

**North Carolina Sea Grant and Albemarle-Pamlico National Estuary Partnership
2022 Final Report Narrative**

Recipient: Stacy Trackenberg, PhD Candidate, Department of Biology, East Carolina University
Project Number: 2020-R/MG-2010
Project Title: Assessing Faunal Community Composition in Newly Restored Seagrass Beds Across a Depth Gradient
Project Award Period: 01-01-2021 – 12-31-2022
Total Expenditures: \$10,000
Total Remaining Balance: \$0

A no cost extension has been approved for this grant as is reflected in a revised timeline for years 1 and 2 (Tables 1 & 2)

Table 1. Revised project timeline based on project progress for year 1 (2021)

<u>Task</u>	<u>Jan</u>	<u>Feb</u>	<u>Mar</u>	<u>Apr</u>	<u>May</u>	<u>Jun</u>	<u>Jul</u>	<u>Aug</u>	<u>Sep</u>	<u>Oct</u>	<u>Nov</u>	<u>Dec</u>
Site selection	X	X	X									
Pre restoration marine fauna monitoring												
Seagrass restoration				X	X							
Deployment of SMURFs						X	X	X	X			
Post-restoration marine fauna monitoring						X	X	X	X	X		
Sample donor bed fauna						X	X	X	X	X		
Characterize seagrass metrics post-restoration					X	X	X	X	X	X		
Characterize seagrass metrics of donor beds					X							
Complete data analysis and submit final report										X	X	X

Table 2. Revised project timeline based on project progress for year 2 (2022; no cost extension)

<u>Task</u>	<u>Jan</u>	<u>Feb</u>	<u>Mar</u>	<u>Apr</u>	<u>May</u>	<u>Jun</u>	<u>Jul</u>	<u>Aug</u>	<u>Sep</u>	<u>Oct</u>	<u>Nov</u>	<u>Dec</u>
Deployment of SMURFs						X	X	X	X			
Post-restoration marine fauna monitoring					X	X	X	X	X	X		
Sample donor bed fauna					X	X	X	X	X	X		
Characterize seagrass metrics post-restoration				X	X	X	X	X	X	X		
Complete data analysis and submit final report	X	X	X	X	X	X	X	X	X	X	X	X

Introduction

Seagrass habitats are beneficial to overall coastal ecosystem health due to the provision of services such as nutrient cycling, sediment stabilization, improvement in water quality and habitat provision. Globally, seagrasses have declined by roughly 30% since initial records in 1879 (Waycott et al. 2009). In North Carolina seagrass beds have generally performed well, however they face threats such as climate change, nutrient-loading from runoff, and increased storminess (NC Department of Environmental Quality 2021). This has led to declines of over 10% of high salinity submerged aquatic vegetation in southern areas of the state between surveys conducted in 2006/2007 and 2013 (Field et al. 2021, NC Department of Environmental Quality

2021). Therefore, the need to develop restoration alternatives to proactively reduce bed losses rather than reactively restore lost or degraded seagrass beds is clear. Seagrass restoration is conducted through seed planting or adult transplanting and has been used to combat these losses globally and within North Carolina estuaries.

Depth is an important environmental variable in predicting fish species richness and density (França et al. 2012, Whitfield 2017). Seagrasses can only exist within a specific depth window due to their light requirements (Duarte 1991, Fonseca and Bell 1998, Duarte et al. 2007), however differences between water depths within this window may be important for seagrass characteristics. Water depth is influenced by the elevation of the seagrass bed and the tidal cycle of the area at the time of measurement. In some areas, species richness is higher in shallower beds and assemblage structure is distinct across depths (Hutchinson et al. 2014), with intertidal seagrass beds acting as an important nursery area (Madi Moussa et al. 2020). At other locations, deeper seagrass beds have more fish than shallower meadows and deeper areas showed a much larger difference in abundances between vegetated and unvegetated habitats than shallower areas (Jenkins et al. 1997). Access to the habitat is also influenced by depth as intertidal seagrass beds will be exposed to the air at low tides, limiting which organisms can remain in the habitat. Low tides occur twice a day in a diurnal tide system such as North Carolina. It is imperative to understand how depth influences restoration and the faunal communities to ensure proper restoration placement and ability to meet goals of faunal recruitment.

North Carolina seagrass meadows are comprised of a mix of one to up to three of the seagrass species, *Halodule wrightii*, *Ruppia maritima*, and *Zostera marina*. Previous restoration efforts in North Carolina's high salinity waters have focused on restoring *Zostera marina*, a species at its southern-most extent in North Carolina. In North Carolina, *Z. marina* reproduces through a mix of annual and perennial reproductive strategies based on location (Jarvis et al. 2012), allowing restoration to occur through transplanting or planting of seed (Zhang et al. 2021). Alternatively, *H. wrightii* is predominantly clonal in growth in North Carolina, with few documentations of flowering plants and low genetic diversity indicating high rates of cloning (Ferguson et al. 1993, Fonseca and Bell 1998, Digiantonio et al. 2020). Therefore, transplanting adult *H. wrightii* shoots is a practical method as seed collection is likely impossible. Due to the range and heat tolerance limitations of *Z. marina*, *H. wrightii* is likely to become increasingly dominant with climate change. Understanding best practices for restoring *H. wrightii* through transplanting is critical should temperatures warm above the tolerance threshold for *Z. marina*, leading to its loss.

We addressed the questions: (1) how does community composition of restored seagrass beds differ between restoration sites of varying depths and (2) how does community composition of restored seagrass beds compare to a nearby natural seagrass bed and nearby bare sand areas. We hypothesize species richness and abundance of fish in intertidal beds will be lower when compared to subtidal beds due to reduced accessibility of the shallower habitat to fish. Additionally, we hypothesize shallower beds will have shorter canopy heights, which will also contribute to lower species richness and faunal abundance when compared to deeper restored beds. We also hypothesize faunal communities in restored plots will be more similar to those in natural seagrass beds than bare sand control plots due to the addition of structure which provides refuge for fauna.

Methods

To evaluate the effects of elevation on seagrass restoration success and faunal community composition, we conducted a field seagrass restoration experiment in Back Sound, North Carolina, United States. In the spring of 2021, we transplanted *H. wrightii* shoots to two elevations within our study location (Figure 1). In April 2021, we used a Trimble R10 RTK GPS to identify sites within our two elevation categories: shallow subtidal (-0.6 m – -0.8 m NAVD88) and intertidal (-0.4 m – -0.55

m NAVD88). Intertidal plots were selected such that they would be exposed at spring tides but remain submerged during neap tides. Elevations were determined based on elevations of natural seagrass beds in Back Sound, NC measured during our observational field experiments during the summer 2020. These elevations were chosen to ensure seagrass can, and does, persist in Back Sound at the chosen elevations. We additionally chose these elevations as they were not the deepest or shallowest elevations of the natural beds to limit the light and temperature stress to the grass after transplantation.

We conducted our restoration April 26th – 28th, 2021. We restored 2 m by 2 m plots of *H. wrightii* in a grid arrangement (Figure 2) using 23 clumps comprised of 15 shoots of *H. wrightii* collected from a nearby (<400 m) seagrass bed (Figure 3). Each clump was on average 0.622 ± 0.056 g and care was taken to ensure intact roots and rhizomes were included within each clump rather than individual, unconnected shoots of *H. wrightii*. Clumps were held down with lawn staples to ensure they were not easily washed away. Due to high winds during our initial planting, we saw a high loss of transplants rapidly at our site. Therefore, we conducted supplemental planting on May 25th, 2021 to replace all lost clumps and ensure 23 total clumps within each plot. In total we restored 11 plots: 5 intertidal and 6 shallow subtidal. An additional 11 bare sand plots (N=5 shallow subtidal, N=6 intertidal) were delineated interspersed with the restoration plots and 5 seagrass reference plots located in the adjacent continuous seagrass bed were marked. All seagrass reference plots were at the intertidal elevation and both bare sand and seagrass reference plots were delineated as 2 m by 2 m.

Bi-monthly from June through October 2021 we counted the number of remaining clumps transplanted in each plot out of the original 23 to determine the proportion remaining in each treatment group as a proxy for patchiness of the seagrass. During our bi-monthly seagrass checks we deployed baited minnow traps in all 27 restored and control plots. We baited the traps with two pieces of dry dog food. Traps were weighted down with pieces of brick and placed in the center of all plots, being careful to avoid placing the brick on seagrass transplants. Traps were deployed within two hours of low tide during the neap tide. Traps were deployed for 24 hours, at which point all fish and crustaceans within traps were identified to the lowest taxonomic level, enumerated, and weighed before being released (Table 1). We standardized the catch in each minnow trap to 24 hours to account for any differences in length traps were deployed. This was quantified as catch per unit effort (CPUE).

Concurrently with our minnow trap deployment we conducted 24-hour squid-pop consumption assays (Duffy et al. 2015). Squid-pops were comprised of a 1.3 cm diameter circular piece of dried squid attached to a garden stake with 5 cm monofilament line (Duffy et al. 2015, Lefcheck et al. 2021). Squid-pops were deployed so that squid was placed 20 cm above the substrate and deployed one hour prior to low tide. Consumption was checked one-, two-, and 24 hours post deployment. Consumption was rated as consumed, partially consumed or intact. Any active predation was also noted. The proportion consumed in each plot across all sampling dates were averaged and the average for each treatment was calculated.

Finally, we conducted two-week deployments of standardized monitoring units for the recruitment of fish (SMURFs; Figure 4) each month to capture larval and juvenile fish and crustaceans entering the study site (Ammann 2004). SMURFs were deployed between plots in the subtidal and within intertidal seagrass reference plots. SMURFs could not be placed within restoration plots as their large size and method of deployment (sand screws) would negatively impact restored plots. SMURFs were constructed with 1.2 x 0.75 m sections of 2.5 cm VEXAR mesh rolled into a cylinder, ends folded over, and closed with zip-ties. Within each tube a 2.5 x 1.2 m section of plastic mesh with 5.0 x 7.5 cm grid was haphazardly folded and inserted with 6 fine mesh onion sacks cut in half placed haphazardly amongst the larger mesh. Once constructed ½ inch

twisted polypropylene rope was added to each SMURF to allow a carabiner to attach to the center of the “bottom” of the SMURF. Buoys were attached with 3/16 inch braided polypropylene rope to the top of the SMURF to ensure flotation above the sediment. After the two-week deployment SMURFs were removed from the water and placed into larger plastic sleds. SMURFs were rinsed with water, and all organisms removed prior to placement in labelled jars for later identification. All organisms collected from SMURFs were identified to the lowest taxonomic level (Table 2), enumerated, and the first 10 of each species per SMURF were measured to the nearest mm (total length for fishes and shrimp; carapace width for crabs; Table 3).

Statistical Analyses

To ensure there were differences between our “shallow” and “deep” plots we conducted two-way analysis of variance (ANOVA) models with the fixed effect of elevation category on the measured elevation of the plots. To ensure there were no differences in elevation between our restored and control plots in the shallow elevations we conducted an ANOVA with the fixed effect of restoration category (restored vs. bare sand vs. seagrass) on the measured elevation of the plots. To ensure there were no differences in elevation between our restored and control plots in the deep elevations we conducted an ANOVA with the fixed effect of restoration category (restored vs. bare sand) on the measured elevation of the plots.

We used a linear mixed model to analyze the fixed effect of plot type (deep vs. shallow), date, and their interaction on the number of clumps remaining during 2021 checks. We included plot number as a random effect. We also used a linear mixed model to analyze the fixed effect of date on the number of clumps remaining in shallow plots in 2022. Plot number was once again included as a random effect. We conducted a one-way ANOVA to determine the effects of plot type (continuous seagrass, intertidal transplanted seagrass, subtidal transplanted seagrass, intertidal bare sand, and subtidal bare sand) on the CPUE of minnow trap catch between our continuous seagrass and shallow and deep restored and bare sand plots. We included the fixed effect of plot type on the measured CPUE. We then used a Tukey’s post-hoc test to assess pairwise differences for any significant result of our ANOVA. A generalized linear model was used to analyze the fixed effects of elevation category, number of clumps remaining in patchy seagrass, and their interaction on the CPUE of our minnow traps. We only included plots that contained patchy grass as we wanted to directly compare levels of patchy seagrass rather than patchy and bare.

Generalized linear models were used to analyze (1) the fixed effects of restoration category (restored vs. bare sand control) and elevation category (shallow vs. deep) on proportion of squid-pops consumed at one and two hours for our restored and bare sand control plots and (2) the fixed effect of habitat type (continuous seagrass, bare sand, restored/patchy seagrass, and oyster reef) on proportion of squid-pops consumed at one and two hours post deployment for all shallow plots. Tukey’s post-hoc tests were used to assess pairwise differences for any significant treatment or interaction effects from our models.

Results

There is a statistically significant difference in elevation, with our “shallow” plots having an elevation of -0.501 ± 0.007 m NAVD88 and “deep” plots have an elevation of -0.684 ± 0.018 m NAVD88 ($p < 0.001$; Table 4). There was no difference between our shallow restored, continuous seagrass, and bare sand control plots ($p = 0.610$; Table 5) or our deep restored and bare sand control plots ($p = 0.763$; Table 6; Figure 5). The number of transplanted *H. wrightii* clumps declined over time from 23 clumps on May 25, 2021, to 8.4 ± 2.8 clumps in intertidal plots and 1.3 ± 0.8 clumps in subtidal plots on October 5, 2021. There was a statistically significant difference in number of clumps remaining based the interaction between date and plot type with intertidal plots declining

slower than subtidal plots over the 2021 sampling period ($p < 0.001$; Table 7; Figure 6). In 2022, we continued to monitor only the intertidal plots as no subtidal plots contained transplanted clumps by April 2022. We saw a significant effect of date on the number of clumps remaining over time with clumps falling from 8.4 ± 2.8 clumps on June 8, 2022, to 0.8 ± 0.8 clumps remaining on October 4, 2022 ($p < 0.001$; Table 8; Figure 6).

There is a statistically significant difference in minnow trap CPUE across our sample plots with continuous seagrass plots having a CPUE of 18.1 ± 0.8 organisms, higher than all other plot types with deep bare sand plots having a CPUE of 1.2 ± 0.3 organisms, deep restored plots having a CPUE of 1.8 ± 0.4 organisms, shallow bare sand plots having a CPUE of 2.6 ± 0.4 organisms, and shallow restored plots having a CPUE of 1.6 ± 0.6 organisms ($p < 0.001$; Figure 7; Table 9, 10). When comparing the CPUE of plots based on the number of clumps remaining in restored plots, we found an increase in CPUE as number of clumps remaining increased ($p = 0.003$), however there was no difference in CPUE between shallow and deep plots ($p = 0.874$) or the interaction of elevation and number of clumps remaining ($p = 0.087$; Figure 8; Table 11).

When comparing our squid-pop consumption, we broke comparisons into two categories: (1) restoration and depth (shallow restored, shallow bare sand, deep restored, deep bare sand) and (2) all intertidal habitat types (continuous seagrass, intertidal restored, intertidal bare sand, and oyster reef). When comparing consumption in our subtidal and intertidal restored and bare sand plots at one hour we found a statistically significant effect of elevation category ($p = 0.01322$) but not restoration category ($p = 0.108$) or the interaction between them ($p = 0.354$ Figure 9A; Table 12) on the proportion of squid pops consumed, with subtidal plots having more squid pops consumed than subtidal plots. When comparing only the intertidal plots (oyster, seagrass, restored, bare sand) we saw a marginally significant difference between all plots at one hour ($p = 0.057$; Figure 9C; Table 13). After two hours, we found a significant interaction between restoration and elevation categories ($p = 0.038$; Figure 9B) when comparing our shallow and deep restored and bare sand control plots, with shallow restored plots having lowest consumption compared to all other plot types (Tables 14, 15). When comparing our shallow plots alone there was a significant difference between plots due to the decreased consumption within intertidal restored plots ($p = 0.038$; Figure 9D; Tables 16, 17). All squid pops were consumed after 24 hours regardless of plot type, so no statistical models were run.

Discussion

Transplanted *H. wrightii* clump survival was higher post-transplanting in shallow, intertidal plots as compared to deeper, subtidal plots (Figure 6). While the mechanism for what caused the decline in subtidal plots post restoration is unknown, it could potentially be due to sedimentation noticed at the site, increased currents in deeper areas, or lowered light availability. Previous studies have found a depth threshold for seagrass restoration (Aoki et al. 2020), which may be different than the depth threshold for the naturally occurring seagrasses (Fonseca et al. 1998). It is unlikely that the depths chosen for restoration were beyond the natural depth threshold of *H. wrightii* in Back Sound, North Carolina. The depth limits of *H. wrightii* varies based on location, however natural seagrass beds containing *H. wrightii* in Back Sound and in the North River near our restoration site are found from intertidal to depths of 1 – 1.5m deep at low tide (Ferguson et al. 1993, Biber et al. 2008, Trackenberg et al. *in prep.*). These natural depths of *H. wrightii* are noticeably deeper than our chosen “deep” treatment of -0.684 ± 0.018 m NAVD88. Plantings are likely more stressed than established beds which may also rely on facilitation from adjoining shoots at the deep edge of natural beds (Fonseca et al. 1998), indicating not all depths found in natural seagrass beds are appropriate for restoration. While not measured, sedimentation was noted anecdotally in both 2021 and 2022, particularly throughout the end of August and all of September.

Lawn staples used to stabilize transplanted clumps were noted to be buried under the sediment when they initially were placed even with the sediments surface. Burial of *H. wrightii* leads to decreases in survival, particularly as the amount of the plant buried increases (Fonseca et al. 1998). The final potential mechanism to explain the loss of subtidal seagrasses is differences in current speed between the deep and shallow plots. Current speed was not measured, however the speed of currents can shape seagrass landscapes, with increased currents leading to decreased percent cover and seedling recruitment (Fonseca and Bell 1998, Fonseca et al. 1998).

While our subtidal restored plots did not persist, our intertidal restored plots continued to persist for over one year post restoration, with one plot still containing transplanted clumps 18 months post restoration. These results suggest that transplantation of *H. wrightii* for restoration may be successful at intertidal depths rather than subtidal despite naturally occurring at these depths. *Halodule wrightii* has the ability to colonize rapidly after a disturbance and at a large scale may be able to colonize new areas (Donaher et al. 2021), a potential mechanism for expansion into deeper, subtidal locations after successful establishment in intertidal areas. Our restoration was conducted at a small scale which may have resulted in our lack of persistence over time as larger scale restorations tend to be more successful (van Katwijk et al. 2016). The median monitoring time for restoration experiments is 12 months, with 67% of seagrass restoration trials containing seagrass after 12 months (van Katwijk et al. 2016). This is encouraging for our intertidal plots were on par with previous efforts, however we only had 52 shoots total remaining at 18 months as compared to a median of 720 shoots remaining across all successful restoration trials (van Katwijk et al. 2016). It is possible that restoring larger clumps or with facilitative species, such as clams, may provide better success in intertidal areas, or, enable success at subtidal elevations (Zhang et al. 2021).

Faunal communities in our restored plots did not reach abundances equal to those in the continuous seagrass bed. Rather, the restored plots had similar abundances to the bare sand control plots. We may be seeing such drastic differences in faunal abundances in the continuous seagrass and restored plots due to the large difference in cover, biomass, and surface area in the continuous seagrass bed as compared to our transplanted areas, which may then increase the beds performance as a nursery area (Orth et al. 1984, Hovel et al. 2002, McCloskey and Unsworth 2015). We also observed faunal abundance increase as number of clumps remaining in a plot increased. This indicates that while low-cover seagrass post-restoration does not match abundances in continuous meadows, a small increase in seagrass cover is important in structuring fish communities, regardless of depth. This further indicates that restoring at a larger scale may be important for habitat provisioning and successful restoration with the goal of restoring ecosystem function.

While we were unable to determine all consumers of our squid-pops, we frequently observed blue crabs or pinfish preying upon the squid-pops during the trials (Duffy et al. 2015, Rodemann and Brandl 2017). At one-hour post-deployment of squid-pops we saw a higher consumption of squid pops in deep plots compared to shallow plots, regardless of plot type. This pattern is likely explained by a lack of habitat access to the shallow plots at the one-hour sampling. We conducted predation assays within one hour of low tide, with our one-hour check occurring at or near low-tide. Fish may leave shallow areas at low tide to prevent stranding, even when beds are not completely exposed (Sogard et al. 1989). Lower water levels also tend to support higher abundances of prey fish as opposed to predators (Davis et al. 2017), which may explain a lack of consumption in shallow water habitats even when water is present. After low tide, as the water rises, fish may arrive back in shallow seagrass beds rapidly (Espadero et al. 2020), with predatory fish abundances rising later in the tidal cycle than prey fish (Davis et al. 2017) allowing higher consumption rates at our two hour check in shallow water. Interestingly, consumption was lowest in shallow, transplanted seagrass when compared to deeper transplanted seagrass, shallow and deep bare sand, seagrass, and oyster reef habitats at the two-hour check. These shallow areas with

transplants may play a role as refuges due to the lowered consumption as compared to other shallow water habitats.

While there are still unknowns regarding the most successful methods of restoring *H. wrightii* in North Carolina, restoring in shallow, intertidal areas using adult transplants may be a step towards recovering lost seagrass beds. Intertidal restoration when combined with methods known to enhance restoration success, such as harnessing both inter- and intraspecific facilitations (Zhang et al. 2021) and planting at a larger scale (van Katwijk et al. 2016) or with higher initial shoot densities (Sheridan et al. 1998) may further enhance the success of restoration. Future restorations, with increased transplant clump size, may be able to provide the critical habitat needed to support faunal communities equivalent to naturally occurring beds.

Outreach and Dissemination

During the project period I mentored an ECU undergraduate, Dawsyn Smith, who completed an Honors project focused on the role of depth and habitat type of the consumption of squid-pops. Dawsyn worked with us during her junior and senior years (2021-2022) including the summer of 2021 where she led the squid-pop consumption assay effort. Throughout her mentoring, Dawsyn learned background information including the role of seagrasses as habitat, their declines and subsequent restoration, and the role of habitat type in the consumption of assays. Dawsyn also learned ecological field methods, sample processing, and data collection both in the field and in the lab. Dawsyn was also mentored in statistical analyses in R programming where she conducted generalized linear models on consumption data and created figures for her honors thesis and our forthcoming publication. In addition to mentoring Dawsyn, I also mentored three technicians over the summer of 2022 who were invaluable in collecting data on the success of the restoration one year post restoration. These technicians included a current undergraduate at the University of Oregon, a recent graduate from North Carolina State University, and a current Masters student at Duke University. These technicians were mentored in project design, data collection, and statistical analysis.

Work from this project was presented at both local and national conferences including the 2022 North Carolina Coastal Conference, the 2022 Benthic Ecology Meeting in Portsmouth, New Hampshire, the Research and Creative Achievement Week at East Carolina University, and the ECU Biology Department's Research in Progress Seminar Series. These presentations included methods and results from our restoration funded through this fellowship. Presentations were well attended and well received by both researchers and practitioners.

Although we had created an activity for the Generating Equity in Science and Technology (GEST) event at UNC-IMS to work with local youth, the event was cancelled in 2021 and 2022 so we were unable to work with local students. I will be attending and working with the Scientific Research and Education Network to create a lesson plan and present to local educators this February, allowing lessons about coastal loss, restoration, and submerged aquatic vegetation to be taught to local K-12 students.

Finally, I am currently writing a manuscript focused on the work completed during this fellowship. This manuscript will highlight the results of the restoration effort as well as the faunal monitoring across depths and habitat types.

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Figures



Figure 1. Map of restoration site. White dots represent intertidal restored (N=5), subtidal restored (N=6), intertidal bare sand control (N=6), subtidal bare sand control (N=5), and seagrass control plots (N=5).

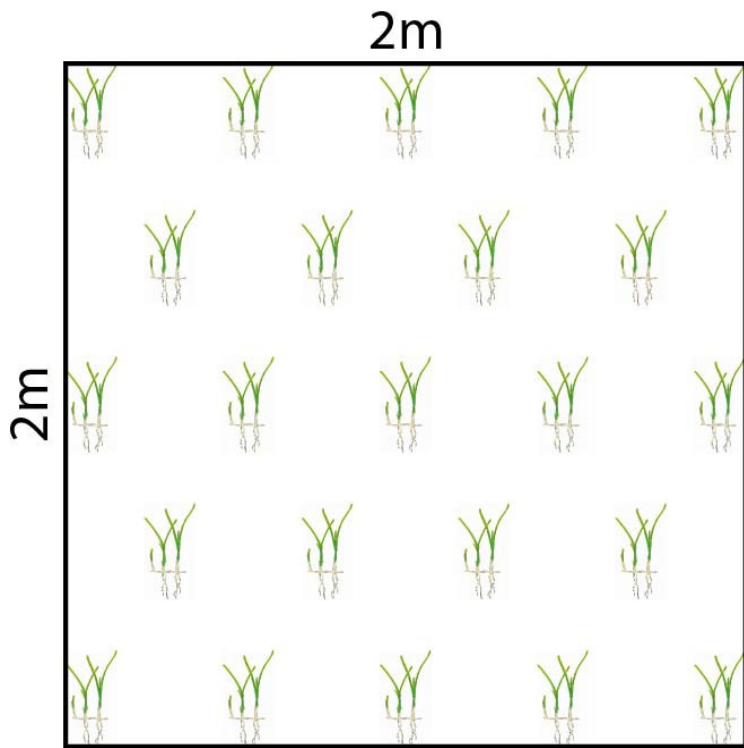


Figure 2: Diagram representing the planting scheme of *H. wrightii* in the manipulative field experiment.

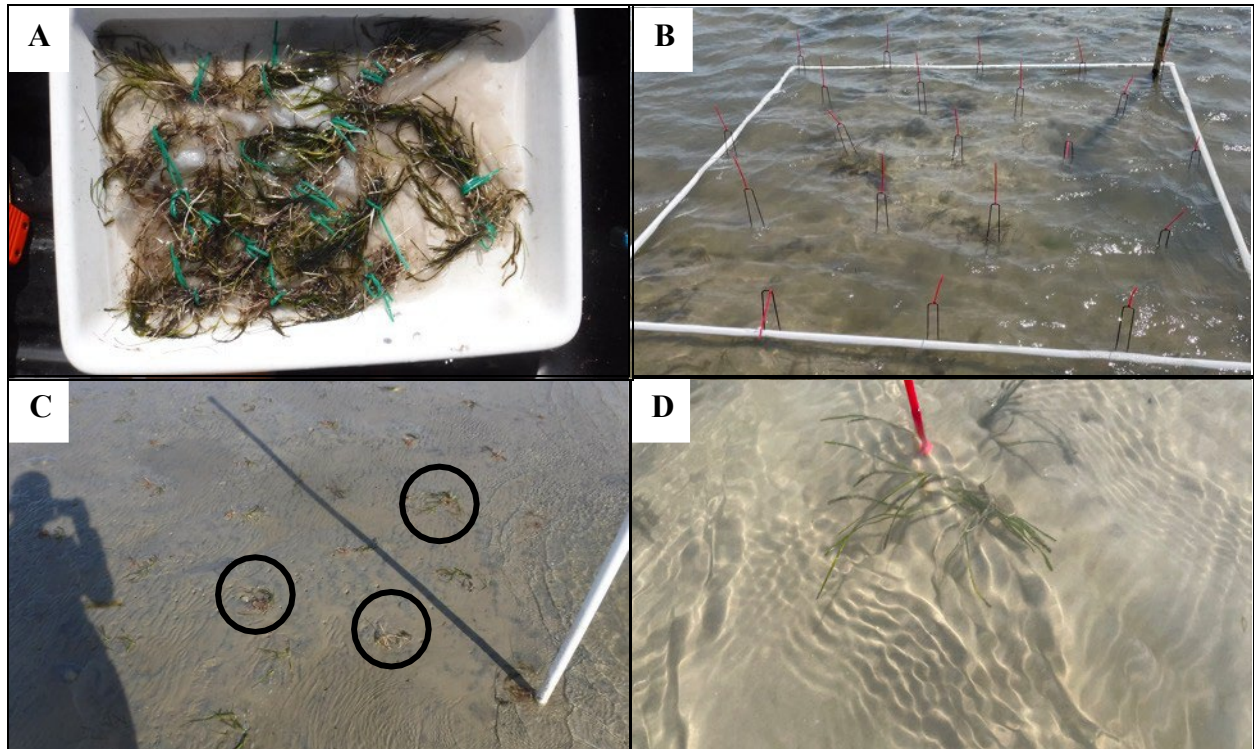


Figure 3. Clumps of 15 shoots of *H. wrightii* to be planted (A) in 2 m x 2 m plot with 23 clumps total marked by lawn staples (B). A restored plot after planting with a few clumps circles for ease of view (C) and a close-up of a planted clump with all roots and rhizomes covered with sand (D).



Figure 4. A standardized monitoring unit for the recruitment of fish (SMURF) deployed in a seagrass control plot.

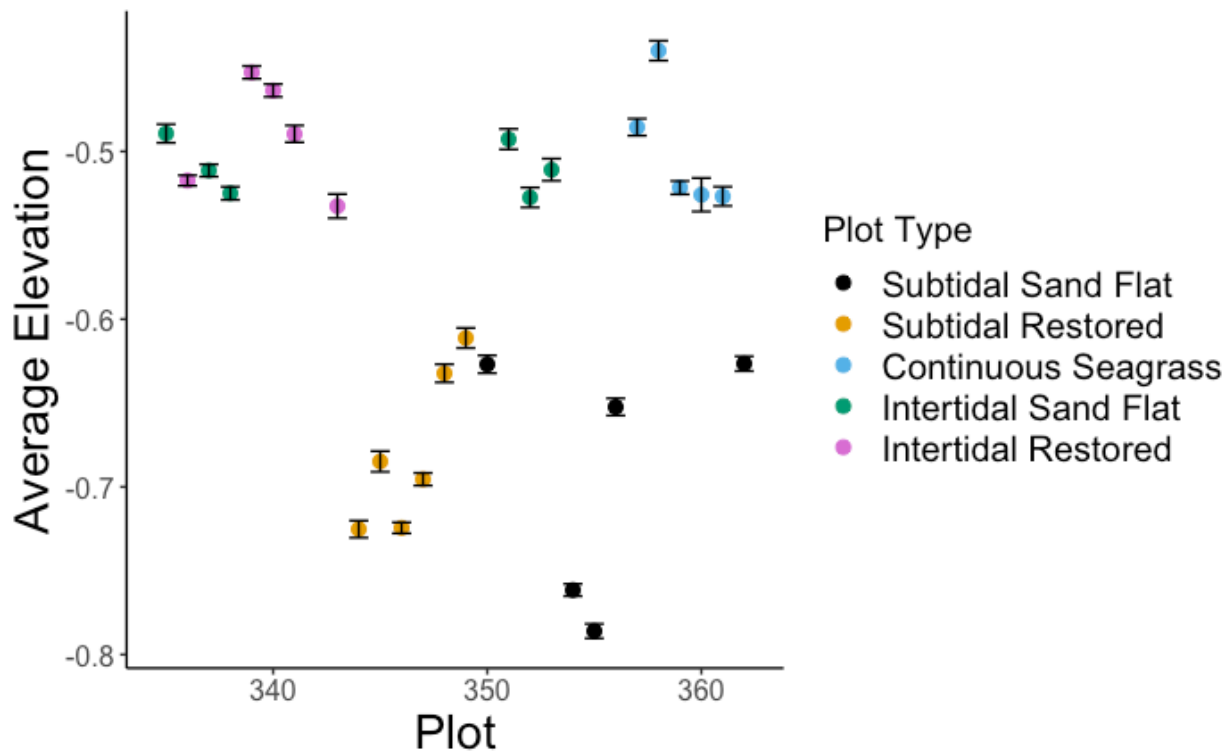


Figure 5: Elevation of each plot in our manipulative field experiment – note that there is no difference within the intertidal or subtidal categories but a difference between the intertidal and subtidal. Error bars denote standard error.

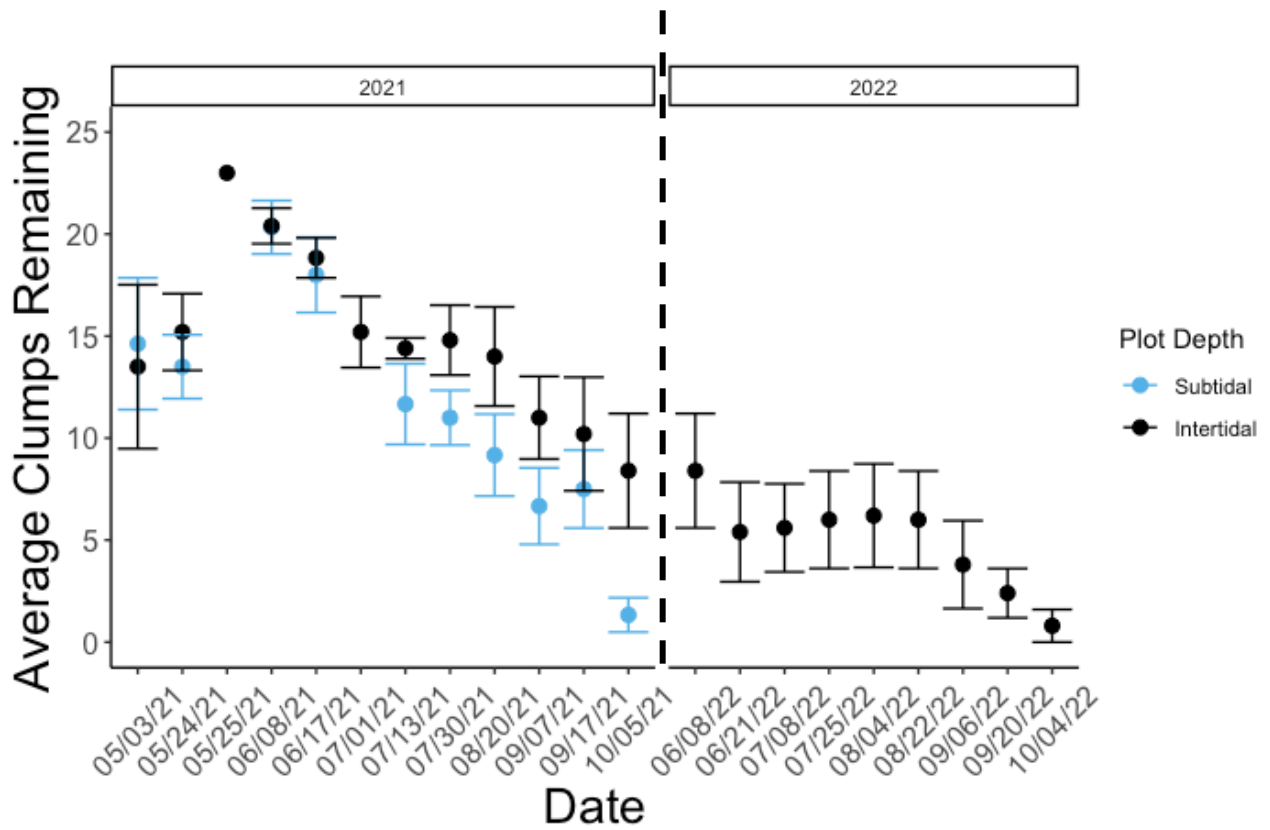


Figure 6. Mean number of clumps remaining for subtidal (blue; n=6) and intertidal (black; n=5) plots over time in 2021 and 2022. Dashed line separates 2021 and 2022 data. Error bars denote standard error.

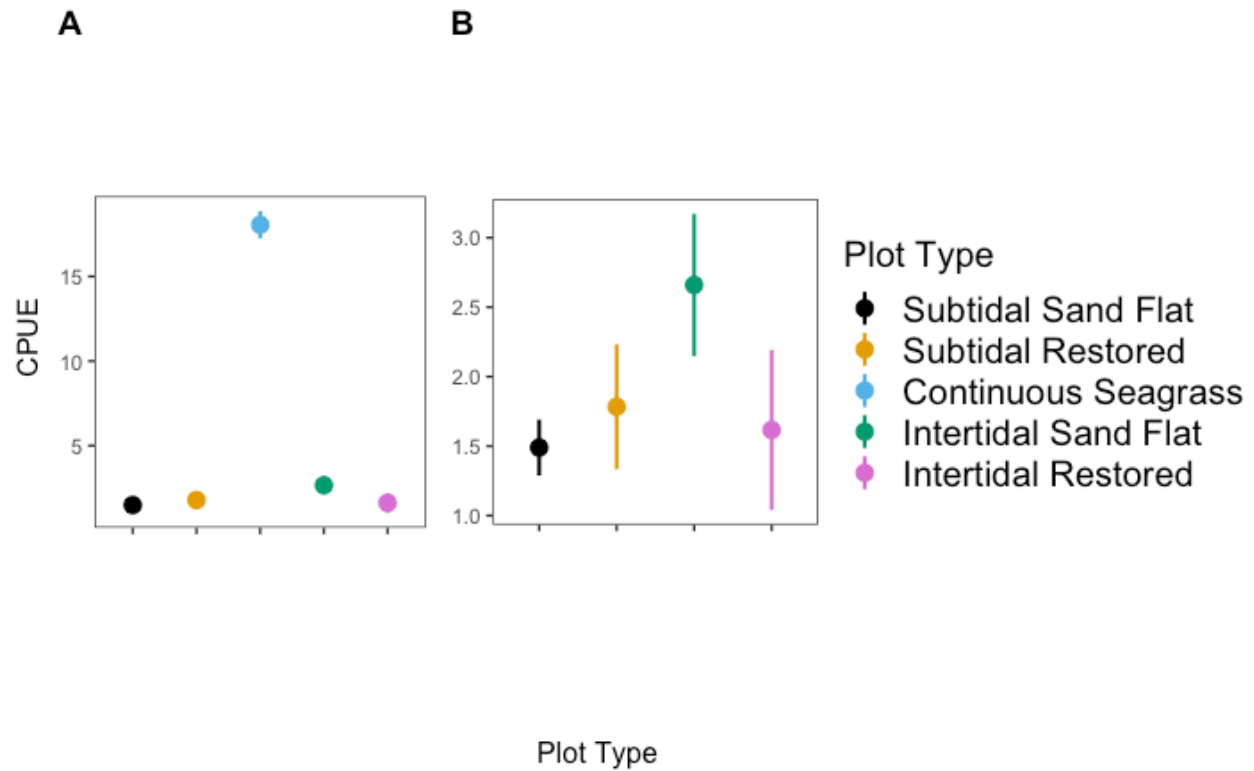


Figure 7: Minnow trap CPUE with and without the continuous seagrass bed plots to allow for better visualization of CPUE for all plots. Error bars denote standard error.

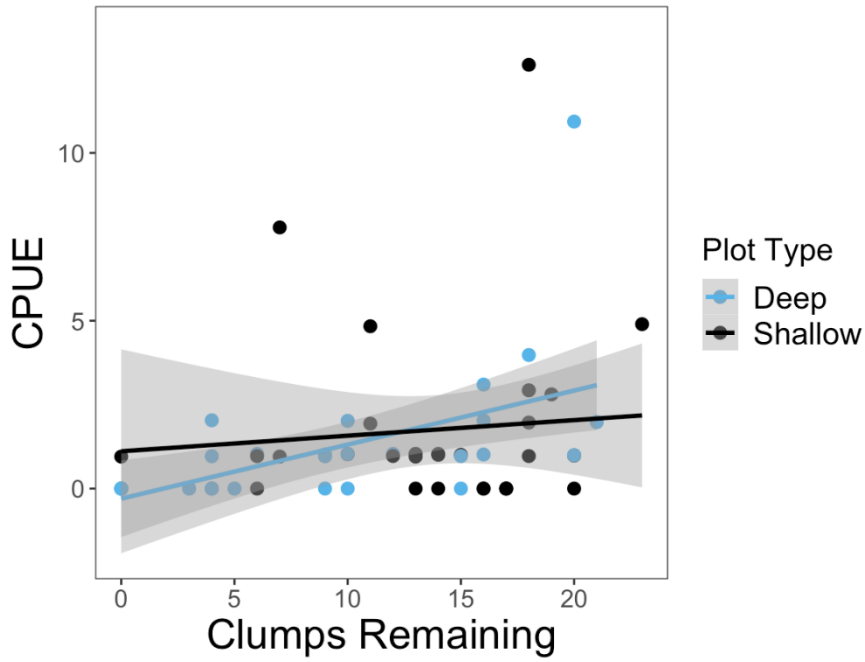


Figure 8: CPUE by number of clumps remaining after seagrass transplanting in deep and shallow plots. Color denotes plot type with our deep in blue and shallow in black. Shading represents the 95% confidence intervals of the linear model

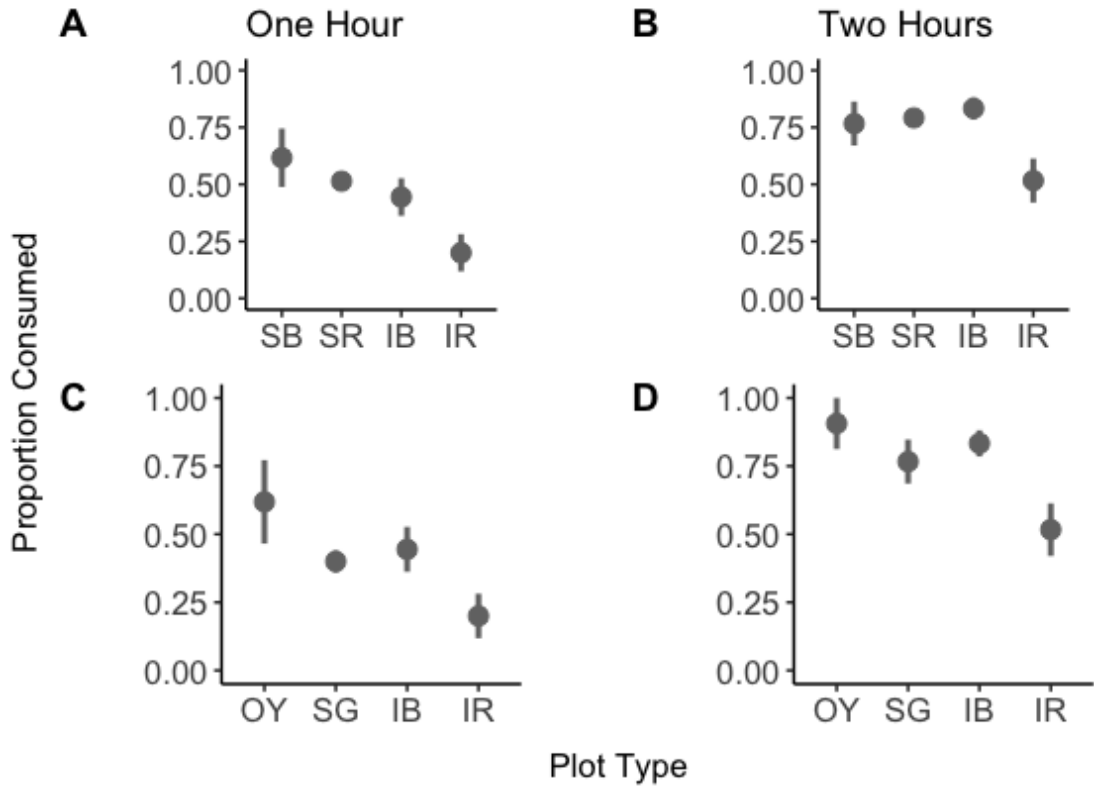


Figure 9: Proportion of consumption assays consumed after one (A,C) and two (B,D) hours for bare sand and restored plots across intertidal and subtidal areas (A,B) and intertidal habitats (C,D). SB denotes subtidal bare sand (n=5), SR denotes subtidal restored (n=6), IB denotes intertidal bare sand (n=6), IR denotes intertidal restored (n=5), OY denotes oyster reef (n=4), and SG denotes continuous seagrass (n=5).

Tables

Table 1: All fauna caught in our minnow traps across all treatment groups. Values here denote mean (standard error) across all sampled plots. Samples were standardized to 24 hours for each sampling event and totaled across dates for each plot

Common Name	Scientific Name	Deep		Shallow		
		Sand	Patchy Seagrass	Sand	Patchy Seagrass	Continuous Seagrass
Fish						
Sheepshead	<i>Archosargus probatocephalus</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.2 (0.2)
Silver Perch	<i>Bairdiella chrysoura</i>	0 (0)	0.32 (0.21)	0.16 (0.16)	0 (0)	0 (0)
Black Sea Bass	<i>Centropristis striata</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.19 (0.19)
Spottail Pinfish	<i>Diplodus holbrookii</i>	0 (0)	0 (0)	0 (0)	0 (0)	2.41 (0.75)
Mummichog	<i>Fundulus heteroclitus</i>	0 (0)	0 (0)	0.16 (0.11)	0.38 (0.38)	1.99 (0.71)
Mojarra	<i>Gerreidae</i>	0.21 (0.21)	0 (0)	0 (0)	0.19 (0.19)	0 (0)
Feather Blenny	<i>Hypsoblennius henz</i>	0.19 (0.19)	0.17 (0.17)	0.17 (0.17)	0.19 (0.19)	1.22 (0.38)
Pinfish	<i>Lagodon rhomboides</i>	4.09 (0.71)	6.05 (2.03)	12.6 (3.43)	7.2 (4.6)	98.34 (8.35)
Mutton Snapper	<i>Lutjanus analis</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.19 (0.19)
Gray Snapper	<i>Lutjanus griseus</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.77 (0.56)
Lane Snapper	<i>Lutjanus synagris</i>	0.2 (0.2)	0 (0)	0 (0)	0 (0)	0.4 (0.24)
Gag Grouper	<i>Mycteroperca microlepis</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.2 (0.2)
Pigfish	<i>Orthopristis chrysoptera</i>	1.17 (0.57)	0.82 (0.53)	0.66 (0.48)	0 (0)	7.75 (1.35)
Gulf Flounder	<i>Paralichthys albigutta</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.2 (0.2)
Planehead Filefish	<i>Stephanolepis hispidus</i>	0.41 (0.41)	0.5 (0.34)	0.16 (0.16)	0.21 (0.21)	3.23 (1.16)
Sea Robin	<i>Triglidae</i>	0 (0)	0.17 (0.17)	0 (0)	0 (0)	0 (0)
Crab						
Blue Crab	<i>Callinectes sapidus</i>	1.16 (0.94)	0.65 (0.21)	0.82 (0.3)	0.97 (0.61)	3.17 (0.58)
Stone Crab	<i>Menippe mercenaria</i>	0 (0)	0.16 (0.16)	0.16 (0.16)	0 (0)	0 (0)
Unknown Swimming Crab	<i>Portunidae</i>	0 (0)	0.17 (0.17)	0 (0)	0 (0)	0.41 (0.41)
Blotched Swimming Crab	<i>Portunus spinimanus</i>	0.8 (0.367)	0.67 (0.34)	0.33 (0.21)	0.6 (0.4)	1.38 (0.86)
Mud Crab	<i>Triglidae</i>	0.2 (0.2)	0.49 (0.33)	0.16 (0.16)	0.4 (0.25)	1.21 (0.74)
Shrimp						
Snapping Shrimp	<i>Alpheidae</i>	0 (0)	0 (0)	0 (0)	0 (0)	0.2 (0.2)
Grass Shrimp	<i>Palaemonetes</i>	1 (0.45)	1.99 (1.23)	3.09 (2.14)	0.78 (0.78)	1.72 (1.72)

Penaeid Shrimp

Penaeidae

1.01 (0.56)

0.33 (0.21)

0.16 (0.16)

0.39 (0.24)

1.18 (0.19)

Table 2: All fauna caught in our SMURF sampling. Values denote average (standard error) across all sampled plots. Samples were standardized to 24 hours for each sampling event and totaled across dates for each plot

Common Name	Scientific Name	Deep	Shallow	Continuous Seagrass
Fish				
Sheepshead	<i>Archosargus probatocephalus</i>	0 (0)	0 (0)	0.2 (0.2)
Unknown Blenny	<i>Blenniiformes</i>	6 (0.71)	0.6 (0.4)	1.2 (0.74)
Skilletfish	<i>Gobiesox strumosus</i>	1.2 (0.49)	1.4 (0.4)	0.4 (0.24)
Slippery Dick	<i>Halichoeres bivittatus</i>	0.2 (0.2)	0 (0)	0 (0)
Crested Blenny	<i>Hypleurochilus geminatus</i>	1.2 (0.49)	2 (0.95)	0.4 (0.24)
Pinfish	<i>Lagodon rhomboides</i>	0.6 (0.4)	0.2 (0.2)	0.8 (0.37)
Gray Snapper	<i>Lutjanus griseus</i>	0.2 (0.2)	0 (0)	0.2 (0.2)
Oyster Toadfish	<i>Opsanus tau</i>	0.2 (0.2)	0.6 (0.24)	0.2 (0.2)
Planehead Filefish	<i>Stephanolepis hispidus</i>	0 (0)	0 (0)	0.4 (0.24)
Unidentifiable Fish	Unidentifiable Fish	0.2 (0.2)	0 (0)	0 (0)
Crab				
Unidentifiable Juvenile Crab	<i>Brachyura</i>	0.2 (0.2)	0.2 (0.2)	0 (0)
Blue Crab	<i>Callinectes sapidus</i>	1.4 (0.4)	1.6 (0.51)	1.8 (0.58)
Say Mud Crab	<i>Dyspanopeus sayi</i>	5.6 (0.93)	3.8 (1.16)	9.2 (1.93)
Common Spider Crab	<i>Libinia emarginata</i>	0.2 (0.2)	0 (0)	0.2 (0.2)
Stone Crab	<i>Menippe mercenaria</i>	0.2 (0.2)	0.4 (0.24)	0 (0)
Atlantic Mud Crab	<i>Panopeus herbstii</i>	2.4 (1.03)	5 (1.22)	0.2 (0.2)
Blotched Swimming Crab	<i>Portunus spinimanus</i>	2.8 (1.02)	3.8 (1.72)	2 (0.32)
Harris Mud Crab	<i>Rhithropanopeus harrisi</i>	1 (0.55)	0.2 (0.2)	3.2 (0.86)
Unidentifiable Mud Crab	<i>Triglididae</i>	0 (0)	0.6 (0.4)	1.4 (1.4)
Shrimp				
Snapping Shrimp	<i>Alpheidae</i>	0.8 (0.37)	1 (0.32)	0 (0)
Grass Shrimp	<i>Palaemonetes</i>	369 (64.91)	467 (41.06)	323.8 (15.62)

Table 3: All lengths/widths of fauna caught in our SMURF sampling. Values denote average length (standard error) across all sampled plots. Fish and shrimp measurements indicate total length, crab measurements indicate carapace width all in millimeters. Note: we do not include the length of the unidentified fish (SI Table 4) in this table as the fish had disintegrated and length was unable to be measured

Common Name	Scientific Name	Deep	Shallow	Continuous Seagrass
Fish				
Sheepshead	<i>Archosargus probatocephalus</i>	0 (0)	0 (0)	38 (0)
Unknown Blenny	<i>Blenniiformes</i>	18.96 (0.68)	16.0 (0)	17.17 (1.17)
Skilletfish	<i>Gobiesox strumosus</i>	15.2 (2.08)	17 (1.1)	12.5 (0.5)
Slippery Dick	<i>Halichoeres bivittatus</i>	70 (0)	0 (0)	0 (0)
Crested Blenny	<i>Hypleurochilus geminatus</i>	27 (2.54)	26.79 (3.25)	20.5 (15.5)
Pinfish	<i>Lagodon rhomboides</i>	41.67 (4.91)	49 (0)	45 (4.49)
Snapper	<i>Lutjanus</i>	0 (0)	0 (0)	0 (0)
Gray Snapper	<i>Lutjanus griseus</i>	55 (0)	0 (0)	40 (0)
Oyster Toadfish	<i>Opsanus tau</i>	14 (0)	39 (19.5)	79 (0)
Planehead Filefish	<i>Stephanolepis hispidus</i>	0 (0)	0 (0)	34 (8)
Crab				
Unidentifiable Juvenile Crab	<i>Brachyura</i>	4 (0)	3 (0)	0 (0)
Blue Crab	<i>Callinectes sapidus</i>	39.43 (3.88)	29.36 (6.54)	33.26 (5.06)
Say Mud Crab	<i>Dyspanopeus sayi</i>	9.92 (0.45)	13.17 (1.48)	10.10 (0.55)
Common Spider Crab	<i>Libinia emarginata</i>	25 (0)	0 (0)	23 (0)
Stone Crab	<i>Menippe mercenaria</i>	8 (0)	8 (1)	0 (0)
Atlantic Mud Crab	<i>Panopeus herbstii</i>	18.29 (1.34)	18.07 (1.49)	21 (0)
Blotched Swimming Crab	<i>Portunus spinimanus</i>	25.11 (2.35)	24.48 (2.46)	33.61 (3.66)
Harris Mud Crab	<i>Rhithropanopeus harrisi</i>	11.1 (1.95)	8 (0)	8.24 (0.37)
Unidentifiable Mud Crab	<i>Triglidae</i>	0 (0)	3.5 (0.5)	2.8 (0)
Shrimp				
Snapping Shrimp	<i>Alpheidae</i>	22 (1.53)	26.75 (1.03)	0 (0)
Grass Shrimp	<i>Palaemonetes</i>	23.36 (0.57)	24.48 (0.62)	24.03 (0.75)

Table 4: Statistical table of ANOVA to test for differences between our shallow and deep elevation categories. Values significant at the 0.05 level are bolded.

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Elevation Category	1	0.219483	0.219483	114.61	<0.0001
Residuals	25	0.047876	0.001915		

Table 5: Statistical table of ANOVA results to test for differences in elevation within our shallow plot categories (continuous seagrass, transplanted clumps, bare sand).

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Habitat Category	2	0.0009123	0.00045615	0.5132	0.6102
Residuals	13	0.0115542	0.00088879		

Table 6: Statistical table of ANOVA results to test for differences within our deep plot categories (seagrass, bare sand).

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Habitat Category	1	0.000376	0.0003761	0.0966	0.7630
Residuals	9	0.035034	0.0038926		

Table 7: Statistical table of linear mixed model results to test for the effect of date, plot type, and their interaction on the number of clumps remaining with plot number as a random effect. Significant values at the 0.05 level are bolded

Scaled residuals:					
Min	1Q	Median	3Q	Max	
-2.02216	-0.539	-0.04706	0.576	2.34488	
Random Effects:					
Groups	Name	Variance	Std. Dev.		
Plot	(intercept)	6	2.45		
Residual		10.12	3.181		
Fixed Effects:					
	Estimate	Std. Error	df	t value	Pr(> t)
(Intercept)	2762	182.8	91.84	15.112	< 0.0001
Plot Type	-911.7	266.9	92.13	-3.416	0.0009
Date	-0.15	0.01	91.83	-15.046	< 0.0001
Plot Type x Date	0.05	0.01	92.14	3.425	0.0009

Table 8: Statistical table of linear mixed model to test for the effect of date on the number of clumps remaining with plot number as a random effect. Significant values at the 0.05 level are bolded.

Scaled residuals:					
Min	1Q	Median	3Q	Max	
-2.8376	-0.3609	0.1494	0.6234	1.3523	
Random Effects:					
Groups	Name	Variance	Std. Dev.		
Plot	(intercept)	17.078	4.133		
Residual		6.995	2.645		
Fixed Effects:					
	Estimate	Std. Error	df	t value	Pr(> t)
(Intercept)	923.99515	196.9778	39.00703	4.691	< 0.0001
Date	-0.04784	0.01025	38.99985	-4.666	< 0.0001

Table 9: Statistical table of ANOVA results for differences between minnow trap catch across plot types. Significant values at the 0.05 level are bolded .

	Df	Sum Sq	Mean Sq	F value	Pr(>F)
Plot Type	4	1065.6	266.401	170.34	<0.0001
Residuals	22	34.41	1.564		

Table 10: Statistical table of Tukey post-hoc test to test for differences in minnow trap catch between plot types. Values significant at the 0.05 level are bolded.

Comparison	Estimate	Std. Error	t value	Pr(> t)
Deep Transplanted Clumps – Deep Bare	0.2937	0.7572	0.388	0.995
Continuous Seagrass – Deep Bare	16.5643	0.7909	20.943	<0.0001
Shallow Bare – Deep Bare	1.1698	0.7572	1.545	0.546
Shallow Transplanted Clumps – Deep Bare	0.1266	0.7909	0.16	1
Continuous Seagrass – Deep Transplanted Clumps	16.2706	0.7572	21.486	<0.0001
Shallow Bare – Deep Transplanted Clumps	0.876	0.722	1.213	0.744
Shallow Transplanted Clumps – Deep Transplanted Clumps	-0.1671	0.7572	-0.221	0.999
Shallow Bare – Continuous Seagrass	-15.3946	0.7572	-20.33	<0.0001
Shallow Transplanted Clumps – Continuous Seagrass	-16.4377	0.7909	-20.783	<0.0001
Shallow Transplanted Clumps – Shallow Bare	-1.0432	0.7572	-1.378	0.647

Table 11: Statistical table from generalized linear model for CPUE by clumps remaining in the transplanted plots. Values significant at the 0.1 level are italicized, significant values at the 0.05 level are bolded.

	Df	Deviance	Residual Df	Residual Deviance	F	Pr(>F)
NULL			60	158.92		
Clumps Remaining	1	27.2367	59	131.68	9.68	0.002909
Elevation Category	1	0.1716	58	131.51	0.061	0.805817
Interaction	1	8.8819	57	122.63	3.1567	<i>0.080954</i>

Table 12: Statistical table from generalized linear model for squid pops at the one hour check comparing shallow and deep restored and bare sand plots. Values significant the 0.05 level are bolded.

	Df	Deviance	Residual Df	Residual Deviance	F	Pr(>F)
NULL			21	5.7224		
Restoration						
Category	1	0.51288	20	5.2095	2.8538	0.1084
Elevation Category	1	1.35747	19	3.852	7.5534	0.01322
Interaction	1	0.16275	18	3.6892	0.9056	0.3589

Table 13: Statistical table from generalized linear model for squid pops at the one hour check comparing all shallow habitats (bare sand, patchy seagrass, continuous seagrass, oyster). Values significant at the 0.1 level are italicized.

	Df	Deviance	Residual Df	Residual Deviance	F	Pr(>F)
NULL			19	5.182		
Habitat Category	3	1.7361	16	3.4459	3.0952	<i>0.05662</i>

Table 14: Statistical table from generalized linear model for squid pops at the two hour check comparing shallow and deep restored and bare sand plots. Values significant at the 0.1 level are italicized, significant values at the 0.05 level are bolded.

	Df	Deviance	Residual Df	Residual Deviance	F	Pr(>F)
NULL			21	4.4283		
Restoration						
Category	1	0.52946	20	3.8989	3.786	<i>0.06747</i>
Elevation						
Category	1	0.3131	19	3.5858	2.2389	0.1519
Interaction	1	0.69975	18	2.886	5.0037	0.03819

Table 15: Statistical table from Tukey post-hoc test to test for differences between treatments at the two hour check comparing shallow and deep restored and bare sand plots. Values significant at the 0.1 level are italicized, significant values at the 0.05 level are bolded.

Comparison	Estimate	Std. Error	z value	Pr(> z)
Shallow Restored – Deep Restored	-1.2683	0.5033	-2.52	<i>0.0564</i>
Deep Sand – Deep Restored	-0.1454	0.5456	-0.267	0.9934
Shallow Sand – Deep Restored	0.2744	0.556	0.494	0.9604
Deep Sand – Shallow Restored	1.1229	0.518	2.168	0.132
Shallow Sand – Shallow Restored	1.5427	0.529	2.916	0.0183
Shallow Sand – Deep Sand	0.4199	0.5694	0.737	0.8817

Table 16: Statistical table from generalized linear model for squid pops at the two hour check comparing all shallow habitats (bare sand, patchy seagrass, continuous seagrass, oyster). Values significant at the 0.05 level are bolded.

	Df	Deviance	Residual Df	Residual Deviance	F	Pr(>F)
NULL			19	5.639		
Habitat Category	3	2.147	16	3.4921	3.56	0.03809

Table 17: Statistical table from Tukey post-hoc test to test for differences between treatments at the two hour check comparing all shallow habitats. Values significant at the 0.1 level are italicized.

Comparison	Estimate	Std. Error	z value	Pr(> z)
Patchy Seagrass – Oyster	-2.202	0.8675	-2.538	<i>0.0525</i>
Sand – Oyster	-0.6592	0.9126	-0.722	0.8858
Continuous Seagrass – Oyster	-1.0791	0.9035	-1.194	0.6245
Sand – Patchy Seagrass	1.5427	0.6342	2.433	<i>0.0689</i>
Continuous Seagrass – Patchy Seagrass	1.1229	0.6211	1.808	0.2639
Continuous Seagrass – Sand	-0.4199	0.6826	-0.615	0.9257