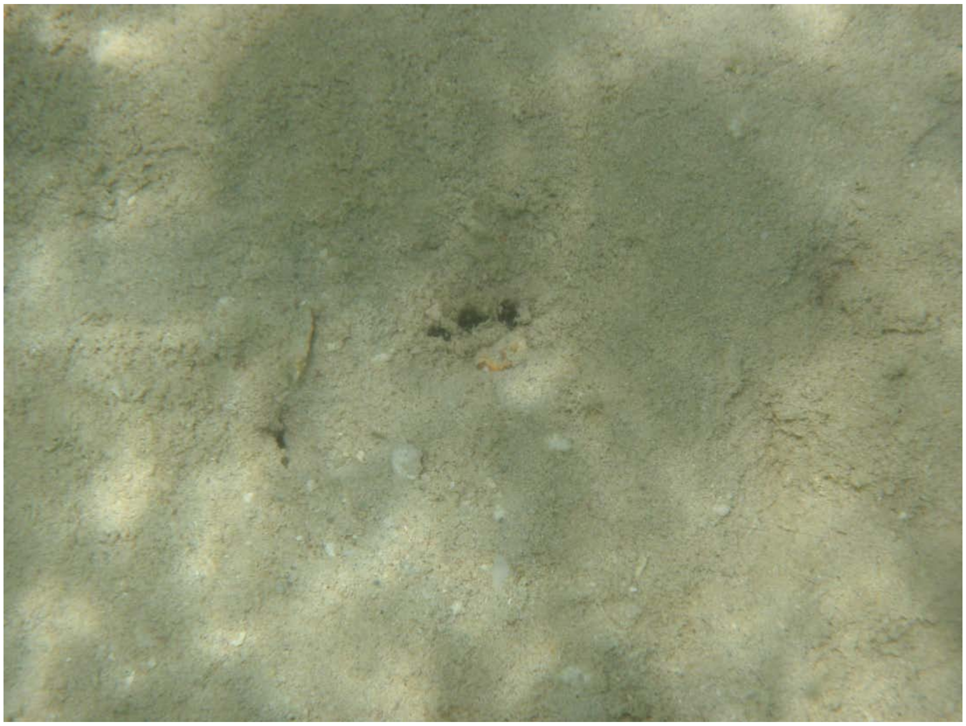
Effects of Salinity, pH, *Opheodesoma spectabilis* and *Gracilaria salicornia* on *L. reevii* Abundance in Kāne‘ohe Bay, Hawai’i (2012)



Cover photo: *L. reevii* siphon burrows in Kāne‘ohe Bay, Hawai‘i

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University of Hawaii at Manoa BIOL 403: Field Research Problems in Marine Biology

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Abstract

Lingula reevii is an inarticulated brachiopod found in Kāneʻohe Bay, Oʻahu, Hawaiʻi. Over the past 40 years, *L. reevii* has been consistently declining due to various anthropogenic factors and was added to the NOAA Species of Concern list in 2004. Field observations were conducted to determine the effects of salinity, pH, *Opheodesoma spectabilis* abundance and *Gracilaria salicornia* percent cover on *L. reevii* abundance. Transect and quadrat surveys were performed at the Sandbar, Pyramid Reef and Fringing Reef J to determine areas of high *L. reevii* abundance. Also, water samples were taken above *L. reevii* burrows to evaluate the influence of pH and salinity on *L. reevii* distribution. T-tests concluded significant differences in salinity and pH between the Sandbar and Pyramid Reef. An ANOVA was also used to determine a significant difference between *L. reevii* abundance and pH as well as *L. reevii* abundance and salinity at the Sandbar. It was concluded that the highest abundance of *L. reevii* was found on Pyramid Reef and abundance on the Sandbar is significantly less than 2010 and 2011 surveys. Future management efforts should include focusing resources on habitat preservation and water quality around Pyramid Reef and the Sandbar as well as continuing *L. reevii* abundance surveys to monitor populations.

Introduction

Lingula reevii, an inarticulated brachiopod, was established as a NOAA Species of Concern (SOC) in March 2004 (Hunter, *et al*., 2008). *L. reevii* is a cryptic organism that burrows vertically into sandy sediments along reef edges (Worcester, 1969). Habitat alteration, overexploitation, pollution and sedimentation, a vulnerable life history as well as limited distribution have led to reduced abundance of *L. reevii* in Kāneʻohe Bay, Hawaiʻi (NOAA SOC, 2007). Habitat degradation has been amplified by increases in human presence in shallow habitats that *L. reevii* is most often found (NOAA SOC, 2007). Sewage diversion as well as introduction of alien algae has led to a reduction in *L. reevii* habitat (NOAA SOC, 2007). *L. reevii* density has significantly declined over the past 50 years from 500 individuals/m^2 to less than 0.32 individual/m^2 in 2010 (Worcester, 1969; Hunter, *et al.*, 2010). Despite extensive surveys, *L. reevii* has not been found elsewhere in the Hawaiian Archipelago, although it has been identified in Ambon, Indonesia and Southern Japan (Cals and Emig 1979; Emig 1997).

L. reevii is identified by its oblong, bilaterally symmetrical turquoise shell (Emig, 1997) (Figure 1). *L. reevii* is characterized by three small holes in the sediment in which it feeds through a filtering lophophore to accumulate particulate matter (Worcester, 1969) (Figure 2).

Figure 1: *L. reevii* outside sediment

Figure 2: *L. reevii* siphon holes in sediment

L. reevii's primary food source consists of diatoms, filamentous algae, rotifers, copepods and oligochaetes (Emig, 1997). In order to understand factors that affect *L. reevii* abundance, water samples were collected to examine salinity and pH. Salinity was measured at sites of *L. reevii*'s presence to compare with Worcester's *in situ* salinity measurements (Worcester, 1969). pH was also measured to determine the presence of a pH gradient conducive to *L. reevii* habitats. Furthermore, the presence of two additional species, *Opheodesoma spectabilis* and *Gracilaria salicornia* were documented (Figure 3 and 4).

Figure 3: *Opheodesoma spectabilis*

Figure 4: *Gracilaria salicornia*

Opheodesoma spectabilis is a holothurian that was observed in high abundances around *L. reevii* habitat during initial Kāne‘ohe Bay surveys (Freeman, 1966). *Gracilaria salicornia,* an invasive alga introduced in the 1970s, has become increasingly abundant in *L. reevii* habitats in Kāne‘ohe Bay (Rodgers and Cox, 1999). *Opheodesoma spectabilis* and *G. salicornia* live in close proximity to *L. reevii* and were assessed to determine potential competitive interactions. The data gathered in this study will provide NOAA with a greater understanding of factors associated with *L. reevii*'s continued population decline and allow for more effective management as well as provide long-term density studies.

Materials & Methods

GPS coordinates were taken from Wedding, *et al. (*2011) and compared against current Google Earth images of Kāne‘ohe Bay to determine if the coordinates were in close proximity to viable *L. reevii* habitats. Three survey sites with known high *L. reevii* abundances were used throughout the duration of the study: the Sandbar, Pyramid Reef and Fringing Reef J (Figure 5).

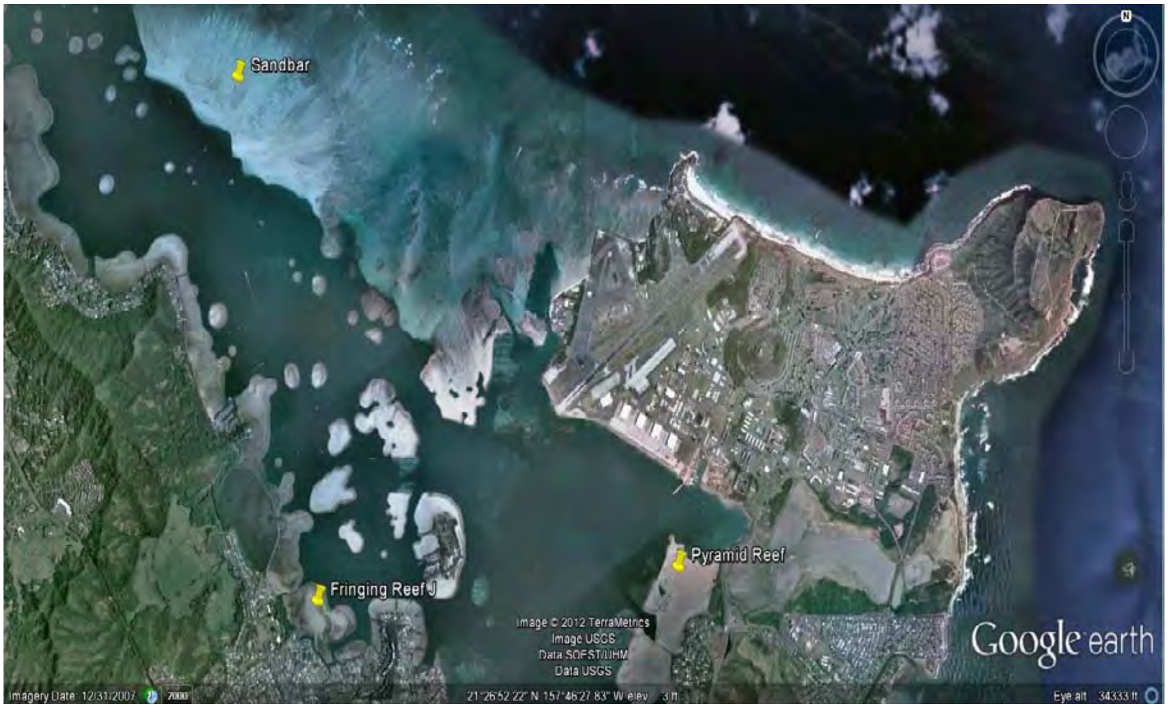

Figure 5: Survey Sites of known *L. reevii* abundance in Kāne‘ohe Bay, Oahu, Hawai'i

Five 50m transects were laid at both the Sandbar and at Pyramid Reef to determine areas of high *L. reevii* abundance (Figure 6 and 7). An additional three transects were laid at Fringing Reef J (Figure 8). Three snorkelers swam along the transects, one 5m to the left, one 5m to the right, as well as one directly above the transect to record *L. reevii* abundance. Once areas of high *L. reevii* abundance were determined, five 0.25m^2 quadrats were centered on *L. reevii* burrows (Figures 6, 7 and 8).

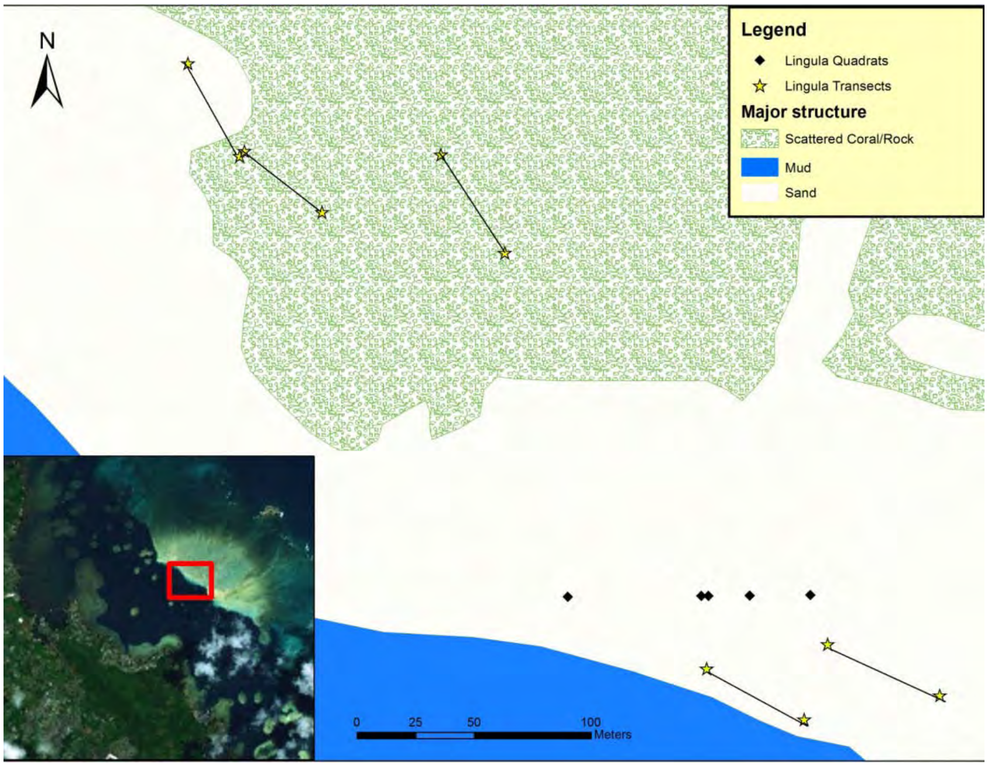

Figure 6: Location of transects and quadrats on the Sandbar.

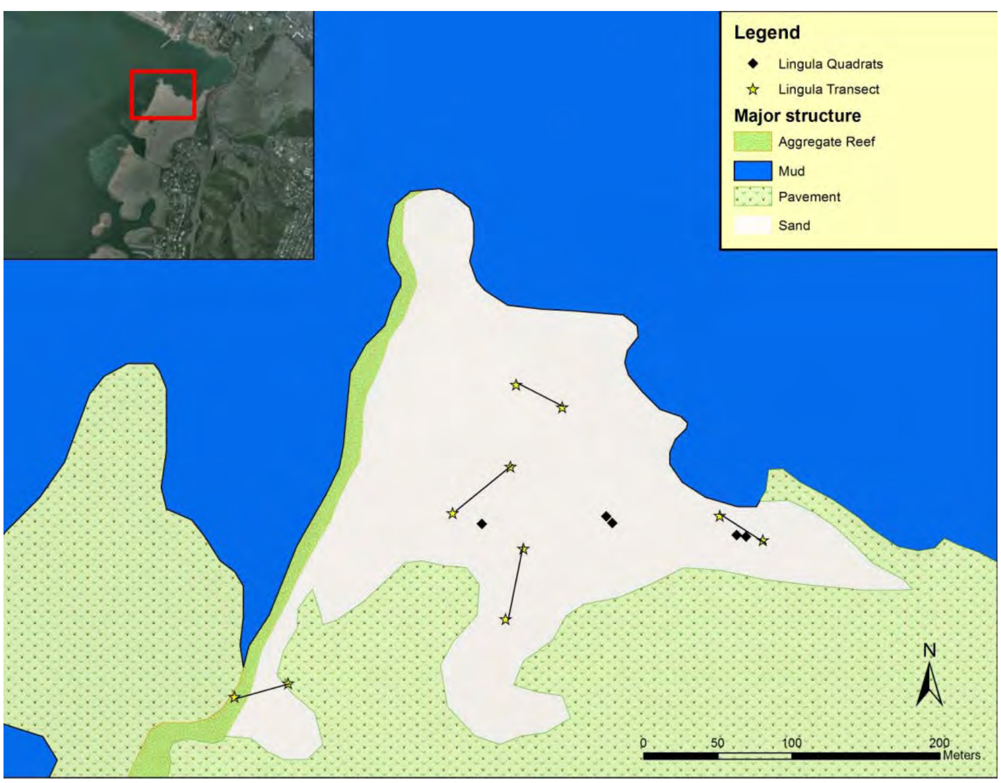

Figure 7: Locations of transects and quadrats on Pyramid Reef

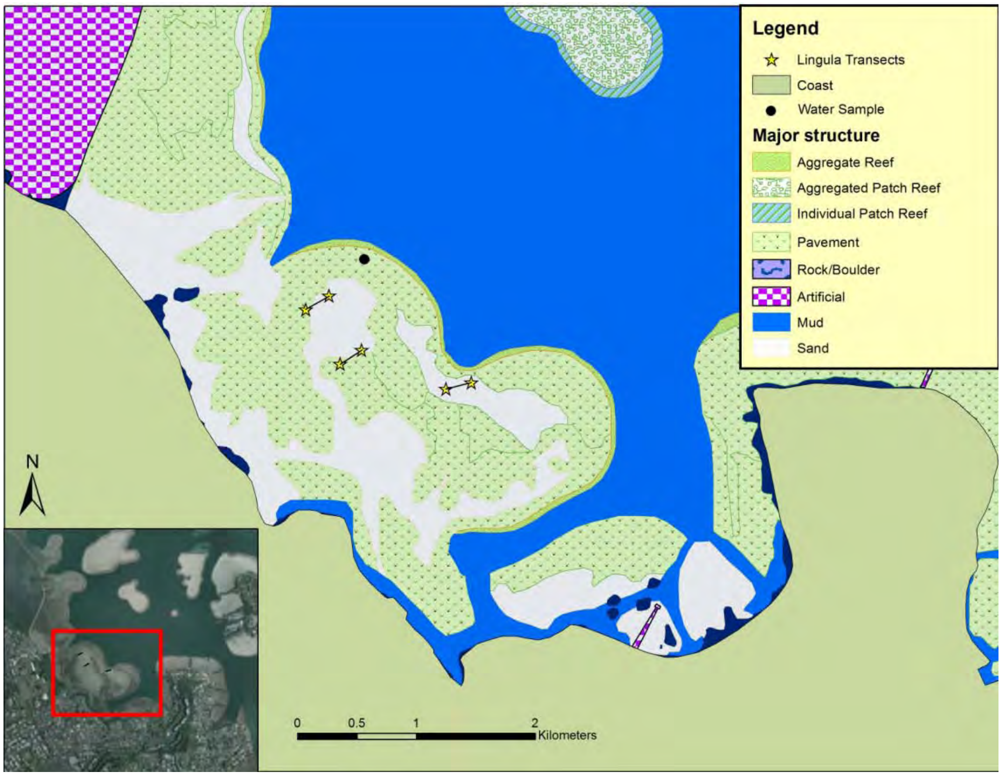

Figure 8: Locations of transect and quadrats on Fringing Reef J

Percent cover of *G. salicornia* and abundance of *O. spectabilis* were recorded at each quadrat to determine if there were any competitive interactions with *L. reevii*. Ten water samples were taken from the Sandbar and Pyramid Reef and two water samples were taken from Fringing Reef J. Eight samples were taken in close proximity of *L. reevii* burrows while two were taken where *L. reevii* were observed to be absent at each site to serve as controls. Salinity and pH were measured from the collected water samples. Salinity was recorded by using a refractometer and a pH meter was used to measure pH. *G. salicornia* percent cover and *O. spectabilis* abundance were assessed with 0.25m^2 quadrats. Salinity and *G. salicornia* percent cover was compared to Hunter *et al.* (2010). One-way ANOVAs were conducted to determine the significance of the relationships between *L. reevii* abundance and *O. spectabilis* abundance, pH, and salinity. T-tests were conducted to assess the significance of the pH and salinity between the Sandbar and

Pyramid Reef. A five minute delay was performed after the transect was laid so that *L. reevii* behavior was as close to natural as possible.

Results

A one-way ANOVA was performed to test whether there was a significant difference in *L. reevii* abundance at different salinities at the Sandbar and Pyramid Reef (Table 1). There was a statistically significant variation in *L. reevii* abundance at salinities of 35, 34.5, and 34 parts per thousand at the Sandbar (F=27.48, P=0.035) (Table 1). *L. reevii* abundance was greatest at a salinity of 35 ppt at the Sandbar, although there were individuals present in areas with 34 and 34.5 ppt (Figure 9). There was no statistically significant variation in *L. reevii* abundance at salinities of 36, 36.5, and 37 parts per thousand at Pyramid Reef (F=14.41, P=0.065) (Table 1). *L. reevii* were most abundant at a salinity of 36.5 ppt at Pyramid Reef (Figure 9). A one-way ANOVA was performed to test whether *L. reevii* abundance varied significantly with pH at the Sandbar and Pyramid Reef. *L. reevii* abundance varied significantly with pH ranging from 8.18 to 8.23 at the Sandbar (F=27.48, P=0.035) (Table 1). The greatest amount of *L. reevii* were found in areas within the Sandbar with a pH of 8.22 (Figure 10). *L. reevii* abundance did not vary significantly with pH ranging from 8.19 to 8.21 at Pyramid Reef (F=0.67, P=0.598) (Table 1, Figure 10).

A one-way ANOVA was performed to test whether there was a significant relationship between *L. reevii* abundance and *G. salicornia* percent cover. *G. salicornia* was only found at Pyramid Reef and was not seen on the Sandbar (Figure 11). There was no significant relationship between *G. salicornia* percent cover and *L. reevii* abundance (F=1.62, P=0.293) (Table 1). A one-way ANOVA was performed to test whether there was a significant relationship between *O. spectabilis* abundance and *L. reevii* abundance. *O. spectabilis* was only observed at the Sandbar

(Figure 12). There was no significant relationship between *O. spectabilis* abundance and *L. reevii* abundance (F=0.14, P=0.924) (Table 1). T-tests determined there was a significant difference in pH between the Sandbar and Pyramid Reef (P=0.01) (Table 2). A significant difference in salinity was found between the Sandbar and Pyramid Reef using a t-test (P<0.00) (Table 2). There was no *L. reevii*, *G. salicornia*, or *O. spectabilis* recorded on Fringing Reef J (Table 2). The mean pH measured at Fringing Reef J was less than the mean pH measured at the Sandbar and Pyramid Reef (Table 1 and 2). The mean salinity measured at Fringing Reef J was greater than the mean salinity at the Sandbar and Pyramid Reef (Table 1 and 2).

Table 1: Mean, variance and p-values of *L. reevii* abundance, *O. spectabilis* abundance, *G. salicornia* percent cover pH and salinity from T-test conducted using water samples from the Sandbar and Pyramid Reef. Significant p-values for *O. spectabilis* abundance, *G. salicornia* abundance, pH and salinity are in bold.

	Location	Sandbar	Pyramid
Variables			
Lingula Abundance	Total	13	37
Opheodesoma Abundance	Mean	2.6	None Found
	Variance	2.302	
	P-Value	**0.924**	
Gracilaria Percent Cover	Mean	None Found	3%
	Variance		0.067%
	P-Value		**0.293**
pH	Mean	8.208	8.026
	Variance	0.026	0.084
	P-Value	**0.035**	**0.598**
Salinity	Mean	34.4	36.5
	Variance	0.418	0.354
	P-Value	**0.035**	**0.007**

Table 2: P-values calculated using T-test for pH and salinity

between the Sandbar and Pyramid Reef.

	Location	Sandbar/Pyramid Reef
Variables		
pH	**P-Values**	**0.01**
Salinity	**P-Values**	**<0.00**

Table 3: *L. reevii* abundance, *O. spectabilis* abundance, *G. salicornia* percent cover as well as pH and salinity mean at Fringing Reef J.

	Location	Fringing Reef J
Variables		
Lingula Abundance		0
Opheodesoma Abundance		0
Gracilaria Percent Cover		0
pH Mean		8.10
Salinity Mean		35.5

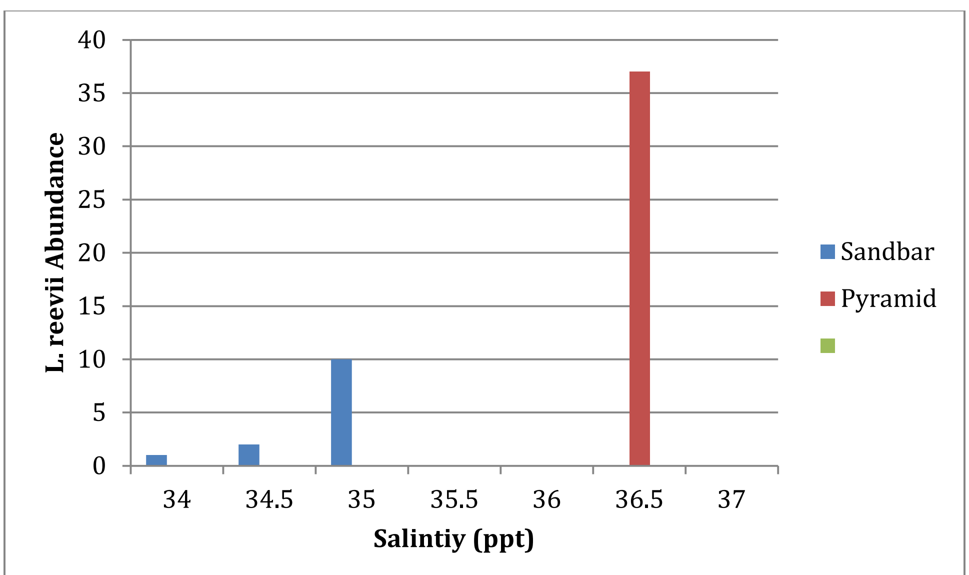

Figure 9: *L. reevii* abundance (m^2) at varying salinities in the Sandbar and Pyramid Reef

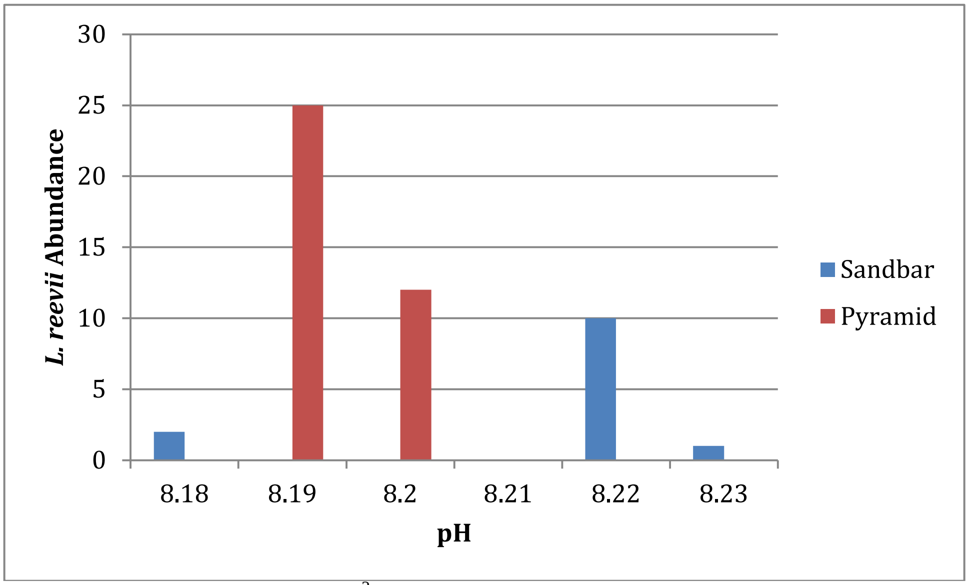

Figure 10: *L. reevii* abundance (m^2) varying with pH at the Sandbar and Pyramid Reef.

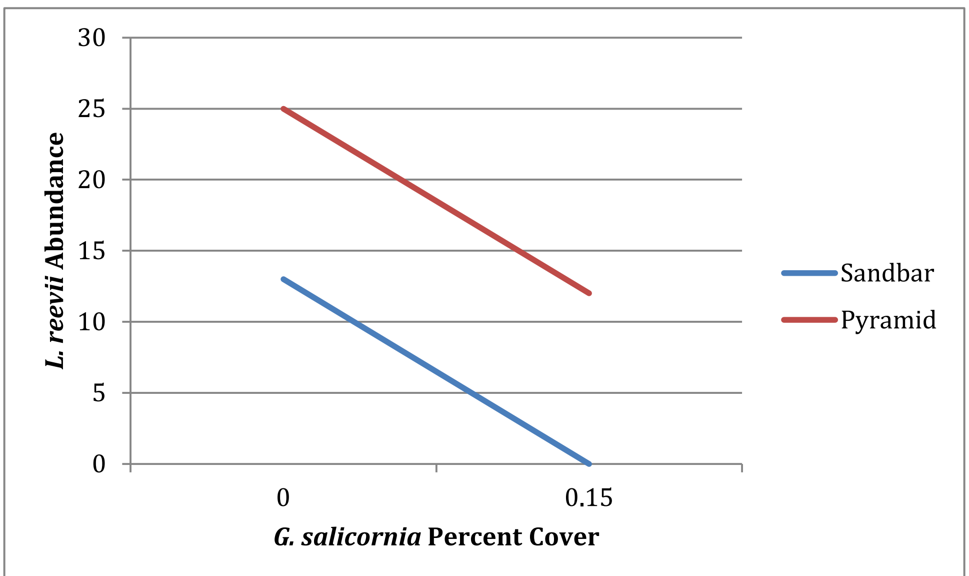

Figure 11: *L. reevii* abundance (m^2) in comparison to *G. salicornia* percent cover on the Sandbar and Pyramid Reef

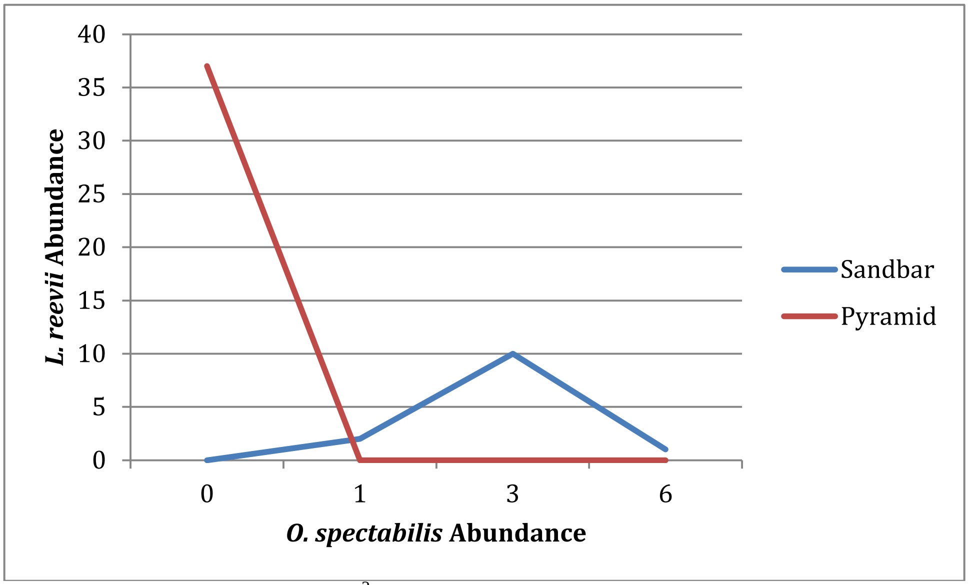

Figure 12: *L. reevii* abundance (m^2) in comparison to *O. spectabilis* abundance on the Sandbar and Pyramid Reef

Discussion

No *L. reevii* were recorded via transect surveys or *in situ* observations at Fringing Reef J. No quadrats were placed at Fringing Reef J since no *L. reevii* were found so *O. spectabilis* abundance and *G. salicornia* percent cover were not recorded (Table 2). Fringing Reef J was highly overgrown with mats of *Eucheuma/Kappaphycus* that covered much of the benthos. This high level of mat-forming algae could be directly affecting *L. reevii*. The brachiopod retracts into the sediment at the slightest disturbance of the surrounding benthos such as large mats of algae dragging over its siphon holes. This could increase the time *L. reevii* is retracted into the sediment and decrease the time spent filter feeding. Even if the high *Eucheuma/Kappaphycus* abundance is not directly affecting *L. reevii*, its presence could indicate environmental factors that are not favorable for the brachiopod. Sand patches that were relatively free of algae still did not have any *L. reevii* abundance which may show that *Eucheuma/Kappaphycus* is simply an indicator species of unfavorable *L. reevii* conditions. No quantifiable data was gathered on *Eucheuma/Kappaphycus* percent cover but *in situ* observations have shown that the brachiopod and algae did not co-occur, at least in noticeable abundances. Survey methodology that was conducted at other locations involved taking water samples at quadrats. Since no quadrats were placed, only the two control water samples were taken from Fringing Reef J.

Worcester (1969) found through experiments conducted in a laboratory setting that optimum salinities for *L. reevii* ranged from 30-35 ppt. Salinities higher than 35 ppt were not tested in the lab. These findings were corroborated through *in situ* work that showed *L. reevii* exists at salinities of 34-36.5 ppt. Salinities ranged to 37 ppt but no *L. reevii* were found at that salinity.

Salinity and pH were both significantly related to *L. reevii* abundance at the Sandbar but not at Pyramid Reef. This discrepancy may be due to the challenges of working with a cryptic species that has a dwindling population. Small sample sizes have led to disparate results and have been difficult to accurately analyze. *L. reevii* was present across a considerable range of pH values, which suggests that *L. reevii* has a tolerance to varying pH. Fringing Reef J was the closest survey site to land, so freshwater input as well as runoff could explain the low pH values that were measured.

There was no significant relationship between *O. spectabilis* abundance and *L. reevii* abundance. The sea cucumber, while present in large numbers at the Sandbar, was not present at Pyramid Reef while *L. reevii* was present at both. An unobserved competitive interaction between *O. spectabilis* and *L. reevii* may exist, but abundance data shows that *L. reevii* are present regardless of whether the sea cucumbers are present.

There was no significant relationship between *G. salicornia* percent cover and *L. reevii* abundance which is consistent with previous studies carried out by Hunter, *et al.,* (2010). The algae were only recorded at Pyramid Reef although *in situ* observations at Fringing reef J saw *G. salicornia* present.

Conclusion

Maps were generated through ArcGIS that depict areas of relatively high *L. reevii* abundance at the Sandbar and at Pyramid Reef. These areas are not all inclusive but do provide future studies with a narrowed search area to focus their efforts. The Sandbar and Pyramid Reef had significantly different salinities and pH. This indicates tolerable ranges of pH and salinity for *L. reevii* since relatively high abundances were found at both locations. Although difficult to measure, a factor that may have a large amount of influence on *L. reevii* abundance is the amount of human traffic and walking on the benthos that occurs. The brachiopod was not found in the shallow areas of the Sandbar where tourism operations often unload customers. Data from previous studies has shown that *L. reevii* numbers continue to decline at the Sandbar (Hunter, *et al.*, 2010; Wedding, *et al.*, 2011). Conversely, Pyramid Reef has few to no people walking around on the sand and relatively high *L. reevii* abundance. Management should be aware that *L. reevii* populations are not facing competitive interactions from *O. spectabilis* or *G. salicornia*. Salinity falls within optimal ranges of the brachiopod. Increasing human presence at the Sandbar is occurring in the same area as *L. reevii* habitat. Management resources should be allocated to the preservation of *L. reevii* habitat and water quality at the Sandbar and Pyramid Reef. Additionally, further *L. reevii* abundance surveys should be conducted to monitor population size.

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Appendix

For future management implications we have created maps of the Sandbar and Pyramid Reef of high *L. reevii* abundance. GPS coordinates are provided with the maps to aid in habitat preservation, future research and reduce search effort for *L. reevii*.

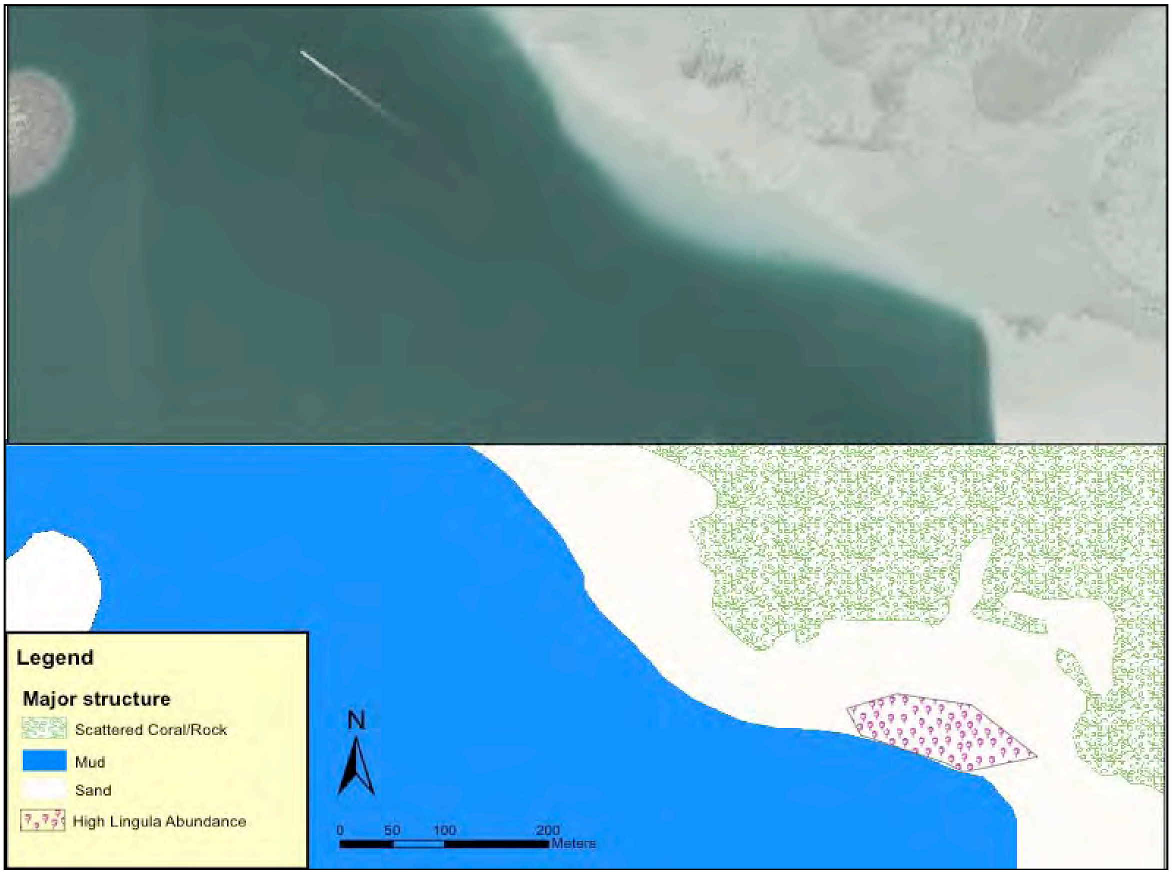

Figure 13: Sandbar management area. GPS Start: 21.46707, -157.81288; GPS End: 21.46671, -157.81267.

Figure 14: Pyramid Reef management area. GPS Start: 21.43246, -157.76341; GPS End: 21.43261; -157.76369.