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Benthos Investigations: Sediment Boxes or Natural Bottom?

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The dispersal of pollutants into aquatic ecosystems has had far-reaching effects and continues to present potential risks to the environment. Aquatic hazard evaluation studies have ranged from expensive multidisciplinary research programs, to single species monitoring, and laboratory-controlled bioassays. In most cases these kinds of studies have fallen short in respect to understanding the more subtle fates and effects of various pollutants as well as in providing decision-makers with appropriate information to aid in their management of aquatic ecosystems.

The benthic habitat of aquatic environments has been the focus of many of these pollution assessment studies because of the sedentary nature of many fauna living there, which provides a good barometer of pollution events. Functional reasons for benthos emphasis include its role in nutrient cycling and secondary production within aquatic ecosystems. Activities of benthic fauna (e.g. bioturbation) regulate such processes as nutrient regeneration, linking the benthos to ecosystem functioning via benthic-pelagic coupling, which often controls overall production of these ecosystems (ROWE 1978). In addition, biomass produced by benthic fauna serves as an important food source to many aquatic living resources. By simply assessing acute toxicity of pollutants, many of the most often used methodologies of pollution monitoring miss these functional roles of ecosystem components such as the benthos.

Many monitoring studies in the past seven years have attempted to more precisely document pollutant effects to the benthos, while also maintaining more control over study design by employing the use of sediment boxes (usually azoic) and the establishment of experimental communities through colonization. These studies have included sediment boxes colonized in the field (e.g. BRUNSWIG et al. 1976, ANDERSON et al. 1978, BOESCH 1979) as well as sediment boxes colonized via flow-through water systems in the laboratory (e.g. HANSEN 1974, SCHOOR & NEWMAN 1976, TAGATZ & TOBIA 1978).

These approaches have proven valuable, 1) because of the increased ease of sampling a difficult and patchy environment, and 2) because one can more accurately measure such factors as community structure, bioturbation, sediment metabolism, and benthic nutrient regeneration. All of these factors, although more subtle than those evaluated by acute toxicity tests, are potentially important in

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respect to detecting integrated impacts and risks associated with the dispersal of marine pollutants in an aquatic habitat. Because sediment boxes are often employed in basic and applied aquatic research to extrapolate results obtained under controlled conditions to the natural environment, we examined the differences in benthic community structure between the natural marine bottom and sediment boxes colonized with fauna under both field and laboratory conditions. Our objective was to gain some insight into problems that may arise in drawing conclusions from an experimental methodology intended to mimic the natural environment.

METHODS

Two sets of experimental sediment boxes, each containing 10 replicate compartments, as described by HANSEN (1974), were filled to depths of 7 cm with azoic sediments that were allowed to dry for 35 days with periodic washings of freshwater. These sediments were of a similar grain size to the bottom sediments of the estuary from which unfiltered water was pumped from 4 m above the bottom to supply continuous flow to our laboratory facilities. One of the two sediment boxes was placed in the laboratory and received this unfiltered continuous water flow at a rate of 200 ml/min. The other box was placed on the estuarine bottom in a water depth of approximately 3 m. Both the laboratory and field systems were allowed to colonize through the settlement of larvae and/or migration of adults for 8 weeks, from 5 December 1979 to 4 February 1980. At the end of this period the field box was covered, retrieved by SCUBA, and placed in the laboratory with the other system on the continuous flow water supply at the same flow rate. Ambient water temperatures and salinities were maintained in the laboratory at all times and a light/dark cycle simulating natural conditions was adhered to.

The two sediment boxes were maintained on the unfiltered seawater system in the laboratory for an additional four weeks. At the end of this period the sediment contents of all experimental compartments were siphoned and washed through a 0.5 mm mesh screen, excluding end compartments to eliminate any potential end-of-row effects.

The natural estuarine bottom was sampled at the same approximate time that the sediment boxes were sampled. Triplicate samples using a Petersen grab were obtained from a bottom habitat in 2-3 m of water that contained sediments similar in grain size to those used in the sediment boxes. This site, although approximately 5 km from the field sediment box placement, was chosen for comparison because of its long history of benthic sampling (7 yr). Community structure of the benthos at this site was shown to be similar to many other physically similar estuarine sites in the area, including the one used for the field sediment box placement. The grab sampler penetrated the bottom to an average depth similar to the depth of the sediment boxes. The same procedures for washing of these samples were followed as described above. All sample statistics were extrapolated to a common surface area (0.18 m^2) for comparative purposes.

Fauna were identified and counted according to methods of FLINT & HOLLAND (1980) and wet weight biomass was measured in each sample. Species lists of each replicate compartment for the laboratory and field-colonized sediment boxes, and the natural sediment samples were compared using cluster analysis, employing the Canberra-Metric similarity index with a flexible sorting strategy. Species diversity of each sample was calculated by the Shannon-Wiener index (PIELOU 1966) using $\log_2$ and equitability (LLOYD & GHELARDI 1964).

RESULTS

Cluster analysis of the species lists for the laboratory-colonized, field-colonized, and the natural estuarine bottom samples fused the three kinds of benthos samples into distinct groupings (Fig. 1). The laboratory-colonized samples were most different from the other groups at a 60% dissimilarity level. The field-colonized cluster and natural sediment bottom cluster differed from one another at 40% dissimilarity. Within each of the cluster groupings (Fig. 1), the highest dissimilarity between replicate samples, 23%, was observed for the laboratory-colonized samples, while the maximum dissimilarity level for the field-colonized replicates was 17% and for the natural bottom samples was 16%. These differences suggested that laboratory-colonized sediments exhibited a patchier distribution of species than either the field-colonized sediments or the natural estuarine bottom sediments.

An inverse cluster analysis using the same similarity measure and sorting strategy, was performed to determine which species groups best characterized the clusters identified in Figure 1. For a species to be included in this analysis it had to represent 1% of the total density for at least one of the comparisons (natural, field, or laboratory). According to the inverse cluster analysis, the dominant fauna observed in the field and laboratory boxes also occurred in high densities in the natural bottom, with the exception of the amphipod *Corophium acherusicum* (Fig. 2). Except for these dominant species, however, the laboratory-colonized replicates exhibited little similarity in species assemblages to the others. In contrast, the field-colonized sediments appeared to attract species that naturally occurred in the estuary relative to the samples from the natural bottom.

Figure 2 further illustrates that in ranking dominant fauna for the natural bottom and field-colonized sediments, there were some similarities and some differences between these two methods for estimating benthic community structure. The most obviuos difference, however, and the one that caused the major dissimilarity between the field-colonized and natural bottom species comparisons in Figure 1, was the overwhelming dominance of the field-colonized sediments by *Balanoglossus* sp. and *Abra aequalis* populations. These differences are further illustrated in Table 1. Not only was the field-colonized total density much higher but total community biomass was also significantly different ($P < 0.05$). Much of this biomass difference was due solely to the biomass of *Abra aequalis* (Table 1).

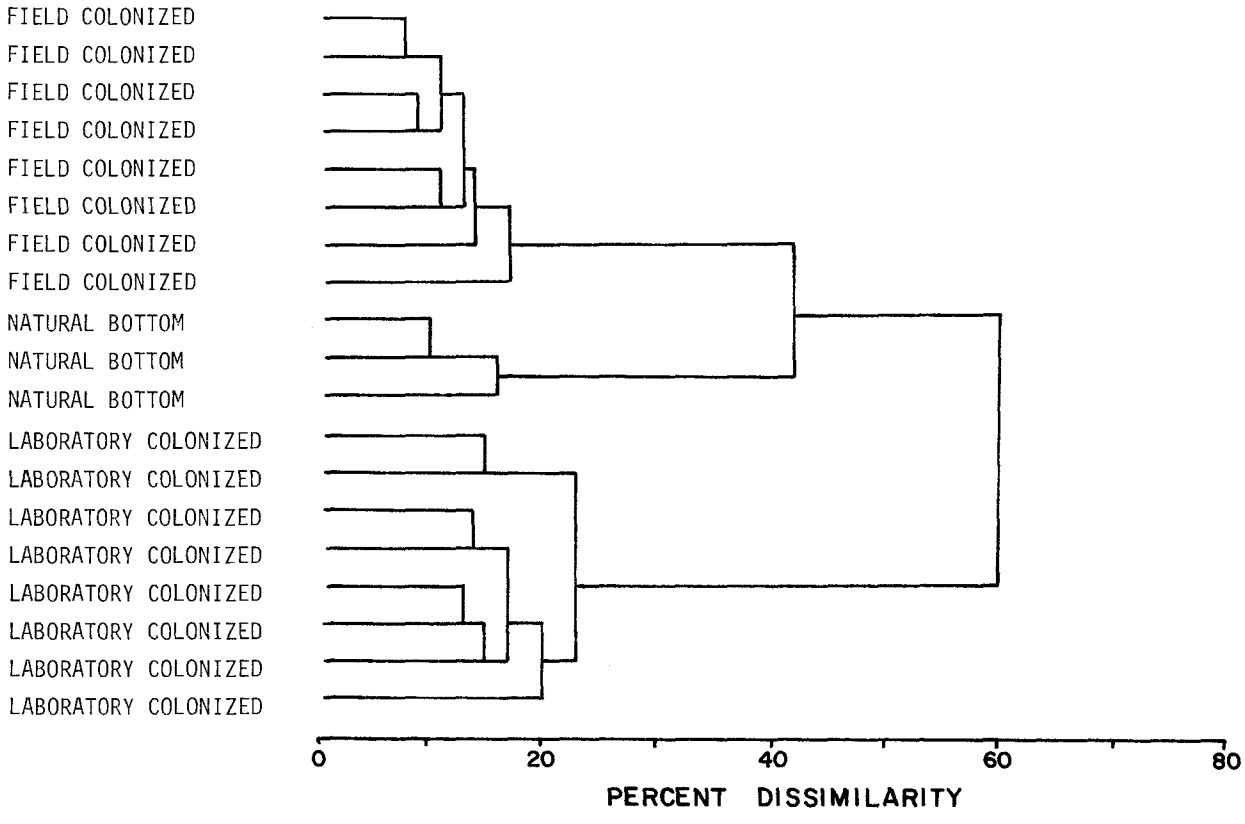

Figure 1: Cluster analysis dendrogram comparing benthic species lists for field-colonized and laboratory-colonized sediment boxes with representative samples from the natural esturarine bottom.

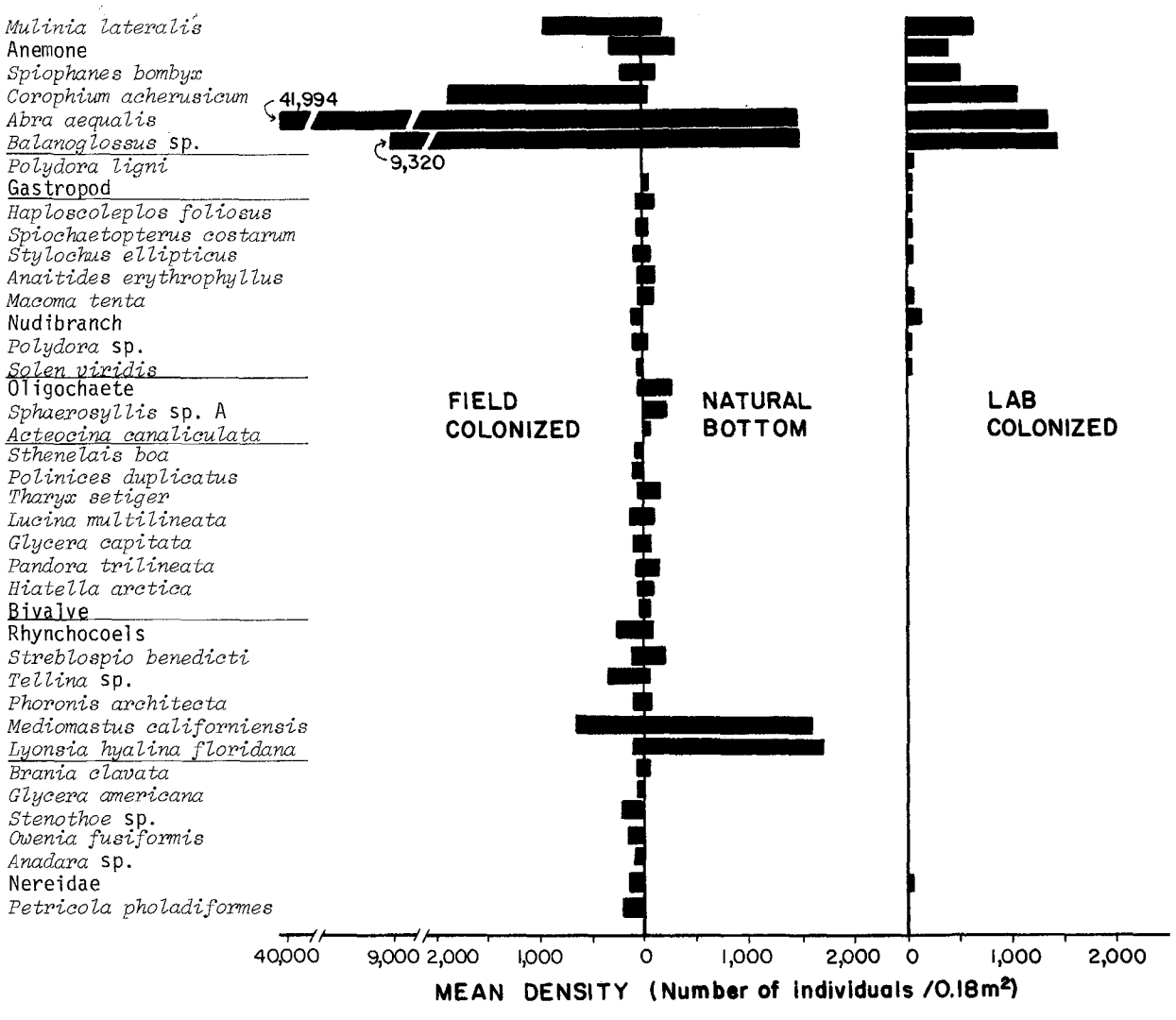

Figure 2: Mean density of dominant benthic infaunal species found in field-colonized sediment

Table 1. Comparison of benthic community variables and individual species biomasses between natural bottom samples, field-colonized sediment boxes, and laboratory-colonized sediment boxes. The numbers in parentheses represent % total abundance in each method for each species.

	Field-Colonized Sediment Boxes	Natural Bottom Sediments	Laboratory Sediment Boxes
Total Density (animals/0.18 m^2)	59,131.5 ± 15,277.9	9,251.3 ± 539.7	5,103.0 ± 1,528.5
Total Biomass (g/0.18 m^2)	39.1 ± 5.6	19.6 ± 6.8	6.1 ± 2.3
Species Number	47.2 ± 4.4	86.3 ± 9.1	19.2 ± 5.6
Species Diversity	1.5 ± 0.3	3.9 ± 0.2	2.6 ± 0.2
Equitability	0.04 ± 0.02	0.16 ± 0.08	0.25 ± 0.09
Abra aequalis biomass (g/0.18 m^2)	25.4 ± 5.5 (72)	2.3 ± 1.1 (16)	3.5 ± 3.1 (24)
Balanoglossus biomass (g/0.18 m^2)	2.9 ± 1.8 (18)	4.4 ± 2.1 (16)	0.3 ± 0.2 (30)
Mulinia biomass (g/0.18 m^2)	2.6 ± 1.3 (2)	1.8 ± 0.9 (2)	0.3 ± 0.2 (13)
Lyonsia hyalina	(1)	(19)	(1)
Mediomastus	(1)	(18)	-
Corophium	(3)	(1)	(17)

A final difference between the estuarine benthos in the three environments comes from examining the size of individual population members of the dominant fauna. For example, the total biomass of *Abra aequalis* in the field-colonized sediments was 25.4 g/0.18 m^2 (Table 1) while the total density for this population was approximately 41,900 individuals/0.18 m^2 (Fig. 2). These data represented an average weight of 0.6 mg/individual for the field-colonized *Abra aequalis* population. In contrast, the natural bottom *Abra aequalis* population comprised 2.3 g/0.18 m^2 total biomass with a total density of 1,461 individuals/0.18 m^2 for an average weight of 1.6 mg/individual. Likewise, *Balanoglossus* sp. showed an average population member weight of 2.9 mg/individual for the natural bottom and 0.3 mg/individual for the field-colonized sediments. *Mulinia lateralis* exhibited a mean of 11.9 mg/individual for the natural bottom and 2.9 mg/individual for the field-colonized sediments. Thus, for those dominant fauna examined, the natural bottom samples exhibited larger-sized populations (biomass per individual) than the field-colonized sediments for the period of year sampled.

DISCUSSION

Investigations of the subtidal marine benthos are complicated by hydrographic variability, substrate patchiness, and capacity to return to the same sampling site. The exposure of sterile sediments and ultimate colonization of benthic fauna is one way of possibly eliminating many of these problems. This method can also potentially contribute to the collection of more precise data on benthic production, turnover ratios, and processes regulated by the infauna, such as bioturbation and nutrient regeneration; all of which can be potentially impacted by pollution disturbances to the seafloor.

According to results presented here, of the two alternatives to investigate benthic communities on the natural seafloor, field-colonized sediment boxes and laboratory-colonized sediment boxes, the field-colonized alternative appears to better mimic the natural species assemblages. This is not to imply that laboratory experiments can not serve a valuable "bioassay" purpose, but if the object is to produce experimental communities more similar to natural communities, the field-colonization experiments are preferred.

Almost all of the numerically important members of the natural community were established in the experimental field boxes (Fig. 2). A few of the species occurring in the boxes, however, exhibited densities several times greater than the natural sediments. After examining the biomass of individuals in these sediment box populations it was obvious that only juveniles of *Abra aequalis*, *Balanoglossus* sp. and *Mulinia lateralis* occurred in contrast to the larger sized population members (adults) of the natural community. Consequently, these high densities in the experimental box were probably an artifact of the phase of colonization and reproductive cycle of the populations that occurred at the time of the experiment. Colonization of these dominant fauna into the field box most likely occurred through reproduction and larval settlement rather than

through adult migration. Since the high densities of a few populations were the major factors responsible for differentiating the field sediment boxes from the natural community in the cluster analysis, we would expect an even better rating of similarity with an increased period of colonization, since the higher experimental box densities were represented by juvenile populations whose community had not yet reached the stability of the natural bottom community.

Based upon the comparison of our results with other investigations, it would appear that field-colonized boxes better mimicked the natural communities than has so far been reported. After a period of 28 months of exposure and faunal colonization BRUNSWIG et al. (1976) found that originally animal-free substrates had not resulted in a species assemblage at all similar to the natural sediments surrounding their experimental boxes. These investigators also noted much higher population biomasses occurring outside the boxes in contrast to our results.

The use of azoic sediment boxes on the Atlantic continental shelf with both 10 and 43 week exposure periods (BOESCH 1979) also exhibited results in colonization different than reported here. The highest similarity observed between the natural seafloor and any experimental boxes was with the 43 week exposure periods. This similarity level was approximately 42% which equalled a dissimilarity level of 58%, in comparison to our 40% dissimilarity level (Fig. 1).

In our studies, 42% of the taxa observed in field and laboratory sediment boxes were common and 80% of the dominant fauna in the natural sediments were observed in the field boxes. In comparison to our success of mimicking the natural environment, TAGATZ et al. (1981) recorded 30% of the total number of taxa as being common between field and laboratory boxes in one experiment and TAGATZ & IVEY (1981) observed 19% of the total species as being common between field and laboratory boxes in another experiment.

In terms of investigating disturbance ecology of the benthic habitat, we feel that, based upon the above experimental results, the use of sterile substrates exposed on the seafloor is worthy of consideration. The use of this methodology, however, must take into account the many natural biological variables of importance in structuring benthic communities such as the size of the species pool, periods of peak reproduction, modes of reproduction of dominants (larval type), and adult mobility. If these factors are considered and an appropriate amount of time employed for maximum colonization to occur, this methodology can lend itself nicely to the study of marine benthos. For example, developing associations of fauna can be studied from the very beginning through increasing stages of complexity and processes of the seafloor that are related to benthic faunal activity, such as nutrient regeneration, bioturbation, or sediment metabolism, can more easily be measured and interpreted in respect to the actual sediment inhabitants. From our results, mimicry of the natural bottom appears possible with the use of sediment boxes

and the most important consideration appears to be a meshing of the period allowed for colonization with the specific objectives of the research that employs this method for studying the natural benthos.

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