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Phenotypic Variation In The Bryozoan *Leioclema Punctatum* (Hall, 1858) From Mississippian Ephemeral Host Microcommunities

By: **Steven J. Hageman** and Jennifer A. Sawyer

Abstract

The morphologic expression of microenvironmental variation is difficult to document in fossil ecosystems and therefore is poorly understood. However, documentation of environmental sources of variation in the phenotype is essential for meaningful studies of microevolution and speciation. A fossil assemblage from the Mississippian (Valmeyeran) Warsaw Formation near St. Louis, Missouri, provides necessary conditions to evaluate microenvironmentally induced phenotypic variation in the Paleozoic trepostome bryozoan Leioclema punctatum (Hall, 1858). Specimens of L. punctatum, found as fragments in 22 discrete piles, were collected in their entirety from a weathered surface. Each pile contained 20-200+ branch fragments of L. punctatum, which were all originally attached to large, soft-bodied hosts (sponges?). Multiple attachment bases were found in most piles, indicating that 1) multiple L. punctatum colonies (genotypes) are represented in each pile, and 2) each pile represents a near contemporaneous, relatively short-lived microcommunity. Morphological characters were measured (four per section) from two branches for each of two specimens from five separate piles. Results from completely random, nested, one-way ANOVA indicate that no highly significant differences exist among microcommunities or between colonies for any measured characters, but that significant variation exists within colonies and among colonies in the same microcommunity (pile). That is, submicroenvironmental variation, within and among colonies, can play a greater role in morphogenesis than environmental heterogeneity within a given environmental setting (undifferentiated facies).

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PHENOTYPIC VARIATION IN THE BRYOZOAN *LEIOCLEMA PUNCTATUM* (HALL, 1858) FROM MISSISSIPPIAN EPHEMERAL HOST MICROCOMMUNITIES

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ABSTRACT—The morphologic expression of microenvironmental variation is difficult to document in fossil ecosystems and therefore is poorly understood. However, documentation of environmental sources of variation in the phenotype is essential for meaningful studies of microevolution and speciation. A fossil assemblage from the Mississippian (Valmeyeran) Warsaw Formation near St. Louis, Missouri, provides necessary conditions to evaluate microenvironmentally induced phenotypic variation in the Paleozoic trepostome bryozoan *Leioclema punctatum* (Hall, 1858). Specimens of *L. punctatum*, found as fragments in 22 discrete piles, were collected in their entirety from a weathered surface. Each pile contained 20–200+ branch fragments of *L. punctatum*, which were all originally attached to large, soft-bodied hosts (sponges?). Multiple attachment bases were found in most piles, indicating that 1) multiple *L. punctatum* colonies (genotypes) are represented in each pile, and 2) each pile represents a near contemporaneous, relatively short-lived microcommunity. Morphological characters were measured (four per section) from two branches for each of two specimens from five separate piles. Results from completely random, nested, one-way ANOVA indicate that no highly significant differences exist among microcommunities or between colonies for any measured characters, but that significant variation exists within colonies and among colonies in the same microcommunity (pile). That is, submicroenvironmental variation, within and among colonies, can play a greater role in morphogenesis than environmental heterogeneity within a given environmental setting (undifferentiated facies). Microenvironmental factors affect the size and shape of mesopores (space-filling structures) more than other morphological characters.

Results are encouraging for the general application of the preserved fossil phenotypes as proxies for biological species. This conclusion is based on the absence of systematic variation at microenvironmental levels, measurable here, but not normally distinguishable in paleontological and sedimentological studies. Correct attribution of fossil species assumes, however, that the source and the relative importance of the low-level (submicroenvironmental) variation on development/ontogeny is recognized and attributed appropriately. Results call for a reevaluation of the application of within versus among colony variation used as a proxy for environmental stability.

INTRODUCTION

SMALL-SCALE, MICROENVIRONMENTALLY induced phenotypic variation in the fossil record is often difficult to quantify. In order to do so, an ancient population or adjacent correlative populations must be preserved. Typically, the fossil record does not provide these conditions, even on a single bedding plane. Studies of microenvironmentally induced variation can, however, be accomplished through detailed morphometric analysis of fossil specimens that accumulated in unusual circumstances, nearly contemporaneous in ecological time. A fossil assemblage from the Valmeyeran Warsaw Formation (340 Ma) near St. Louis, Missouri, provides an unusual opportunity to evaluate phenotypic variation using the trepostome bryozoan *Leioclema punctatum* (Hall, 1858).

Microevolution is the accumulation of genetic changes that occur within a single species up to and including the appearance of a new species (biological definition of a species being the potential to interbreed, producing fertile offspring). Because we cannot directly observe reproductive habits of ancient organisms, and genetic material is not typically preserved, paleontologists rely almost entirely on the phenotype of the preservable hard-parts for distinctions among species. Morphology is generally used, therefore, as a paleontological proxy for genetic composition. Complexity is introduced in the process of the recognition of ancient species, however, because the phenotype is controlled only partially by the genotype. The phenotype can be greatly impacted by environmental influences. If environmental effects are significantly greater than the genetic signal, this may cause fossil organisms to be incorrectly assigned at the species level of taxonomic organization (Simpson, 1953; Mayr, 1963; Levinton, 2001). A fundamental question for the systematics of fossil organisms: What is the degree to which genetics controls an individual organism's phenotype versus the degree to which the environment affects an individual's phenotype?

The colonial nature of bryozoans (modules of identical genotype within colonies) provides an excellent model for evaluation of sources of phenotypic variation (Abbott, 1973; Farmer and Rowell, 1973; Schopf, 1976; Pachut, 1982; Key, 1987; Hageman, 1995; Holdener and Hageman, 1998). The purpose of this paper is to document microenvironmentally induced phenotypic variation in the trepostome bryozoan *Leioclema punctatum* from the Mississippian Warsaw Formation at three hierarchical levels. This study does not address the overall limits of variation for this species or interspecific variation among its congeners. The aim of this study is an evaluation of genetically versus environmentally induced variation within a near contemporaneous local assemblage (i.e., a population).

METHODS AND MATERIALS

Study organism.—The organism used in this study is the middle Mississippian bryozoan *Leioclema punctatum.* This species was selected based on its abundance in well-preserved discrete microcommunities on a well-exposed outcrop of planar extant, and the availability of suitable morphometric characteristics (Fig. 1). Conspecificity of these specimens was presumed based on 1) the distinctiveness of the characteristics among all described *Leioclema* species, and 2) the presence of a single morphotype of *Leioclema* Ulrich, 1882 at the study site based on general inspection. However, the documented presence of "cryptic species" in some Modern cheilostome bryozoans (Cheetham et al., 1993, 1994, 1995) warrants caution in any assumption of conspecificity. If multivariate analysis of detailed morphometric data collected from specimens from this study yields multiple, distinctive groups, then the presence of potential "cryptic species" would need to be addressed. As shown in the Results section herein, this is not the case. Therefore, there is no evidence to support the rejection of the assumption that all specimens in this study belong to the same biological species.

Leioclema punctatum is a member of the class Stenolaemata

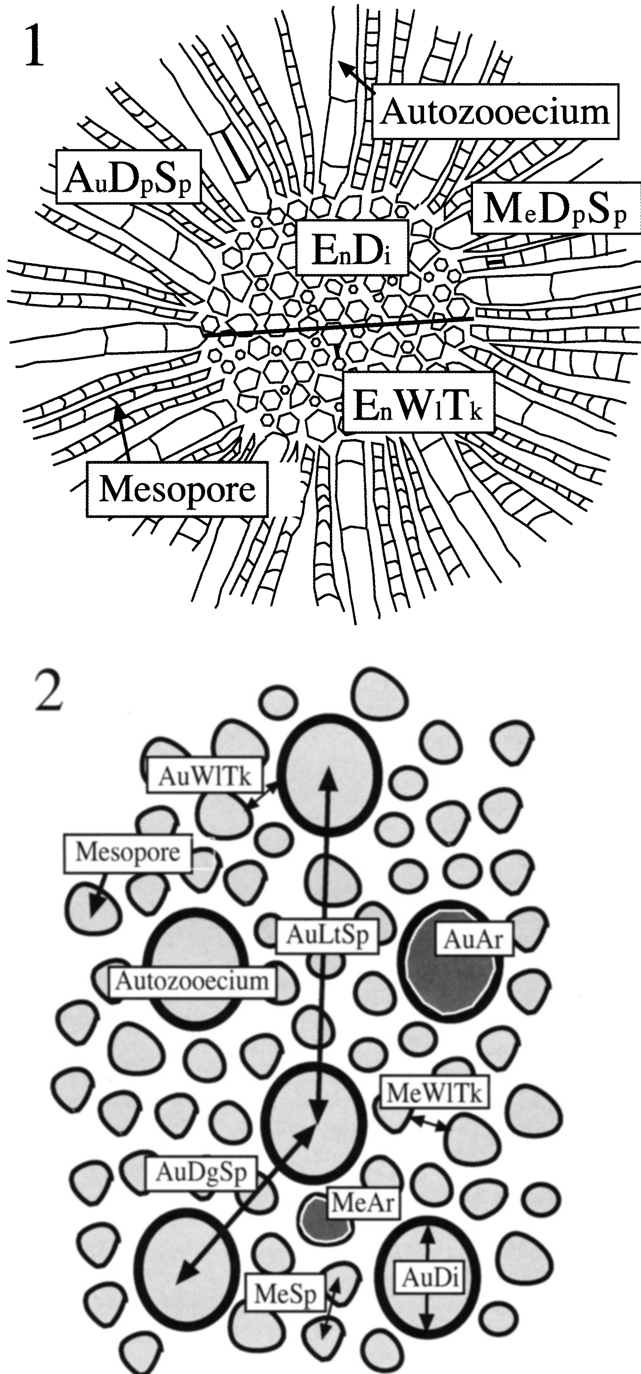

FIGURE 1—*1,* Reconstruction of transverse section of *Leioclema punctatum* (Hall, 1858), ×20, showing position of measurements. Explanation of abbreviations: EnDi = endozone diameter, EnWlTk = endozone wall thickness, AuDpSp = autozooecium diaphragm spacing, MeDpSp = mesopore diaphragm spacing. *2,* Reconstruction of tangential section of *Leioclema punctatum*, ×36.5, showing position of measurements. Explanation of abbreviations: AuDi = autozooecium diameter, AuAr = autozooecium area, AuDgSp = autozooecium diagonal spacing (within quincunx), AuLtSp = autozooecium lateral spacing (across quincunx), MeSp = mesopore spacing, MeAr = mesopore area, AuWlTk = autozooecium wall thickness, MeWlTk = mesopore wall thickness.

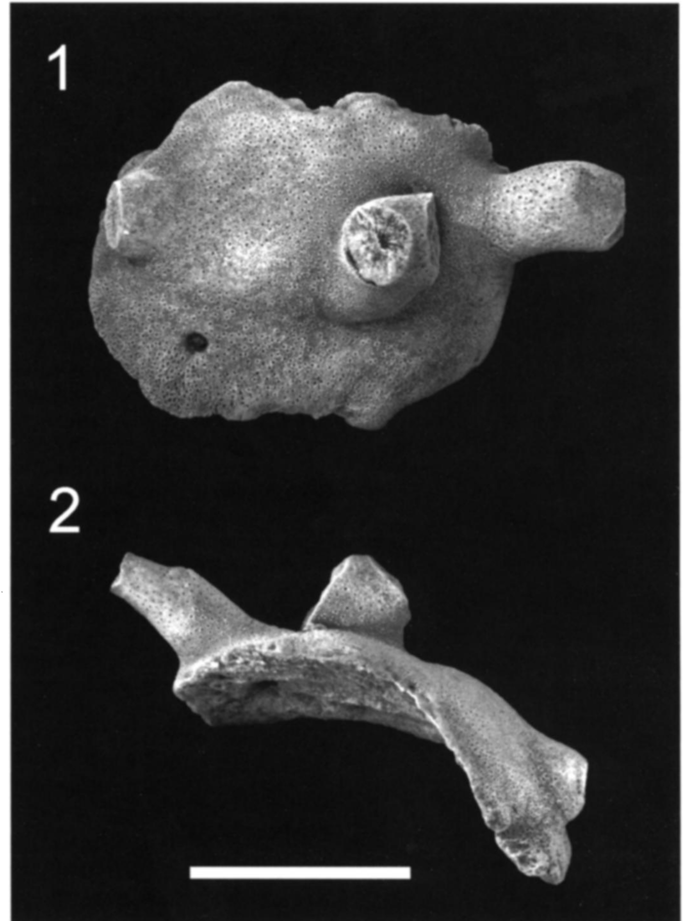

FIGURE 2—Colonial base piece from a single colony of *Leioclema punctatum*, ×3. *1,* Branch fragments for this study were taken from bases such as these. *2,* The curvature of the bases suggests that specimens were growing on a columnar host, such as a sponge or algal frond. Scale bar = 1.0 cm.

and the order Trepostomata. Kenozooids are common in this order (McKinney and Jackson, 1991), and are represented by mesopores in *Leioclema*. *Leioclema* is a member of the suborder Halloporoidea Astrova, 1965 and the family Heterotrypidae Ulrich, 1890 (Wyse Jackson, 1996). Members of the genus *Leioclema* can be encrusting, erect branching, or massive (Bassler, 1953). Diaphragms are complete in autozooecia and mesopores and are tabulate in mesopores (Bassler, 1953) (Fig. 1.1).

Leioclema punctatum was described by Ulrich (1890) as "parasitic" (grew on some soft-bodied host). Based on the morphology and curvature of attachment bases (Fig. 2) of *L. punctatum*, this host is assumed to be cylindrical in shape with stalk/body diameters of 1–2 cm. The basal attachment surfaces of five well-preserved colonies were studied using latex molds in an attempt to determine the identity of the host substrate or other bioimmured organisms. Although detailed, the surface texture of the basal attachment did not provide evidence sufficient for identification of the host. Possible hosts for *L. punctatum*, however, include sponges or algal stems.

Geological and ecological setting.—Specimens used in this study were collected by D. B. Blake in 1968 from the Warsaw Formation (Valmeyran, Mississippian) (Fig. 3), from a weathered surface, parallel to near horizontal bedding at a then-recently excavated road cut surface located in the center of the southwest

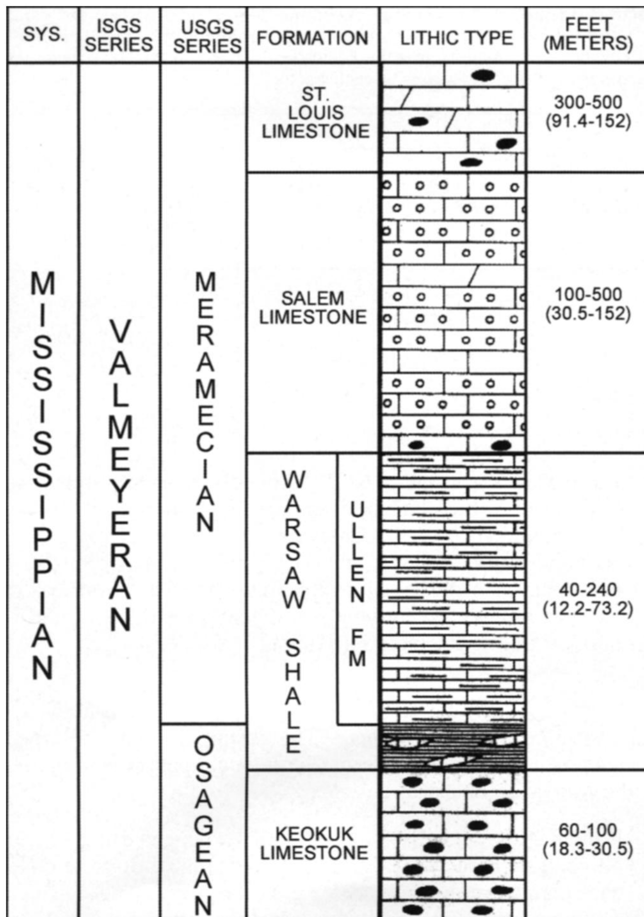

FIGURE 3—Stratigraphic column showing the Valmeyeran Warsaw Formation, after Snyder (1991, fig. 2, p. 13).

cloverleaf connecting the outer belt of St. Louis (junction of Interstate 270 with Interstate 44).

At this site, a thin interval of calcareous fossils was present in a mudstone. Fortuitously, the area had been graded during construction to a level just above the fossiliferous interval. The surface of the mudstone later eroded uniformly, leaving a lag of calcareous fossils. Specimens of *L. punctatum* were collected from discrete piles separated by approximately 1 m laterally. Individual, scattered fossil fragments between piles were not collected. Each discrete pile represents the remnants as a relatively short-lived microcommunity that grew on an ephemeral host substrate such as a sponge or alga (Hageman et al., 2000). Each pile consisted of the fragments of colonies that collapsed, but were otherwise untransported after the death of the host. Collectively, these piles represent a near contemporaneous assemblage in ecological time. This is supported by the relatively undisturbed nature of the piles in a setting above storm wave based (Snyder, 1987) on the presence of otherwise normal benthic marine fauna (Blake, personal commun., 2005). Multiple attachment bases were found for most piles, suggesting that multiple colonies and therefore multiple discrete genotypes are represented by each pile.

Materials used in study.—Twenty-two piles, each containing 20–200+ branch fragments and attachment bases, were collected in their entirety, each representing one discrete microcommunity. Two representative attachment bases were chosen from each suitable microcommunity. Suitability of specimens for inclusion in

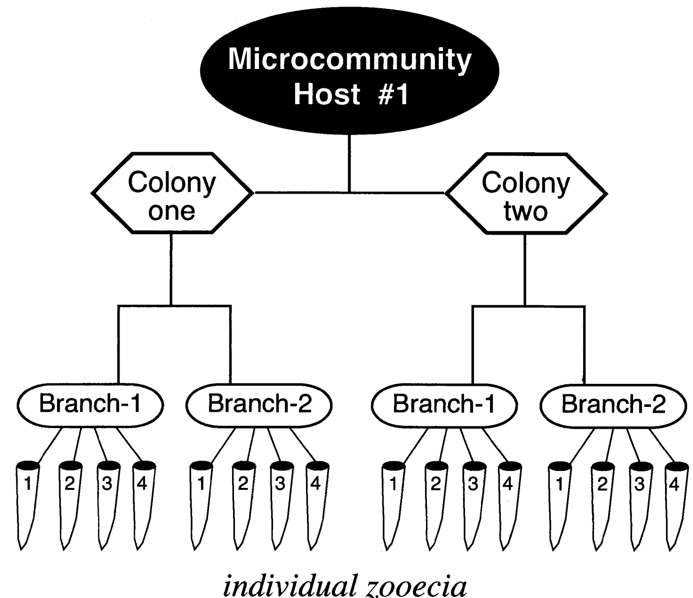

FIGURE 4—Experimental design for completely random, nested ANOVA design used in this study (one of five microcommunities shown). Colonies are nested within microcommunities and branches are nested within colonies and microcommunities. Four observations were made for each character (Fig. 1). N = Five microcommunities, a = two colonies per microcommunity, b = two branches per colony, c = four observations measured per branch (see Table 1).

the study was based on: 1) the availability of attachment bases bearing two or more branches with measurable characters within the microcommunity; 2) the use of branches that did not show deformation or crushing; and 3) colony bases with all zooecial outlines diverging from a single point (indicative of a single ancestrula rather than fused colonies of multiple genotypes). In all, five microcommunities were chosen, each contributing two attachment bases, that each contributed two branches (Fig. 4). Although not quantified, all specimens selected had approximately the same thickness of exozones, thereby minimizing ontogenetic differences among colonies.

Methods.—Colonial attachment bases were cut into two pieces with one branch each. Each branch was embedded in Buehler Epo-Thin low viscosity epoxy. The branches were cut, sanded, and polished to expose transverse and tangential sections. Specimens were etched in 0.05% formic acid for approximately one minute and imprinted on acetate slides using methods of Nye et al. (1972) and Snyder (1991).

Morphometric data were collected using images collected with an Olympus CC12™ digital video camera and analysis with Olympus MicroSuite™-Basic. Eight characters were measured from tangential sections, illustrating spacing, diameter, and area of mesopores and autozooecia, as well as their wall thicknesses (Fig. 1.2). Four independent measurements were collected from each section for each character (Fig. 4). These measurements were autozooecium area (AuAr), autozooecium diameter (AuDi), mesopore area (MeAr), mesopore spacing (MeSp), autozooecium lateral spacing (AuLtSp), autozooecium diagonal spacing (AuDgSp), autozooecium wall thickness (AuWlTk), and mesopore wall thickness (MeWlTk). Four characters were measured from transverse sections (Fig. 1.1), representing endozone diameter (AuDi) and wall thickness (AuWlTk), as well as spacing of diaphragms of autozooecia (AuDpSp) and mesopores (MeDpSp).

TABLE 1—Sum of squares table from nested Analysis of Variance for endozone diameter (EnDi in μm, Fig. 1), including degrees of freedom and F-ratio determination for all factors random and "c-branch" nested within "b-colony," and "b-colony" nested within "a-microcommunity" from Zar (1999, appendix 7) (df = degrees of freedom, SoS = sum of squares, MS = mean SoS, *P*-value = probability that the Factor/level is due to random effects, Var. = variance accounted for by each Factor/level, %Var. = the percentage of the total variance accounted for by each Factor/level).

Factor/level	df calc.	df	SoS	MS	F calc.	F	*P*-value	Var.	% Var.
Microcommunity	a-1	4	1099274	274,819	MSa/MSb	0.47	0.7605	−19,707	0
$Colony_{[microcom]}$	a(b-1)	5	2950628	590,126	MSb/MSc	3.04	0.0633	49,513	42.14
$Branch_{[colony]}$	ab(c-1)	10	1940249	194,025	MS/MSn	7.47	<0.001	42,011	35.75
Residual	abc(n-1)	60	1558763	25,979				25,979	22.11
Total	abcn-1	79	7548915	95,556				117,503	100

a = # of Microcommunities = 5.
b = # of Colonies/Microcommunity = 2.
c = # of Branches/Colony = 2.
n = # of Residuals/Branch = 4.
EnDi Mean = 1410.75.
Standard Error = 58.61.

Data were analyzed using a completely random, nested ANOVA model with SAS Statistical Software (version 8.2). Table 1 shows the appropriate degrees of freedom and F-value determinations for a completely random, nested design (Zar, 1999). It should be noted that a *random model* addresses the question as to whether the significance for sources of variation can be generalized to the entire statistical population (here, all specimens of *Leioclema punctatum* from this locality), as compared to a *fixed* model, which would consider the significance only of the specimens used in this study. In general, a random model requires a higher degree of differentiation among group means than a fixed model to achieve equivalent levels of significance. Therefore, patterns reported here are very strong and in theory have broader applicability beyond just the specimens studied. The null hypotheses for this study (Fig. 4) are:

H_0: no significant difference exists among the group means for the factor *Microcommunity*.
H_1: no significant difference exists between the group means for the factor *Colony* within *Microcommunity*.
H_2: no significant difference exists between the group means for the factor *Branch* within *Colony*, nested within *Microcommunity*.

Critical F-values for the ANOVA model were acquired using SISA <http://home.clara.net/sisa/signhlp.htm> (Uitenbroek, 2004). Principal components analysis with varimax rotation (Fig. 5) was applied to the entire data set (omitting AuDgSp due to missing data) in order to help visualize the distribution of the variance within and among levels in the study and to account for correlations among characters (SysStat, v. 5.2.1).

RESULTS

Conspecificity of specimens.—The assumption that all specimens studied belong to the same biological species is supported by the following observations.

1. Principal components analysis of all specimens using 11 morphometric characters (Fig. 5.1) did not reveal discrete differences among colonies.
2. The observed variation within single colonies (known, identical genotype) is only slightly less than the variation among colonies (different genotypes) (Fig. 5.1). If cryptic species such as those documented by Cheetham et al. (1993, 1994, 1995) were present in these specimens, differentiation of groups would be observed in plots such as 5.1.

TABLE 2—Summary of characters based on *P*-value and percent of total variance for each character accounted for by each factor. Percent variance across a character through all levels plus residual sums to 100% (e.g., EnDi = 0.0% + 42.1% + 35.8% + 22.1% = 100.0%). *P*-values in bold are significant at the level of ≤ 0.05.

	(1) Microcommunity		(2) Colony		(3) Branch		(4) Residual
	(A) *P*-val	(B)% var	(A) *P*-val	(B)% var	(A) *P*-val	(B)% var	% var
AUTOZOOECIUM							
Diameter (AuDi)	0.426	2.5	0.543	0.0	**<0.001**	63.6***	33.8
Area (AuAr)	0.292	11.3*	0.461	0.3	**<0.001**	59.8***	28.7
Lateral Spacing (AuLtSp)	0.251	17.0*	0.112	22.4*	**0.001**	22.7*	37.8
Diagonal Spacing (AuDgSp)	0.082	34.2**	0.281	7.4	**0.001**	22.5*	35.9
Diaphragm Spacing (AuDpSp)	0.576	0.0	0.612	0.0	**<0.001**	53.2***	46.8
MESOPORE							
Area (MeAr)	0.632	0.0	**0.026**	42.8**	**0.010**	16.7*	40.5
Spacing (MeSp)	0.527	0.0	**0.033**	39.9**	**0.011**	17.0*	43.1
Diaphragm Spacing (MeDpSp)	0.855	0.0	0.286	12.4*	**<0.001**	43.7**	43.8
WALL THICKNESS							
Autozooecium (AuWlTk)	0.190	22.8*	0.075	22.4*	**0.019**	14.0*	40.8
Mesopore (MeWlTk)	0.454	2.0	0.182	22.4*	**<0.001**	41.4**	34.3
Endozone (EnWlTk)	0.415	4.3	0.220	18.3*	**<0.001**	43.0**	34.4
ENDOZONE							
Diameter (EnDi)	0.761	0.0	0.063	42.1**	**<0.001**	35.8**	22.1
Average % variance per Factor		7.8		19.2		36.1	36.8

* Factor accounts for > 10% variance for this character.
** Factor accounts for > 25% variance for this character.
*** Factor accounts for > 50% variance for this character.

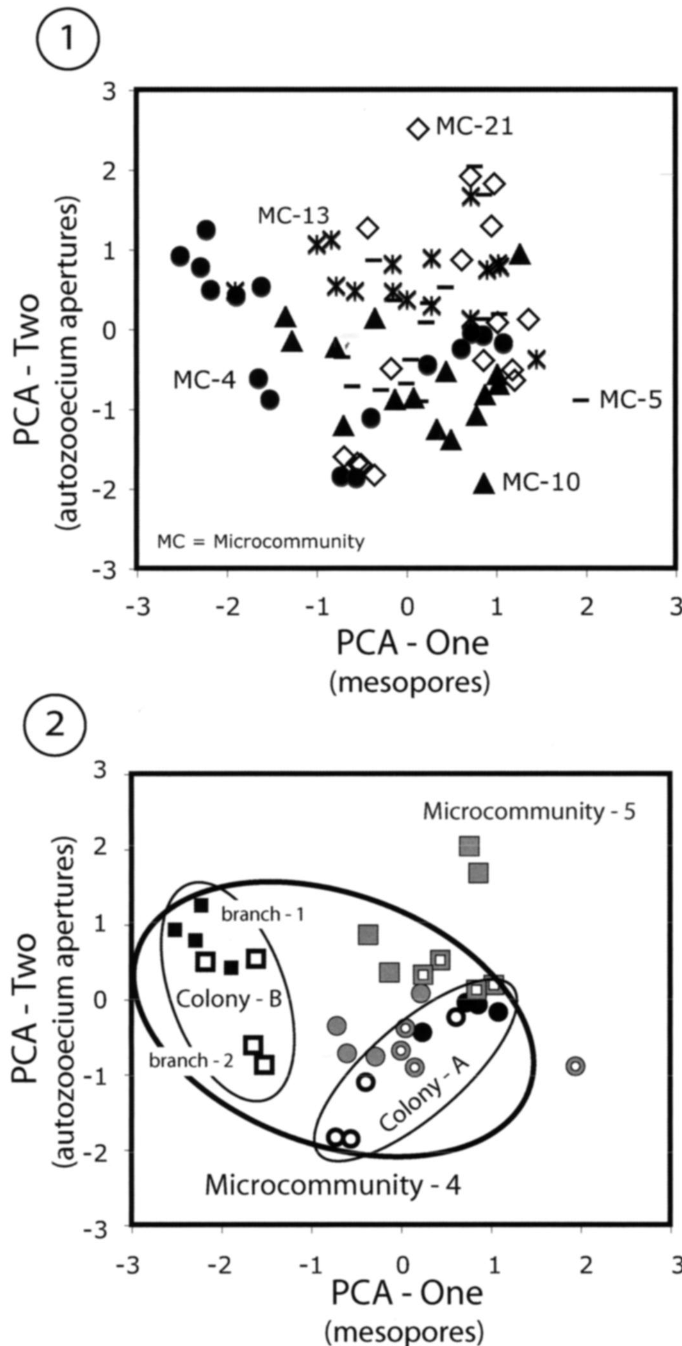

FIGURE 5—Data plotted on the first two axes of principal components, which account for 36.7% of the total variance. Each symbol represents a suite of 11 measured characters for each zooecia. *1,* Symbols represent microcommunities (Mc); circle = Mc-4; dash = Mc-5; triangle = Mc-10; diamond = Mc-21; and cross = Mc-13. *2,* Same principal component space as *1,* but with only microcommunities 4 and 5, indicated with black and gray symbols, respectively. Microcommunity 4 is highlighted by a large oval. Two colonies (A and B) of microcommunity 4 are outlined in smaller ovals. In each colony, four zooids measured from the same branch are highlighted with a white center. Four zooids measured from the other branch within the same colony have solid fills.

Variation among microcommunities (Fig. 6.3).—No significant difference was detected among microcommunities for any measured character (Table 2, Column 1A). Characters for which the factor *Microcommunity* accounted for more than 10% of the variation include: autozooecium area and wall thickness, autozooecium lateral, and diagonal spacing. Across all characters, the factor of *Microcommunity* accounted for, on average, 7.8% of the total variance. For the factor *Microcommunity* the null hypothesis (H_0: no significant difference exists among the group means for the factor *Microcommunity*) fails to be rejected for all characters observed at $P \leq 0.05$. Results from principal components analysis, below, support this conclusion.

Variation between colonies (Fig. 6.2).—Between colonies, within microcommunities, only two characters, mesopore area and spacing, were found to be significant using a *P*-value of $\leq$0.05 (Table 2, Column 2A). No characters between colonies were found to be highly significant (*P*-value $\leq$ 0.01). The nested factor of *Colony* accounted for more than 10% of the variation for all characters except autozooecium diameter, diagonal spacing, and diaphragm spacing. The nested factor of *Colony* accounted for 25%–50% of the variance for the following characters: mesopore area and spacing and endozone diameter (Table 2, Column 2B). For the factor of *Colony*, the null hypothesis (H_1: no significant difference exists between the group means for the factor *Colony* within *Microcommunity*) fails to be rejected for all characters observed, using a critical *P*-value of $\leq$ 0.01. The characters of mesopore area and spacing are significant for the factor of *Colony* at the $P \leq 0.05$ (Table 2, Column 2A). Results from principal components analysis, below, indicate that overall differences among colonies do exist.

Variation among branches (Fig. 6.1).—Between branches, within colonies, autozooecium wall thickness, diagonal and lateral spacing, and mesopore area and spacing are significant using a *P*-value of $\leq$0.05 (Table 2, Column 3A). Autozooecium area, diaphragm spacing and diameter, mesopore diaphragm spacing and wall thickness, and endozone wall thickness are each significant with a *P*-value of $\leq$0.001 (Table 2, Column 3A). The nested factor of *Branch* accounted for more than 10% of the variation for all characters (Table 2, Column 3B). The nested factor of *Branch* accounted for 25%–50% of the variance for the following characters: endozone wall thickness and mesopore diaphragm spacing and wall thickness (Table 2, Column 3B). The nested factor of *Branch* accounted for >50% of the variance for the following characters: autozooecium diameter, area, and diaphragm spacing. Across all characters, the factor of *Branch* accounted for on average 36.1% of the total variation. The null hypothesis (H_2: no significant difference exists between the group means for the factor *Branch* within *Colony*, nested within *Microcommunity*) is rejected at $P \leq 0.001$ for all but two characters (Table 2, Column 3A). Results from principal components analysis, below, support this conclusion.

Residual variance—The residual (error) is the variance not accounted for by the ANOVA model used in the study. On average, 36.8% of the variation found in this study was not accounted for in the model used (Table 2, Column 4). Residuals ranged from 22.1% for endozone diameter to 46.8% for autozooecium diaphragm spacing.

Principal component analysis.—Patterns highlighted in ANOVA results are evident in a plot of the first two principal components (Fig. 5), which account for only 36.7% of the total variance. Although some differences among the five microcommunities are evident, no systematic difference among means of microcommunities were detectable (Fig. 5.1). Principal component axis one (varimax rotation) is dominated by mesopore characters (Table 1, Column PCA-1). Axis two, however, is more

strongly associated with autozooecium aperture size (Table 1, Column PCA-2).

Visual inspection of Figure 5.2 suggests that overall differences do exist among the means of colonies within microcommunities (compare Microcommunity 4, Colony A vs. Colony B and Microcommunity 5, Colony A vs. B). In addition, differences exist between branches within the same colony (compare open vs. closed symbols for the same colony, Fig. 5.2). Principal component axis three is dominated by exozone characters and axis five is dominated by autozooecium spacing (Table 3).

DISCUSSION

Sources of morphological variation.—The distribution of variance observed in *Leioclema punctatum* can be summarized as follows.

1. There is significant morphological variation between branches, within single colonies of *L. punctatum* (Fig. 6.1).
2. Differences among colonies within microcommunities (Fig. 6.2) are minor for most characters, with the exception of the mesopore spacing and area (Table 2, Column 2A, bold). These differences do contribute to observable, overall variation in principal component analysis.
3. No significant differences were found among microcommunities of *L. punctatum* at this locality (Fig. 6.3).
4. Overall, as much variation occurs between branches within single colonies of *L. punctatum* as occurs between some colonies or among microcommunities.

These results lead to the question: Why is there systematic variation between branches on single colonies, but less systematic variation between colonies or among microcommunities (i.e., microenvironmental variation)? One possible explanation may be that variation between branches is expected because the growth of individual zooids is partially controlled by the microenvironment in which they develop through time (Boardman and Cheetham, 1973; Hageman, 1995). Adjacent zooids experience the same or more similar microenvironmental conditions during development than do zooids developing on other areas of the colony. This means that it is possible to have more variation between branches of a single colony than between branches in similar regions of different colonies (Hageman, 1995). Differences experienced by the colonies during development, such as nutrition levels, temperature, salinity, and predation, may have affected their phenotypic output. To test for this, systematic measurements of sections along branches could be obtained. It may be possible to correlate areas on different branches that grew at the same time based on similarities in morphometric analysis of different zones.

A second explanation for systematic variation among branches within colonies is that branches within colonies were affected in very small-scale spatial distributions by slight variations in their microenvironment. The growth of new branches may have slightly redirected water flow away or towards other branches. Some branches analyzed in this study may have grown nearer the host on which the colony was living than other branches within the

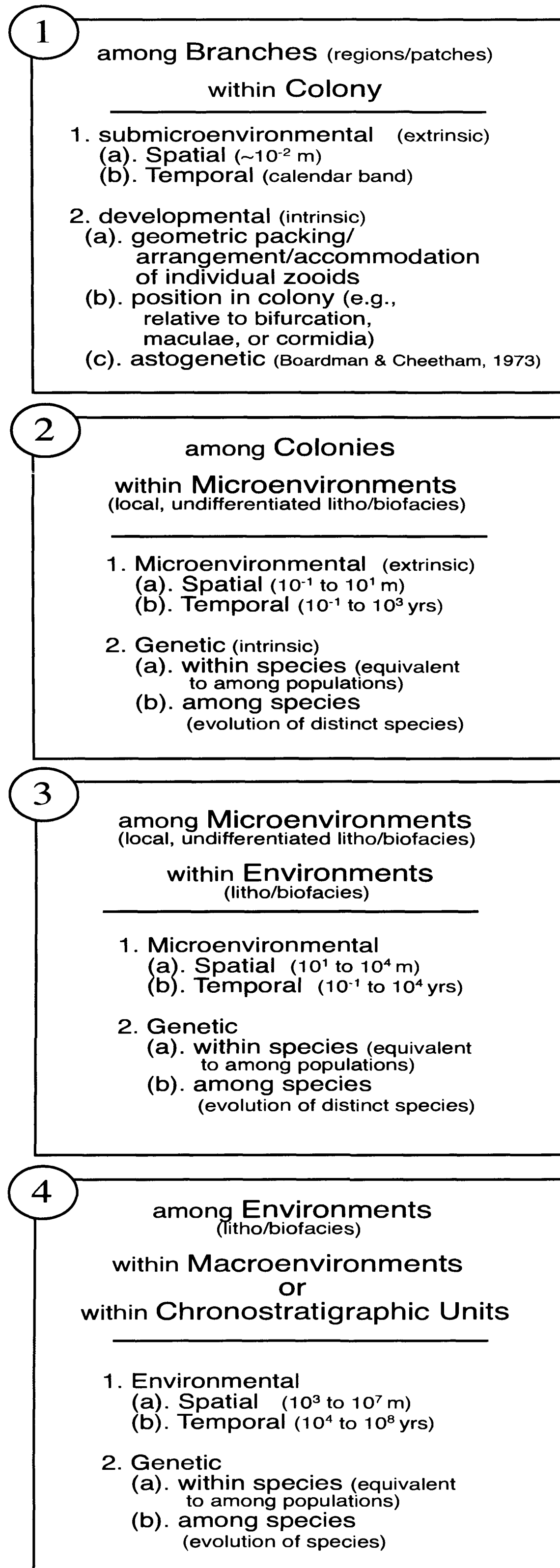

FIGURE 6—Potential sources (causes) of morphological variation at several hierarchical levels. Each level of environmental variation may be a result of either changing conditions through space (position within environment), or changing conditions through time, or both. *1,* Variation within a genetic clone (colony) may be due to very small-scale environmental variation or intrinsic controls on development of single genotype. *2,* Variation among colonies (very close in space and time) may be due to small-scale environmental heterogeneity in space and/or time, or genetic variation among colonies, the effects of which are observable (Hageman et al., 2001). *3,* Variation among groups of colonies (time correlative) may be due to small-scale environmental heterogeneity in space and/or time or genetic variation among colonies. *4,* Variation among larger scales of environmental hierarchies (space and/or time) can be envisioned, but were not tested within this study.

TABLE *3*—Summary loadings for the first five Principal components (varimax rotation). Bold values indicate the characters that are most correlated with each axis.

Character	PCA-1	PCA-2	PCA-3	PCA-4	PCA-5	Abbreviation
Mesopore Area	**0.7788**	−0.0044	−0.2144	−0.2254	0.2865	MeAr
Mesopore Spacing	**0.7705**	0.1045	0.2712	0.2055	0.0627	MeSp
Mesopore Wall Thickness	**0.7667**	−0.0406	−0.0401	0.2691	0.2693	MeWlTk
Autozooecium Diameter	−0.0648	**0.9562**	0.0014	−0.0221	0.1204	AuDi
Autozooecium Area	0.1292	**0.9295**	0.0534	−0.0294	0.1915	AuAr
Endozone Wall Thickness	−0.0793	−0.1090	**−0.8691**	0.1897	−0.0175	EnWlTk
Endozone Diameter	−0.1089	−0.1588	**0.5604**	0.1872	0.4947	EnDi
Mesopore Diaphragm Spacing	0.0457	−0.1129	−0.1593	**0.8559**	0.1035	MeDpSp
Autozooecium Wall Thickness	0.4144	0.1408	0.1098	**0.5505**	0.2488	AuWlTk
Autozooecium Lateral Spacing	0.2363	0.2033	0.0900	0.0680	**0.8193**	AuLtSp
Autozooecium Diagonal Spacing	0.1816	0.2008	0.0216	0.1537	**0.7511**	AuDgSp

Percent of total variance accounted for by each Principal component axis (cumulative): PCA-1 = 19.0 (19.0%), PCA-2 = 17.7 (36.7%), PCA-3 = 11.3 (48.0%), PCA-4 = 11.8 (59.8%), PCA-5 = 16.0 (75.8%).

same colony. If this host was a sponge or other filter-feeder, nutrient levels may have decreased for some branches, leaving different phenotypes. It is also possible that some branches benefited from closer proximity to the host, by intercepting food particles that the host attracted. To test for this, systematic measurements should be taken along branches within the colonies.

A third explanation for significant variation between branches within colonies is methodological error in data collection. If present, however, such error would increase overall variation. Additional variation would not be expected to be distributed systematically among factors (i.e., correlation of error with certain factors would not be expected).

Variation of specific characters.—Significant differences did exist between colonies within microcommunities for mesopore area and spacing. This suggests that these characters are affected more strongly than the others by the microenvironmental influences these specimens encountered. This pair of characters is correlated by their definition. The primary purpose of mesopores is inferred to be space-filling, that is, mesopores form secondarily around autozooids (the primary feeding structures). The spacing of the autozooids is optimized in order to provide adequate space for the food-gathering lophophores. Mesopores fill the space in between, shown here by increasing the area of the mesopore opening, which consumes less resources than filling the space with solid skeleton.

Residual variation in the model.—The residual is the amount of variation not accounted for in a statistical model (ANOVA). Residuals in this study range from 22.15% to 46.8%, which is typical for studies of morphological variation in Bryozoa (Hageman et al., 2001). A large amount of residual variance accounts for packing and nearest-neighbor effects within the colony (Hageman et al., 2001), and could be accounted for in an experimental design created to do so (nested ANOVA). The residual may also include microenvironmental and developmental factors not controlled for in the experimental design.

Broader significance.—Results from this study have significance for both paleoecological analysis at the microenvironmental scale and for studies of patterns and processes of microevolution and speciation. Previous studies have tested the hypothesis that in stable environments (minimal perturbation though ecological time) within-colony variation should be greater than among-colony variation (Farmer and Rowell, 1973; Schopf, 1976; Schopf and Dutton, 1976; Pachut and Anstey, 1979; Pachut, 1982; Key, 1987). This stems from the argument that phenotypic plasticity, induced by short-term environmental change and/or heterogeneity of microenvironments, will increase when an organism develops in unstable environments (Farmer and Rowell, 1973; Schopf, 1976). The argument has also been made that genetic polymorphism (taxonomic diversity) should increase with increased stability of the environment (Pachut, 1982). However, this increase may be more a consequence of proportional change than a change in absolute degree.

Applying the variation/stability model to this study (high within-colony variation for *L. punctatum* and low among-colony variation observed here) suggests an unstable environment for the shale facies of the Warsaw Formation. However, this conclusion is confounded by the observation of a low degree of variation among microcommunities. That is, there was more systematic variation between branches of single *L. punctatum* colonies (Fig. 6.2) than was observed among the groups of colonies that lived on separate soft-tissue hosts living meters and/or years apart. The magnitude of variation was greater among microcommunities than within branches (larger clouds in Fig. 5), but at the microcommunity level the variation was not systematic (microcommunities were not distinguished from each other). These results suggest that microenvironmental variation increases the overall morphologic variance, but does not differentiate the group means systematically (e.g., more scatter but not segregation in data points).

Results also support the assertion that morphometric data collected for taxonomic studies should be collected in a manner that samples broadly across a colony (Hageman et al., 2001). Concentration of a large number of measurements/observations, on a small portion of a colony (or colony fragment), will not represent the entire variation present within a phenotype (Fig. 5.2, cf. solid- vs. white-centered data points from the same colony). Results from this study also demonstrate that in studies of microenvironmental variation, multiple colonies should be measured in order to document environmental variation (Fig. 5.2, cf. colony 4-A vs. 4-B).

In this study, however, there does not appear to be an intermediate level of environmental variation between microcommunities and the assemblage from the same general environment (Fig. 5, little systematic variation among the five microcommunities). Thus, very small-scale (submicro-) environmental variation apparently plays a greater role in morphogenesis than environmental variation within the scale of a single environmental setting, i.e., undifferentiated litho/biofacies. These results are important for paleoecological, microevolutionary studies because they do not suggest the presence of major phenotypic variation at a level between an observable facies and above the directly observable intercolonial level. These results need to be tested further, but are consistent with those of Hageman (1994, 1995).

Extrapolation of results to solitary organisms.—Although results cannot be extrapolated directly, the following analogies to solitary organisms can be made. Submicroenvironmental effects

(Fig. 6.1) in a solitary organism are the equivalent of changes in environmental conditions during the ontogeny of an individual. This is the equivalent of identical twins (two members of a clone) living their entire lives in the same, otherwise undifferentiated environment, but one of which suffered a severe pathogen during its development. Different adult morphologies between the two members of the clone could be the end results.

Results from this study suggest that phenotypic influences at the submicroenvironmental (ontogenetic developmental) level play a more confounding role in the application of phenotype to taxonomic identification than undifferentiated (unrecognized) micro- to mesoenvironmental variation.

SUMMARY

1. Because the reproductive habits of fossil organisms cannot be observed directly, and genetic material is not typically preserved, species concepts for fossil organisms rely greatly upon hard-part skeletal morphology. Recognition of fossil species is complicated by uncertainties associated with the relative degree to which the formation of the skeletal phenotype is controlled by the organism's genotype and/or the environmental conditions in which it lived.
2. Detailed morphometric analysis of colonial fossil organisms, sampled over very short intervals in stratigraphic sections, can document microenvironmentally induced phenotypic variation in the fossil record. The trepostome bryozoan *Leioclema punctatum* was chosen for this study based on its abundance and distribution, and preservation of characters suited to morphometric analyses. Colony fragments of *Leioclema punctatum* were found within discrete piles. Multiple attachment bases were found in each of these piles suggesting that multiple, unique genotypes were represented by each pile, and that each pile represents a relatively short-lived microcommunity.
3. No significant differences occurred among microcommunities (meter scale and 10^2 year distributions). Mesopore area and mesopore spacing were significant between colonies at a *P*-value of ≤ 0.05. No characters were highly significant between colonies, within microcommunities. All 12 characters were significant between branches, within colonies.
4. More variation occurs within colonies of *L. punctatum* than among microcommunities. Therefore, in any study of morphologic variation (among species, populations, individuals) care must be taken so as not to misattribute the source of variation (i.e., appropriate level in the hierarchy).
5. Mesopore spacing and area are affected more strongly by microenvironmental influences than are other characters used in this study.
6. Very small-scale environmental variation, within and among colonies, plays a greater role in morphogenesis than environmental variability within a given environmental setting (e.g., undifferentiated litho/biofacies). Results therefore suggest that factors that control phenotypic variation are minimal at levels below an observable facies but above the directly observable variation among colonies (i.e., no systematic variation among groups of colonies within a local environment).

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REFERENCES

ABBOTT, M. 1973. Intra- and intercolony variation in populations of *Hippoporina neviani* (Bryozoa–Cheilostomata), p. 223–245. *In* R. S. Boardman, A. H. Cheetham, and W. A. Oliver Jr. (eds.), Animal Colonies: Development and Function Through Time. Dowden, Hutchinson and Ross, Stroudsburg.

ASTROVA, G. G. 1965. Morfologiya, istroriya razvitiya i sistema Ordovikskikh i Silurijskik mshanok. Akademi Nauk SSSR Paleontologicheski Institut, 106, 52 p.

BASSLER, R. S. 1953. Stenoporidae, p. G101–G105. *In* R. C. Moore (ed.), Treatise on Invertebrate Paleontology. Pt. G. Bryozoa. Geological Society of America and University of Kansas Press, Lawrence.

BOARDMAN, R. S., AND A. H. CHEETHAM. 1973. Degrees of colony dominance in stenolaemate and gymnolaemate Bryozoa, p. 121–220. *In* R. S. Boardman, A. H. Cheetham, and W. A. Oliver Jr. (eds.), Animal Colonies: Development and Function Through Time. Dowden, Hutchinson and Ross, Stroudsburg.

CHEETHAM, A. H., J. B. C. JACKSON, AND L. A. C. HAYEK. 1993. Quantitative genetics of bryozoan phenotypic evolution. I. Rate tests for random change versus selection in differentiation of living species. Evolution, 47:1526–1538.

CHEETHAM, A. H., J. B. C. JACKSON, AND L. A. C. HAYEK. 1994. Quantitative genetics of bryozoan phenotypic evolution. II. Analysis of selection and random change in fossil species using reconstructed genetic parameters. Evolution, 48:360–375.

CHEETHAM, A. H., J. B. C. JACKSON, AND L. A. C. HAYEK. 1995. Quantitative genetics of bryozoan phenotypic evolution. III. Phenotypic plasticity and the maintenance of genetic variation. Evolution, 49:290–296.

FARMER, J. D., AND A. J. ROWELL. 1973. Variation in the bryozoan *Fistulipora decora* (Moore and Dudley) from the Beil Limestone of Kansas, p. 377–394. *In* R. S. Boardman, A. H. Cheetham, and W. A. Oliver Jr. (eds.), Animal Colonies: Development and Function Through Time. Dowden, Hutchinson and Ross, Stroudsburg.

HAGEMAN, S. J. 1994. Microevolutionary implications of clinal variation in the Paleozoic bryozoan *Streblotrypa*. Lethaia, 27:209–222.

HAGEMAN, S. J. 1995. Observed phenotypic variation in a Paleozoic bryozoan. Paleobiology, 21:314–328.

HAGEMAN, S. J., M. BAYER, AND C. D. TODD. 2001. Partitioning phenotypic variation: Implications for morphometric analyses (Bryozoa), p. 131–140. *In* P. N. Wyse Jackson, C. Buttler, and M. Spencer Jones (eds.), Bryozoan Studies 2001. Swets and Zeitlinger, Lisse.

HAGEMAN, S. J., N. P. JAMES, AND Y. BONE. 2000. Cool-water carbonate production from epizoic bryozoans on ephemeral substrates. Palaios, 15:33–48.

HALL, J., 1858. Report on the Geological Survey of the State of Iowa: Embracing the Results of Investigations Made During Portions of the Years 1855, 56 & 57, Pt. 2, Paleontology. Iowa Geological Survey, Des Moines, 251 p.

HOLDENER, E. J., AND S. J. HAGEMAN. 1998. Implications of intracolonial variation in a Paleozoic bryozoan. Journal of Paleontology, 72:809–818.

KEY, M. M., JR., 1987. Partitioning of morphological variation across stability gradients in Upper Ordovician trepostomes, p. 145–152. *In* J. R. P. Ross (ed.), Bryozoa: Present and Past. Western Washington University, Bellingham.

LEVINTON, J. S. 2001. Genetics, Paleontology, and Macroevolution. Cambridge University Press, Cambridge, 617 p.

MAYR, E. 1963. Animal Species and Evolution. Belknap Press, Cambridge, Massachusetts, 797 p.

MCKINNEY, F. K., AND J. B. C. JACKSON. 1991. Bryozoan Evolution. The University of Chicago Press, Chicago, 238 p.

NYE, O. B., D. A. DEAN, AND R. S. HINDS. 1972. Improved thin section techniques for fossil and recent organisms. Journal of Paleontology, 46:271–275.

PACHUT, J. F. 1982. Morphologic variation within and among genotypes in two Devonian bryozoan species: An independent indicator of paleostability? Journal of Paleontology, 56:703–716.

PACHUT, J. F., AND R. L. ANSTEY. 1979. A developmental explanation of stability-diversity-variation hypotheses: Morphogenetic regulation in Ordovician bryozoan colonies. Paleobiology, 5:168–187.

SCHOPF, T. J. M. 1976. Environmental versus genetic causes of morphologic variability in bryozoan colonies from the deep sea. Paleobiology, 2:156–165.

SCHOPF, T. J. M., AND A. R. DUTTON. 1976. Parallel clines in morphologic and genetic differentiation in a coastal zone marine invertebrate. Paleobiology, 2:255–264.

SIMPSON, G. G. 1953. The Major Features of Evolution. Columbia University Press, New York, 434 p.

SNYDER, E. M. 1987. Bryozoan succession in the Warsaw Formation (Valmeyeran, Mississippian) of the Mississippi Valley, USA, p. 245–252. *In* J. R. P. Ross (ed.), Bryozoa: Present and Past. Western Washington University, Bellingham.

SNYDER, E. M. 1991. Revised taxonomic procedures and paleoecological applications for some North American Mississippian Fenestellidae and Polyporidae (Bryozoa). Palaeontographica Americana, Number 57, 275 p.

UITENBROEK, D. 2004. SISA online statistical analysis: Research and Statistical Consultancy. The Netherlands <http://home.clara.net/sisa>.

ULRICH, E. O. 1882. American Paleozoic Bryozoa. Journal of Cincinnati Society of Natural History, 5:121–175.

ULRICH, E. O. 1890. Paleozoic Bryozoa. Illinois Geological Survey, 1: 283–688.

WYSE JACKSON, P. N. 1996. Bryozoa from the Lower Carboniferous (Visean) of County Fermanagh, Ireland. Bulletin of the Natural History Museum, 52:119–171.

ZAR, J. H. 1999. Biostatistical Analysis (fourth edition). Prentice-Hall, Upper Saddle River, New Jersey, 929 p.

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APPENDIX—Summary of the means, standard deviations (stdev), and number of observations (n) per factor/level. Measurements are in mm. Abbreviations defined in Figure 1 and Table 2.

	EnDi			MeDpSp			EnWlTk			AuDgSp		
Factor	mean	stdev	n	mean	stdev	n	mean	stdev	n	mean	stdev	n
Microcommunity-1	1.242	0.325	16	0.061	0.020	16	0.0081	0.0028	16	0.317	0.067	16
Colony-A	1.429	0.349	8	0.075	0.009	8	0.0066	0.0013	8	0.357	0.069	8
Branch-1	1.187	0.093	4	0.068	0.005	4	0.0060	0.0006	4	0.308	0.031	4
Branch-2	1.670	0.346	4	0.083	0.005	4	0.0071	0.0016	4	0.407	0.059	4
Colony-B	1.055	0.158	8	0.047	0.018	8	0.0095	0.0032	8	0.278	0.037	8
Branch-1	0.922	0.072	4	0.051	0.013	4	0.0121	0.0022	4	0.256	0.033	4
Branch-2	1.189	0.073	4	0.042	0.024	4	0.0070	0.0012	4	0.299	0.030	4
Microcommunity-2	1.324	0.145	16	0.071	0.015	16	0.0108	0.0032	16	0.383	0.072	16
Colony-A	1.273	0.146	8	0.079	0.014	8	0.0132	0.0023	8	0.383	0.080	8
Branch-1	1.339	0.098	4	0.076	0.014	4	0.0114	0.0014	4	0.358	0.094	4
Branch-2	1.207	0.169	4	0.082	0.015	4	0.0151	0.0011	4	0.408	0.068	4
Colony-B	1.375	0.133	8	0.062	0.011	8	0.0084	0.0020	8	0.383	0.068	8
Branch-1	1.476	0.083	4	0.061	0.008	4	0.0090	0.0028	4	0.344	0.043	4
Branch-2	1.275	0.088	4	0.064	0.015	4	0.0079	0.0007	4	0.423	0.068	4
Microcommunity-3	1.512	0.484	16	0.057	0.015	16	0.0090	0.0022	16	0.266	0.070	16
Colony-A	1.892	0.406	8	0.056	0.016	8	0.0091	0.0024	8	0.319	0.051	8
Branch-1	2.170	0.407	4	0.046	0.004	4	0.0072	0.0011	4	0.361	0.032	4
Branch-2	1.615	0.115	4	0.066	0.018	4	0.0110	0.0015	4	0.277	0.021	4
Colony-B	1.133	0.092	8	0.058	0.015	8	0.0089	0.0021	8	0.213	0.039	8
Branch-1	1.105	0.066	4	0.047	0.013	4	0.0078	0.0014	4	0.180	0.009	4
Branch-2	1.161	0.116	4	0.068	0.007	4	0.0100	0.0023	4	0.246	0.025	4
Microcommunity-4	1.415	0.238	16	0.065	0.025	16	0.0086	0.0013	16	0.387	0.090	16
Colony-A	1.404	0.259	8	0.056	0.015	8	0.0085	0.0006	8	0.407	0.104	8
Branch-1	1.197	0.081	4	0.044	0.003	4	0.0084	0.0006	4	0.478	0.065	4
Branch-2	1.611	0.190	4	0.068	0.011	4	0.0086	0.0007	4	0.335	0.087	4
Colony-B	1.425	0.232	8	0.074	0.030	8	0.0088	0.0018	8	0.367	0.076	8
Branch-1	1.279	0.176	4	0.049	0.014	4	0.0086	0.0015	4	0.377	0.084	4
Branch-2	1.572	0.193	4	0.099	0.015	4	0.0090	0.0022	4	0.357	0.078	4
Microcommunity-5	1.560	0.110	16	0.064	0.017	16	0.0070	0.0024	16	0.425	0.051	16
Colony-A	1.510	0.083	8	0.057	0.015	8	0.0076	0.0030	8	0.442	0.061	8
Branch-1	1.503	0.073	4	0.064	0.015	4	0.0098	0.0028	4	0.430	0.077	4
Branch-2	1.517	0.103	4	0.049	0.012	4	0.0055	0.0012	4	0.455	0.048	4
Colony-B	1.610	0.115	8	0.071	0.016	8	0.0063	0.0014	8	0.408	0.036	8
Branch-1	1.709	0.039	4	0.069	0.013	4	0.0061	0.0011	4	0.384	0.036	4
Branch-2	1.512	0.062	4	0.073	0.021	4	0.0065	0.0018	4	0.433	0.011	4

	AuLtSp			AuDi			AuAr			MeSp		
	mean	stdev	n	mean	stdev	n	mean	stdev	n	mean	stdev	n
Microcommunity-1	0.532	0.171	16	0.148	0.028	16	0.0112	0.0032	16	0.070	0.025	16
Colony-A	0.634	0.161	8	0.128	0.017	8	0.0105	0.0030	8	0.092	0.014	8
Branch-1	0.618	0.237	4	0.137	0.001	4	0.0127	0.0014	4	0.093	0.018	4
Branch-2	0.651	0.059	4	0.119	0.020	4	0.0083	0.0023	4	0.091	0.013	4
Colony-B	0.430	0.115	8	0.168	0.022	8	0.0120	0.0035	8	0.047	0.007	8
Branch-1	0.377	0.045	4	0.176	0.011	4	0.0147	0.0014	4	0.046	0.005	4
Branch-2	0.483	0.147	4	0.159	0.028	4	0.0093	0.0025	4	0.049	0.008	4
Microcommunity-2	0.620	0.065	16	0.153	0.028	16	0.0140	0.0048	16	0.089	0.019	16
Colony-A	0.651	0.060	8	0.135	0.014	8	0.0103	0.0016	8	0.086	0.020	8
Branch-1	0.630	0.047	4	0.138	0.016	4	0.0110	0.0013	4	0.074	0.008	4
Branch-2	0.672	0.072	4	0.131	0.011	4	0.0095	0.0016	4	0.097	0.023	4
Colony-B	0.588	0.057	8	0.171	0.026	8	0.0177	0.0039	8	0.093	0.019	8
Branch-1	0.590	0.031	4	0.190	0.020	4	0.0206	0.0030	4	0.085	0.025	4
Branch-2	0.586	0.081	4	0.153	0.016	4	0.0149	0.0024	4	0.100	0.010	4
Microcommunity-3	0.427	0.138	16	0.132	0.028	16	0.0106	0.0036	16	0.090	0.017	16
Colony-A	0.532	0.115	8	0.142	0.028	8	0.0121	0.0036	8	0.083	0.016	8
Branch-1	0.624	0.067	4	0.151	0.021	4	0.0139	0.0026	4	0.075	0.015	4
Branch-2	0.440	0.061	4	0.132	0.034	4	0.0104	0.0038	4	0.092	0.012	4
Colony-B	0.321	0.045	8	0.123	0.027	8	0.0091	0.0032	8	0.096	0.017	8
Branch-1	0.294	0.030	4	0.109	0.009	4	0.0076	0.0010	4	0.099	0.015	4
Branch-2	0.348	0.043	4	0.137	0.032	4	0.0105	0.0042	4	0.094	0.021	4
Microcommunity-4	0.654	0.154	16	0.172	0.016	16	0.0167	0.0013	16	0.081	0.019	16
Colony-A	0.679	0.177	8	0.174	0.014	8	0.0162	0.0015	8	0.070	0.016	8
Branch-1	0.824	0.085	4	0.172	0.020	4	0.0161	0.0017	4	0.084	0.006	4
Branch-2	0.534	0.099	4	0.176	0.008	4	0.0164	0.0016	4	0.057	0.008	4
Colony-B	0.628	0.135	8	0.170	0.018	8	0.0172	0.0010	8	0.092	0.015	8
Branch-1	0.594	0.112	4	0.168	0.024	4	0.0175	0.0012	4	0.097	0.020	4
Branch-2	0.662	0.163	4	0.172	0.015	4	0.0170	0.0008	4	0.087	0.008	4
Microcommunity-5	0.638	0.088	16	0.155	0.045	16	0.0140	0.0058	16	0.099	0.017	16
Colony-A	0.663	0.068	8	0.162	0.033	8	0.0151	0.0044	8	0.102	0.013	8
Branch-1	0.611	0.038	4	0.140	0.008	4	0.0132	0.0018	4	0.097	0.011	4
Branch-2	0.716	0.042	4	0.183	0.035	4	0.0170	0.0057	4	0.107	0.014	4
Colony-B	0.613	0.104	8	0.148	0.057	8	0.0129	0.0071	8	0.096	0.021	8
Branch-1	0.566	0.043	4	0.095	0.002	4	0.0064	0.0006	4	0.081	0.007	4
Branch-2	0.660	0.132	4	0.202	0.007	4	0.0194	0.0024	4	0.110	0.021	4

APPENDIX—Extended.

Factor	MeAr			AuWlTk			MeWlTk		
	mean	stdev	n	mean	stdev	n	mean	stdev	n
Microcommunity-1	0.0015	0.0006	16	0.051	0.021	16	0.029	0.015	16
Colony-A	0.0020	0.0004	8	0.065	0.016	8	0.043	0.007	8
Branch-1	0.0022	0.0002	4	0.075	0.013	4	0.043	0.009	4
Branch-2	0.0017	0.0003	4	0.056	0.014	4	0.043	0.005	4
Colony-B	0.0010	0.0002	8	0.036	0.015	8	0.016	0.006	8
Branch-1	0.0010	0.0003	4	0.024	0.004	4	0.011	0.002	4
Branch-2	0.0010	0.0002	4	0.048	0.009	4	0.021	0.004	4
Microcommunity-2	0.0021	0.0003	16	0.078	0.014	16	0.043	0.009	16
Colony-A	0.0020	0.0003	8	0.084	0.013	8	0.043	0.012	8
Branch-1	0.0020	0.0002	4	0.083	0.011	4	0.036	0.008	4
Branch-2	0.0021	0.0004	4	0.085	0.016	4	0.051	0.012	4
Colony-B	0.0023	0.0003	8	0.072	0.014	8	0.043	0.005	8
Branch-1	0.0023	0.0003	4	0.062	0.010	4	0.045	0.004	4
Branch-2	0.0022	0.0004	4	0.081	0.010	4	0.041	0.005	4
Microcommunity-3	0.0019	0.0004	16	0.058	0.011	16	0.040	0.011	16
Colony-A	0.0019	0.0005	8	0.058	0.007	8	0.037	0.006	8
Branch-1	0.0015	0.0003	4	0.059	0.009	4	0.037	0.008	4
Branch-2	0.0023	0.0002	4	0.057	0.006	4	0.037	0.004	4
Colony-B	0.0019	0.0002	8	0.058	0.015	8	0.043	0.014	8
Branch-1	0.0018	0.0003	4	0.053	0.004	4	0.031	0.011	4
Branch-2	0.0020	0.0001	4	0.064	0.021	4	0.055	0.002	4
Microcommunity-4	0.0021	0.0006	16	0.079	0.016	16	0.042	0.011	16
Colony-A	0.0020	0.0006	8	0.075	0.018	8	0.044	0.012	8
Branch-1	0.0024	0.0005	4	0.072	0.009	4	0.053	0.005	4
Branch-2	0.0017	0.0006	4	0.077	0.026	4	0.035	0.009	4
Colony-B	0.0022	0.0006	8	0.084	0.014	8	0.041	0.010	8
Branch-1	0.0024	0.0007	4	0.080	0.018	4	0.042	0.013	4
Branch-2	0.0019	0.0003	4	0.087	0.009	4	0.039	0.005	4
Microcommunity-5	0.0020	0.0007	16	0.077	0.018	16	0.047	0.010	16
Colony-A	0.0025	0.0006	8	0.067	0.018	8	0.046	0.011	8
Branch-1	0.0023	0.0004	4	0.073	0.020	4	0.051	0.012	4
Branch-2	0.0026	0.0007	4	0.061	0.017	4	0.040	0.007	4
Colony-B	0.0014	0.0003	8	0.086	0.013	8	0.048	0.010	8
Branch-1	0.0014	0.0001	4	0.075	0.006	4	0.039	0.004	4
Branch-2	0.0015	0.0004	4	0.097	0.007	4	0.056	0.005	4