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Determinate Growth and Modularity in a Gorgonian Octocoral

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Abstract. Growth rates of branches of colonies of the gorgonian *Pseudopterogorgia elisabethae* were monitored for 2 years on a reef at San Salvador Island, Bahamas. Images of 261 colonies were made at 6-month intervals and colony and branch growth analyzed. Branch growth rates differed between colonies and between the time intervals in which the measurements were made. Colonies developed a plumelike morphology through a pattern of branch origination and determinate growth in which branch growth rates were greatest at the time the branch originated and branches seldom grew beyond a length of 8 cm. A small number of branches had greater growth rates, did not stop growing, and were sites for the origination of subsequent "generations" of branches. The rate of branch origination decreased with each generation of branching, and branch growth rates were lower on larger colonies, leading to determinate colony growth. Although colonial invertebrates like *P. elisabethae* grow through the addition of polyps, branches behave as modules with determinate growth. Colony form and size is generated by the iterative addition of branches.

Introduction

Taxa ranging from algae to higher plants, and from cnidarians to protochordates, grow through the iterated replication of individual modules to form large, integrated individuals or colonies (Jackson *et al.,* 1985). Such modular organisms are ubiquitous and are often dominant members of a wide variety of animal and plant communities (Jackson, 1977; Hughes, 1989). Although the concept of modularity is applicable to many groups, the basic unit of replication, the module, is sometimes difficult to identify. This is perhaps best illustrated in plants where meristems, leaves, branches, and leaf-branch systems can each be characterized as the module from which the individual is constructed (Harper and White, 1974). Many of the same difficulties exist in the study of colonial invertebrates that grow through the iterative replication of polyps and zooids. The polyps of a coral—or the zooids of a tunicate—are undoubtedly "modules," but in many taxa these modules are then organized into structures that are themselves iteratively replicated, such as the branches of gorgonians or the inhalant-exhalent systems of colonial tunicates. Colony growth and development, known as astogeny, can be characterized by the replication of these larger units as readily as by the demography of polyps and zooids (Lasker and Sánchez, 2002; Rinkevich, 2002). Although the history of colony growth can be portrayed using either polyps or branches, it is unclear which level of organization should be studied to understand the processes controlling colony size and form.

The construction of colonies from modules is closely associated with indeterminate growth, that is, continuous growth throughout an organism's lifetime. Indeed, modular organisms are known to live for centuries and to attain sizes of tens of meters. The continuous production and addition of individuals leads to colonies that are remarkably variable in size and form. This mode of growth is central to a colony's ability to survive and recover from both natural and anthropogenic disturbances (Done, 1987, 1988). However, modular growth does not necessarily lead to indeterminate growth. Many species can be characterized by a distinct colony form and a maximum colony size. The attribution of

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a definable colony form and maximum size suggests that growth of the modules is in some fashion constrained, which suggests determinate growth.

The focus of the current study is to identify the pattern of colony and branch growth among colonies of the Caribbean gorgonian octocoral *Pseudopterogorgia elisabethae*. We show that branches on these colonies behave as modules, with a developmental sequence that leads to side branches of similar length throughout the colony. We show how both branches and entire colonies of *Pseudopterogorgia elisabethae* exhibit determinate growth.

Materials and Methods

A colony of *Pseudopterogorgia elisabethae* starts developing when a planula larva settles and metamorphoses into a polyp. A single, vertical branch is produced as polyps are added alternately on both sides of the tip, thereby extending the branch (Fig. 1A). Formation of new branches occurs several centimeters below the branch tip (Figs. 1B and 2A). This pattern is apparent in Figure 2A, in which branches 32 through 35 developed on branch 1 between December 1998 and July 1999. Each new branch then grows in the same fashion as the original branch, and some give rise to additional branches (Fig. 1). This pattern of side branches themselves becoming sources of new side branches is illustrated by the growth of branches 2 and 5 in Figure 2A.

This mode of branching and growth in *P. elisabethae* leads, eventually, to plume-like colonies that characteristically have a maximum height of less than 1 m (Fig. 2B). The pattern of branching creates a hierarchy among the branches, and we characterize branch order using an ordering system that we term generational ordering (Lasker and Sánchez, 2002; Sánchez, 2002; Sánchez *et al.*, 2003), in which branches are ranked on the basis of the number of branching events away from the original, primary branch (Fig. 1; Lasker and Sánchez, 2002). The original branch has a rank of 1; each branch that it produces is assigned a rank of 2; branches that grow off of the secondary branches are assigned a rank of 3; and so on. Each branch tip is assigned a rank, and the rank of any single branch remains constant. While having some similarities to ordering systems previously adapted to colonies (*i.e.*, Brazeau and Lasker, 1988), generational ordering has very different properties (Sánchez *et al.*, 2003). We also make a distinction between branches that give rise to additional branches, termed mother branches, and those from which no additional branches originate, termed daughter branches. Since there are no other morphological differences between mother and daughter branches, the distinction is based solely on the presence of side branches. The terminology is homologous to "sources and tributaries" used previously (Brazeau and Lasker, 1988; Lasker and Sánchez, 2002), but has the advantage of portraying the temporal relationships among the branches (Sánchez, 2002).

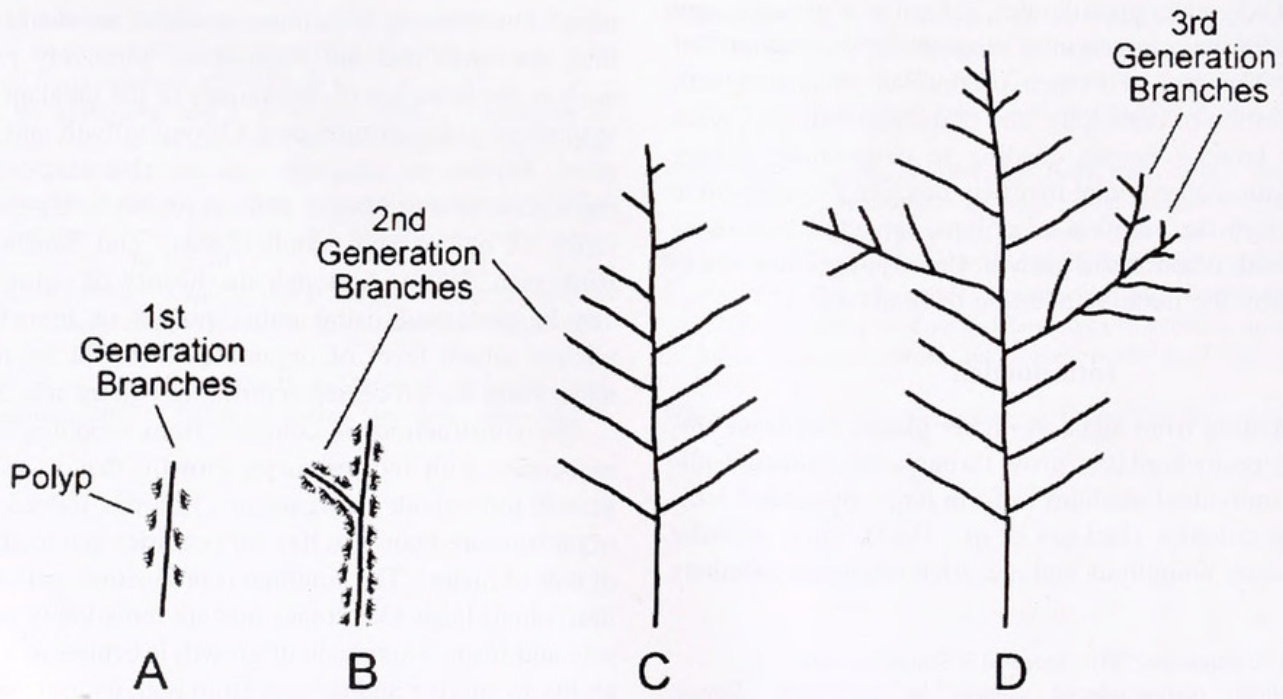

Figure 1. Colony development of *Pseudopterogorgia elisabethae*. (A) Initial development and subsequent branch elongation occurs through the addition of polyps at the branch tip. (B) The primary, "first generation" branch generates a side branch subapically (the second generation). (C) The colony continues to add a second generation of branches as the primary branch grows. (D) Two of the second-generation branches give rise to a third generation of branches. Polyps are no longer depicted in C and D. (After fig. 5, Lasker and Sánchez, 2002).

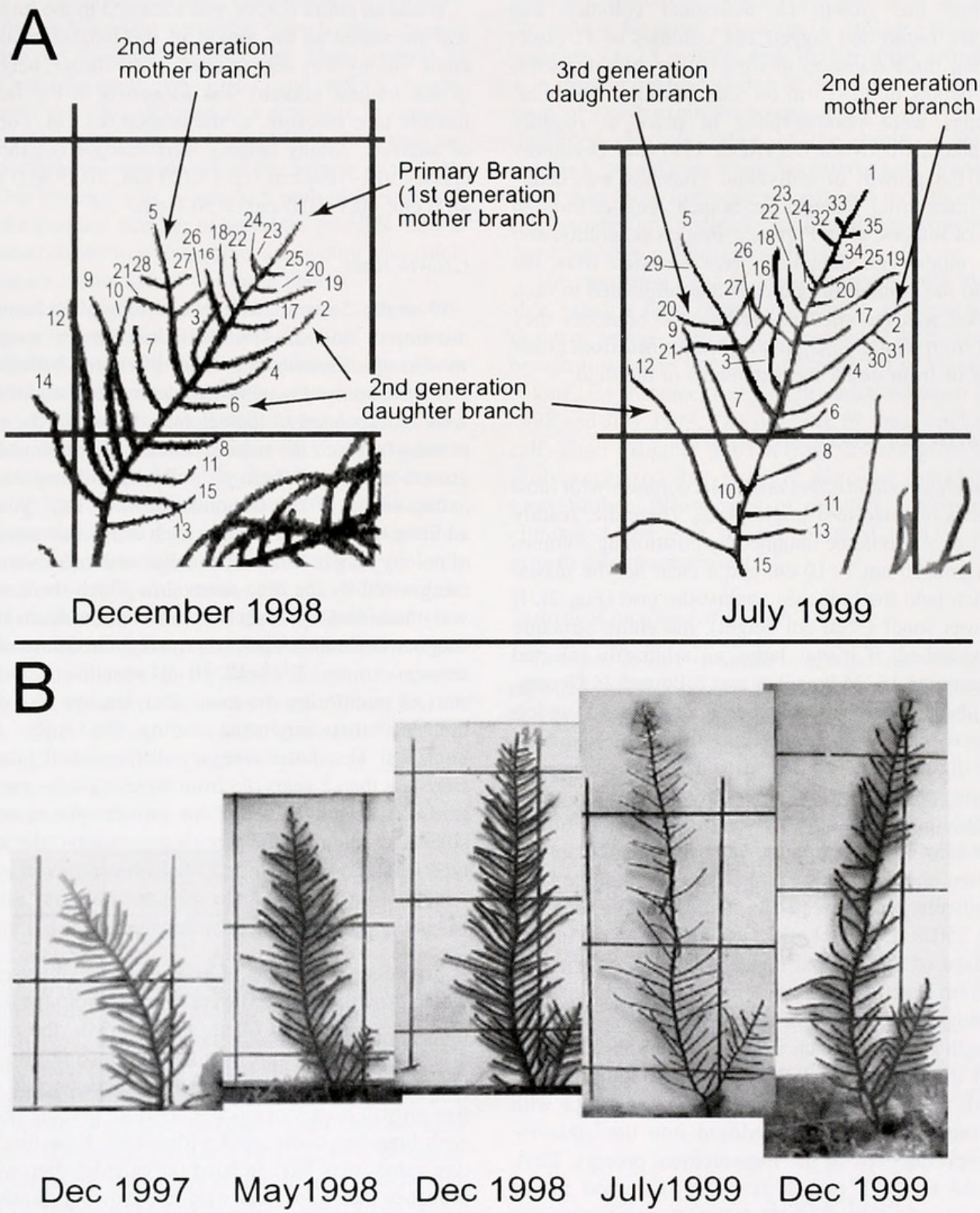

Figure 2. (A) Close-up photographs of a *Pseudopterogorgia elisabethae* branch in December 1998 and July 1999 showing branch growth and the system for numbering branches, which enabled successive measurements. Representative branches are labeled on the basis of type (mother, daughter) and order. Note that only the most distal (*i.e.*, youngest) daughter branches grew during the interval between photos, and that mother branches continued to extend and to branch regardless of position. Branches 30–35 originated, and 10 and 15 died, during the interval between photographs. Grid lines in the photos are 10 cm apart. See text for explanation of branch order. (B) Photographs of a *P. elisabethae* colony from San Salvador, Bahamas, taken over a 2-year period. All of the photographs have been normalized to the same scale, and the grid is 10 × 10 cm.

To follow the growth of individual colonies and branches, we found and tagged 261 colonies of *P. elisabethae* along three transects totaling 70 m^2 of substratum located at depths of 12–15 m on San Salvador, Bahamas. The colonies were photographed in place at roughly 6-month intervals between December 1997 and December 1999; and the growth of individual branches was determined by measuring changes in branch lengths through sequences of images, as in Figure 2. Branch generation and type (*i.e.,* mother *vs.* daughter) were assessed from the images, and the number of branches that originated in each time interval was recorded as well as the branches they developed from. Colony height was determined from either the images or from direct measurements in the field.

Image analysis and branch measurements

Pseudopterogorgia elisabethae forms colonies with most side branches oriented in a single plane. Therefore, readily measured images could be obtained by positioning colonies between a grid 10 cm × 10 cm and a clear acrylic plastic cover, which held the branches against the grid (Fig. 2). If a colony was small (<20 cm height), the entire structure was photographed; if it was large, an arbitrarily selected branch containing 15–25 branches was followed. In December 1997, photographs were taken with a Nikonos V underwater camera and Kodachrome 200 film. Those images were later digitized and converted to TIFF. In subsequent observations, photographs were taken at a resolution of 640 × 480 bits with a Sony Mavica digital camera (either MVC-7 or MVC-83) in an underwater housing. Distortion created when photographs were shot at a slight angle from the perpendicular was corrected in Photoshop (Version 4.0, Adobe). A 250 × 250 pixel grid was overlaid on the image, and the shape of the original image was adjusted with the free transform function of the program until the 10-cm grid in the photograph matched the 250-pixel grid.

The length of each branch in the photograph was measured with the program SCION (Scion Corporation, Frederick, MD). Although the program measures distance with high accuracy, variation is introduced into the measurements by several steps in the measurement process. First, although the tip of a branch is easily discerned in the images, it was also necessary to define its point of origin. We chose, as the point, the intersection of the branch with the line running along the middle of its mother branch, and that point had to be identified in each image. Second, the branches are curvilinear structures and were measured as segmented lines. Small differences in measurements are created by variation in the number and placement of those line segments. To assess the magnitude of measurement variation, three different observers measured each of 130 branches. The between-measurement standard deviation was 0.3 cm.

When an entire colony was included in the image, height was measured as the length of the longest branch. If the entire colony was not included in the image, height (length of the longest branch) was measured in the field with a flexible tape measure, to the nearest 0.1 cm. For purposes of analysis, colony heights were categorized into six size classes: 0.1–10.0 cm, 10.1–20.0 cm, 20.1–30.0 cm, 30.1–40.0 cm, 40.1–50.0 cm, >50.1 cm.

Growth rates

Over the 2 years, 261 colonies with 5870 branches were monitored, and 23,478 individual length measurements were made. Growth rate—the difference between successive measurements—was then determined, and those values were extrapolated to annual rates based on the number of months between the measurements. Measurement error for growth rates was 1.2 cm y^{-1}, which combines the effects of extrapolation of the 6-month intervals to 1 year and the additive effect of variance in each of the two measurements of colony length. The individual growth measurements were categorized by the time interval in which the measurement was made and by branch age, based on when the branch originated. Branch ages were categorized into one of five classes: <6, 6–12, 13–18, 19–24 months, or present at the start of monitoring. In some analyses, we also designated branches that originated during the study as "new" branches. This latter category distinguished branches that were less than 2 years old from those branches present at the start of the study. Each of the growth rates was also classified according to the branch's generation order and branch type (mother or daughter).

Negative growth rates

To reduce the effects of grazing on the analyses, cases in which growth was <0.0 were dropped from the data set. By rejecting these cases of negative growth, the most severe effects of grazing were eliminated. Grazing also may have reduced the observed growth of some branches with positive growth rates, but since scars from grazing heal rapidly, such branches could not be identified. Rejecting the negative values may have inflated the calculated growth rates of branches that were otherwise not growing. Since the measurement error was 1.2 cm y^{-1}, some branches that had not grown would, through measurement error alone, have small negative growth rates, and some would have small positive growth rates. Exclusion of branches with growth less than 0.0 cm y^{-1} would have eliminated the underestimates of the zero growth branches but not the overestimates.

Statistical analyses

Growth rates were compared by analysis of variance (ANOVA, functions UNIANOVA and MANOVA, SPSS

version 10.1). In those analyses, branches were classified using the five independent variables: branch order, branch type (mother or daughter), branch age, time interval in which the measurement was made, and colony height. Branch length was also included as a covariate in some of the analyses. Due to the size and complexity of the data set, it was impossible to examine all of the effects in a single analysis. Our strategy was to conduct multiple tests, each including the greatest number of variables possible, and to use Bonferroni corrections to significance testing when multiple tests were conducted on the same data.

Between-colony variation and time interval effects. The same branches were measured multiple times, so a repeated-measures ANOVA was the most appropriate design. However, because of the large number of branches nested within colonies, an ANOVA that included all five categorical variables could not be computed within a single repeated-measures design. We therefore conducted a repeated-measures ANOVA that tested for the random effects of branches within colonies and time interval (the 6-month interval in which the measurement was made). The effects of inter-colony variation were again examined in an ANOVA of the growth of branches that were less than 6 months old (*i.e.*, growth during the time interval in which the branch had originated). Growth rates in this analysis were compared with respect to colony and time interval (Table 1A). A simple two-way ANOVA was used for this analysis, as data from no single branch was included in more than one observation in this analysis.

Between-colony variation and branch age. Next, a series of analyses that simultaneously considered colony and age of the branch were conducted (Table 1B through 1E). The analysis was repeated for each of the four time intervals, which again led to a single branch being used only once in each of the analyses.

Multi-way ANOVA. Finally, a multi-way ANOVA that included all of the positive growth rates was conducted using branch type, branch order, branch age, and colony height as the independent variables (Table 2).

All of the ANOVA results are reported for untransformed data. Growth rates were heteroscedastic even after a variety of transforms (Levene's, Bartlett's or F_{max} tests of homogeneity of variances, depending on the structure of the data set). ANOVA results were almost always concordant with a parallel ANOVA using the rank transformed data and with nonparametric analyses, which could only consider a single factor at a time.

Results

General observations

Growth of *Pseudopterogorgia elisabethae* colonies was not indeterminate. Branches exhibited a distinct developmental cycle in which they arose, grew rapidly, and then grew at dramatically slower rates. This is evident in Figure 2A, where the rapid extension of daughter branches (*e.g.*, branches 22, 23, and 24) near the tip of the primary branch is in marked contrast to that of branches closer to the base, many of which exhibited no apparent growth (11, 13, 14). A simplified characterization of the results is that mother branches grew faster than daughters, new branches grew faster than those that were more than 6 months old (Fig. 3), and branches on small colonies had higher growth rates than those on large colonies (Fig. 4A).

The measured rates of branch growth were highly variable, ranging from negative values to 17.8 cm y^{-1}. Large negative values were often associated with almost total branch loss, as in the case of branch 10 in Figure 2A; this colony, for instance, lost two branches between December 1998 and July 1999. The presence of some uncharacteristically short daughter branches that did not grow during the study suggested that severely damaged branches did not grow further. When cases of negative growth rates were eliminated, the data set was reduced to 8341 individual growth rate measurements.

Statistical analyses of branch growth

Between-colony variation and time-interval effects. The repeated-measures analysis of variance was restricted to the 608 branches for which measurements were available from

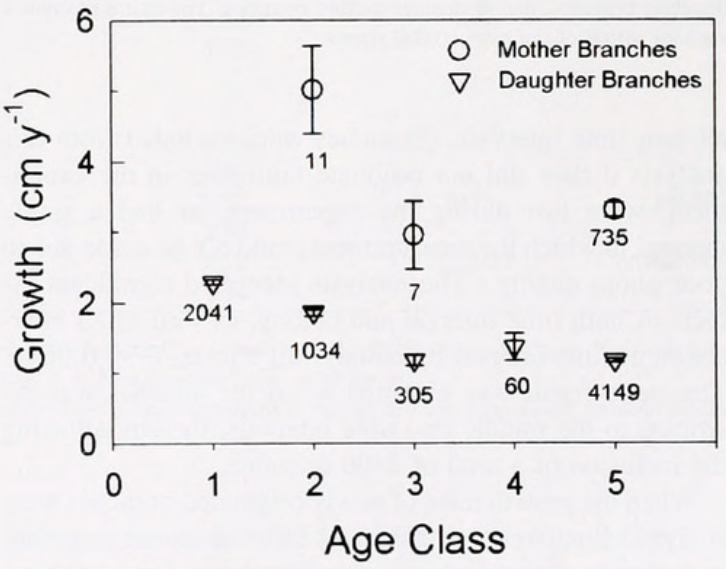

Figure 3. Branch growth rates for *Pseudopterogorgia elisabethae* colonies from San Salvador, Bahamas (mean ± standard error) plotted as a function of branch age class. Branches in age classes 1 through 4 originated during the course of the monitoring: class 1, 6 months; 2, 6–12 months; 3, 12–18 months; 4, 18–24 months. Branches in class 5 were present at the start of the 2-year monitoring program. Since mother and daughter branches cannot be distinguished when they originate, there are no age-class 1 (<6 months old) mother branches. The data set also did not include any age-class 4 (18–24 months old) mother branches. Only non-negative growth rates were used in the analysis, and the number of measurements for each class is indicated above or below the error bar.

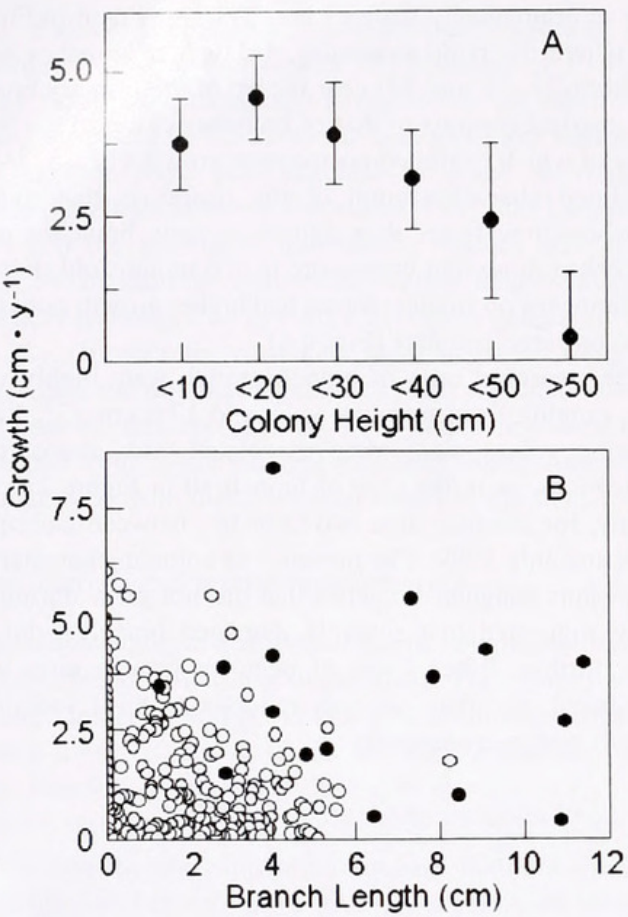

Figure 4. Growth rates of *Pseudopterogorgia elisabethae* branches from San Salvador, Bahamas, as a function of colony height (A) and branch length (B). Values in (A) are means ± standard error. (B) ○ denotes daughter branches, and ● denotes mother branches. The figure presents a random subset of the over 10,000 points.

all four time intervals. (Branches were excluded from this analysis if they did not originate until later in the experiment, were lost during the experiment, or had a single interval in which the measurement could not be made due to poor photo quality.) The analysis identified significant effects of both time interval and colony, as well as an interaction of time interval by colony (all effects, $P < 0.001$). The same result was obtained when the analysis was restricted to the middle two time intervals, thereby allowing the inclusion of a total of 2496 branches.

When the growth rates of newly originated branches were analyzed separately, growth rates differed among colonies, and there was also an interaction between colony and time interval, but no significant effect of time interval alone (Table 1A). These analyses indicate that the growth rates differed between colonies, and that the magnitude of differences between colonies varied between time intervals, but there was no simple additive difference in variance based on time interval alone. For instance, one colony may have had its highest growth rates in one time interval, while a different colony had its lowest growth in the same time interval.

Between-colony variation and branch age. Despite the effects of colony and time interval on growth, significant fixed effects were detected when the data were partitioned into subsets based on branch type. Separate analyses of the growth of daughter branches during each of the time intervals (Table 1, B–E) identified a significant effect of branch age in three of the four time periods (C–E). There were significant interactions between colony and branch age in all four intervals, and significant colony effects in two of the time intervals (Table 1, D and E).

Multi-way ANOVA. The effects of branch type, generation, age, and colony height were compared in this large analysis (Table 2). Branch growth rates were significantly affected by branch type (growth rates of mother branches > daughter branches, Fig. 3), branch age (younger > older, Fig. 3), and colony height (short > tall, Fig. 4A). In addition, there were significant interactions between branch type and age, branch type and colony height, and in the three-way interaction between branch type, order, and colony height. The significant interactions reflect the nonlinearity of the patterns seen in Figures 3 and 4; that is, the effects of the different factors were not additive.

Variation between colonies was differentiated in these analyses due to the computational limits of incorporating colonies as a fourth independent variable with 260 degrees of freedom (*i.e.*, 261 colonies). Branches from the different colonies were represented in almost all of the combinations of branch type and age, which should have reduced the confounding effects of not incorporating colony identity as an independent variable. Time interval was not tested in these analyses because it is confounded with the age of the branches (*i.e.*, the later intervals for a given branch also record the growth of an older branch). Generation, which was marginally not significant, was confounded with branch type because first-generation branches are by definition mother branches. When daughter and mother branches were analyzed separately in an analysis that was otherwise identical to that in Table 2, generation was not a significant factor ($P = 0.52$ and 0.40, respectively).

The effect of branch age on growth rates of the new branches is underestimated in Figure 3 because it assumes that new branches grew over a 6-month period. If we assume that branches originated continuously over the 6-month interval, then the average age of a new branch would be 3 months, and the mean growth rate would be twice that reported in Figure 3. Furthermore, as noted in Materials and Methods, excluding negative values from the analysis has the effect of slightly inflating growth estimates when the true value is near zero. When cases of negative growth were included, older (>12 mo) daughter branches had growth rates close to zero.

With branch length as a covariate, growth rates of daughter branches were analyzed separately for the effects of branch age. Both increasing age and increasing branch length (Fig. 4B) had significant negative effects on branch

Table 1

Analysis of variance tables testing colony and temporal variation in growth rates of Pseudopterogorgia elisabethae *at San Salvador, Bahamas*

Source of variation		df	Mean square	*F*	*P*[1]
A. Growth of newly originated branches during their first 6 months over four successive time intervals					
COLONY		166.0	5.27	1.52	**0.002**
	Error	207.4	3.46		
TIME INTERVAL		3.0	6.25	2.11	0.099
	Error	343.3	2.96		
COLONY × TIME INTERVAL		159.0	3.87	1.93	**0.000**
	Error	1702.0	2.00		
B. Growth of daughter branches as a function of branch age Dec 1997–June 1998					
COLONY		62.0	7.76	1.59	0.075
	Error	33.0	4.88		
AGE[2]		1.0	7.55	2.29	0.135
	Error	68.9	3.30		
COLONY × AGE		39.0	4.34	2.27	**<0.001**
	Error	790.0	1.91		
C. Growth of daughter branches as a function of branch age June 1998–Dec 1998					
COLONY		125.0	5.49	1.48	0.015
	Error	120.3	3.71		
AGE		2.0	34.82	14.16	**<0.001**
	Error	193.0	2.46		
COLONY × AGE		121.0	3.68	3.36	**<0.001**
	Error	2131.0	1.10		
D. Growth of daughter branches as a function of branch age Dec 1998–July 1999					
COLONY		153.0	5.48	2.12	**<0.001**
	Error	231.7	2.58		
AGE		3.0	85.86	40.23	**<0.001**
	Error	435.8	2.13		
COLONY × AGE		199.0	2.75	1.87	**<0.001**
	Error	2248.0	1.47		
E. Growth of daughter branches as a function of branch age July 1999–Dec 2000					
COLONY		122.0	6.08	2.13	**<0.001**
	Error	258.6	2.86		
AGE		4.0	26.77	10.73	**<0.001**
	Error	373.9	2.49		
COLONY × AGE		195.0	3.28	2.10	**<0.001**
	Error	1303.0	1.56		

[1] Significant *P* values listed in bold. Since some of the same branches are tested in each of the time intervals (B–E), Bonferroni correction of significance levels would suggest that $P = 0.0125$ be used as the highest significant *P* value. *F* = Mean square/Error mean square.

[2] Age classes were 0–6, 7–12, 13–18, 19–24, and >24 months. The number of classes present increased as the experiment progressed, those branches aged, and new branches originated.

growth rates. However, the two variables are confounded, and cases of long, young branches do not exist, so determining whether age or branch size is the functional factor is difficult.

Patterns of branch and colony growth

Mother-daughter comparisons. Mother branches were not identified until they had produced a side branch, but their growth rates suggest that their growth behavior changes before their daughter branches are produced. A retrospective analysis of branches that eventually became mother branches indicates that they had higher growth rates than daughter branches 1 year prior to the start of branching (Fig. 5). During the 6 months in which branching was first observed, these branches had the greatest growth rates of any of the groups of branches that we distinguished. In contrast to the mother branches, daughter branches exhibited decreasing growth rates as they aged and elongated (Fig. 5). After a year of growth, rates were near 1 cm y^{-1}, and when negative growth is incorporated into the analysis, the rates were not significantly different from zero. The

Table 2

Analysis of variance table of branch growth rates as a function of branch and colony characteristics

Source of variation	df	Mean square	*F*	*P*
Branch age	**4**	**11.95**	**5.06**	**0.000**
Generation	3	6.05	2.56	0.053
Branch type	**1**	**68.44**	**29.00**	**0.000**
Colony height	**5**	**13.14**	**5.57**	**0.000**
Branch age × Generation	7	2.26	0.96	0.459
Branch age × Branch type	**4**	**16.14**	**6.84**	**0.000**
Order × Branch type	2	7.01	2.97	0.051
Branch age × Generation × Branch type	3	2.59	1.10	0.349
Branch age × Colony height	18	1.42	0.60	0.902
Generation × Colony height	**15**	**5.96**	**2.52**	**0.001**
Branch age × Generation × Colony height	21	3.25	1.38	0.117
Branch type × Colony height	**5**	**6.16**	**2.61**	**0.023**
Branch age × Branch type × Colony height	5	2.43	1.03	0.399
Generation × Branch type × Colony height	**5**	**7.45**	**3.16**	**0.008**
Error	8236	2.36		
Total	8335			

Significant *P* values listed in bold. *F* = Mean square/Error Mean Square.

different growth trajectories for the two branch types, perhaps from their origination, also suggests that the estimated growth rates of daughter branches may have been inflated by the behavior of mother branches that had not started branching and were thus misidentified.

The existence of a determinate developmental sequence in the growth of daughter branches is again suggested by the distribution of branch sizes observed at San Salvador. If branch growth were indeterminate, then continuing growth of branches on a colony would have led to a steady accumulation of ever-larger branches, as well as to the presence of many branches with intermediate branch length. Most branches ceased growing when they reached a length of 5–8 cm (Figs. 4B and 6B). With time, colonies added height and new branches at their distal end (Fig. 2B), but colonies did not widen continuously at their base. The development of a plumelike morphology requires that the side elements of the plume show determinate growth. The few branches that became larger (Fig. 6B) were mother branches.

Colony size. Branch formation and extension control both the form and size of the colony, and colony size was also subject to determinate growth. Branching did not occur indefinitely among *P. elisabethae* branches. Colonies almost never had more than fourth-order branches (we have observed only one colony with fifth-order branches), and the number of branches produced by mother branches decreased with branch order. Branch origination rates were 4.7 (standard error = 0.2) new branches for each 6-month monitoring period on first-order branches, and 3.2 (0.2) and 2.4 (0.5) on second- and third-order branches, respectively. Origination rates were significantly different between branch generations (two-way analysis of variance of natural-log-transformed data, $F_{2,8.9} = 15.9$, $P = 0.001$). Neither the time interval nor interaction effects were significant ($P = 0.1$ and 0.29, respectively). Branch growth also declined as a function of colony height independent of branch generation (Fig. 4A). The net effect of these processes was that, on San Salvador, colonies seldom exceeded 50 cm in height (Fig. 6A), and all of the monitored colonies were less than 70 cm in height.

Discussion

The results lead to conclusions in three interrelated areas, all of which suggest that *Pseudopterogorgia elisabethae,* and by extension other gorgonians, have body plans that are under greater developmental control than generally considered in discussions of modularity. First, although colonies are built through the iterative generation of polyps, both the branch and the whole colony behave as integrated units. Second, the tempo and mode of branch origination and growth generate branches and colonies of finite size, as well as predictable form. Third, branches on *P. elisabethae* colonies appear to fall into two distinct developmental catego-

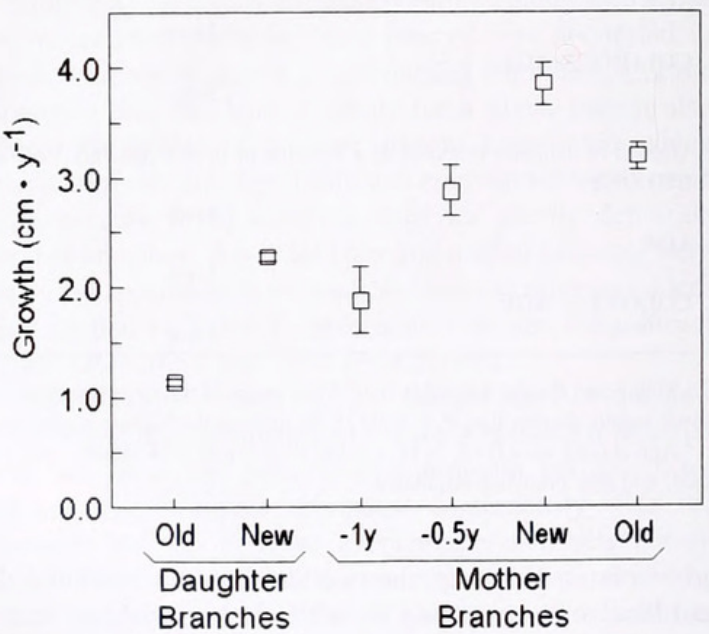

Figure 5. Growth of *Pseudopterogorgia elisabethae* branches from San Salvador, Bahamas. Daughter branches were divided into those present at the start of observations (old daughter) and those that were <0.5 y (new daughter). Branches that transformed from daughter to mother branches are included as mother branches, and their growth rates at 1 y and 0.5 y before they started branching, are denoted as −1 and −0.5 y, respectively. "New" mother branches started branching during the 6 months in which the growth rate was determined. Branches that were already mother branches at the start of the observations are labeled "Old." All values are means ± standard error.

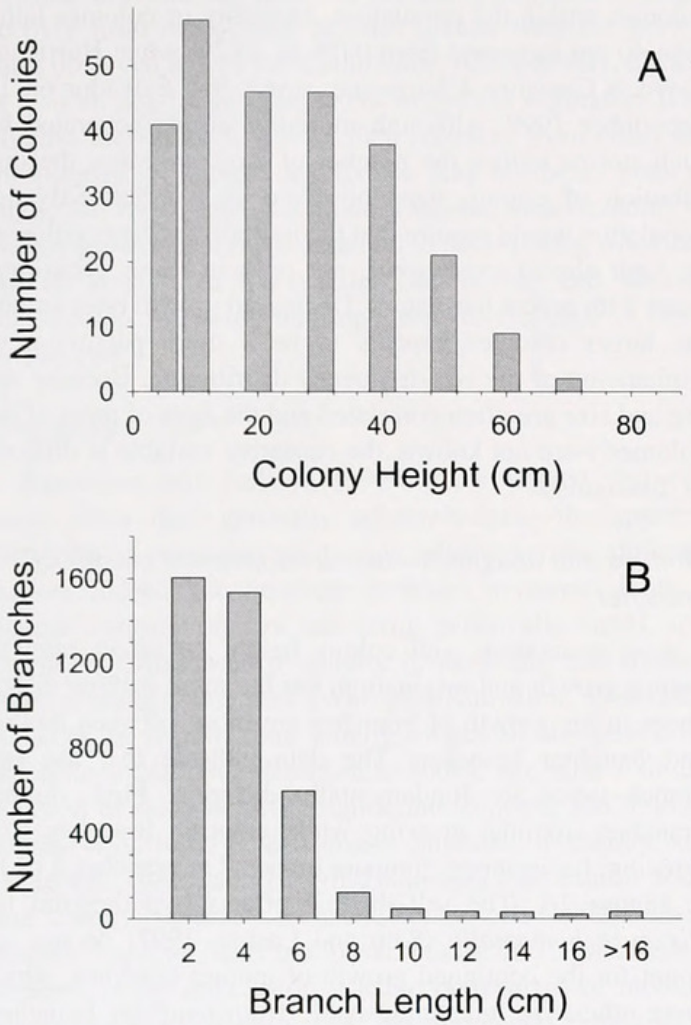

Figure 6. Size-frequency distribution of colonies (A) and branches (B) from 261 *Pseudopterogorgia elisabethae* found within 70 m^2 of belt transects at 12–16 m depth at San Salvador, Bahamas. Heights were measured as the length of the tallest branch, and the measurement was made either from photos or in the field with a tape measure. Height classes are centered on the upper limit of the size class; 0–5 cm and >50 cm classes were lumped with adjacent size classes in the statistical analyses. Branch lengths were determined from digital images either of entire colonies or, in the case of large colonies, from digital images of an arbitrarily selected subset of branches. Branch measurements are from December 1999. Branches include both daughter and mother branches.

ries, mothers and daughters, which exhibit different growth characteristics from the time they first originate. In addition to those findings, the results also have implications for colony regeneration following disturbance, and in a final section, we discuss the implications of the study for the harvest of *P. elisabethae,* which is already being conducted in the Bahamas.

Branches as modules

Growth of *P. elisabethae* colonies is best described in terms of branches, and each branch behaves as an integrated unit, or module. Daughter branches follow a predictable developmental sequence in which they first grow rapidly, then slow as they age, and eventually stop growing. Mother branches follow a sequence in which they grow and generate both daughter and mother branches, but their growth and the rate at which they generate new branches also slow as the colony grows.

Concepts of integration and modularity have been used in describing the development and evolution of suites of morphologic features among solitary organisms (Pagliucci, 2002). This approach emphasizes the developmental relationships of traits. While not all groups of integrated traits need be "modules" as used in the invertebrate and plant literature, the modules that make up invertebrate colonies should exhibit the statistical correlations indicative of integrated development and evolution (*i.e.,* Magwene, 2001). Our analysis of branch growth rates suggests that both branches and colonies develop as integrated units. Similarly, correlations among five traits in 21 gorgonian species also differentiated polyp-level traits from those at the branch or colony level (Sánchez and Lasker, 2003). In the context of this broader definition of modularity, three levels of modularity or integration can be recognized in *P. elisabethae:* polyps, branches, and the colony. The polyp has always been recognized as a distinct unit with a well-defined ontogeny and determinate growth. Our data on branch growth suggest that the branches of *P. elisabethae* also exhibit a well-defined ontogeny. The colony must also be considered as a level of organization, because growth of the branches is also dependent on colony-level traits, that is, on colony height and generational order.

Determinate growth among modular organisms

Daughter branches on *P. elisabethae* stop growing as they age, and the growth rates decrease as the colony increases in height. Furthermore, origination rates of branches decrease with branch generation. In concert, these changes in developmental rates conserve a colony's size and form, a pattern that is functionally equivalent to determinate growth. The pattern of determinate growth observed in *P. elisabethae* colonies could be generated by set developmental programs or through predictable responses to microenvironmental variation around the colony. Both are, at some level, growth in response to cues and are not mutually exclusive. We argue that a developmental program is the principal factor controlling the determinate growth of branches, while the size of whole colonies probably reflects a mix of both developmental effects and environmental and historical factors (Rinkevich, 2000).

Branches have a clear developmental cycle which, among the daughter branches, leads them to stop growing long before they reach 10 cm in length. Kim and Lasker (1997) report that interior branches of the gorgonian *Plexaura homomalla* have lower growth rates than those on the perimeter of the colony, a pattern they attributed to nutrient

supply and self-interference. A number of realistic models of form in modular organisms have been developed, in which growth is controlled by the local responses of the individual modules to their local environment (Braverman, 1974; Graus and MacIntyre, 1976; Colasanti and Hunt, 1997; Kaandorp and Kübler, 2001; Oborny *et al.,* 2001). However, the decrease in branch growth among *P. elisabethae* colonies is best described as an age-dependent decrease. Thus, although the smallest branches had the greatest growth rates, many small branches also exhibited low growth (Fig. 5). Furthermore, daughter branches stop growing while adjacent mother branches continue to grow, which indicates that position alone does not control branch growth.

Colony size in *P. elisabethae* also appears to be determinate, and observations of determinate growth have been reported among a wide range of colonial taxa. As noted, octocoral descriptions often include maximum colony sizes (*i.e.,* Bayer, 1961), and decreasing growth with increasing colony size has been reported for a number of gorgonians (Grigg, 1974; Velimirov, 1975; Mitchell *et al.,* 1993; Coma *et al.,* 1998; Cordes *et al.,* 2001). Among scleractinian corals, determinate growth of independently growing branches generates colonies with determinate form and size (Rinkevich, 2002). Among botryllid tunicates, groups of zooids, referred to as systems, undergo synchronous senescence (Sabbadin, 1969), and whole colonies, including isolated explants from a common source, undergo simultaneous senescence (Milkman, 1967; Rinkevich *et al.,* 1992). In addition, graptolite colonies are believed to have had determinate growth leading to distinct species-specific forms and sizes (Mitchell, 1988).

Maximum size alone does not demonstrate determinate growth. In a manner functionally equivalent to determinate growth, modular organisms may also stop growing, not according to a genetically determined developmental plan, but due to size-dependent interactions between the colony and environment, such as the balance between nutrient uptake and metabolic rates. Size-dependent change in colony growth that is mediated by metabolic rate and resource capture has been modeled by Sebens (1982, 1987) and by Kim and Lasker (1998). Taxa that exhibit growth patterns consistent with these simple models have been reported in octocorals (McFadden, 1986; Kim and Lasker, 1997) and tunicates (Holyoak, 1997).

Ecological processes such as size-dependent mortality also could generate a maximum colony size and the appearance of determinate growth. Colonies are susceptible to being knocked over because drag forces increase with colony size and bioerosion weakens the substratum around the holdfast (Birkeland, 1974). During most of this study, however, mortality decreased with colony height (mortality per 6 months: 0–10 cm, 0.146; 11–20 cm, 0.108; 21–30 cm, 0.071; 31–40 cm, 0.048; >40 cm, 0.000). That pattern of mortality would have led to the accumulation of large colonies within the population. Mortality of colonies taller than 40 cm increased from 0.0% to 40.2% when Hurricane Floyd, a Category 4 hurricane, struck San Salvador on 13 September 1999. Although mortality events generated by such storms reduce the number of large colonies, the distribution of colony sizes observed in the San Salvador population would require that the mortality of large colonies be high almost every year, not only in those occasional years with severe hurricanes. Decreased growth rates among the larger colonies appears to be a more parsimonious explanation of the size-frequency distribution. Because aging and size are often correlated and the ages of most of the colonies were not known, the causative variable is difficult to distinguish.

Mothers and daughters—two developmental classes of branches

Age, generation, and colony height all affect rates of branch growth and origination, but the most striking differences in the growth of branches are those between mother and daughter branches. The data indicate that the two branch types are fundamentally different. First, mother branches continue growing while adjacent branches stop growing; for instance, compare branch 2 to branches 4 or 17 in Figure 2A. The self-shading effects hypothesized for *Plexaura homomalla* (Kim and Lasker, 1997) do not account for the continued growth of mother branches, which were otherwise indistinguishable from daughter branches. Second, mother branches exhibit high growth rates as soon as they originate, well before they have produced their first daughter. However, the two branch types are not immutable. When colonies are damaged, branches that were previously daughter branches begin to extend and generate new branches (Castanaro and Lasker, 2003). Understanding whether and how branches "become" mother branches will be essential to our understanding of developmental processes among these colonial organisms.

Applications

Modular growth is an especially advantageous growth strategy, when transplants are used to remediate populations (Rinkevich, 2000), when explants are used as stock in mariculture, and for the sustained harvest of wild populations where colonies are cropped at regular time intervals (Castanaro and Lasker, 2003). Understanding the pattern of growth of *P. elisabethae* colonies is particularly important because this species is harvested and extracted for a class of natural products called pseudopterosins (Mayer *et al.,* 1998). Material for extraction is collected by cropping branches from colonies. If growth is inversely related to the size of the colony, then reductions in colony size will enhance growth and productivity. Among harvested populations, this suggests that colonies can be maintained at an

optimal size, and that naturally occurring populations might recover from disturbance at rates greater than the growth rates observed before the disturbance. Alternatively, if there is also an age-based component to growth regulation (*i.e.,* Hughes and Connell, 1987), then recovery from either anthropogenic or natural disturbance may not be as great as suggested by colony size alone. Detailed understanding of colony growth patterns is essential to determining whether a species is suitable for sustained harvesting and whether remediation following anthropogenic disturbance is likely to succeed.

Conclusions

Knowlton and Jackson (1994) have argued that—far more often than generally acknowledged—the apparent plasticity of coral reef cnidarians reflects genetic differentiation. Indeed, the literature provides numerous hints of genetic controls on size and form: genetically based, species-level differences in colony form within the *Montastraea annularis* complex (Weil and Knowlton, 1994); differences in regeneration among clones of the reef coral *Stylophora pistillata* (Rinkevich, 2000); age effects on the survival of stony corals (Hughes and Connell, 1987); graptolites with highly determinate patterns of colony size (Mitchell, 1986); and botryllid tunicates that exhibit zooid and colony senescence (Rinkevich *et al.,* 1992). Those studies, together with our observations of *P. elisabethae,* underscore the conclusion that the body plans of modular organisms are constrained by developmental programs as well as by the environment. If the potential for indeterminate growth that modularity seemingly confers is not realized, then, "Why not?" becomes a valuable question. How does constraining the size of branches affect the array of forms that can be realized by the whole colony? Since reproductive output is a function of colony size (Beiring and Lasker, 2000), can determinate colony growth be explained by trade-offs between current and future reproduction? Are there hydrodynamic or feeding advantages to the plumelike form of *P. elisabethae,* and are the advantages dependent on branch size, on colony size? Although colonial organisms are far more plastic than unitary forms, their growth should be studied as an internally regulated process that generates colony form. Both the form and the processes by which it is realized affect fitness and are subject to natural selection.

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Literature Cited

Bayer, F. M. 1961. *The Shallow Water Octocorallia of the West Indian Region.* Martinus Nijhoff, The Hague.

Beiring, E. A., and H. R. Lasker. 2000. Egg production by colonies of a gorgonian coral. *Mar. Ecol. Prog. Ser.* **196:** 169–177.

Birkeland, C. 1974. The effects of wave action on the population dynamics of *Gorgonia ventalina* Linnaeus. *Stud. Trop. Oceanogr.* **12:** 115–126.

Braverman, M. 1974. The cellular basis of morphogenesis and morphostasis in hydroids. *Oceanogr. Mar. Biol. Annu. Rev.* **12:** 129–221.

Brazeau, D. A., and H. R. Lasker. 1988. Inter- and intraspecific variation in gorgonian colony morphology: quantifying branching patterns in arborescent animals. *Coral Reefs* **7:** 139–143.

Castanaro, J., and H. R. Lasker. 2003. Effects of clipping on growth of colonies of the Caribbean gorgonian *Pseudopterogorgia elisabethae. Invertebr. Biol.* **122:** 299–307.

Colasanti, R. L., and R. Hunt. 1997. Resource dynamics and plant growth: A self-assembling model for individuals, populations and communities. *Funct. Ecol.* **11:** 133–145.

Coma, R., M. Ribes, M. Zabala, and J.-M. Gili. 1998. Growth in a modular colonial marine invertebrate. *Estuar. Coast. Shelf Sci.* **47:** 459–470.

Cordes, E. E., J. W. Nybakken, and G. Van Dykhuizen. 2001. Reproduction and growth of *Anthomastus ritteri* (Octocorallia: Alcyonacea) from Monterey Bay, California, USA. *Mar. Biol.* **138:** 491–501.

Done, T. J. 1987. Simulation of the effects of *Acanthaster planci* on the population structure of massive corals in the genus *Porites:* evidence of population resilience? *Coral Reefs* **6:** 75–90.

Done, T. J. 1988. Simulations of the recovery of pre-disturbance size structure in populations of *Porites* spp. damaged by crown-of-thorns starfish. *Mar. Biol.* **100:** 51–61.

Graus, R. R., and I. G. MacIntyre. 1976. Light control of growth form in colonial reef corals: computer simulation. *Science* **193:** 895–898.

Grigg, R. W. 1974. Growth rings: annual periodicity in two gorgonian corals. *Ecology* **55:** 876–881.

Harper, J. L., and J. White. 1974. The demography of plants. *Annu. Rev. Ecol. Syst.* **5:** 419–463.

Holyoak, A. R. 1997. Patterns and consequences of whole colony growth in the compound ascidian *Polyclinum planum. Biol. Bull.* **192:** 87–97.

Hughes, R. N. 1989. *A Functional Biology of Clonal Animals.* Chapman and Hall, London.

Hughes, T. P., and J. H. Connell. 1987. Population dynamics based on size or age? A reef-coral analysis. *Am. Nat.* **129:** 818–829.

Jackson, J. B. C. 1977. Competition on marine hard substrata: the adaptive significance of solitary and colonial strategies. *Am. Nat.* **111:** 743–767.

Jackson, J. B. C., L. W. Buss, and R. E. Cook, Eds. 1985. *Population Biology and Evolution of Clonal Organisms.* Yale University Press, New Haven.

Kaandorp, J. A., and J. E. Kübler. 2001. *The Algorithmic Beauty of Seaweeds, Sponges and Corals.* Springer-Verlag, Heidelberg.

Kim, K., and H. R. Lasker. 1997. Flow-mediated resource competition

in the suspension feeding gorgonian. *Plexaura homomalla* (Esper). *J. Exp. Mar. Biol. Ecol.* **215:** 49–64.

Kim, K., and H. R. Lasker. 1998. Allometry of resource capture in colonial cnidarians and constraints on modular growth. *Funct. Ecol.* **12:** 646–654.

Knowlton, N., and J. B. C. Jackson. 1994. New taxonomy and niche partitioning on coral reeds—jack of all trades or master of some? *Trends Ecol. Evol.* **9:** 7–9.

Lasker, H. R., and J. A. Sánchez. 2002. Allometry and astogeny of modular organisms. Pp. 207–253 in *Reproductive Biology of Invertebrates. Vol. XI. Progress in Asexual Reproduction,* R. N. Hughes, ed. John Wiley & Sons, New York.

Magwene, P. M. 2001. New tools for studying integration and modularity. *Evolution* **55:** 1734–1745.

Mayer, A. M. S., P. B. Jacobson, W. Fenical, R. S. Jacobs, and K. B. Glaser. 1998. Pharmacological characterization of the pseudopterosins: Novel anti-inflammatory natural products isolated from the Caribbean soft coral, *Pseudopterogorgia elisabethae. Life Sci.* **62:** 401–407.

McFadden, C. S. 1986. Colony fission increases particle capture rates of a soft coral—advantages of being a small colony. *J. Exp. Mar. Biol. Ecol.* **103:** 1–20.

Milkman, R. 1967. Genetic and developmental studies on *Botryllus schlosseri. Biol. Bull.* **132:** 229–243.

Mitchell, C. E. 1986. Morphometric studies of *Climacograptus* (Hall) and the phylogenetic significance of astogeny. Pp. 119–129 in *Palaeoecology and Biostratigraphy of Graptolites,* C. P. Hughes and R. B. Rickards, eds. Geological Society Special Publication 20, Geological Society of America, Boulder, CO.

Mitchell, C. E. 1988. The morphology and ultrastructure of *Brevigraptus quadrithecatus* n. gen., n. sp. (Diplograptacea), and its convergence upon *Dicaulograptus hystrix* (Bulman). *J. Paleont.* **62:** 448–463.

Mitchell, N. D., M. R. Dardeau, and W. W. Schroeder. 1993. Colony morphology, age structure, and relative growth of two gorgonian corals, *Leptogorgia hebes* (Verrill) and *Leptogorgia virgulata* (Lamarck), from the northern Gulf of México. *Coral Reefs* **12:** 65–70.

Oborny, B., T. Czaran, and A. Kun. 2001. Exploration and exploitation of resource patches by clonal growth: a spatial model on the effect of transport between modules. *Ecol. Model.* **141:** 151–169.

Pagliucci, M. 2002. Touchy and bushy: phenotypic plasticity and integration in response to wind stimulation in *Arabidopsis thaliana. Int. J. Plant Sci.* **163:** 399–408.

Rinkevich, B. 2000. Steps towards the evaluation of coral reef restoration by using small branch fragments. *Mar. Biol.* **136:** 807–812.

Rinkevich, B. 2002. The branching coral *Stylophora pistillata:* contribution of genetics in shaping colony landscape. *Isr. J. Zool.* **48:** 71–82.

Rinkevich, B., R. J. Lauzon, B. W. M. Brown, and I. L. Weissman. 1992. Evidence for a programmed life-span in a colonial protochordate. *Proc. Natl. Acad. Sci. USA* **89:** 3546–3550.

Sabbadin, A. 1969. The compound ascidian *Botryllus schlosseri* in the field and laboratory. *Publ. Stn. Zool. Napoli* **37**(suppl.): 67–72.

Sánchez, J. A. 2002. Dynamics and evolution of colony form among branching modular organism, Ph.D. dissertation. University at Buffalo, State University of New York, Buffalo, NY. 137 pp.

Sánchez, J. A., and H. R. Lasker. 2003. Patterns of morphologic integration in marine modular organisms: supra-module organization in branching octocoral colonies. *Proc. R. Soc. Lond.* **DOI:** 10.1098/rspb.2003.2471.

Sánchez, J. A., W. Zeng, V. R. Coluci, C. Simpson, and H. R. Lasker. 2003. How similar are branching networks in nature? A view from the ocean: Caribbean gorgonian corals. *J. Theor. Biol.* **222:** 135–138.

Sebens, K. P. 1982. The limits to indeterminate growth: an optimal size model applied to passive suspension feeders. *Ecology* **63:** 209–222.

Sebens, K. P. 1987. The ecology of indeterminate growth in animals. *Annu. Rev. Ecol. Syst.* **18:** 371–407.

Velimirov, B. 1975. Growth and age determination in the sea fan *Eunicella clavolinii. Oecologia* **19:** 259–272.

Weil, E., and N. Knowlton. 1994. A multi-character analysis of the Caribbean coral *Montastraea annularis* (Ellis & Solander, 1786) and its two sibling species, *M. faveolata* (Ellis & Solander, 1786) and *M. franksi* (Gregory, 1895). *Bull. Mar. Sci.* **55:** 151–175.