

BEST DRESSED TEST: A STUDY OF THE COVERING BEHAVIOR OF THE COLLECTOR URCHIN *TRIPNEUSTES GRATILLA*

MORGAN A. ZIEGENHORN

Department of Integrative Biology, University of California, Berkeley, California 94704 USA

Abstract. Many sea urchin genera exhibit cryptic covering behaviors. One such behavior has been documented in the sea urchin *Tripneustes gratilla*, and previous studies have theorized that the behavior serves as protection from UV radiation. However, other hypotheses have been presented such as protection from predators or added weight to help *T. gratilla* resist strong currents. A field study was conducted to assess urchin cover in the natural habitat, and found that urchins partially underneath rocks cover more, and with more algae, than urchins totally underneath rocks. This result supported the hypothesis that urchin cover is related to light intensity. A series of 30 minute experimental trials were run on 10 individuals in bright and dim conditions. Individuals were given opaque and clear plastic, and percent cover of each was recorded. These tests were repeated once fifty percent of spines had been removed from the urchin. Urchins had a distinct preference for cover that best protects them from UV radiation. Spine loss did not affect urchin ability to cover, and urchins with removed spines still preferred opaque cover. General protection, rather than UV radiation, might be a motivation for covering for urchins that have lost spines. Individual preference was found to be statistically significant, which has not been noted before in *T. gratilla*. However, no knowledge of why individual preference exists was found. This study adds to the body of knowledge on sea urchin covering behavior by demonstrating more fully the extent of the relationship between light intensity and cover in *T. gratilla* urchins. It also provides previously unknown knowledge about sea urchin resilience after spine loss and individual behavior. Understanding urchin covering behavior more aids in the understanding of the interconnectivity of cryptic behavior across species.

Key words: cryptic; coral reefs; sea urchin; Moorea; individual behavior; spine loss, UV radiation

INTRODUCTION

Crypsis, the process of avoiding observation or detection by other species, is a widely seen phenomenon in the animal kingdom (Stevens and Merilaita 2009). Many organisms including various mollusks, flies, and crabs exhibit cryptic behaviors such as camouflage and mimicry (Portmann 1959). Crypsis can serve many functions: to protect species from predation, harmful environmental factors, or conceal species from prey while hunting.

In sea urchins (class Echinoidea), cryptic behavior involves using tube feet in conjunction with spines to hoist and secure materials to the aboral surface (Adams 2001), or, in the case of floating materials, seizing objects directly with tube feet (Millot 1956). Though this behavior is exhibited by several different urchin species, it remains a poorly understood phenomenon (Dumont *et al.* 2007), and reasons for covering are thought to differ

between species. Some species, like *Stronglyocentrotus drobachiensis*, cover to a higher degree when exposed to wave surges (Dumont *et al.* 2007), while others such as *Evichinus chloroticus* cover as an aid in food capture (Dix 1969).

In the case of the urchin *Tripneustes gratilla*, commonly known as the “collector urchin”, many possible hypotheses for covering behavior have been explored, including protection from predators, protection from light exposure, and protection from strong currents (Park and Cruz 1994). In several studies, a correlation between light intensity and urchin cover was noted (Dumont *et al.* 2007, Park and Cruz 1994), and it has been postulated that *Tripneustes* covering behavior is a form of protection from the sun (Kehas *et al.* 2004). This conclusion is bolstered by the urchin’s ability to sense and respond to light via photo-sensitive tube feet (Millot 1975).

Prior studies have concluded that collector urchins relinquish cover in lower light conditions because darkness is a trigger for nocturnal foraging, during which the urchins are more mobile and weighing less is more energetically favorable (Park and Cruz 1994). However, little work has focused on preference between covering materials with regards to light intensity. If sunlight is the main factor that influences covering behavior, urchins should prefer materials that best shield them from light. A consideration of cover preferences would therefore provide insight into the role of light, leading to a better understanding of how and why this behavior evolved.

Though the mechanism of attaching materials to the test is known and urchin spines are considered an important part of this process (Adams 2001), no previous research has examined how covering ability is affected or damaged by spine loss. Spine loss is a common phenomenon in urchins, and can occur at very high levels when it is a symptom of disease. One such disease that remains undefined has been observed in Hawaiian reefs (Abercrombie and Aila 2014). However, spine breakage and regrowth remains undocumented in *T. gratilla*, though it has been thoroughly detailed for other urchin species, and the mechanism of regrowth across echinoderms is well understood (Dubois and Ameys 2001). Spine breakage most likely occurs from urchins being tossed by strong currents or from non-lethal interactions with predators such as pufferfish and humans that try to pry them out from under rocks. Understanding how spine loss affects important behaviors in urchins such as covering will aid in determining the likelihood that injured urchins are able to survive in their environment until their spines regrow.

Beyond a need to explore preferences for different covering materials under different light regimes, no work has explored individual variation in covering behavior in urchins. Outside of primates, research in animal personalities is a relatively new field (Sih 2004), one that often involves testing the repeatability of various behaviors. In terms of urchin covering behavior, urchin 'personalities' might lead certain individuals to cover themselves more or less often, regardless of stimuli such as light, type of cover available, or presence of predators.

The overall goal of this study was to characterize *T. gratilla* covering behavior as related to habitat (underneath rocks vs.

partially exposed), light intensity, and spine loss, as well as variation in covering behavior among individuals. Specifically, I addressed the following four questions: (1) how is covering behavior in the urchin *T. gratilla* represented in the field, (2) to what extent is covering behavior affected by differing light conditions, (3) how is covering behavior affected by spine loss, and (4) do individuals change their covering behavior in different conditions? I conducted a field study to understand the influence of habitat and a lab study to understand the influence of cover type, light conditions, spine loss, and individual preference. I predicted that covering behavior in the field would be linked to location, as urchins already protected from light by being underneath coral would not need to cover themselves. I did not expect to see a preference for particular covering materials. For the lab study, I predicted that there would be a statistically significant preference for opaque covering materials in bright light conditions, but not in dim light conditions where danger from light exposure was not a serious threat. I did not expect this trend to change once spines were removed, though I did expect to see less covering behavior as spines were considered important for securing materials to the urchin's test. In terms of personalities, I predicted that individuals would not show significant variation between trials pre and post spine removal, due to the effects of individual covering preferences.

METHODS

Field study

Field surveys took place in the channel between the motus Tiahura and Fareone on the northwest side of the island Moorea, French Polynesia (17°29'44"S, 149°49'61"W) once a week from October 12, 2015 to November 10, 2015 (Fig. 1). A 30 meter by 50 meter site was surveyed by choosing 15 coral rocks on which *T. gratilla* urchins were seen. Each rock was thoroughly examined and the following data collected: location of urchin on rock (partially or totally underneath), percent cover of algae, and percent cover of coral rock/shells. Possible replication of some rocks in subsequent weeks was considered insignificant due to the assumption that, since urchins are not sessile, surveying the same rocks would not necessarily mean surveying the same urchins.

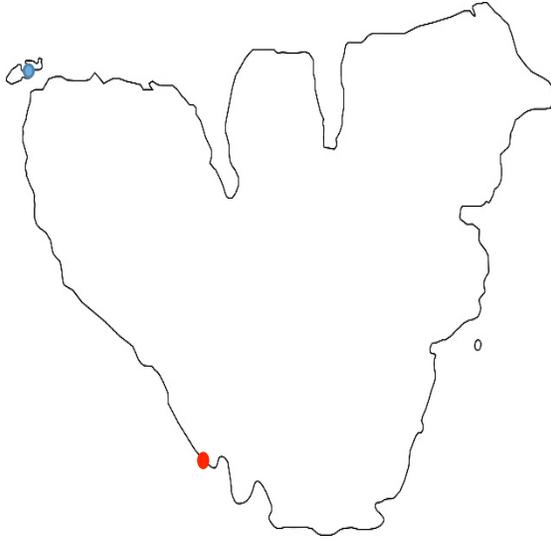


FIG 1. Map of Moorea, French Polynesia with field site between the motus Tiahura and Fareone (blue dot) and collection spot at Haapiti (red dot) both noted.

Experimental procedure

Experiments were conducted at Gump Station in Cooks Bay, Moorea, French Polynesia from October 16, 2015 to November 9, 2015. Ten *T. gratilla* urchins were collected from the reef at Haapiti on the southwestern side of Moorea (Fig 1). Urchins were kept in a tank with constant seawater flow from Cook's Bay and fed a variety of macroalgal species, most notably *Sargassum sp.* and *Turbinaria sp.* Urchins were distinguished from one another via morphological characteristics such as test size, tube feet coloration, and spine coloration. A detailed description was recorded for each urchin and was used from then on to distinguish urchins from one another.

The lab experiments involved testing urchins in two light conditions: bright sunlight and dim sunlight, and observing their covering behavior. For the bright light tests, two urchins were placed in one tub in direct sunlight with no available shade (Fig. 2). This tub was divided in half so urchins could not reach each other. All cover was removed from the urchins prior to each test, and they were presented with eight pieces of hard red plastic and eight pieces of hard clear plastic, all of similar sizes. Cover of each type of plastic was estimated every minute for ten minutes, and then every five minutes for a total of one half hour. After this, plastic cover was removed from the urchins and they were returned to the larger tank.

This test was repeated on the urchins in the dim light condition after a rest period of at least forty-five minutes. Water was replaced in the test tub when it began to heat, as higher temperatures stress the urchins and lead to unwanted spawning (personal observation). The tests were repeated five times on each urchin, with no urchin going through more than one cycle of tests (one bright and one dim) in one day. Individual urchins were tested every other day. To control for the fact that urchins going through a second test in



FIG 2. Photograph of urchin test setup in the bright light condition.

one day might be fatigued and cover less because of this, urchins were tested in dim light first some days and in bright light first on others.

Then, spines were removed from the urchin's test in order to test the effect of spine loss on covering behavior. Spines were clipped as close to the test as possible, and care was taken to not remove tube feet. Spines were removed over half the urchin in a semicircle that included both oral and aboral sides. Urchins were put through a dim light test of the same procedure as explained above directly after their spines were removed. Later on, they were again subjected to bright light conditions. These tests were done in accordance with the method above a total of five times for each urchin, with every other day being a rest day for the individuals tested on the day prior. After the conclusion of these tests, urchins were returned to the reef off of the Gump Station at Cook's Bay, where they were present (personal observation). Urchins were not returned to Haapiti because of a lack of available transport.

Statistical analysis

Three Kruskal-Wallis rank sum tests (Ambrose et. al 2002) were performed in R (R 2013) in order to test the significance of location on rock, cover type, and the combination of these two on total percentage cover in the field surveys. These tests were used in lieu of a two-way Analysis of Variance (ANOVA) because the field data did not fit the assumptions of normality required by parametric tests. A bar plot of percent cover of each cover type in each location was created using the ggplot2 package in R (Wickham 2009).

For the experimental data, a repeated measures ANOVA was conducted in R using the lme4 package (Bates et. al 2015) to test the significance of light condition and cover type on percentage cover of the urchins. Percent cover of each cover type was plotted against time in each light condition using the ggplot2 package in R (Wickham 2009). Data was averaged to provide one line for each cover type. The percent cover at the end of each test (thirty minutes) was compared in each of the two light conditions using an unpaired t-test (Ambrose et. al 2002) to determine if there was a significant difference between final percentage of red cover and clear cover in each light condition.

A series of Friedman rank sum tests (Ambrose et. al 2002) were run to test the significance of cover type, light condition, and the combination of the two on total percent cover of the urchins after 50% spine removal. The Friedman test was necessary because the data did not fit the requirements of the parametric equivalent. Individual was used as the blocking variable as each urchin was tested more than one time. Percent cover of each type was plotted against time for each light condition in R, using the ggplot2 package (Wickham 2009). Final percent cover of each cover type in each light condition was compared using a series of Wilcoxon rank sum tests (Ambrose et. al 2002), and final percent cover of the same cover type was compared for both light conditions to what was seen prior to spine loss using sign tests for related samples (Ambrose et. al 2002). To compare total percent cover of red and clear plastic before and after spine loss, a bar plot was created using the ggplot2 package in R (Wickham 2009).

Percent cover amongst individuals was compared using a one-way ANOVA (Ambrose et. al 2002) in R for the data

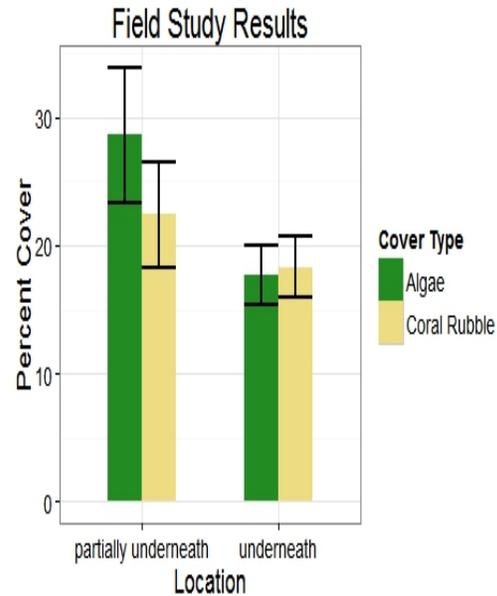


FIG 3. Percent cover of algae and coral rubble on urchins in found partially underneath or totally underneath rocks in the motu Tiahura reef.

taken before spine removal. Total percent cover for each individual was plotted before and after spine loss using the ggplot2 package in R (Wickham 2009). For the data taken after spine removal, another series of Kruskal-Wallis rank sum tests were used (Ambrose et. al 2002) to test for statistically significant differences in percent cover amongst individuals.

Phylogeny

The evolution of covering behavior in sea urchins was examined using an existing phylogeny of echinoderms (Littlewood and Smith 1995). Urchin genera from the phylogeny presented by Littlewood and Smith were searched on the internet both through images and articles to determine if they exhibited covering behaviors. This information was mapped onto the phylogeny using Mesquite (Maddison and Maddison 2015) by creating a character matrix of all genera included in the phylogeny in which covering behavior was given a value of one, and no covering behavior a value of zero. This character matrix was analyzed and plotted onto the given phylogeny using the "Parsimony Ancestral States" option within the "Trace Character History" function.

RESULTS

Field study

In the field, urchins partially underneath rocks covered themselves with algae and coral rubble more than urchins totally underneath rocks, with a difference of 25.6 percent cover partially underneath rocks versus 18.0 percent cover totally underneath rocks. This difference of 7.6 percent was statistically significant (Kruskal-Wallis rank sum test, chi-squared=10.571, $P < 0.01$). In terms of cover type, there was a minor overall preference of algae over coral rubble, but this was not statistically significant (Kruskal-Wallis rank sum test, chi-squared=0.086739, $P > 0.05$) (Fig. 3). However, urchins partially underneath rocks did prefer algal cover over coral rubble cover, with a total percentage of algal cover of 28.6 percent versus a total coral rubble cover of 22.4 percent, a difference of 6.2 percent (Fig. 3). This interactive effect between location and cover type was statistically significant (Kruskal-Wallis rank sum test, chi-squared = 12.476, $P < 0.01$).

Experimental study

In the bright light conditions, urchins collected more red cover over time than clear cover, with average percentage cover at the end of thirty minutes of 34.8 percent for red cover and 20.1 percent for clear cover (Fig. 4a). In the dim light conditions, the two cover types had nearly no difference, with an average percent cover of 19.3 percent for both cover types at the end of thirty minutes (Fig. 4b). The difference of 16.3 percent in total percent cover between the two light conditions was statistically significant (repeated measures ANOVA, $T_{27.241} = 2.509$, $P < 0.05$). The observed preference for red cover over clear cover across individuals (Fig. 5) was also statistically significant, and this preference was correlated with the bright light condition (repeated measures ANOVA, $T_{27.241} = 11.473$, $P < 0.0001$, $T_{27.241} = -8.610$, $P < 0.001$). The average percentage of red cover at the end of

thirty minutes in bright light conditions was 34.8 percent, while that of clear cover in the

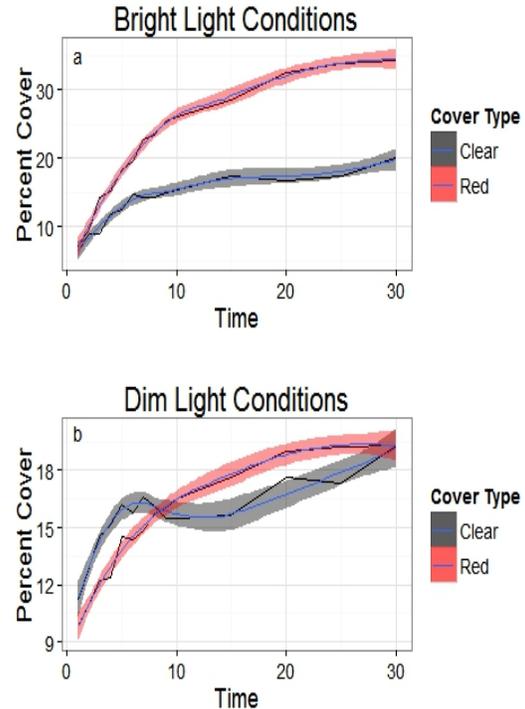


FIG 4. Average values of percent cover versus time in the bright light condition (4a) and the dim light condition (4b). 'Red' refers to opaque cover. Lines of best fit, calculated from the geom_smooth function in R as conditional means, are included in blue, and error margins are represented by shaded areas.

same condition was 20.1 percent (Fig. 4a), and this difference of 14.7 percent was statistically significant (Welch two sample t-test, $T_{76.241} = 4.1442$, $P < 0.0001$).

In the dim light conditions (Fig. 4b), the average percentage clear cover and red cover at the end of thirty minutes were both 19.3 percent, and so no statistically significant difference existed between them (Welch two sample t-test, $T_{17.416} = 0$, $P > 0.05$).

For the tests conducted post-spine removal, light condition no longer had a statistically significant effect on total percent cover, which was 16.1 percent in bright conditions and 17.4 percent in dim conditions at the end of thirty minutes, a difference of only 1.3 percent (Friedman rank sum test, chi-squared= 0.4, $P > 0.05$), but a statistically significant preference for red cover was still observed across individuals (Friedman rank sum test, chi-squared= 6.4, $P < 0.05$) (Fig. 5).

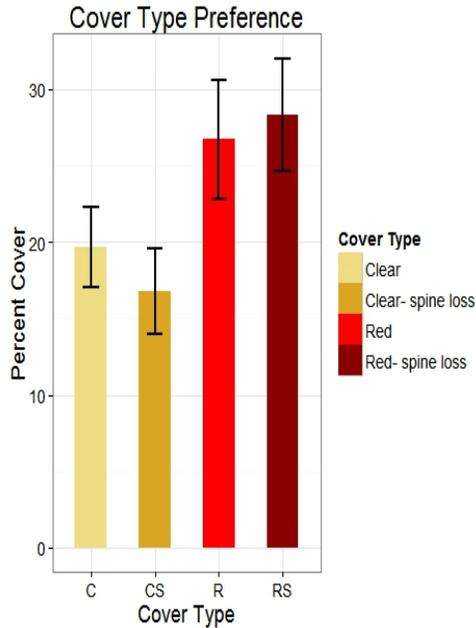


FIG 5. Average percent cover of both cover types comparing pre and post spine loss data. Both light conditions and all individuals were averaged into one column for each cover type. Cover percentages were taken at the end of the thirty -minute test period.

On average, percentage cover of red plastic increased after spines were removed, while percentage cover of clear plastic decreased (Fig. 5). However, these differences were slight (Fig. 5). The average red cover at the end of thirty minutes was 30.1 percent and for clear cover was 17.4 percent in the bright light condition (Fig. 6a). In the dim light condition, the average percentage red cover was 26.4 percent, and for clear cover was 16.0 percent, at the end of thirty minutes (Fig. 6b). These differences of 12.7 and 10.4 percent respectively were statistically significant in both light conditions (Wilcoxon rank sum tests, $W = 1535$, $P < 0.01$, $W = 1647$, $P < 0.01$). The difference between the average final red cover percentages pre and post spine loss in

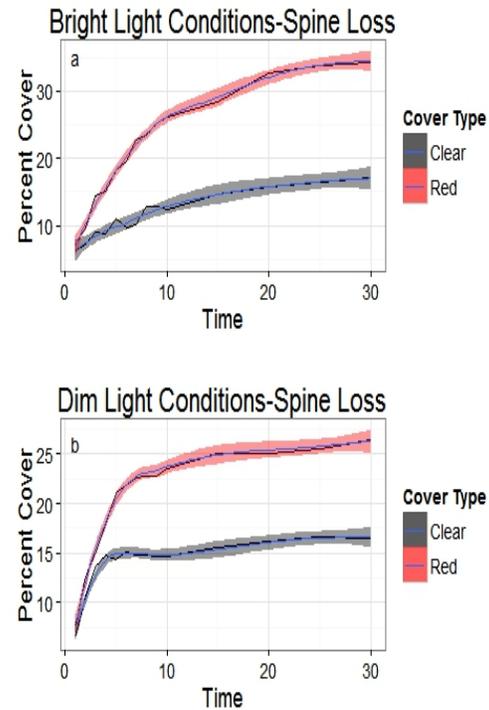


FIG 6. Average values of percent cover versus time in the bright light condition (6a) and the dim light condition (6b) after spines were removed. 'Red' refers to opaque cover. Lines of best fit, calculated from the geom_smooth function in R as conditional means, are included in blue, and error margins are represented by shaded areas.

the bright light condition was 4.7 percent and in the dim light condition was 7.1 percent. The difference between the average final clear cover percentages (pre and post spine loss) was 2.7 percent in the bright condition and 3.3 percent in the dim condition. However, none of the differences between slopes of the same type were compared to slopes before spines were removed (ie, comparison of red cover slope in the bright light conditions before and after spine loss) were statistically significant (sign tests, $T = 21$, $P > 0.05$, $T = 15$, $P > 0.05$, $T = 24$, $P > 0.05$, $T = 18$, $P > 0.05$).

A difference of 20.2 percent in total percent cover (cover of both red and clear plastic) existed between the individual which covered the most on average (31.1 percent cover) and the individual that covered the least on average (10.9 percent cover) prior to spine loss (Fig. 7). Differences in covering behavior were statistically significant amongst individuals (two-way ANOVA, $F = 23.009$, P

< 0.01). However, all individuals preferred red cover over clear cover (Fig. 5), and this covering preference was statistically significant (two-way ANOVA, $F_1 = 0.442$, $P > 0.05$).

Post spine removal, total percent cover varied by 16.5 percent between the most covered individual on average (29.0 percent cover) and least covered individual on average (12.5 percent cover), and the differences amongst individuals in terms of total percent cover were still statistically significant (Kruskal-Wallis rank sum test, chi-squared = 19.799, $P < 0.05$). For most individuals, average percent cover differed little before and after spine loss, though for one individual average percent noticeably decreased after spine loss (Fig 7). Individual preference for red cover was also still present, and statistically significant, after spines were removed (Kruskal-Wallis rank sum test, chi-squared = 49.354, $P < 0.001$) (Fig. 5).

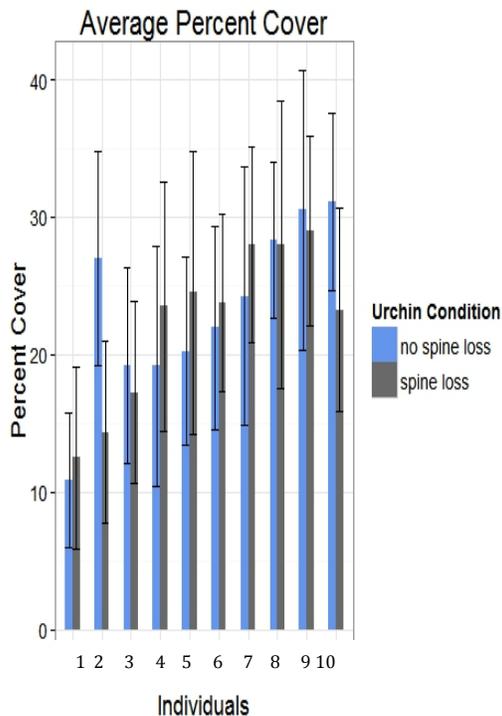


FIG 7. Average percent cover of individuals before and after spine loss. Averages were created by combining data points for both light conditions and cover types at the end of each thirty-minute test.

Phylogeny

Covering behavior was successfully mapped onto a pre-existing phylogeny courtesy of Littlewood and Smith (1995) (Appendix A). Covering behavior was found in the genera *Glyptocidaris* and *Stronglyocentrotus* (Zhao et. al 2014), *Temnopleurus* (Yoshida 1966), *Mespilia* (Yanagisawa 1972), *Salmacis* (Belleza et. al 2012), *Echinus* (Forster 1959), *Psammechinus* (Milligan 1915), *Paracentrotus* (Crook et. al 1999), *Sphaerechinus* (Unger and Lott 1993), and *Lytechinus* (Amato et. al 2008). The behavior evolved once, and was lost in the genera *Glyptocyphus* (Smith 1988), *Colobocentrotus* (Thet et. al 2004), and *Heliocidaris* (Pederson and Johnson 2006). The other genera included in the phylogeny did not exhibit covering behavior. Covering behavior was also found in other genera within the order Camarodonta that were not included in the phylogeny. These genera were *Pseudoboletia* (Ogden et. al 1989), *Toxopneustes* (James 2000), *Pseudechinus* (Dayton et. al 1977), and *Genocidaris* (Pawson and Pawson 2013).

DISCUSSION

The results of the field study indicated that urchins partially underneath rocks covered themselves more than urchins totally underneath rocks. This supported my hypothesis that urchins cover themselves to protect from sunlight, as urchins that are only partially under rocks are more vulnerable to light than urchins totally underneath rocks. However, this did not rule out other covering explanations such as camouflage from predators (Agatsuma 2001), or protection from strong currents (Park and Cruz 1994, James 2000).

A previous study of *T. gratilla* has suggested that urchin cover is random with respect to the environment (Park and Cruz 1994). However, in the present study a link was found between cover type and location in that partially exposed urchins had more algal cover than coral cover. This result refuted the hypothesis that urchins cover to weigh themselves down, as in this case heavier coral cover, which can be more closely held to the test, would be more advantageous than algae to urchins that are more exposed (James 2000). Algal cover preference also supported the hypothesis that urchins cover themselves as a way of carrying food (Lewis 1958). This result also refuted the idea that urchins cover

themselves as camouflage from predators, as heterogeneity of cover is a requirement for camouflage (Endler 1978). However, this study did not quantify whether coral rubble and algal cover were equally available in the environment, so it cannot be said what percentage of algae and what percentage of coral could have been considered a true random sample of the environment.

Urchins subjected to bright light did prefer cover that protected them from the sun, and in dim conditions had no significant preference in cover type. This result supported the hypothesis that urchin cover in *T. gratilla* urchins is a response to sunlight, an idea supported by previous literature (Lewis 1958, Moore 1966, Park and Cruz 1994). If covering behavior was mainly influenced by another factor, such as predators or currents, no significant difference should have been seen between the two light conditions.

Once spines were removed, urchin covering behavior was altered. Urchins no longer responded differently in the two light conditions, covering themselves to an equal extent regardless of bright or dim light. This result refuted the hypothesis that spines are a crucial part of urchin covering behavior; in fact, it appeared that only tube feet were necessary for successful covering (personal observation). Though urchin locomotion was impaired by spine loss it was not severe enough to limit their ability to find cover. The lack of difference between light conditions post-spine loss also refuted the idea that light is the only trigger for covering behavior, as if this were the case no difference would have been seen between the data taken pre and post spine removal. As urchins covered themselves to an equal extent in dim light post-spine loss, it seems covering is also related to general protection. Once spines were removed, urchins sensed that they were more vulnerable to their environment, and possibly predation, and so covered more to protect themselves from threats. This result supported the hypothesis that urchin cover in some species is related to protection from predators (Zhao et. al 2014).

It was also found that individuals preferred opaque cover to clear cover, which was evidence of the ability of *T. gratilla* to sense the better cover type using their phototactic tube feet (Millot 1975) even when the light was not as strong. It also is further support of covering being related to light intensity because there was not another reason for urchins to prefer the opaque cover, as both

plastics were very similar. The ability of individuals to choose cover in this way, and the significant difference in total percent cover between individuals, was evidence of the existence of individual covering behavior, which has been noted in previous studies (Millott 1956), though the reasons for individual preference are not known. This result also supported the hypothesis that spine loss would not have an effect on covering material preference.

From the phylogeny (Appendix A) it appeared that covering behavior evolved in sea urchins in the common ancestor of *Glyptocidaris* and its sister group. This behavior was subsequently lost twice: once in the genus *Glyptocyphus* and again in the *Colobocentrotus*-*Heliocentrotus* clade, possibly because these clades evolved other protection methods and no longer needed to cover themselves for protection.

Conclusion

Overall, the results of this study supported the hypothesis that covering behavior in *T. gratilla* urchins is heavily influenced by light condition, and that a primary motivation urchins have for covering themselves is protection from sunlight. Though in the field, preference for covering materials was not statistically significant on its own, the lab studies showed that individual preference is a factor in covering behavior in *T. gratilla*, a result that has not been previously noted for this species. Spine loss does affect urchin behavior by making urchins more likely to cover themselves, perhaps as a way of compensating for the loss of spines which protect them from predators. Spine loss does not impede covering behavior, and though it does impede movement, does not seem to put urchins at great risk in their environment while their spines regrow. This knowledge is important in that it demonstrates how robust this species is, which is important in its consideration as a biological control agent. It also means urchins with lost spines can still successfully protect themselves from predators via covering, which has been shown to reduce predation over short periods of time (Zhao et. al 2014).

The observance of individual behavioral differences is an important consideration for others conducting studies on this species, as it represents a confounding variable in studies that focus on determining the reasons for urchin behavior. Previous studies have noted that differences in covering behavior between

individual gastropods are due to the lack of a needed cover as they grow larger and age (Portmann 1959), and postulated that individual variance in *T. gratilla* is due to this phenomenon (Park and Cruz 1994). However, in this study the two smallest, and hence youngest, urchins covered less than larger, older ones.

My results provided new support for the link between *T. gratilla* covering behavior and UV radiation. I also showed that *T. gratilla* may cover more when injured to protect themselves from predation and other threats. This study also found individual variation in *T. gratilla* covering behavior, which has not been noted before. Better definition of both covering and individual behavior in sea urchins adds to the body of literature in existence on echinoderm behavior, which has mainly focused on spawning and aggregating behaviors.

Confirming that urchin behavior is strongly related to light intensity is important in that future studies can use this knowledge to compare covering behaviors between genera and species and see what patterns might arise. This information could lead to a better understanding of the phylogeny of covering behavior evolved in sea urchins. This knowledge will also help compare urchins not only to each other but to the other various species on the planet that exhibit cryptic behaviors.

Future directions

Further study of the urchin *T. gratilla* could include a more detailed field study that compared locations with varying currents, as this study did not quantify what, if any, portion of the urchin's covering behavior has to do with current resistance. It would also be helpful to do a more in depth study simply on spine loss to see when, or if, urchins begin to cover less again, and see if that corresponds to a certain level of spine regrowth. To test the true importance of spines and tube feet to locomotion and covering, it might also be helpful to remove spines from the entire urchin test, or to remove tube feet, and observe urchin behavior.

It would be interesting to know what the reasons are for individual variation in covering percentages, and to see if there seems to be any sort of selection in urchins for urchins that cover more, or urchins that cover less. It would also be interesting to test other urchin species for individual preference, to compare them to *T. gratilla*.

Urchins also responded poorly to higher water temperatures, often spawning when they felt threatened (personal observation). Further study could therefore also examine when urchins feel the need to spawn, and if there is a certain temperature at which they spawn. This information might be useful in determining how urchins would respond to rising ocean temperatures.

ACKNOWLEDGEMENTS

I would like to thank the UC Berkeley Gump station and its staff for their hospitality, capability, and resilience. This class would also not be possible without the professors Jonathon Stillman, Brent Mishler, Stephanie Carlson, Cindy Looy, Vince Resh, and Patrick O'Grady. Their knowledge and support was unending. I would also like to acknowledge our graduate student instructors Eric Armstrong, Camilla Souto, and Dave Kurz. They were our friends, our champions, and our gurus. Finally, I would like to thank Chandler Schaeffer for her invaluable help as a field partner, as well as the rest of the Moorea class of 2015.

LITERATURE CITED

- Abercrombie, N., and W. J. Aila Jr. 2014. DLNR leads management team to investigate sea urchin disease. Department of Land and Natural Resources, News Release 5/16/14. <<<http://dlnr.hawaii.gov/blog/2014/05/16/nr14-061/>>>
- Adams, N. L. 2001. UV radiation evokes negative phototaxis and covering behavior in the sea urchin *Strongylocentrotus droebachiensis*. Marine Ecology Progress Series 213:87-95.
- Agatsuma, Y. 2001. Effect of the covering behavior of the juvenile sea urchin *Strongylocentrotus intermedius* on predation by the spider crab *Pugettia quadridens*. Fisheries Science 67:1181-1183.
- Amato, K. R., S. L. Emel, C. A. Lindgren, K. M. Sullan, P. R. Wright, and J. J. Gilbert. 2008. Covering behavior of two co-occurring Jamaican sea urchins: differences in the amount of covering and selection of covering material. Bulletin of Marine Science 82(2): 255-261.
- Ambrose, H. W., K. P. Ambrose, D. J. Emlen, K. L. Bright. 2002. A Handbook of Biological Investigation, sixth edition.

- Hunter Textbooks Inc., North Carolina, USA.
- Bates D., M. Maechler, B. Bolker, and S. Walker. 2015. Fitting linear mixed-effects models using lme4. *Journal of Statistical Software* **67**(1): 1-48.
- Belleza, D. F. C., R. S. A. Abao Jr., C. A. Taguba, and D. T. Dy. 2012. Effects of UV-C on the masking behavior of the green urchin *Salmacis sphaeroides* (Linnaeus, 1758). *The Philippine Scientist* **49**: 34-43.
- Crook, A. C., E. Verling, and D. K. A. Barnes. 1999. Comparative study of the covering reaction of the purple sea urchin, *Paracentrotus lividus*, under laboratory and field conditions. *Journal of the Marine Biological Association of the UK* **79**: 1117-1121.
- Dayton, P. K., R. J. Rosenthal, L. C. Mahen, and T. Antezana. 1977. Population structure and foraging biology of the predaceous Chilean asteroid *Meyenaster gelatinosus* and the escape biology of its prey. *Marine Biology* **39**(4): 361-370.
- Dix, T. G. 1970. Covering response of the echinoid *Evechinus chloroticus* (Val.). *Pacific Science* **24**:187-194.
- Dubois, P., and L. Ameye. 2001. Regeneration of spines and pedicellariae in echinoderms: A review. *Microscopy Research and Technique* **55**(6):427-437.
- Dumont, C. P., D. Drolet, I. Deschenes, and J. H. Himmelman. 2007. Multiple factors explain the covering behaviour in the green sea urchin, *Strongylocentrotus droebachiensis*. *Animal Behavior* **73**(6):979-986.
- Forster, G. R. 1959. The ecology of *Echinus esculentus* L quantitative distribution and rate of feeding. *Journal of the Marine Biological Association of the United Kingdom* **38**: 361-367.
- James, D. W. (2000). Diet, movement, and covering behavior of the sea urchin *Toxopneustes roseus* in rhodolith beds in the Gulf of California, México. *Marine Biology* **137**(5): 913-923.
- Juinio-Menez, M. A., H. G. Bangi, M. C. Malay, and D. Pastor. 2008. Enhancing the recovery of depleted *Tripneustes gratilla* stocks through grow-out culture and restocking. *Reviews in Fisheries Science* **16**(1-3):35-43.
- Kehas, A. J., K. A. Theoharides, and J. J. Gilbert. 2004. Effect of sunlight intensity and albinism on the covering response of the Caribbean sea urchin *Tripneustes ventricosus*. *Marine Biology* **146**(6):1111-1117.
- Lewis, L. B. 1958. The biology of the tropical sea urchin *Tripneustes esculentus* Leske in Barbados, British West Indies. *Canadian Journal of Zoology* **36**:607-621.
- Littlewood, D.T.J., and A.B. Smith. 1995. A combined morphological and molecular phylogeny for sea urchins (Echinoidea: Echinodermata). *Philosophical Transactions of the Royal Society B: Biological Sciences* **347**(1320): 213-234.
- Maddison, W. P., and D. R. Maddison. 2015. Mesquite: a modular system for evolutionary analysis. Version 3.04. <<<http://mesquiteproject.org>>>.
- Milligan, H. N. 1915. Observations on the foreign objects carried by the purple sea urchin. *Zoologist* **894**: 441-453.
- Millot, N. 1956. The covering reaction of sea urchins. *Experimental Biology* **33**(3): 508-523.
- Millott, N. 1975. The photosensitivity of echinoids. In: Russell FS, Yonge M (eds) *Advances in Marine Biology*. Academic Press, New York. 1-52.
- Ogden, N. B., J. C. Ogden, and I. A. Abbott. 1989. Distribution, abundance and food of sea urchins on a leeward Hawaiian reef. *Bulletin of Marine Science* **45**(2): 539-549.
- Moore H. B. 1966. *Ecology of Echinoids. Physiology of Echinodermata*. New York, Jown Wiley & Sons, Inc. 73-87.
- Park, I., and C. Cruz. 1994. Masking behavior and distribution of the tropical sea urchin *Tripneustes gratilla*. *Biology and Geomorphology of Tropical Islands* **4**:22-47.
- Pawson, D. L., and D. J. Pawson. 2013. Bathyal sea urchins of the Bahamas, with notes on covering behavior in deep sea echinoids (Echinodermata: Echinoidea). *Deep Sea Research Part II: Topical Studies in Oceanography* **92**: 207-213.
- Pederson, H. G., and C. R. Johnson. 2006. Predation of the sea urchin *Heliocidaris erythrogramma* by rock lobsters (*Jasus edwardsii*) in no-take marine reserves. *Journal of Experimental Marine Biology and Ecology* **336** (1): 120-134.
- Portmann, A. 1959. *Animal Camouflage*. University of Michigan.
- R Development Core Team. 2013. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna Austria. Retrieved from<<<http://www.R-project.org/>>>.

- Sih, A., A. Bell, and J. C. Johnson. 2004. Behavioral syndromes: an ecological and evolutionary overview. *Trends in Ecology and Evolution* **19**:372-378.
- Smith, A. 1988. Phylogenetic relationship, divergence times, and rates of molecular evolution for camarodont sea urchins. *Molecular Biology and Evolution* **5**(4): 345-365.
- Stevens, M., and S. Merilaita. 2009. Animal camouflage: current issues and new perspectives. *Philosophical Transactions of the Royal Society*. **364**(1516):423-427.
- Stimson, J., T. Cunha, and J. Philippoff. 2007. Food preferences and related behavior of the browsing sea urchin *Tripneustes gratilla* (Linnaeus) and its potential for use as a biological control agent. *Marine Biology* **151**(5):1761-1772.
- Thet, M. M., M. Noguchi, and I. Yazaki. 2004. Larval and juvenile development of the echinometrid sea urchin *Colobocentrotus mertensii*: emergence of the peculiar form of spines. *Zoological Science* **21** (3): 265-274.
- Unger, B., and C. Lott. 1993. In-situ studies on the aggregation behaviour of the sea urchin *Sphaerechinus granularis* Lam. (Echinodermata: Echinoidea). *Echinoderms through time. Proceedings of the Eighth International Echinoderm Conference, Dijon, France* 913-919.
- Valentine, J. P., and G. J. Edgar. 2010. Impacts of a population outbreak of the urchin *Tripneustes gratilla* amongst Lord Howe Island coral communities. *Coral Reefs* **29**(2):399-410.
- Wickham, H. 2009. ggplot2: elegant graphics for data analysis. R package, <<<https://cran.r-project.org/web/packages/ggplot2/>>>.
- Yanagisawa, Y. 1972. Preliminary observations on the so-called heaping behaviour in a sea urchin, *Hemicentrotus pulcherrimus* (A. Agassiz). *Publications of the Seto Marine Biology Laboratory* **19**(6): 431-435.
- Yoshida, M. 1966. Photosensitivity. Pages 435-463 in R. A. Boolootian, editor. *Physiology of Echinodermata*. John Wiley & Sons, New York, USA.
- Zhao C, N. Ji, B. Zhang, P. Sun, W. Feng, J. Wei, and Y. Chang. 2014. Effects of covering behavior and exposure to a predatory crab *Charybdis japonica* on survival and *HSP70* expression of juvenile sea urchins *Strongylocentrotus intermedius*. *PLoS ONE* **9**(5):1.
- Zhao, C. H., Zhou, X. Tian, W. Feng, and Y. Chang. 2014. The effects of prolonged food deprivation on the covering behavior of the sea urchins *Glyptocidaris crenularis* and *Strongylocentrotus intermedius*. *Marine and Freshwater Behaviour and Physiology* **47**(1): 11-18.

APPENDIX A

Phylogeny of urchin species via Littlewood and Smith (1995), modified in Mesquite. Covering behavior is noted in black.

