. Such an alteration in spatial configuration affects diffusion characteristics and other important organism behaviors, which in turn regulate transport reaction processes and ultimately sediment-seawater fluxes.

A general decrease in pore-water heterogeneity of oxygen and pH was found with increased species richness (Fig. 9). This result is contradictory to studies that show increased variance with greater infaunal abundance, community complexity, or both (e.g., Aller et al. 1997). Clearly, some configurations serve to reduce heterogeneity and rates of flux. Gilbert et al. (2003) found that intermediate diffusion scales between oxic and anoxic sediments corresponding to intermediate abundances of infauna increased nitrification-denitrification rates relative to low or high abundances. Likewise, Marinelli and Williams (2003) found the strongest effect on ammonia fluxes at an intermediate infaunal density. Although previous studies show that abundance is an important "optimizing" parameter (sensu Gilbert et al. 2003), this

study underscores that diversity (i.e., infaunal activities that occur in a multispecies context) can also be an important additional moderating or optimizing parameter.

Clear and observable differences in fluxes and profiles were found between the three single-species treatments, representing two different "functional groups," with functionality (here) defined primarily by feeding behavior. For example, *Spio* is a surface deposit feeder that mixes the shallow depths of sediment and likely interacts more closely with the aerobic microbial community. Accordingly, *Spio* was associated with significantly higher fluxes of oxygen and phosphate across the sediment-water interface (Figs. 2, 5), significantly lower oxygen penetration within the sediment (Fig. 7), lower pH in the upper sediment column, and higher pH in the lower sediment column (Fig. 6). Conversely, *Clymenella* is a deep-dwelling, head-down deposit feeder affecting a deeper portion of the sediment column. *Clymenella* significantly increased pH throughout its feeding depth range (Fig. 6), maintained an intermediate oxygen flux relative to the other two species (Fig. 2), had slightly higher oxygen in the pore water (Fig. 7), and had almost no apparent effect on phosphate flux (Fig. 5) relative to the Control. Similar to *Clymenella*, *Leitoscoloplos* is a head-down feeder, although it is motile and resides in intermediate to upper layers of the sediment column. It had relatively low oxygen flux (Fig. 2), elevated phosphate flux (Fig. 5), pore-water oxygen levels intermediate to the other two single-species treatments (Fig. 7), and little effect on pH of pore water. Although *Clymenella* and *Leitoscoloplos* might be feeding and moving particles in the same general direction, their differences (also compared with *Spio*) in depth of bioirrigation and tube building appear to have measurable effects on the resulting geochemical properties of the sediment column. Irrigation rates, general behavior, and actual depth of burrows were not directly measured in this experiment. However, observations suggest that differences in irrigation behavior exist. On completion of the experiment, consolidated, well-formed burrow structures were found in treatments containing *Spio* and *Clymenella*, with *Clymenella* tubes having a larger area of lighter sediment around burrows (indicative of oxygen penetration) compared with the *Spio* tubes. Conversely, *Leitoscoloplos* treatments were characterized by poorly formed, friable tubes, rather than a well-formed burrow, that had little to no light grey (oxic halo) around the tubes, indicative of less active ventilation and motility. This evidence leads us to speculate that *Clymenella* had the greatest irrigation effect on transport relative to reaction, followed by *Spio* then *Leitoscoloplos*, with respect to oxygen. This analysis suggests that traditional functional groupings of infauna based on feeding groups might have limited usefulness for prediction of faunal effects on geochemical processes. Functional groupings for geochemical processes based on the intensity of nonlocal exchange processes, as well as feeding rates and pathways of particle motion, might more profitably reflect functional diversity effects.

Identification of geochemical keystone species or species that have a significant effect as physical ecosystem engineers (sensu Jones et al. 1997) could provide valuable understanding of the relationship between biodiversity and ecosystem

function in sediments. For example, the deep burrowing behavior of *Clymenella* and reflected changes in pore-water pH and oxygen (Figs. 6, 7), compared with the other species in the current study, could lend it to be classified as a geochemical keystone species in microcosms. The increased pH at depth is notable because of the important role pH plays in many diagenetic reactions in nearshore systems (Marinelli and Boudreau 1996). The increased oxygen at depth not only affects diagenetic reactions, microbial distributions, and colonization patterns of meiobenthos (Meyers et al. 1987) but also has an important link to phosphate dynamics. Phosphate adsorption is enhanced in the presence of oxygen (Krom and Berner 1980), and changes in adsorption will affect flux rates. Indeed, the data show that for *Clymenella*, oxygen fluxes are marginally higher than the Control (Fig. 2) and oxygen penetration into sediments comparatively deep (Fig. 7) relative to other microcosms. However, phosphate fluxes (Fig. 5) in the *Clymenella* treatments are low and comparable to the Controls. In this case, the low phosphate flux might not be a result of lack of activity (as could be the case in the Control), but of the depth enhancement of oxygen penetration by *Clymenella* and subsequent lowering of phosphate in the pore water because of adsorption (Fig. 8)—hence, the underyielding. *Praxilella*, another maldivid polychaete, has been targeted as a keystone species by Levin et al. (1997) for its significant effects on organic matter movement and burial in continental shelf sediments. One could argue that *Spio* could also be classified as a keystone species because of its large flux rates measured in monoculture, but these effects are not as readily manifested and observed in the assemblage as the effects of *Clymenella* are. Therefore, *Spio* is less noteworthy as a candidate to be a geochemical keystone species. It should also be noted that our classification of *Clymenella* as a geochemical keystone species is dependent on the scale of observation and the particular function being observed. These studies underscore the need to examine species and community composition more closely as facilitators of geochemical processes, and to consider a different suite of characteristics for categorizing bioturbational effects on sediment geochemistry.

The designation of *Clymenella* as a geochemical keystone species invokes the need to consider the problem of sampling effects (also known as selection effects) and the extent to which disproportional species effects are actually biodiversity effects or simply an artifact of experimental design (Wardle 1999). Wardle (1999) states that in order to examine the possibility of sampling effects, it is important that each species is evaluated in monoculture and that the species used are in fact found to co-occur. Although we were unable to mimic all elements of complexity in natural benthic communities, we were able to assess monoculture and mixed assemblages that were consistent with naturally occurring densities. Although the deep irrigating effects of *Clymenella* seem to be disproportionately higher than the other species and were in part responsible for changes in phosphate flux rates, they cannot account for all the underyielding between expected and measured assemblage fluxes. Furthermore, recent works seem to indicate that sampling effects are in fact true biodiversity effects, but further discussion here is be-

yond the scope of the current treatment (see Kinzing et al. 2001).

We have shown in this study that changes in the species composition, as well as changes in functional diversity within experimental microcosms, will significantly alter measurable ecosystem parameters; however, it remains to be demonstrated whether the magnitude of these changes are enough to effect ecosystemwide changes. To illustrate potential effects, we apply the results from the current study to an estuary scale model to represent how basinwide decreases in functional diversity caused by eutrophication (Smith et al. 2000) or fishing practices (Thrush et al. 2001) might affect entire ecosystems and provide feedback effects. We do recognize the pitfalls in extrapolating results from this limited laboratory study to the field. Given the close proximity of the sample site at the mouth of the Poquonock River to Long Island Sound, the low flow nature of this river, and the representative nature of the species evaluated here with regard to Long Island Sound benthos, we feel that the application of the current data to the model is informative and illustrative of important, negative effects. Torgersen et al. (1997) used ^{222}Rn as a tracer of water circulation in Long Island Sound and found that oxygen dynamics within the Sound were in a delicate balance between physical (supply) and biogeochemical (demand) processes. In other words, the supply of oxygen-rich waters because of tidal mixing with the Atlantic Ocean was very closely balanced with the oxygen demand (biological and chemical) of the water column and sediments. Sediment oxygen uptake rate constants used by Torgersen et al. (1997) were $0.018\text{--}0.007\text{ d}^{-1}$ for spring and summer, respectively. The net summer rate constant (sum of all rate constants) for their study area of Long Island Sound was 0.006 d^{-1} . Sediment oxygen consumption rate constants calculated from this study ranged from 0.019 d^{-1} (for *Spio*) to 0.014 d^{-1} (for *Leitoscoloplos*) and 0.016 d^{-1} for the three-species assemblage. More important, the range of oxygen uptake rates calculated from this study ($0.019 - 0.014 = 0.005\text{ d}^{-1}$) compared with the net summer rate constant (0.006 d^{-1}) in Torgersen et al. (1997) illustrates that changes in benthic species composition could have a significant effect on basinwide effects on oxygen dynamics in Long Island Sound. Although application of the results obtained from laboratory microcosms to field studies should be made with caution, these results do indicate the balance of biological and physical controls on hypoxia in Long Island Sound might be susceptible to functional changes of benthic macrofauna.

Coastal benthic habitats fill a variety of ecosystem functions, and the anthropogenic effects of pollution, hypoxia, and exotic species introduction on species and functional diversity will undoubtedly shift the composition of resident benthic communities. This study illustrates that properties of benthic sediment habitats resulting from the interactions of functionally different organisms are emergent and not easily predicted from knowledge of each given species. Results add to the growing body of literature supporting the role biodiversity and functional diversity has in regulating ecosystem function. Many challenges exist in unraveling the complexity that lies at the intersection of ecology and geochemistry in marine sediments, and more work is needed to understand

the potential consequences the loss of biodiversity could have in coastal marine ecosystems (Smith et al. 2000; Levin et al. 2001).

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