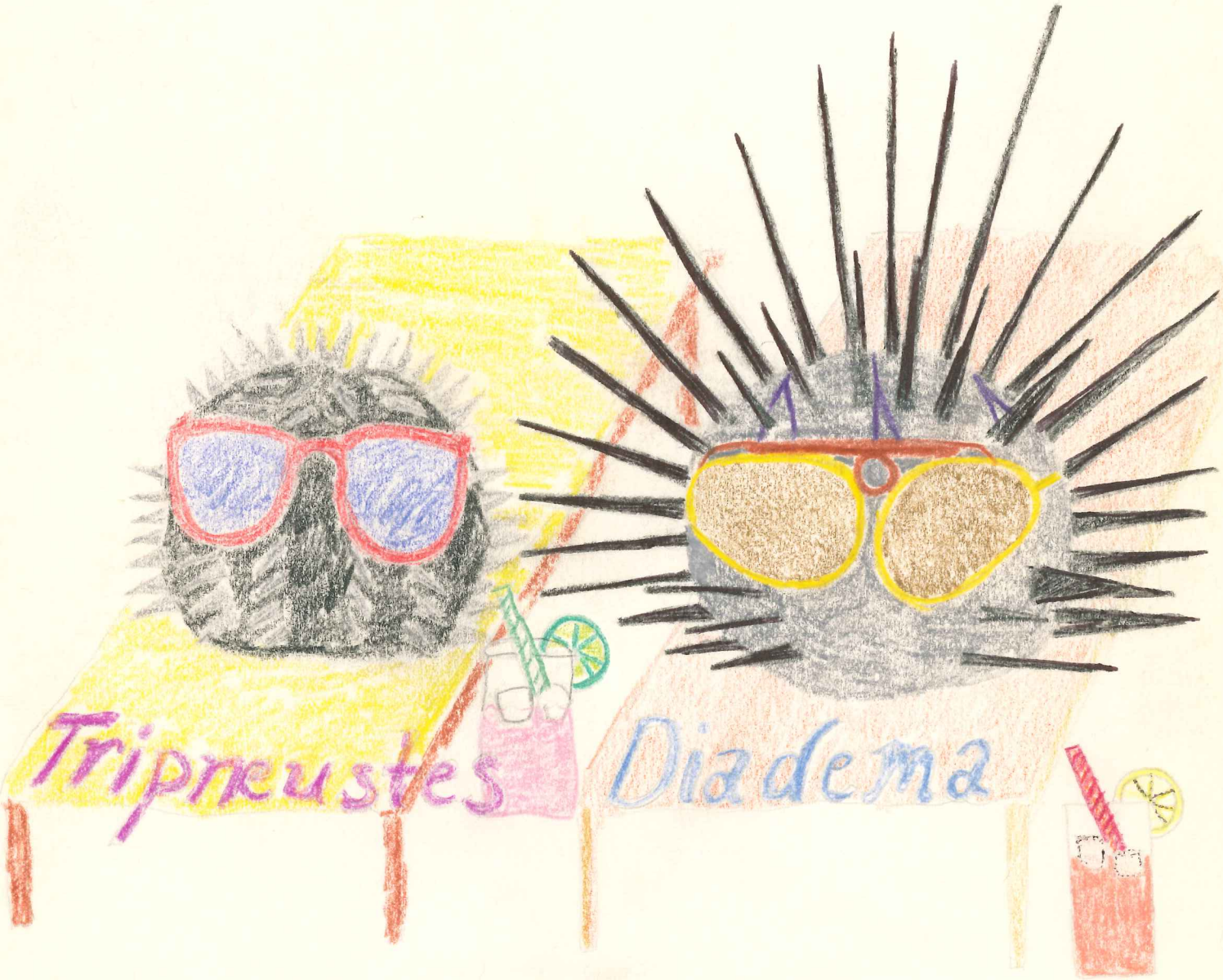
Tripneustes ventricosa and his partner Diadema antillarum on premium Lobophora variegata grazing.

(A study by Andrea Dols and Liz Walter on behalf of the Bartles and James Premium Wine Cooler Co.)



Biology FSP
February 26 - March 5, 1989
Discovery Bay Marine Lab, Jamaica

ABSTRACT (LW)

Sea urchins are known to exert strong grazing pressure on the algal populations on coral reefs. Through their grazing, they regulate the abundance of algae growing on these reefs. In our experiment, we examined the density of the sea urchins *Diadema antillarum Philippi* and *Tripneustes ventricosa* on the fore reef and how these densities related to the percent cover of the brown macro algae *Lobophora variegata*. As expected, fore reef densities of *T. ventricosa* were very low, as were *D. antillarum* abundances when compared with pre die-off densities. However, we still found a trend where densities of *D. antillarum* sharply decreased with depth, while the percent

cover of Lobophora variegata increases with depth. From grazing experiments, we found that D. antillarum grazing was significantly greater during the night than during the day. Tripneustes ventricosa showed no significant differences between day/night grazing. When given a choice, both T. ventricosa and D. antillarum preferred L. variegata over Thalassia. Overall, T. ventricosa grazed more heavily on L. variegata and Thalassia testudinum than did D. antillarum

INTRODUCTION (AD):

Grazing is considered the main factor in structuring coral reef algal communities (de Ruyter van Steveninck and Breeman, 1987). In fact, grazing is of great importance to the entire coral reef community. An undergrazing of algae would cause degradation of the coral reef through the resulting unregulated growth of fleshy and filamentous algae causing the death of crustose corallines covered by them and the death of corals by the entrapment of sediments (Van den Hoek, et. al, 1975). ~~Furthermore~~ More directly, grazing plays an important role in controlling the standing-crop of primary producers on coral reefs (Barnes, 1983). Therefore, grazers play an integral role in the coral reef system.

Sea urchins are grazers on benthic algae and coral tissue as well as a major bioeroder of hard substrata (Liddell and Ohlhorst, 1986).

Most tropical sea urchins appear to be generalist feeders which consume algae, seagrass, and even invertebrates. However, Sammarco 1972 and Ogden 1976 concluded that urchins may indeed favor certain macroalgae species (Barnes, 1983).

There are many various species of urchins found on or near coral reefs. Diadema antillarum Philippi, the long-spined black sea urchins, are normally ubiquitous in shallow Caribbean reefs. Their grazing exerts an important control on benthic algal populations (Liddell and Ohlhorst, 1986). In areas like Discovery Bay, Jamaica, that are heavily fished and undergoing recovery from hurricanes, there is increased influence on Diadema antillarum's regulatory role of algal populations. Tripneustes ventricosus, commonly referred to as the salt-and-pepper urchin, is another abundant urchin on coral reefs. Unlike the ubiquitous D. antillarum,

T. ventricosus populations are concentrated in the back reef. They also appear in small numbers on the fore reef, but do not appear to have as great an impact as D. antillarum.

At Discovery Bay, Jamaica, D. antillarum were was essentially eliminated from fore reef sites during August 1983 by an unidentified waterborn pathogen. With this great decline in D. antillarum, there was no substantial increase in other echinoid species' abundances. Algal biomass and cover did increase however (Hughes, et. al, 1987), and many studies were performed on those dominant macroalgae commonly encountered on the fore reef, yet absent on the back reef. Lobophora variegata is one such foliose brown macroalga which exhibits a distinct gradient distribution due to increasing grazing pressure with decreasing depth (de Ruyter van Steveninck and Breeman, 1987). In fact, by studying

post die-off systems, de Ruyter van Steveninck and Breeman (1987) found that herbivore damage to *L. variegata* was positively correlated with *D. antillarum* densities and inversely related to *L. variegata* cover.

In our study, we examined the current fore reef abundances and grazing patterns of *D. antillarum* and *T. ventricosus*. We formulated the following hypotheses:

i) Urchin densities decrease sharply with increasing depth, while *L. variegata* cover follows an opposite pattern.
ii) Fore reef densities of *T. ventricosus* are expected to be low, as are *D. antillarum* abundances in comparison with pre-die-off densities.
iii) Both urchin species exhibit greater nocturnal grazing than diurnal grazing.
iv) *D. antillarum* grazes more heavily than *T. ventricosus* on *L. variegata*.
v) Both species prefer *Thalassia testudinum* over *L. variegata* as a food source.

Methods (LW)

All manipulations and measurements were taken at Discovery Bay, Jamaica, West Indies. (18°30'N: 77°20'W) from February 26, 1989 through March 5, 1989. The weather remained consistent for the entire study period, with the mornings being warm and sunny with generally calm waters, and breezy afternoons characterized by choppy surface waters.

URCHIN DISTRIBUTION AND ABUNDANCE

Density of *Diadema antillarum* and *Tripneustes ventricosa* were made at 09:00 on 28 February, 1 March, and 3 March. at Discovery Bay on the West Fore Reef due South of Mooring 1. We counted *D. antillarum* and *T. ventricosa* in 10, 1*10m strips at depths of 10, 20

and 30 feet. To determine the density of the normally noctural, benthic D. antillarum, special care was taken to look between coral crevices and underneath ledges.

LOBOPHORA VARIEGATA DENSITY (McBride and Gangadharan, 1989)

Densities of L. variegata were determined by randomly selecting 6 sites at depths of 10, 20 and 30 feet. Measurements of percent cover of L. variegata were recorded by placing a grid over the substratum and counting the number of squares in which L. variegata occurred. The 6 sites were then averaged to determine the mean percent cover of L. variegata for each depth. This data was used in conjunction with our urchin density findings.

GRAZING EXPERIMENTS

For the grazing manipulations, we constructed ~~0.5 meter~~ cages of quarter inch wire mesh. (Dimensions, 0.5 m * 0.5m * 0.5m). These dimensions were chosen because of previous experiments indicating the *D. antillarum* foraging of aproximately 0.5-1.0 m^2 (Carpender, 1986). These cages were secured to the sandy substrate in 20 feet of water in the Blue Hole of Discovery Bay by attaching them to concrete blocks.

unclear! per unit time?

The *L. variegata* used in our experiment was collected on the rubble from the West Fore Reef at Mooring One at a depth of 30 feet. All coral fragments with *L. variegata* were brought to the lab where

sketches were drawn to document the amount and location of L. variegata as well as any previous herbivory. Also, all other macro algae was removed by scraping with a razor blade. This was to ensure that the sea urchins would be grazing only on the desired macroalgae. We then tagged the coral fragment with the cage number and date. Care was taken to equalize the amount of L. variegata among cages as well as to standardize the amount of L. variegata throughout all 8 trials. We determined the amount of grazing by comparing the number of bite marks before we placed the L. variegatus in the enclosures and after it had been left in the cages for 12 hours. We defined 1 bite mark as being a ragged edge approximately 1cm long.

D. antillarum and T. ventricosa for the cage experiments were collected daily (3/1- 3/5) from the West Back Reef at approximately 14:30. The

D. antillarum ~~were~~ was found in crevices in the coral heads, while T. ventricosa ~~were~~ was found in the Thalassia testudinum beds approximately 25 Meters from the reef crest.

At 17:30 on 1 March, ~~we placed~~ using SCUBA, we placed the tagged L. variegata in the center of each of the 8 cages. Next, we introduced one D. antillarum per cage to enclosures 1 through 4 and one T. ventricosa per cage to enclosures 5 through 8. We returned at 06:30 the following morning to collect the L. variegata. At the same time, we placed the L. variegata for the daytime experiment in the cages. This schedule of removal and replacement of L. variegata was chosen to coincide with the sunrise and sunset in order to

establish any nocturnal or diurnal grazing patterns.

D. antillarum and T. ventricosa were replaced at 17:30 after the completion of 1 night/day cycle. (24 hours). We then placed the sea urchin from each cage in a marked bag and brought them to the lab to record the weight of each individual. After this, they were returned to their respective habitats.

On the second day of the experiment (3/2 at 17:30), we modified the methods to ensure more accurate, quantifiable results. First, we introduced 3 ungrazed blades of 12 centimeter long T. testudinum into each enclosure by using a clothes pin and anchoring it into the sandy substrate. Introduction of the T. testudinum was intended

to be a control to ensure that both D. antillarum and T. ventricosa were grazing and also to determine if there was a choice preference between T. testudinum and L. variegata. Secondly, we introduced 2 individuals of the same species into each cage. This ensured an amount of grazing that we could accurately quantify. Other than these two modifications, we followed the same procedures and schedules for the introduction and removal of D. antillarum, T. ventricosa, L. variegata, and T. testudinum.

Upon the completion of the 8 trials, cages and cement blocks were delicately removed and safely stored for future experimentation by Dartmouth Biology FSP Students

Is this difference significant?

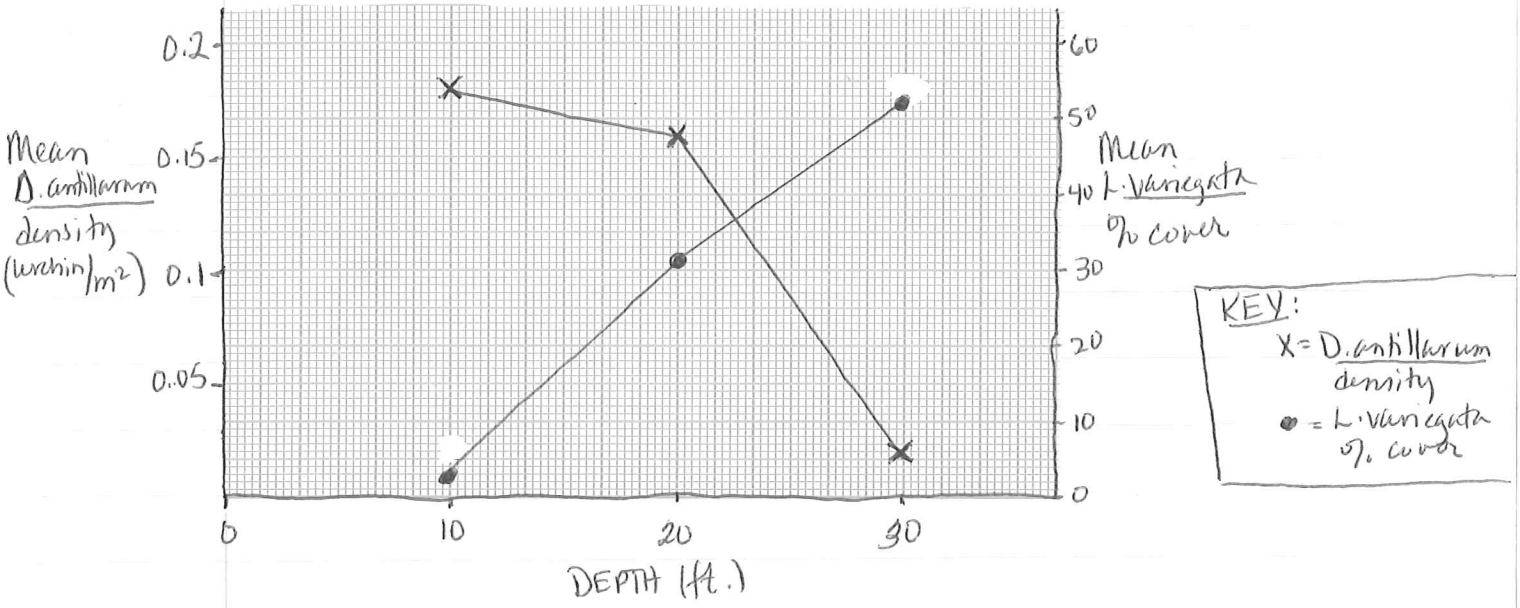
RESULTS (AD):

Urchin Distribution and Abundance

In our urchin census on the fore reef, we found that D. antillarum density decreased with increasing depth: averaging 0.18/m² at 10ft, 0.16/m² at 20ft, and 0.02/m² at 30ft.. There was no such trend in T. ventricosus distribution, as we found only one T. ventricosus in the 300 m² surveyed. Therefore, they are not as common on the fore reef as D. antillarum.

In a study by Gangadharan and McBride (1989) on the same fore reef, L. variegata percent cover was found to follow an opposite pattern of the D. antillarum abundance. That is, percent cover of L. variegata increased greatly as depth increased: averaging 0.24% cover at 10ft., 31.67% cover at 20ft., and 51.90% cover at 30ft.. Figure 1 illustrates the inverse trends of D. antillarum density and L. variegata percent cover with depth.

FIGURE 1:
Mean D. antillarum density and L. variegata % cover versus depth



Therefore, D. antillarum densities followed the expected trend with depth.

When comparing our findings with densities found by T. P. Hughes et al. at approximately the same depth and location before and soon after the die-off, our D. antillarum densities are still relatively low (Table 1).

TABLE 1:
D. antillarum densities at 10.5 m depth in Mooring 1, Discovery Bay, Jamaica

	1978	1983	1986	1989*
Density (urchin/m²)	12.2 ± 4.3	0	0	0.02 ± 0.01

* our results taken at 30 ft ≈ 9.2 m depth.

Urchin Grazing Experiments

There was a significant difference in *D. antillarum* grazing on *L. variegata* during the day (06:30 - 17:30) and night (17:30 - 06:30). The nighttime grazing ($\bar{x}_N = 2.3 \pm 2.1$ bites/urchin) was significantly greater than that during the daytime ($\bar{x}_D = 0.66 \pm 0.85$ bites/urchin) (Student's t-test, $t = 3.0143$, $p < 0.01$, $df = 15$). On the other hand, *T. ventricosus* did not exhibit a significant difference between their grazing during the day ($\bar{x}_D = 5.4 \pm 2.2$ bites/urchin) and the night ($\bar{x}_N = 4.6 \pm 2.7$ bites/urchin) on *L. variegata* (Student's t-test, $t = 0.8998$, $p > 0.4$, $df = 15$). Once again using a Student's t-test, we compared the number of bites on *L. variegata* per urchin between the two species during both a day and night cycle. We found a very significant difference, with *T. ventricosus* averaging 5.0 ± 2.5 bites/urchin during the 24 hour cycle while

D. antillarum averaged only 1.5 ± 1.8 bites/urchin (Student's t-test, $t = -6.573$, $p < 0.001$, $df = 31$).

We also tested for food preferences within each urchin species. We examined whether L. variegata or T. testudinum was eaten more often when presented to the urchins during the same time period. There was a significant difference between the grazing on L. variegata and T. testudinum by D. antillarum (Chi-Square Analysis, $\chi^2 = 13.94$, $p < 0.001$, $df = 1$). 75% (18/24) of the L. variegata presented showed signs of herbivory, whereas only 8.3% (2/24) of the T. testudinum samples presented did. Furthermore, the two times the T. testudinum was eaten, it was in miniscule amounts (a few, unmeasurable bites) compared to the amounts of L. variegata eaten (1-12 bites). T. ventricosus

also exhibited a significantly different grazing frequency on *L. variegata* and *T. testudinum* (Chi-Square Analysis, $\chi^2 = 14.27$, $p < 0.001$, $df = 1$). Everytime *L. variegata* was presented to *T. ventricosus*, it incurred at least one bite and the range of herbivory was 1-20 bites per trial. *T. testudinum* was also grazed during a majority of the *T. ventricosus* trials (54.2% or 13/24), but less frequently than the *L. variegata*. With regard to *T. testudinum*, *T. ventricosus* grazed more heavily on it than did *D. antillarum*, and even occassionally completely removed the presented blades.

By combining the bites on *L. variegata* / *D. antillarum* for the day and night trials of the sixteen experimental replicates, we calculated the mean *D. antillarum* herbivory rate as 2.9 ± 2.3 bites/urchin/24 hours

Using this mean grazing rate and the previously established D. antillarum densities, we calculated the effect of D. antillarum on L. variegata at the three examined depths (Table 2).

TABLE 2:

Diadema antillarum grazing effects on Lobophora variegata

Depth (ft)	D. antillarum density ($\frac{urchin}{m^2}$)	Mean herbivory rate (bites/urchin/24hrs)	Daily grazing on L. variegata (bites/m^2/24hrs)
10	0.18	2.94	0.53
20	0.16	2.94	0.47
30	0.02	2.94	0.06

Therefore, the daily grazing effect on L. variegata by D. antillarum increases with a decrease in depth.

During experimentation, a few interesting behavioral observations were made. D. antillarum were most often found grouped together in the corner of their cages, while T. ventricosus seemed to separate to different parts of the cages, usually attaching

to the cage side or top. Also, T. ventricosus were sometimes found in physical contact with the L. variegata covered coral, and occassionally they were found with either L. variegata leaves or the clothespin of T. testudinum blades on their backs.

Discussion (LW + AD)

Our results indicate that the density of D. antillarum decreases with depth while the density of L. variegata increases with depth. The inverse relationship between L. variegata and D. antillarum over depth agrees with pre die-off studies. This continuing opposite pattern suggests that D. antillarum grazing pressure may have affected the L. variegata distribution on the West Fore Reef. We found the daily grazing affect of D. antillarum dramatically decreases with increasing depth, especially between 20 and 30 feet. However, the grazing rates were extremely small.

When comparing our low D. antillarum densities with previous densities before and soon after the die-off,

we can conclude the D. antillarum population was devestated by the die-off, but may now be starting to slowly recolonize the fore reef. The long term effects of the reduced D. antillarum population and the possible algal increase, will depend on the ability of D. antillarum to reestablish itself on the fore reef.

During our grazing experiments, we found that D. antillarum did graze on L. variegata and did so in the expected nocturnal pattern, however, the observed grazing occurred in small quantities. This finding, observed by de Ruyter van Stevenick and Breiman in 1987, predicted that L. vanegata was relatively insusceptable to grazing due to the presence of

secondary metabolites and certain morphological characteristics such as a thick leathery form. These features act as defense mechanisms to guard against herbivory. Therefore, considering these two findings, it will take high densities of D. antillarum to impact the L. variegata population. Thus, not until D. antillarum reaches pre die-off status will it's regulatory role in algal density resume.

Discuss: Can urchins explain the absence of Lobophora at shallow depths?

Although T. ventricosa ~~are~~ is not as important a grazer on the fore reef, as speculated by the very low abundances, in our cage experiments, they appeared to be heavier grazers on both L. variegata and T. testudinum. The heavier grazing may be

explained by two behaviors not associated with feeding. T. ventricosa seemed more active and therefore may have encountered the food items more frequently than D. antillarum. Secondly, T. ventricosa exhibits a covering behavior where ~~they~~ it uses debris such as T. testudinum blades and L. variegata leaves in a hypothesized attempt to become more cryptic or less succeptable to ultraviolet light. Therefore, we cannot be sure that removed T. testudinum or L. variegata was actually ingested, but reguardless of the purpose, T. testudinum or L. variegata is being removed from the substratum.

Although T. testudinum is the preferred food

of both T. ventricosa and D. antillarum (Ogden, 1976), in our experiments, we found that both species grazed most heavily on L. variegata. This finding may be explained by the presentation of T. testudinium as well as the higher encounter rate of L. variegatus. The T. testudinum was presented on clothes pins where an inch of wood separated the substrate from the exposed T. testudinum blade. This may have effectively deterred grazing due to the presumed absence of edible material. On the other hand, L. variegata was presented on a low-lying natural substrate which may have facillitated the investigation and eventual ingestion of the L. variegata. Therefore, the difference

between the presentation of the two food items may have introduced an artificial preference. Furthermore, the area on which the L. variegata was presented was much greater than the area in which the T. testudinum was presented. Therefore, the encounter ~~et~~ rate may have been higher for L. variegata than T. testudinum. Again, it is possible that this difference may have caused the greater grazing on L. variegata.

While the methods we used were adequate, improvements could be made to increase the accuracy, reproducibility and validity of our findings. The most crucial aspect of this study, yet the most difficult to quantitatively determine,

was the amount of herbivory on the L. variegata.

We feel a more accurate assessment could be made in two ways. First, photographic documentation before and after placement in the enclosures would allow for quick and precise herbivory evaluations.

Secondly, one sample of L. variegata could remain in the enclosure for a period of 4-5 days, while maintaining the same 24 hour schedule of changing the sea urchins. In this way, cummulative herbivory would allow for a large enough difference between initial and final percent cover of L. variegata. That photographs could be overlayed and the amount of L. variegata removed from the substrate

accurately measured.

The next phase of this study requires that these manipulations be conducted on the fore reef with T. ventricosa collected from the backreef and D. antillarium collected from the fore reef. The enclosures should all have a hard bottom, so that organisms colonizing the underlying substrate do not interfere with the normal behavior of the sea urchins. Also, the sea urchins should be allowed a period of acclimitization before experimentation begins (4-5 hours). In this study, we also recommend that a choice preference between the commonly occurring fore reef algae - such as Halimeda, Dictyota

and Lobophora be studied as well as the other factors examined in our experiment. Finally, we recommend that all sea urchins be divided into weight classes so that data about differences in preferrence with weight and in the amount consumed with weight may be examined.

Possibilities for future studies include investigating why L. variegata is not found on the back reef. To do this, L. variegata could be transplanted onto the back reef. ~~and see what occurs.~~ Is it grazed to exclusion by T. ventricosa or other reef herbivores, or is it physiologically unable to survive in the back reef environment? Logically,

T. ventricosa could be moved to areas on the fore reef with all other sea urchins removed, with only D. antillarum present, and with all sea urchins present. In this way, the interactions, the physical limitations and the natural occurences of these influential reef grazers may be better understood.

A-

Liz/Andrea:

A good study and write-up. In the discussion, you should have used your bite/urchin/day data to consider the ability of urchins to exclude Lobophora from shallow depths.

John

References (AD):

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