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Evaluating the Capacity of Ascophyllum nodosum Habitats to Act as Carbon Sinks in the Gulf of Maine

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EVALUATING THE CAPACITY OF ASCOPHYLLUM NODOSUM HABITATS TO ACT AS CARBON SINKS IN THE GULF OF MAINE

by

Megan K. Sinclair

A Thesis Submitted in Partial Fulfillment of the Requirements for a Degree with Honors (Wildlife Ecology)

The Honors College
University of Maine
May 2021

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Some coastal ecosystems are defined as being carbon sinks for their ability to absorb more carbon than they release as a result of their high primary productivity. There has been support for the claim that macroalgal communities can act as carbon sinks and reduce levels of CO$_2$ in seawater through photosynthesis and potentially mitigate some local effects of climate change (Chung et al., 2011; Chung et al., 2013; Hill et al., 2015; Sondak et al., 2017). Within the state of Maine, rocky intertidal zones are coastal ecosystems that are particularly vulnerable to climate change and dominated by *Ascophyllum nodosum* (rockweed) communities (Fong 2008; Letcher 2015). The objective of this study was to evaluate the ability of rockweed to act as a carbon sink in the Gulf of Maine and mitigate local effects of climate change. The hypothesis for this experiment was photosynthesis, not respiration, would be the dominant process observed in treatments with rockweed present. Productivity was estimated through calculating the departure from oxygen saturation of each treatment. Three habitat complexity treatments were observed: a control with only salt water, rockweed submerged in salt water, and rockweed and invertebrates submerged in salt water. It was predicted the rockweed treatment would have the highest productivity, seconded by the invertebrate and rockweed treatment, and the control treatment would experience neither productivity nor respiration. Results showed little to no oxygen was produced during either trial in any treatment, suggesting that respiration was the dominant process in the experiment. This experiment does not support the claim that rockweed has the ability to act as a carbon sink. Other literature contradicts these findings which suggests this experiment would benefit from replication or further expansion.
DEDICATION

I dedicate this work to my mother, Stacey. I could have never come this far without your unconditional love and kindness. Thank you for sharing your sense of wonder and appreciation for all living things with me, especially pelicans and okapis.
ACKNOWLEDGMENTS

I would like to thank my advisors, Amanda Klemmer and Hannah Mittelstaedt, for their continued support and guidance in this research effort. I am so thankful my volunteer experience with Hannah would lead to the creation of a thesis two years later. I am thankful for Jasmine Saros for her suggestions and help in the data analysis process, and Dudu Meireles and Jordan LaBouff for their support and assistance in this thesis project. I would also like to thank Hamish Greig and Isaac Shepard for their help in allowing me to borrow equipment and taking the time to show me how to use them. I am thankful for Tamara Levitsky and her help in securing vital supplies for this experiment. I would like to thank everyone who helped in lending equipment or assisted with finding working probes for this thesis. I am so thankful for my wonderful friends in sharing our unique thesis experiences and their help in collecting rockweed on a Saturday morning before sunrise. I would like to thank my family for their constant love and support. I am very fortunate to have the opportunity to conduct and defend a thesis through the University of Maine and the Honors College.
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Table 1. One way ANOVA on habitat complexity treatments predicting response variables: dissolved oxygen departure from saturation for a) trial 1 and b) trial 2; temporal variation of dissolved oxygen departure from saturation for c) trial 1 and d) trial 2.
Coastal ecosystems are one of the most ecologically and economically important habitats in the world as they provide many services for wildlife and humans. Aquatic vegetation in coastal environments provide nutrient cycling, carbon storage, and nursery habitats for valuable species (Schmidt et al., 2011). Coastal ecosystems are sources for goods, such as food for humans, salt, minerals, and construction materials, and provide crucial services to humans that include shore protection from extreme weather events, erosion control, and waste treatment and purification (Martínez et al., 2007). The beauty and accessibility of coastal ecosystems make them valuable places for residency, recreation, and tourism. These qualities of coastal ecosystems make them one of the most populated places in the world (Martínez et al., 2007).

Coastal ecosystems are threatened by anthropogenic effects like harvesting, pollution, and coastal development. These threats have increased over recent years and cause injury to organisms and environment degradation or loss. The continual rise of anthropogenic atmospheric carbon dioxide, as a result of the burning of fossil fuels, industrialization, deforestation, and other land-use changes (Guinotte and Fabry 2008), will create new threats and worsen established impacts on coastal ecosystems. These threats include rising sea levels, rising ocean temperatures, and ocean acidification (Kaplanis et al., 2019; Guinotte and Fabry 2008). Ocean acidification is the change in ocean chemistry caused by anthropogenic atmospheric CO$_2$ (Guinotte and Fabry 2008). The oceanic absorption of carbon dioxide makes oceans more acidic due to the presence of excess of hydrogen ions. Ocean acidification is harmful to many marine organisms, especially calcifying organisms as they rely on carbonate ions to form their skeletons and
shells (Jones et al., 2018). In acidic conditions, carbonate ions bind with excess hydrogen ions resulting in a reduction of available carbonate for calcifying organisms to build and maintain their structures. These structures can even dissolve if the pH is too low.

However, not all marine organisms are negatively affected by ocean acidification.

Algae and seagrasses are primary producers and perform photosynthesis, where CO₂ is absorbed and fixed for growth, like in terrestrial plants (Hill et al., 2015) and could potentially benefit from higher CO₂ conditions in oceans, primarily through increased photosynthetic rates (Duarte et al., 2017; Guinotte and Fabry 2008; Koch et al., 2012). In addition to benefitting from the effects of ocean acidification, studies have shown that algae and other primary producers can mitigate the effects of climate change (Hill et al., 2015; Chung et al., 2011; Sondak et al., 2017; Duarte et al., 2017).

Certain vegetated coastal ecosystems like mangrove forests, seagrass beds, and salt marshes, are known as carbon sinks for their ability to store carbon (Hill et al., 2015; Sondak et al., 2017). These ecosystems sequester carbon when CO₂ is absorbed from the atmosphere through photosynthesis while oxygen is released. A fraction of the carbon absorbed is released during oxidation and respiration, but some assimilated carbon remains in the form of living biomass and contributes to organic carbon stored in sediment (Sondak et al., 2017). Macroalgal communities have recently been classified as short-term carbon sinks due to their ability to reduce the levels of dissolved inorganic carbon (DIC) in oceans (Chung et al., 2011; Chung et al., 2013; Hill et al., 2015; Sondak et al., 2017). Macroalgae communities utilize (DIC) as CO₂ in seawater that diffuses through cellular membranes and as bicarbonate which is actively pumped into the cell using a carbon concentrating mechanism (CCM) (Sondak et al., 2017). The uptake of
DIC and its transformation into organic carbon can decrease the amount of CO₂ in seawater (Sondak et al., 2017). Macroalgal communities differ from ecosystems known to sequester carbon because macroalgal communities lack sediment where carbon can be stored in the form of organic carbon (Chung et al., 2013; Hill et al., 2015). Yet, studies have shown that seaweed aquaculture beds can mitigate the effects of climate change in oceans (Sondak et al., 2017; Chung et al., 2011). For example, commercially harvested seaweeds remove 0.7 million tons of carbon from oceans every year (Sondak et al., 2017). Also, evidence suggests seaweed aquaculture beds have the ability to raise the pH in their immediate vicinity and buffer their local habitat from ocean acidification (Xiao et al., 2021). Macroalgal communities function as carbon sinks because they are very productive autotrophs and generally produce more organic matter than is consumed through respiration (Duarte et al., 2017).

One type of vegetated coastal ecosystem that is dominated by productive macroalgal communities are rocky intertidal zones (Fong 2008). Intertidal zones act as the interface between land and sea that experiences extreme environmental gradients as a result of wave and tidal actions. These habitats are defined by having large, daily fluctuations in temperature and underwater submersion (Pfaff et al., 2018). Waves and tides create horizontal bands of distinct algal and invertebrate communities along the coast that are defined by the amount of time per day the area is submerged or susceptible to heavy wave activity. Intertidal zones range from the high shore, which is exposed to air for most of the day except during high tide, the mid-shore, that is dominated by barnacles and some macroalgae, and the low shore, which is only exposed to air during low tide (Pfaff et al., 2018). The hardy organisms that reside in intertidal zones mitigate
the harsh conditions of their habitat through behavioral and physiological adaptations. Many invertebrate species in intertidal zones are sessile and are permanently attached to rocky substrates or other organisms. Common sessile invertebrates include those belonging to the phylums Cnideria (e.g., hydroids and sea anemones), Arthropoda (e.g., barnacles), Mollusca (e.g., mussels and clams) and Annelida (e.g., sessile polychaetes) (Petraitis et al., 2008). Most of these organisms use suspension feeding to ingest plankton found in the water column and contribute to secondary production of this habitat by transferring ocean productivity to the shore (Petraitis et al., 2008). Mobile invertebrates include species from Crustacea (e.g., crabs, shrimp, amphipods, and isopods), Annelida (e.g., errant polychaetes), Gastropoda (e.g., snails), and Echinodermata (e.g., sea urchins and sea stars) (Petraitis et al., 2008). Vertebrates that utilize the intertidal zone include fish, birds, and mammals that rely on these habitats for food and protection from predators. Unfortunately, rocky intertidal zones are particularly sensitive to the effects of climate change given these ecosystems connect terrestrial and marine environments and display some of the fastest responses to climate change of any other habitat (Letcher 2015). Organisms occupying these already hostile habitats will face additional challenges such as sea and air temperature changes, extreme desiccation, fluctuations in ocean chemistry, and other threats to their survival (Letcher 2015).

The coast of Maine is mostly composed of rocky intertidal zones. Those habitats along with other marine ecosystems in the state have experienced negative impacts of climate change. The average temperatures of the Gulf of Maine have steadily increased, the pH of the Gulf has been reduced, and Maine is susceptible to more frequent flooding due to rising sea levels (Fernandez et al., 2020). Maine’s economy is heavily reliant on
marine fisheries with lobster (*Homarus americanus*) being the most valuable export in the industry (Fernandez et al., 2020). Studies suggest that climate change events, such as ocean acidification and warmer temperatures, will severely threaten lobster growth, reproduction, and survival (Klymasz-Swartz et al., 2019; Keppel et al., 2012).

Maine rocky intertidal zones are dominated by the macroalgae, rockweed. Rockweed belongs to the brown algae group Phaeophyta and is a multicellular, brown alga found in rocky intertidal ecosystems on either side of north Atlantic (Phillippi et al., 2014). Rockweed plays a vital role in the rocky intertidal community as it provides habitat, food, and protection from desiccation and predation for fish, invertebrates, and waterfowl (Phillippi et al., 2014). In addition to its ecological importance, rockweed is also economically important in the state of Maine. Rockweed has several commercial uses such as fertilizer, animal feed, and alginate extracts (Phillippi et al., 2014). About 5.4-6.8 tons of rockweed is harvested in Maine each year, however there is little evidence this removal of algae negatively impacts the intertidal community (Phillippi et al., 2014). Rockweed has a relatively high growth rate (~20 cm per year) which implies the algae can recover rapidly after harvest (Phillippi et al., 2014).

Rockweed’s resilience to harvesting is a product of its adaptive morphologies for survival in its unforgiving, wave swept environment. Rockweed utilizes a holdfast to anchor the alga to a rocky surface and a stipe to support and protect the alga to allow it to withstand the forces of breaking waves (Petraitis et al., 2008). Rockweed and other large brown algae use pneumatocysts, gas-filled floats, to buoy blades, structures that perform gas exchange and capture light for photosynthesis, close to the surface of water where light intensity is greatest (Petraitis et al., 2008). Rockweed performs the second highest
rate of primary productivity in Phaeophyta and is capable of productivities per unit area substrate over 1,000 gC m$^{-2}$year$^{-1}$ (Chung et al., 2010). The photosynthetic gas-exchange responses of rockweed involve a CCM (Koch et al., 2012). CCM is an adaption to help maximize an organism’s ability to photosynthesize in aquatic environments characterized by unfavorable conditions for photosynthesis like low CO$_2$ levels and is common in algae (Singh et al., 2014). Evidence suggests rockweed would benefit from increased CO$_2$ environments due to its photosynthetic adaptation to maximize productivity in low CO$_2$ level environments and high productivity rates. These qualities, in addition to a fast growth rate, make rockweed an ideal candidate in understanding if wild, unmanaged macroalgal communities have any effect in alleviating local climatic change stressors and have the ability to function as carbon sinks in the Gulf of Maine.

My study aims to evaluate the capacity of rockweed to act as a mitigator for local ocean acidification effects in the Gulf of Maine by using departure from oxygen saturation values to determine if the macroalgae could function as a carbon sink. Studies differentiated carbon sinks from sources by using a ratio of production : respiration (Hill et al., 2015). A production : respiration ratio of 1 suggested the system removed and stored more carbon than was released and was a carbon sink (Hill et al., 2015). Departure from oxygen saturation reveals the consumption or production of oxygen in a system and shows if photosynthesis or respiration processes dominated in the system. Systems in which photosynthesis dominant overall processes would be oversaturated with oxygen and systems in which respiration dominated overall processes would be undersaturated with oxygen. It is inferred that a system that is predominantly oversaturated would be a carbon sink.
The objectives of the study were to evaluate the ability of rockweed to act as a carbon sink by analyzing departure from oxygen saturation values as a proxy for productivity in tanks with only rockweed, and rockweed and *Littorina littorea* (common periwinkle, abundant in rocky intertidal areas dominated rockweed), and only saltwater. Oxygen saturation was measured in tanks with rockweed and common periwinkles to show the oxygen relationship between an autotroph and a heterotroph and mimic the algae’s interactions within the intertidal community. Other tanks only measured the oxygen saturation of rockweed so its ability to absorb carbon as an isolated system was fully recorded. It was hypothesized the tanks of only rockweed would have the highest rates of productivity due to the algae’s efficient ability to absorb carbon and produce oxygen. The control tanks were predicted to have the lowest rates of productivity due to the presence or absence of an autotroph, and lack of oxygen expected to be introduced into the system. The tanks of rockweed and invertebrates were expected to have lesser rates of productivity compared to tanks of only rockweed because the periwinkles should consume some of the oxygen the algae produces.
METHODS

Field Methodology

Rockweed and periwinkles were collected from the Mitchell Marsh Preserve located on Mount Desert Island in Tremont, Maine (Figure 1). The 35-acre preserve features an extensive coastal marshland and rocky intertidal zone. Permission was granted by the Maine Coast Heritage Trust for the removal of rockweed individuals for this experiment. The low tide zone was visited twice in November 2020. Collection occurred in the same general area in the low intertidal zone during low tide hours. Collected rockweed individuals were cut above the holdfast. Rockweed individuals were no longer than one meter from the point where branching started from the holdfast and were recently exposed during the receding tide. Nine rockweed individuals were collected for the first trial and twelve rockweed individuals were collected for the second trial. Collected individuals were stored in plastic bags with ocean water and kept in a cooler. Thirty common periwinkles were collected in the same area for each experiment. Preference was given to larger snails that with shells roughly 2.5 cm wide. Snails were removed from rocky substrates and were placed in a plastic container with ocean water. Rockweed individuals and snails were kept in a cooler with ice packs for approximately two hours during the drive from the study site to the experiment room. Salinity of ocean water was recorded with a refractometer during each site visit.

Lab Experimental Design

The experiment was conducted in a room with artificial lighting in Libby Hall at the University of Maine, Orono. Two trials were performed for this thesis experiment.
The first trial occurred on November 7, 2020 and the second trial on November 21, 2020. Eleven 38-liter fish tanks were prepared for the first trial (the twelfth tank broke during the trial) and fourteen tanks were prepared for the second trial. All tanks for both trials were completely filled with Instant Ocean solution. Instant Ocean Solution was made by combining 38 liters of tap water and 1,360 g of Instant Ocean salt for each tank. The salinity of all tanks was measured using a refractometer to ensure salinity was within the range of the ocean water of the field site. The salinity of the ocean water was recorded at 32.5 ppt at each field collection event (November 7, 2020; November 21, 2020) and the tanks’ salinity ranged from 32.5 to 35 ppt and deemed suitable for the experiment. There were three habitat complexity treatments in this experiment: control, rockweed, and invertebrates (periwinkles) and rockweed. Control treatments consisted of tanks with Instant Ocean solution and no rockweed (Figure 2). The rockweed treatment had the Instant Ocean solution and one rockweed individual per tank that was thoroughly washed with tap water, to remove microscopic invertebrates, before being placed in the tank (Figure 3). Invertebrates and rockweed treatments included tanks with Instant Ocean solution, one rockweed individual, and six common periwinkles per tank (Figure 4). Periwinkles were contained by weighted plastic mesh baskets to prevent escapement. Rockweed individuals in all treatment tanks were weighted down and tethered so they were completely submerged for the duration of the trials. The first trial consisted of two control tanks, four rockweed tanks (one was lost mid-experiment), and five invertebrate and rockweed tanks. The second trial had two control tanks, six rockweed tanks, and six invertebrate and rockweed tanks.
Lab Experiment Procedure

All measurements for each tank were recorded roughly every two hours for about twenty-four hours for each trial. Dissolved oxygen percentage (DO %) and dissolved oxygen in milligram per liter (DO mg/L) were recorded using a YSI 55 DO probe. During the first trial, the YSI 55 DO probe was left still within the tanks and this was corrected in the second trial where the probe was constantly moved back-and-forth at the surface in each tank until it generated a steady reading. Conductivity (µS/cm), temperature (°C), and salinity (ppt) were recorded using a DO200 probe. The DO200 probe was submerged and untouched until it generated a consistent reading. The pH was recorded using litmus paper. Lights within the experimental room were synchronous with sunrise and sunset times and tanks were exposed to only a headlamp at night for short periods of time while measurements were being taken. Light values (lumens) were automatically recorded every hour by HOBO loggers submerged in each tank for both trials.

Statistical Methodology

Dissolved oxygen departure from saturation (O2 Dep-Sat) was used as a proxy for the relative influence of photosynthesis versus respiration in each tank in both trials. Theoretical 100% oxygen saturation (mg/L) for each tank was calculated using mean tank temperature, mean tank specific conductance, and barometric pressure of 760 mm Hg. These values were inputted into the USGS ‘Dissolved oxygen solubility table’ online tool (USGS 2019). Dissolved oxygen departure from saturation (O2 Dep-Sat) was then
calculated for every DO reading across the 24-hour trials for each tank using the equation:

\[ O_2 \text{ Dep} - \text{Sat} = (\text{obsDO} - \text{sat100}) \times 32 \]

Where \( O_2 \text{ Dep-Sat} \) is the dissolved oxygen departure from saturation, \( \text{sat100} \) is the theoretical 100% oxygen saturation (mg/L) at each measurement recording within a trial, \( \text{obsDO} \) is the observed dissolved oxygen (mg/L) at each measurement recording within a trial, and 32 is the molecular weight of \( O_2 \).

Two summary statistics were calculated for each tank: tank \( O_2 \text{ Dep-Sat} \) and temporal variation. Tank \( O_2 \text{ Dep-Sat} \) was calculated by taking the mean of all measurements through time of dissolved oxygen departure from saturation of each tank in their respective treatments. Temporal variation was calculated by taking the standard deviation of all measurements through time of dissolved oxygen departure from saturation of each tank and averaged to produce the standard deviation of all treatments in both trials. One-way analysis of variance (ANOVA) was performed separately for the first and second trial with habitat complexity treatments as the predictor variable and tank \( O_2 \text{ Dep-Sat} \) or temporal variation as the response variable. All analyses of variance were conducted using R version 3.6.2 (R Core Team 2019).
RESULTS

Salinity (ppt) fluctuated less than 0.5 ppt over the course of 24 hours and ranged from 32.5 ppt to 35 ppt (Figure 5). Specific conductivity ranged between 44,000 µS/cm to 47,000 µS/cm (Figure 6). Specific conductivity values fluctuated in each treatment in the first trial but remained constant in the second trial. The pH of each treatment in both trials was recorded at 8 throughout the experiment. Temperature ranged from 18°C to 20.2°C over the course of both trials (Figure 8). Temperature readings during the second trial were roughly 1°C lower than readings from the first trial but were more consistent between treatments. Dissolved oxygen (mg/L) ranged between 5.8 mg/L to 1.6 mg/L over 24 hours in the two trials (Figure 7). Dissolved oxygen declined over time in all treatments from both trials except for the control treatment in the first trial where dissolved oxygen increased in both tanks. Treatments from the second trial showed a stronger trend in declining dissolved oxygen values than in the first trial.

Control treatments in both trials had the smallest tank O$_2$ Dep-Sat (Figure 9). Overall, the second trial had lower O$_2$ Dep-Sat than the first trial (Figure 9). In the first trial, the invertebrate and rockweed treatment had a higher tank O$_2$ Dep-Sat than the rockweed treatment (Figure 9). Interestingly, in the second trial, the invertebrate and rockweed, and rockweed treatments had nearly the same tank O$_2$ Dep-Sat (Figure 9). The tank O$_2$ Dep-Sat differed significantly across treatments in the first trial ($p<0.001$, Table 1) and in the second trial ($p=0.003$, Table 1).

The invertebrate and rockweed treatment in the first trial had the lowest temporal variation, showing the least change in O$_2$ Dep-Sat through time over the course of the trial (Figure 10). The rockweed treatment in the first trial had the next lowest temporal
variation (Figure 10). The invertebrate and rockweed treatment in the second trial had the highest temporal variation (Figure 10). The temporal variation of the invertebrate and rockweed, and rockweed treatments in both trials were very different, yet the control treatment temporal variation was nearly the same in each trial (Figure 10). The temporal variation was different across treatments in the first trial ($p=0.021$, Table 1). The second trial had a significantly different temporal variation across treatments ($p=0.003$, Table 1).
DISCUSSION

Coastal ecosystems are experiencing worsened effects from climate change, such as ocean acidification. Rocky intertidal zones are a particularly sensitive coastal ecosystem that are dominated by the macroalgae rockweed in Maine. Some ecosystems have the ability to act as carbon sinks and feature vegetation that absorbs DIC in seawater resulting in lower CO$_2$ concentrations in their environment. The purpose of this experiment was to evaluate the capacity of rockweed habitats to act as carbon sinks and mitigate local effects of climate change in the Gulf of Maine. Departure from oxygen saturation values were calculated to estimate the rate of productivity occurring in each treatment. It was predicted that treatments with rockweed present would exhibit high productivities due to overwhelming evidence for the high productivity of macroalgal communities and rockweed habitats. Results showed respiration was the dominant process in all treatments in the experiment indicating that rockweed would not have the ability to act as a carbon sink.

**Rockweed Productivity**

In the first trial, the invertebrate and rockweed treatment had a higher O$_2$ Dep-Sat than the rockweed treatment (Figure 9) and both treatments had nearly the same O$_2$ Dep-Sat in the second trial (Figure 9). Further, the temporal variation values of the second trial show a greater change of dissolved oxygen over time than the first trial (Figure 10). Temperature, salinity, pH, and specific conductivity values remained constant across all treatments and both trials (Figures 5-8), so it is likely they did not affect O$_2$ Dep-Sat or temporal variation in this experiment. The first trial supports the prediction that the
invertebrate and rockweed treatment would have a higher O$_2$ Dep-Sat than the rockweed treatment. However, this trial likely suffered from human error and did not produce accurate results, which will be explained in greater depth later in this discussion. So, it is unusual that the rockweed, and invertebrate and rockweed treatment in the second trial produced nearly identical tank O$_2$ Dep-Sat values. One possible reason for the similar tank O$_2$ Dep-Sat of these treatments is that the periwinkles and microscopic invertebrates in the invertebrate and rockweed treatments did not require enough oxygen to have created a detectable deficit in the system compared to the rockweed treatment. This implies that the tank O$_2$ Dep-Sat of the second trial was primarily impacted by rockweed productivity as with the rockweed treatment. Since O$_2$ Dep-Sat was used as a proxy for productivity, the results suggest that there was little to no photosynthetic activity in both trials and respiration was the driving process in the experiment. My unexpected findings of low rockweed productivity contradict other studies that recorded high productivity in rockweed and other macroalgae. The high primary productivity of macroalgae communities is well documented (Sondak et al., 2017; Duarte and Cebrián 1996; Chung et al., 2013). Chung et al. (2011) found rockweed to be one of the most productive algae in Phaeophyte and can exceed a productivity of 1,000 gC m$^{-2}$year$^{-1}$ while Vadas et al. (2004) found rockweed has a maximum productivity of 894 gC m$^{-2}$year$^{-1}$.

Rockweed is characterized, like most organisms occupying rocky intertidal zones, as being robust and able to withstand a wide range of environmental conditions (Stengel and Dring 1997). One study found transplanted rockweed individuals that were moved from the high intertidal zone to the low intertidal zone and vice versa photosynthetically acclimated to their new location and were as productive as resident algae in the same
respective zone (Stengel and Dring 1997). The same study removed rockweed by tearing part of it from its original attachment site and saw no evidence of damage to the rockweed as a result of removal (Stengel and Dring 1997). It is likely that the physical cutting of rockweed from its holdfast for transport in this experiment did not injure or damage the algae. Thus, it is very unexpected that the rockweed was unable to photosynthesize for the duration of this experiment. There are several possible reasons that explain the lack of productivity recorded in this study. The most obvious explanation for my unexpected findings implies the rockweed individuals used in the experiment were unable to photosynthesize due to stress. It is important to note that the rockweed individuals in the experiment were weighted down and tethered, so they were completely submerged in the Instant Ocean solution. This was done so any gas exchange would be between the algae and water, allowing for the most accurate measurements and best estimation of whether photosynthesis or respiration occurred more often over time. Additionally, some evidence claims that most intertidal algae grow best in constant submersion with ample light (Schonbeck and Norton 1980). Rockweed is typically found in the low to mid intertidal zone where it is known to grow the fastest among intertidal zones (Stengel and Dring 1997). However, even rockweed occupying the lowest intertidal zones, that receive the smallest amount of sunlight, are not submerged in seawater for 24 hours at a time like the rockweed in this experiment. Evidence suggests emersion is important for the photosynthetic activity of algae. Dring and Brown (1982) found brown algae can experience up to 25% of increased photosynthetic activity when the algae is recently emersed before the algae is exposed to dry conditions for too long. It is possible that the rockweed in this experiment was unable to perform photosynthesis
due to a lack of emersion. Although very little or no photosynthetic activity occurred in this experiment, this data could provide knowledge in understanding environmental conditions and limits that prevent the establishment of rockweed communities in the lowest levels of the intertidal zone where emersion is not facilitated.

It was hypothesized the control treatments would have an $O_2$ Dep-Sat value of zero since neither respiration nor photosynthesis should occur. Unexpectedly, the control treatments experienced the least respiration and highest productivity between the other treatments in both trials (Figure 9). Additionally, the temporal variance of the control treatments was very similar between both trials indicating they experienced the same rate of oxygen loss over time (Figure 10). One possible explanation for the low respiration of the control treatments could be the lack of organic matter decaying in the control treatments compared to others. Bacteria and other microorganisms decompose organic matter in water and consume oxygen during the process (Mesner and Geiger 2010). Another theory is the bacteria that likely existed on the rockweed individuals from the collection site could have grown in their population and consumed additional oxygen in the water as they respirated and decomposed the organic algal matter in their tanks (Waksman and Carey 1934). The decomposition of algae could have occurred in the rockweed, and invertebrate and rockweed treatments but the control treatments did not include algae, so there would have been no organic matter to support the activities of bacteria. However, it seems unlikely for the algae to decompose in less than four hours since their removal and transport from the study site.

There are several technical reasons due to experimental design and execution that could explain some of my unexpected results described above. The difference in $O_2$ Dep-
Sat values and temporal variation between rockweed, and invertebrate and rockweed treatments in the first and second trial is most likely due to human error in recording measurements. In the first trial, I left the YSI 55 DO probe untouched in each tank until a steady reading was generated after about ten minutes. The probe must consume oxygen to measure dissolved oxygen and if left still it consumes the oxygen in its immediate vicinity and produces a lower, inaccurate reading for the system. This measuring technique was corrected in the second trial where the probe was moved at a speed of ~0.3 m/s until a steady reading was generated. The second trial was believed to have yielded more accurate results based on the uniform dissolved oxygen trends measured between treatments. A technical reason explaining the strangely low O\textsubscript{2} Dep-Sat values measured in the control treatments could be a result of human error in failing to clean the experimental fish tanks thoroughly. Tanks were washed with tapwater and wiped down but perhaps there could have been residue bacteria or other microorganisms consuming oxygen that were left in the tanks from previous use. Another cause could have been letting the water in the tanks sit for prolonged periods of time before the experiments began. The Instant Ocean salt took time to dissolve, even when vigorously stirred, and was often left for several hours before the experiment began. Lastly, the artificial lights in the room where the experiment was conducted could have failed to facilitate rockweed photosynthesis. Rockweed can tolerate low-light conditions in its environment but perhaps the lights in the experimental room did not have the capacity to promote photosynthesis.
Rockweed Habitats as Carbon Sinks

Since respiration was the dominant process occurring in the experiment, the hypothesis that rockweed habitats can act as a carbon sink is not supported by these experimental results. This deliberation was made on the knowledge of carbon sinks being characterized as areas with high primary productivity that absorb more carbon through photosynthesis than is lost through respiration (Hill et al., 2015). Although other research evaluating the ability of rockweed to act as a carbon sink is severely lacking, as is the case with most information on wild algal communities, the results of this experiment are still very unexpected. As mentioned before, rockweed has been documented as having a relatively high productivity rate and is abundant in the rocky intertidal zones of Maine, even though it is a commercially harvested species. Thus, the failure to classify rockweed habitats as potential carbon sinks is surprising, given the abundant literature that documented other macroalgal communities having high productivity and the capacity to act as carbon sinks. Duarte et al. and Sondak et al. (2017) and Chung et al. (2013) have stated wild seaweed communities are important to ecosystems due to their abilities to remove DIC from seawater which can lower the amount of CO$_2$ in the area and potentially mitigate local effects of climate change. Some studies only classify macroalgal communities as short-term CO$_2$ sinks (Hill et al., 2015) due to their inability to sequester carbon. Carbon sequestration is the photosynthetic fixation and long-term storage of carbon that occurs in carbon sinks (Hill et al., 2015). Unlike other carbon sink ecosystems, like mangrove forests, seagrass beds, and salt marshes, macroalgae grows on rocky substrates and do not develop significant carbon deposits, like soft sediment (Duarte et al., 2013; Hill et al., 2015). If macroalgae has any ability to store carbon, it
would be in the form of above ground living biomass which is not as efficient in carbon sequestration as below-ground stores of carbon seen in other carbon sinks (Hill et al., 2015).

However, recent evidence has found macroalgal communities indirectly contribute to carbon sequestration as being “carbon donors” (Hill et al., 2015; Sondak et al., 2017). Hill et al. (2015) defined “carbon donors” as autotrophs that donate carbon to another receiver habitat that has the capacity to bury carbon, such as salt marshes or seagrass beds. Sondak et al. (2017) supports this claim in stating macroalgae absorb CO₂ in photosynthesis and use it to increase their biomass, as autochthonous carbon, that has the potential to be transferred and deposited to other ecosystems, as allochthonous carbon. Rockweed appears to be an ideal candidate to function as a carbon subsidy between the shores of rocky intertidal zones and carbon sinks due to its abundance in its ecosystem, large biomass, and high productivity described in literature. Yet, this experiment does not support that claim because of the low productivity observed in rockweed.

**Future Research**

Future studies exploring the potential of wild macroalgal communities acting as carbon sinks should measure productivity in both controlled laboratory conditions and the natural habitats of rockweed. This would account for possible stressors to the algae related to the experiment, like removal and transportation. Studying algal productivity in natural environments would provide a more accurate representation of how algae interact with heterotrophs on a community scale. This would be beneficial to better understanding the productivity of the rocky intertidal community and the role of macroalgae within this
system. Conducting an experiment in the algae’s natural environment could show the direct effects of climate change such as warming oceans, more acidic conditions, and higher sea levels on the health of the algae and other organisms occupying the rocky intertidal zone. An experiment set in the rocky intertidal zone could also shed information on the effects of rockweed harvesting on the productivity of the immediate crop area and how it compares to protected areas. Parameters to be measured in natural environments should include algae coverage of an area, net primary productivity, growth rate, biomass turnover, and other variables recorded in other literature (Hill et al., 2015). Future research in laboratory settings should allow the algae to float freely in its system, instead of being submerged, to mimic how rockweed floats on the surface of seawater in its natural environment. It would be interesting to observe the effects of drying the tanks for portions of the day during the experiment to imitate emersion that rockweed would experience in the rocky intertidal zone. Also, the pH of the experiment should vary between acidic and slightly basic conditions to compare the performance of rockweed in different settings. At the very least, future studies should utilize more sophisticated lighting known to support photosynthesis.
CONCLUSION

The results of this study do not provide evidence supporting the claim that rockweed has the capacity to function as a carbon sink. Respiration was observed to be the dominant process occurring in all treatments of the experiment and suggests rockweed habitats would be a poor candidate for a carbon sink. However, results of this experiment potentially point to stress mechanisms surrounding constant submersion that could explain the limits of rockweed establishment in the lowest levels of the intertidal zones. Therefore, there are several aspects of the experiment that would benefit from replication, adjustment, or expansion in future studies. Numerous studies have documented the ability of commercial macroalgae communities to act as carbon sinks and sequester carbon through harvesting, but little research has been done to examine the potential of wild, unmanaged macroalgae communities to act as carbon sinks and alleviate the effects of climate change. Further research should be conducted to better understand the abilities of algae to store carbon as a possible mitigator for the impacts of climate change on vulnerable ecosystems.
TABLES AND FIGURES

Figure 1. Rockweed bed at Mitchell Marsh Preserve where collection of algae and snails occurred.

Figure 2. Photograph of a control treatment tank filled with Instant Ocean solution and containing a HOBO logger.
Figure 3. Photograph of rockweed treatment tank filled with Instant Ocean solution and containing a HOBO logger and a weighted down rockweed individual.

Figure 4. Photograph of invertebrate and rockweed tank filled with Instant Ocean solution and containing a HOBO logger, weighed down rockweed individual and five periwinkle snails contained by a basket.
Figure 5. Salinity (ppt) values of control (black), invertebrate and rockweed (light gray), and rockweed (dark gray) treatments in Trial 1 (dotted line) and Trial 2 (solid line). Salinity was measured in all tanks and averaged by treatment.

Figure 6. Conductivity (µS/cm) values of control (black), invertebrate and rockweed (light gray), and rockweed (dark gray) treatments in Trial 1 (dotted line) and Trial 2 (solid line). Conductivity was measured in all tanks and averaged by treatment.
Figure 7. Dissolved oxygen (mg/L) values of control (black), invertebrate and rockweed (light gray), and rockweed (dark gray) treatments in Trial 1 (dotted line) and Trial 2 (solid line). Dissolved oxygen was measured in all tanks and averaged by treatment.

Figure 8. Temperature (˚C) values of control (black), invertebrate and rockweed (light gray), and rockweed (dark gray) treatments in Trial 1 (dotted line) and Trial 2 (solid line). Temperature was measured in all tanks and averaged by treatment.
Figure 9. Departure from oxygen saturation (μM) for control, invertebrate and rockweed, and rockweed treatments with Trial 1 (dark gray) and Trial 2 (light gray) analyzed separately. Bars represent mean departure from oxygen saturation and error bars represent standard error of the mean.

Figure 10. Standard deviation of departure from oxygen saturation value (μM) for control, invertebrate and rockweed, and rockweed treatments with Trial 1 (dark gray) and Trial 2 (light gray) analyzed separately. Bars represent mean departure from oxygen saturation and error bars represent standard error of the mean.
Table 1. One way ANOVA on habitat complexity treatments predicting response variables: dissolved oxygen departure from saturation for a) trial 1 and b) trial 2; temporal variation of dissolved oxygen departure from saturation for c) trial 1 and d) trial 2.

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<th>Response</th>
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<td>c) Temporal Variation (T1)</td>
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