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# Evaluating in situ Grazing Patterns of *Lytechinus variegatus* and their Effects on Seagrass Beds of *Thalassia testudinum*

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**Evaluating *in situ* Grazing Patterns of *Lytechinus variegatus* and their Effects on  
Seagrass Beds of *Thalassia testudinum***

**ABSTRACT:**

The sea urchin *Lytechinus variegatus* is a known grazer of the seagrass *Thalassia testudinum*, and has been known to denude vast stands of seagrass beds at high densities. Outside of these denudation events, the effects of sea urchins on seagrass are poorly understood. This study examines the effects of *L. variegatus* on *T. testudinum* *in situ*, to understand how sea urchins are affecting seagrasses *in situ*. Results indicate that urchins were found in the offshore portion of the seagrass bed at densities up to 4 urchins/m<sup>2</sup>. Changes in temperature and sediment size in the bay indicate that there is a greater exchange rate bay water in the offshore portion of the seagrass bed, and bay water may act as a temperature buffer for urchins in that part of the bed. Urchin movement experiments and dispersion patterns indicate that urchins move more where seagrass cover is low. Field surveys and lab choice experiments indicate that urchins tend to be detrital consumers of seagrass blades rather than herbivores on live seagrass tissue. Together, these results suggest that *L. variegatus* densities observed in this study do not appear to have strong negative effects on seagrass beds as has been previously seen in field enclosure experiments and denudation events.

## INTRODUCTION

*Thalassia testudinum*, turtlegrass, is a submerged marine angiosperm that is one of the most dominant seagrass species in subtropical areas of the Gulf of Mexico (Darnell and Heck 2013). It is a vital component of marine ecosystems. *T. testudinum* beds are helpful in maintaining fisheries as well as crucial in preventing of erosion of sediments (Valentine and Duffy 2006). While an array of marine organisms such as pinfish, blue crabs, sea urchins, and a host of others use *T. testudinum* beds for refuge from predation (e.g. Heck and Valentine 2006), the value of seagrass as a food source to the overall grazer community is not clear.

Though *T. testudinum* and other seagrasses are highly productive, seagrass has traditionally been thought to be “unpalatable” and of low nutritional quality to grazers (e.g. Heck and Valentine 2006). In response to direct grazing, *T. testudinum* can trigger a cascade of plant defenses that can change the composition of the blade itself. These responses can include an increase of chemical and structural deterrents such as lignins, as well as an increase in epibiotic factors such as epiphytes (Verges et al. 2010). In response to herbivory, nitrogen in seagrass tissue becomes lower making it less nutritious to organisms that consume them. Because of this, researchers accepted for decades past that seagrasses are not a primary food source for herbivores, and that grazing rates are low (e.g. Klumpp et al. 1992). However, more recent literature challenges that paradigm. Reviews of literature on seagrass herbivory indicate that that herbivores can remove and consume between 5-90% of *T. testudinum* in any given time frame (Cebrian and Duarte 1998), and that between ~3% and 100% of above ground seagrass production reaches heterotrophs via the grazing pathway (Heck and Valentine 2006). However, the degree to which senesced seagrass tissue is consumed relative to live tissue is unclear.

There are several documented instances of overgrazing by sea urchins. Studies suggest that urchin grazing may have a larger role in the decline of seagrass beds than previously thought (Cebrian and Duarte 1998; Eklof et al., 2008). *Diadema antillarum*, the long spined urchin, can create barren “halos” of sand around reefs (Heck et al. 2000, Ogden et al. 1973). *Actinopyga rudiata* has been found to completely eliminate sea grass beds in

waters surrounding Japan (Heck et al. 2000). *Tripneustes ventricosus*, found in subtropical and tropical Atlantic areas, is also known for completely denuding seagrass beds (Heck et al. 2000, Ogden et al. 1973, Moses and Bonem 2001).

*Lytechinus variegatus* (Lamarck, 1816), the green or variegated urchin, is an abundant inhabitant of subtropical and tropical seagrass beds. *L. variegatus* has occasionally been observed to completely denude areas of seagrass beds (Heck and Valentine 1995; Camp et al. 1973), as well as affect species abundance and composition of a seagrass bed within enclosures (Heck et al. 2000). In the laboratory, *L. variegatus* can rapidly consume live *T. testudinum* tissue (pers. obs.), though it will feed on senesced seagrass tissue as well as living tissue (e.g. Greenway 1995). In addition to grazing on seagrass, *L. variegatus* also consumes prey such as small gastropods and the mussel, *Madiolus americanus*, sessile invertebrates such as hydrozoans, ectoprocts, barnacles, and macroalgae in order to maximize growth (Cobb and Lawrence 2005, Hammer et al. 2006, Klumpp et al. 1992, Watts et al. 2007). In general, urchins are thought to be opportunistic feeders, though more recent laboratory studies indicate that *L. variegatus* has the ability to be selective using chemoreceptors located on hundreds of tube feet covering both oral and aboral surfaces to sense food items (Pisut 2004, Klinger and Lawrence 1995). While Klinger and Lawrence (1984, 1995) concluded that *L. variegatus* can only locate food (particularly *T. testudinum*) at distances up to 8 cm, and therefore rely on random encounters for feeding strategies, Pisut (2004) found that *L. variegatus* has the ability to sense food to distances up to 1 meter in the laboratory, but only under conditions of turbulent flow. Despite numerous diet studies, field observations, and enclosure studies (reviewed below), it remains unclear the ways through which *L. variegatus* affect seagrass populations.

When *L. variegatus* abundances are unusually high (364 – 636 urchins/m<sup>2</sup>), they completely denude portions of seagrass beds (Rose et al. 1998; Camp et al. 1973). Evidence that *L. variegatus* herbivory influences seagrass populations comes not only from observation of denuding at high densities, but also from field experiments enclosing urchins at lower densities. Heck and Valentine (1995) investigated the conditions

necessary for urchins to create permanently unvegetated plots in seagrass beds as well as how varying intensities of grazing affect the above and below ground biomass of *T. testudinum*. Urchins containing 40 urchins/m<sup>2</sup>, a density seen towards the upper range of urchin densities during the year, in St. Joseph's Bay, Florida, were placed in enclosures for 4 months. The enclosures were buried 15 centimeters to sever the rhizome/root layer of the seagrass. Urchins that were allowed to graze continuously created plots that remained permanently unvegetated even 3.5 years after the experiment. Intermittent grazing plots resulted in seagrass growing back at about half the biomass of the ungrazed plot. Researchers concluded that by severing the rhizomes where the stored energy for regrowth was, seagrass could not replace the above ground biomass at a rate greater than or equal to the rate at which it was being consumed. This experiment also explored the variable effects of urchin grazing over seasons and locations. Success of recovery differed by season. Experiments started at the beginning of winter, during low plant productivity, resulted in permanently unvegetated plots due to exhaustion of rhizomes. Overall, the results of this study were consistent with observations of denuding in the field.

Valentine et al (1997) explored the effects of grazing during the growing season. Using 1-m<sup>2</sup> enclosures, three different densities (0, 10 and 20 urchin/m<sup>2</sup>) of urchins were placed in enclosures for 3 weeks and excluded them for 1 week from May to August. During the week with no urchins, *T. testudinum* production and abundance as well as blade width were measured. All parameters decreased, including blade width and production by individual short shoots, but independently of urchin presence. However, above ground biomass increased across all treatments. Grazing treatments had a 40% increase in above ground short shoots relative to the no urchin control plots. In contrast, during fall through spring grazing manipulations, urchins completely denuded seagrass beds and left permanently unvegetated areas in seagrass beds. The researchers concluded that grazing can increase shoot density and biomass during different seasons, and that this dynamic could ultimately explain the persistence of seagrass beds even at high urchin densities. This study indicates that enclosure experiments do not always yield negative effects of urchin grazing on seagrasses

Valentine et al. (2000) also explored the idea that urchins control seagrass abundance seasonally, as well as influence the composition of seagrass beds. They looked at the effects of varying intensities of grazing by urchins on the abundance of *T. testudinum*, as well as seagrass's ability to compensate for biomass losses to urchin grazers at varying depths of water. Grazing effects during two different seasons – February to May, and June through August were examined as well. An ungrazed control was compared to three different grazing intensities using a constant density of 20 urchins/m<sup>2</sup> in 5 m<sup>2</sup> enclosures. In the first treatment, urchins were allowed to graze for 1 month. In the second, they were allowed to graze for 2 months. In the third and last treatment, they were allowed to graze for 3 months. After urchins were removed, above ground and below ground biomass was measured. Overall, they found that the effects of grazing varied seasonally and were not the same across the different treatments. The first and second treatments yielded a decrease in above ground biomass. In the summer treatments, there was not a significant difference in the 2-month treatment compared to the ungrazed control. Additionally, Moore and McPherson (1965) found that urchins in enclosures had much higher consumption rates of *T. testudinum* in the summer than in the winter season. However, when this was tested in Jamaica where there is limited seasonality, it was found that urchins do not have a difference in consumption rates (Greenway 1995). This suggests that urchin seagrass interactions are complex.

Interpreting the influence of urchins on seagrasses from enclosure experiments is difficult because urchin enclosures prevent the import of potential food items, and limit natural movement patterns of urchins. Rather than allowing them to potentially move away from a less palatable food source, the enclosure confines them to one area, forcing them to consume the food source within the boundaries. In laboratory aquaria, *L. variegatus* can move 4.92 meters per hour on average, up to 8.2 meters per hour at their top speeds (Parker 1936). Once urchins consume the more easily accessible or more nutritionally suitable blade tissue, they might move. By having urchins enclosed, they are forced to consume blades perhaps to prevent starvation, and might even consume more blades here to compensate for lower nutritional quality (Heck and Valentine 2006). In addition, it is possible that urchins move on in response to grazer-induced deterrents produced by

seagrasses (reviewed below), or to grazer-induced reduction in seagrass cover. Heck and Valentine (1995) noted that tethered urchins on bare sand were consumed more frequently than those within seagrass. They speculate that when urchin densities are high enough, seagrass cover is reduced, leading to a reduction in the number of urchins from predation pressures. Reduction in the number of sea urchins present can allow seagrasses to recover from grazing pressures. They also found that larger urchins (>30 mm) were preyed upon more than the smaller urchins (<30 mm), especially when there was reduced cover. Movement out of areas where seagrass density is low may be an adaptive behavior. Leaving seagrass beds prior to denudation events occurring may be a trade off to high predation pressures. However, the extent to which seagrass is injured and consumed before urchins move on has not been documented in natural settings where urchin movement is unrestricted. Overall, restricting urchins in enclosures may cause an over-estimation of urchin grazing to seagrass beds.

Urchins may not be grazing on all parts of seagrass indiscriminately. *T. testudinum* blades grow vertically in shoots, with the youngest tissues being at the bottom of the blade on the interior of the shoot. As blades grow taller, and are in the water column longer, distal ends accumulate epiphytes. These distal portions of blades eventually senesce, noted by green tissue turning to brown. Brown, senesced portions of blades become weaker as the decaying process progresses, making them susceptible to breaking off in the water column due to wave action and tides (Larkum et. al 2006). Senesced, broken off portions of seagrass blades accumulate on the bottom of the seagrass bed among shoots (pers. obs.). If *L. variegatus* is consuming senesced detrital blade tissue, then the effect they have on seagrass beds may be less negative than the direct effects herbivores have on live tissue.

Whether urchins are consuming senesced tissue or live tissue, the literature remains unclear on the relative nutritional quality of young seagrass tissue versus older blade tissue. Literature suggests that younger portions of marine plants such as *T. testudinum* are more chemically defended, increasing fitness. With the younger parts of the plants more defended and in turn less grazed, that part of the plant can survive to produce more

tissue while the older, perhaps senescing part of the plant is consumed (Verges et. al 2010). Because of this, live, younger blades tissue should be infrequently ingested. Greenway (1995) found that detrital blades are consumed more based on gut analyses of field urchins.

In addition to being less defended, detrital blades are easier to consume because they are not as structurally defended as live blades. Live blades contain lignins and carbohydrates (fibers) that make them hard, stiff and overall difficult to consume (McConnell et. al 1982). These lignins also bind important nutrients making them less accessible to grazers. A small percentage of grazers have adaptations that allow grazers to access nutrients that are stored in seagrass, but most grazers cannot (Klumpp 1992). Because of this, seagrasses such as *T. testudinum* may not be a useful food source for many herbivores until they have undergone partial decomposition. Partial decomposition can reduce chemical deterrents and blade fiber content, making it easier to digest (Zieman et al 1984). However, younger, quicker growing blades have less lignin than older blades (Cebrian and Duarte 1998). Because of this, grazers might preferentially feed on younger blades that allow easier access to the nutrients within seagrass. Overall though, different grazers may be responsive to different defenses and the net effect of the traits that influence palatability may be important, ultimately making it difficult to predict what tissue type should be preferred in a given situation.

While younger ungrazed blades were found to be less nutritious (higher C/N ratios) than their senesced counterpart by Cebrian and Duarte (1998), Heck et. al (2000) found that decomposed detached blades (detrital) or attached blades that are partially decomposed (senesced) have lower nitrogen concentrations than live blades, potentially making them a poorer quality food item than live blades. Senesced detached and senesced attached blade portions may only become nutritious when they have collected microbial communities through decomposition (Heck Jr. and Valentine 2006). Live blades that are younger and grow faster could have better nutritional quality due to their high nitrogen levels despite containing more lignins that would bind nutrients. Therefore, these blades should be preferentially fed upon (Cebrian and Duarte 1998). This difference in grazing

selection based on nitrogen content has lead Valentine and Heck (2001) to suggest that the lowering of nitrogen content might be used as a defense against herbivory.

Grazing is known to enhance production of physical and chemical deterrents in seagrasses (for example McConnell et al. 1982, Ziemen et al. 1984). In laboratory experiments between choice and no choice, Darnell and Heck (2013) found that *L. variegatus* has a greater preference towards ungrazed blades. Urchins collected from St. Joseph's Bay and St. Andrew's Bay in Florida. A single urchin was then given ungrazed seagrass shoots or grazed seagrass shoots (no choice), or a combination of grazed and ungrazed shoots. The urchins were allowed to graze for up to 8 hours before the shoots were removed. Researchers found that the no choice experiments yielded similar amounts of tissue consumed, but in the choice experiment, urchins consumed significantly more ungrazed seagrass than grazed. They suggest that this was because ungrazed blades have lower lignins, less chemical deterrents and more nitrogen than grazed blades.

It also appears that some urchins, including *Tripneustes gratilla*, *Salmacis sphaeroides*, and *L.variegatus*, in laboratory settings, have a preference towards detrital blades over live blades (Greenway 1995, Watts et al. 2007, Klumpp et al 1993). Klumpp (1993) examined food preferences in the laboratory using the species *Tripneustes gratilla* and *Salmacis sphaeroides*. When presented five equally weighted bunches of identified important food sources (live seagrass, dead seagrass, red algae *Amphiroa fragilissima*, and brown algae *Sargassum crassifolium*), *Tripneustes gratilla* consumed more live seagrass than detrital seagrass while *Salmacis sphaeroides* consumed equal amounts of live seagrass tissue and detrital tissue.

Field studies also support preference for senesced blades. In the field, Greenway (1995) found that 60% urchins had more than 50% detrital blade content in their stomachs rather than live blade or epiphyte material. This was also noticed in the laboratory by Valentine and Heck (2001) showed that urchins preferentially ate lower nitrogen food sources than nitrogen enriched (i.e. detritus vs. live blades), but consumed more of the nitrogen poor sources than nitrogen rich to compensate for poorer nutritional quality.

Overall, these studies suggest that the interaction between the degree of decomposition, amount epiphyte cover, and defense responses by the plant are complex. Laboratory studies may not provide an accurate picture of how urchins are grazing in the field. Laboratory studies offer a variety of advantages that field studies do not. First, they can allow the setting to be more controlled so that there is a lower possibility of confounding variables such as differences in resource availability. Next, it allows researchers to easily manipulate systems on smaller scales than those seen in field settings. Lastly, it allows researchers to closely observe behaviors. However, results may differ in field settings where urchin movement is unrestricted, food availability is more complex, and rate and pattern of water movement differ. Laboratory studies have demonstrated food choice between grazed and ungrazed blades, detrital blades versus live blades, and blades with epiphytes and those without, though it is not clear whether such “preferences” are ultimately important in affecting seagrasses in the field.

### **The Central Objective**

Although interactions between *T. testudinum* and *L. variegatus* have been investigated both in the laboratory and within field enclosures, there is still much unknown about *in situ* effects these urchins have on seagrasses. As discussed above, field enclosures indicate that *L. variegatus* has the ability to affect *T. testudinum* productivity and standing crop, though the effects of *L. variegatus* are not consistent across locations and seasons. It has not been clearly demonstrated whether feeding behaviors documented in laboratory experiments have observable effects in the field. The extent to which urchins concentrate feeding on live tissue or senesced tissue, and concentrate their grazing within a given area likely influence the physiology and mortality of seagrass. This thesis examines 1) the degree to which *L. variegatus in situ* are herbivores on living seagrass versus detrital consumers of senesced blade tissue, and 2) the extent to which these plant-consumer interactions occur over multiple spatial scales. Field observations and laboratory studies on feeding will be examined in context of urchin distributions and movement patterns in order to gain a better understanding of how urchins influence seagrass beds.

This study will examine the effects of urchins on seagrass on three scales by investigating these questions:

**1. Large scale (100's of meters):**

What portion of the seagrass bed are urchins potentially influencing? The direct impact of urchins will be less if the distribution of and grazing by urchins are restricted to certain areas of the seagrass bed.

**2. Medium scale (meters):**

Do changes in seagrass resources affect urchin movement in beds? The direct impact of urchins will be less if urchin tend to move as seagrass tissue is diminished by grazing.

**3. Small scale (centimeters):**

What type of tissue are urchins consuming along the blade? The direct impact of urchins will be less if urchin tend to consume senesced tissue over living tissue.

## METHODS

### Study Site

All field data were collected in St. Joseph's Bay, in the northwestern Gulf of Mexico along the Florida panhandle (USA). St. Joseph's bay is a protected bay bound by Port. St. Joe and St. Joseph's Bay Buffer Preserve on the east and St. Joseph's peninsula on the west (Figure 1). The bay receives no major freshwater input and has 1/6<sup>th</sup> of its bottom (an estimated 9,669 acres of 73,000 total acres) covered in seagrass, mostly along narrow (typically <500 m), shallow (<2m) shelves adjacent to the shoreline of the bay ("St. Joseph's Bay Aquatic Preserve"). Water depth increases dramatically on the bayward side of these shelves (typically to 5-10 m depth over most of the open bay). The dominant seagrass is *Thalassia testudinum* (turtle grass), but other species including *Halodule wrightii* (shoal grass) and *Syringodium filiforme* (manatee grass) also inhabit the bay.

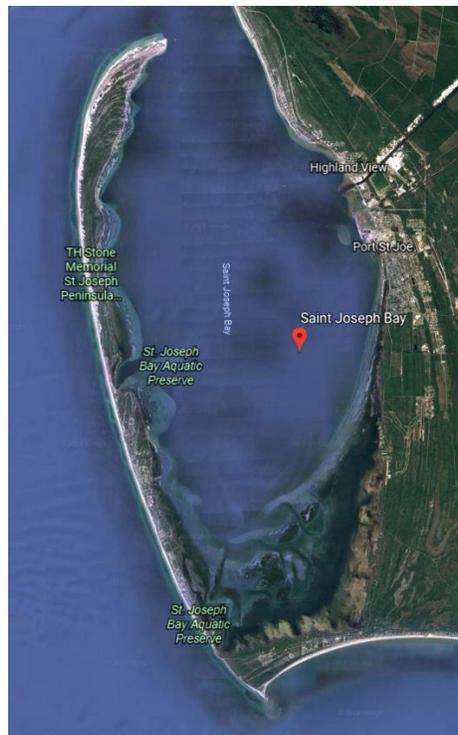


Figure 1: Aerial view of St. Joseph Bay, Florida, USA, where all sampling in this study occurred.



Figure 2: Aerial view of Mosquito Point where all sampling occurred. The long transect extends 500 m, from shore across the bed, until the water drops off. Short transects were sampled parallel to shore in this area.

**1. Large scale effects** - What portion of the seagrass bed are urchins potentially influencing?

To assess trends in urchin abundance and other potential factors related to urchin abundance, a 500 m transect was established perpendicular to shore starting from the shore of Mosquito Point (29°46'45N 85°23'50W), north of Eagle Bay Harbor, with sampling occurring over 25-m intervals (Figure 2, 3). The number of urchins within 0.5 meters along each 25-m transect interval were tabulated. Percent cover of seagrass was also collected through visual estimation to the nearest 20 percent along each 25-m transect interval. Three shoots were sampled at every 25-m interval; one at the beginning of the transect, one in the middle, and one at the end to examine patterns of injury and senescence over the entire transect. The shoots were haphazardly selected in each area (haphazardly being defined in this study as selecting samples without scientific reasoning, removing as much bias as possible). All shoots sampled had the meristem intact. Detached senesced blades were also collected at designated points along the transect. A 0.5 m<sup>2</sup> quadrat was placed near the end of the offshore end of each 25-m interval. Detached blades were collected by gentle raking with a hand through the entirety of the quadrat until all detached blades were removed. Blades collected were placed in

labeled bags, placed on ice and transported to Kennesaw State University for further analysis.

Several physical parameters including water temperature, depth, sediment grain size and organic matter were also examined at the end of each 25-m transect interval. Temperature was collected September 2017 with a thermometer. Depth was measured by placing a meter stick in the water at the end of the 25-m interval (August and September 2017). Sediment for analysis of grain size and organic matter samples were collected in September 2017. At the end of designated meter intervals, sediments from the top ~3 inches were collected by hand. Sediments were placed in labeled bags, put on ice and transported to Kennesaw State University for further analysis.

#### **Live blade Sample Analysis:**

To examine patterns of injury and senescence across the seagrass bed, live blades that were uprooted along the long transect were placed on ice for 24 hours and transported back to Kennesaw State University. Blades from each section of the transect were photographed on lined (1-cm between lines) waterproof paper. Photographs were then uploaded to a computer and analyzed using Image J picture software. Whole blade length and width measurements were recorded as well as presence of injury and senescence at each 1-cm section along the blade. Injury position on the blade was also recorded (one side or both). These measurements were used to calculate mean number of centimeters per blade with injury and with senescence for each 25-m interval.

#### **Detached Senesced Blade Sample Analysis:**

Detached senesced blades (i.e. blade segments found detached from seagrass shoots in the field, typically brown in color) were placed into pre-weighed, labeled aluminum foil bags. Bags were then placed in a drying oven set to 60°C and allowed to dry for 36 h, or until mass remained constant between weighs. Samples were then reweighed, allowing for blade weight to be calculated.

**Sediment Grain Size Analysis:**

To examine how different sized sediments were distributed across the seagrass bed, sediment samples were collected in the field were frozen upon return to Kennesaw State University. Once sediments were thawed, they were homogenized in their respective bags. Each bag was then separated equally into two separate weigh tins. All samples were placed into a drying oven set at 60°C for 36 h, or until constant mass was obtained; this mass was recorded. From there, each sample was placed into a mortar and pestle, where all large aggregates of sediment were broken up until sediment consistency was uniform. Each sample was then separately placed into a sieve series (4000 µg to <63 µg mesh) and sieved for 1 minute, allowing the sediment grain sizes in each sample to separate. The sediment in each sieve was carefully placed into a pre-weighed, labeled aluminum tin and weighed. Percent error for each sample was then calculated. Once each sieve was emptied and weighed, the sieve series was dusted to prevent cross contamination between sediment samples.

**Organic Matter Analysis:**

To examine how organic matter was distributed across the seagrass bed, sediment samples collected for organic matter analysis were homogenized, dried until mass was constant, weighed, and then placed in a muffle furnace. The muffle furnace was set to 500°C for 4 hours to allow any organic matter present to be removed through loss on ignition. After the samples had cooled, the samples were reweighed, and percent organic matter was calculated.

**2. Medium Scale - Do changes in seagrass resources affect urchin movement in beds?****Short Transect Survey:**

To examine urchin dispersion and as well as urchin abundance relative to seagrass cover, a total of 5 short (5-m) transects were established. All short transects were located the

same distance from shore (~450 m from shore) in an area of high urchin density as determined from the long transect survey. The location of transects were chosen so that transect included areas that varied in seagrass cover. Two 0.5 m<sup>2</sup> quadrats were laid side by side on either side of the transect (East and West) at 10 consecutive 0.5-m points along each of the five 5-m transect (n=100 quadrats) (Figure 3). Total number of urchins in the quadrat were enumerated, location within the quadrat noted, and each urchin was gently turned over to determine what the urchin was feeding on (seagrass closest to the urchins' mouth either directly in the mouth cavity or laying across the mouth cavity was noted). Effort was made not to tear urchins from attached blades. Percent cover in the quadrat and number of detached senesced blades in each plot were estimated, and 3 shoots of seagrass were sampled. Seagrass shoots were haphazardly sampled within the quadrat and placed into labeled bags. Detached senesced blades were estimated by raking one's hand diagonally across the quadrat once, carefully to minimize detachment of any live tissue. These blades were then counted and placed in a pre-labeled bag.

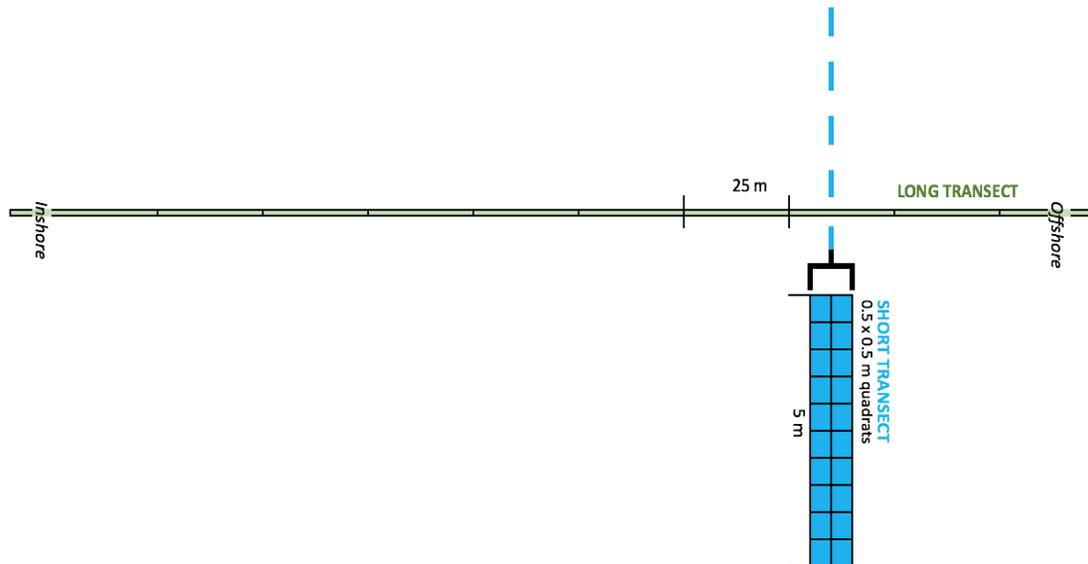


Figure 3: Short Transect Diagram. Short transects were sampled parallel to shore. Size of the short transect is enlarged relative to long transect. Dashed blue lines indicate position of the 5 short transects.

### **Behavioral Movement Experiments:**

To further investigate the movement of urchins in response to changes in seagrass resource, experiments were conducted in August, September and November 2017. Three observers haphazardly selected 8 *L. variegatus* each (24 urchins total), then divided urchins into 6 replicate trials with 4 urchins per trial. Each group of 4 urchins was placed around the base of a flag ~2 cm apart to ensure they were not touching each other or the flag. A minimum of half an hour was lapsed before the trials were concluded. Urchins were checked periodically in order to keep track of them. Stop time for each trial was recorded along with total distance each urchin moved from the flag measured. There were 4 experimental treatments:

- a. Undisturbed – Densely covered, undisturbed seagrass
- b. Clipped – Attached seagrass clipped to “urchin height”. Blades clipped still have the structural obstacle, but the light showing through to the bottom is manipulated.
- c. Clipped without detached senesced blades – The same initial clipped plot was used, but detached senesced grass blades at the bottom were removed so urchins could not cover themselves.
- d. Previously Unvegetated – A nearby sand patch containing absolutely no seagrass prior to trials. If the urchin traveled out of the bare area into the seagrass bed, the time was recorded.

Each date these experiments was conducted, the set up above was replicated, but the experimental design was modified:

- August – Treatments were run in consecutive order with the same eight urchins per person used across all treatments. Urchins were not randomized between trials, but kept in the same two groups of four for the duration of the experiment. The order of treatments was undisturbed, clipped, clipped without detached senesced, and previously unvegetated. The same plots were used for the first three treatments, but urchins were moved to a different area for previously unvegetated trials.
- September – Experimental design from August was replicated in September, except that an undisturbed without detached senesced blades treatment was added, and clipped without detached blades was not replicated. The order of treatments

was undisturbed, undisturbed without detached senesced blades, clipped, clipped without detached senesced and previously unvegetated. The same plots were used for the first two treatments. Urchins were moved to new plots for the clipped and clipped without detached senesced blades, then again to new plots for previously unvegetated trials.

- November – Possible differences in movement seen in August and September trials could have been due to temporal effects (i.e. change in rate of urchin movement is a function of time since initial disturbance or as a function of a previous treatment). To eliminate this possibility, trials in November were ran simultaneously with 4 urchins in each trial (undisturbed, undisturbed without detached senesced, and clipped). Each person had 12 urchins, 4 for each of 3 treatments. Each person ran each treatment trial at the same time so that there were three trials of the same treatment being conducted at the same time in different areas of the bed. No urchin was used more than once. Treatments were randomized in their order:

### **3. Small Scale - What type of tissue are urchins consuming along the blade?**

All of the following procedures were used to assess the degree to which urchins consume senesced versus living seagrass tissue:

#### **Blade and Senescence along Blade Analysis:**

Blades collected along 500-m transects (described in the large-scale methods section) were used to examine trends in injury and senescence along individual blades (n = 255 for May, n = 271 for August). Presence or absence of injury and senescence was recorded every centimeter along the blade.

#### **Urchin Fecal Pellet Study:**

Six urchins from each short transect (n=30 individuals) were collected for fecal pellet analysis – two from the endpoints of the transect, and two from the midpoint (n=6). If

there was no urchin present, the nearest urchin directly outside of the quadrat was collected. The urchins sampled had food cleared from their mouth, all items on their spines removed, and were washed off in seawater. They were then placed into a single labeled jar, containing filtered seawater. A small piece of mesh wire and a rubber band was used to cover the opening of each jar in order to allow oxygen to be accessible to the urchin. All 30 urchins were placed in a cooler with aeration, and allowed to defecate for a minimum of 18 hours. Urchins were removed from the jars the next day and released. Fecal pellets in the jar were photographed, then preserved in a 70% ethanol solution for transport for further analysis. In the laboratory, these photographs were analyzed by drawing a single line down the center of the photo, then counting how many fecal pellets touched the line. Fecal pellets were classified as brown or green in color. To assess whether tissue color changes from green to brown as it passes through the gut, urchins in the laboratory were given only green blades. Urchins given only green seagrass yielded fecal pellets that were 99-100 % green, so that percent brown pellets collected from field urchins are considered as the percent brown (senesced) seagrass tissue consumed.

### **Oral Cavity Survey:**

On each of three sampling dates (October 2016, August 2017, September 2017), ~200 urchins were haphazardly sampled in the field ~450 m from shore. Urchins were sampled while observers moved in a single direction parallel to shore to ensure no urchin was sampled twice. Urchins were gently lifted and flipped over to examine what food source was present in the mouth (brown seagrass, green seagrass, other, or nothing). Urchins that were attached to the substrate were examined close up by the observers completely submerging themselves and examining underneath the urchin.

### **Choice Laboratory Studies:**

Urchins in the laboratory were subject to choice experiments in which they were given the choice between two food sources. Urchins were denied food for a minimum of 24 hours prior to trials to standardize hunger levels. Urchins were then placed in individual beakers (1000 mL). They were then given equal amounts of green and brown seagrass (detached senesced) that had been dry blotted and weighed. Urchins were then allowed to

feed for one hour. After an hour, blades were removed, blotted to remove water and reweighed to measure how much tissue had been consumed.

## RESULTS

### 1. Large scale effects - What portion of the seagrass bed are urchins potentially influencing?

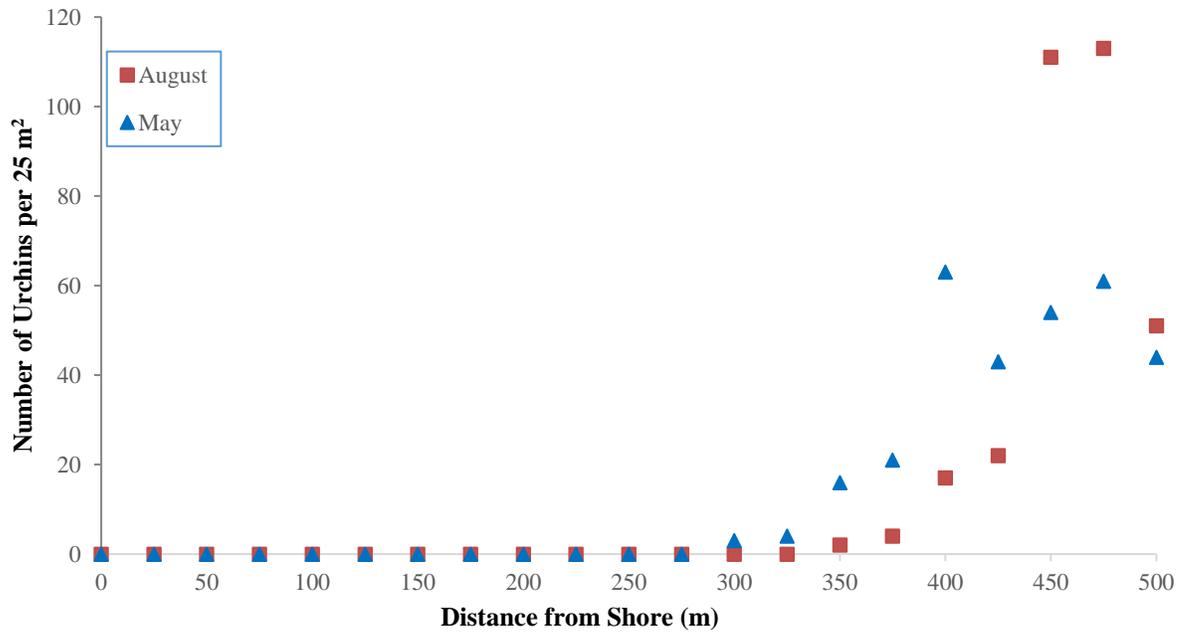


Figure 4: Distribution of *L. variegatus* along the long transect (500 m).

Urchin densities are low inshore, with no urchins being observed inshore.

Densities dramatically increase after ~ 300 meters. The maximum number of urchins recorded in May was 63 urchins/ 25 m<sup>2</sup> while the maximum number of urchins recorded in August was 113 urchins/ 25 m<sup>2</sup> (Figure 4).

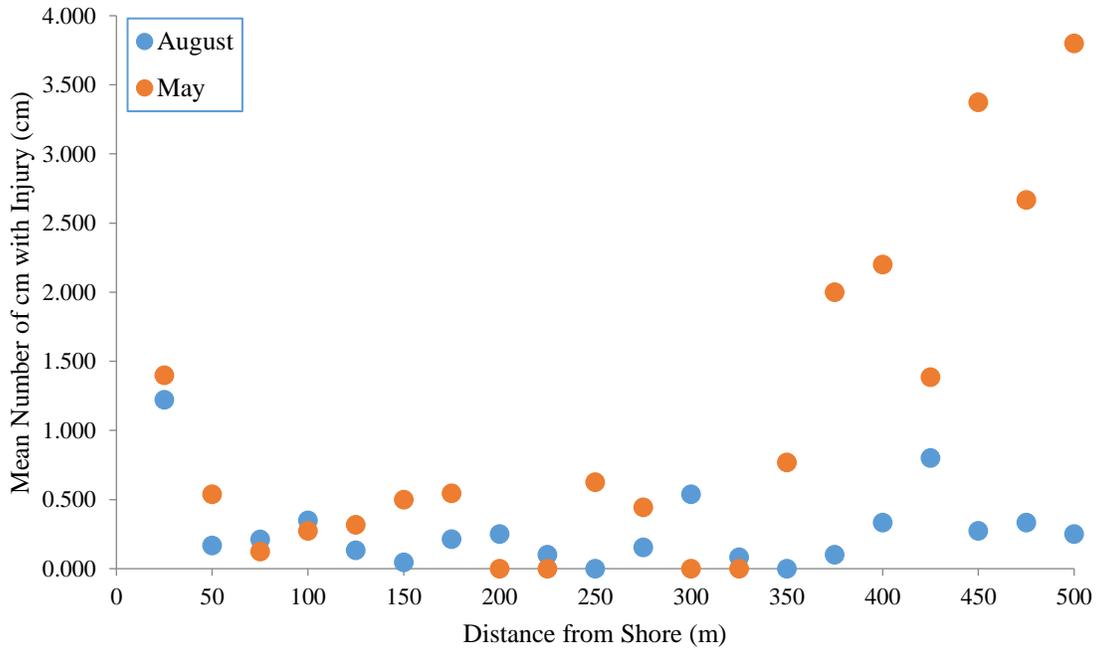


Figure 5: Mean number of centimeters per blade with injury present.

Inshore locations (<350 m) generally had low incidences of injury along blades, except injury closest to shore. Mean number of centimeters with injury dramatically increase offshore after 350 m in May, though not in August. Injury was positively correlated with urchin density in May ( $r = 0.8515$ ;  $p < 0.0001$ ;  $n=20$ ), and not significantly correlated in August when injury tended to be less common across the transect ( $r = 0.0796$ ;  $p > 0.05$   $n=20$ ) (Figure 5).

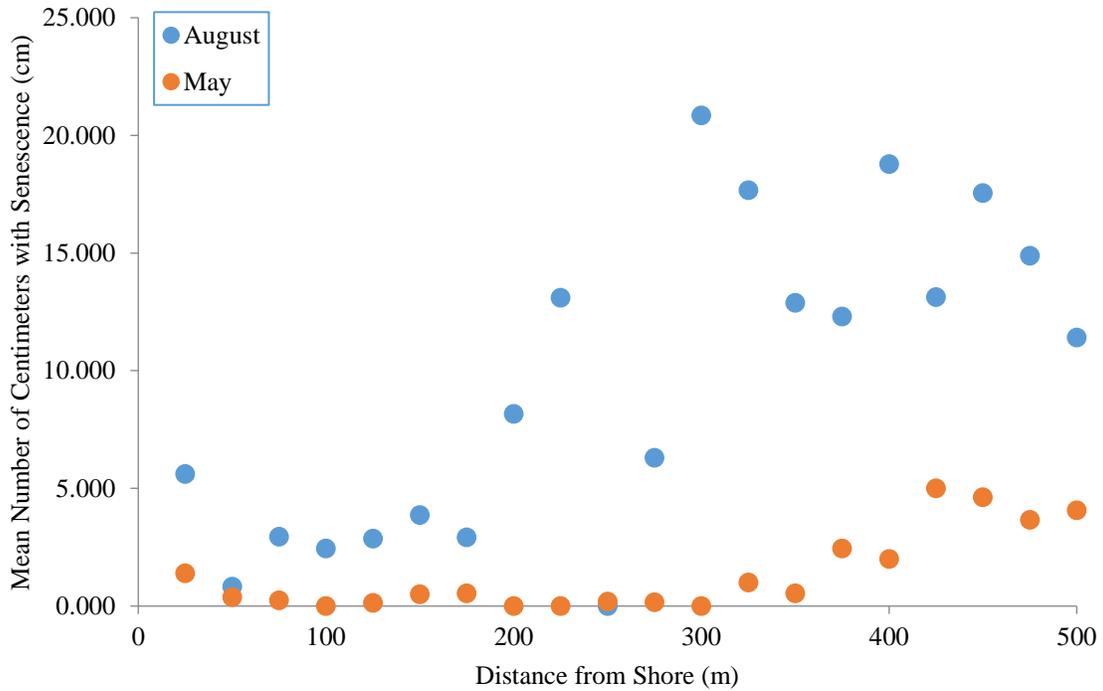


Figure 6: Mean number of centimeters per blade with senescence present.

Senescence is low within 200 meters of the shore, but increases to < 3 fold after 200 meters. Blades in May had a similar pattern, but overall senescence was lower. Senescence was correlated with urchin density in May ( $r = 0.8868$ ;  $p < 0.0001$ ;  $n=20$ ), and not significantly correlated in August ( $r = 0.4271$ ;  $p > 0.05$ ;  $n=20$ ) (Figure 6).

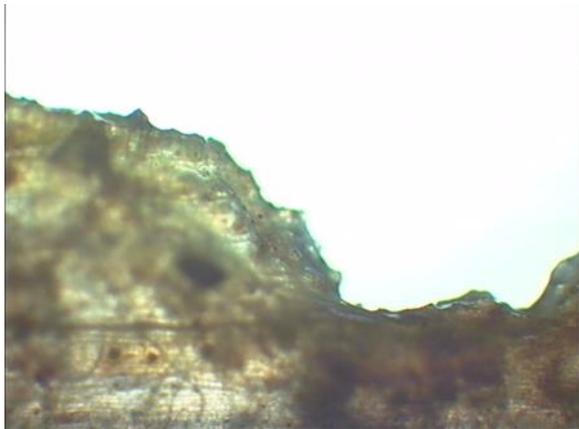
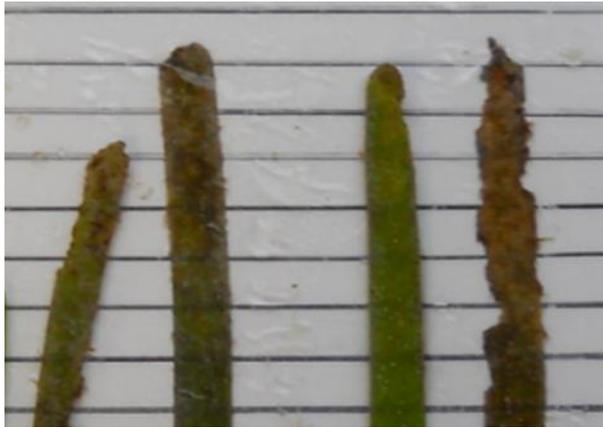
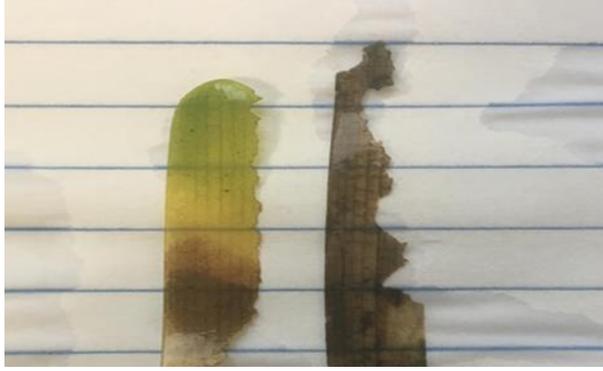


Figure 7: Top: Injury induced by urchins in the laboratory shows jagged, torn edges. Horizontal lines are 1 cm apart on images on the left; Images on the right show damage seen under the microscope (10X). Bottom: Injury observed in situ.

Patterns of injury observed in the field were similar to patterns of injury induced by urchins in the laboratory. Both sets of blades showed jagged tears along the edges of the blades (Figure 7).

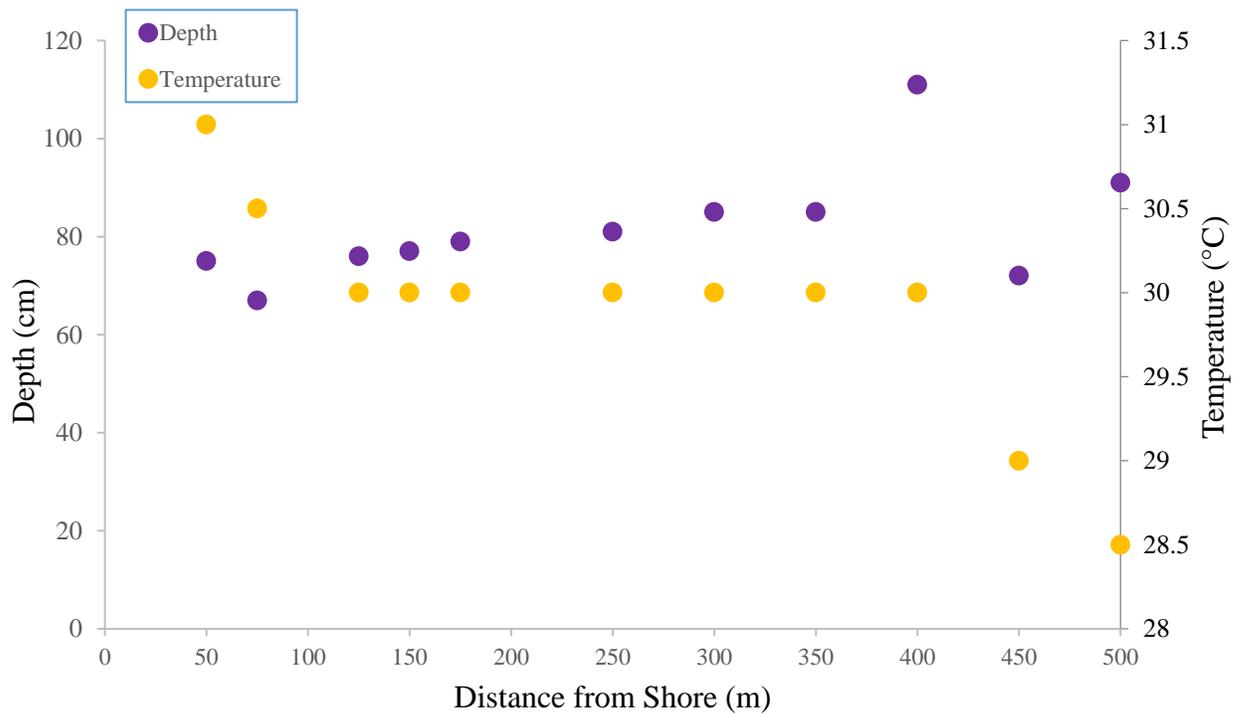


Figure 8: Temperature and depth along the long transect (500 m) of September 2017.

Several abiotic and biotic parameters were measured across the seagrass bed (along the transect), in order to examine factors that might affect urchin distribution. Temperature along the transect varies ~2.5 degrees, with the highest recorded temperatures being inshore (before 100 m), and the lowest temperatures being furthest offshore (after 400 m) (Figure 8). Temperatures in the middle of the transect, from ~125 to ~400 meters remain stable at 30 degrees Celsius. Depth along the transect slightly increases from inshore to offshore, and is more variable after ~350 meters (Figure 8).

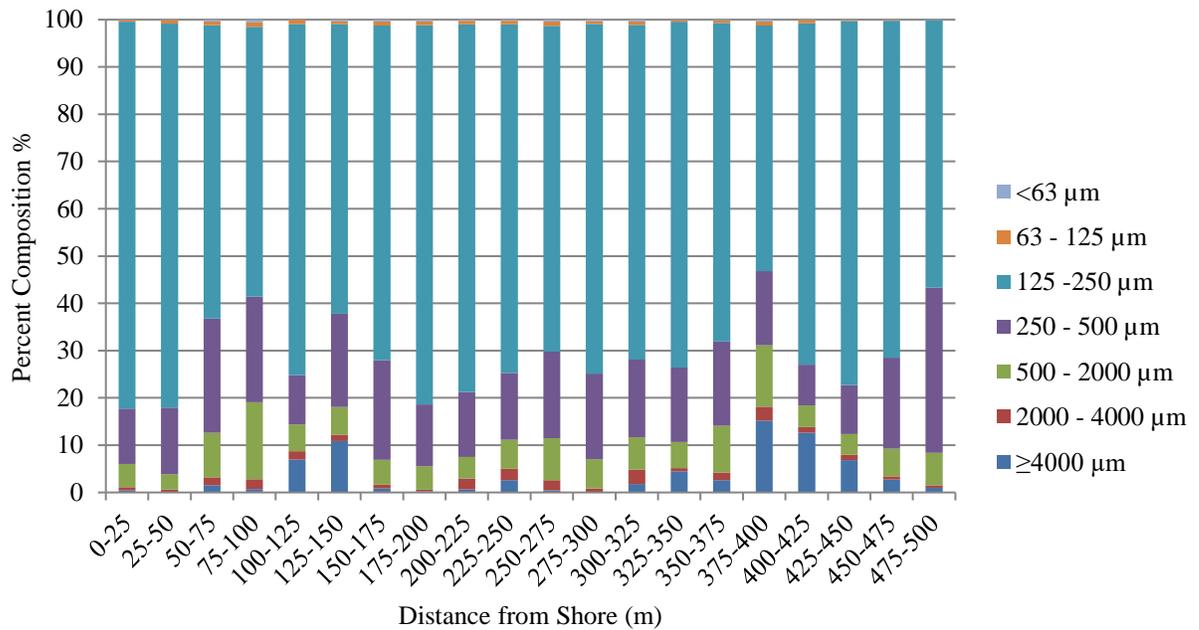


Figure 9: Sediment grain size distributions along the long transect (500 m) September 2017.

Sediment grain size varied along the entire transect. The majority of all samples are comprised of sediment that is 125-250 μm (Figure 9). Larger sediments ( $\geq 4000 \mu\text{m}$ ) are generally more abundant further offshore. Overall, fine sands composed most of the sediments across the transect.

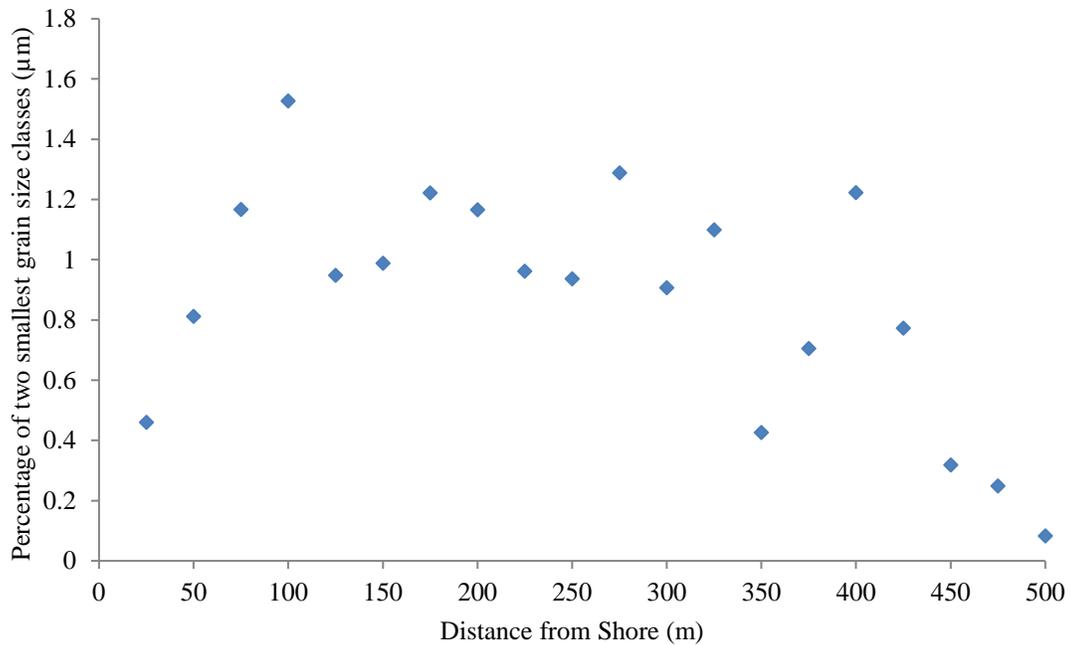


Figure 10: Sediment grain size of 2 smallest grain classes (<63  $\mu\text{m}$  and 63-125  $\mu\text{m}$ ).

Sediment grain size of the smallest two classes is highest inshore, but decreases offshore (~300 m) and were lowest at the furthest three offshore sites (Figure 10).

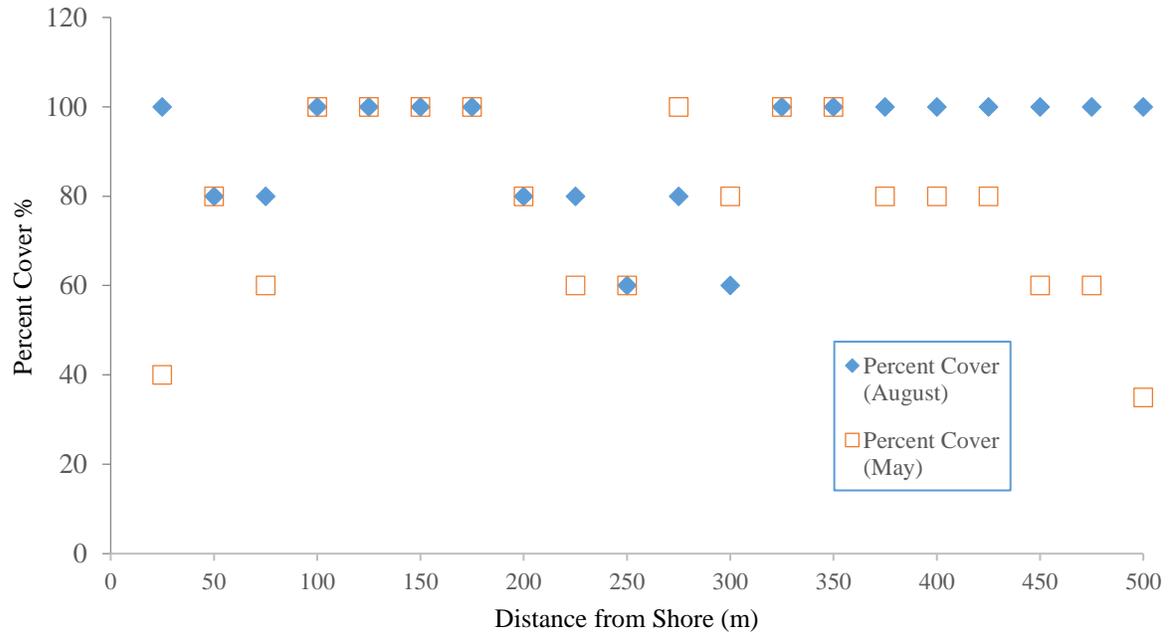


Figure 11: Percent cover of *T. testudinum* (nearest 20 percent) along the long transect (500 m).

Percent cover varied from 40 to 100 percent over the transect, but tended to be low after ~ 325 meters in May (Figure 11). In August, the percent cover tended to be consistently thick after (100 percent cover) ~ 350 meters, indicating there is more seagrass present.

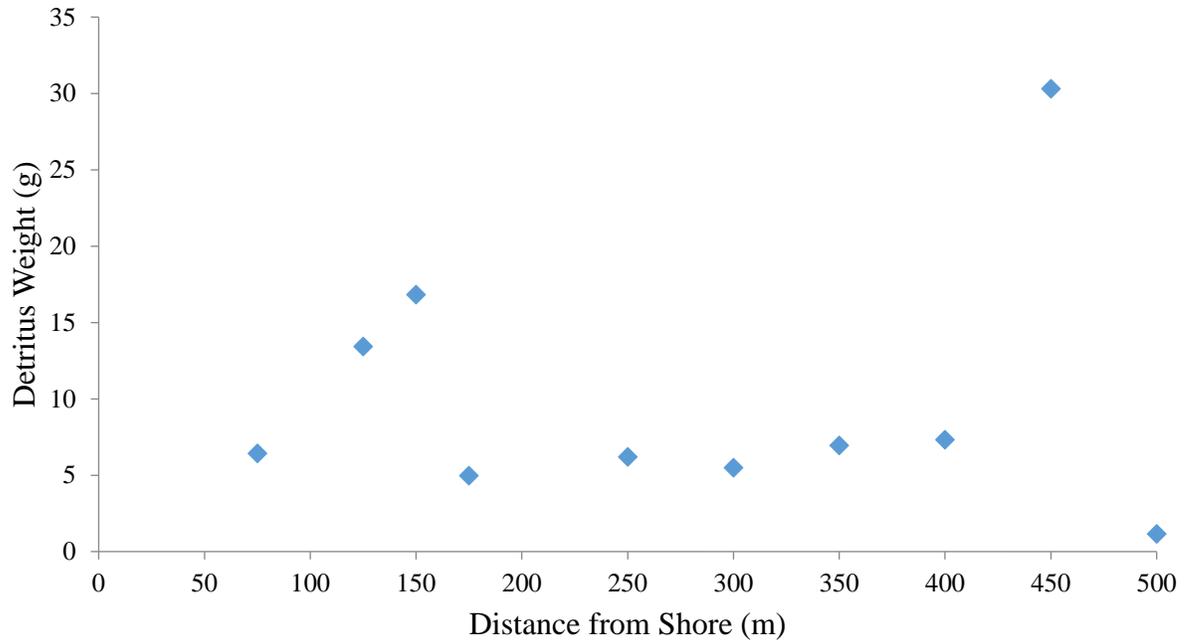


Figure 12: Detrital seagrass blade dry weight along long transect (500 m).

Detrital seagrass blades are present at all sites, but are variable in amounts over the transect. Dry blade weight increases inshore to offshore from ~75 meters to 150 meters, then decreases and doesn't vary until ~450 meters (Figure 12).

## 2. Medium Scale - Do changes in seagrass resources affect urchin movement in beds?

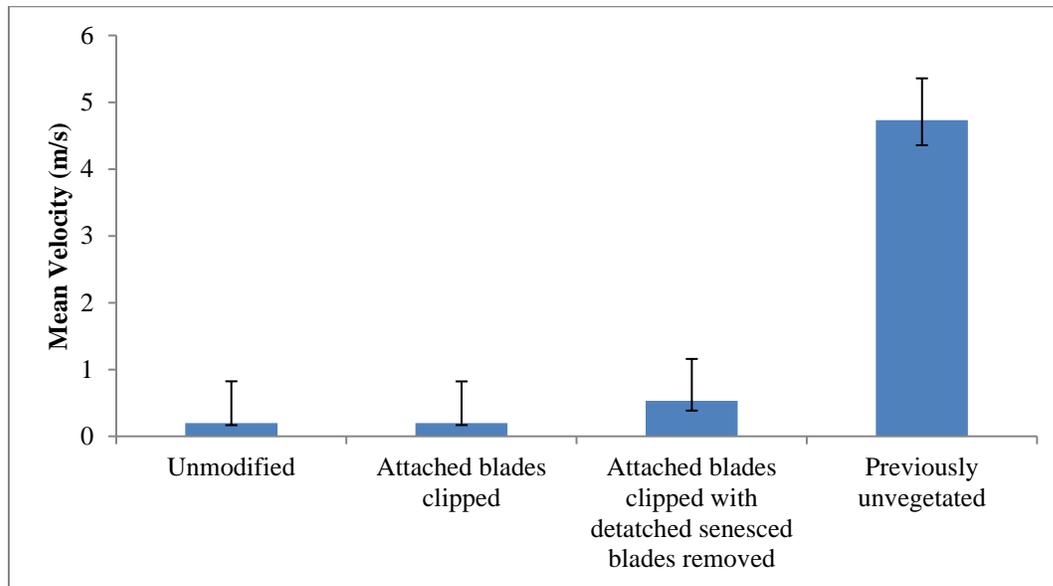


Figure 13: Mean velocity (cm/min) of *L. variegatus* across four different treatments in August. Error bars represent  $\pm 1$  standard error. Order of trials are indicated from left to right where the right bar (previously unvegetated) is the final trial.

Field experiments of urchin movements also suggest that urchins move as a result from changes in seagrass resources. Mean velocity of urchins in previously unvegetated areas was 5 times greater than in unmodified. Urchin velocity differed among treatments (ANOVA;  $p < 0.001$ ) (Figure 13).

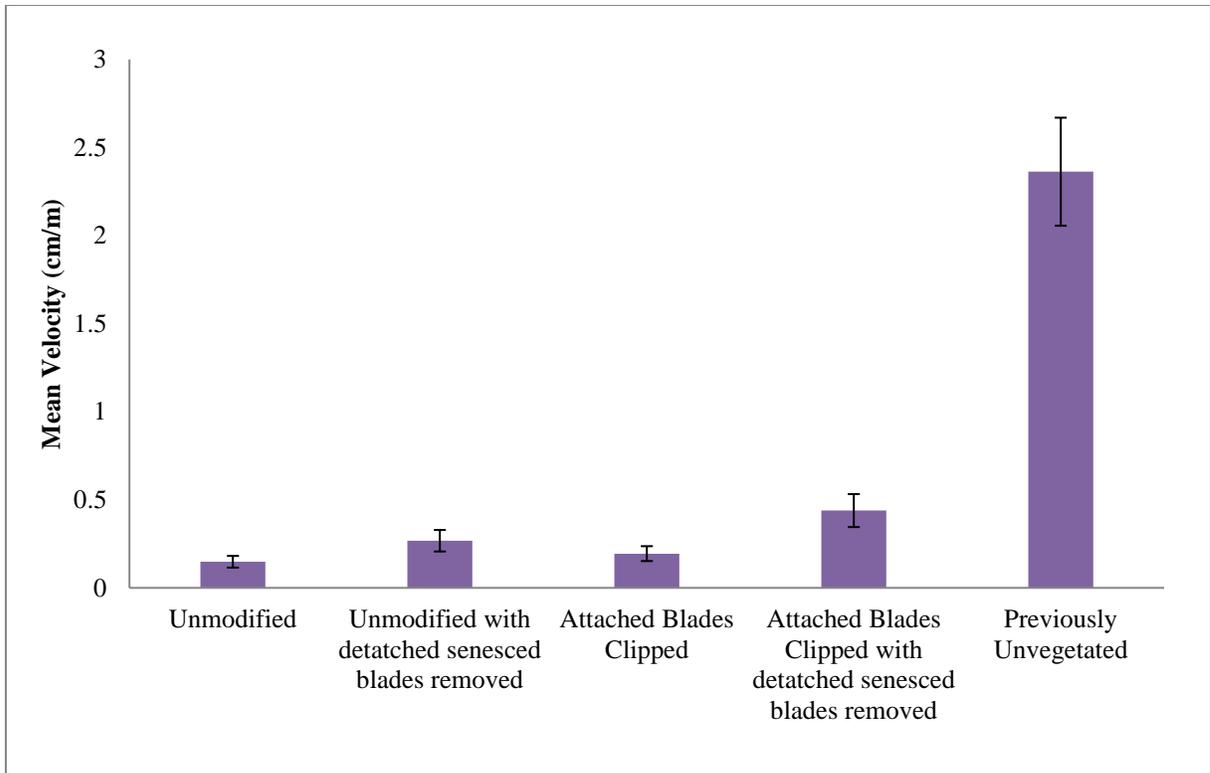


Figure 14: Mean velocity of *L.variegatus* across 5 treatments in September. Error bars represent  $\pm 1$  standard error. Order of trials are indicated from left to right where the right bar (previously unvegetated) is the final trial.

In the September experiments, mean velocity of urchins in previously unvegetated areas was more than an order of magnitude greater than in unmodified. Urchin velocity differed among treatments (ANOVA;  $p < 0.001$ ) (Figure 14).

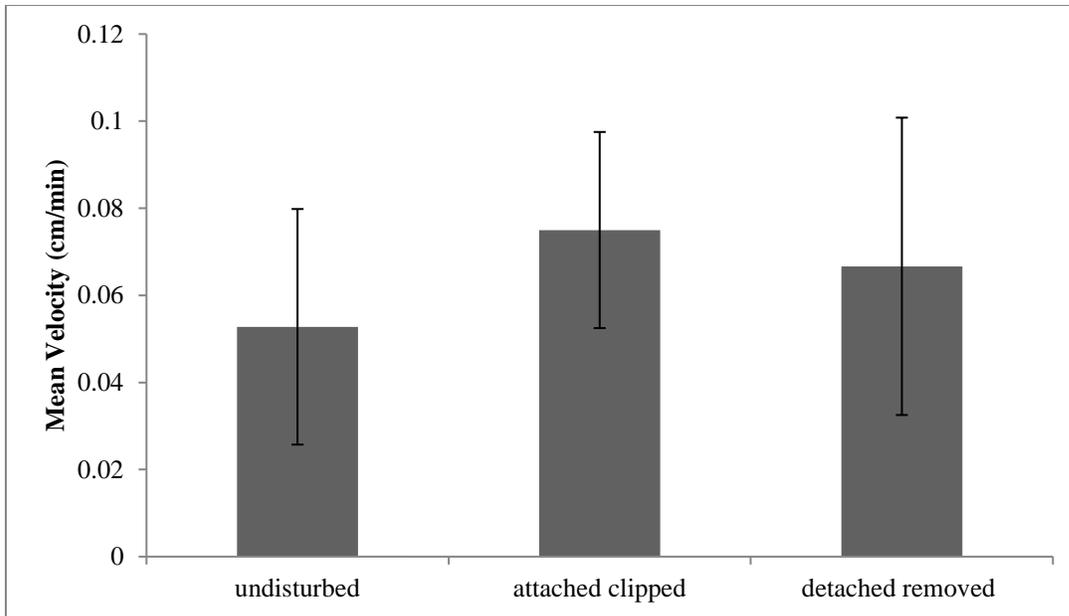


Figure 15: Mean velocity of *L. variegatus* across 3 simultaneous treatments in November. Error bars represent  $\pm 1$  standard error.

The trend of lowest velocity in the undisturbed treatment is similar to the other two treatments, but is not significant (Figure 15) (ANOVA;  $p > 0.05$ ). Velocity of urchins were generally an order of magnitude lower in November than in August and September as water temperature declined.

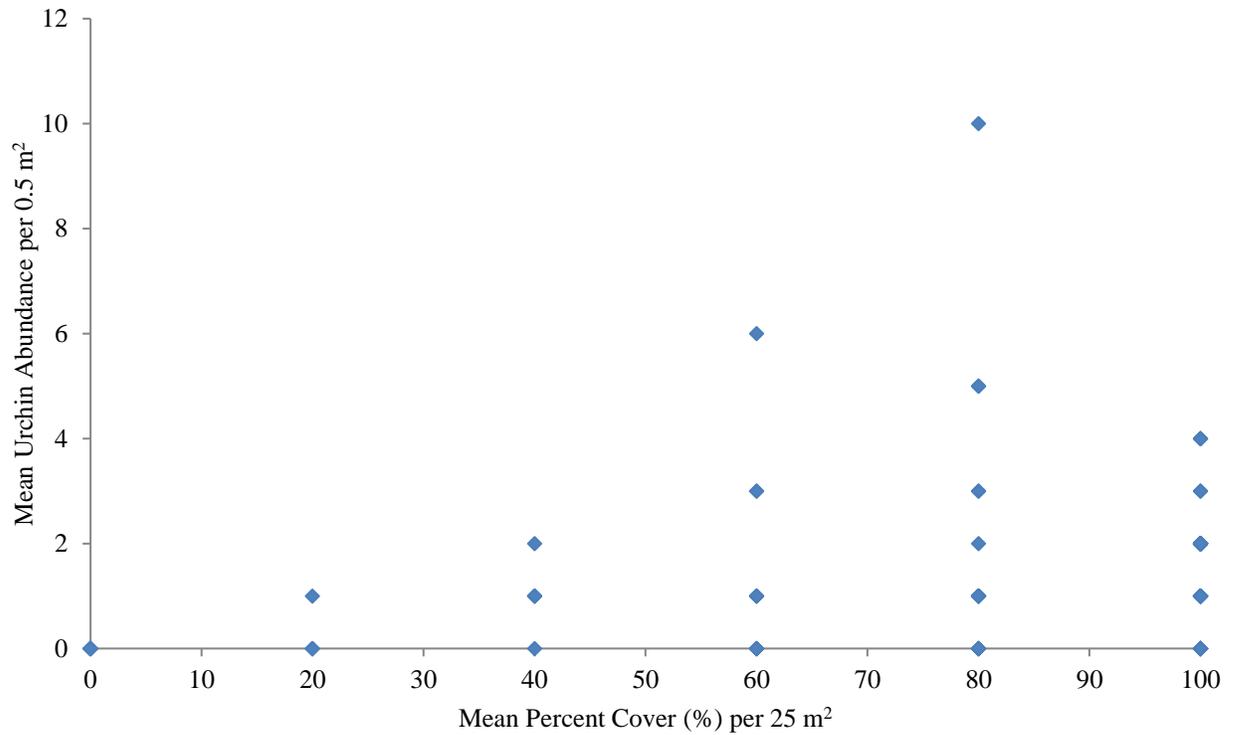


Figure 16: Mean number of sea urchins versus mean percent cover of *Thalassia testudinum* (nearest 20%) per 25 m<sup>2</sup> quadrat.

Where percent cover is low (0%), mean number of urchins is also low (0). As the percent cover increases, the mean number of urchins present becomes more variable. ( $R = 0.1865$ ). This was not a significant correlation ( $p > 0.05$ ). Correlation analysis between urchin density and mean number of segments with blade injury was not significant ( $p > 0.05$ ,  $r = 0.001$ ) (Figure 16).

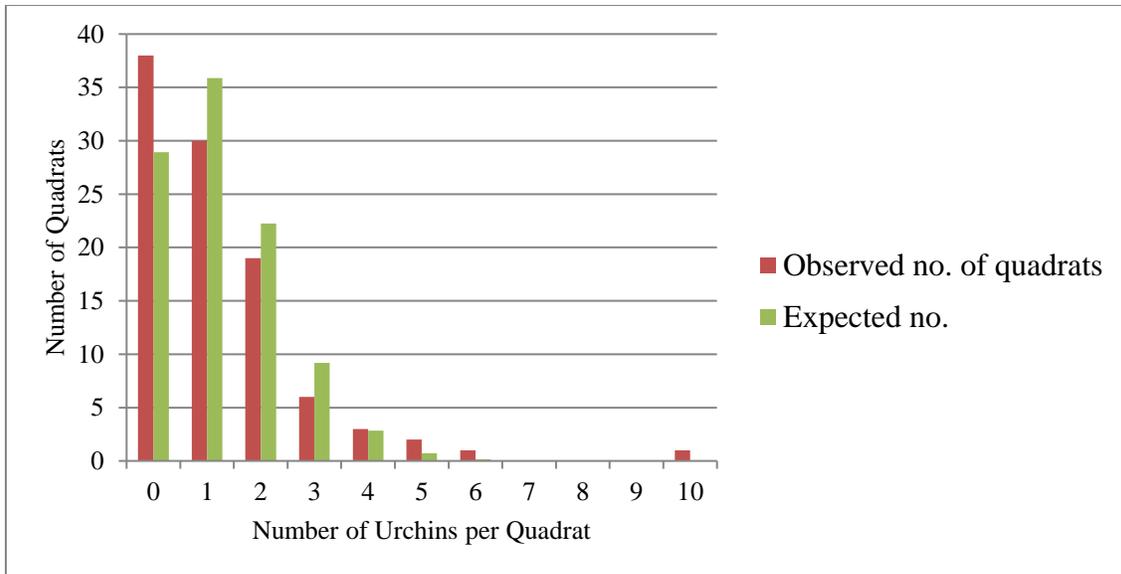


Figure 17: The expected frequency of urchins per quadrat and observed frequency of urchins per quadrat.

Chi Square analyses of urchins indicated clumped dispersion pattern (chi sq = 14601.32,  $p < 0.001$ ). However, the tendency toward clumped was driven by a single plot with 10 urchins (no other plot had more than 6 urchins) (Figure 17). This plot was in a transect that crossed a bare area and a patch of *Halodulli wrightii*. Removal of this transect from the analysis indicated urchin dispersion was not significantly different from random (chi sq = 8.96,  $p > 0.05$ ).

### 3. Small Scale - What type of tissue are urchins consuming along the blade?

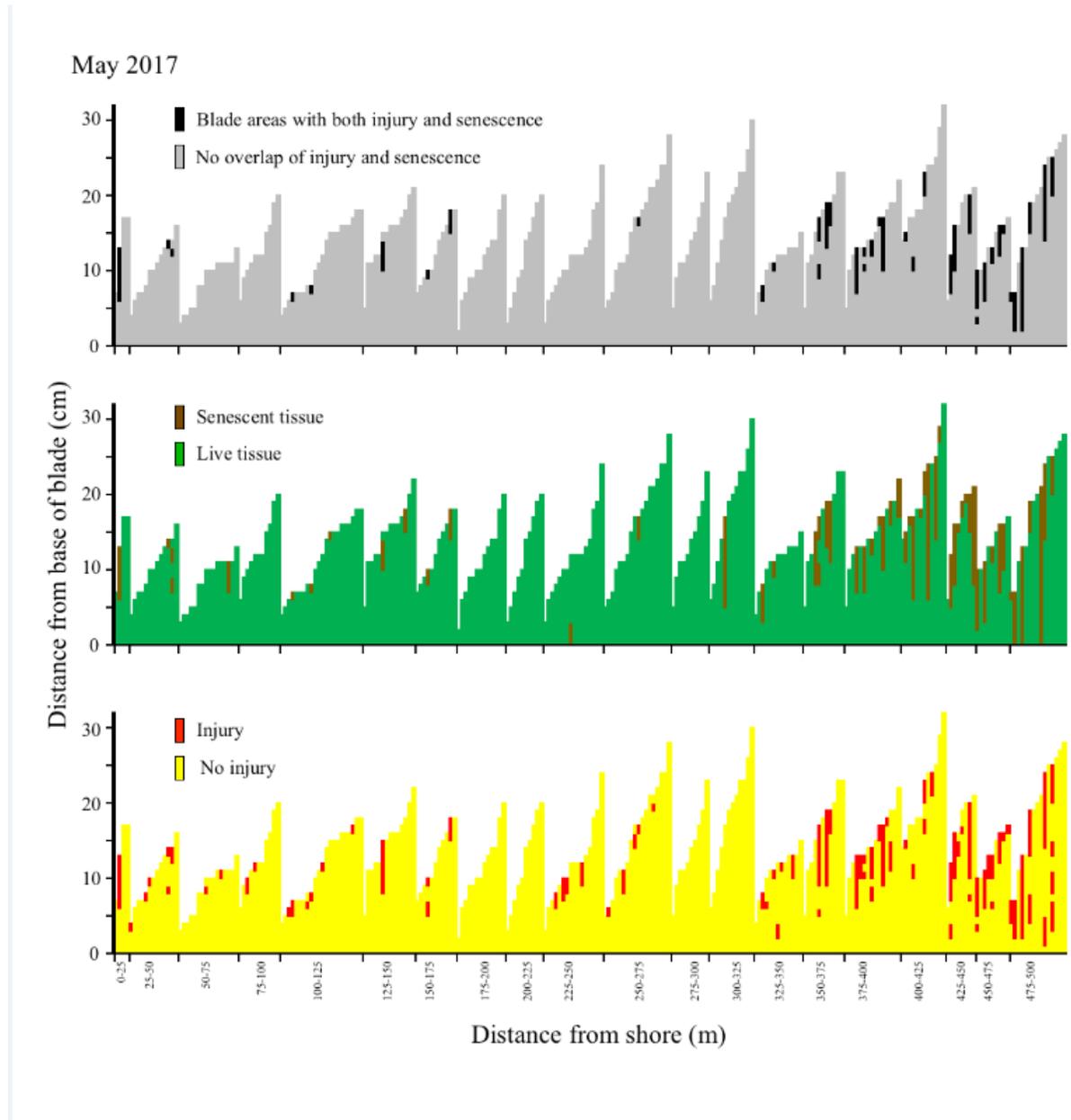


Figure 18: Distribution of injury (bottom panel), senescence (middle panel), and areas where both injury and senescence coincide (top panel) along the long transect in May 2017. Blades are ranked from shortest to tallest (left to right) within each 25 m section.

Most of the injury is concentrated at the distal portion of blades, furthest away from the base (Figure 18, bottom panel). Of 253 blades examined, 60 had injury reaching to the distal end, whereas only 6 injured blades had no injury at the distal end. On many blades, injury occurs over several consecutive centimeters especially along blades sampled

further from shore. Senescence displays a similar pattern (represented by brown), with the majority of senescence being concentrated at the distal portion of blades (Figure 18, middle panel). Inshore, there are few areas where both injury and damage coincide (Figure 18, top panel), but the frequency of both drastically increase after ~325 meters. The percent of senesced blade tissue that is injured (44.3%) is more than ten-fold greater than the percent of live blade tissue injured (3.5%). Chi-square analysis indicate that injury and senescence are more likely to co-occur than by chance alone ( $p < 0.0001$ ) (Table 1).

Observed

		Injured	
		absent	present
Senesced	absent	3081	110
	present	165	131

Predicted

		Injured	
		absent	present
Senesced	absent	2970	221
	present	276	20

Table 1: Comparison of the observed and expected occurrence of injury and senescence along blades in May 2017.

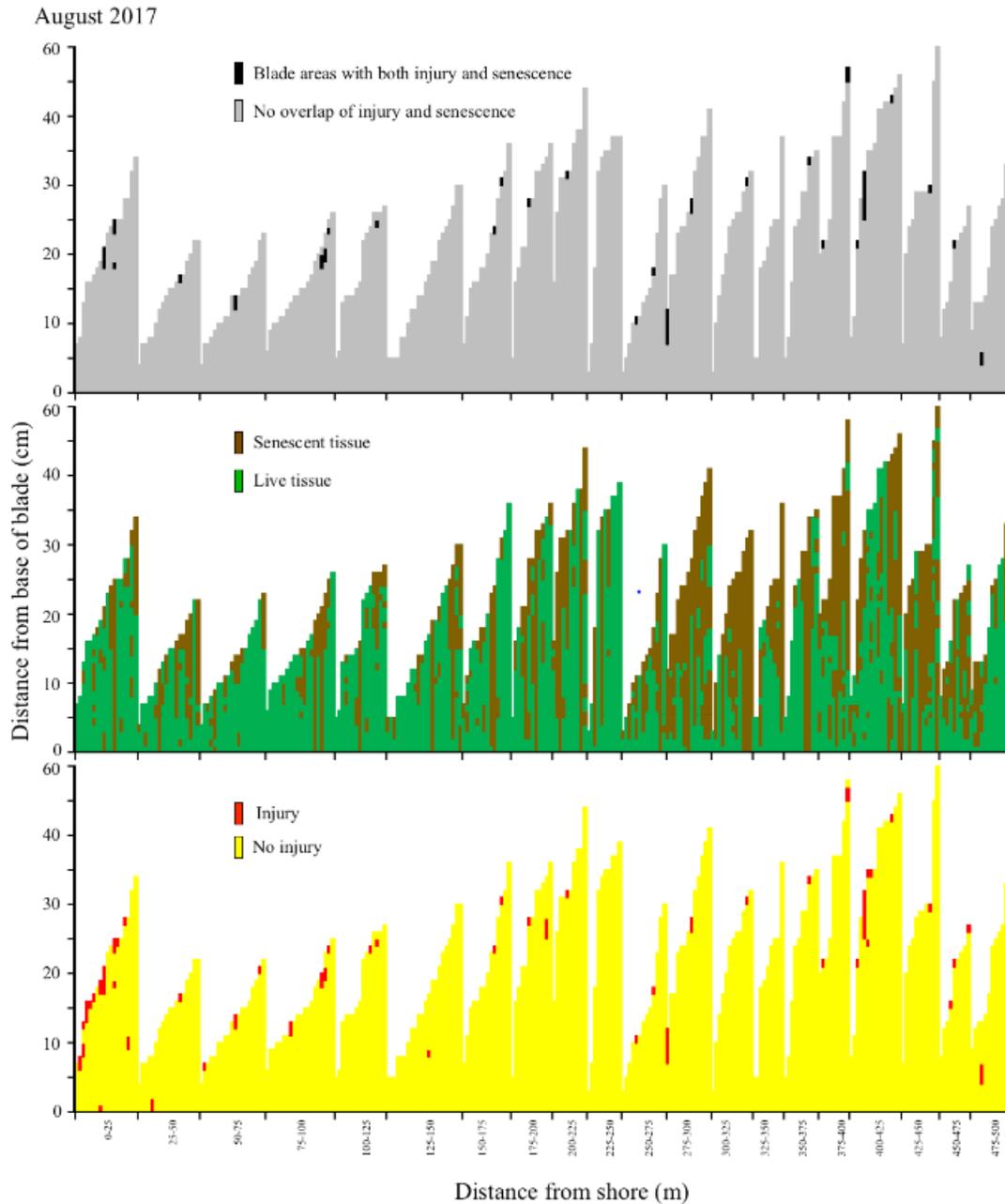


Figure 19: Distribution of injury (bottom panel), senescence (middle panel), and areas of both injury and senescence present (top panel) along the long transect in August 2017. Blades are ranked from shortest to tallest (left to right) per each 25 meter section.

As in May, most of the injury is concentrated at the distal portion of blades, furthest away from the base. Injury overall is much less frequent and is present more consistently over the entire transect than in May (Figure 19). Of 271 blades examined, 38 had injury reaching to the distal end, whereas only 7 injured blades had no injury at the distal end. Senescence displays a similar pattern (represented by brown), with the majority of

senescence being concentrated at the distal portion of blades. Senescence is present over the entire transect, with more present after ~ 300 meters. The percent of senesced blade tissue that is injured (2.1%) is more than twice the percent of live blade tissue injured (1.0%). Chi-square analysis indicates that injury and senescence are more likely to co-occur than by chance alone ( $p < 0.01$ ) (Table 2) (Figure 19).

Observed

		Injured	
		absent	present
Senesced	absent	3449	35
	present	2195	46

Predicted

		Injured	
		absent	present
Senesced	absent	3435	49
	present	2209	32

Table 2: Comparison of the observed and expected occurrence of injury and senescence along blades in August 2017.

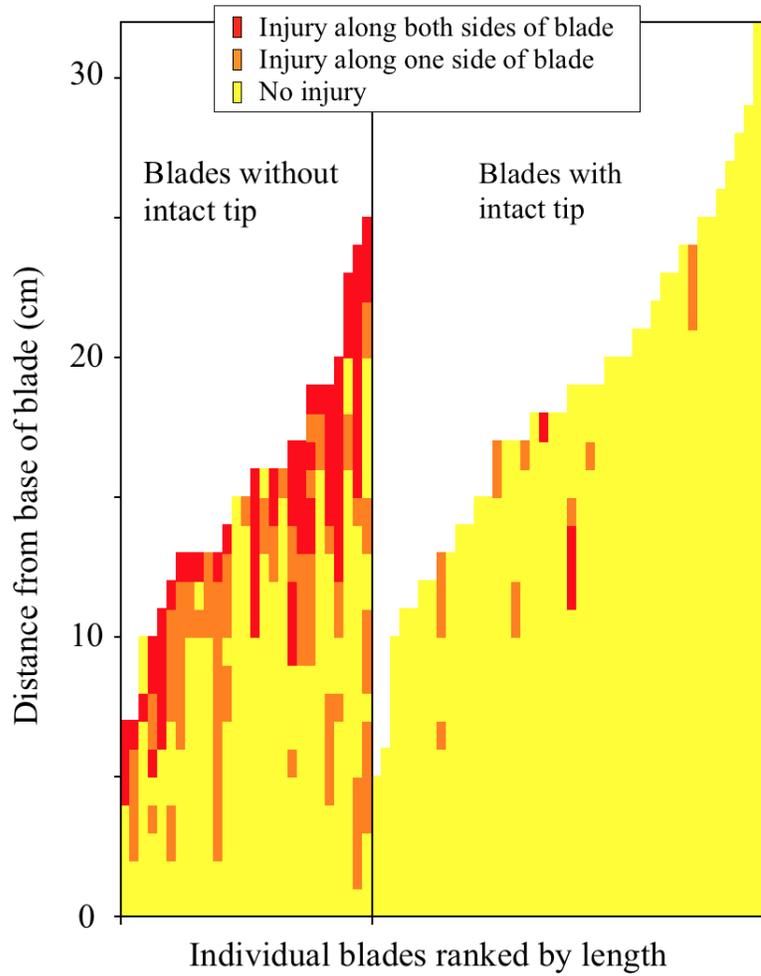


Figure 20: Detailed representation of injury along blades from 350-500 m section of long transect in May.

Blades without tip intact tend to have injury along both sides of the blade. Blades that have their tip intact have less injury present than blades without tips (Figure 20).

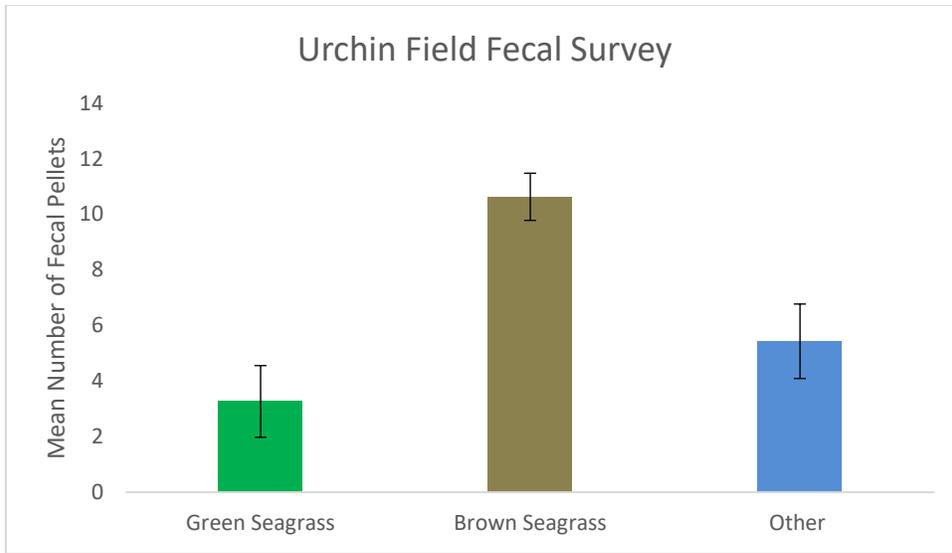


Figure 21: Urchin Field Fecal Survey of *L.variegatus* (August 2017). Error bar represents  $\pm 1$  standard error.

Laboratory and field experiments suggest that urchins key in on senesced tissue. Of the 30 urchins sampled in the field, there were significantly (2 times more,  $p < 0.001$ ) more brown fecal pellets than green and other (Figure 21).

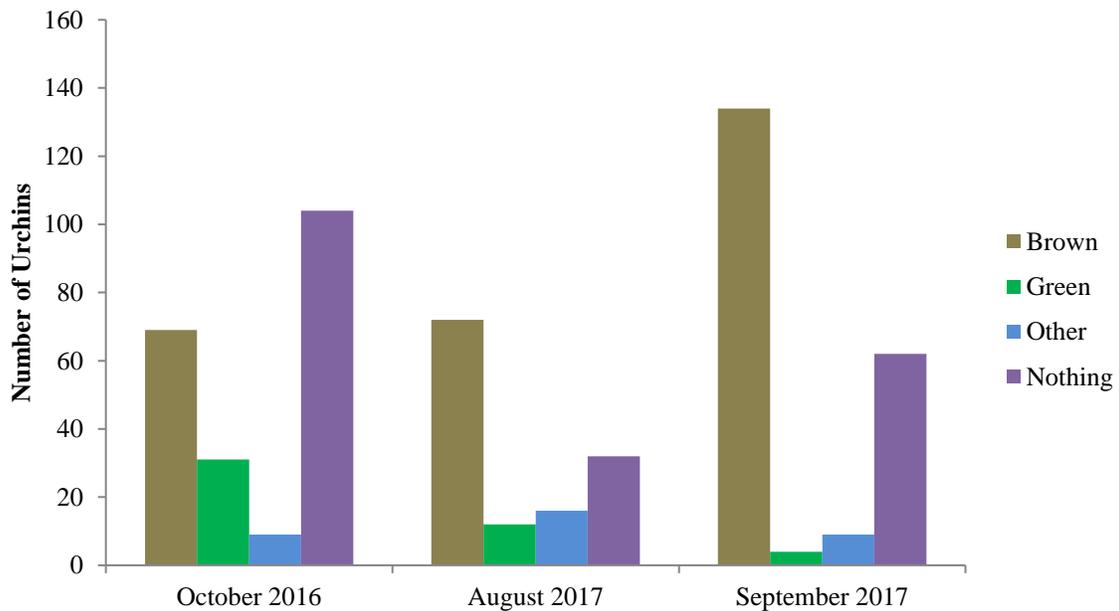


Figure 22: Urchin field oral opening survey. Each color represents a different food source present in the oral opening of the urchin sampled.

All three sampling dates reveal that of urchins that had something in their oral opening, almost 2 times more urchins had brown seagrass in their mouth than green seagrass or other food items, mostly bryozoans and macroalgae (Figure 22).

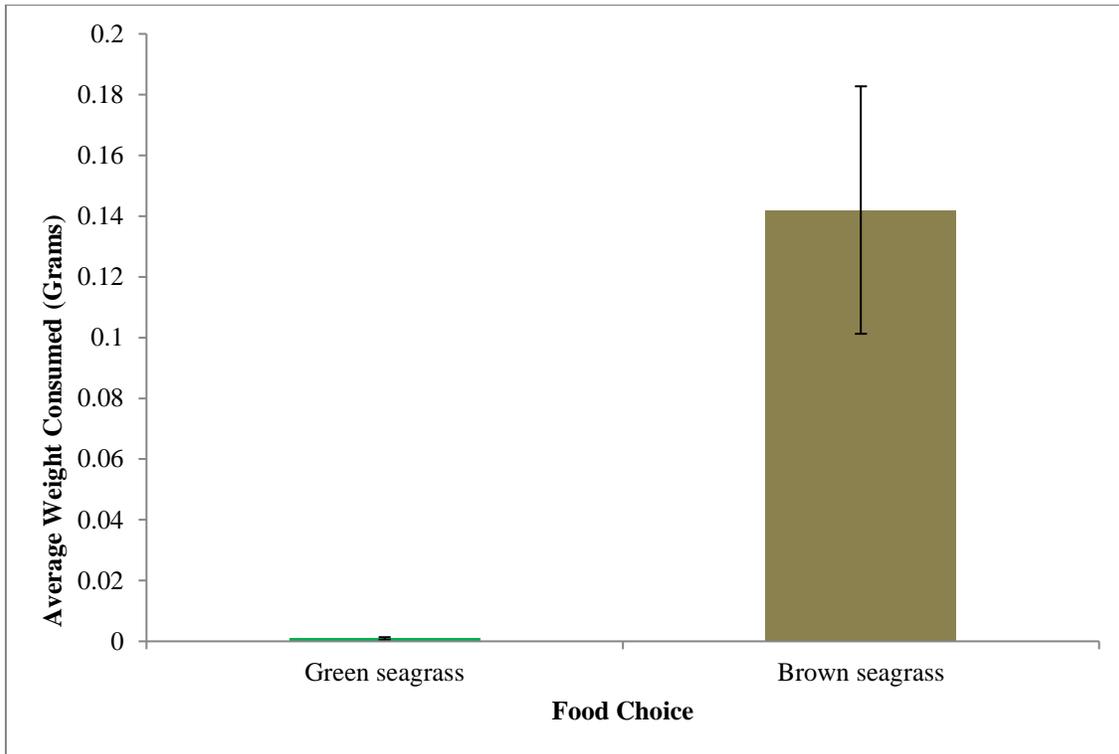


Figure 23: Average weight of blade choice consumed. Error bar represents  $\pm 1$  standard error ( $n = 6$ ).

In controlled laboratory settings where urchins are presented with brown and green seagrass, *L. variegatus* consumed significantly more brown seagrass (paired t-test,  $n = 6$ ,  $p < 0.05$ ) than green seagrass. Trials where urchins did not consume either food choice were not included in statistical analyses (Figure 23).

## **DISCUSSION:**

**Large scale effects** - What portion of the seagrass bed are urchins potentially influencing?

Based on the distribution of *L. variegatus* along the 500 m transect, urchins do not appear to affect the entire seagrass bed equally. No urchins were detected within 300 m of shore, however there is a dramatic increase in sea urchin densities (maximum of 113 urchins/25 m<sup>2</sup> in August, 63 urchins/25 m<sup>2</sup> in May within the next 200 m). Similar distributions of urchins from inshore to offshore were observed in 2015 at 6 locations from the north end of Cape San Blas to the south end of Saint Joseph Bay. No urchins were found inshore (within 150 m), and highest urchin densities (comparable to densities found in this study) occurred further from shore at all locations (unpublished data, J. Dirnberger).

Spatial patterns in several abiotic and biotic factors examined in this study do not correspond to changes in urchin density across the seagrass bed, including water depth, detritus, and percent cover. Depth along the transect does not change drastically along the length of the transect. Detached senesced blades are present at all sites, but is variable along the transect. Percent cover of seagrass inshore was variable, with trends similar in May and August. Offshore, percent cover after about 300 m declined bayward in May, but was consistently high (100%) in August.

Water temperature and depth, however, might offer some insight to the urchin distribution along the long transect. The lowest temperatures recorded farthest offshore where urchin densities were highest, possibly due to mixing with bay water. Though depth varied only by ~0.5 m over the entire 500-m transect, depth increased to greater than 10 m within the next few hundred meters beyond the transect. Deeper bay water offshore is likely to be of different temperatures when compared with peninsula shelf waters inshore. Deep water is less influenced by light and conductive transfer of heat between air and water, moderating temperature fluctuations in offshore waters. Although temperatures observed in May and August may not directly account for the sea urchin distributions, it does provide evidence that the hydrologic dynamics at the offshore end of the seagrass bed are different than inshore, where offshore water masses appears to mix

only partially into the seagrass bed. The idea is supported by the trends seen in the sediment characteristics (Figure 9 and 10), where there is a decrease in finer sediments (<125  $\mu\text{m}$ ) after about 300 m (corresponding to where urchin densities begin to increase) and little organic matter after 400 m. Distribution of sediment particles can be directly related to the amount of water movement energy in the surrounding system. Significant water movement associated with the exchange of water between the open bay and the seagrass bed would prevent finer sediments and organic matter from settling out of the water column. Areas of higher exchange with the open bay appear to be associated with the ability of urchins to populate an area.

While it is not within the experimental design of this thesis to elucidate the factors that determine urchin distribution, a reasonable explanation might involve summer and winter temperature extremes. Watts et al. (2013) noted that *L. variegatus* demonstrate temperature tolerance behavior, existing in water temperatures ranging from at least 11° up to 35 °C. However, outside of this range, mass mortality events have been documented when waters were shallow and temperatures became more extreme. In St. Joseph Bay, areas of bay water and peninsula shelf water mix and may create a “buffer” along the offshore areas of the seagrass bed where temperatures do not change drastically. This can create areas where sea urchins can exist comfortably, rather than inshore where temperature conditions may be less favorable for urchins to inhabit during annual periods of extreme low and high temperatures.

Distribution of injury along the long transect suggests direct grazing effects of *L. variegatus* on *T. testudinum* is limited spatially across the seagrass bed and temporally across seasons. Injury seen along the transect correlates with urchin densities and increases dramatically after about 300 m in May. In August, urchin densities had a similar trend as May, but the amount of injury remained low over the entire seagrass bed. This, along with the increase in percent cover offshore in August is suggestive of a seasonal shift in diet away from live blades. This seasonal shift in grazing patterns among urchins, along with the distribution of urchins across the seagrass bed, support the idea that urchin are not affecting entire seagrass beds but only a portion of them.

While patterns of injury across the seagrass bed corresponded to urchin distribution, it is possible that injury was due to other factors such as other grazers or mechanical injury. However, jagged, torn edges were present both on blades that were injured in the laboratory by urchins and on most of the injury observed on blades collected from the field (Figure 7). Injury patterns such as those are classic examples of urchin injury documented in literature (e.g. Ogden et al. 1973). These observations support the idea that the injury on blades that was seen in the field was most likely due to urchin grazing. In addition, injured blades collected in the field frequently had injury present on both sides rather than one. It would seem unlikely that blades torn by mechanical forces such as wave action would have injury on both sides nor have long stretches of injury along blade margins. Urchins observed in laboratory often demonstrated grazing behaviors that produced these patterns. When given blades, urchins climb up blades then consume from the top down, sometimes grazing only part of the blade and other times consuming the whole blade. Blades that are not completely consumed usually have grazing injury on both sides.

Data collected from these surveys suggest that sea urchins are affecting only about 1/3 of the seagrass bed sampled. Impacts from enclosure studies that suggest urchins can affect seagrass productivity and biomass (Valentine et al. 1997, 2001, Zieman et al. 1984, Heck and Valentine 2001) cannot be applied across the entire seagrass bed. Because of this, more research needs to be conducted, mapping *L. variegatus* distributions in order for researchers to better understand their grazing behaviors and why distributions shift.

## **2) Medium scale effects / Short transects and urchin movement experiments - Do changes in seagrass resources affect urchin movement in beds?**

Within the offshore areas where the highest urchin densities were observed, urchins did not appear to concentrate feeding on whole seagrass shoots in a way that would denude patches within the seagrass bed. This is suggested by the movement of urchins in

response to changes in blade resources, scarcity of urchins in low cover areas, and the rarity of urchins to clump over the scale of several meters.

In experiments measuring the rate of urchin movement in the field for August and September, velocities were greater where seagrass resources were lower. This supports the idea that reduced cover could increase predation pressures (as does scarcity of urchins where percent cover is low as measured across the scale of the short transects). Heck and Valentine (1995) found that urchins tethered in bare patches were consumed at high rates relative to those tethered within stands of seagrass. Urchins that browse lightly and move on rather than denude a patch should experience less predation. Removal of detached senesced blades resulted in increases in movement, and may also be a response to predation as urchins were frequently observed to cover themselves with these blades. *L. variegatus* tend to cover themselves with items such as seagrass, shells, and algae possibly as a way to avoid predation detection (Watts et al. 2013).

Overall, these data support the idea that urchins move over the scale of several meters in seagrass beds, rather than concentrating their grazing efforts in one area. The lack of significant correlation between urchin abundance and injury also supports the idea that urchins are moving on to other blades, rather than focusing their grazing efforts in an area until seagrass is depleted.

If urchins are concentrating grazing efforts in a single area of the seagrass bed, dispersion patterns might be expected to be strongly clumped. Existing literature on urchin grazing events that resulted in denudation of a bed (Camp et al. 1973, Rose et al. 1999) noted that urchins in these events were highly clumped, creating high grazing pressures in those areas of seagrass beds. Densities in these instances were unusually high (636 per m<sup>2</sup> in Camp et al 1973, and over 300 in Rose et al. 1999), and attributed to highly successful spawning seasons (Rose et al. 1999). Urchins at the densities observed in the present study were two orders of magnitude lower (maximum of 4 urchins per m<sup>2</sup>) and do not appear to denude entire areas of seagrass beds as previously reported in literature, but instead injury blades and move on.

The ability of urchins to move over the scale of meters as suggested by observations in this study is relevant to the interpretation of studies utilizing field enclosures that suggest urchins influence seagrass biomass and productivity (e.g. Valentine et al. 1997, 2001, Zieman et al. 1984, Heck and Valentine 2001). Field enclosures restrict urchins to certain areas, potentially causing unnatural grazing pressures in that area. By restricting urchin movements, urchins may be forced to switch to stationary, growing seagrass tissues rather than rely on other more “preferred” food sources such as detached blades that cannot be renewed as quickly because enclosure wall prevent their import. Urchin densities typically used in these enclosure experiments may also have resulted in changes to seagrass biomass that would not have occurred at densities observed in this study (up to an order of magnitude difference).

### **3) Small scale effects / individual blades - What type of tissue are urchins consuming along the blade?**

The minimal and diffuse effects of *L. variegatus* on seagrass as observed in this study is supported by preferential consumption of older, senesced portions of blades. This is suggested by the co-occurrence of injury and senescence, more senesced tissue present in feces, and urchins consuming significantly more senesced tissue in the field and laboratory.

Blades tended to have more injury present along the distal, older portions (Figure 19). Senescence was also concentrated furthest from the base of the blade and tended to coincide with injury in both May and August. However, from this data, it cannot be said that injury causes senescence or vice versa. Regardless of cause and effect, these trends suggest that urchins are grazing on more distal, older tissue along attached blades. Evidence from the surveys of urchin oral cavities fecal pellets and in the field as well as choice experiments conducted in laboratory settings indicate that urchins not only consume older tissue but tissue that has already senesced. This suggests that urchins may

be more detrital consumers of decomposing seagrass than grazers of live seagrass tissue as suggested by enclosure studies. Marco-Méndez et al. (2012), found that *L. variegatus* consumed more senesced seagrass through choice experiments as well as gut content analyses. Vadas et al (1982) found that the majority of sea urchins surveyed in the field had senesced seagrass tissue in their guts. Greenway (1995) also found that 60 percent of the urchins sampled in the field during surveys conducted had more than 50 percent of detrital blades in their guts, suggesting urchins are consuming more senesced tissue than live tissue. Urchins could be preferentially consuming senesced blades because they are less chemically defended as well as less structurally defended than live blades (Zieman et al. 1984). This make nutrients in senesced tissue easier to access as well as blades easier to consume than live blade counterparts.

As discussed above, blades that sustain injury from urchin grazing typically have injury that occurs over several consecutive centimeters along the distal portion of blades, often on both sides of the blade. Injury such as this might cause tips of blades to be removed or weakened causing them to break off. There is less injury present on blades that have their tips intact versus those that do not have their tip intact. There are a couple plausible ideas as to why there is more injury present on blades without their tips. The first is that urchins are severing blades when they are grazing, causing the blades to lose their tips. Another idea is that urchins are keying in on broken blades because broken blades would hypothetically already have senescence present from the previous damage done. This idea seems to be more likely given injury was frequently seen along both sides of blades. Regardless, urchins in this study have a minimal effect on the young, basal growing tissue.

The results of the fecal pellet and feeding studies support the idea that *L. variegatus* have the ability to key in on senesced blade tissue, and “preferentially” consume this tissue in their natural habitat settings, as is also suggested by greater frequency of injury where senescence occurs along the transect as well as along blades. Based on evidence at all three scales, *Lytechinus variegatus* appear to affect seagrass beds through pathways other than direct herbivory on living *T. testudinum* tissue.

Urchins at densities observed in this study do not appear to have strong negative affects on live seagrass tissue, as has been previously seen in field enclosure experiments and denudation events. Future studies should examine at what densities urchins begin to exert direct negative effects on seagrass biomass, and what factors contribute to such urchin densities. By understanding factors that cause urchin densities to reach high numbers such as those in denudation events, effective management and conservation practices can be developed to assist in preserving seagrass ecosystems.

## WORKS CITED

- Camp, David K., Stephen P. Cobb, and Jack F. van Breedveld. "Overgrazing of Seagrasses by a Regular Urchin, *Lytechinus variegatus*." *BioScience* 23.1 (1973): 37-38.
- Cebrián, Just, and Carlos M. Duarte. "Patterns in leaf herbivory on seagrasses." *Aquatic Botany* 60.1 (1998): 67-82.
- Cobb, Janessa, and John M. Lawrence. "Diets and coexistence of the sea urchins *Lytechinus variegatus* and *Arbacia punctulata* (Echinodermata) along the central Florida gulf coast." *Marine Ecology Progress Series* 295 (2005): 171-182.
- Darnell, Kelly M., and Kenneth L. Heck. "Species-specific effects of prior grazing on the palatability of turtlegrass." *Journal of Experimental Marine Biology and Ecology* 440 (2013): 225-232.
- Eklöf, J. S., et al. "Sea urchin overgrazing of seagrasses: a review of current knowledge on causes, consequences, and management." *Estuarine, Coastal and Shelf Science* 79.4 (2008): 569-580.
- Greenway, Margaret. "Trophic relationships of macrofauna within a Jamaican seagrass meadow and the role of the echinoid *Lytechinus variegatus* (Lamarck)." *Bulletin of Marine Science* 56.3 (1995): 719-736.
- Hammer, Hugh, et al. "The effect of dietary protein on consumption, survival, growth and production of the sea urchin *Lytechinus variegatus*." *Aquaculture* 254.1 (2006): 483-495.
- Heck, Kenneth L., and John F. Valentine. "Sea urchin herbivory: evidence for long-lasting effects in subtropical seagrass meadows." *Journal of Experimental Marine Biology and Ecology* 189.1-2 (1995): 205-217.
- Heck, Kenneth L., and John F. Valentine. "Plant-herbivore interactions in seagrass meadows." *Journal of Experimental Marine Biology and Ecology* 330.1 (2006): 420-436.
- Klinger, Thomas S., and John M. Lawrence. "Distance perception of food and the effect of food quantity on feeding behavior of *Lytechinus variegatus* (Lamarck)(Echinodermata: Echinoidea)." *Marine & Freshwater Behaviour & Phy* 11.4 (1985): 327-344.

- Klumpp, David W., J. T. Salita-Espinosa, and M. D. Fortes. "Feeding ecology and trophic role of sea urchins in a tropical seagrass community." *Aquatic Botany* 45.2-3 (1993): 205-229.
- Larkum, Anthony, et al., editors. *Seagrasses: Biology, Ecology and Conservation*. Springer Netherlands, 2006.
- McConnell, Oliver J., et al. "Effects of secondary metabolites from marine algae on feeding by the sea urchin, *Lytechinus variegatus*." *Journal of chemical ecology* 8.12 (1982): 1437-1453.
- Moore, Hilary B., et al. "The biology of *Lytechinus variegatus*." *Bulletin of Marine Science* 13.1 (1963): 23-53.
- Moore, Hilary B., and B. F. McPherson. "A contribution to the study of the productivity of the urchins *Tripneustes esculentus* and *Lytechinus variegatus*." *Bulletin of Marine Science* 15.4 (1965): 855-871.
- Moses, Christopher S., and Rena M. Bonem. "Recent population dynamics of *Diadema antillarum* and *Tripneustes ventricosus* along the north coast of Jamaica, WI." *Bulletin of marine science* 68.2 (2001): 327-336.
- Ogden, John C., Richard A. Brown, and Norman Salesky. "Grazing by the echinoid *Diadema antillarum* Philippi: formation of halos around West Indian patch reefs." *Science* 182.4113 (1973): 715-717.
- Parker, George Howard. *Direction and means of locomotion in the regular sea-urchin *Lytechinus**. Musée royal d'histoire naturelle de Belgique Koninklijk Natuurhistorisch Museum van België, 1936.
- Pisut, Daniel Peter. *The distance chemosensory behavior of the sea urchin *Lytechinus variegatus**. Diss. Georgia Institute of Technology, 2004.
- Rose, C. D., et al. "Overgrazing of a large seagrass bed by the sea urchin *Lytechinus variegatus* in Outer Florida Bay." *Marine Ecology Progress Series* 190 (1999): 211-222.
- "St. Joseph's Bay Aquatic Preserve." *Florida Department of Environmental Protection*, 26 Oct. 2017,  
[floridadep.gov/fco/aquatic-preserve/locations/st-joseph-bay-aquatic-preserve](http://floridadep.gov/fco/aquatic-preserve/locations/st-joseph-bay-aquatic-preserve).  
Accessed 3 May

2018.

- Valentine, John F., et al. "Experimental evidence that herbivory increases shoot density and productivity in a subtropical turtlegrass (*Thalassia testudinum*) meadow." *Oecologia* 112.2 (1997): 193-200.
- Valentine, John F., and K. L. Heck. "The role of leaf nitrogen content in determining turtlegrass (*Thalassia testudinum*) grazing by a generalized herbivore in the northeastern Gulf of Mexico." *Journal of Experimental Marine Biology and Ecology* 258.1 (2001): 65-86.F
- Valentine, John F., and J. Emmett Duffy. "The central role of grazing in seagrass ecology." *SEAGRASSES: BIOLOGY, ECOLOGY AND CONSERVATION*. Springer Netherlands, 2007. 463-501.
- Valentine, John F., et al. "Role of sea urchin *Lytechinus variegatus* grazing in regulating subtropical turtlegrass *Thalassia testudinum* meadows in the Florida Keys (USA)." *Marine Ecology Progress Series* 200 (2000): 213-228.
- Vergés, Adriana, Teresa Alcoverro, and Javier Romero. "Plant defences and the role of epibiosis in mediating within-plant feeding choices of seagrass consumers." *Oecologia* 166.2 (2011): 381-390.
- Watts, Stephen A., James B. McClintock, and John M. Lawrence. "The ecology of *Lytechinus variegatus*." *Developments in Aquaculture and Fisheries Science* 32 (2001): 375-393.
- Zieman, J. C., S. A. Macko, and A. L. Mills. "Role of seagrasses and mangroves in estuarine food webs: temporal and spatial changes in stable isotope composition and amino acid content during decomposition." *Bulletin of Marine Science* 35.3 (1984): 380-392.