SUCCESSIONAL MECHANISM VARIES ALONG A GRADIENT IN HYDROTHERMAL FLUID FLUX AT DEEP-SEA VENTS

Lauren S. Mullineaux,[1] Charles H. Peterson,[2] Fiorenza Micheli,[3] and Susan W. Mills[1]

[1]*Biology Department, Woods Hole Oceanographic Institution, Woods Hole, Massachusetts 02543 USA*
[2]*University of North Carolina at Chapel Hill, Institute of Marine Sciences, Morehead City, North Carolina 28557 USA*
[3]*Hopkins Marine Station, Stanford University, Pacific Grove, California 93950 USA*

Abstract. Invertebrate communities inhabiting deep-sea hydrothermal vents undergo substantial succession on time scales of months. Manipulative field experiments assessed the relative roles of environmental state and biotic interactions in determining temporal succession along a spatial gradient in vent fluid flux at three vent sites near 9°50′ N on the East Pacific Rise (2500 m water depth). Species colonization patterns on cubic basalt blocks (10 cm on a side) deployed by the submersible Alvin revealed both positive (facilitation) and negative (inhibition) biological interactions, in the context of established succession theory. Over a series of four cruises from 1994 to 1998, blocks were exposed to colonists for consecutive and continuous intervals in short-term (5 + 8 = 13 mo) and longer-term (8 + 29 = 37 mo) experiments. Colonists grouped into a mobile functional group were less abundant in the continuous interval (13 mo) than in the synchronous pooled-consecutive intervals (5 + 8 mo) of the short-term experiment, indicating that early colonists inhibited subsequent recruitment. Colonists grouped into a sessile functional group exhibited the opposite pattern, indicating facilitation. Similar trends, though not statistically significant, were observed in the longer-term experiment. The character of species interactions varied along a gradient in hydrothermal fluid flux (and inferred productivity), with inhibitory interactions more prominent in zones with high temperatures, productivity, and faunal densities, and facilitative interactions appearing where temperatures, productivity, and densities were low. Analyses of primary succession on introduced basalt blocks suggest that biological interactions during early vent community development strongly modify initial patterns of settlement, even in the absence of sustained temporal change in the vent fluid flux.

Key words: deep sea; facilitation; hydrothermal vent; inhibition; Lepetodrilus; *limpet;* Ophryotrocha; *polychaete; productivity gradient; succession; tubeworm; vestimentiferan.*

INTRODUCTION

The pathways of ecological succession, defined as a directional change in community structure following a perturbation that creates new or modifies existing habitat, may be fundamental characteristics of most animal, plant, and microbial communities. Uncovering the mechanisms driving these temporal sequences of species replacements and community composition shifts is essential for a full understanding of the structure, diversity, and dynamics of biotic systems. Although a few frequently cited early studies suggested that succession in some systems might progress unidirectionally toward an inevitable and fixed climax (e.g., Clements 1916; reviewed in Odum 1969), most subsequent empirical and theoretical investigations indicate that the trajectories vary and can be difficult to predict. For instance, during primary succession in benthic marine systems, very different outcomes can occur, depending on the identity of initial colonists (Sutherland and Karlson 1977), the relative importance of positive and neg-

ative interactions among species (Sousa 1979*a, b,* Lubchenco 1983), and the role of indirect effects of consumers (Hixon and Brostoff 1996). Further complications occur when the abiotic environment varies unpredictably, or during secondary succession after a disturbance that has not completely eradicated the existing community (i.e., leaving a biological legacy that influences the course of species reestablishment, as occurred at Mt. St. Helens after the 1980 eruption; Franklin and MacMahon 2000).

Succession is particularly apparent and important in systems where episodic disturbances induce large changes in availability of primary substratum or food. Such dramatic disturbances occur at deep-sea hydrothermal vents, where catastrophic events such as volcanic eruptions and tectonic events (quakes) create or destroy entire habitats. On fast-spreading mid-ocean ridges, catastrophic perturbations occur on time scales of years to decades (MacDonald et al. 1980, Haymon et al. 1993), making disturbance and primary succession dominant characteristics of the ecosystem. The physical and chemical nature of the habitat continues to be dynamic after eruptions, with rerouting of hydrothermal fluid sources due to tectonic shifts or geo-

chemical clogging, changes in chemical composition of fluids (von Damm 1995, Butterfield et al. 1997) and rock falls or basaltic pillow collapses (Haymon et al. 1993). These physical perturbations modify the habitat, and the chemical variations (i.e., changes in hydrothermal fluid flows that support the chemoautotrophic-based food web) alter supplies of the primary nutritional source. The responses of colonizing species at deep-sea vents to these perturbations, either directly through physiological tolerances and nutritional requirements, or indirectly through biological interactions, are largely unknown.

The mechanisms of succession in the vent environment may parallel those in forests, streams, or shallow marine environments, or they may differ greatly due to the unique combination of habitat transience and patchiness, species endemism, and steep gradients in temperature, toxic chemicals, and chemoautotrophic production. Given the insights that successional studies have provided into the ecology of intertidal (Dayton 1971, Sousa 1979*a, b*), subtidal (Witman 1987), and deep-sea (Grassle and Morse-Porteous 1987) communities, understanding successional change in hydrothermal vent communities is necessary to explain their spatial and temporal patterns and may simultaneously provide broader insights into the general nature of ecological succession.

Previous studies of invertebrate colonization at hydrothermal vents have documented a distinct and consistent successional sequence over the lifespan of vents on the East Pacific Rise (Fustec et al. 1987, Hessler et al. 1988). The initial visibly dominant sessile metazoan in sites with moderate-temperature fluids (i.e., <30°C anomaly; not high-temperature black smokers) appears to be the small vestimentiferan tubeworm *Tevnia jerichonana* (Shank et al. 1998). This species then is replaced by the larger tubeworm *Riftia pachyptila*, frequently over a period of less than one year. Later in the sequence, the mussel *Bathymodiolus thermophilus* colonizes and may, in some cases, displace the vestimentiferans (Hessler et al. 1988). Time-series observations suggest that this sequence can correspond to a change in the temperature and chemistry of vent fluids (Shank et al. 1998), although Hessler et al. (1988) and Mullineaux et al. (2000) document species succession under conditions of constant venting and conclude that biological interactions also play a role. Because most studies of vent ecology are observational and tend to target the larger species, the mechanisms causing observed successional replacements remain speculative, and patterns for a vast number of smaller, but numerically important, species (i.e., gastropods, polychaetes) are unexplored. The alternative approach is controlled manipulative experimentation, which has proven useful in elucidating mechanisms of succession in other marine environments (e.g., Dayton 1971, Sutherland and Karlson 1977, Sousa 1979*a, b,* Farrell 1991, Benedetti-Cecchi 2000).

The objective of our study was to characterize and quantify changes over time in the invertebrate communities colonizing new surfaces at hydrothermal vents along a gradient in vent fluid flux, and to examine what mechanisms may be effecting these changes. Incorporating successional theory from marine and terrestrial systems (starting with Connell and Slatyer 1977; modified with subsequent studies as reviewed in McCook 1994), and documenting colonization of new habitat during staggered, but overlapping, observation intervals (as in Ambrose 1984), we reasoned that faunal changes at vents are driven by one or more of the following processes:

1) Inhibition. The initial invertebrate colonists interact with later arrivals in a way that deters subsequent colonization. Under these conditions, we expect that colonists of a new habitat exposed over an extended interval would be fewer than the sum of colonists on similar habitats exposed sequentially over the same time period. The extreme case for inhibition is preemption, in which the initial colonists prevent future settlers or immigrants from becoming established (as described in Underwood and Denley 1984), and maintain their dominance for an extended period. In this case, the numbers and species composition of colonists on habitats exposed at the same time would be similar, regardless of the duration of exposure (presuming the absence of any external perturbation to restart succession).

2) Facilitation. The initial colonists interact with later arrivals in a way that enhances subsequent colonization. In this case, we expect that colonists on a habitat exposed over an extended interval would be more numerous and more diverse than the sum of colonists on similar habitats exposed sequentially over the same interval. The extreme case for facilitation is obligate facilitation (Odum 1969), in which a secondary species could colonize only after a pioneer species became established.

3) Tolerance. Initial colonists interact weakly or not at all with subsequent arrivals, so colonists accumulate as propagules arrive independently of the species composition or abundance of pioneers (Egler 1954). In this case, we expect that colonists on a habitat exposed over an extended interval would be equivalent to the sum of colonists on similar habitats exposed sequentially over the same interval. This scenario serves as our null model.

In addition to the above inhibition and facilitation models, which assume that species interactions drive the successional sequence toward a specific trajectory (canalized succession; Berlow 1997), the sequence of species replacement might vary in space and time depending on the variation in the physical characteristics of the environment and in the composition of the available larval pool (externally driven succession; Berlow 1997). In the highly dynamic vent habitat, successional changes may reflect species' responses to variation in

the environment (i.e., any of the thermal, geochemical, or nutritional properties that covary with vent fluid flux), as well as species' interactions with each other. Because it is important to distinguish between these mechanisms (e.g., Callaway and Walker 1997), we monitored both hydrothermal vent fluid flux (using temperature as a proxy; Johnson et al. 1988, 1994) and the ambient faunal community throughout the study. Also, because environmental and faunal variation occurs among, as well as within, individual vent fields, we conducted our studies at several different sites with distinctly different community structure, and over a gradient of vent fluid flux at each of these sites. In addition, we carried out the experiments over both short (months) and longer (years) time scales in order to evaluate interactions that may occur on different temporal scales. This temporal perspective becomes important when different mechanisms operate at different stages of succession (as reviewed by Caswell 1976).

It is important to recognize that change in species composition over the course of colonization of new habitat may be mediated by mobile predators that are not typically monitored as part of a successional study. For instance, an increase in abundance of a colonizing species may result from interactions with a facilitating species, or indirectly by reduction of a competing species through predation (as in Hixon and Brostoff 1996). Although the activities of large mobile predators were not monitored directly in the present study, information on their activities and diets is available from associated projects in vent habitats (Micheli et al. 2002; C. H. Peterson, *unpublished data*; G. Sancho, *unpublished data*).

MATERIALS AND METHODS

Study sites

Studies were conducted at vents near 9°50′ N, 104°17′ W along the East Pacific Rise (EPR; Fig. 1) at a water depth of 2500 m. This location was selected because nearby vent communities had been monitored frequently since 1991, when a well-documented volcanic eruption occurred (Haymon et al. 1993). The three vent fields selected appeared to be at different stages in the colonization sequence (as defined by Shank et al. 1998), with Worm Hole at an early stage (vestimentiferan clumps dominated by *T. jerichonana*), East Wall at a midstage (vestimentiferan clumps co-occupied by *T. jerichonana* and *R. pachyptila*; see Plate 1), and Biovent at a later stage (vestimentiferan clumps inhabited by *R. pachyptila* with co-occurring mussels).

At each site, the community inhabited an area that spanned a spatial gradient of hydrothermal fluid flux. With the exception of organisms inhabiting one focused "black smoker" orifice at Biovent (where hydrothermal fluids reached temperatures of 403°C, and contained concentrations of H_2S of up to 30 mmol/L; von Damm 2000), our three study sites experienced vent
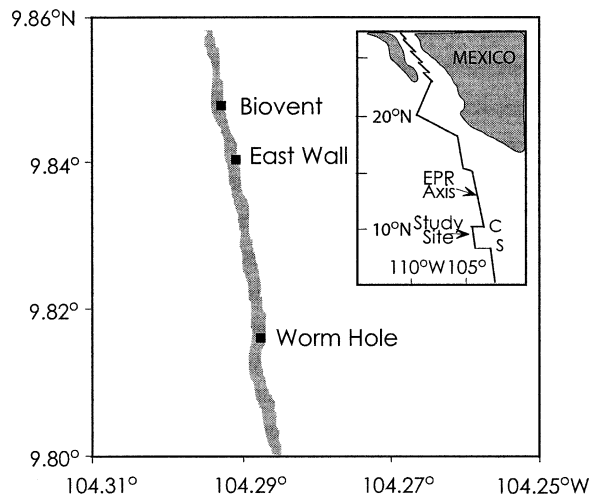

FIG. 1.   Map of East Pacific Rise axis (shaded corridor) near 9°50′ N, showing Biovent, East Wall, and Worm Hole vent sites between the Clipperton (C) and Sequieros (S) fracture zones.

fluids that had been released from the seafloor after mixing with ambient seawater (2°C) and exhibited temperature anomalies in the range of 30°C to <1°C. Because the high-temperature fluids were characterized by low pH and potentially toxic concentrations of both H_2S and heavy metals, a gradient in fluid flux represents a gradient in potential thermal and chemical stresses. The fluid flux gradient also is assumed to represent a productivity gradient because chemosynthetic production depends on the reduced chemicals in the vent fluids. This assumption is supported by the positive relation observed between vent fluid flux and faunal density and cover (C. Fisher, *unpublished data*).

The faunas at these sites displayed a striking biological zonation along the vent fluid flux gradient that is characteristic of vents in this region of the EPR (Hessler et al. 1985): alvinellid polychaetes and associated species inhabit areas of high-temperature vent flow (>25°C anomaly); vestimentiferan tubeworms occur in vigorous flow (up to 25–30°C above ambient); bivalves (mussels and clams) inhabit regions with moderate flow (up to 10°C above ambient); and various suspension feeders, including barnacles and serpulid polychaetes, inhabit weak flow (<2°C above ambient). The Biovent site exhibited all these zones, whereas the East Wall and Worm Hole sites did not exhibit black smokers (or the associated alvinellid residents) and at Worm Hole, the suspension-feeders were intermixed with sparse, juvenile mussels. For our colonization study, we excluded the black smoker habitat and designated the other distinct faunal zones by the prominent signature species as vestimentiferan, bivalve, and suspension feeder. At Worm Hole, the region with mixed bivalves and suspension feeders experienced moderate vent flows, but lacked the dense beds of mature mussels, so was designated as an intermediate bivalve/suspension-

PLATE 1.   Recovery of a basalt colonization block by the submersible Alvin from an East Pacific Rise vent community in a mid-successional stage (i.e., vestimentiferan zone dominated by the tubeworm *Riftia pachyptila* with no overgrowth by mussels). (Left) Limpets and polychaetes were included in analyses of colonization; large transient organisms such as the crab *Bythograea thermydron* were not. Limpets are also visible on the surface of vestimentiferan tubes in the background. (Right) The manipulator claw of the submersible Alvin is used to conduct studies of ecological succession at deep-sea hydrothermal vents. Clumps of the tubeworm *Riftia pachyptila* are typical inhabitants of mid-successional communities inhabiting zones of moderate-temperature venting (5°–25°C). Markers identifying experiments are visible in the background.

feeder zone. We also defined at all sites a periphery zone that lacked a detectable temperature anomaly and generally was occupied by deep-sea invertebrates with no known affinity to vents, except the vent-endemic foraminifer species *Abyssotherma pacifica*.

Experimental design

We tested for positive (facilitative) and negative (inhibitory) species interactions by quantifying colonization of vent species on cubic basalt blocks roughly 10 cm on a side. The submersible Alvin was used to position and recover the blocks in the appropriate habitats on the seafloor. For each of five deployment intervals, replicate blocks ($n = 3$) were introduced into four zones (vestimentiferan, bivalve, suspension feeder, periphery) at each of three vent fields (Biovent, East Wall, and Worm Hole; at Worm Hole, the bivalve and suspension-feeder zones were intermixed) located along a 2-km section of the axial valley of the ridge. ''Interval'' treatments were clustered in three replicate plots within each zone (Fig. 2). We were aware that the number of replicates was low, and might compromise our ability to detect treatment effects. However, given our lack of a priori knowledge of the natural history of the vent ecosystem, we chose to spread our studies over multiple sites and zones, with the intent of discovering trends that occurred generally over the temporal and spatial mosaic of vents on the EPR.

The fluid microhabitat of each block was characterized on deployment and recovery by measuring water temperature with the Alvin temperature probe at the lowermost extremity of the block (as in Mullineaux et al. 2000). Longer-term (months to years) temperature measurements were obtained with an internally recording ''Hobo'' temperature probe (Onset, Pocasset, Massachusetts, USA) at each zone for the duration of the block deployments. Ambient temperature of the bottom water was measured as 1.8°C by the Alvin probe, and as 2.0°C by the lower-resolution ''Hobo'' probe.

Sets of blocks were deployed and recovered on four cruises (in November 1994, April 1995, December 1995, May 1998) in a series of overlapping time intervals (November 1994 to April 1995, 5 mo; April 1995 to December 1995, 8 mo; November 1994 to December 1995, 13 mo; April 1995 to May 1998, 37 mo; December 1995 to May 1998, 29 mo). Blocks from the longest possible interval (November 1994 to May 1998, 42 mo) are not included in analyses because recovery was poor. The purpose of these overlapping deployments was to test for positive or negative biological interactions, in the context of succession theory, as modified from Connell and Slatyer (1977). The rationale followed that of Ambrose (1984): if the sum of the colonists on blocks in two consecutive (i.e., 5 and 8 mo) intervals is the same as the number in the continuous (13 mo) interval, then initial colonists would appear to have no detectable effect on later arrivals, supporting our null model (tolerance) of no effect of biological interactions on the successional sequence. If
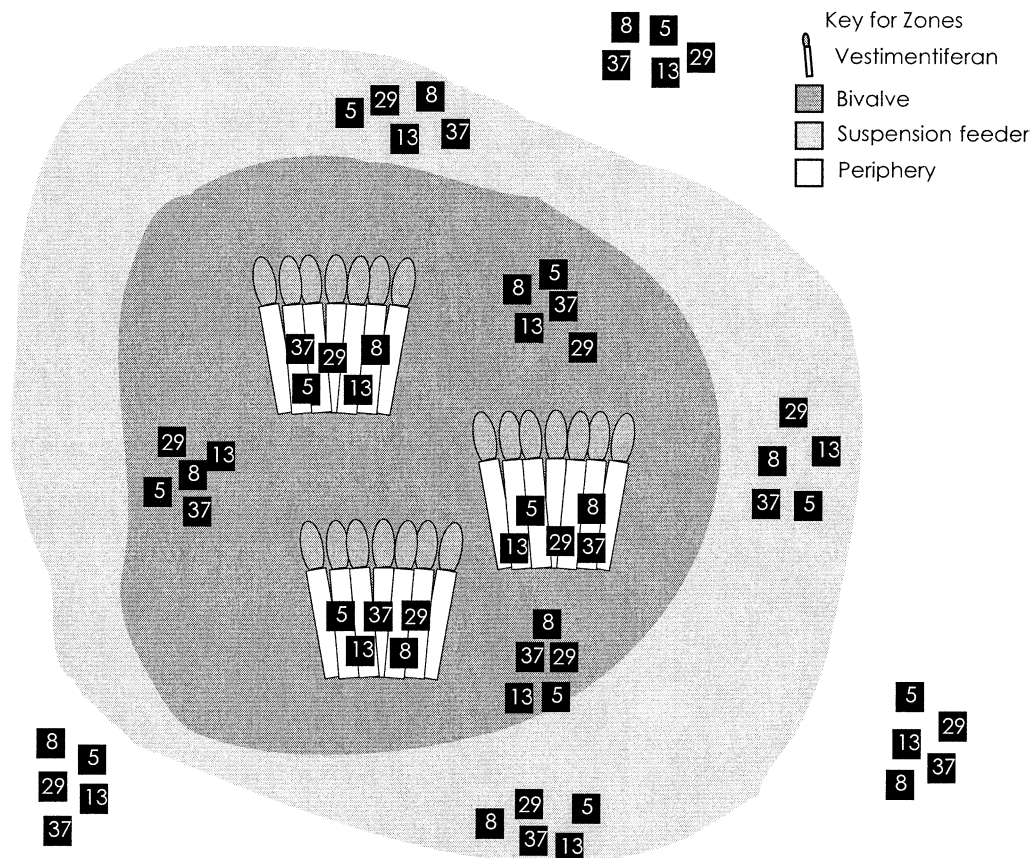
Fig. 2. Diagram (not to scale) of experimental design at a single vent site showing plots of colonization blocks with various ''interval'' treatments (5, 8, 13, 29, and 37 mo) within each zone.

the sum of colonists across the consecutive intervals is less than the numbers accumulated over the continuous interval then positive, facilitative interactions would be indicated. If, however, the sum of the colonists in the consecutive intervals is greater, inhibitory interactions, such as space preemption or competition for nutritional resources, would be suggested. Separate analyses were conducted for shorter (13 mo) and longer (37 mo) study intervals.

Blocks were recovered by the submersible Alvin and placed in individual collection compartments for transport back to the ship, as in Mullineaux et al. (2000). On shipboard, blocks and their attached colonists were preserved in 80% ethanol, as were any detached individuals from the compartment retained on a 63-μm sieve. In the laboratory, each block was examined under a dissecting microscope; all metazoan and selected protozoan (foraminifer and folliculinid ciliate) colonists were enumerated and identified (to species when possible). Folliculinid abundances were estimated from subsamples when densities exceeded 100 individuals per block face. From the sieved samples, all individuals >1 mm in length were counted and identified. The smaller individuals, mostly juvenile gastropods and

polychaetes, were very numerous but too difficult to identify to species using standard morphological techniques.

Species were grouped into mobile and sessile functional groups, based on their morphologies and on known behaviors of related species (Table 1). Pooling individual species into functional categories decreased variability among replicate blocks and increased the power of statistical tests. Analysis of functional groups in the short-term (13 mo) experiment was performed with a three-way nested MANOVA (Systat version 10, Systat, Evanston, Illinois, USA), testing effects of ''interval,'' ''zone,'' and ''site'' on sessile and mobile species groups. Intervals were categorized into two levels: pooled-consecutive, with colonists summed from the 5-mo and 8-mo blocks within each plot, and continuous (13 mo). A split-plot analysis (with intervals clustered into plots within zones) was originally planned for this design, but a nested approach was used instead because of the complexity of the multifactor MANOVA. Preliminary tests comparing the nested and split-plot approaches within individual sites showed no difference in detection of significant interval and zone effects. The zone factor had three levels (vestimentiferan, sus-

TABLE 1.   Mean numbers of individuals in taxa colonizing blocks over the short-term exposures (5-, 8- and 13-mo) at Biovent, East Wall, and Worm Hole, and long-term exposures (8-, 29- and 37-mo) at East Wall.

Taxon	Taxonomic group	Fn Grp	Biovent 13 mo ($n = 33$)	East Wall 13 mo ($n = 30$)	Worm Hole 13 mo ($n = 27$)	East wall 37 mo ($n = 33$)	ANOVA
Metafolliculina sp.	ciliate	S	54.45	451.57	818.31	834.21	prominent
Lepetodrilus elevatus	gastropod	M	65.24	24.27	186.39	22.27	prominent
Abyssotherma pacifica	foraminiferan	S	16.79	26.00	51.67	146.61	signature†
Amphisamytha galapagensis	polychaete	S	23.15	16.80	25.64	68.61	prominent
Vestimentiferans	vestimentiferan	S	16.55	24.87	38.61	20.00	signature†
Laminatubus alvini	polychaete	S	2.73	2.73	19.97	15.27	signature†
Lepetodrilus pustulosus	gastropod	M	8.76	8.40	19.08	3.24	prominent
Ophryotrocha akessoni	polychaete	M	2.24	4.83	4.42	8.18	prominent
Cyathermia natacoides	gastropod	M	8.97	12.17	4.89	3.30	
Ventiella sulfuris	amphipod	M	5.00	17.87	4.58	2.21	
Bathymodiolus thermophilus	bivalve	S	2.03	1.37	3.42	8.61	signature
Lepetodrilus ovalis	gastropod	M	1.39	0.57	9.33	8.55	⋯
Clypeosectus delectus	gastropod	M	1.33	2.23	5.08	3.82	⋯
Archinome rosacea	polychaete	M	1.33	1.23	1.61	5.94	⋯
Eulepetopsis vitrea	gastropod	M	0.36	0.30	5.33	3.24	⋯
Paralvinella spp?	polychaete	S	2.06	4.07	0.00	0.12	⋯
Gorgoleptis spiralis	gastropod	M	0.15	0.10	2.25	0.52	⋯
Lepetodrilus cristatus	gastropod	M	1.03	1.20	0.00	0.52	⋯
Nereis sp.	polychaete	M	0.15	0.47	0.92	0.85	⋯
Polynoid polychaetes	polychaete	M	0.27	0.47	0.53	0.79	⋯
Bathymargarites symplector	gastropod	M	0.36	0.33	0.14	1.00	⋯
Hesiospina vestimentifera	polychaete	M	0.09	0.00	0.00	1.42	⋯
Neolepas zevinae	barnacle	S	0.18	0.17	0.42	0.70	⋯
Nicomache sp.	polychaete	S	0.06	0.00	0.19	1.03	⋯
Unsegmented worm	mainly platyhelminths	M	0.12	0.10	0.81	0.18	⋯
Galapagomystides aristata	polychaete	M	0.82	0.00	0.00	0.06	⋯
Ophiuroids	ophiuroid	M	0.12	0.27	0.33	0.12	⋯
Helicoradomenia acredema	aplacophoran	M	0.06	0.03	0.14	0.55	⋯
Lepidonotopodium spp.	polychaete	M	0.00	0.13	0.50	0.09	⋯
Tanaids	crustacean	S	0.09	0.40	0.08	0.12	⋯
Bathypecten vulcani	bivalve	S	0.12	0.23	0.06	0.15	⋯
Iphionella risensis	polychaete	M	0.15	0.13	0.00	0.21	⋯
Cnidarian	cnidarian	S	0.00	0.47	0.00	0.00	⋯
Brown papillated worm	polychaete	M	0.03	0.00	0.14	0.18	⋯
Branchipolynoe sp.	polychaete	M	0.09	0.00	0.03	0.24	⋯
Gorgoleptis emarginatus	gastropod	M	0.00	0.17	0.00	0.18	⋯
Barnacle cyprids	barnacle	S	0.00	0.30	0.03	0.00	⋯
Glycera sp.	polychaete	M	0.03	0.07	0.06	0.09	⋯
Dorvilleid polychaete	polychaete	M	0.03	0.03	0.11	0.03	⋯
Anemone	anemone	S	0.00	0.03	0.08	0.09	⋯
Rhynchopelta concentrica	gastropod	M	0.00	0.00	0.00	0.18	⋯
Provanna sp.	gastropod	M	0.00	0.00	0.00	0.12	⋯
Prionospio sandersi	polychaete	M	0.00	0.07	0.00	0.06	⋯
Isopods	crustacean	M	0.03	0.07	0.00	0.00	⋯
Peltospira delicata	gastropod	M	0.00	0.03	0.00	0.00	⋯
Depressigyra planispira	gastropod	M	0.00	0.00	0.00	0.03	⋯
Hesionid polychaete	polychaete	M	0.00	0.03	0.00	0.00	⋯
Small gastropods		M	3.48	17.87	33.03	0.03	⋯
Small limpets		M	1.64	10.07	3.36	36.52	⋯
Gastropod juveniles	gastropod	M	5.12	27.93	36.39	36.55	⋯
Unidentified gastropod	gastropod	M	0.00	0.00	0.03	4.79	⋯
Polychaete juveniles	polychaete	M	0.06	0.60	0.53	0.24	⋯
Unidentified polychaetes	polychaete	M	0.09	0.03	0.08	0.12	⋯
Unidentified foraminifers	foraminiferan	S	0.06	0.73	0.28	0.52	⋯
Leptostracans	crustacean	M	0.00	0.00	0.19	0.00	⋯
Total mobile		M	98.45	86.27	107.67	279.11	Fn Group
Total sessile		S	118.27	529.83	1096.03	958.75	Fn Group
Total individuals			216.73	616.10	1203.70	1237.86	

Notes: Taxa are categorized into functional mobility groups (Fn Grp), based on inferences of predominant lifestyle made from morphology: M, mobile; S, sessile. Prominent colonizing taxa (among top 10 in at least three of the four site/exposure combinations) and signature species for each zone (vestimentiferans, *Bathymodiolus thermophilus*, *Laminatubus alvini*, and *Abyssotherma pacifica*) were analyzed individually by ANOVA.

† Signature species also prominent.

pension feeder, and periphery), with blocks of different intervals grouped into three plots within each zone. This analysis treated the bivalve/suspension-feeder zone at Worm Hole as comparable to a suspension-feeder zone, because it lacked the distinct three-dimensional habitat structure provided by the deep bivalve cover at the other two sites. The bivalve zones of Biovent and East Wall were excluded from the analysis because no comparable zone occurred at Worm Hole. A two-way nested MANOVA was used for the longer-term (37 mo) study at East Wall, with interval (two levels) and zone (four levels) as factors. Univariate ANOVAs were used to test the effect of factors that were significant in the MANOVAs. To determine which species contributed to the patterns exhibited by the functional groups, similar univariate ANOVAs were used to test treatment effects on abundant individual species. Prior to statistical analysis, the data were plotted and variances tested for homogeneity (using Cochran's tests at $\alpha = 0.05$) and for normal distribution; logarithmic transformations, $\ln(x + 1)$, were performed if necessary. Treatment means were compared using Tukey's post hoc comparisons. Sign tests (Sokal and Rohlf 1995) were used to evaluate Interval effects in selected zones.

RESULTS

Colonization in continuous vs. pooled-consecutive intervals

Of the 112 blocks recovered and analyzed, all but five had been colonized by vent-associated invertebrates, with metazoan densities (excluding swimming crustaceans) ranging up to 2271 individuals per block, and total densities (including foraminifers and ciliates) reaching >7000. On most blocks, less than half the available surface area was colonized, although a few were very heavily colonized by vestimentiferans or mussel byssal threads and had >90% of the available surface covered. Individuals were separated into 55 species or species-groups (Table 1) and assigned to functional mobility groups based on field observations, functional morphology or knowledge of related species. Colonists of the mussel *B. thermophilus* were assigned to the sessile group because they are attached with byssal threads, and did not function as mobile grazers like the other taxa assigned to the mobile group.

In the short-term experiment, the mobile and sessile groups showed very different patterns in response to interval and zone treatments. MANOVA revealed that the interval, zone, and site effects on the functional groups were significant ($P < 0.05$; Table 2). ANOVA showed a significant interval effect for the mobile group, which was more abundant in the pooled-consecutive than the continuous intervals ($P = 0.013$; Table 2). In contrast, the sessile group tended to be more abundant in the continuous than in pooled-consecutive intervals (Fig. 3), but this result was not significant

(Table 2). The interval effect on each functional group was most consistent in the zone where that group was most abundant; i.e., the mobile group in the vestimentiferan zone and the sessile group in the suspension-feeder zone (Fig. 3). ANOVA also showed significant zone and site effects and a zone × site interaction in both mobile and sessile groups. Post hoc Tukey tests showed that abundances of both groups generally decreased with decreasing fluid flux, with significantly higher abundances in the vestimentiferan zone than in the periphery (Table 2). Abundances in the vestimentiferan zone were significantly higher than in the suspension-feeder zone at Biovent and East Wall, but not Worm Hole.

In the longer-term experiment, MANOVA detected a significant effect of zone ($P = 0.02$; Table 2), but not interval, on the functional groups. ANOVA showed that this zone effect was significant for both mobile and sessile groups, with abundances in the periphery zone lower than were those in the other zones. ANOVA also showed that sessile invertebrate abundances differed significantly among the plots.

Nine taxa were selected (Table 1) to evaluate their individual contribution to the interval effects. Taxa were selected for analysis if they were among the 10 most abundant colonizing species in at least three of the four site/exposure data sets of Table 1 (the limpets *Lepetodrilus elevatus* and *Lepetodrilus pustulosus*, the polychaetes *Ophryotrocha akessoni*, *Laminatubus alvini*, and *Amphisamytha galapagensis*, vestimentiferans, the foraminifer *Abyssotherma pacifica*, and the ciliate *Metafolliculina* sp.), or if they were a signature adult resident of a zone (the mussel *Bathymodiolus thermophilus*). The "vestimentiferans" were treated as a group because small (<1 mm) individuals of the three species, *Riftia pachyptila*, *Tevnia jerichonana*, and *Oasisia alvinae*, could not be identified reliably to species using morphological techniques. Species level identification of small individuals is possible using molecular genetic techniques (as in Mullineaux et al. 2000), but that approach was not feasible for the present study because of the large number of vestimentiferan colonists.

In both the short- and longer-term experiments, a few individual species showed interval effects that were very similar to their corresponding functional group (Figs. 4–6). In the short-term study, three mobile invertebrate species, the limpets *L. elevatus* and *L. pustulosus* and the polychaete *O. akessoni* (except at Biovent), tended to be more abundant in the pooled-consecutive (5 + 8 mo) than the continuous (13 mo) intervals (Fig. 4). Two sessile invertebrate species, the protozoans *A. pacifica* and *Metafolliculina* sp., demonstrated the opposite pattern, usually being more abundant in the continuous (13 mo) than the pooled-consecutive (5 + 8 mo) interval. The polychaetes *A. galapagensis* and *L. alvini*, the vestimentiferans, and the mussel *B. thermophilus* did not show interval pat-

TABLE 2. MANOVAs comparing abundance of individuals in mobile and sessile groups (A) between continuous (13-mo) and pooled-consecutive (5 + 8-mo) intervals, among three zones (vestimentiferan, suspension-feeder, and periphery), and among three sites (Biovent, East Wall, and Worm Hole) in the short-term experiment; and (B) between continuous (37-mo) and pooled-consecutive (8 + 29-mo) intervals, among four zones (including bivalve) at East Wall in the longer-term experiment.

Source	df	Univariate ANOVA, mobile			Univariate ANOVA, sessile			MANOVA		
		MS	F	P	MS	F	P	Wilks' λ	F	P
A) Short-term (13 mo)										
Interval	1	5.23	6.71	0.02	0.22	0.16	0.69	0.73	5.08	0.01
Zone	2	90.56	116.18	<0.01	79.18	59.06	<0.01	<0.01	66.23	<0.01
Site	2	11.84	15.19	<0.01	41.47	30.94	<0.01	0.25	13.37	<0.01
Interval × Zone	2	0.50	0.65	0.53	1.83	1.36	0.27	0.90	0.73	0.58
Zone × Site	4	3.42	4.39	0.01	4.03	3.00	0.04	0.48	2.99	0.01
Interval × Site	2	0.75	0.96	0.40	1.23	0.92	0.41	0.85	1.17	0.37
Interval × Zone × Site	4	0.71	0.91	0.48	0.72	0.54	0.71	0.81	0.77	0.63
Plot (Zone)	6	1.19	1.53	0.21	0.78	0.59	0.74			
Error	26	0.78			1.34					
Total sum of squares	49	256.46			306.17					
B) Long-term (37 mo)										
Interval	1	2.02	1.50	0.27	<0.01	0.03	0.87	0.80	0.63	0.57
Zone	3	9.39	6.98	0.02	8.01	63.21	<0.01	0.14	3.90	0.02
Interval × Zone	3	0.60	0.45	0.73	0.07	0.55	0.67	0.70	0.33	0.91
Plot (Zone)	8	1.09	0.81	0.62	2.28	17.96	<0.01			
Error	6	1.35			0.13					
Total sum of squares		48.80			43.24					
Tukey test	v s b p				s v b p					

Notes: N = 3 for most cases; exceptions are displayed in Fig. 3. Logarithmic transformations ln(x + 1) were performed on data when necessary to obtain homogeneity of variances. For tests with a significant zone, site, or interval effect (P < 0.05), post hoc Tukey tests show means that are not significantly different (P > 0.05) connected with underlines (sites are designated as B, E, and W; zones as v, b, s, and p; intervals as C and P).

Tukey tests:

Short-term mobile WvP EvP BvC BvP WvC WsP WsC EvC EsP BsP EsC EpP WpP BsC WpC BpP EpC BpC

Short-term sessile Ws Wv Ev Bv Bs Es Wp Ep Bp

Long-term mobile v s b p

Long-term sessile s v b p

terns that were consistent among sites. ANOVA indicated that the interval effect was significant for *O. akessoni* (P < 0.05 without Bonferroni correction; Table 3), as well as site and zone effects, and interval × zone and zone × site × interval interactions. The interval interactions occurred because abundances were significantly higher in pooled-consecutive than continuous intervals in the suspension-feeder zone at all sites and in the vestimentiferan zone at East Wall and Biovent, but the opposite pattern occurred in the vestimentiferan zone at Biovent, and the effect was not significant in the periphery (Tukey tests, P < 0.05; Table 3). Interval effects were not significant for any of the other taxa.

Results from the longer-term experiment at East Wall (Figs. 4–6) were qualitatively similar to those of the short-term studies. Three mobile species (*L. elevatus*, *L. pustulosus*, and *O. akessoni*) and the vestimentiferans tended to be more abundant in the pooled-consecutive (8 + 29 mo) than in the continuous (37 mo) intervals. The sessile protozoan *A. pacifica* showed the opposite pattern in the suspension-feeder zone, where it tended to be more abundant in the continuous interval, whereas *Metafolliculina* sp. did not differ detectably between the intervals. The three other prominent

taxa (*L. alvini*, *A. galapagensis*, and *B. thermophilus*) tended to be more abundant in the pooled-consecutive intervals in the Vestimentiferan zone, and more abundant in the continuous interval in the suspension-feeder zone (Figs. 4–6).

The power of our analyses of variance was low due to limited replication and high variation among replicates. A sign test was used as an alternative method to evaluate Interval trends that were consistent across sites but not significant in the ANOVAs. In the suspension-feeding zone, mean sessile-group abundances were higher in the continuous than the pooled-consecutive intervals at all three sites and during both experimental durations. The probability of this pattern occurring in the absence of an interval effect would be P = 0.0625. The same pattern occurred for the sessile species *Metafolliculina* sp., whereas the opposite pattern (abundances higher in pooled-consecutive than continuous intervals in all four cases) occurred in the vestimentiferan zone for the mobile species *L. elevatus* and *L. pustulosus*. The low probabilities of these patterns occurring in the absence of an Interval effect suggest that our ANOVAs may have failed to detect a real effect for these groups.
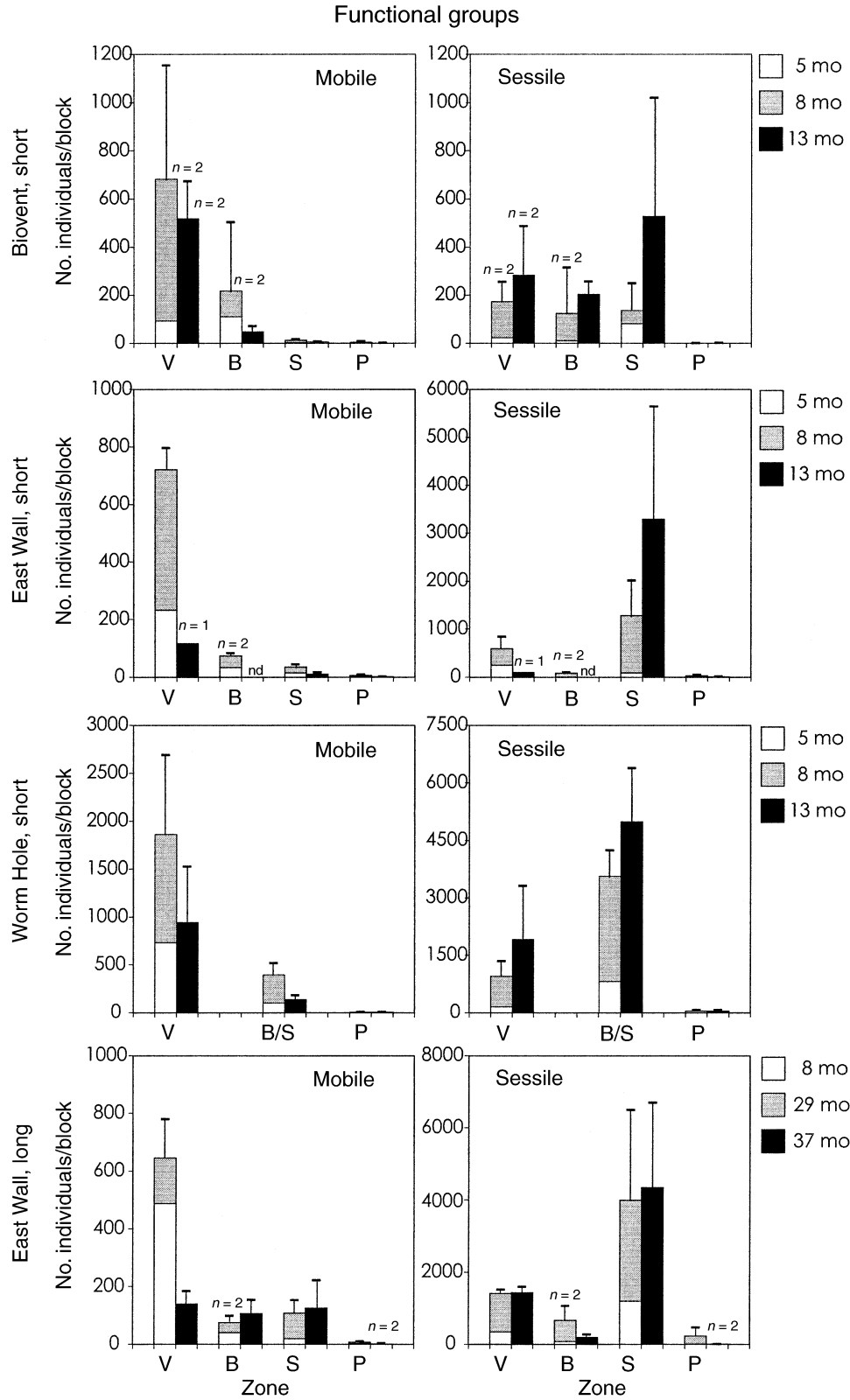
FIG. 3. Abundance of mobile and sessile species-groups on blocks from pooled-consecutive (short-term, 5 + 8 mo; longer-term, 8 + 29 mo) and continuous (short-term, 13 mo; longer-term, 37 mo) intervals, at four zones (vestimentiferan, bivalve, suspension-feeder, and periphery) in three sites (Biovent, East Wall, and Worm Hole). At Worm Hole, two zones were merged into a bivalve/suspension-feeder zone. Error bars represent 1 SE for $n = 3$, except as noted on figure (nd, no data). Results from a three-way MANOVA and post hoc Tukey tests are in Table 2.
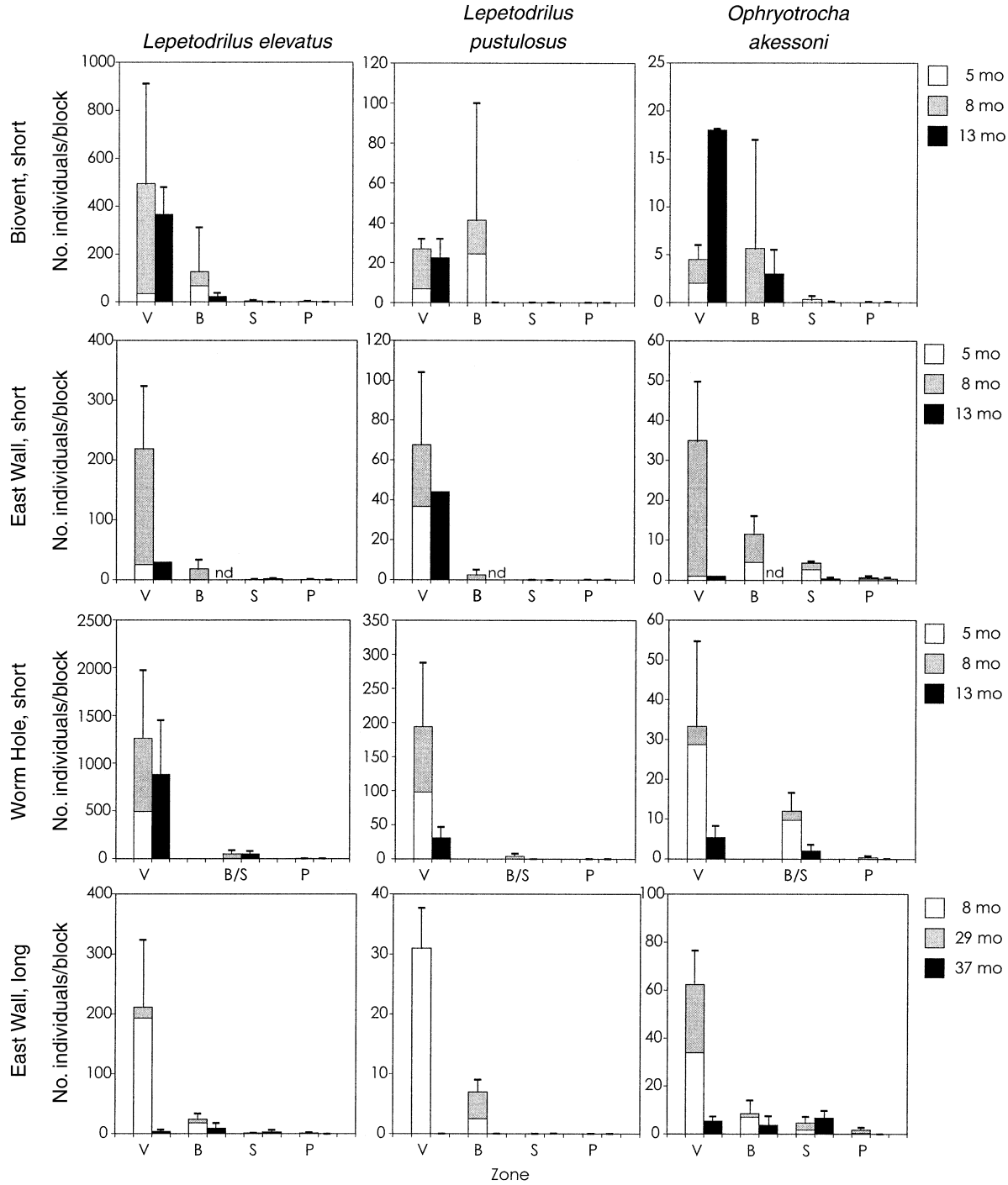
FIG. 4. Abundance of gastropods *Lepetodrilus elevatus* and *Lepetodrilus pustulosus* and the polychaete *Ophryotrocha akessoni* on blocks from pooled-consecutive (short-term, 5 + 8 mo; longer-term, 8 + 29 mo) and continuous (short-term, 13 mo; longer-term, 37 mo) intervals, at four zones (vestimentiferan, bivalve, suspension-feeder, and periphery) in three sites (Biovent, East Wall, and Worm Hole). Error bars represent 1 SE for $n = 3$, except as noted in Fig. 3. Results from a three-way MANOVA and post hoc Tukey tests are in Table 3.

Variation of colonization among zones and sites

Colonists of two of the zonal signature species, vestimentiferans and the serpulid polychaete *L. alvini*, settled in spatial patterns similar to the adults. Vestimen-tiferan colonists were found almost exclusively in the vestimentiferan zone; the exceptions occurred on four blocks in the bivalve or bivalve/suspension-feeder zones (Fig. 4). Video images of the exceptional blocks
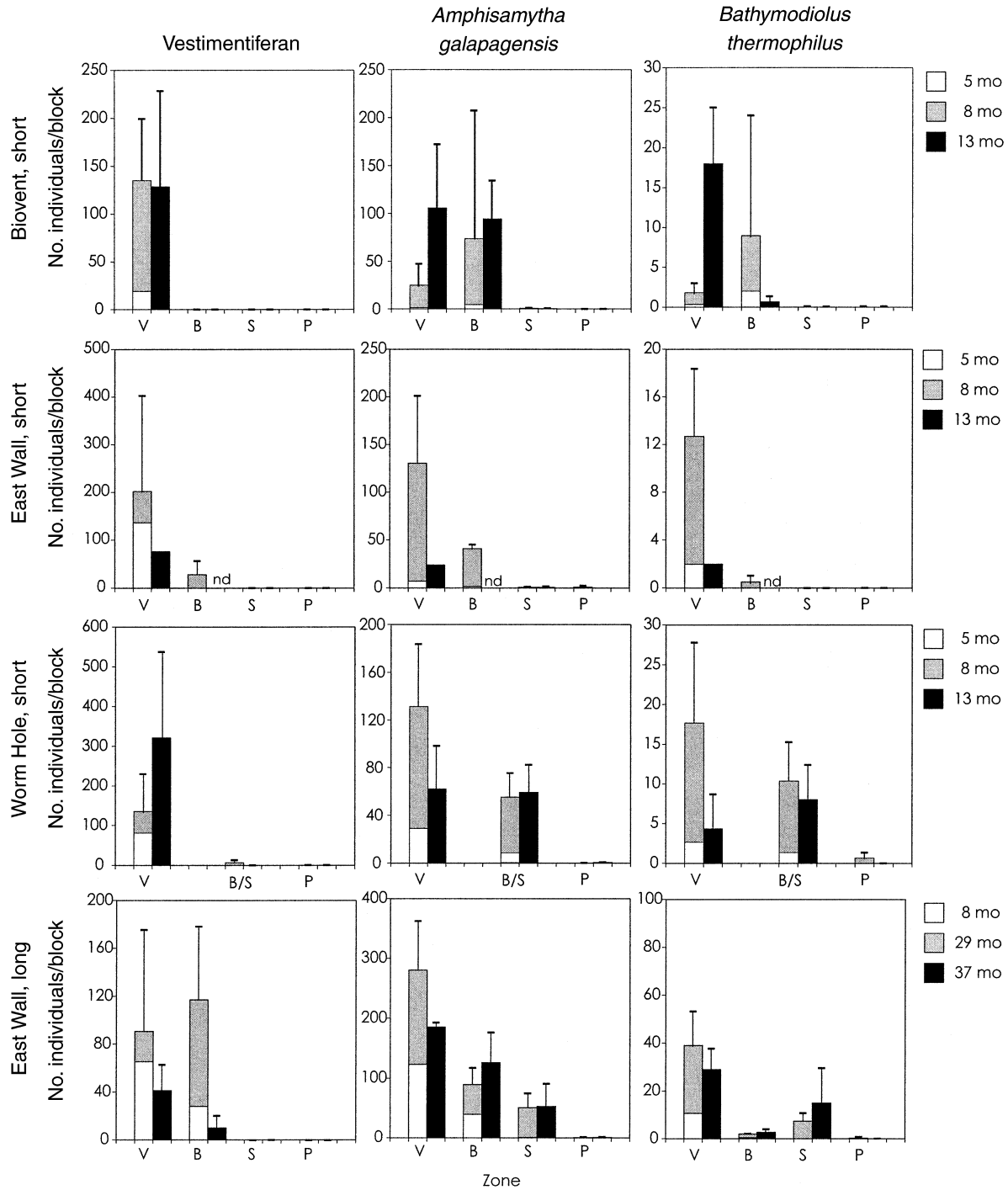
FIG. 5. Abundance of the polychaete *Amphisamytha galapagensis* and the mussel *Bathymodiolus thermophilus* on blocks from pooled-consecutive (short-term, 5 + 8 mo; longer-term, 8 + 29 mo) and continuous (short-term, 13 mo; longer-term, 37 mo) intervals, at four zones (vestimentiferan, bivalve, suspension-feeder, and periphery) in three sites (Biovent, East Wall, and Worm Hole). Error bars represent 1 SE for *n* = 3, except as noted in Fig. 3. Results from a three-way MANOVA and post hoc Tukey tests are in Table 3.

at East Wall revealed previously undetected resident vestimentiferans within 1 m of each block, suggesting that these blocks may have been placed in an anomalous microhabitat within that zone. The exceptional

block at Worm Hole had a measured temperature of 5.0°C, which was substantially higher than temperatures adjacent to the other blocks in the bivalve/suspension-feeder zone (1.8–3.8°C), and within the range
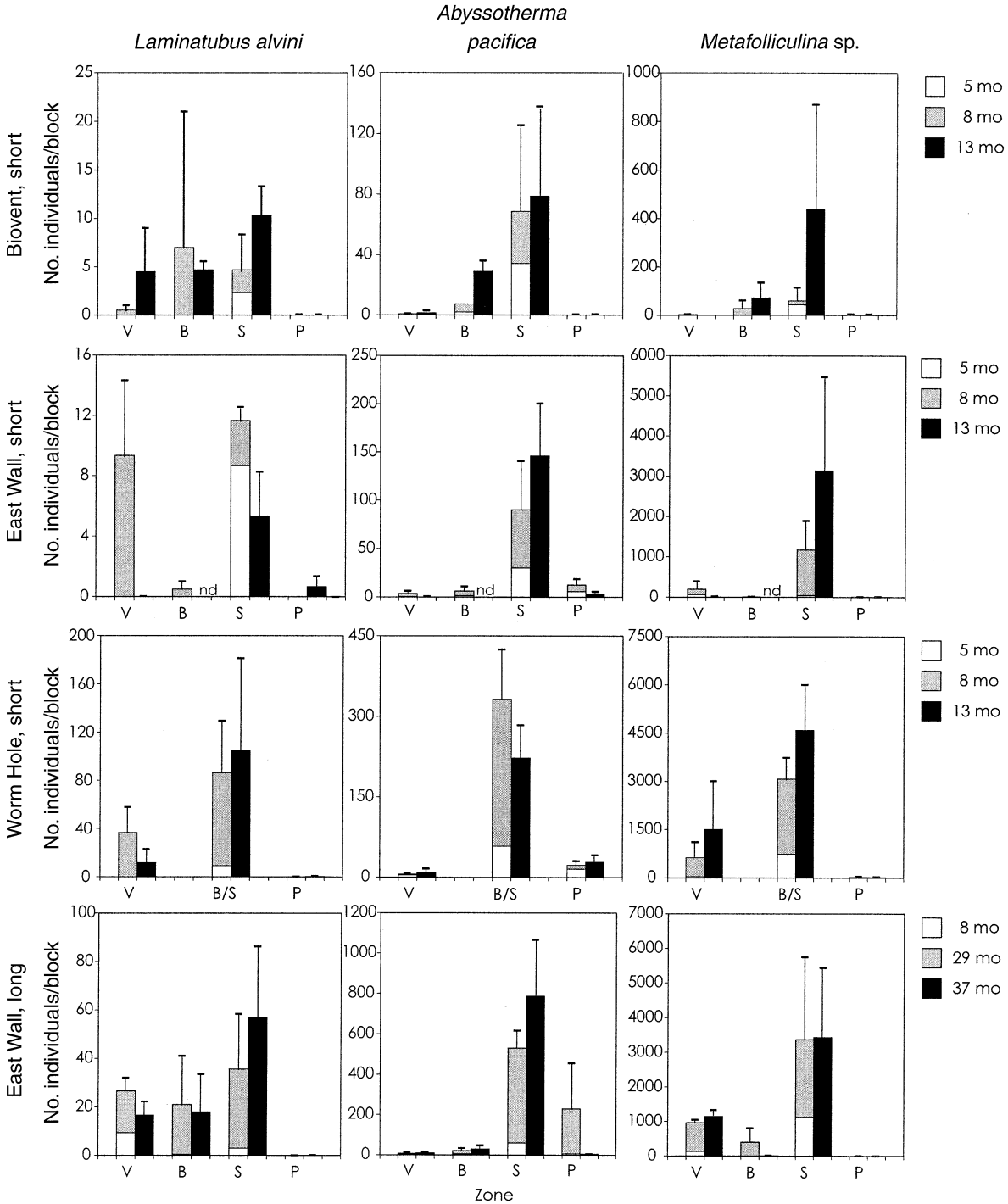
FIG. 6. Abundance of the polychaete *Laminatubus alvini*, the foraminifer *Abyssotherma pacifica*, and the ciliate *Metafolliculina* sp. on blocks from pooled-consecutive (short-term, 5 + 8 mo; longer-term, 8 + 29 mo) and continuous (short-term, 13 mo; longer-term, 37 mo) intervals, at four zones (vestimentiferan, bivalve, suspension-feeder, and periphery) in three sites (Biovent, East Wall, and Worm Hole). Error bars represent 1 SE for $n = 3$, except as noted in Fig. 3. Results from a three-way MANOVA and post hoc Tukey tests are in Table 3.

measured in the vestimentiferan zone at that site (1.9–10.9°C). Colonists of the signature species for the suspension-feeder zone, the serpulid polychaete *L. alvini*, were more abundant in that zone than elsewhere.

In contrast, the signature species of the bivalve zone, *B. thermophilus*, did not colonize in highest numbers there (Fig. 5). This anomalous pattern was most distinct at East Wall, where mussel adults were large and abun-

dant in the bivalve zone, but where mussel colonists were notably less abundant than in the vestimentiferan zone. At Worm Hole, where adult mussels were small and sparse, mussel colonization in the bivalve/suspension zone was not distinctly different from that in the vestimentiferan zone. Finally, colonists of the signature species for the periphery zone, *A. pacifica*, were more abundant in the suspension-feeder zones than the periphery (Fig. 6).

In the short-term experiment, colonist abundances varied significantly among sites (Table 2). Abundances of mobile individuals were highest at Worm Hole (Fig. 3), where this group was dominated by the lepetodrilid gastropods (Fig. 4). Abundances of sessile individuals also were highest at Worm Hole, where the ciliate *Metafolliculina* sp. was most numerous. Species abundant at any one site (i.e., those with mean abundances greater than five individuals per block) were widespread over all three sites (Table 1).

Temporal variation in environment and resident biota

Over the course of the colonization studies, there was little detectable trend in the thermal habitat measured by "Hobo" probes at any site, and (with one notable exception) little change in the resident faunas. During the 13-mo experiments, tidal variation in temperature on daily and fortnightly time scales was evident in the vestimentiferan zones of each site (Mullineaux et al. 2000). However, for periods longer than 1 mo, mean temperatures remained consistent over the interval with the exception of the final six weeks at Biovent, where temperatures increased by roughly 2°C (Fig. 3 in Mullineaux et al. 2000). Temperatures in the bivalve zones also displayed no longer-term trends (Fig. 7), despite the distinct fluctuations on time scales of days to weeks, and no temperature anomalies or trends were detected by "Hobo" probes in the suspension-feeder zones (data not shown; temperature anomalies of <0.1°C were below the detection limits of "Hobo" probes, and anomalies >0.1°C were detected with the Alvin probe at only three of 24 blocks in this zone). No change in relative dominance of resident species within zones, or movement in zonal boundaries of the resident fauna was detected in repeated video surveys at any of the sites over the 13-mo study (C. H. Peterson, *unpublished manuscript*; C. Fisher, *unpublished data*).

Characterization of temperature variation during the last 29 mo of the longer-term study was compromised by the loss of the "Hobo" temperature probe for that interval in the vestimentiferan zone, and the misplacement of the "Hobo" probe in the bivalve zone into a microhabitat with no temperature anomaly. Thus, the only record of temperature change at East Wall for this interval was from point measurements by the Alvin probe during placement and recovery of colonization blocks. A summary of these sparse measurements from the vestimentiferan and bivalve zones (Table 4) shows large variation but suggests no consistent decline or increase of vent fluid flow during this time. However, in May 1998 we noticed that some areas of the vestimentiferan zone that had been vigorously venting in December 1995 (as evidenced by shimmering water) had cooled to ambient temperatures.

Despite the absence of a detectable change in vent fluid flows at East Wall, the distribution of adult mussels expanded dramatically during the 29-mo interval between December 1995 and May 1998, with individuals appearing in the vestimentiferan zone in substantial numbers. Thus, by the end of the present study, the fauna inhabiting vigorous vent flow at East Wall became similar to the fauna in the comparable habitat at Biovent, where mussels also coexisted with vestimentiferans.

DISCUSSION

Negative and positive interactions along a gradient in hydrothermal fluid flux

The null model for our study was an expectation that early colonists would have no effect on later ones, leading to no difference in colonization between blocks exposed for continuous intervals and the sum of those exposed consecutively. Our results demonstrate that colonists of the Mobile functional group, which were more abundant in the Vestimentiferan zone, deviated significantly from this expectation, allowing us to reject the null (tolerance) model of succession for that group. Abundances of individuals in the mobile group were higher in pooled-consecutive than continuous intervals (Table 2), indicating that direct negative (inhibitory) interactions had occurred. These biological interactions resulted in faunal changes over time, even in the absence of consistent, long-term changes in the thermal (and inferred chemical) environment.

In contrast, colonists of the Sessile functional group tended to be more abundant in continuous than pooled-consecutive intervals in the Suspension-feeder zone where they were most numerous. This pattern was consistent across sites and experimental durations, although it was not significant ($P > 0.05$) in the ANOVA. A sign test indicated that such a pattern was unlikely to occur ($P = 0.065$) in the absence of an interval effect. Thus, we suggest that the patterns exhibited by the sessile group in the suspension-feeder zone are indicative of a positive (facilitative) influence of early colonists on later arrivals. Indeed, the MANOVA on joint abundances of sessile and mobile functional groups showed a significant shift with interval.

Several prominent individual species appear largely responsible for inhibitory or facilitative patterns exhibited by the functional groups to which they belong. For three mobile species, the limpets *Lepetodrilus elevatus* and *Lepetodrilus pustulosus* and the polychaete *Ophryotrocha akessoni* (except at Biovent), coloniza-

TABLE 3. ANOVAs comparing abundance of individuals between the continuous and pooled-consecutive intervals for the short-term (13-mo) and longer-term (37-mo) experiments.

Source	*L. elevatus* F	*L. elevatus* P	*L. pustulosus* F	*L. pustulosus* P	*O. akessoni* F	*O. akessoni* P	Vestimentiferan F	Vestimentiferan P
Short term								
Interval	0.21	0.65	0.87	0.36	11.04	<0.01†	4.85	0.04
Site	7.28	<0.01	0.99	0.39	3.87	0.03	0.30	0.74
Zone	70.67	<0.01	86.02	<0.01	37.74	<0.01	89.50	<0.01
Int × Zone	0.16	0.85	2.82	0.08	10.93	<0.01	3.13	0.06
Int × Site	0.18	0.84	0.25	0.79	2.54	0.10	6.63	0.01
Zone × Site	2.50	0.07	0.77	0.55	2.42	0.07	1.03	0.41
Zone × Site × Int	0.85	0.51	1.17	0.35	3.62	0.02	1.62	0.20
Plot (Zone)	1.37	0.26	0.99	0.45	0.68	0.67	10.28	<0.01
Error MS	1.32		0.66		0.39		0.85	
Sum of squares	269.6		144.61		69.01		256.81	
Longer term								
Interval	20.52	<0.01‡	266.25	0.00§	28.66	<0.01‖	15.41	0.01¶
Zone	10.68	0.01	113.17	0.00	22.45	<0.01	47.27	<0.01
Int × Zone	5.13	0.04	113.17	0.00	10.79	0.01	15.02	<0.01
Plot (Zone)	1.92	0.22	0.75	0.66	5.47	0.03	6.00	0.02
Error MS	0.80		0.03		0.22		0.39	
Sum of squares	71.22		32.25		39.00		99.43	

Notes: For the short-term experiment, three-way ANOVA included interval, zone (three levels; vestimentiferan, suspension-feeder, and periphery), and site (three levels; Biovent, East Wall, and Worm Hole). In the longer-term experiment, two-way ANOVA included interval and zone (four levels; vestimentiferan, bivalve, suspension-feeder, and periphery). Degrees of freedom as in Table 2. $N = 3$ for most cases; except as displayed in Fig. 3. Logarithmic transformations $\ln(x + 1)$ were performed on data when necessary to obtain homogeneity of variances. For tests with a significant interval effect ($P < 0.05$), post hoc Tukey tests shows means that are not significantly different ($P > 0.05$) connected with underlines (sites are designated as B, E, and W; zones as v, s, b, and p; intervals as C and P).

† Tukey test: EvP WvP BvC WsP EsP WvC BvP WsC EvC EpP EpC EsC BsP WpP WpC BsC BpC BpP

‡ Tukey test: vP bP bC vC pP sC sP pC

§ Tukey test: vP bP vC bC sC sP pP pC

‖ Tukey test: vP bP sC vC sP bC pP pC

¶ Tukey test: bP vC vP bC sC sP pC pP

tion in pooled-consecutive intervals was consistently greater than in continuous intervals in the vestimentiferan zone (Fig. 4). In the sessile protozoan *Metafolliculina* sp., the opposite pattern was apparent, with colonization greater in continuous than pooled-consecutive intervals in the suspension-feeder zone. Although these interval patterns were statistically significant in the ANOVAs only for *O. akessoni* in the short-term experiments at East Wall and Worm Hole, the sign test on the other three species indicates that the patterns were unlikely to occur ($P = 0.065$) in the absence of an interval effect. Statistical analyses at the species level were less powerful than at the functional group level, but these four species were likely the main contributors to the group responses.

The strength of inhibitory and facilitative interactions varied along a gradient of hydrothermal fluid flux at each site, which corresponded to a gradient in faunal cover and metazoan density on the colonization blocks. Inhibitory interactions among the mobile group were particularly evident in the vigorous fluid flux zone (vestimentiferan), where individuals were most abundant (Fig. 3). Facilitative interactions were evident among the sessile group only in the weak fluid flux (suspension-feeder) zone. These patterns suggest that com-petition may have been especially important in the vestimentiferan zone where metazoan densities were high, whereas facilitation was more prevalent in the suspension-feeder zone where densities were lower. Facilitation for the sessile group in the suspension-feeder zone was barely evident in the longer-term experiment at East Wall. In that case, the abundance pattern of the sessile group was driven largely by patterns of the numerically dominant *Metafolliculina* sp., whereas other sessile taxa in the group (e.g., *Bathymodiolus thermophilus*, *Laminatubus alvini*, and *A. pacifica*) showed the more typical trend of facilitation. The lack of detectable facilitation in *Metafolliculina* in the longer-term exposure suggests that, for this species, processes operating on periods of years may obliterate the patterns generated by facilitation during the early months of colonization.

The quantitative demonstration of inhibition of mobile species and facilitation of sessile ones in multiple vent sites that differed in successional stages suggests that these patterns can be generalized beyond a single type of vent ecosystem. Similarly, the consistency in the patterns among species within each functional mobility group allows us to predict that similar interactions will occur in other vent systems inhabited by these

Table 3. Extended.

A. galapagensis		B. thermophilus		L. alvini		A. pacifica		Metafolliculina	
F	P	F	P	F	P	F	P	F	P
0.21	0.65	0.20	0.66	2.05	0.16	0.59	0.45	1.37	0.25
20.43	<0.01	6.62	0.01	8.96	<0.01	15.91	<0.01	11.48	<0.01
88.16	<0.01	26.89	<0.01	30.09	<0.01	54.52	<0.01	25.53	<0.01
2.51	0.10	7.27	<0.01	1.94	0.16	0.58	0.57	0.43	0.66
0.01	0.99	0.11	0.90	2.43	0.11	0.55	0.59	2.40	0.11
12.80	<0.01	6.17	0.00	2.64	0.06	1.23	0.32	1.06	0.40
2.44	0.07	3.82	0.01	0.85	0.51	0.56	0.69	0.39	0.82
1.43	0.24	2.18	0.08	1.46	0.23	0.28	0.94	1.16	0.36
0.53		0.40		0.99		1.10		4.78	
169.728		64.27		136.2		196.8		572.2	
0.68	0.44	0.17	0.70	0.08	0.79	4.80	0.07	1.04	0.35
70.68	<0.01	17.45	<0.01	8.09	0.02	93.25	<0.01	21.44	<0.01
0.78	0.55	0.20	0.89	0.26	0.85	0.50	0.70	1.01	0.45
5.00	0.03	1.96	0.21	1.18	0.43	15.38	<0.01	1.11	0.46
0.29		0.58		1.35		0.25		2.47	
75.62		43.67		54.60		105.30		205.97	

functional groups, even if they do not host the same species. However, the difference in the relative strengths of inhibitory and facilitative interactions along a gradient in vent fluid flux (and inferred gradient in productivity) suggests that predicting which interaction will dominate at a specific location requires knowledge of the local hydrothermal fluid flux. Our ability to generalize and predict from these deep-sea experiments demonstrates the value of conducting manipulative studies over a broad range of environmental conditions, even in environments where such approaches are especially challenging.

Mechanisms for succession along a gradient in vent fluid flux

Any mechanisms proposed to account for the observed biological interactions must explain both the difference between responses of mobile and sessile functional groups to initial colonists, and the difference in the intensity and direction of these patterns along a gradient in vent fluid flux, productivity and faunal density. We suggest that the following processes contribute to successional changes in the vent community: (1) mobile species exert negative influences on both mobile and sessile colonists by removing ("bulldozing") new settlers; (2) sessile species are relatively immune to removal once firmly attached and relatively large; (3) sessile species exert negative influences on mobile colonists by occupying their grazing space; and (4) sessile species exert positive effects on sessile colonists by providing spatial refuge from bulldozing by mobile grazers, and/or a settlement cue. We expect negative interactions to be more prominent in regions of high
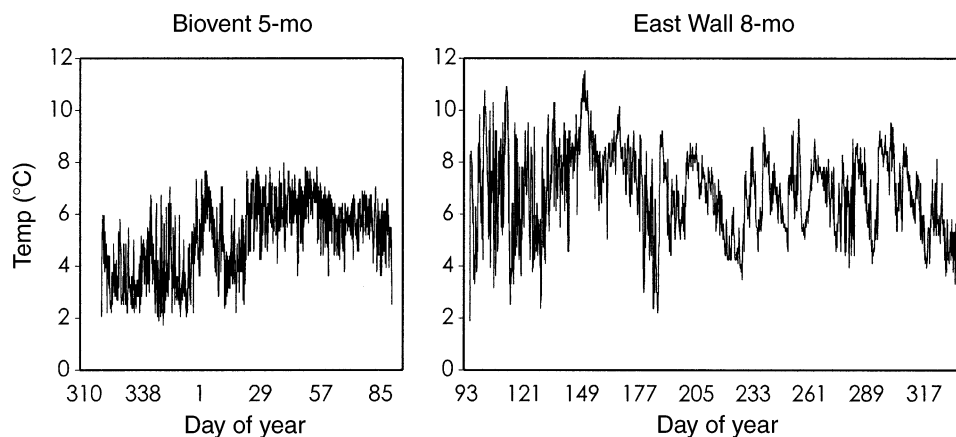

Fig. 7. Temperatures recorded in the bivalve zone over a 5-mo interval (November 1994–April 1995) at Biovent and over an 8-mo interval (April 1995–December 1995) at East Wall. Records for the 5-mo interval at East Wall are not shown because the temperature probe was not recovered. Records for the 5-mo interval at Worm Hole and the 8-mo intervals at Biovent and Worm Hole are not shown because no temperature anomaly was recorded at the probe locations.

TABLE 4. Temperature anomalies recorded at the base of blocks deployed at East Wall in April and December 1995 and recovered May 1998 (37-mo and 29-mo intervals).

Zone	Interval (mo)	Deploy temp. (°C)	Recover temp. (°C)	Change temp. (°C)
Vestimentiferan	29	1.5	5.4	3.9
	29	4.1	1.1	−3.0
	29	2.0	1.5	−0.5
	37	5.5	1.1	−4.4
Bivalve	29	2.9	0.0	−2.9
	37	5.3	6.6	1.3
	37	1.0	0.0	−1.0

Note: Only blocks with temperatures recorded successfully on deployment and recovery are shown.

faunal density where disruption by mobile grazers is frequent and grazing space limited. Positive interactions are more likely to occur in regions of lower faunal density where bulldozing is less frequent, allowing sessile species to become established and more effectively provide a refuge or settlement cue to subsequent sessile colonists. A diagram of these processes (Fig. 8) shows the relative importance of negative interactions decreasing and positive interactions increasing along a gradient of decreasing vent fluid flux and metazoan faunal density.

Support for these proposed interactions among and between mobile and sessile species comes from a mechanistic understanding of faunal interactions in other marine communities and from natural history observations of vent species. Mobile grazers such as limpets are known to impede attachment of other mobile or sessile species by physically dislodging (bulldozing) their recruits (Connell 1961, Stimson 1970, Dayton 1971, Menge 1976, Dixon 1978) and potentially can interfere with each other's feeding activities. Similarly, the presence of established sessile organisms may physically limit the size of, or access to, grazing areas of mobile individuals (note that although large vestimentiferan tubes can ultimately provide grazing area for some limpet species, tubes remained small in the present study). Their influence on the physical habitat is analogous in some ways to the effects of plants in terrestrial systems as "ecosystem engineers" (Jones et al. 1997). When new habitat is exposed at vents, the small mobile grazers have the potential to impede establishment of sessile species by removing newly settled individuals. However, it appears that at least some sessile individuals are able to recruit, and, once they grow to sufficient size, may begin to restrict the grazing territories and abilities of mobile species, thereby limiting the number of mobile colonists and causing the inhibition detected in our experiments. Alternatively, at high densities, mobile grazers also may remove enough food (expected to be surface-attached microbes; Hessler and Smithey 1983) to limit the food available for other grazing individuals. Thus, the inhibition of mobile individuals in the vestimentiferan zone could be due to direct interference from estab-

lished mobile or sessile colonists, and/or resource limitation among the mobile species.

Recruitment of sessile individuals may be facilitated by established sessile species through two different mechanisms. Gregarious settlement mediated by chemical or physical cues produced by the adults is common among tubiculous species (reviewed in Pawlik 1992), suggesting a potential mechanism for facilitative recruitment of sessile vent species (Mullineaux et al. 2000). Alternatively, established sessile species could facilitate other sessile individuals by interfering with grazing activities of mobile species and providing a refuge from bulldozing for new sessile recruits. One way to test for gregarious settlement on our colonization blocks would be to look for small-scale (millimeters to centimeters) aggregations in the colonists. However, such an analysis was outside the scope of our study, and thus we cannot use our colonization data to evaluate directly which process (provision of settlement cues or refuge from bulldozing) provides the most plausible explanation for the observed facilitation of sessile species. Although facilitation via a refuge from bulldozing may be important where mobile grazers are abundant, facilitation via a settlement cue seems a more likely explanation for facilitation in the suspension-feeder zone where mobile grazers are rare.

The prevalence of inhibitory interactions at the high end of the fluid flux gradient, where productivity and faunal densities are high, and of facilitative interactions at the low end, where productivity and densities are low, is consistent with our proposed mechanisms of species interactions. We expect negative interactions to be more intense in high-productivity regions, where faunal densities are high and mobile individuals can frequently bulldoze subsequent mobile settlers, or limit their foraging efficiency. Interference of mobile colonists by sessile individuals also is more likely at high faunal densities, where space is limited and sessile species not easily avoided. However, inhibition of sessile species in the high fluid flux region was not detected, suggesting that sessile settlers were less susceptible to bulldozing than mobile ones and/or that facilitation by established sessile individuals (via chemical cues or refuges from bulldozing) more than compensated for
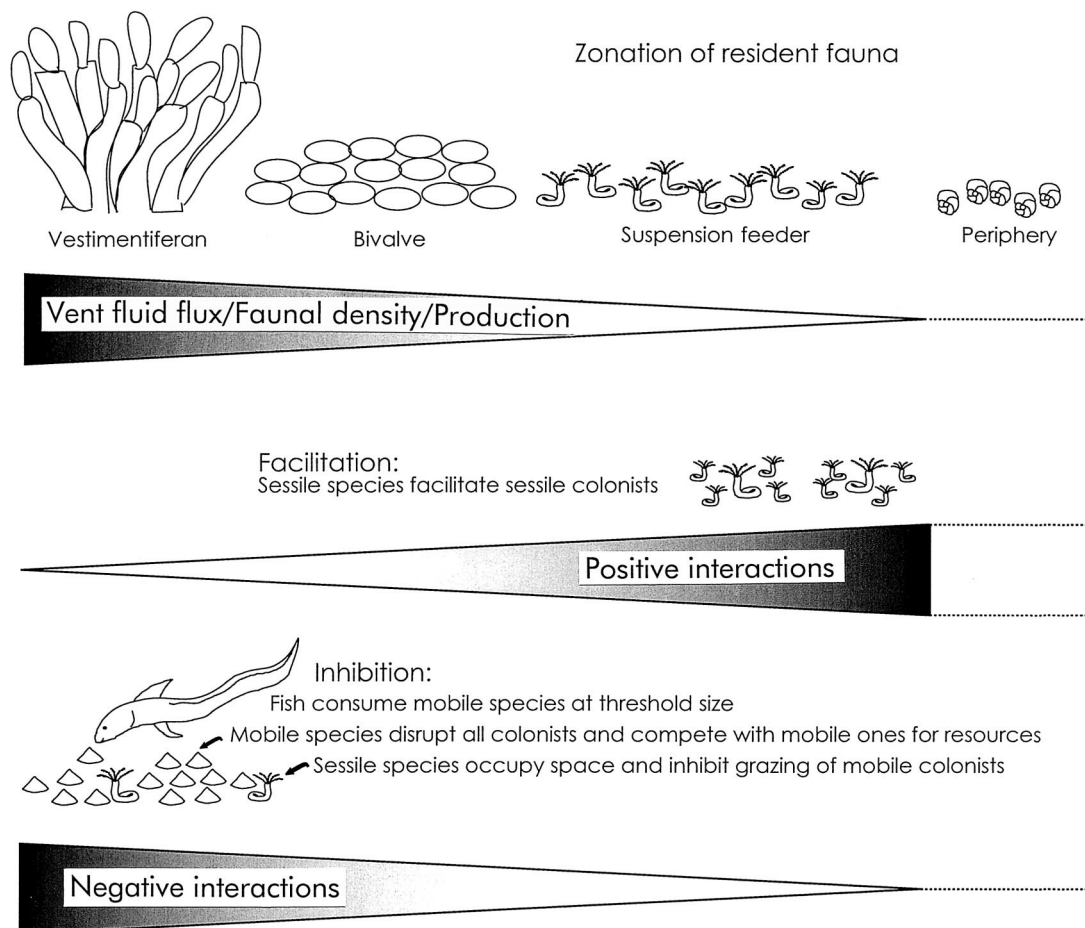
FIG. 8.   Diagram showing proposed mechanisms for species interactions along a production gradient at hydrothermal vents. Wedges indicate the relative intensity of hydrothermal fluid flux (as inferred from temperature measurements), and negative interactions and positive interactions (as indicated by interval effects in colonization experiments). Zonation of resident fauna indicates position of signature species for the vestimentiferan zone (vestimentiferans, including *Riftia pachyptila*, *Tevnia jerichonana*, and *Oasisia alvinae*), bivalve zone (vent mussel, *Bathymodiolus thermophilus*), suspension-feeder zone (serpulid polychaete, *Laminatubus alvini*), and periphery zone (foraminifer, *Abyssotherma pacifica*) along the fluid flux gradient. Supplementary evidence from vents and other environments suggests that facilitation of sessile species may be due to settlement cues and/or refuges from disturbance, whereas inhibition of mobile species may be due to disruption and competition from other mobile individuals, impediments to grazing imposed by sessile individuals, and/or indirect effects of size-selective consumption by fish. Dotted lines in the periphery zone indicate absence of detectable vent fluid flux and uncertainty about biological interactions due to small numbers of colonists.

interference by mobile ones. We expect positive interactions to be most prominent at the low end of the fluid flux gradient, where densities of mobile species are low, and their potential to interfere with sessile settlers minimal. In this region, facilitation of sessile settlers by established sessile individuals (most likely via chemical or physical settlement cues) should be readily detectable because it is not negated by inhibitory interactions. Furthermore, in the regions of weak vent fluid flux, vent fluid chemical and thermal cues are diluted, and biogenic cues may provide the best strategy for avoiding erroneous settlement into the food-poor suspension-feeder and periphery zones (Mullineaux et al. 2000).

Indirect effects on succession

Although our discussion of species interactions has concentrated mainly on direct effects of invertebrate colonists on each other, successional changes also could have been mediated by mobile predators that alter abundances of colonizing species. A series of independent observations on the behavior of the vent fish *Thermarces cerberus*, which is an abundant predator at vents along the northern East Pacific Rise (Geistdoerfer 1996), suggests that it potentially has a strong influence on the population density and size structure of limpets at vents. Gut contents show that a prominent component of this fish's diet are limpets, including *Lepetodrilus* spp. (Geistdoerfer 1996). Caging experi-

ments have demonstrated that gastropods attain higher densities when *T. cerberus* has been excluded (Micheli et al. 2002). Recent feeding observations indicate that *T. cerberus* concentrates its feeding on larger limpets, rather than the smaller size classes that are early colonists of newly cleared habitat (G. Sancho, *unpublished data*). Considered together, these observations suggest that *T. cerberus* may reduce the population sizes of *Lepetodrilus* spp. and other limpets at vents, but in a size-selective manner that gives the small, newly recruited individuals a size refuge.

Given these observations on the trophic role of *Thermarces cerberus*, we suggest that its predatory activities may be contributing to the inhibition pattern observed in mobile species in the present study (Fig. 8). When new habitat becomes available, mobile species colonize and appear to be immune to fish predation while they are small. Once the limpets reach a certain size, they become prey for the fish *T. cerberus*, and are cropped to lower levels. This process would result in lower limpet abundance in continuous experimental exposures where limpets reach larger sizes than in pooled-consecutive exposures. The effect of the vent fish is expected to be important in the vestimentiferan zone, but not in the other zones, where they rarely are observed feeding (L. Mullineaux, C. Petersen, F. Micheli, and S. Mills, *personal observations*) and have little effect on limpet populations (Micheli et al. 2002).

Microhabitat variation

Any interpretation of interval and zone effects on colonization must take into account the substantial thermal and chemical variation within zones, which can occur over scales as small as a few centimeters (Johnson et al. 1988). In the present study, this variation was documented in the form of water temperature measurements at individual blocks (complete data not shown). For instance, at East Wall, block temperatures differed from each other by as much as 22.7°C within the vestimentiferan zone, 14.4°C within the bivalve zone, and 0.9°C within the suspension-feeder zone (no anomaly or variation was detected in any of the periphery zones). This variation in vent fluid flow on small spatial scales (centimeters to meters) is likely responsible, in large part, for the observed high variability in colonization among blocks within a treatment (interval or zone) and the occasionally significant variations among plots within zones (e.g., for the sessile group in the longer-term experiment; Table 2). Microhabitat variation does not introduce bias into our analyses because variation among zones was much greater than variation within zones (Table 2), but it does illustrate that although our zones appear relatively homogeneous in terms of dominance by prominent signature species, they are not homogeneous environmentally.

The substantial variation in vent fluid flux over very small spatial scales was a surprise to us, and compro-

mised our experimental design. A split plot design clearly is not ideal in a habitat where environmental variability is high within plots. Furthermore, to increase power of experimental tests in such systems, the level of replication needs to exceed three (substantially) or a suite of temperature recorders must be employed to provide temperature covariates for each experimental unit to reduce error variance. Such experimentation is becoming possible in the deep sea as manipulative, observational and sampling capabilities improve, and background information on the natural history and dynamics of deep ecosystems becomes available.

Interpreting prior successional observations at vents

Vestimentiferan colonization patterns were not studied at the species level in the present study, but our results do contribute to a better understanding of the mechanisms causing the successional transition from vestimentiferans to bivalves observed in prior studies (Hessler et al. 1988, Shank et al. 1998). The mussel *Bathymodiolus thermophilus* colonized new habitat in the vigorous flow (vestimentiferan) zone quickly (in ≤5 mo) and in substantially higher numbers than in the other zones (Fig. 5). This result is surprising because the mussel is the signature adult species of the moderate fluid flux (bivalve) zone, and has not been observed as a pioneer colonist in photo- and video-based analyses of succession in the vigorous fluid flux zones of other vent sites along the East Pacific Rise (Hessler et al. 1988, Shank et al. 1998). One scenario consistent with all these observations is that mussels are early colonists of the Vestimentiferan zone but remain cryptic until they reach an age of >13 mo. Another possibility is that mussels are facilitated by the structure provided by mature vestimentiferans, perhaps via the increased surface area provided by their tubes. The impact of the mussels on the much larger vestimentiferans is unlikely to be substantial until they reach a size or population density that can divert flow from the vestimentiferan plumes (as proposed in Johnson et al. 1994) or otherwise disrupt them mechanically. Thus, we suggest that the transition from vestimentiferans to mussels at EPR vent sites involves facilitation of mussels by the sessile pioneer, which is subsequently outcompeted when the mussels reach a critical size or density.

Are vent communities controlled by biological or physical processes?

In summary, temporal change in colonization patterns of species at deep-sea hydrothermal vents appears to reflect inhibition of mobile species by initial colonists (both mobile and sessile), and possibly facilitation of sessile species by initial sessile colonists. The relative intensity of negative and positive interactions varies along a production gradient, with negative interactions most prominent where vent fluid flux and

faunal densities are highest. Our results suggest that biological interactions at the functional (mobility) group level can shape succession in a general, repeatable sequence, in the absence of consistent environmental change. Although physiological constraints and nutritional requirements of individual species no doubt set limits to their occurrence in the patchy and changeable vent environment (Fisher et al. 1988*a, b*), we conclude that their interactions with each other and with mobile predators have a strong influence on succession.

The development of our understanding of community structure at hydrothermal vent habitats reveals a parallel with ecological study in rocky intertidal habitats. Both systems exhibit strong gradients in the physical or chemical environment, and are characterized by faunal zonation that correlates with the gradients. In the rocky intertidal, manipulative studies have demonstrated that biological interactions are important in the dynamics and community structure of the ecosystem (reviewed in Connell 1972, Paine 1977). Comparable studies in deep-sea vent communities have the potential to produce a similarly enhanced perspective on the complex set of biological processes involved in establishing zonation patterns during community succession. As remote underwater technologies are developed, and ecological questions focused, it will become possible to design experiments with sufficient replication to test our proposed successional mechanisms and resolve current controversies over the relative importance of physical and biological processes at vents.

LITERATURE CITED

Ambrose, W. G. J. 1984. Influence of residents on the development of a marine soft-bottom community. Journal of Marine Research **42**:633–654.

Benedetti-Cecchi, L. 2000. Predicting direct and indirect interactions during succession in a mid-littoral rocky shore assemblage. Ecological Monographs **70**:45–72.

Berlow, E. 1997. From canalization to contingency: historical effects in a successional rocky intertidal community. Ecological Monographs **67**:435–460.

Butterfield, D., I. Jonasson, G. Massoth, R. Feely, K. Roe, R. Embley, J. Holden, R. McDuff, M. Lilley, and J. Delaney. 1997. Seafloor eruptions and evolution of hydrothermal fluid chemistry. Philosophical Transactions of the Royal Society of London, Series A **355**:369–386.

Callaway, R. M., and L. R. Walker. 1997. Competition and facilitation: a synthetic approach to interactions in plant communities. Ecology **78**:1958–1965.

Caswell, H. 1976. Community structure: a neutral model analysis. Ecological Monographs **46**:327–354.

Clements, F. E. 1916. Plant succession: an analysis of the development of vegetation. Carnegie Institution of Washington, Washington, D.C., USA.

Connell, J. H. 1961. Effects of competition, predation by *Thais lapillus*, and other factors on natural populations of the barnacle *Balanus balanoides*. Ecological Monographs **31**:61–104.

Connell, J. H. 1972. Community interactions on marine rocky intertidal shores. Annual Reviews of Ecology and Systematics **3**:169–172.

Connell, J. H., and R. O. Slatyer. 1977. Mechanisms of succession in natural communities and their role in community stability and organization. American Naturalist **111**:1119–1144.

Dayton, P. K. 1971. Competition, disturbance, and community organization: the provision and subsequent utilization of space in a rocky intertidal community. Ecological Monographs **41**:351–389.

Dixon, J. D. 1978. Determinants of the local distribution of four closely-related species of herbivorous marine snails. Dissertation. University of California, Santa Barbara, California, USA.

Egler, F. E. 1954. Vegetation science concepts I. Initial floristic composition. A factor in old-field vegetation development. Vegetatio **4**:412–417.

Farrell, T. 1991. Models and mechanisms of succession: an example from a rocky intertidal community. Ecological Monographs **61**:95–113.

Fisher, C. R., et al. 1988*a*. Microhabitat variation in the hydrothermal vent mussel *Bathymodiolus thermophilus*, at Rose Garden vent on the Galapagos rift. Deep-Sea Research **35**:1769–1792.

Fisher, C. R., et al. 1988*b*. Microhabitat variation in the hydrothermal vent clam, *Calyptogena magnifica*, at Rose Garden vent on the Galapagos rift. Deep-Sea Research **35**:1811–1832.

Franklin, J., and J. MacMahon. 2000. Messages from a mountain. Science **288**:1183–1185.

Fustec, A., D. Desbruyères, and S. K. Juniper. 1987. Deep-sea hydrothermal vent communities at 13°N on the East Pacific Rise: Microdistribution and temporal variations. Biological Oceanography **4**:121–164.

Geistdoerfer, P. 1996. L'ichthyofaune des écosystèmes associés à l'hydrothermalisme océanique: état des connaissances et résultats nouveaux. Oceanologica Acta **19**:539–548.

Grassle, J. F., and L. S. Morse-Porteous. 1987. Macrofaunal colonization of disturbed deep-sea environments and the structure of deep-sea benthic communities. Deep-Sea Research **34**:1911–1950.

Haymon, R. M., et al. 1993. Volcanic eruption of the mid-ocean ridge along the East Pacific Rise crest at 9° 45–52′N: direct submersible observations of sea-floor phenomena associated with an eruption event in April, 1991. Earth Planetary Science Letters **119**:85–101.

Hessler, R. R., and W. M. Smithey. 1983. The distribution and community structure of megafauna at the Galapagos

Rift hydrothermal vents. Pages 735–770 *in* P. S. Rona, K. Bostrom, L. Laubier, and K. L. Smith, editors. Hydrothermal processes at seafloor spreading centers. NATO Conference Series. Plenum Press, New York, New York, USA.

Hessler, R. R., W. M. Smithey, M. A. Boudrias, C. H. Keller, R. A. Lutz, and J. J. Childress. 1988. Temporal change in megafauna at the Rose Garden hydrothermal vent (Galápagos Rift; eastern tropical Pacific). Deep-Sea Research **35**:1681–1709.

Hessler, R. R., W. M. Smithey, and C. H. Keller. 1985. Spatial and temporal variation of giant clams, tubeworms and mussels at deep-sea hydrothermal vents. Bulletin of the Biological Society of Washington **6**:465–474.

Hixon, M. A., and W. N. Brostoff. 1996. Succession and herbivory: effects of differential fish grazing on Hawaiian coral-reef algae. Ecological Monographs **66**:67–90.

Johnson, K. S., J. J. Childress, C. L. Beehler, and C. M. Sakamoto. 1994. Biogeochemistry of hydrothermal vent mussel communities: the deep-sea analogue to the intertidal zone. Deep-Sea Research **41.**

Johnson, K. S., J. J. Childress, R. R. Hessler, C. M. Sakamoto-Arnold, and C. L. Beehler. 1988. Chemical and biological interactions in the Rose Garden hydrothermal vent field, Galapagos spreading center. Deep-Sea Research **35**:1723–1744.

Jones, C. G., J. H. Lawton, and M. Shachak. 1997. Positive and negative effects of organisms as physical ecosystem engineers. Ecology **78**:1946–1957.

Lubchenco, J. 1983. *Littorina* and *Fucus*: effects of herbivores, substratum heterogeneity, and plant escapes during succession. Ecology **64**:1116–1123.

MacDonald, K. C., K. Becker, F. N. Spiess, and R. D. Ballard. 1980. Hydrothermal heat flux of the ''black smoker'' vents on the East Pacific Rise. Earth and Planetary Science Letters **48**:1–7.

McCook, L. J. 1994. Understanding ecological community succession: causal models and theories, a review. Vegetatio **110**:115–147.

Menge, B. A. 1976. Organization of the New England rocky intertidal community: role of predation, competition, and environmental heterogeneity. Ecological Monographs **46**:355–393.

Micheli, F., C. H. Peterson, L. S. Mullineaux, C. R. Fisher, S. W. Mills, G. Sancho, G. A. Johnson, and H. S. Lenihan. 2002. Predation structures communities at deep-sea hydrothermal vents. Ecological Monographs **72**:365–382.

Mullineaux, L. S., C. R. Fisher, C. H. Peterson, and S. W. Schaeffer. 2000. Vestimentiferan tubeworm succession at hydrothermal vents: use of biogenic cues to reduce habitat selection error? Oecologia **123**:275–284.

Odum, E. P. 1969. The strategy of ecosystem development. Science **164**:262–270.

Paine, R. T. 1977. Controlled manipulations in the marine intertidal zone, and their contributions to ecological theory. Pages 245–270 *in* The changing scenes in natural sciences, 1776–1976. Academy of Natural Sciences, Philadelphia, Pennsylvania, USA.

Pawlik, J. 1992. Chemical ecology of the settlement of benthic marine invertebrates. Oceanography and Marine Biology: an Annual Review **30**:273–335.

Shank, T. M., D. J. Fornari, K. L. von Damm, M. D. Lilley, R. M. Haymon, and R. A. Lutz. 1998. Temporal and spatial patterns of biological community development at nascent deep-sea hydrothermal vents (9°50′ N, East Pacific Rise). Deep-Sea Research **45**:465.

Sokal, R. R., and F. J. Rohlf. 1995. Biometry. W. H. Freeman, New York, New York, USA.

Sousa, W. P. 1979*a*. Disturbance in marine intertidal boulder fields: the non-equilibrium maintenance of species diversity. Ecology **60**:1225–1239.

Sousa, W. T. 1979*b*. Experimental investigations of disturbance and ecological succession in a rocky intertidal algal community. Ecological Monographs **49**:227–254.

Stimson, J. 1970. Territorial behavior of the owl limpet, *Lottia gigantea*. Ecology **51**:113–118.

Sutherland, J. P., and R. H. Karlson. 1977. Development and stability of the fouling community at Beaufort, North Carolina. Ecological Monographs **47**:425–446.

Underwood, A. J., and E. J. Denley. 1984. Paradigms, explanations, and generalizations in models for the structure of intertidal communities on rocky shores. Pages 151–180 *in* D. R. Strong, D. Simberloff, L. G. Abele, and A. B. Thistle, editors. Ecological communities: conceptual issues and the evidence. Princeton University Press, Princeton, New Jersey, USA.

von Damm, K. L. 1995. Controls on the chemistry and temporal variability of seafloor hydrothermal fluids. Pages 222–247 *in* S. E. Humphris, R. A. Zierenberg, L. S. Mullineaux, and R. E. Thomson, editors. Seafloor hydrothermal systems: physical, chemical, biological, and geological interactions. Geophysical Monograph Series. American Geophysical Union, Washington, D.C., USA.

von Damm, K. L. 2000. Chemistry of hydrothermal vent fluids from 9°–10° N, East Pacific Rise; ''time zero,'' the immediate posteruptive period. Journal of Geophysical Research, B, Solid Earth and Planets **105**:11,203–11,222.

Witman, J. K. 1987. Subtidal coexistence: storms, grazing, mutualism, and the zonation of kelps and mussels. Ecological Monographs **57**:167–187.