

have similar distribution patterns in some cases.

LITERATURE CITED

Balser, Teri C. and Sheryl L. Soucy. 1992. The effects of *Diadema antillarum* on two Jamaican reefs: an examination of community response. This volume.

Goreau, Thomas F., and Nora I. Goreau. 1973. The ecology of Jamaican Coral Reefs, II. Geomorphology, zonation, and sedimentary phases. *Bulletin of Marine Science* 23:2, pp. 399-464.

Hay, Mark E. 1981. The functional morphology of turf-forming seaweeds: persistence in stressful marine habitats. *Ecology* 62:3 pp. 739-750.

Littler, Mark M. and Diane S. Littler. 1973. The evolution of thallus form and survival strategies in benthic marine macroalgae: field and laboratory tests of a functional form model. *The American Naturalist* 116(1), pp. 25-44.

Littler, Mark M., Diane S. Littler, and Phillip R. Taylor. 1983. Evolutionary strategies in a tropical barrier reef system: functional-form groups of marine macroalgae. *Journal of Phycology* 19?, pp. 229-237.

Padilla, Dianna K. 1984. The importance of form: differences in competitive ability, resistance to consumers and environmental stress in an assemblage of coralline algae. *Journal of Experimental Marine Biology and Ecology* 79, pp. 105-127.

Appendix A. Example of the contingency table format used for G-tests between quadrat types. (Comparison of *A. stellata* in quadrat types A and B)

	Plot type	
	A (45ft, sand)	B (45ft, hard)
#of plots with <i>A. stellata</i>	0	6
#of plots w/o <i>A. stellata</i>	7	4

DISTRIBUTION AND BEHAVIOR OF *BUNODEOPSIS ANTILLIENSIS*

Anthony L. Guerrero

Abstract. I investigated the distribution and abundance of *Bunodeopsis antilliensis* in the field and several aspects of its behavior in the laboratory. *Bunodeopsis* was found only on the leaves of *Thalassia testudinum*. Its density in *Thalassia* fields increased with depth to a maximum at three meters, after which no *Thalassia* and no *Bunodeopsis* were found. It showed a diel behavior pattern of expanding its tentacles at night and contracting them during the day. I found it does try to avoid predation by *H. canunculata* (fireworm), but the response does not seem mediated by a long distance chemical cue. It did however respond to being touched with a piece of fireworm and fireworm homogenate significantly more than to a control, suggesting a tactile cue, or a combination of tactile and contact chemical cue. The anemones did not respond to external stimuli of predator or prey (*Antem ia*) when contracted.

In nature *Bunodeopsis* is found mostly on the tips of *Thalassia* leaves. Settling experiments showed this is not due to random settling, but that the highest points are actively chosen by these anemones. Further experiments revealed that there appears to be an influence of both light intensity and flow rates when making this choice. (ALG)

INTRODUCTION (ALG)

Bunodeopsis antilliensis is an anemone with a highly toxic sting found attached to leaves of *Thalassia testudinum* throughout the Caribbean (Humann 1992). It is most often attached at the tips of the blades (pers. obs.). The maximum length of its base ranges up to ~2cm (pers. obs.) and when extended it has long, thin, opaque tentacles attached to a long column. Surrounding the column base are columnar vesicles. These vesicles hold most of *Bunodeopsis*' zooxanthellae. During the day *Bunodeopsis* is contracted and the columnar vesicles are inflated. At night the anemone extends its tentacles and deflates the vesicles. *Bunodeopsis* is phototactic and will orient itself on the upper surface of an object (Sebens and DeRiemer 1977). Besides movement to reach the upper surface, these anemones are reported to release from a *Thalassia* leaf in response to predation by *Hermodice carunculata* (John Gilbert, pers. comm.).

Here I will attempt to describe their distribution and some of their behaviors using both field components and laboratory observations and experiments.

Field components. I will attempt to determine if *Bunodeopsis* is located only on *Thalassia* blades. I will also look at their distribution with depth. I predict that density will be low at shallow depths due to high ultra-violet intensity and turbulence, low at deep depths due to light attenuation, and maximum at some intermediate depth. Also I predict these anemones will decrease the amount of herbivory suffered by *Thalassia* on or near blades colonized because fish will avoid these blades due to the highly toxic sting.

Laboratory components. I will look at the diel activity pattern of *Bunodeopsis*. Based on the field observations of Sebens and DeRiemer, I predict that *Bunodeopsis* will be extended at night and contracted during the day.

I predict that the anemones will respond to predation by fireworms because of communications with J. Gilbert, although I do not have a prediction as to what the cue for this response will be. Therefore, I will perform several different experiments to determine, i) if there is indeed a response and ii) the cue for this response.

I will look at the difference in responses between contracted and extended anemones. I predict that anemones with their tentacles extended will respond more often and more readily to stimuli.

Finally, I will look at the settling behavior of *Bunodeopsis*. As stated above, Sebens and DeRiemer have determined that these anemones will position themselves on the upper surface of an object. I intend to see if they will move to a different object with higher exposure if presented the opportunity, and if the choice is based simply on light intensity or if there is some consideration for water flow. I predict that both of these factors are important for determining the microhabitats selected because greater light intensity will increase photosynthetic rates and higher flow will presumably cause a higher probability of encountering zooplankton. I will also attempt to investigate if the anemones are present on the tips of *Thalassia* due to random settlement or if there is some active choice for these sites. Since my experience in collecting these anemones has shown they can readily move, I predict there is a strong component of anemone choice in picking these high sites.

METHODS (ALG)

Field components. Field observations were made in Jamaica on the western side of the Discovery Bay back reef from 28 February to 3 March 1992.

To search for *Bunodeopsis* on various substrates I ran 4 snorkel transects haphazardly through the back reef area. Each transect was 100m long and 15cm wide, and passed over sand, algae, coral, rubble, and *Thalassia* beds. Depths from 1 to 3+ meters were sampled, although the entire range was not included in every transect. I swam the transects approximately 15cm above the substrate at approximately 10cm/sec, searching for *Bunodeopsis* visually and by touch. I checked my speed periodically by timing how long it took me to cover 1m.

For density measurements I ran 2 non-intersecting transects in the back reef using SCUBA. I placed my transects in the *Thalassia* beds at the far west end of Discovery Bay away from areas where students had been collecting *Thalassia*. Each transect was 100m long. Every 3m I placed a 1m² square and thoroughly searched it, visually and by touch, for *Bunodeopsis*. I measured depth with my depth gauge and body lengths. I did not search any areas which did not contain *Thalassia* due to the results of the snorkel transects (presented below).

I attempted to measure grazing on colonized and uncolonized *Thalassia* leaves but this proved extremely difficult because: i) I could not find sufficiently standardized blades colonized by *Bunodeopsis* and therefore could not set up the assays in the field, and ii) *Bunodeopsis* which had colonized standardized leaves in the lab released from the leaves when placed

back in the ocean. This experiment was therefore discontinued.

Laboratory components. *Bunodeopsis* used for the laboratory tests were collected from the field on 26-28 February and 1 March. While in the lab they were kept in a fish tank with anchored *Thalassia* and running seawater. They were fed brine shrimp every 24 to 48 hours. Those anemones being used for the settling experiments described below were not fed during those experiments.

The fireworms used were captured on 27 and 29 February and kept in the lab in a bucket of running seawater containing sand and rocks. They were not fed except for the *Bunodeopsis* they ate during predation trials.

Laboratory experiments were run on four aspects of behavior:

1) *Diel activity cycle.* Ten anemones were placed in a separate tank (with running water) and anchored *Thalassia* for 60 hours. The tank was covered with black plastic from 1900 to 700 and uncovered from 700 to 1900. At various times throughout the day I recorded the number of anemones extended and contracted.

2) *Response to predation.*

I. I placed a *Bunodeopsis* in a glass finger bowl with 200ml of seawater. After allowing the anemone to attack to a floating *Thalassia* leaf or the side of the bowl. I subjected it to one of four treatments: a) control, inject 20ml of plain seawater; b) fireworm, inject 20ml of seawater in which a fireworm had been sitting for at least 1 hour to see if the anemones respond to a chemical released by the fireworms; c) fireworm and *Bunodeopsis*,

inject 20ml of seawater in which a fireworm had eaten a *Bunodeopsis* in the last 10min to see if the anemones respond to an alarm pheromone released by another *Bunodeopsis* or a chemical released by the fireworm when it is feeding; d) *Bunodeopsis* extract, inject 20ml of 200ml seawater in which an anemone has been chopped up to see if the anemones are responding to a chemical from the *Bunodeopsis* only.

The treatments were injected at 1.5-2ml/sec using a syringe. When injecting, I placed the syringe as far away from the anemone as possible so that an influx of liquid would not disturb the anemone. Trials included one test of each treatment and were run at night using extended anemones. Finger bowls were washed out and refilled with fresh seawater after each treatment and the *Thalassia* leaves were changed. I ran 10 trials over three nights. A response to the treatment was defined as releasing hold within five minutes of injection.

II. I again placed anemones in glass finger bowls with 200ml of seawater, although this time I also let them attach to the bottom of the finger bowl as well. I subjected the anemones to one of 3 treatments: a) control, touch with a Q-tip soaked in seawater; b) fireworm homogenate, touch with a Q-tip soaked in fireworm homogenate to see if the anemones are responding to a chemical present in the fireworm; c) fireworm, touch with severed head (about Q-tip size) of a fireworm to see if the anemones are responding to being touched by an actual fireworm.

The probe or head was held about 3mm from the anemone's column and wiggled back and forth about

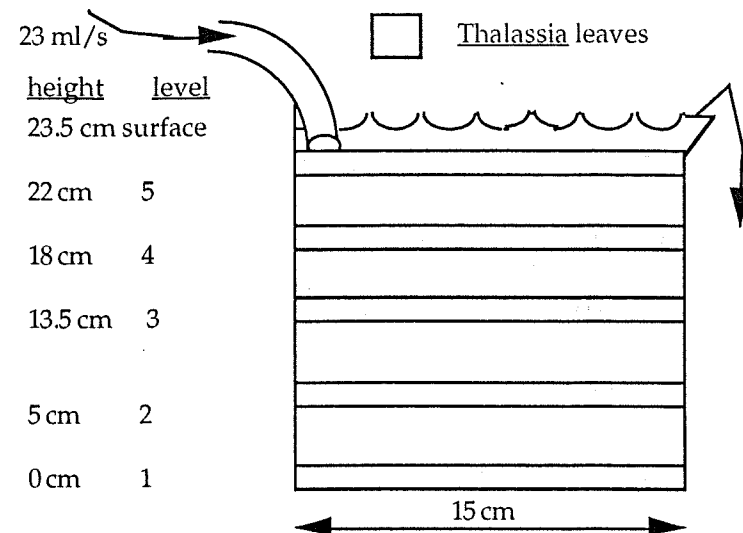


Figure 1. Top flow experimental set-up.

3mm to each side for one minute. A response was defined as releasing hold within that one minute. Ten trials, each consisting of running one of each treatment, were conducted with extended anemones over two nights. The bowls were washed out and the water replaced between treatments.

III. The final part of the predation study consisted of placing fireworms into the *Bunodeopsis* holding tank and observing the interactions. I observed for approximately 2 hours in one afternoon.

3) Responses of *Bunodeopsis*

I. I dropped 12 contracted and 8 extended anemones one by one into a plastic cup containing fireworms and sea water and observed the interactions. The contracted anemones did not extend when dropped into the cups.

II. Using a pipette I injected live brine shrimp at 17 contracted and 11 extended anemones and observed their reactions.

4) Settling experiment

I set up a bucket with *Thalassia* leaves at 5 heights off the bottom. The leaves were 15cm long and attached at each end by clothspins which were tied to the side of the bucket. Running seawater entered the bucket 5cm below the surface at approximately 23ml/sec. and exited over the top on the other side so flow was primarily across the water's surface (Figure 1). I placed weighted *Thalassia* blades colonized by anemones at the bottom of the bucket. I observed

the distribution of the anemones on the leaves and the bucket surface after 12 and 24 hours. This was repeated once ($n_1=10$ anemones, $n_2=9$ anemones). For analysis I assigned each level a number from 1 to 5 and included anemones attached to the side as being on the level whose distance most closely matched their distance from the bucket's bottom.

The second bucket I set up was slightly smaller in height and diameter so there were only 3 levels of *Thalassia*. The blades were still 15cm long but were buckled. I covered the outside of the bucket with black plastic so that light conditions would be similar to the first, grey bucket. In contrast to the first bucket though, flow extended at the bottom of the bucket and exited through a hole covered with screening at the bottom of the bucket (Figure 2). Flow rate was similar to that in the first bucket. A tiny drip of water did exit the bucket over the top lip, but the vast majority of water exited through the hole. I did this to be sure I would not return 12 hours later to find my bucket empty. Anemones were poured into the bucket so they were

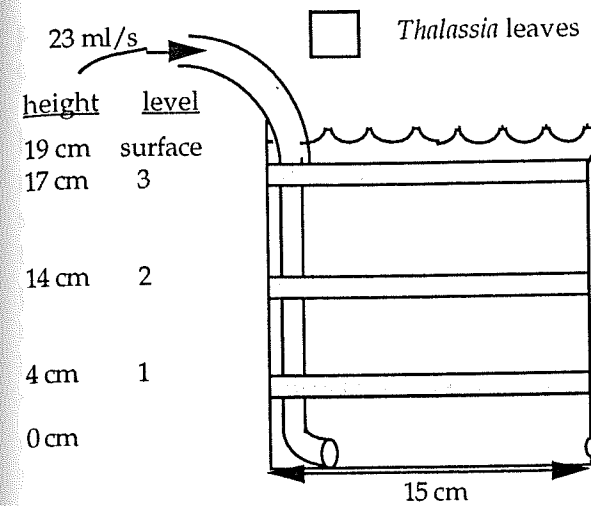


Figure 2. Bottom flow experimental set-up.

scattered throughout the water column at the start of the experiment. I observed their distributions at 12 and 24 hours. This was also repeated once ($n_1=7$ anemones, $n_2=10$ anemones). Analysis was conducted as explained above for the first bucket.

The first bucket described was named the "top flow" experimental setup and the second bucket was named "bottom flow".

RESULTS (ALG)

Field components. In the four snorkel transects I found 57 *Bunodeopsis* on *Thalassia* blades and zero on all other substrates. Density versus depth measurements did not exhibit the curvilinear relationship expected. Instead, there was a significant positive correlation between depth and anemone density ($r=0.58$, $n=52$, $p<0.001$) to a depth of 3m where the density then dropped to zero because no *Thalassia* were found below this depth (Figure 3).

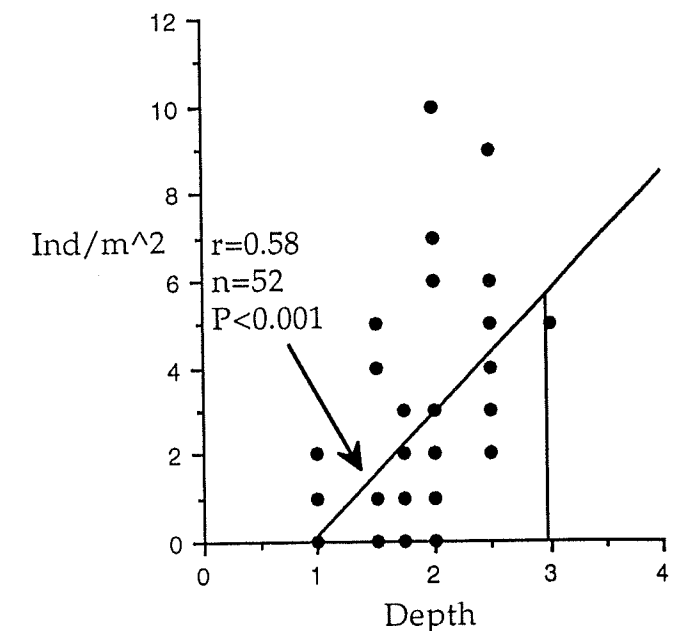


Figure 3: Anemone density versus depth

Lab components. A general trend was found for *Bunodeopsis* to be contracted during the day and extended at night, although there was some deviation from this pattern (Figure 4).

There was no significant difference between anemones responding to injected seawater (0 responses in 10 trials), water in contact with a fireworm (1 response in 10 trials), water in which a *Bunodeopsis* had just been eaten by a fireworm (3 responses in 10 trials), and anemone extract (3 responses out of 10 trials; Table 1; $G_{adj}=4.31$, $0.5>p>0.1$). There was a significant difference between anemones

Table 1. Response of <i>Bunodeopsis</i> to a long distance chemical cue from <i>H. carunculata</i> .				
Treatment	control (a)	(b)	(c)	(d)
Response	0	1	3	3
No response	10	9	7	7
See text for explanation of treatments.				

responding to being touched with a Q-tip (1 response in 10 trials), a Q-tip soaked in fireworm homogenate (5 responses in 10 trials), and a piece of fireworm (8 responses in 10 trials;

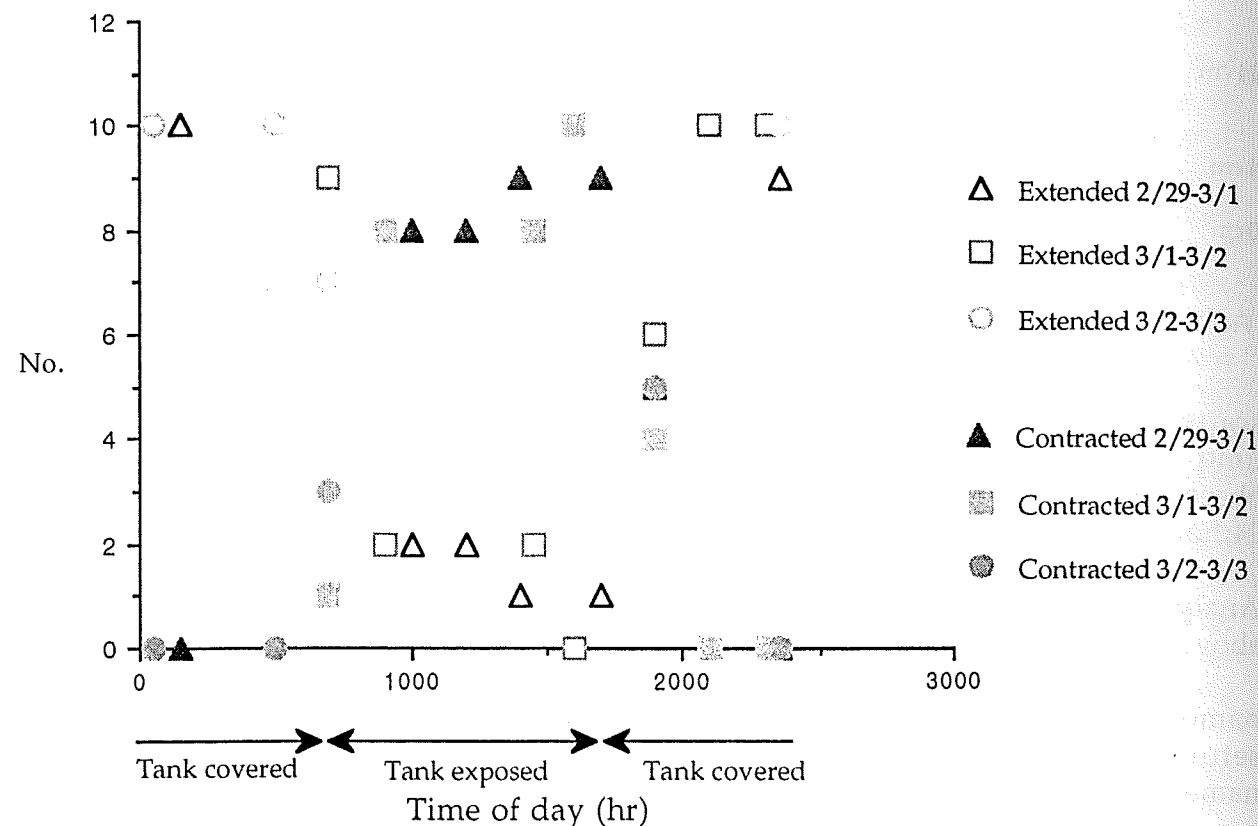


Figure 4: Number of anemones extended and contracted versus time of day.

Table 2. Response of *Bunodeopsis* to contact and/or short distance chemical cue from *H. carunculata*.

Treatment	control (a)	(b)	(c)
Response	1	5	8
No response	9	5	2

See text for explanation of treatments.

Contracted anemones had no response to brine shrimp being injected at them (17 anemones used), while all the extended anemones exhibited normal prey capture and feeding behaviors (11 anemones used). Of the 12 contracted anemones used for

Table 2; $G_{adj}=15$, $p<0.005$). There was no difference between the later two touching treatments ($G_{adj}=2.01$, $0.5>p>0.1$).

the predation segment, none responded until the fireworm had wrapped itself around it and began to feed. At this point the anemones would extend their tentacles but all were eaten despite this. All 8 of the extended anemones attempted to beat off the fireworm. Some of the larger ones managed to avoid being eaten for several minutes and 2 managed to climb up the side of the cup. I did not test either of these for significance because the appearance of zeros in two of the cells makes a G-test inap-

propriate, although the trend is obvious.

In the flow experiments, after 12 hours the distribution of the anemones in the "top flow" buckets differed significantly from a random distribution for both trials ($G_{adj1}=30.18$, $p_1<0.005$; $G_{adj2}=13.48$, $p_2<0.001$), being strongly grouped near the top of the bucket (Table 3). Anemones in the "bottom flow" buckets did not have a distribution significantly different from random after 12 hours for both trials ($G_{adj1}=3.19$, $0.5>p_1>0.1$; $G_{adj2}=0.5$, $0.9>p_2>0.5$). Although I did not analyze it these distributions did not change appreciably over the second twelve hours.

DISCUSSION (ALG)

In the field I found *Bunodeopsis* solely on *Thalassia* blades, and in the holding tank they settled only on *Thalassia* and the smooth, glass walls. Over the course of the whole study I noticed only three out of over eighty anemones in the tank attached to rocks and all three had found new holds within two hours. This suggests that a smooth, flat surface is necessary for *Bunodeopsis* colonization. *Thalassia* apparently provides the only suit-

Table 3. Distribution of *Bunodeopsis* in bucket after 12 hours of "top flow" treatment.

Level	5	4	3	2	1
Trial 1	8	0	0	0	1
Trial 2	6	1	1	0	1

Table 4. Distribution of *Bunodeopsis* in bucket after 12 hours of "bottom flow" treatment.

Level	3	2	1
Trial 1	2	5	0
Trial 2	4	4	2

able flat surface in the natural environment and hence *Bunodeopsis* distribution is tied to *Thalassia* distribution. When examining the density of individuals I did find density increasing with depth; however, I never reached a threshold depth where the density began to decrease because no *Thalassia* was found below three meters. In Discovery Bay, *Bunodeopsis*' depth limit does not seem to be a function of light attenuation, but lack of suitable habitat provided by *Thalassia*.

The low density of individuals in shallow water could be explained by ultraviolet light stress or wave stress. I did notice that there seemed to be more individuals in shallow water where the grass is short than where it is long. This might suggest that wave stress is the more limiting of the two factors because individuals on short blades are less likely to be swept around and bashed into the substratum or other blades and would spend less time shaded by other blades. I also noticed the density seemed to be greater near the edges of the *Thalassia* patches. If this were true it might mean recruitment from outside sources is greater than the reproductive rates of members already in the patch.

There does not seem to be any aggression among anemones. Several times in the field I found two or three closely spaced individuals, and in the holding tanks I often found over five *Bunodeopsis* on the same leaf and never observed any aggressive interactions.

I would like to see the herbivory assays done if someone could work out the methods, perhaps with models of *Bunodeopsis*. I noticed that many of the anemones were at the tip of their leaves because it had been grazed

down to the anemone, but the anemone itself had not been bitten. This would suggest the fish are actively avoiding parts of blades with *Bunodeopsis*.

My findings on the diel activity cycle confirms the field work done by Sebens and DeRiemer. *Bunodeopsis* have the majority of their zooxanthellae located in the columnar vesicles. Their calculations show that with such an arrangement of zooxanthellae this cycle (contracted during the day, expanded at night) serves to conserve energy and limiting nutrients, such as nitrogen. Further studies could try artificially manipulating the day/night exposure to see if this behavior is a response to light or is based on an internal clock. Interestingly, the diel activity cycle was not held to exactly by all anemones. An estimated 10-20% of the anemones in the holding tank were extended during the day. It seemed that these were mostly the larger individuals. In the field this trend was reduced to only a handful of anemones. Perhaps the attenuated light levels in the lab made photosynthesis alone an insufficient source of energy and the anemones were attempting to supplement this with captured prey.

My results of *Bunodeopsis* response to *H. carunculata* predation did not support a long distance chemical cue as a warning system for the anemones. One might be tempted to say the data suggest a trend with three responses being obtained for the *Bunodeopsis* being eaten and the *Bunodeopsis* extract treatments; however, all but one of all responses obtained for this particular experiment occurred on a single night. Thus, I would dismiss these responses as anemone rest-

lessness on one night. The results of the contact cue experiments do support a warning mechanism based on tactile cues and, perhaps, contact chemical cues. Although the results do not show a significant difference between the number of responses for touching with a Q-tip dipped in fireworm homogenate and touching with a piece of fireworm, there was a large difference in the anemones' behavior before moving away. Upon first encountering the piece of fireworm, all of the tentacles immediately concentrated between the fireworm and the anemone and those tentacles near the sides began to repeatedly hit the fireworm. This is in contrast to the Q-tip, where only one of the anemones attacked it so violently. Perhaps the violent attacks are a response to being pierced with fireworm bristles, and on the particular Q-tip which was attacked a bristle happened to poke the anemone. Other anemones who encountered the fireworm homogenate just reached out to touch the Q-tip and seemed to be feeding on the particles imbedded in the cotton. In one case I saw the anemone pull a large chunk of flesh off the Q-tip. Why then the anemones would move away from a source of food is unknown. An interesting extension of this part of the experiment would be to see if the anemones respond to being touched with a Q-tip covered with needle-like spines in the same way they responded to fireworm head. It could be that anemones respond to their cell walls being pierced.

Predation observations in the holding tank supported the above findings. There was no mass release when the seven fireworms entered the tank, as one would expect if long dis-

tance chemical cues were at work, and several times a fireworm passed within less than a centimeter of a *Bunodeopsis* with no response by the anemone. When the fireworm did find a *Bunodeopsis*, the anemone's response was similar to those caused by a piece of fireworm in the experimental trials: the anemone would interpose its tentacles between its columnar vesicles and the fireworm and release its hold on the substrate. Escape seemed a function of anemone size and position in the tank. Larger anemones seemed more successful at keeping the fireworm's mouth away from the column and columnar vesicles, and anemones higher up in the tank seemed to be able to fall away to safety while those near the substrate were usually pinned down and eaten. One anemone escaping from or being eaten by a fireworm did not seem to affect other anemones' behavior, supporting the results that there is no anemone warning pheromone.

If the fireworms are commonly passing within centimeters of anemones without zeroing in on them, sitting still might be the most advantageous strategy. If the anemone were to let go when a fireworm is sweeping its head back and forth a few centimeters below it, the anemone would very likely bump into the fireworm. However, once the fireworm has zeroed in on an anemone, leaving should increase fitness. Thus, imprecise fireworm prey homing abilities could lead to selection for such behavior.

When contracted these anemones do not respond at all to external stimulus. A contracted anemone would not respond to a fireworm until the fireworm had begun eating it, and similarly it would not extend its

tentacles to feed even though brine shrimp were swimming throughout its columnar vesicles. Extended anemones reacted normally to both stimuli. This, and the observation that an anemone contracted except for the tip of one tentacle, fully extended all tentacles when I injected brine shrimp at it, suggests that only the tentacles contain receptors for predator and prey stimuli.

The results of the flow experiments were particularly interesting. The original question was if the anemones are found on the tips of the *Thalassia* simply because that is where a pelagic organism will settle first. The first flow experiments, "top flow", suggested that anemones were active in choosing a microhabitat. If the anemones settled on the first flat substrate they encountered, one would expect the anemones to be clumped near the bottom of the bucket since they started there. However, they all ended up clumped near the top. This distribution could have simply been a response to water flow since for the "top flow" trials the flow of water was mostly across the top of the bucket, so I designed the second experiment, "bottom flow," where the anemones were subjected to two opposing tugs: Phototaxis would pull them upwards, while positioning themselves in the greatest flow would pull them downwards. The result was a random distribution. Since the "top flow" experiment showed that the anemones do move from their initial points of contact, I believe it is safe to assume the random distribution was not a function of their random positions in the water column at the start of the experiment.

In nature the areas of highest flow and greatest light both exist at the tips of the *Thalassia*. This experiment suggests there is a consideration for both: high light would provide greater photosynthetic activity, while high flow would presumably result in greater zooplankton capture. This plus the fact that anemones positioned higher off the substrate have a better chance of avoiding fireworm predation, make the tips of the *Thalassia* leaves the best choice of microhabitat. Further work should attempt to sepa-

rate out the importance of these three variables in microhabitat selection.

LITERATURE CITED

- Humann, P. 1992. *Reef creature identification*. New World Publications, Inc.
- Sebens, K.P. and K. DeRiemer: Diel cycles of expansion and contraction in coral reef anthozoans; *Marine Biology* 43: 247-256 (1977).