Cost-Benefit Analysis of Macroalgal Harvesting for Nitrogen Abatement

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Abstract

This project explored seaweed (macroalgae) harvesting as a means to remove nitrogen from eutrophic estuaries and compared the costs of suction harvesting to other nutrient abatement approaches. Elevated nutrients in estuaries can trigger macroalgal blooms, which can suffocate and kill marine organisms. Harvesting macroalgae removes nitrogen from estuaries and can improve the health of aquatic ecosystems. Macroalgal biomass and nitrogen surface water data were collected to try to identify an optimal macroalgal harvest window. A macroalgal harvesting pilot study was conducted to determine the costs of suction harvesting and how much macroalgae and thus nitrogen could be harvested on a dollars/kg basis.

The costs for nitrogen abatement measures, such as reducing fertilizer use or wastewater treatment upgrades, range from $1 to $16,000/kg of nitrogen removed. This study found the cost for removing nitrogen using suction macroalgal harvesting to range from $62 to $3,271/kg, depending on macroalgae condition and wages. The costs per kg of nitrogen removed for macroalgal harvesting were on the higher end of the range of interventions. However, this approach has a low point of entry cost of roughly $1000 compared to some interventions that require millions of dollars of investment. In addition, there is potential to improve the efficiency of harvesting and lower costs through refinement of harvesting methods.

Macroalgae harvesting is unlikely to remove enough nitrogen to resolve nitrogen-loading problems in eutrophic estuaries. However, it presents a useful option for

communities to reach their water quality goals in conjunction with other measures and may provide some benthic habitat benefits in the short to medium term.

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Introduction

Coastal development in the United States has increased dramatically in the past forty years and nitrogen (N) inputs into coastal waters have more than doubled (Howarth, 1998). Most coastal counties in New England have seen population increase at more than double the national average between 1960 and 2008 (USCB, 2010). Development of the coastal zone often results in elevated nutrient loading to coastal waters. Elevated nutrients in bays and estuaries can degrade water quality and trigger seaweed (macroalgae) blooms, which can foster hypoxic and anoxic conditions that decrease the abundance of invertebrates and fish, replace seagrass meadows and suffocate aquatic organisms (Fox, Stieve, Valiela, Hauxwell, & McClelland, 2008).

Methods to reduce nutrients before they reach the water, such as wastewater treatment plants, are expensive and require considerable space. Because macroalgae concentrate N into their tissues as they grow, harvesting macroalgae presents a novel method to remove N from estuaries. Macroalgal harvesting has been conducted in a few locations for aesthetics and to prevent odors and anoxic conditions from rotting seaweed. The only known empirical investigation into harvesting of macroalgae for N removal was conducted in Prince Edward Island (PEI), Canada (Crane & Ramsay, 2012). The PEI study used large macroalgal harvester machines and the main goal of the study was to prevent anoxia. The PEI study had the support of multiple regional government agencies and personnel and a budget of at least $170,000.

Macroalgal harvesting has taken place for generations in coastal communities, mostly as a seasonal resource in conjunction with fishing. More recently macroalgal harvesting for certain species has become more intense. For example, harvesting of kelps in Canada and Norway has developed in recent decades into a large commercial industry. In Italy removal of nuisance seaweed from Venice Lagoon has taken place since the late 1980s to prevent the effects of seaweed proliferation and decomposition (Caliceti, Argese, Sfriso & Pavoni, 2002). However, harvesting in Venice is done mainly for aesthetic purposes and uses large and expensive harvesting machines.

Small-scale suction harvesting of macroalgae from eutrophic estuaries presents a new approach to help address the impacts from excess N loading and opportunistic macroalgal blooms with a much smaller capital investment. Analyzing tissue N of the harvested macroalgae biomass allows for an accounting of the mass (kg) of N removed from the estuary by harvesting and allows for a direct cost comparison to other N abatement methods.

Research Significance and Objectives

To date there have been no known studies investigating the feasibility of small scale harvesting of nuisance macroalgae as a N reduction management tool. To appraise the potential for macroalgae harvesting to remove N, a macroalgal suction harvest pilot study was conducted and the costs of harvesting, including labor costs, equipment used such as pumps and tubing and end use were tallied and compared to other N abatement measures.

This thesis provides information about how much N can be removed from an estuary by suction harvesting macroalgae and what the approximate costs would be to allow communities to compare macroalgal suction harvesting to other N removal methods and develop N abatement plans that are acceptable to all stakeholders. Removing N from eutrophication estuaries will improve water quality, reduce anoxic stress on estuarine ecosystems and help attain N goals put forth in water quality regulations.

Background

Development and human activity can increase nutrient loading into nearby lakes, streams and estuaries. Elevated nutrients in estuaries can trigger macroalgal blooms, which can suffocate benthic organisms, resulting in fish kills and impacts on shellfish harvests (Valiela et al., 1992). Nutrient pollution is recognized as a growing problem in estuaries across the United States (U.S.), ranging from Long Island Sound to the plume of the Mississippi River in the Gulf of Mexico (Howarth, Sharpley, & Walker, 2002). Compared to relatively pristine reference ecosystems, human activity in the U.S. has increased N inputs into coastal waters by 280%, and by as much at 800% along the northeastern U.S. coast (Howarth, 1998).

The total N load for a water body consists of all the N inputs into the watershed including wastewater treatment effluents, septic systems, atmospheric deposition and fertilizers to name a few. Estuaries have a limited ability to assimilate N. When this limit is exceeded it can alter the aquatic ecosystem by causing reduced water quality and macroalgal blooms, which can cause anoxic conditions and kill aquatic organisms.

Eutrophication and Nutrient Loading

Eutrophication can broadly be expressed as an increase in any nutrient in water. Nitrogen is important in coastal marine ecosystems because it is the nutrient most often limiting primary productivity (Rose, Bricker, & Ferreira, 2015). In areas where excess N makes its way into surface water the primary productivity will often flourish, typically in plankton and macroalgal blooms. This is the case for many estuaries along the eastern U.S. where coastal development and wastewaters have been causing increases in N loading for decades. For many shallow estuaries the macroalgal biomass can have adverse impacts on native flora and fauna.

As communities began to notice large mats of seaweed growing over and replacing shellfish and seagrass beds they began to analyze what was happening to these important resources. It became apparent that anthropogenic nutrients were getting into the water and stimulating primary productivity, so communities with eutrophic waters began to try to control their nutrient loading. Unfortunately, many of the steps required to reduce nutrient loading are expensive, so progress dealing with eutrophication and nutrient loading has been limited.

Communities must address eutrophic issues to comply with the Federal Clean Water Act (CWA, 2002). Section 303(d) of the CWA provides mechanisms to restore and protect aquatic resources. The tool used to assess the efforts by communities to improve aquatic habitats is the Total Maximum Daily Load (TMDL), which sets water quality goals for communities to reach in order to maintain healthy aquatic resources (Howes et al., 2012). A TMDL represents the greatest amount of a pollutant that a waterbody can tolerate and still be considered protective of public health and maintaining beneficial uses of drinking, swimming, recreation and fishing. The TMDLs are meant to be attained using adaptive methods to allow flexibility for local communities to decide what works best for them to reach their TMDL goals.

The Massachusetts Surface Water Quality Standards (310 CMR 4.00) set water quality benchmarks for surface waters in the state (MSWQS, 2013). The surface water regulations state that: "surface waters shall be free from nutrients in concentrations that would cause or contribute to impairment of existing or designated uses and shall not exceed the site specific criteria developed in a TMDL" (MSWQS, 2013, p.15). In addition to nutrients, the Surface Water Quality Standards require waters of Massachusetts to meet other standards such as pH and dissolved oxygen. For example, all surface waters in Massachusetts must have dissolved oxygen concentrations of at least 5.0 mg/l to comply with water quality standards.

As a part of the CWA planning, Massachusetts established the Massachusetts Estuaries Project (MEP) in 2001 to assess the health of coastal ecosystems in southeastern Massachusetts (Howes et al., 2012). The purpose of the MEP was to identify N thresholds and determine what nutrient reductions will be needed to support healthy estuaries. The MEP provides an assessment of the nutrient status of an estuary, identifies all nutrient sources, sets N threshold concentrations to maintain water quality, analyzes N reductions needed to meet the TMDL and a modeling tool to evaluate N abatement alternatives.

Coastal Development Leads to Elevated Nutrient Loading

The population growth rate of Cape Cod (Barnstable County) in Massachusetts between 1960 and 2008 was seven times the national average (USCB, 2010).

Development of the coastal zone on Cape Cod continues and has resulted in elevated nutrient loading to estuaries, predominantly in the form of N from residential septic wastewater. Septic systems with leaching fields only remove about 10 – 20% of the N from the wastewater (Passeport, et al., 2013), so a large portion of the nutrients leach into the groundwater and make their way towards surface water.

Much of the geology of Cape Cod is comprised of sandy glacial moraine which means these wastewaters often travel quickly through the porous soils, limiting how much of the nutrients can absorb onto organic matter before reaching surface waters. Hauxwell, Cebrian and Valiela (2003) found that as the number of houses in a watershed increased, the estuarine N load generally increased (Table 1).

Table 1. Land-derived nitrogen loading rates modified from Hauxwell et al. (2003).

Variable	Estuary						
	Timms	Sage Lot	Hamblin	Jehu	Eel	Quashnet	Childs
Nitrogen loading rate (kg N/hectare/year)	5.3	7.6	28.4	30.1	62.7	298	407
Houses present in watershed	0	0	0	529	718	767	1233
Depth of estuary (meters)	1.3	1.3	1.3	1.7	1.4	0.8	1.4

Nitrogen Loading and Macroalgal Blooms

In many aquatic systems excess N loading can trigger plankton and macroalgal blooms. Macroalgal blooms can have many negative impacts on aquatic ecosystems and the environmental services these habitats provide. The impacts of nuisance macroalgal blooms on estuarine ecosystems are detailed below.

Impacts of Macroalgal Blooms on Eelgrass

Historically the dominant feature of the estuaries in Falmouth has been bottom cover of the seagrass *Zostera marina*, or eelgrass. In the past 30-50 years nutrient loading from nearby homes and septic systems has increased N loading. Elevated nutrient loading has resulted in increases in macroalgae and declines in eelgrass across Cape Cod. During the summer months macroalgal biomass can accumulate into mats as thick as 75cm or more (Rivers & Peckol, 1995a). In some cases nutrient loading has caused estuaries to lose 100% of their historical seagrass cover.

Eelgrass provides food for a variety of organisms, a nursery for developing fish and invertebrates, sequesters carbon and stabilizes the sediment. A loss of seagrass typically results in lower shellfish landings, reduced water quality and declines in fish population (Fox et al., 2008). Eelgrass losses in Massachusetts appear to be particularly hard on the Bay Scallop (*Argopecten irradians*) population (Valiela et al., 1992) (Figure 1). However, studies have found that seagrass habitat can recover in areas where anthropogenic nutrient levels have been reduced (Vaudrey, Kremer, Branco, & Short, 2010).

The status of eelgrass in Waquoit Bay has been severely impacted and the downward trend is related to development and nutrient loading into the estuary and eelgrass habitat loss. Hauxwell et al. (2003) determined eelgrass bed area for four Waquoit sub-estuaries subject to different N loading rates by collecting measurements on

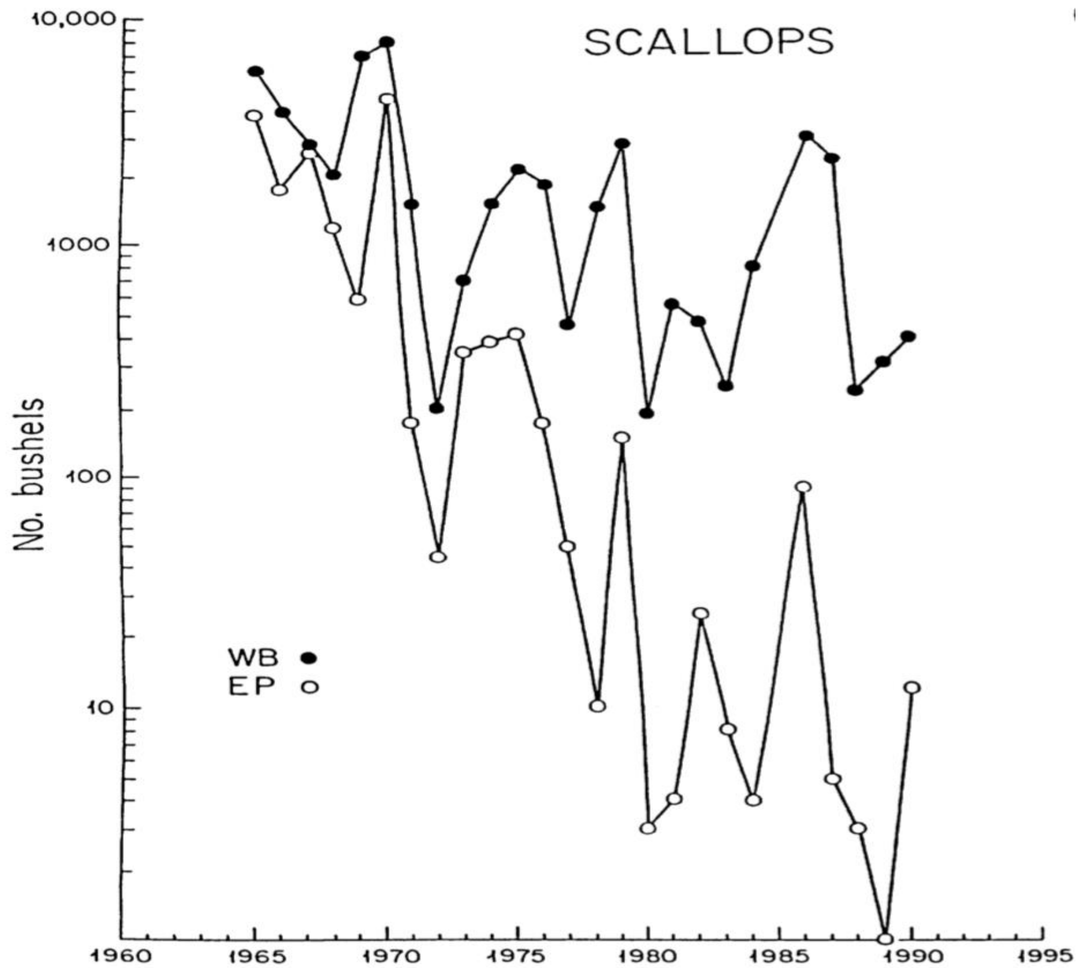


Figure 1. Reported scallop catch from Waquoit Bay and Eel Pond (Valiela et al., 1992).

eelgrass shoot densities, shoot biomass and various growth rates to compare to N loading rates. They found: "substantial eelgrass loss (80 to 96%) at loads of approximately 30 kg N/ha/yr. and total disappearance of eelgrass at loads ≥60 kg N/yr." (Hauxwell et al., 2003, p.59).

Deegan et al. (2002) conducted an experiment that assessed the effects of N loading and macroalgal biomass on eelgrass, benthic invertebrates and fish populations in three Waquoit Bay sub-estuaries. They found that as N loading increased, macroalgal biomass increased and eelgrass biomass and shoot density decreased. However, when they removed macroalogae from some of the test plots in areas with eelgrass and high macroalgal biomass they found increased eelgrass abundance as well as increased oxygen concentrations. They also found that lower macroalgal biomass resulted in higher fish and decapod abundance and biomass.

Removal of macroalgae may have more than just short-term benefits. Deegan et al. (2002) found that removal of macroalgae resulted in a: "denser cover of eelgrass that persisted into the following spring" (p.203). For eelgrass plots where macroalgae was removed, the shoot density the following spring was four times greater than control plots and 10 times greater than the high macroalgal biomass treatment plots. Low macroalgal biomass treatment plots had twice as much oxygen at the surface as did the other treatments. They concluded that removal of macroalgae increased abundance of eelgrass and suggested that excessive macroalgae interferes with eelgrass growth through light limitation.

Dissolved Oxygen Impacts of Macroalgal Blooms

Organic matter associated with macroalgal biomass stimulates sediment oxygen demand by increasing benthic respiration, which reduces the bottom dissolved oxygen (D.O.) concentration making it less hospitable for a variety of organisms (Green, Sutula, & Fong, 2014). D'Avanzo and Kremer (1994) measured considerable diel swings in oxygen concentrations on the bottom in the Childs River with night concentrations down to a low of 1-3 mg/l. During sunny days the D.O. would reach 10-15 mg/l due to photosynthesis (Figure 2).

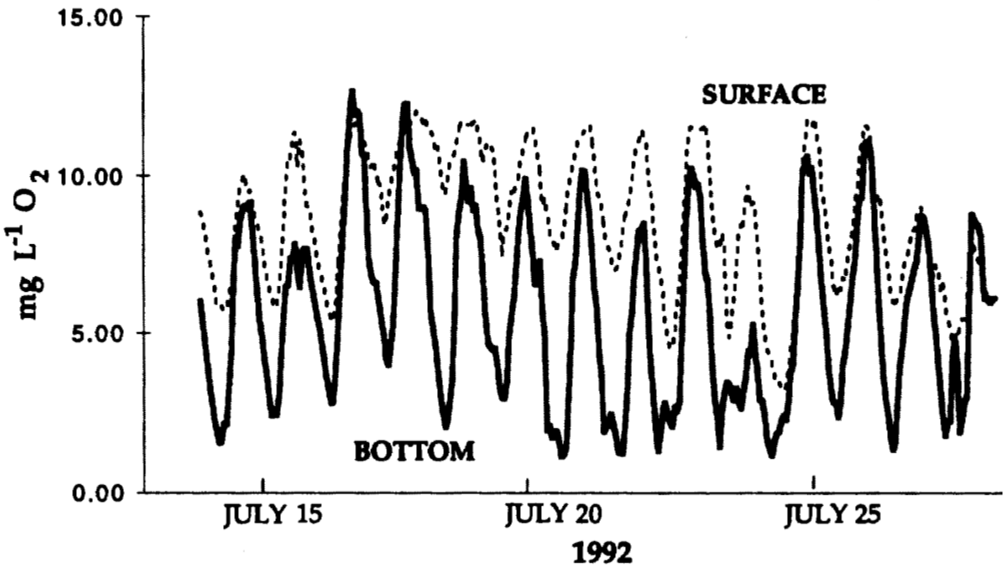


Figure 2. Continuous O_2 measurements of surface (dotted line) and bottom water (solid line) in the Childs River (D'Avanzo & Kremer, 1994).

Macroalgae can smother organisms and habitats by developing dense mats over the bottom, which can result in hypoxic or anoxic bottom conditions and kill benthic flora and fauna. Macroalgal mats can create dramatic oxidation/reduction transition zones. For example, Peckol and Rivers (1996) found a steep decline in oxygen levels within *Cladophora vagabunda* mats, falling to anoxic conditions within a few centimeters. Anoxic conditions caused by large accumulations of macroalgae impact macrophytes like eelgrass as well as invertebrates. Reduced oxygen concentrations and anoxia alters the oxidation/reduction (redox) status of the bottom, which can trigger the release of sulfide and ammonium concentrations, which can be toxic to many estuarine species.

Opportunistic, fast growing macroalgae can take up large amounts of nutrients in eutrophic waters and allow them to dominate slower growing macroalgae. The quick growth of opportunistic macroalgae can result in boom and bust cycles with rapid biomass growth followed by smothering. If growing conditions turn less favorable, such as several cloudy days, a portion of the biomass then dies and quickly decomposes on the bottom. This increases the organic content of the sediment and provides a ready supply of nutrients that then can re-enter the nutrient cycle and stimulate more macroalgae growth.

Invertebrate Impacts of Macroalgal Blooms

Hypoxic and anoxic conditions from decomposing macroalgae can cause sulfide and ammonium concentrations to rise, which can be toxic and cause the diversity and abundance of sediment invertebrates to decline. Benthic invertebrate declines have impacts on estuarine birds and fish that feed on the invertebrates. An example of the impacts of macroalgal blooms on invertebrates is reported in a study in the Baltic Sea by Norkko & Bonsdorff (1996). They set up 50 cm by 50 cm macroalgal plots and stocked them with approximately 2 kg wet weight/m^2 (ww/m^2) of macroalgae. After nine days the sediment surface under the macroalgal treatments were completely black due to anoxia and adult *Macoma balthica* clams were noted on the sediment surface. Impacts to the *M. balthica* stocks could affect flounder populations since adult *M. balthica* are a major food source for flounder.

Norkko & Bonsdorff (1996) observed complete community breakdown after 16 to 21 days of algal cover and only opportunistic and hypoxia tolerant species, such as the opportunistic polycheate *Capitella capitata* remained under the algae. Invertebrate abundance was reduced 87% in areas under the macroalgae compared to controls without macroalgae. Invertebrate biomass was reduced by 94% in the macroalgae treatments. They estimate that one hectare of uncovered sand would sustain 242 million individuals and one ton of benthic macrofauna, versus 31 million individuals and only seven kg of benthic macrofauna for one hectare covered with macroalgae.

Reduced Larval Settlement from Macroalgal Blooms

The information about how macroagal blooms effect larval settling suggests it negatively impacts at least some larval organisms. Bolam, Fernandes, Read and Raffaelli (2000) implanted *Enteromorpha* onto meter-squared plots in the well-oxidized Firth of Forth in Scotland at rates of one to two kg/m^2 ww. This caused marked changes in the macrobenthos as well as significant changes in all of the measured sediment variables. They conclude that the negative effect of *Enteromorpha* was due to larval filtering, suggesting macroalgae is: "likely to have detrimental effects on population maintenance of most species that rely on planktonic larval recruitment" (Bolam et al., 2000, p.123).

Similar adverse effects of high macroalgal biomass have been seen by other researchers; for example, Norkko & Bonsdorff (1996, p.154) found that: "macroalgal mats form a physical barrier that impacts the initial phase of recruitment by the filtering of settling larvae". Bonsdorff (1992) found that algal mats with a biomass of 832 + 60 g/m^2 dry weight (dw) filtered more than 70% of settling *Macoma balthica* larvae when compared to bare sandy bottoms. Wennhage and Pihl (1994) found that juvenile flat fish preferred to recruit to bare sand compared to plots with dense macroalgal cover.

Reduced Fisheries Stocks from Macroalgal Blooms

Empirical information on the effects of macroalgal blooms on fisheries stocks is limited. However, there are a few statements in the literature that suggest macroalgal blooms adversely affect some fish populations. For example, Deegan and Buchsbaum (1991) observed that short-term anoxic events (<24 hours) in eutrophied areas can kill an entire year-class of fish, especially those with high site fidelity such as winter flounder. More broadly, Sutula (2011) suggests that macroalgal blooms can cause fish kills, reduce fish larval settlement and result in declines in fish stocks.

At a minimum it is conceivable that excessive macroalgal biomass that cover the bottom in shallow estuaries has the potential to impact larval settling for a variety of species due to the alterations in sediment chemistry. The reduced oxygen levels and elevated sulfide and ammonium concentrations may make areas beneath macroalgal mats inhospitable for settlement for certain larval species, including bottom species such as flounder.

Localized Acidification from Eutrophication and Macroalgae

Over the past 200 years our carbon emissions have resulted in empirically measured reductions in pH (Orr, et al., 2005). Roughly one-third to one-half of the carbon dioxide emitted into the atmosphere makes its way into ocean waters. Ocean acidification has broad implications for aquatic life because it can reduce calcium availability potentially depriving calcium to aquatic organisms that require it for survival.

In addition to ocean acidification from carbon dioxide, researchers have found that localized eutrophication can also result in acidification of surface waters (Borges & Gypens, 2010; Cai et al., 2011). Eutrophic waters often have spikes in biological primary productivity in the form of plankton or macroalgae. When that biomass dies, most of it settles to the bottom. Benthic microbial communities consume the resulting detritus. The carbon dioxide generated by the microbial respiration results in localized reduction in pH and increased acidity (Cai et al., 2011). This can adversely impact species that have limited mobility and may impact larval settling, especially for organisms with critical calcium needs during their settling phase, such as mollusks. An example of this phenomenon was captured in data collected by Sfriso, Pavoni, Marcomini and Orio (1992) in Venice Lagoon (Figure 3). Note the drop in pH and D.O. in early June and mid-July.

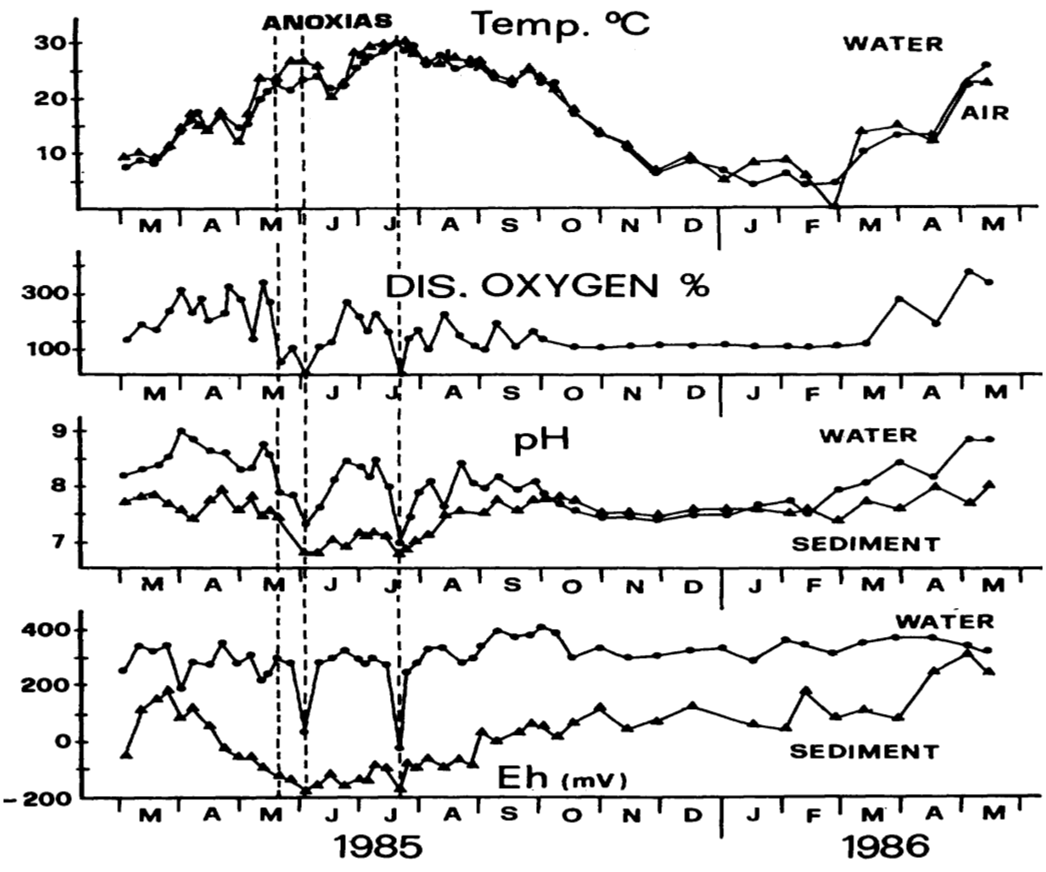


Figure 3. Temperature, D.O., pH and redox potential (Eh) of Lido Station Venice Lagoon showing anoxic system collapse (Sfriso et al., 1992).

Estimating a Macroalgal Biomass Threshold

Macroalgae in a balanced aquatic ecosystem can be an important part of a healthy ecosystem. However, when nutrients are elevated and aquatic systems become overrun with opportunistic macroalgal species, macroalgal blooms can pose a number of ecosystem problems. From a management perspective it would be useful to know if there is a threshold of macroalgal biomass where adverse effects begin to become apparent. Table 2 provides an overview of some studies that have considered this problem.

Table 2. Summary of macroalgal biomass adverse effects thresholds.

Location	**Effect**	**Dry weight** g/m2	**Wet weight** g/m2	**Reference**
California	Reduced macrofauna	110 - 120	840 - 930	Green, Sutula & Fong, 2014
California	Reduced D.O.	175 - 358	1,400 - 2,900	Sutula et al., 2014
Virginia	Anoxia, reduced biomass & diversity	>100	>830	Thomsen, McGlathery & Tyler, 2006
Venice Italy	Adverse Effects	90	700	Bona, 2006
United Kingdom	Management action warranted	70	500	Scanlan, 2007

Green, Sutula and Fong (2014) conducted an experiment in California that looked at the impacts of adding varying densities of macroalgae to treatment plots and measured the effects to invertebrates. They found that mats of *Ulva sp*. one cm deep, equivalent to 110-120 g dw/m2 or 840-930 g ww/m2 resulted in the reduction of macrofaunal abundance by at least 67% and species richness by at least 19% within two weeks.

Sutula, Green, Chicchetti, Detenbeck and Fong (2014) developed a model in an attempt to correlate biomass with redox potential and estimated that macroalgae biomass impacted the redox conditions between 175 g dw/m^2 and 358 g dw/m^2 (roughly 1.4 to 2.9 kg ww/m^2) with a median of 319 g dw/m^2 (2.6 kg ww/m^2). Their model suggests that at macroalgal biomasses exceeding 300 g dw/m^2 (2.5 kg ww/m^2) surface sediments will begin turning anoxic. Thomsen et al. (2006) reported observing several summer anoxic events in Chesapeake Bay in areas with macroalgal biomass reached over 100 g/m^2 dw (830 g/m^2 ww) with associated reductions in faunal biomass and diversity. Appendix 1 provides a summary of the effects of macroalgal blooms on fauna, modified from Sutula et al. (2014).

Bona (2006) conducted a study in the Venice Lagoon, Italy looking at apparent redox potential discontinuity (aRPD) as it relates to benthic habitat quality and macroalgal biomass. The aRPD method delineates between the light colored aerobic sediments and the grey to black hypoxic or anoxic sediments using sediment profile imaging. Bona found adverse effects to benthic macro invertebrates at 700 g ww/m^2 (90 g dw/m^2) in the Venice Lagoon. She also reported that locations with >700 g ww/m^2 always had seaweed cover greater than 40%. While not explicitly stated by Bona, this suggests that the >40% cover field observation could potentially be used as a rough metric for identifying areas where macroalgal growth may be negatively impacting sediment conditions. However, field cover percentage estimates pose a number of complications.

Scanlan et al. (2007) proposed a macroalgal assessment tool that uses biomass and percent cover to categorize estuary ecological status from bad to moderate to high. This tool identifies macroalgal biomass of greater than 500 g ww/m^2 m^2 (70 g dw/m^2) as indicative of a moderately impacted benthic invertebrate community.

Nitrogen Loading Abatements

There are many methods that can be used to reduce nutrient loading to coastal waters (Table 3). However, many of the interventions are expensive and require large amounts of space to implement. For example, wetland construction and restoration can be used to remove N, but this requires considerable space near the water, which may already be developed or expensive to purchase. The costs for N removal range from $0.22 to $16,742/kg ($0.10 to $7,610 per pound) N removed. N removal cost estimates in Table 3 were compiled from across the U.S. and Europe.

Table 3. Cost ranges of nitrogen removal options (modified from Rose et al., 2015).

Strategy	Cost per # Nitrogen Removed	Cost per kg Nitrogen Removed
Shellfish	$5.70 - $150	$12.54 - $330
Agricultural	$0.10 - $470	$0.22 - $1,034
Urban Stormwater	$30 - $3,629	$66 - $7,984
Wastewater treatment upgrades	$0.50 - $7,610	$1.10 - $16,742
Wetlands	$0.60 - $214	$1.32 - $471
Other	$2.80 - $218	$6.16 - $480
Macroalgal suction harvesting	$28 - $1,487	$62 - $3,271

A few coastal towns, including Falmouth, are removing N from estuaries with oyster aquaculture. Oysters filter and consume phytoplankton, which are abundant in eutrophic estuaries. When oysters are harvested the N in their shells and tissues are removed from the estuary, helping to reduce the nutrient load. Shellfish harvesting as a N removal method can range in cost from $5.70 to $150 per pound of N removed (Rose et al., 2015).

The Town of Falmouth has been promoting shellfish culturing and propagation since 1974 and has an ongoing shellfish propagation program (Falmouth, 2017). Initially the town focused on raising quahogs, but more recently oyster aquaculture has been used to help remove N from several eutrophic estuaries in town. Quahog production is still important for Falmouth with an average of 3.5 million seed quahogs planted every year from 2013-2016. The town has estimated that oyster and quahog culturing in suitable town estuaries could remove 3,455 kg N per year (Falmouth, 2017, T.14).

Shellfish Stimulate Macroalgae Growth

Aquaculturists have noted that the macroalgal population in and around aquaculture leases and the associated infrastructure tends to flourish. One theory posed as to why this occurs is that the macroalgae thrive as a result of the structure and support of the aquaculture gear such as nets, pens and bags, which also brings any macroalgae growing on the structure closer to the surface light. Another theory is that the macroalgae are growing at culturing sites due to the nutrients excreted by the shellfish. It may be that both of these factors are stimulating the growth of macroalgae around aquaculture sites.

Researchers have noted that molluscs excrete nutrients such as nitrates, ammonia and phosphorus as a part of their metabolism (Bartoli, Naldi, Nizzoli, Roubaix, & Viaroli, 2003; Murphy, Anderson, & Luckenbach, 2015). These nutrients can stimulate the growth of fast growing, opportunistic macroalgal species such as *Ulva*. Bartoli et al. (2003, p.158) reported: "excretion by clams measured in laboratory experiments range from 0.16-13 umol ammonium (NH_4) g/dw/h". At a minimum this suggests clam excretions could be a source of N for macroalgae in culturing areas. Ammonium is the preferred source of N for many opportunistic macroalgal species including for *Cladophora vagabunda* and *Gracilaria tikvahiae* (Peckol et al., 1994).

Bartoli et al. (2003) found that *Ulva rigida* growth was greater in areas where clams were present than in areas without clams. They noted that the daily difference between growth rates of the two areas was small (0.22/day vs. 0.20/day). However, this small difference in growth rate produced a +18% increase in total *Ulva rigida* biomass at the end of the experiment.

Another possible contributing factor to biomass spikes of macroalgae near shellfish beds and aquaculture sites is that the increased organic and nutrient loading in areas around the shellfish can stimulate the nitrification process and increase benthic nutrient recycling. For example, Gilbert, Souchu, Bianchi and Bonin (1997) found that a shellfish farming location had enhanced nitrification rates (12-fold for NH_4 oxidation; 3-fold for NO_2 oxidation) when compared to the control location.

Opportunistic macroalgae can pose problems for aquaculturists. These macroalgae can foul the nets and cages used to grow the shellfish and limit water exchange between the shellfish and surrounding waters. Equipment fouling can limit shellfish growth and in extreme cases could suffocate the shellfish. Murphy et al. (2015) suggest that macroalgal harvesting may be an effective means to negate the localized eutrophication spike caused by clam cultivation and remove unwanted nutrients from the ecosystem.

This unique set of circumstances makes for a potentially mutually beneficial management option to harvest macroalgae from shellfish culturing locations. This provides a benefit to the culturist in that it removes some of the nuisance macroalgae and would likely improve water circulation. It concentrates the macroalgal biomass in an area that may make it easier to harvest, which would benefit the macroalgae harvester. In addition, *Ulva rigida* cultured with clams not only had a higher growth rate but also: "maintained higher nitrogen content than *Ulva rigida* in bare sediment chambers" (Bartoli et al., 2003, p. 147). So, harvesting macroalgae around aquaculture sites may provide good N removal potential.

Harvesting Macroalgae

Globally the majority of macroalgal harvesting is undertaken for commercial purposes. There is a great deal of literature that covers the species collected, the most productive areas for growing and collecting, the products made and the markets served with commercial seaweed operations. This thesis is focused on the collection and harvest of macroalgae for non-commercial purposes as a means to reduce nutrient loading and/or improve ecosystem health. While all commercial macroalgal harvesting is not inherently bad for habitats, there are some instances where harvest practices have negative ecosystem impacts.

Most non-commercial macroalgal harvesting efforts target weeds that are invasive species, are considered a nuisance, or foul waterways. Removal of nuisance macroalgae often involves the use of large aquatic weed harvesting machines, which are essentially boats with a conveyor belt and a cutting mechanism to cut and pull the macroalgae out of the water and onto the boat (Figure 4).

Figure 4. Example of *Ulva* bloom and aquatic weed harvester (Crane & Ramsay, 2012).

Information on harvesting of macroalgae ranges from general information such as the amount a free diver in Japan can gather in a day to large multi-year harvesting operations funded by regional governments. Appendix 2 provides an overview of macroalgal harvesting methods. Benefits to removing excess or blooming seaweed include improved aesthetic value of the estuary, preventing noxious odors from developing due to biomass decomposition, improved water quality and improving benthic ecosystems by reducing or preventing anoxic stress.

Macroalgae Harvesting in Venice, Italy

The lagoons of Venice consist of a wide shallow basin covering 549 km^2 or 54,923 hectares (Sfriso et al., 1992). The lagoons produce approximately 1,000,000 tons (wet weight) of *Ulva rigida* biomass annually (Cuomo, Perretti, Palomba, Verde, & Cuomo, 1995). The City of Venice conducted a macroalgal harvesting program in the 1980s to address eutrophication in the estuary. This project was based on the removal of *Ulva rigida* from lagoons using aquatic weed harvesting boats. From the late 1980s to the early 1990s the Venice Water Authority harvested macroalgae between April and July using boats in shallow waters (Curiel, Rismondo, Bellemo, & Marzocchi, 2004).

Several years of macroalgal mechanical harvesting in the late 1980s and early 1990s removed approximately 90% of the macroalgal biomass from the lagoon, led to increased oxygenation of the sediment and changed the sediment from soft and black to firm and pale. In 1990 roughly 50,000 m_3 of *Ulva rigida* was collected from the lagoons. Biomass declined from 34,500 m_3 in 1991 to 3,500 m_3 in 1996. As biomass declined harvesting was carried out less frequently. By the early 1990s the macroalgal blooms diminished significantly and eelgrass began to colonize sediments that were no longer covered by extensive *Ulva* beds. The cost of these removal programs does not appear to be available in the published literature.

Macroalgae Harvesting in Orbetello, Italy

On the west coast of Italy there have been a number of studies investigating the effects of macroalgae harvesting in the Orbetello Lagoon. The Orbetello Lagoon is a 2,600-hectare estuary located in the southern coast of Tuscany. Coastal development has resulted in elevated nutrient loading, which has resulted in increased macroalgal biomass and deterioration of the traditional habitats in the lagoon. Eutrophication of this estuary has caused a reduction of eelgrass beds and significant macroalgal blooms, especially during the summer tourist season.

In 1987 the local government purchased an aquatic weed harvesting boat to remove some seaweed and improve the quality of the lagoon. Seaweed harvesting operations were conducted in the winter and spring months and harvested predominantly *Chaetomorpha sp*. Lenzi (1992) reports that 437,000 kg, or 437 metric tons (MT) of macroalgae was harvested in 1987. Macroalgae harvests increased up to 3,000 MT in 1989. Harvested macroalgae were dumped in open fields away from the town. Harvesting operations reportedly were conducted for as little as 2,300 lire per quintal (100 kg ww), or roughly $1.90 per quintal at 1992 prices. Assuming the macroalgae removed contained 2% N and the percent dw was 12%, this would amount to an estimated $7.92/kg of N harvested. This intervention reportedly influenced about 20% of the standing crop of macroalgae and resulted in a reduction in fermentation (anoxic) phenomena.

Macroalgae Harvest Modeling in the Po River Delta, Italy

A macroalgal harvesting program in the Sacca di Goro in the Po River Delta was modeled to estimate the costs and benefits of removing excess *Ulva rigida*, which had been causing anoxic conditions and heavy mortality of cultured shellfish in the area (Cellina, De Leo, Bartoli, & Viaroli, 2002). Cellina and colleagues estimated that the annual cost of mechanical weed harvesters was approximately $100,000 per harvester. They estimated that macroalgal harvesting in the Sacca di Goro resulted in an annual savings of approximately $3,000,000 per year. Estimated costs of doing nothing to address eutrophication with macroalgal blooms followed by collapse and subsequent clam death were as high as $1,000,000 per year. These estimates only considered losses to cultured clam sales and did not include other ecosystem services or benefits. Details pertaining to estimated total biomass removed and N content were not published.

Macroalgae Harvesting in Prince Edward Island, Canada

A pilot project harvesting sea lettuce (*Ulva lactuca*) using mechanical weed harvesters (Figure 4) was conducted in three eutrophic estuaries in Prince Edward Island (PEI), Canada in 2011. This study attempted to determine if it was possible to reduce or eliminate anoxic events in eutrophic estuaries and how much sea lettuce would need to be collected to reduce anoxia (Crane & Ramsay, 2012). The three eutrophic estuaries studied had heavy seasonal *Ulva lactuca* growth that frequently collapsed and decomposed after reaching maximum biomass. Using aquatic weed harvesting machines, a total of 146.2 MT of seaweed were harvested during the study. All harvested sea lettuce was either spread on agricultural land or composted.

The reported costs for this study were $581/hour of harvesting and $322/MT of harvested sea lettuce, although the report suggests these costs could be reduced by improving efficiency (Crane & Ramsay, 2012). The *Ulva* harvested reportedly contained between 0.49-0.60% N ww. Based on the average percent N of roughly 0.55%, the 146.2 MT of *Ulva* harvested would have removed approximately 804 kg of N or 5.5 kg N per MT of harvested *Ulva*. At the reported cost of $322/MT, the approximate cost per kg of N removed during this project was $58.55/kg N harvested (i.e., $322/5.5).

The report estimated that between 12 and 14 harvesters would be required to harvest all the impacted estuaries on PEI at a cost of $2,000,000 to $3,400,000. One limitation of the weed harvester machines is that they can only harvest from a set depth. For this project the harvesters were only able to collect *Ulva* at depths between 0.6 to 1.6 meters. Macroalgae outside of this depth range was not collected. This collection program focused on removal of *Ulva* mats that float to the surface, which does not occur in all areas where *Ulva* blooms.

Based on the available information for the PEI site it appears that total N removed was based on estimated weight of the number of truckloads of *Ulva* harvested multiplied by the percent N content. However, this is only a fraction of the total N load for the three estuaries and the amount of sea lettuce harvested was insufficient to prevent anoxia from occurring in the three estuaries. The provincial government does not regard sea lettuce harvesting as a solution to nutrient enrichment and are currently directing resources towards the reduction of nutrients at their source (C. Crane, personal communication, February 6, 2019).

Macroalgae Harvesting in Western Australia

In Western Australia one approach that is sometimes used to remove macroalgae is the harvesting of seaweed that washes up on the shore. A beach removal program in the Peel-Harvey region removed an average of 13,000 cubic meters of algae mostly from beach collections at a projected annual cost of $328,500. This project did not include total macroalgal biomass or N data to allow for a per kilogram N removal cost estimate. However, using estimates from a separate project, Crane and Ramsay (2012), estimated that 15 cubic meters of wet macroalgae weighs roughly 6000 pounds or 2.7 MT. Using this estimate, one cubic meter of macroalgae would weigh approximately 180 kg (i.e., 2700/15= 180). Assuming one cubic meter of macroalgae weighs 180 kg, 13,000 cubic meters would amount to 2,340 MT macroalgae collected from the beaches. If this macroalgae was 12% dw, that would amount to 280.8 MT dw harvested. Assuming the dried harvested macroalgae contained 2% N, this would amount to 5,616 kg N harvested off the beach or roughly $58.49/kg N removed. However, this is a crude estimate and nutrients may leach out of macroalgae once it is deposited on the shoreline.

Macroalgae Harvesting in the Caribbean and the Gulf of Mexico

Since 2011 massive blooms of the pelagic macroalgae *Sargassum natans* and *S. fluitans* have been causing problems in the Caribbean and the Gulf of Mexico when dense floating mats of seaweed drift onto beaches, sometimes covering miles of beach with seaweed a foot deep or more. These blooms appear to originate from the Central Atlantic Ocean as a result of upwelling of nutrients off the west coast of Africa and nutrient runoff from the Amazon River (Wang et al., 2019). Wang and colleagues estimate the June 2018 Sargassum belt from Africa to the Caribbean Sea contained more than 20 million metric tons of Sargassum biomass.

Beaches along the Mexican Riviera, including Cancun, have been intermittently inundated with thick mats of Sargassum, which stinks as it decomposes, posing problems for coastal ecosystems and the tourism industry. In 2015 the Mexican government spent $9.2 million on removal efforts in the state of Quintana Roo along the eastern coast of the Yucatan Peninsula (EFE, 2015). The 2015 Sargassum removal included hiring 4,600 workers to remove the seaweed from tourist beaches using backhoes and bulldozers to scrape the seaweed from beaches. The state of Quintana Roo has over 200 miles of eastern shoreline that has been impacted to differing degrees, yet from just one popular stretch of beach in Cancun, workers hauled away more than 1,000 truckloads of seaweed, estimated at half a million cubic feet (Partlow & Martinez, 2015). The Mexican navy has been called in to help deal with stinking piles of Sargassum on tourist beaches and is reportedly testing out a hydraulic suction pump to remove the seaweed before it reaches land. No detailed costs associated with the harvest methods could be located in the literature.

There is some agreement that it is best to collect Sargassum before it reaches the beach. However, this approach requires mechanical harvesters, which are expensive, costing upwards of $150,000. In most instances the seaweed is removed with rakes, shovels and wheel barrels after the Sargassum has beached. In some areas booms or netting have been used to prevent seaweed mats from damaging sensitive habitats or getting into small harbors.

The Mexican government has used a seaweed harvesting machine called the Sarganeitor to help clean up the Sargassum problem before it reaches shore (San Pedro Sun, 2015). The Sarganeitor is like a floating combine, which can harvest up to 7.5 tons of Sargassum per day and is similar to the aquatic weed harvesters used in Canada and elsewhere (Figure 4). It works well for removing Sargassum before it hits the beaches, but costs around $300,000, so it is not an option for most communities.

Many Caribbean Islands are having Sargassum problems. Some islands are trying to find ways to use the beached seaweed as a resource, but in most cases it is taken from the beaches with rakes and wheel barrels and either composted or landfilled in areas away from tourists. Details on harvesting methods and costs are limited, but it is estimated to have cost the Caribbean community at least $120 million dollars in 2018 to deal with this issue (Caricom, 2019).

The City of Miami is dealing with large amounts of Sargassum seaweed that washes ashore in the summer. City officials have estimated that it would cost $45 million per year to remove the seaweed from the entire 15 miles of coast affected (Hanks, 2019). The city plans to use bulldozers, front-end loaders and trucks to haul away the seaweed from beaches with the most seaweed. The cost for this targeted removal is estimated to be $350,000 per month. Operations to remove the seaweed are complicated by the fact that some beaches are protected as sea turtle nesting areas.

Macroalgae Harvesting in Harwich, Massachusetts

In 1998 the Harwich Natural Resources Department attempted to remove the opportunistic macroalgae *Ulva lactuca* from Round Cove to reduce smothering and improve water quality in the cove by increasing D.O. This effort consisted of providing commercial shellfishermen with bins to gather any *Ulva* they encounter during harvesting. Shellfishermen would then bring the bins to the Natural Resources Department for disposal. According to town personnel, all *Ulva* recovered was disposed of at the town dump and no further details about biomass removed or N content were collected (H. Proft, personal communication, February 12, 2019).

Macroalgae Harvesting in Jamaica Bay, New York

In 2010 the New York City Department of Environmental Protection (NYCDEP) conducted a pilot project to harvest sea lettuce (*Ulva lactuca*). Recurring macroalgal blooms were decomposing and creating noxious odors, suffocating benthic invertebrates and suppressing spawning and nesting activities (NYCDEP, 2012). Using trash skimmer boats, which are similar to aquatic weed harvesters, they collected 2.5 cubic yards (1.9 m^3) of *Ulva* in 90 minutes of harvesting. An additional 300 gallons of sea lettuce were collected by hand. NYCDEP report that a portion of the harvested *Ulva* was successfully converted to butanol using fermentation. Costs for this project are not available.

Aquatic Weed Harvesting in Fresh Waters

In a number of areas in the U.S. towns and non-profit organizations are attempting to remove invasive weeds in fresh water lakes and ponds. Removal of weeds often targets the invasive European Milfoil (*Myriophyllum spicatum*), which can overwhelm native aquatic plants. Several removal programs have used Diver Assisted Suction Harvest (DASH) methods to pull up and remove invasive weeds. While these plants are different from detached floating macroalgae found in estuarine systems, the DASH method is similar to macroalgal suction harvesting conducted for this study.

The DASH collection method typically involves a Venturi-designed pump system to create water suction with which a SCUBA diver can use to target and remove invasive weeds. The advantage of a Venturi pump is that the materials suctioned up from the water never enter the pump housing and impeller, which allows for the collection of larger and more fibrous materials without clogging and damaging the pump. One complication with Venturi pumps is that they are custom built and more expensive than standard water or semi-trash pumps. There is no published literature available on the use of DASH harvesting methods conducted in marine environments.

The New York State Department of Environmental Conservation (NYSDEC) has prepared a draft primer on aquatic plant management (NYSDEC, 2005), which targets invasive and exotic freshwater plants. This report estimates that DASH equipment costs between $20,000 to $30,000 dollars and that labor costs for harvesting would range between $500-$1,000/day. NYSDEC estimate that costs of vegetation removal could range from $1,000-$25,000 per acre, not including harvesting equipment costs. The NYSDEC estimates that mechanical harvesting machines (Figure 4) range from $100,000 - $200,000 and cost $800 - $2,400 per acre to operate.

Potential Adverse Effects of Macroalgal Harvesting

Lavery, Bootle and Vanderklift (1999) investigated the potential ecological impacts of harvesting macroalgae from the shoreline in the Peel Estuary in Western Australia. Nuisance macroalgae in this area were harvested with a variety of methods to remove seaweed before it decomposed and caused foul odors in recreational areas. They found an initial decrease in the densities of epifauna and fish, but densities returned to baseline within two months of harvesting. This study found no effect on the densities or richness of benthic infauna. However, the authors point out that the most commonly used harvest method in this area consists of pulling macroalgae out of the area using front-end loaders and that the effects of harvesting macroalgae depend on the harvesting technique, frequency and intensity of the harvesting as well as the vulnerability of the species or habitat present. They conclude that the long-term effects of harvesting macroalgae appear to be positive.

A shellfish and sediment survey was conducted during a mechanical weed harvesting project in PEI, Canada in 2011 (Crane & Ramsay, 2012). This survey determined that there were no demonstrated impacts from the mechanical harvesting equipment. A concurrent study of bycatch reported that a variety of small fish, shrimp, clams, mussels and invertebrates were inadvertently collected during harvesting.

Macroalgae Harvesting as an Economic Resource

Macroalgal harvesting of economically valuable seaweeds occurs in many countries. As of 2015 the seaweed industry cultured or collected 30.4 million metric tons of wet macroalgae (Ferdouse, Holdt, Smith, Murua, & Yang, 2018), 97% of which was cultured. The seaweed industry provides a variety of products and had an estimated global annual value of $10.6 million in 2015. Most of the value of the industry is in food products for human consumption such as nori. As of 2003 there were 35 countries that had active commercial macroalgal harvesting operations.

In Canada, an industry revolves around the harvest and processing of Rockweed (*Ascophyllum nodosum*). Rockweed harvesting has been taking place since at least the 1960s in Nova Scotia (Doty, Caddy, & Santelices, 1987). Traditional hand and rake harvesting methods were replaced in the 1970s by mechanical methods. Mechanical harvesting typically uses aquatic weed harvesting boats that have a cutting blade to separate the fronds from the stipe and a conveyor belt that brings the Rockweed onto the boat. There are some concerns in communities where Rockweed harvesting occurs that over harvesting could have negative impacts on intertidal ecosystems.

In a few instances harvested macroalgae has been considered as a feedstock for biogas or biodiesel production. For example, in Prince Edward Isle, Canada harvested *Ulva lactuca* was considered as a potential feedstock biomass for biogas production (Crane & Ramsay, 2012). The estimated value of harvested sea lettuce for biogas production ranged from $45-$62 per ton of seaweed.

Nitrogen Removal via Macroalgal Aquaculture

Lindell, Yarish and Kim (2015) cultured *Gracilaria tikvahiae* in Falmouth, Massachusetts between 2012-2013 and found that they could use a portion of their harvest as a food item. Using rope-culturing methods, they harvested 132 kg ww of *G. tikvahiae* containing an estimated 264 grams of N. Lindell and colleagues estimated that as much as much as 75 kg of N per hectare could be removed by culturing *G. tikvahiae* over one growing season. However, costs associated with this effort were not estimated.

While culturing *Gracilaria tikvahiae* has potential in the region, there are complications with macroalgae aquaculture. The primary limitation to broader *G. tikvahiae* cultivation is cost. A considerable amount of time and money are required to culture seaweed in a laboratory before it can be planted in a natural setting to grow to maturity. Once in the estuaries the cultured plants become potential settling sites for all manner of settling planktonic organisms, or epibionts. Research has shown that the growth of epiphytes on macroalgae is probably the greatest constraint on commercial seaweed culture (Fletcher, 1995). As organisms settle on the cultured macroalgae they often reduce productivity and give the cultured macroalgae a less than marketable appearance, thereby reducing the market value of the cultured macroalgae.

Nitrogen Removal via Macroalgal Harvesting

While macroalgal blooms are often considered a nuisance, they can also be viewed as a way to concentrate N in an estuary. The macroalgae and associated N can be harvested and thereby removed from the estuary. While harvesting macroalgae is unlikely to resolve TMDL exceedances in highly eutrophic areas, it may present a useful option for communities to reach their TMDL goals in conjunction with other interventions and may provide short-term improvement to localized water quality and provide some benthic habitat benefits in the short to medium term.

The N content of macroalgae is variable depending on the species harvested, the concentration of N in the waters and the time of year. For the red algae *Gracilaria tikvahiae*, the dw N concentration typically ranges between 1.75-5% of dw (Lindell et al., 2015). The N content of the other dominant macroalgae in the area, *Cladophora vagabunda*, tends to range from 2.5% to 5.5% N (Thompson & Valiela, 1999).

Recent studies indicate that the invasive macroalgae *Gracilaria vermiculophylla* is present in the Childs River (Lindell et al., 2015). *G. vermiculophylla* is indistinguishable from *G. tikvahiae* to the naked eye and genetic testing is required to confirm species identification. Nettleton, Mathieson, Thornber, Neefus and Yarish (2013) reported the presence of *G. vermiculophylla* in five different Massachusetts estuaries in 2000, so it is possible *G. vermiculophylla* is present elsewhere in Waquoit Bay. Due to resource limitations, this study did not distinguish between the two *Gracilaria* species. *G. vermiculophylla* has broad tolerances to salinity, nutrients, sediment burial, grazing and temperatures. Tyler and McGlathery (2006) found that *G. vermiculophylla* had N tissue concentrations in the range of 2% to 4%.

To optimize N removal via macroalgal harvesting, the timing of the harvest should consider trends in macroalgal N tissue concentrations and biomass density. In general, the higher the N concentration in surface water, the higher the percent N in the macroalgal tissue. For example, Peckol et al. (1994) found that *Cladophora vagabunda* in high N water (Childs River) maintained higher N tissue concentration throughout the year. This is one of the advantages of conducting macroalgal harvest potential in the Childs River. The high N loading in the Childs should result in slightly higher tissue N concentrations and therefore better N removal rates.

When macroalgal biomass reaches maximum densities the N tissue concentrations and biomass begin to decline due to shading, suffocation and decomposition. For example Viaroli, Naldi, Bondavalli and Bencivelli (1996) found that the growth rate of *Ulva rigida* begins to decline when biomass reaches around 400 g/m_3 dw (~3,333 g/m_3 ww) and growth rates become negative at biomass densities over 1000 g/m_3 dw (~8,333 g/m_3 ww) (Fig 5).

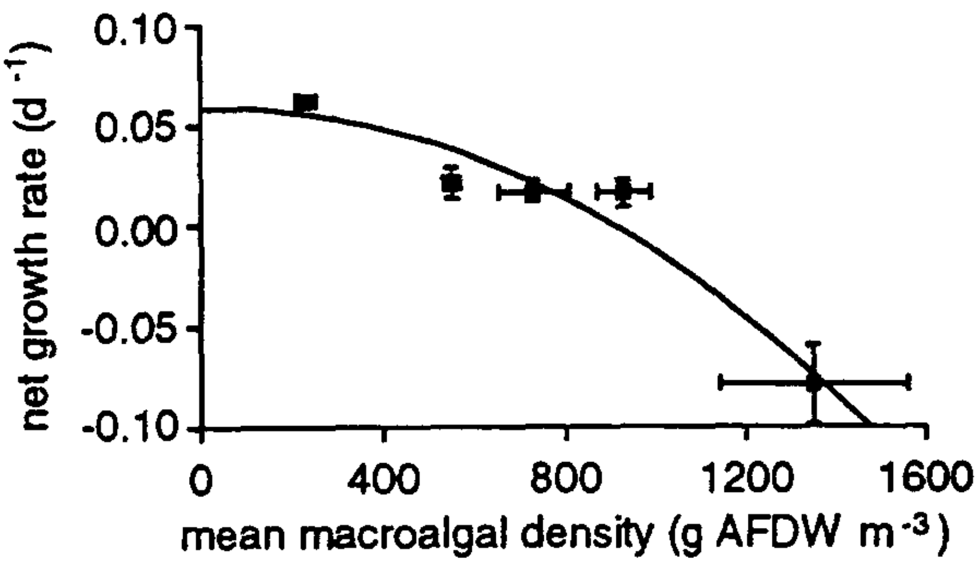


Figure 5. *Ulva rigida* growth rate vs. dry weigh biomass (Viaroli et al., 1996).

While macroalgal harvesting has had some success in areas where macroalgae often float on the surface and can be collected using harvester machines, there is very limited information about benthic or water column macroalgal harvesting in the literature. According to Ohno (1997) suction pumps are used for harvesting the brown macroalgae (*Cladosiphon sp.*) in Japan in many locations. However, details about these aquaculture operations are not available in the published literature.

Nitrogen Estuarine Impacts Case Study: Falmouth, Massachusetts

Falmouth, Massachusetts has 14 estuaries with MEP reports and N TMDL targets. This thesis focused on the largest estuary in Falmouth, Waquoit Bay. Waquoit Bay is a National Estuarine Research Reserve and totals approximately 1,632 acres (CCC, 2017). Waquoit Bay consists of the main bay, the Childs and Quashnet Rivers as well as four salt ponds (Eel, Hamblin, Jehu and Sage Lot). Most of the waters in the Waquoit Bay system are shallow well-mixed waters with an average depth around one meter. Nitrogen loading in Waquoit Bay has increased steadily since 1940, predominantly the result of residential development and wastewater from residential septic systems (Figure 6).

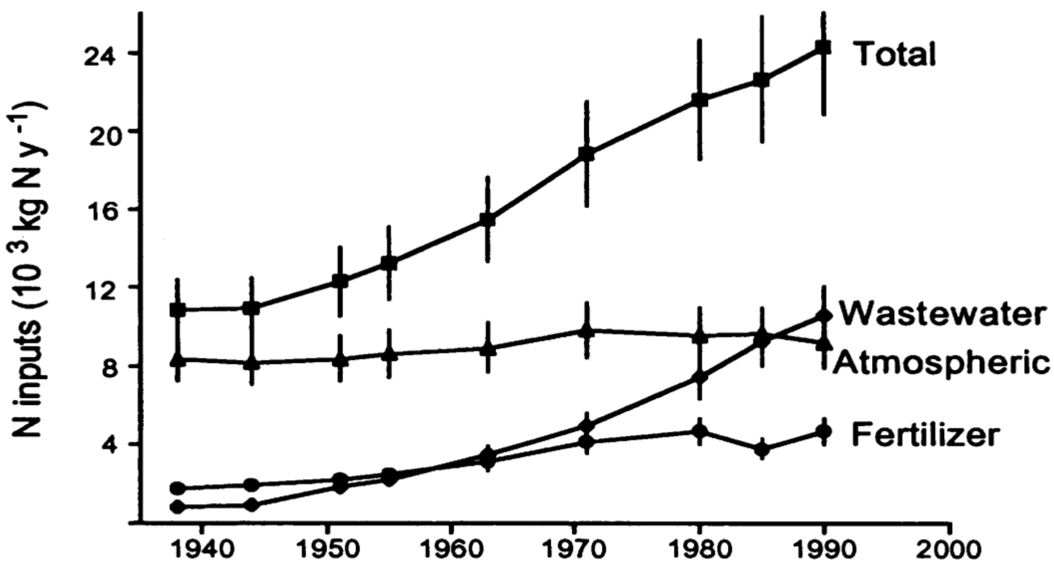


Figure 6. Nitrogen load increase for Waquoit Bay over time (Bowen, & Valiela, 2004).

The CWA N targets for Waquoit Bay are geared towards the restoration of eelgrass habitat, improved water clarity, and improved shellfish and fisheries resources (Howes et al., 2012). Improved benthic habitat quality is considered a secondary condition. The Waquoit Bay watershed load is 39,655 kg-N/year, which is well above the estimated N load capacity of 15,440 kg-N/year (Cape Cod Commission, 2017). Based on the TMDL loading estimates, Waquoit Bay is receiving more than double the tolerable limit for N. Elevated N loading has resulted in impaired water quality, dense macroalgal blooms, fish kills, loss of seagrass and reduced shellfish landings. Macroalgae biomass density ranges considerably in Falmouth, but can exceed two kg ww/m_2.

The estuary with the highest N load and macroalgal densities in the Waqouit Bay and Falmouth is the Childs River (Figure 7).

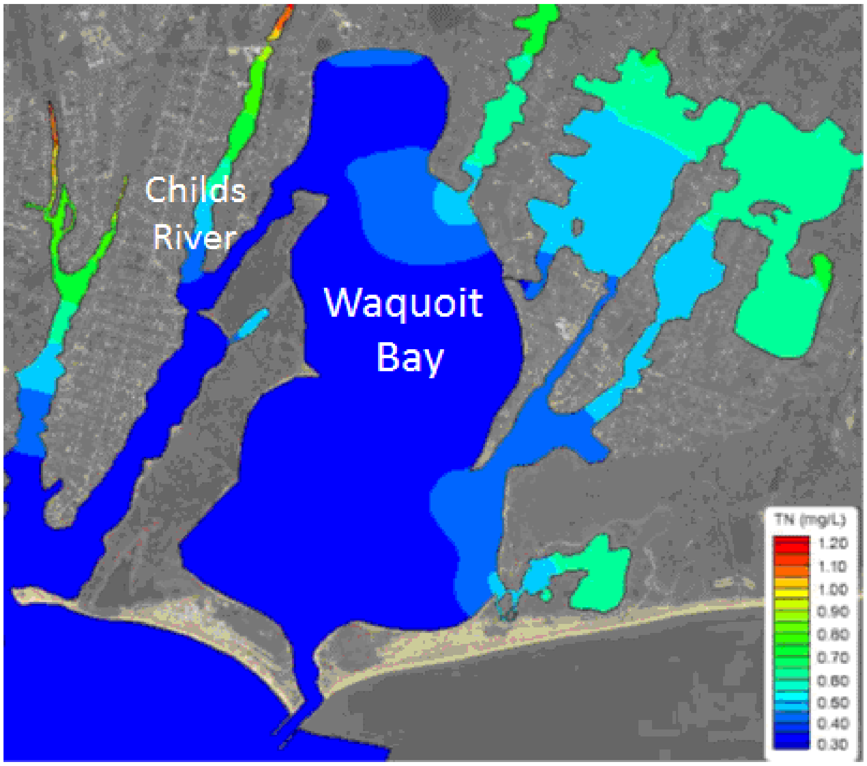


Figure 7. Nitrogen loading & macroalgal harvesting area (Howes et al., 2012).

Because the Childs River has high nutrient loading and high macroalgal densities, this area was targeted for macroalgal suction harvesting. The Childs River receives approximately 624 kg N/ha/yr (Hauxwell, McClelland, Behr, & Valiela, 1998).

For Cape Cod towns, the septic N loads represent the most significant N inputs to their estuaries (Figure 8). For example, the primary contributor to the N load in Waquoit Bay is wastewater (75%), followed by impervious surfaces (13%) and fertilizers (12%) (Howes et al., 2012).

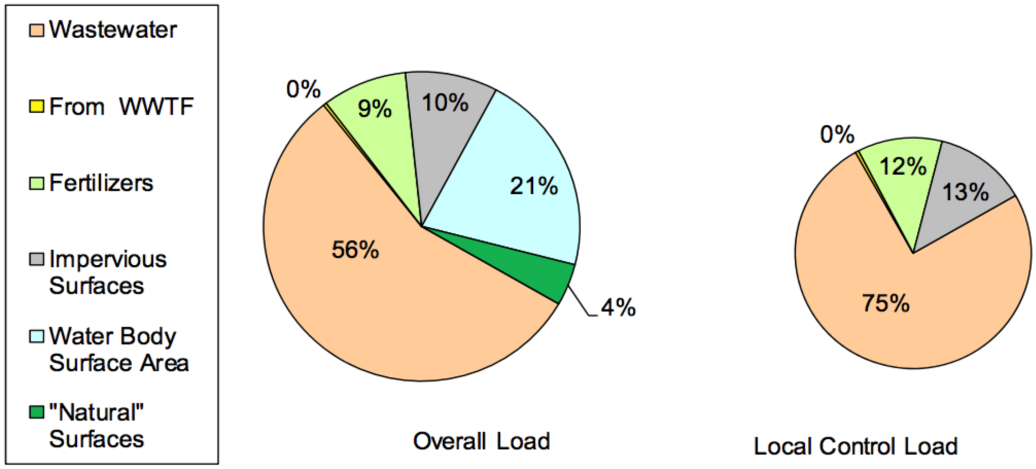


Figure 8. Land use nitrogen loads for Waquoit Bay (Howes et al., 2012).

The Town of Falmouth has been wrestling with eutrophication for decades and has implemented a number of programs to reduce nutrient loading and improve water quality. Falmouth is interested to pursue additional means to improve water quality. This makes Falmouth a good case study to appraise the potential application of macroalgae harvesting.

Research Questions, Hypotheses and Specific Aims

This study was conducted to determine the costs to remove N from estuaries by harvesting macroalgae using suction pump methods and to determine if macroalgal harvesting is a cost-competitive means to reduce estuarine N loading by addressing the following two questions and hypotheses:

1. How much N can be removed from eutrophic estuaries via macroalgal suction harvesting?

- H1: A crew of three workers can remove 2 kg N equivalents in one day of suction harvesting.

2. What are the costs per kg N removed of harvesting macroalgae compared to more traditional options?

   - H2: Macroalgal harvesting is a cost-competitive N abatement approach on a dollar/kg N removed basis compared to traditional N abatement methods.

Specific Aims

The specific aims of this research relating to the hypotheses required me to:

1. Identify macroalgal harvest areas within the Childs River and/or Waquoit Bay with the highest macroalgal N and biomass concentrations.
2. Harvest macroalgae and measure kg N harvested.
3. Determine the cost of macroalgae suction harvesting on a per kg N removed basis.
4. Develop a cost range estimate for estimated kg N removed by suction harvesting.
5. Compare macroalgae harvesting N abatement costs against other available N abatement measures.

Chapter II

Methods

In an effort to better understand the amount of nitrogen (N) that can be removed by harvesting macroalgae from a eutrophic estuary using modest suction harvesting equipment, the biomass and N tissue concentrations of macroalgae were measured from the Childs River within the Waquoit Bay estuary in Falmouth, Massachusetts. The Childs River consistently has the highest N loads in Falmouth. The Waquoit Bay Massachusetts Estuary Program (MEP) report (Howes et al., 2012) estimated the watershed N load for the Childs River as 12 kg/day. This is well in excess of what the estuary is estimated to tolerate, or the target load of 4 kg/day. The Childs River has high macroalgae biomass densities. Fox et al. (2008) reported macroalgal biomass of 165 grams dw per square meter (1.3 kg ww). These conditions make the Childs River a good location to test the feasibility of harvesting macroalgae for N abatement.

Macroalgal Data Collection

Field sampling and data collection occurred in the Childs River roughly 1000' north of where the Childs River joins the Seapit River. GPS coordinates for this location are 41.572, -70.534, or 41°, 34', 19.8" North, 70°, 32', 0.29" West. Data were collected from the eastern shore of the river along a stretch of the shore without any jetties or docks. This location is between the Waquoit Bay National Estuarine Research Reserve (WBNERR) sample locations CR166 and CR167 collected in 2004, 2007 and 2011 and is virtually identical to sample location CR6, collected in 2016 by researchers in the Valiela Lab at the Marine Biological Lab (MBL, 2016), which was at 41.571, -70.534. Macroalgal samples were collected from areas predominantly one meter deep or less.

For each of the five sampling dates, macroalgal biomass sampling consisted of five replicates of 0.25 m^2 quadrats in the selected location. Quadrat replicate locations were determined by haphazardly tossing the quadrats within 50 feet of the shoreline along a 300 foot stretch of the eastern shore of the river (Figure 9).

Figure 9. Haphazard quadrat sampling method.

This method has the advantage of being reasonably easy to implement as well as allowing for comparison to other macroalgal biomass studies (Hauxwell et al., 2003; Deegan et al., 2002; Thybo-Christensen, Rasmussen & Blackburn, 1993).

Ideally, all data would have been collected at the same phase of the tide. However, other practical considerations (weather, daylight hours, etc.) made this impractical. This added a layer of variability to the data because water samples were not always collected from the same phase of the tide and N concentrations are affected by the tidal cycle. Fujita (1985) found that ammonium concentrations can fluctuate from three to four fold between high and low tidal cycles in the nearby Bournes Pond estuary (Figure 10).

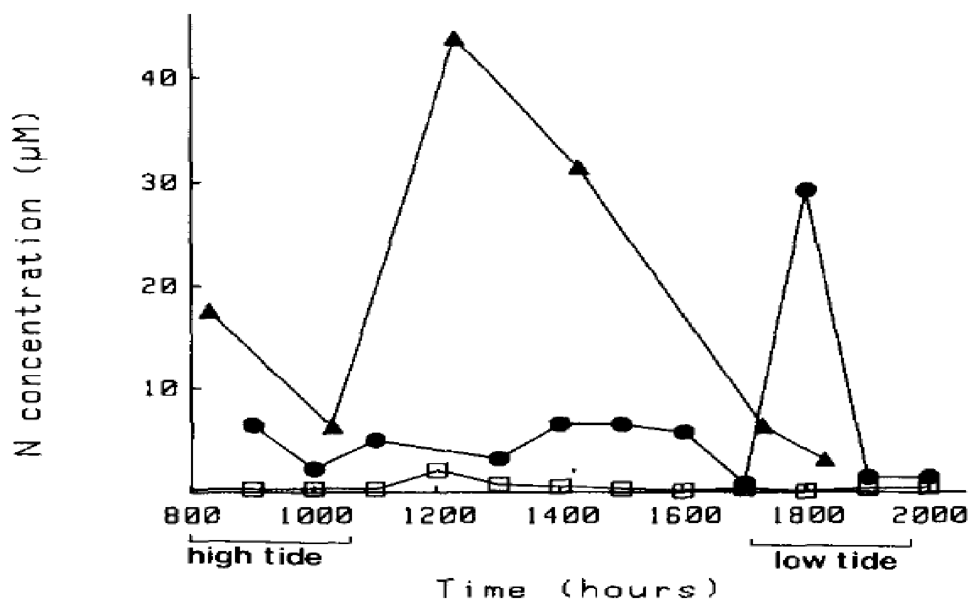


Figure 10. Tidal cycles of NH_4 (●), NO_3 (□) concentrations in Bournes Pond July 1984: NH4 concentration (▲) in August 1984 (Fujita, 1985).

Many seaweeds prefer ammonium as a N source (McHugh, 2003). Ammonium is the preferred N source for *Cladophora vagabunda* and *Gracilaria tikvahiae* in Waquoit Bay (Peckol et al., 1994). Pedersen and Borum (1997) found that several opportunistic macroalgae, including *Cladophora* and *Ulva*, were able to exploit pulses of high concentrations of ammonium by taking up ammonium at enhanced rates. Laboratory N uptake studies with *Gracilaria vermiculophylla* found a preference for ammonia as a N sources (Abreu, Pereira, Yarish, Buschmann, & Sousa-Pinto, 2011) as well. Because N concentrations can fluctuate with the tide and opportunistic macroalgae can incorporate nutrients very quickly, it is at least theoretically possible that tidal phase could have some effect on macroalgal tissue N status.

After the quadrat settled on the bottom free diving equipment and a fine mesh net were used to collect the macroalgae within the quadrat. All quadrat samples had 100% coverage and most samples had 5-15 cm macroalgal mats typically composed of *Gracilaria* and *Ulva* (Figure 11). Macroalgal samples were then transferred to bins on the boat and samples were then sorted to remove any macroinvertebrates or debris from the macroalgae sample. Samples were then spun in a salad spinner to remove excess water and placed in labeled plastic sample bags to measure the ww biomass for each quadrat.

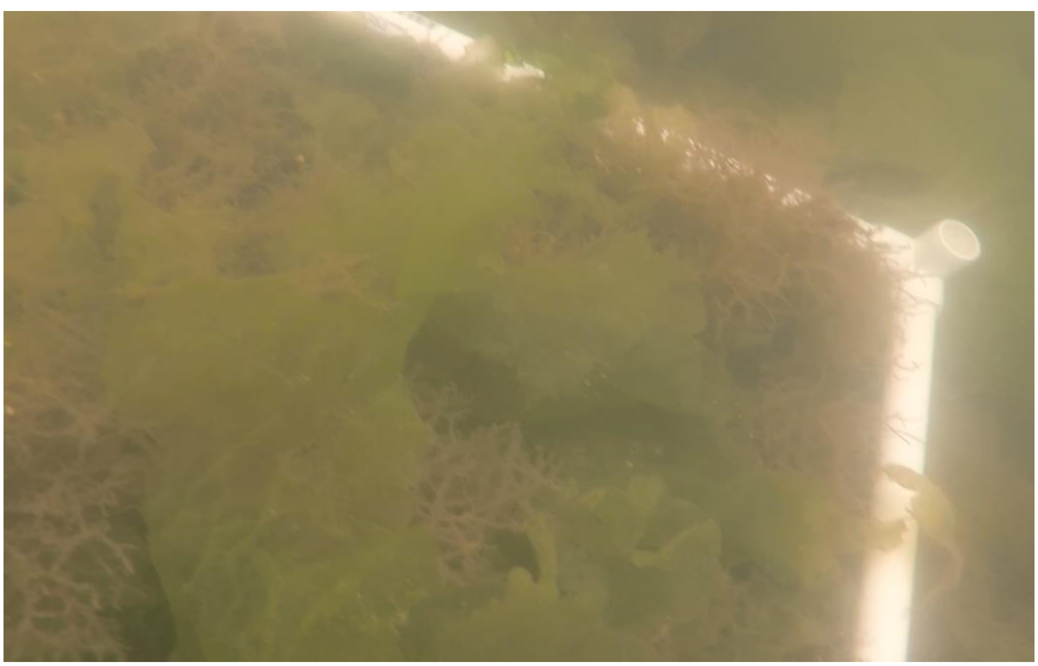


Figure 11. Close-up of one corner of a typical 0.25 square meter quadrat.

Kim, Kraemer and Yarish (2014) found that the salad spinner method is effective at removing excess water from the macroalgae. Samples were then placed on ice in a cooler for transport to the laboratory.

Macroalgal data collection began in May and continued into July 2019 on a roughly biweekly basis in an effort to capture data showing biomass and N trends during the rapid spring growth phase. Data collection focused on macroalgal biomass, water N concentration and macroalgal N concentration. The main objective of this data time series was to determine if an optimal harvesting time window could be identified. Theoretically, the optimal window for macroalgal harvesting would be when both N tissue concentrations and biomass are high. Identifying an optimal harvest window should improve the N yield of the harvested macroalgae and improve the effectiveness of this N abatement tool.

Macroalgal harvesting took place on July 15, 2019. This was later in the season than the original target date of late June. However, conditions for harvesting were good because bottom D.O. concentrations were still healthy at 5 mg/L. In addition, the condition of the macroalgae on July 15 showed no notable black anoxic sediment conditions in the shallow regions, suggesting that a major die off or collapse had not yet occurred.

Conditions deteriorated in the region shortly after the harvest date as the Falmouth Shellfish Constable observed a hypoxic event in the main channel of Waquoit Bay on July 19th with a D.O. reading of 0.94 mg/L at 9:30 a.m. (personal communication, C. Martinsen, 7/19/19). This reading was taken four hours after sunrise (i.e., 5:24 a.m.), which suggests D.O. concentrations were even lower during the night.

Macroalgae embedded in sediment were avoided during data collection and harvesting to reduce bycatch and prevent damaging the pump. Avoiding the macroalgae embedded in sediment during data collection and harvesting resulted in leaving a portion (~0-20%) of the macroalgae in situ. Sediments observed during data collection were consistently soft, fine and apparently highly organic. Virtually all of the samples had embedded macroalgal biomass, with the exception of the few samples collected in very shallow water. The genus observed intermixed with the sediment was predominantly *Gracilaria sp*. In addition, many of the samples had numerous snails apparently feeding on the benthic detritus at the sediment surface (Figure 12).

Figure 12. Snails apparently feeding on detritus at sediment surface.

Macroalgal Suction Harvesting

Macroalgal suction harvesting employed methods similar to those used by Mozuku (*Cladosiphon okamuranus*) farmers in Okinawa Prefecture Japan (Jacquemin, 2018). Mozuku farmers typically use a four to six horsepower gas-powered water or semi-trash pumps for suction harvesting (H.Tome, personal communication, February 19, 2019). These pumps are attached to long flexible tubing sometimes up to 100 feet in length. Mozuku farmers typically use SCUBA gear to assist in suction harvesting Mozuku in clear, shallow tropical waters.

For this study, suction harvesting used a small, shallow draft boat as a platform with a semi-trash pump and three field personnel. One person suctioned up macroalgae with the suction tubing in the water while the other two people operated the pump and collected and bagged the harvested macroalgae on the boat.

The four stroke 165 cm^3 engine is a semi-trash pump capable of generating 4.8 horsepower (3.6 kW) at 3,600 rpms and is capable of tolerating solids up to 5/8-inch in diameter (Figure 13). This pump is capable of 42 pounds per square inch (PSI) pressure, has 23 feet of potential suction lift and can pump 15,850 gallons per hour. The suction field around the mouth of the suction hose was approximately 10 cm during harvesting. While this pump may provide more suction than is necessary for macroalgal suction harvest applications, it is among the smaller pumps available on the market that will tolerate solid materials.

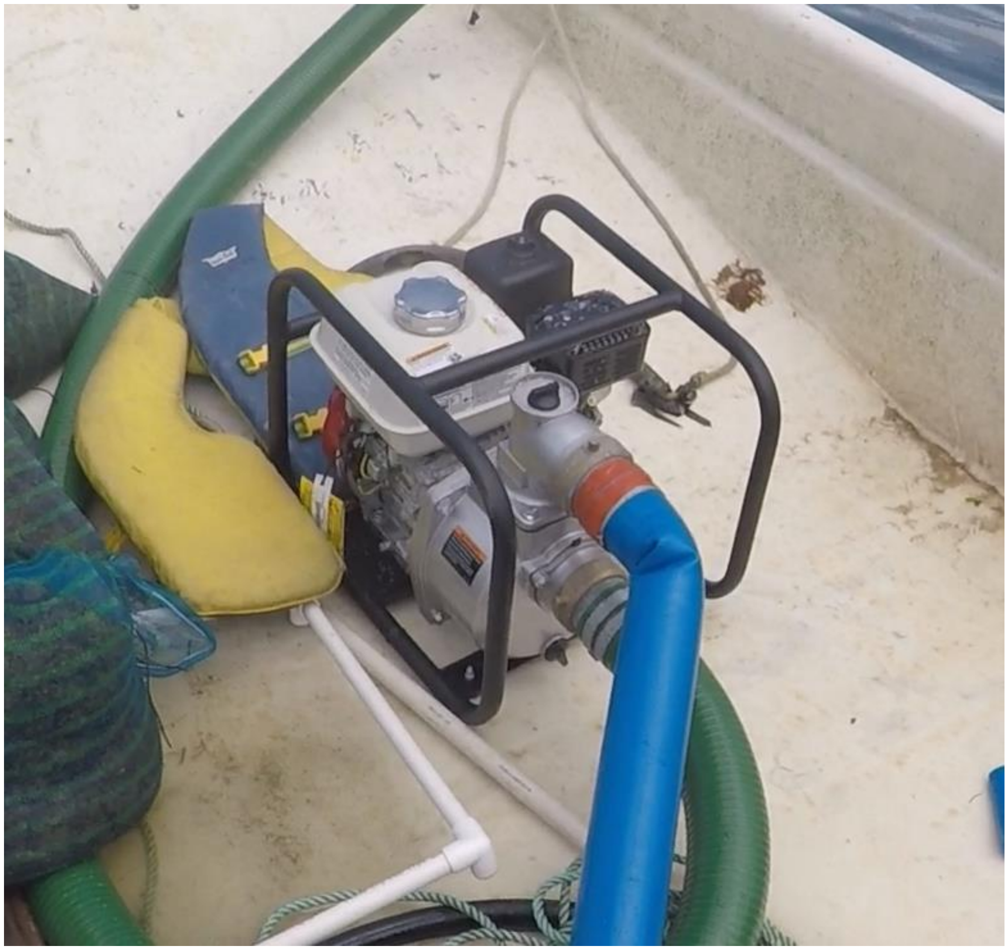


Figure 13. Honda GX-160 3-inch semi-trash suction pump used to harvest macroalgae

Modifications were made to the pump fittings and hoses for collecting macroalgae in situ. The standard pump fitting configurations were three inch National Pipe Thread (NPT) tubing. To reduce the size of the hosing used to collect macroalgae near the bottom and make the tubing more manageable for the working in the water, a reducing couple was used to constrict the intake port from three inch hosing to two inch hosing. This allowed for the use of a two inch suction hose port at the front of the pump line. This made the hose easier to manipulate in the water and easier to harvest macroalgae.

Approximately 20 feet of three inch suction tubing was attached to the pump intake. A reducing couple was used to reduce the tubing down to two inch diameter and approximately 50 feet of two inch PVC clear discharge hosing. This hosing has a 125 psi rating so that suction force from the pump would not collapse the tubing. The pump exit port was fitted with a 3-inch NPT discharge hose that was approximately 12' long, which was just enough to run the tubing from the pump to over the rail of the boat.

Harvesting took place after collection of surface water triplicates and five macroalgal quadrat samples. Macroalgal biomass data was also collected on the harvest date to tie into the previous data collected. After the surface water and biomass data were collected, the pump was set up and fitted with the tubing.

Hosing was connected to the pump and a free diver moved around in the water to locate areas of heavy macroalgae biomass for harvesting. The suction end of the tubing was kept approximately 10-20 cm off the bottom and macroalgae were swept towards the suction end of the hose. A float was tied to the end of the suction tubing to prevent the hose from hitting the bottom. Selective harvesting was done by holding the hose above the bottom with one hand while bringing seaweed from the bottom or near bottom up to the hose with the other hand. The disturbance and movement with the non-suction hand on the bottom caused most fish and invertebrates to move away from the area and appeared to reduce the number of fish and invertebrates that were inadvertently sucked up with the macroalgae. While this appeared somewhat successful at encouraging organisms to move away from the suction, it did not eliminate all bycatch. Once the macroalgae is in the hose it was pumped to the boat. The macroalgae then travels through the pump impeller housing and exits the three inch exhaust port.

The discharge for the pump was placed over the side of the boat into mesh bags to catch the algae, allowing the water to drain, but capturing the macroalgae. Mesh bags used for harvesting were approximately one mm mesh polyethylene aquaculture bags (Figure 14). The bags held up well to the strain of the macroalgae biomass. However, it did appear that a portion of the macroalgae may have been passing through the mesh pore size due to grinding as the macroalgae passed through the pump impeller. This would be especially likely for the more delicate macroalgal species, such as *Cladophora vagabunda.*

Figure 14. One mm mesh bags used to collect harvested macroalgae.

Triplicate surface water samples were collected from the mid-water column immediately prior to biomass data collection and analyzed for total N and total phosphorus. After harvesting, the macroalgae were allowed to drain and air dry for at least one hour to remove excess water similar to Conover (1958). The harvested macroalgae were then weighed to attain total ww using a 50-pound portable digital scale sensitive to +/- two ounces (56 grams). Suction harvest operations were timed during the actual in water time to determine the ww mass that can be removed per hour. Macroalgae and surface water samples were put on ice and delivered to the laboratory at the Center for Coastal Studies (CCS) in Provincetown, Massachusetts. The Macroalgae were sub-sampled and dried for percent dw, total N and total phosphorus concentrations. The percent dw was applied across the total wet biomass of macroalgae to estimate total harvested dw, which was then used to derive the total N harvested by multiplying macroalgal dw by percent N.

Three small sub-samples were collected from each macroalgae biomass quadrat sample to form a composite sample for analysis. For the harvested macroalgae, a grab sample was collected from each of the six harvested bags of macroalgae. The six grab samples were placed in a bag, thoroughly mixed and three composite sub-samples were collected from the six-bag composite.

Analytical Methods

For the macroalgal biomass, 10-12 grams of ww macroalgae were weighed and then dried in a 60 degree Celsius oven for at least 48 hours to remove moisture. The samples were then weighed again to determine the percent dw. After drying, macroalgal samples were homogenized using a mortar and pestle, encapsulated in a pre-weighed tin capsule and weighed again to determine the weight of the sample before combustion. Dried samples were then analyzed for total N and total phosphorus using a Thermo

Scientific Flash 2000 organic elemental analyzer. Elemental analyzers have been used for macroalgal tissue total N concentrations in a number of other studies (Rivers & Peckol, 1995; Thybo-Christensen, 1993). Dr. Amy Costa and her staff at CCS conducted all laboratory and analytical procedures.

Triplicate surface water samples were collected from the Childs River location from the mid-water column. Brown polyethylene 120 ml bottles were rinsed three times in the water prior to sample collection and filled and capped underwater, removing air bubbles from the sample. Samples were put on ice and then delivered to CCS. At the CCS surface water samples underwent a digestion process using a modified version of the USGS persulfate digestion method (USGS, 2003). This method used an alkaline persulfate digestion, which oxidizes all forms of N to nitrate and hydrolyzes all forms of phosphorous to ortho-phosphate. After digestion, samples were analyzed with an Astoria 2 autoanalzyer.

Macroalgal Harvesting Costs

The cost for removing N from a eutrophic estuary using suction macroalgal harvesting was estimated for a range of different harvesting scenarios, and compared to other methods. To estimate the cost on a per kilogram of N removed basis, material costs such as pumps, bags, couplings and tubing were tallied. For this suction harvesting operation, costs came to $1,055 (Table 4). The total capital costs did not include the costs of owning and operating a small boat, which would be needed in most instances. The assumption is that most communities with estuaries are likely to own a small vessel to monitor and regulate the resource.

Table 4. Capital costs of macroalgal harvesting.

Item	Cost
Pump: 160cc self-priming semi-trash water pump	$502.00
Suction tubing: 50 feet of 2" dia. clear 125 psi polypropylene hose	$158.31
15 feet 3" suction polypropylene hose	$97.50
20 feet 3" discharge polypropylene hose	$92.49
Hose coupling: Banjo coupler, 2", 125 psi, polypropylene	$14.92
Hose coupling: Banjo coupler, 3", 75 psi, polypropylene	$14.75
Banjo reducing coupling, 3" to 2", 125 psi, polypropylene	$37.25
Harvest bags: 25 polypropylene 0.75 mm mesh oyster spat bags *	$100.00
Funnel to safely pour gas into pump on boat	$6.25
Fish scale to weigh harvested macroalgae: Berkeley digital	$31.86
Total capital costs	**$1,055.33**

Note. * https://ketchamsupply.com/products/spat-bags

Because most of the time involved with this work focused on the collection of macroalgal data and working through the logistics of suction harvesting, only one proper test run of the suction harvesting method was completed. For cost estimate purposes the time and analytical costs of the pre-harvesting biomass data collection were not used in the macroalgal harvesting cost calculation.

The amount of data required for the purposes of N removal were significantly less than what is presented here. The minimum information needed for macroalgal harvest N removal would be the wet weight (ww) of the harvested macroalgae, the percent dry weight (dw), and percent N concentration of a macroalgae sub-sample of the harvested macroalgal. Macroalgal sub-sampling should consist of several replicates or a composite sample to provide a reasonably reliable estimate of the mean percent N and percent dw. If precision of the mean for these parameters are of great concern, incremental sampling should be considered. Incremental sampling is designed to provide an unbiased, statistically valid estimate of the mean value of an analyte (ITRC, 2012).

For purposes of the harvesting cost estimate, the analytical costs assumed that samples cost $10 per sample and that three samples will be submitted for each round of harvesting for a total of $30 to derive a mean macroalgal tissue N concentration and mean percent dw. This information was theused to calculate the total N harvested with the use of the ww data.

Present Value Analysis of Harvesting Scenarios

A present value (PV) analysis was conducted to estimate the cost of suction harvesting based primarily on information provided in the Cape Cod Commission's (CCC) 2019 Cape Cod Area Wide Water Quality Management Plan update (CCC, 2019). The PV analysis assumed that the suction pump used for macroalgal harvesting will last for four years before requiring replacement or five times over the 20-year planning period used by the CCC. The pump used for the suction harvesting has a three year limited warranty. The assumption is that the pump can operate one year past the warranty before needing to be replaced.

Costs incorporated into the PV analysis included the capital costs of the pump equipment, labor costs, the harvest yield, percent N and percent dw to derive a range of estimates of the cost per kg N removed via suction harvesting. Static costs for conventional N removal methods compared to macroalgal suction harvesting were summarized in Table 3.

Each harvesting scenario assumed that one day of harvesting would consist of four hours of suctioning. Four hours was chosen because even when wearing a wetsuit or drysuit exposure to waters in the 60 degrees Fahrenheit range for four hours can be difficult and exhausting. In addition, the 3.3-quart gas tank on the pump has a run time of approximately two hours, according to the manufacturer. So, this would mean for a four-hour session, there would need to be at least one stop to refuel the pump. For each scenario five different intensities of harvesting effort are projected. For example, a minimal intensity effort would consist of one day of harvesting for four hours. The most intense level of effort estimated projects results based on 80 hours of suction harvesting per year, or 20 outings at four hours per outing. While 80 hours per year is the maximum presented in Table 6, the total amount of harvest potential will be site or community specific and it is conceivable that more than 20 outings per year could be productive.

Labor costs assumed that one town employee or staff would conduct the suction harvesting with the assistance of two volunteers. Labor costs were assumed to be $25/hour for one worker. While this may be on the low end of the cost range for communities that don't have ample volunteers to assist in harvesting, it should be viewed in light of the fact that labor costs for some N abatement approaches are either not incorporated into the price per kg N removed or the contribution of labor costs are not transparent, making direct comparisons impossible. It should also be noted that the percent dw and percent N values are not static and will vary widely depending on location and season. While percent dw may go down, percent N may go up and vice versa.

Chapter III

Results

Macroalgae biomass from the Childs River site was dominated by *Gracilaria sp*. constituting roughly two-thirds of the biomass across sample dates and quadrats. There are two species of *Gracilaria* in the area, the native *Gracilaria tikvahiae* and the invasive *Gracilaria vermiculophylla*. The two species are not discernable to the naked eye and genetic testing is required to speciate them. Due to the difficulty in determining between *Gracilaria* species, the species of macroalgae collected for this study were not determined.

Approximate relative proportions of composition by genera were reported for each sample. After *Gracilaria*, the next most common genus present was *Ulva*. Based on field observations it appears that *Ulva lactuca* is the predominant species of *Ulva* in the area. The only other species of macroalgae observed or collected in the Childs River was *Cladophora vagabunda*, which was occasionally intermixed with other species but never comprised more than 25% of any quadrat sample. Every quadrat sample had 100% macroalgal bottom cover. Macroalgal mat depth was not measured, but was typically five to 10 cm deep, and occasionally deeper.

Macroalgal Biomass

Macroalgal biomass showed considerable variability and ranged from 748 g/m^2 ww to a maximum of 2,155 g/m^2 with an average biomass of 1,407 g/m^2 across all dates and samples. Macroalgal biomass data is presented in Appendix 3. A boxplot summary of the macroalgal biomass data is presented in Figure 15.

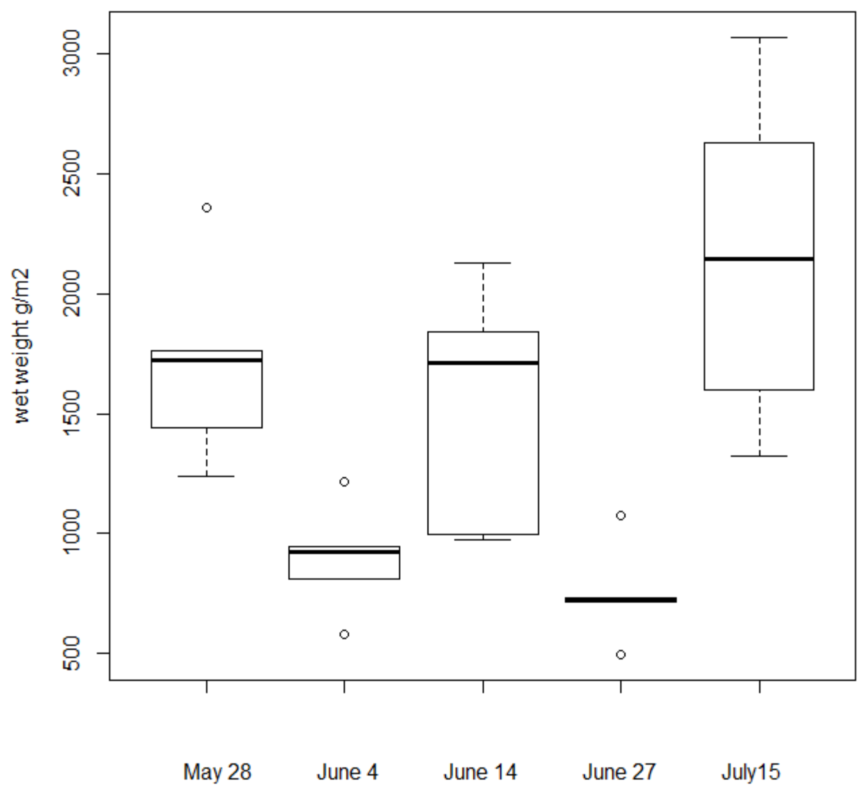


Figure 15. Boxplot of macroalgal biomass data from the Childs River. Boxplot reports minimum, 25th percentile, median, 75th percentile and maximum values.

Macroalgal Percent Dry Weight and Nitrogen Content

Percent dry weights for macroalgae collected for this study in the Childs River had a maximum of 34% and a mean of 28% (Table 5). These values were greater than percent dry weights reported in the literature. According to Morand and Briand (1996), dw ratios typically range between 5% and 20% depending on the species and time of year, but generally is approximately 12%. Macroalgal data collected in 2016 by the Marine Biological Laboratories (MBL) at the Childs River, the Quashnet River and Sage

Lot Pond (MBL, 2016) found the average percent dw to be 13%. The 2016 MBL macroalgal data reported the average percent dw for Gracilaria at 22%.

Table 5. Mean percent dry weight and percent nitrogen of macroalgae from Childs River.

Date	Mean % dry weight	Tide status	Mean % N
28-May	13%	Ebbing	1.76
4-Jun	34%	Rising	1.91
14-Jun	29%	Rising	1.56
27-Jun	30%	Ebbing	1.96
15-Jul	32%	Peak	1.72

It is unclear why macroalgae collected from the Childs River had high percent dry weights. However, the fact that *G. vermiculophylla* is present in the area and that both in 2016 and 2019 macroalgae in the Childs River had high percent dry weights (22% and 28% respectively) leads to the question of whether *G. vermiculophylla* may have high percent dry weights compared to other macroalgal species. This could have implications for future harvesting scenarios as well as for macroalgal research. At a minimum, variable percent dry weights suggest that macroalgal studies and harvesting efforts should measure percent dw instead of relying on percent dry weights to estimate N mass removal taken from existing literature.

Macroalgal tissue percent N ranged from 1.16% to 2.51% with a mean across all sampling events of 1.78% (Figure 16). Percent N values published for macroalgae in the area are generally in this range. For example, Teichberg, Heffner, Fox and Valiela (2007) reported mean macroalgal values in the Childs River ranging from 0.7% to 2.5% N.

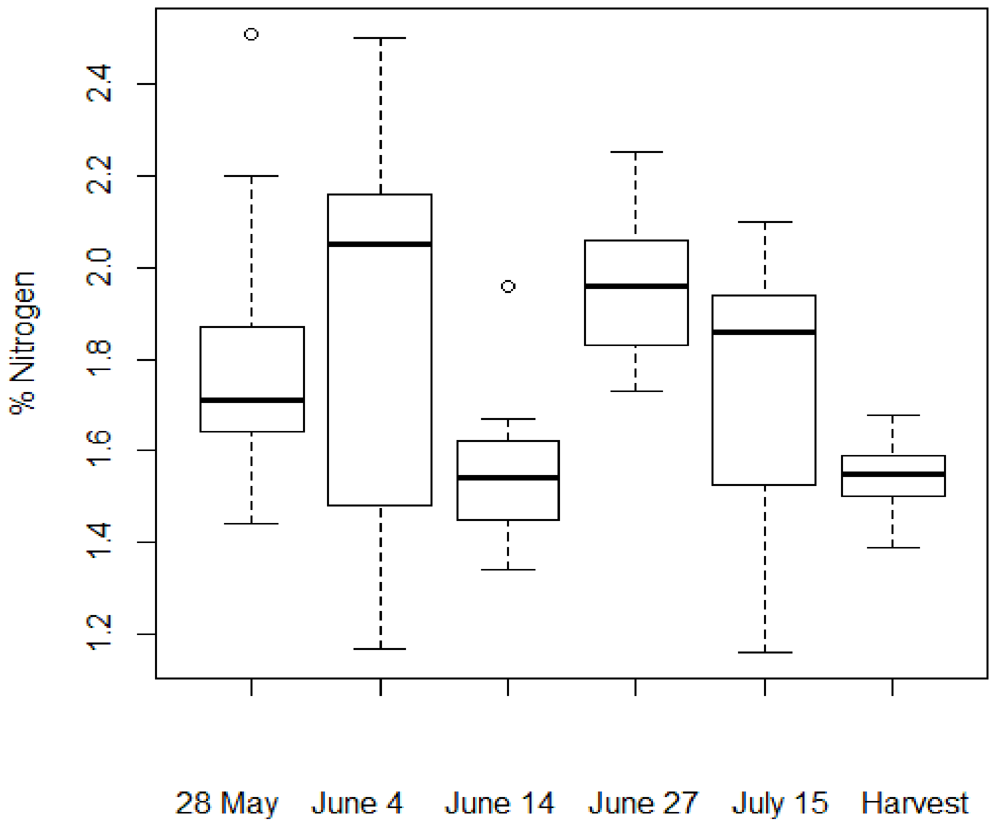


Figure 16. Boxplot of macroalgae percent nitrogen from the Childs River. The Harvest data were also collected on July 15, but represents three homogenized composite samples of all the harvested macroalgae.

Surface Water Nitrogen Data

Surface water collected for this study had total N concentrations ranging from 1.0 to 3.5 mg/L with a mean across the five sampling dates of 1.8 mg/L (Table 6). A portion of the variability of these values is likely related to the variation of the sampling period within the tidal cycle. The 2012 MEP for Waquoit Bay reported N concentrations in the Childs River ranged from 0.89 to 1.19 mg/L (Howes, et al., 2012). Results from data presented here suggests that N surface water concentrations have increased in the Childs River since the 2012 MEP report.

Table 6. Surface water nitrogen concentrations at sample location over time.

Sample ID	Collection Date	Total Nitrogen	
		(µM/L)	(mg/L)
CR-SW-1	28-May	21.3	1.3
CR-SW-2		19.4	1.2
CR-SW-3		15.6	1.0
Mean		**18.8**	**1.2**
CR-SW-1	4-Jun	17.5	1.1
CR-SW-2		17.3	1.1
CR-SW-3		18.5	1.1
Mean		**17.8**	**1.1**
CR-SW-1	14-Jun	34.8	2.2
CR-SW-2		34.0	2.1
CR-SW-3		31.1	1.9
Mean		**33.3**	**2.1**
CR-SW-1	27-Jun	25.3	1.6
CR-SW-2		28.4	1.8
CR-SW-3		26.0	1.6
Mean		**26.6**	**1.6**
CR-SW-1	15-Jul	46.8	2.9
CR-SW-2		56.2	3.5
CR-SW-3		53.9	3.3
Mean		**52.3**	**3.2**

Macroalgal Suction Harvest Data

During the harvest period (34 minutes) a total of six mesh bags were filled with macroalgae that had a total weight of 24.6 kg (Table 7). The rate of harvesting worked out to 43.5 kg/hr. Limited visibility makes it difficult to avoid the bottom and organisms that might be in the area. Partly due to this issue, suction harvesting lasted only 34 minutes when a green crab (*Carcinus maenas*) was accidentally sucked into the hosing. While the crab was freed, it shortened the harvest period. The intent was to harvest for at least one hour. This brings into question the feasibility of longer harvesting operations.

Table 7. Harvested macroalgae weights.

	weight 1	weight 2	ave. pounds	ave. kg
Bag 1	12# 10 oz	12# 12 oz	12# 11 oz	5.75
Bag 2	5# 5 oz	5# 7 oz	5# 6 oz	2.44
Bag 3	7# 6 oz	7# 6oz	7# 6oz	3.35
Bag 4	12# 7oz	12# 8 oz	12# 7.5 oz	5.65
Bag 5	9# 13 oz	9# 13 oz	9# 13 oz	4.45
Bag 6	6# 9 oz	6# 14 oz	6# 11.5 oz	3.05
Total	54# 2oz	54# 12 oz	54# 7 oz	24.69

Macroalgal Suction Harvesting Bycatch

Based on observations in the Childs River during data collection and macroalgal harvesting, it was apparent that a number of macrofauna reside in and among the macroalgae. Similar to Deegan et al. (2002), the Grass Shrimp (*Paleomonetes pugio*) was the most commonly observed macrofauna during all sampling events. No measurements of macrofauna bycatch were collected for this work to quantify bycatch. This information would be useful information to collect in future work.

Other macrofauna observed in the macroalgal quadrat samples included mud snails (*Tritia obsoleta*), one juvenile eel (*Anguilla rostrata*), several hairy sea cucumbers (*Sclerodactyla briareus*) and one 3-spined stickleback (*Gasterosteus aculeatus*). Comb jellies (*Mnemiopsis leidyi*) and green crabs (*Carcinus maenas*) were also observed in the area. Efforts were made to minimize bycatch during suction harvesting.

Statistical Analyses

Statistical analyses were performed with R version 3.6.1 statistical software (R Foundation for Statistical Computing, Palo Alto, CA, USA). Regression analysis was conducted in an attempt to determine if macroalgal biomass is related to water N concentrations in the hopes that water N concentrations could be used as a proxy to select optimal macroalgal harvest dates. Macroalgae tissue ww (g/m^2) was not significantly correlated with surface water total N concentrations (uM/L), with an adjusted R-squared value of 0.278 and a p-value of 0.209. With such a small sample size in the context of a system with inherently high variability, it is not surprising that a strong correlation was not observed. However, macroalgae tissue dw (g/m^2) was significantly correlated with surface water total N (uM/L); the adjusted R-squared value was 0.825, with a p-value of 0.021. Regression plots, residuals and R syntax are available in Appendix 4.

For the purposes of future macroalgal harvesting efforts, the statistical analysis suggests that surface water total N concentrations correlate with macroalgal biomass dw, which is important because the dw biomass is used to derive the total N harvested from a system. However, this conclusion should be tempered with the realization that the sample size for this analysis is small. Additional supporting data would help to better define the confidence in this conclusion.

Regression residuals were reviewed to evaluate the validity of the regression assumptions. Small sample size makes it difficult to assess the residuals. The residuals versus fitted values for the ww versus water N concentration regression show the data to be scattered for three data points at lower water N concentrations. The heteroscedasticity for these values brings into question the suitability of the regression model and suggests that the relationship between these two variables may not be linear.

The Normal Q-Q residual values are close to the 45 degree line suggesting the data is normal or near normally distributed. The Scale-Location residuals have too few points to discern any pattern. The Residuals versus Leverage plot show sample one and five to be at the Cook's Distance, which suggests they are possible outliers and may be strongly influencing the linear regression.

Residuals for the dw versus water N concentration regression show the data to have a bit less scatter at lower water N concentrations. The fact that there is less heteroscedasticity for these residuals suggests that the relationship between these two variables appears to be somewhat linear and raises the confidence in the suitability of the regression model for this comparison.

The Normal Q-Q residual values are close to the 45 degree line, but show more deviation than the Q-Q residual for the ww regression, suggesting that the data may not be normally distributed. Similar to the ww regression, the Scale-Location residuals have too few points to discern any pattern. The Residuals versus Leverage plot shows samples two and five are right at the Cook's Distance, which suggests they are possible outliers and may be strongly influencing the linear regression.

Analysis of Macroalgal Harvesting Scenarios

Table 8 provides several potential scenarios for the costs to implement macroalgal suction harvesting for removing N from a eutrophic estuary. Costs per kg N were estimated to range between $62/kg to as much as $3,271/kg, depending on a few variables. The data and information collected in this study suggest that macroalgal harvesting may provide a suitable intervention for some communities. While the costs of macroalgal harvesting are on the higher end of the range of interventions on a dollar per kg of N removed basis, this approach has a low point of entry cost of roughly $1000,

Table 8. Macroalgal suction harvesting estimated cost per kg scenarios.

Worst Case	Labor costs	Analytical	Fuel costs	Operating cost	Yield (kg/hr.)	Harvest (kg)	kg N	cost/kg N
4 hrs./yr.	$100	$30	$10	$1,195	43.5	174	0.4	$3,271
8 hrs./yr.	$200	$60	$20	$1,335	43.5	348	0.7	$1,827
20 hrs./yr.	$500	$150	$51	$1,756	43.5	870	1.8	$961
40 hrs./yr.	$1,000	$300	$102	$2,457	43.5	1740	3.7	$672
80 hrs./yr.	$2,000	$600	$203	$3,858	43.5	3480	7.3	$528
Median	Labor costs	Analytical	Fuel costs	Operating cost	Yield (kg/hr.)	Harvest (kg)	kg N	cost/kg N
4 hrs./yr.	$100	$30	$10	$1,195	43.5	174	1.2	$1,006
8 hrs./yr.	$200	$60	$20	$1,335	43.5	348	2.4	$562
20 hrs./yr.	$500	$150	$51	$1,756	43.5	870	5.9	$296
40 hrs./yr.	$1,000	$300	$102	$2,457	43.5	1740	11.9	$207
80 hrs./yr.	$2,000	$600	$203	$3,858	43.5	3480	23.8	$162
Best Case	Labor costs	Analytical	Fuel costs	Operating cost	Yield (kg/hr.)	Harvest (kg)	kg N	cost/kg N
4 hrs./yr.	$100	$30	$10	$1,195	50.0	200	1.5	$802
8 hrs./yr.	$200	$60	$20	$1,335	50.0	400	3.0	$448
20 hrs./yr.	$500	$150	$51	$1,756	50.0	1000	7.4	$236
40 hrs./yr.	$1,000	$300	$102	$2,457	50.0	2000	14.9	$165
80 hrs./yr.	$2,000	$600	$203	$3,858	50.0	4000	29.8	$129
All Volunteer	Labor costs	Analytical	Fuel costs	Operating cost	Yield (kg/hr.)	Harvest (kg)	kg N	cost/kg N
4 hrs./yr.	$0	$30	$10	$1,095	50.0	200	1.5	$735
8 hrs./yr.	$0	$60	$20	$1,135	50.0	400	3.0	$381
20 hrs./yr.	$0	$150	$51	$1,256	50.0	1000	7.4	$169
40 hrs./yr.	$0	$300	$102	$1,457	50.0	2000	14.9	$98
80 hrs./yr.	$0	$600	$203	$1,858	50.0	4000	29.8	$62

Note. Worst-case scenario assumes 1.75% N and 12% dry weight.
1.75% N represents the geometric mean of data collected for this study.
12% dry weight is based on a published average (0.12) (Morand, 1996).

Median-case scenario assumes 1.75% N and 39% dry weight.
39% dry weight based on macroalgae harvested on July 15, 2019 (0.39).
Best-case assumes 1.91% N, 39% dry weight and harvest improvement to 50 kg/hr.
1.91% N represents the highest mean across all sampling datas for this study.
All volunteer scenario = Best-case and no labor costs.
Analytical costs assume 3 replicates for every 4 hours of harvesting @$10/sample.

assuming towns already operate a boat, compared to some interventions that require millions of dollars of investment. While this approach is unlikely to be used as the primary N reduction approach for a eutrophic estuary, it may help some communities reach their TMDL N goals.

If the mean annual biomass of the standing stock of seaweed was harvested from Waquoit Bay, 15-66 kg of N could be removed per hectare per year (Kim et al., 2014). To put this into perspective, one simulation presented in Table 8 would require 80 hours of suction harvesting to remove 29.8 kg of N using best-case assumptions. So, this study shows that Kim's estimates are attainable using suction harvesting methods.

Cost estimates included several assumptions including the amount of fuel used to operate the pump and a boat, some labor costs, as well as fixed costs such as the pump and associated tubing and couplings (Table 8). For most of the cost estimate scenarios the measured macroalgal harvest yield of 43.5 kg/hr. via suction harvesting were used. However, for the best-case scenario and for an all-volunteer labor scenario, an improved yield estimate of 50 kg/hr. was used. The two parameters with the greatest influence on the cost/kg N removal were labor costs and the harvest yield. Based on limited testing there appears to be potential to improve on the 43.5 kg macroalgae harvested per hour of suctioning. Suction harvesting field observations suggest that improved yields could be obtained with the use of smaller pore sized mesh bags to collect and secure suctioned macroalgae. Improvements in this area could potentially double the harvest yield. A more modest estimate of harvest improvement to 50 kg/hr was used in Table 6 to represent best-case scenario estimates.

The present value (PV) cost comparison of conventional N abatement methods to suction harvesting is presented in Table 9. The PV analysis puts macroalgal suction harvesting costs roughly in the middle of the range ($40.18) for costs to abate N loading on a present value kg per dollar basis. The harvest scenario used for this comparative PV analysis is the median estimate of potential macroalgal harvest for 40 hours of harvesting per year at a cost of $2,457 as presented in Table 8.

Table 9. Present value nitrogen abatement cost comparison.

Abatement method	Project cost	Useful life	nper	Present Value (PV)	Kg N removed	PV cost/kg N removed
Fertilizer management	$105	20	1	$101	43	$2.35
Dredging	$231	25	1.25	$274	60	$4.57
Aquaculture- shellfish	$63,000	20	1	$60,001	4990	$12.02
Fertigation wells - Turf	$2,310	20	1	$2,201	85	$25.89
Conventional WW treatment	$28,048,800	10	0.5	$13,519,482	448761	$30.13
Macroalgal suction harvesting	$2,457	4	0.2	$478	11.9	$40.18
Constructed wetlands	$714,000	20	1	$680,001	8803	$77.25
PRB*	$1,092	20	1	$1,041	12	$86.75
Composting toilets	$14,000	20	1	$13,334	72	$185.20
Stormwater BMPs	$107,800	20	1	$102,668	299	$343.37

Note. Inputs for this comparison based on values in the Cape Cod Comm. Tech Matrix.
https://www.capecodcommission.org/our-work/technologies-matrix/
nper= the useful life of the technology divided by the plan period.
Plan period for this analysis is 20 years.
Harvesting inputs based on the central tendency (40 hrs. of harvesting in 10 days)
Project costs = construction costs + 40%.
Interest rate assumed for this comparison is 5% (0.05).
*PRB- Permeable Reactive Barrier, 30 foot trench method.

Chapter IV

Discussion

The objective of this work was to determine if harvesting macroalgae is a cost-competitive method for removing nitrogen (N) from eutrophic estuaries compared to other N abatement methods. A macroalgal suction harvesting pilot study was conducted to determine costs and potential N yields. Macroalgal biomass and N surface water data were collected with the hope of identifying an optimal macroalgal biomass harvest window. Data collected during this study showed that suction macroalgal harvesting is a potential tool for removing excess N from eutrophic estuaries. However, logistical and equipment issues need refinement if this method is to have broad application.

Macroalgal Harvest

Macroalgal harvest yields were below the projected biomass. Suction harvesting was estimated to yield 300 kg macroalgal ww per hour of operation. The actual harvest yield was 43 kg ww/hour. The cost range for macroalgal harvesting ($62 - $3,271/kg N removed; Table 8) reflects the complexity of macroalgal system and some of the unknowns of the application of the method. Costs could be reduced if all the work were to be conducted by volunteers or if there is a major improvement in harvest yields.

The amount of macroalgae harvested suggests there is either a major flaw in the harvest estimate or an issue with the method of collection, perhaps both. Visual observations during harvesting indicated that a portion of the suctioned macroalgae was passing through the mesh netting. These observations suggest that the mesh bags used to capture the suctioned macroalgae had too large a pore size and allowed a portion of fine macroalgae to pass through the mesh. This issue could be resolved by using a finer mesh to capture the smaller macroalgal fractions. Several different mesh size trials may be necessary to discern the best mesh size for effectively capturing all the harvested macroalgae. Deegan et al. (2002) found that a mesh of 500 micrometer was sufficient for collecting *Cladophora vagabunda*, but their study used more delicate collection methods compared to a semi-trash suction pump.

One of the difficulties of the suction harvest method was that when the bottom gets disturbed after a few minutes of operation, the visibility drops to less than one foot. This is related to the fine soft sediments at the pilot study location. One approach that can help with this issue is to harvest when the tide is not slack. A gentle tidal current can help to remove some of the particulates that get suspended during bottom harvesting, which can help with visibility.

Based on the methods used in this study, the range of potential N removal via suction harvesting for eutrophic estuaries is expected to range from 5 to 30 kg N/year depending on the effort and macroalgae conditions. This is only a fraction of the N load for some eutrophic estuaries. For example, the Childs River receives over 600 kg N per hectare per year (Hauxwell et al., 1998). However, it is worth noting that macroalgal harvesting may also help improve some estuarine resources on a small scale by reducing anoxic stress and death in benthic habitats.

Metrics Used to Identify Optimal Harvest Period

Researchers have shown that opportunistic macroalgae species in eutrophic areas tend to have a period of rapid growth in the spring with a biomass peak in late spring or early summer followed by a decline or collapse in the summer. Timing of macroalgal harvesting should occur in spring and potentially in fall to avoid system collapse and lower biomass that often occurs in the summer (Figure 17).

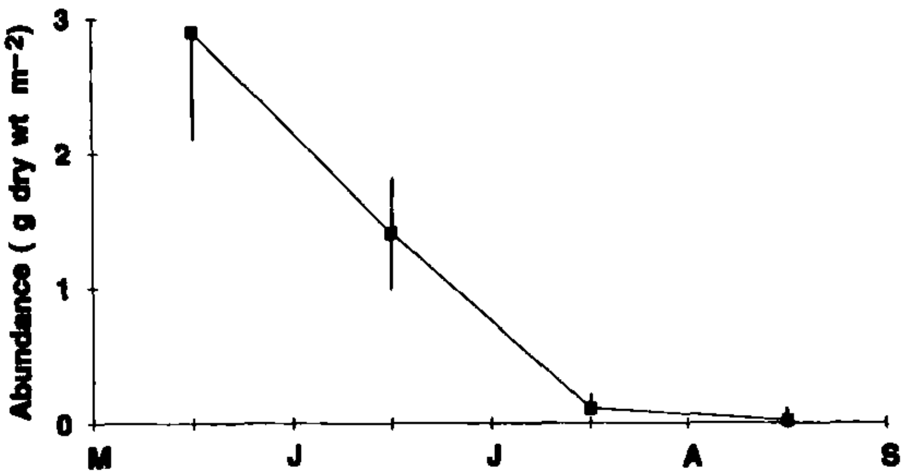


Figure 17. Mean g dry wt/m2 of *Ulva lactuca* - Childs River (Rivers & Peckol, 1995b).

Factors that are believed to cause the summer macroalgal decline include shading due to a thick macroalgal mat, high water temperature and low light conditions due to cloud cover. In many instances a collapse is the result of more than one of these variables. As biomass builds the macroalgae mat tends to shade biomass on the bottom of the mat reducing the light to the bottom. The trigger for decline is often either high temperature, which reduces growth rates for some macroalgae and increases bottom respiration, or several days with low light conditions, which can turn the bottom into a hypoxic or anoxic zone due to respiration. A drop in D.O. is often followed by macroalgal decomposition, which quickly releases nutrients back into the system. Nutrient cycling in macroalgal-dominated systems can be very quick because macroalgae have little structural material and decompose rapidly (Buchsbaum, Valiela, Swain, Dzeirzeski, & Allen, 1991).

Water temperature is an important parameter to consider when deciding when to harvest opportunistic macroalgae. As the waters warm in spring the macroalgae growth increases until sometime in the summer when either macroalgal nutrient reserves become limited or water temperatures reach into the upper seventies Fahrenheit and the growth rate of some macroalgae begins to decline. Rivers and Peckol (1995a) observed a notable decline in photosynthetic efficiency of *Ulva lactuca* at 25 Celsius (77 Fahrenheit). Prolonged periods of high temperatures can be a trigger for a system collapse, especially if combined with other factors such as low light conditions.

Kim, Yarish and Pereira (2016) found that *Graciliaria vermiculophylla* has a higher maximum temperature tolerance than *Ulva lactuca* and has growth rates of 5-10% greater than *G. tikvahiea* at temperatures over 22 Celsius (72 Fahrenheit). Higher temperature tolerance may be one of the features of *G. vermiculophylla* that gives it an advantage over some of the other macroalgal species in the area. This is especially relevant considering that average temperatures are continuing to climb regionally due to climate change.

While an optimal harvest temperature is not yet discernible, it appears that by the time water temperatures in the Falmouth area reach 70 degrees Fahrenheit, the optimal harvest window may have passed because the opportunistic, bloom forming macroalgae are growing quickly and using the N reserves they may have. This rapid growth uses up surplus N and will result in lower percent N tissue concentrations in the harvested biomass. In addition, by the time temperatures reach 70 degrees Fahrenheit, the macroalgal biomass is often significant, which poses some risk of system collapse for the macroalgal mat if a few days of cloudy conditions occur.

Below 60 degrees Fahrenheit most opportunistic macroalgae have limited growth. For example, Lapointe, Rice and Lawrence (1984) found that *G. tikvahiae* did not grow below 15 Celsius (59 Fahrenheit), so the optimal water temperature harvest window for macroalgae present in the Falmouth area appears to be between 60 and 70 degrees Fahrenheit (15.5–21 Celsius).

Surface water temperature data from Falmouth indicates that temperatures are generally in the low 60s in early June and rise into the upper 60s by the end of the month (NOAA, 2019). Water temperatures in mid-September are generally in the upper 60s and decline down to 60 degrees by mid-October. In conclusion, surface water temperature data for the Falmouth area suggests that macroalgal harvesting should take place in June and again from mid-September to mid-October.

In addition to temperature considerations, macroalgal harvesting should target when biomass and N tissue concentrations are high to maximize N removal. However, macroalgal biomass and tissue N concentrations are often at odds because as biomass increases, macroalgae tends to use up the N reserves it may have to stimulate tissue growth. Thus, macroalgal harvesting should occur before N tissue concentrations decline. A decline in tissue N concentrations was observed in three sub-estuaries of Waquoit Bay between May and June by Thompson and Valiela (1999) (Figure 18), suggesting that macroalgal harvesting should occur in May or early June to avoid the drop in tissue N concentrations.

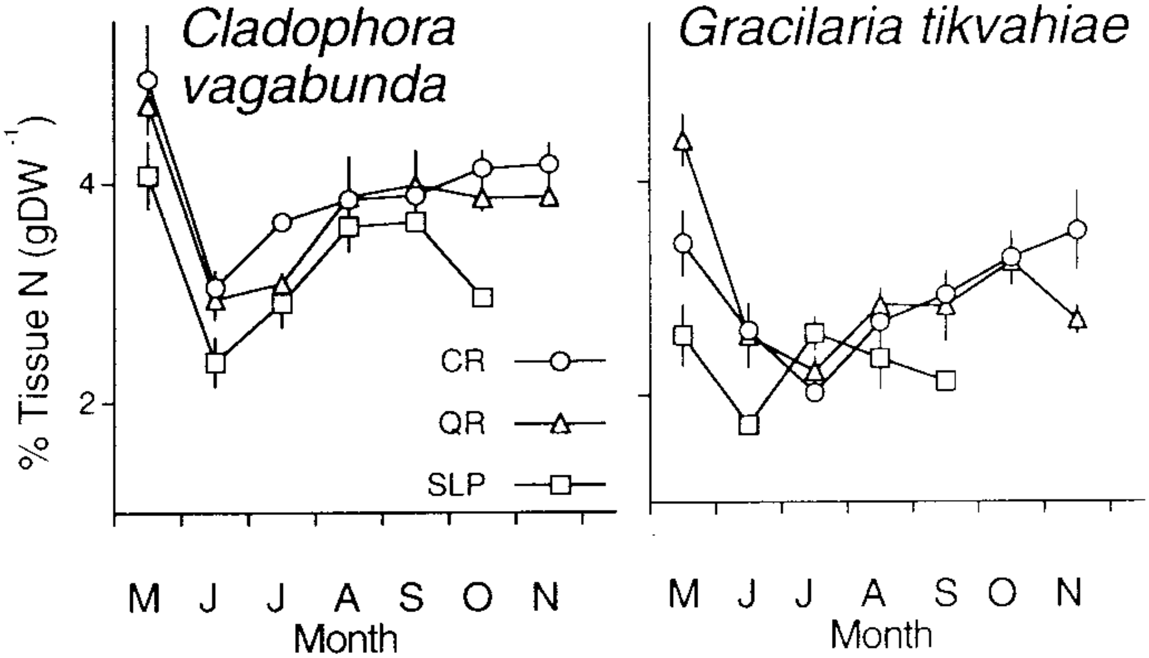


Figure 18. Seasonal macroalgal tissue % nitrogen trends. Childs River (CR- circles), Quashnet River (QR-triangles), Sage Lot Pond (SLP-squares) (Thompson & Valiela, 1999).

Factors affecting macroalgal N concentrations include the macroalgal species, the time of year and the N concentration in surface water. Duarte (1992) reported macroalgal percent N for 46 species ranged from 0.4% to 4.4% with an average of 1.9%. Rivers and Peckol (1995a) found N concentrations in *Ulva lactuca* collected from Waquoit Bay to have 2% N in spring, declining to 1% in late summer through autumn. Fujita (1985) found percent N for in the nearby Bourne's Pond in mid- July to range from 1.02% for *Ulva lactuca* to 1.59% for *Enteromorpha sp*. Nejrup and Pedersen (2010) reported that for *G. vermiculophylla*, tissue N concentrations were higher in the spring and late fall (3-

3.5%) and lower in the summer (~1.5%). The fact that *G. vermiculophylla* is likely present at the Childs River location and that macroalgal percent N averaged 1.78%, suggests that the harvest date of July 15 may have been too late in the season.

Surface water N trends can be important for macroalgal harvesting because macroalgal N tissue concentrations generally correlate with water N concentrations. Thompson and Valiela (1999) found that macroalgal biomass and percent N tissue concentrations increased as N loading increased. Gordon, Birch & McComb (1981) found that for the genus *Cladophora*, tissue N concentrations increased with water N increases up to approximately 1 mg/L N, after which tissue N concentrations plateau (Figure 19).

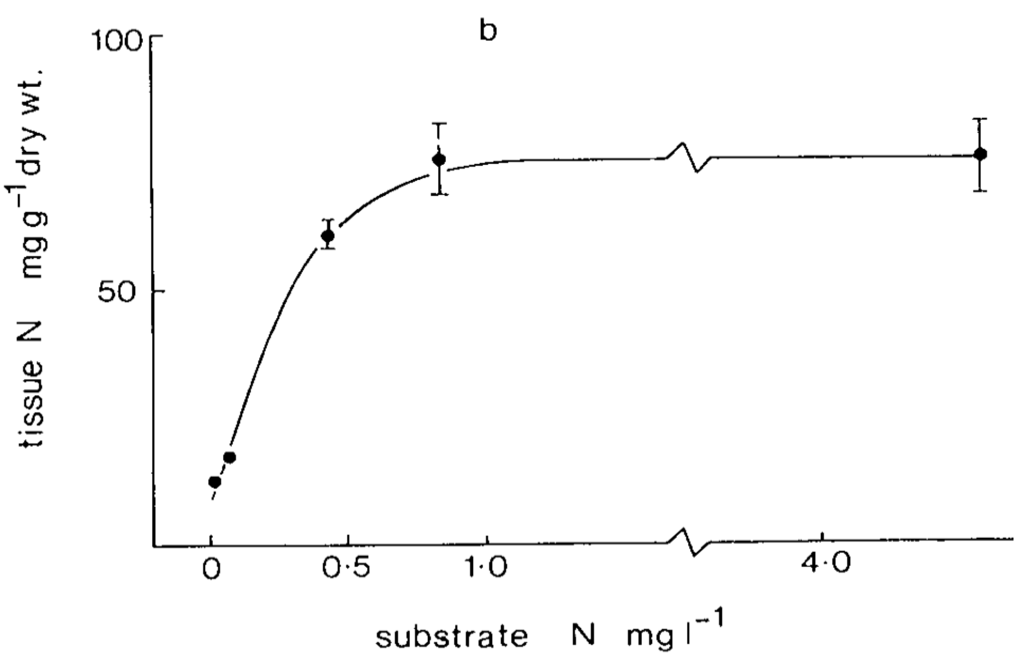


Figure 19. *Cladophora* nitrogen as a function of water nitrogen (Gordon, Birch & McComb, 1981).

The surface water data collected for this study in the Childs River suggests that for at least *Cladophora*, N surface water concentrations are not a limiting factor to macroalgal growth because the surface water N concentration was 1.0 mg/L or greater for all fifteen samples over five dates and as high as 3.5 mg/L for one sample.

Low light conditions over just a couple of days could be impacting macroalgal biomass. To discern a possible cause of the relatively low biomass on June 4 and June 27 (Figure 14), weather conditions for two days prior to each sample date were reviewed based on the theory that low light conditions could result in slow growth or even breakdown of macroalgae biomass. Interestingly, the light conditions for the two days preceding the low biomass collection dates of June 4 and June 27 were overcast. The remaining three sampling dates were preceded by at least one sunny day in the two days leading up to data collection.

While it is only a working theory, it is plausible that good light conditions two days prior to harvesting could stimulate macroalgal growth and lead to higher biomass in eutrophic estuaries. While it may not always be practical, it would appear that future macroalgal harvesting should occur after at least one day of sunny conditions and ideally two days.

Adverse Effects of Macroalgal Harvesting

While attempts were made to limit bycatch, the reality is that it is unlikely that suction harvesting can be done without some organisms being accidentally sucked up with the macroalgae. However, many organisms in the macroalgal mats will be impacted if the bottom turns anoxic and removal of macroalgae from eutrophic areas has been shown to improve benthic oxygen status (Deegan, et al., 2002).

End Use of Harvested Macroalgae

End use options considered for the harvested macroalgae included using the macroalgae as a food item, fermentation into alcohol, anaerobic digestion of macroalgal biomass into methane, composting and as a soil amendment. Other potential end uses not considered for this project include drying the macroalgae and using it as a fertilizer or plant growth stimulator and agar production.

While there appears to be a market for edible high quality *Gracilaria tikvahiae,* the quality of the macroalgae harvested for this project was not food grade. Lindell et al. (2015) had success culturing *Gracilaria tikvahiae* in the area, but the cultivation cost and the growth of epiphytes on macroalgae were constraints on its market viability.

There have been a few examples in the literature of using macroalgae biomass for alcohol production. The NYCDEP conducted a pilot project in 2010 harvesting *Ulva lactuca* (NYCDEP, 2012). Approximately 300 gallons of harvested Sea Lettuce was sent to the University of Arkansas for processing into biofuel. Researchers were able to produce one liter of butanol from this pilot study.

Massachusetts has a food waste ban, which has stimulated the development of anaerobic digestion facilities in Massachusetts. While it would be possible to incorporate harvested macroalgae into an anaerobic biomass feedstock, it is not clear that digestion operators would be willing to take occasional macroalgal biomass because anaerobic digesters tend to operate better with a consistent feed stock. There is also some concern about the salt content of macroalgae having negative effects on anaerobic metabolism. If a consistent harvesting effort were to be undertaken, it is conceivable that anaerobic digestion could handle some or all of the harvested biomass if a suitable partner relationship can be developed.

Sea Lettuce (*Ulva*) has approximately the same N content as pig manure with the added benefit of micronutrients (Crane & Ramsay, 2012). The PEI Department of Agriculture recommends application rates of Sea Lettuce at 14.5 tons/hectare, which is similar to pig manure. A field trial by Agriculture Canada (Rodd, Henry, Mills, Grimmett, & Gentile, 2011) suggested crop yields increased 60% for forage crops from Sea Lettuce application at these rates of application. Researchers in Italy reported that farmers in coastal regions had been using macroalgae such as *Ulva* and *Cladophora* as fertilizer for generations (Cuomo et al., 1995). Macroalgal biomass typically is high in N and requires the addition of some carbon source such as wood shavings or straw for proper composting.

The Town of Falmouth indicated that macroalgal biomass could be incorporated into their ongoing composting operations. There are also several small farms in the area that could use the macroalgae in their composting and organic farming operations. Pariah Dog Farm in Falmouth is a small farm that focuses on identifying and utilizing local waste streams for fuels, fodor and compostables. After discussing the macroalgae harvesting project with farmer Matt Churchill, he agreed that Pariah Dog would accept the harvested biomass and incorporate it into their composting operations and eventually their soils (personal communication 4/18/19).

Research Limitations

This study did not measure or quantify the potential ecological costs and benefits to the estuarine system as a result of the macroalgal suction harvesting. This study only makes qualitative claims about the potential benefits and impacts to the ecosystem from macroalgal removal. While these issues are important, they are beyond the scope of this investigation.

The small sample size obtained for the data in this study limits the confidence in the results. The data collected shows considerable variability, which was expected given the complex nature of ecological systems. High variability means that unless a signature is very strong, a large sample size will be needed to effectively identify trends in the data. The collection of 25 data points across five sampling dates may not represent adequate replication to adequately capture the variability of the macroalgae in these estuaries.

Several assumptions about the potential harvesting yield were incorrect. One incorrect assumption was that 100% of the macroalgae present could be suctioned off the bottom. This assumption was made before field data collection began and it became apparent that some macroalgae was imbedded in the sediment. During harvesting only a portion of the total biomass present was suctioned from the bottom to avoid harvesting snails and sediment. The decision to avoid macroalgae embedded in sediment during data collection and harvesting resulted in leaving a portion (~0-20%) of the macroalgae in situ.

Typically, quadrat macroalgal data collection would gather all of the macroalgae within the boundaries of the quadrat. Some studies use benthic grab samplers (e.g., Ponar, Ekman) to collect macroalgal biomass data and in these cases the macroalgae would be collected with the sample along with a portion of the sediment due to the method of collection. This may bias the macroalgal biomass data low compared to the actual macroalgae present. This may have some relevance when comparing biomass data for this project to other macroalgal biomass studies. However, it is unlikely to affect the objective of the study, which was to observe trends in biomass and N concentrations to determine an optimal macroalgal biomass harvest window.

There are several additional parameters that were not accounted for in this study that could impact macroalgal biomass in estuaries. One potentially important variable is the depth of the sample. Sample depth data were not collected for this work because water depth was strongly influenced by tidal phase and this study could not control for sampling period within the tidal phase. The tidal range for the Childs River is approximately 0.5 m and the majority of samples collected were from less than 1.5 m.

Another parameter that this work did not control was the effect of winds on the movements of macroalgae at the sample site. While prevailing wind direction was noted on the day of sampling, wind data can be very localized making it a difficult parameter to assess how it effects macroalgae mats in a specific location. Due to the narrow width of the Childs River at the sample location (~300'), the area was somewhat protected from easterly and westerly winds.

On the June 14 sampling event there was a southerly wind (~20 mph SSW), which affected conditions at the surface of the sample location. The wind developed small wave action and may have delayed the turn of the tide, but no discernable impacts on bottom macroalgal mat density or rafting were noted. While winds can increase D.O. concentrations of surface waters, they do not appear to play a major role on the conditions of the macroalgal mat at the Childs River location. Some studies have found that sustained winds can have an effect on estuarine flushing (Thornber, et al., 2017), which could alter macroalgal densities in some circumstances.

One unforeseen issue with the use of surface water N concentrations as a proxy for macroalgal tissue N is that the procedure for analyzing total N in surface water requires several days. So, by the time the N results are available to make a projection of macroalgal tissue N concentrations, the conditions in the field will likely have changed due to factors such as sunlight, wind, etc.

Future Research

One area of research that would be informative for the purposes of macroalgal harvesting is an empirical study of the macrofauna bycatch associated with macroalgal harvesting. An empirical bycatch study would help to frame the potential ecological costs of suction harvesting against some of the benefits.

In addition to removing N, macroalgal harvesting could provide some habitat relief to areas in an estuary where N loading and associated organic matter deposition are gradually altering the benthic habitat. Deegan et al. (2002) investigated potential benefits to estuarine habitat from macroalgal removal and found considerable benefits. It would be informative to conduct a before and after habitat status investigation relating to suction macroalgal harvesting to get a better sense of the ecological impacts and benefits from this method. A study of the effects of macroalgal harvesting on bottom D.O. concentrations could be one avenue to pursue this question.

Relatively high macroalgal percent dry weights were measured in the Childs River during this study as well as for data collected by the MBL in 2016. These data suggest that an investigation into the percent dry weights of *G.tikvahiae* as compared to the invasive *G. vermiculophylla* may be useful to allow the use of percent dry weights as a simple way to differentiate between these two species, which occupy similar habitats.

Conclusions

Elevated nutrients in estuaries can trigger seaweed (macroalgal) blooms, which can suffocate benthic organisms, kill fish and impact shellfish harvests (Valiela et al., 1992). Macroalgae biomass density ranges considerably based on nutrient loading, time of year and location, but can exceed two kg ww/m_2 in the Childs River. Removing macroalgae removes N from an estuary. Removing excess N can improve water quality, reduce anoxic stress on benthic habitats and help attain N goals put forth in water quality regulations. Removing excess macroalgae has also been shown to increase eelgrass, fish and decapod abundance (Deegan et al., 2002).

The Childs River in Falmouth, MA was chosen as the pilot study area because it has very high nutrient loading and macroalgal biomass. Macroalgal data collection began in May and continued into July on a biweekly basis in an effort to capture data showing biomass and N trends during the rapid growth phase. The main objective of this data collection was to determine if an optimal harvest time window could be identified. Theoretically, the optimal time for harvesting macroalgae would be when both macroalgal N concentrations and biomass are high.

Regression analysis of dw macroalgal biomass versus total N surface water concentrations indicated these two parameters were correlated with an adjusted R-squared value of 0.825 and a p-value of 0.021. This correlation suggests surface water N concentrations could be used as a proxy for macroalgal biomass. However, the time required for handling and processing surface water samples is several days. By the time analytical results are available, conditions in the field may have changed.

Rose et al. (2015) reported the cost for N abatement measures range from $0.22/kg to $16,742/kg for measures ranging from altered agricultural practices to wastewater treatment upgrades. However, not all of the cost estimates in this range include labor costs. The cost for removing N using suction macroalgal harvesting in this study were estimated to range from $62 to $3,271/kg, depending on the percent N and the percent dw of the macroalgae harvested as well as wages, all of which can all vary considerably.

Based on the information collected in this study, harvest efficiency is the area that needs the most refinement. Future harvests should use finer mesh to ensure that all the macroalgae suctioned are actually collected and bagged on the boat. Several trials of differing mesh size will likely be needed to identify the optimal mesh pore size.

Macroalgal harvesting should avoid macroalgal system collapse that can occur in the summer. The ideal water temperature harvest window for macroalgae in the Falmouth area is believed to be 60 - 70 degrees Fahrenheit (15.5–21 Celsius). Surface water temperature data for the Falmouth area suggests that harvesting should take place in June and again from mid-September to mid-October.

Macroalgal suction harvesting is best conducted in the lower range of the tidal cycle with light tidal current movement. Because harvesting activities disturb the bottom and kick up some debris and particulates, a slight current can help move some of this material out of the harvest area and improve visibility. Preliminary data suggests that harvesting should occur after at least one day of sunny conditions and ideally two days. Observations of eutrophic estuaries in Falmouth suggest that there should be little difficulty in finding areas in spring with ample macroalgal biomass for harvesting.

Macroalgal suction harvesting presents a novel approach to removing excess N from eutrophic estuaries. However, this approach has several limitations that need refinement if it is to have broad application. While the costs per kg of N removed for macroalgal harvesting are on the higher end of the range of N abatement interventions, this approach has a low point of entry cost of roughly $1000 compared to some interventions that require millions of dollars of investment. Macroalgal suction harvesting is unlikely to remove enough N to resolve N loading problems in highly eutrophic estuaries. However, it presents a useful option for communities to reach their TMDL goals in conjunction with other measures and may provide some benthic habitat benefits in the short to medium term.

Appendix 1

Summary of Effects of Macroalgal Blooms

Location	Source	Treatment abundance	duration	Observation	Comments
Baltic Sea	Norko & Bonsdorff, 1996	2 kg ww/m2	34 days	Reduced abundance of most macrobenthic inverts.	~280 g dw/m2 single treatment level- one time algal applic.
Australia	Cummins et al., 2004	4.5 kg ww/ m2	12 weeks	Reduced macrobenthic species abundance	~640 g dw/m2 single treatment level- one time algal applic.
Portugal	Cardoso et al., 2004	0.3 kg ww/m2 no effect	4 weeks	Reduced macrobenthic species abundance species specific response	~30 g dw/m2 multi treatment levels- one time algal applic.
California	Green, 2010	0.5 cm (60 g dw/m2) NOAEC after 2-8 weeks, 8 weeks; 1.5 cm (186 g dw/m2) adverse effect after 4 weeks; 3.0 cm (416 g dw/m2) adverse effect after 2 weeks.	2 - 8 weeks	Increased biomass reduced surface deposit feeders and increased subsurface deposit feeders	multi treatment levels - maintained algal treatment level biweekly
Scotland	Hull, 1987	3 kg ww/m2 adverse effects species specific	22 weeks	After 10 weeks some surface deposit feeders decreased while some subsurface feeders increased. After 22 weeks pattern similar	~420 g dw/m2 - multi treatment levels- one time algal applic.
Scotland	Raffaelli, 1999	No biomass treatment after 10 weeks increase is species specific. 3 kg ww/m2 after 10 weeks adverse effects were species specific. Equivalent abundances of both species in all treatments after 22 weeks	22 weeks	High abundances result in increase of subsurface deposit feeders, decrease in surface deposit feeders after 10 weeks	~420 g dw/m2 - multi treatment levels- one time algal applic.
Sweden	Osterling & Pihl, 2001	1.2 kg ww/m2 adverse effect on all taxa after 21 days	36 days	Macroalgal impacts to all macrofauna @ start. Subsurface detritivores and carnivores positively affected @36 days.	~160 g dw/m2 - multi treatment levels- one time algal applic.

Massachusetts	Deegan et al., 2002	Treatments included control, low macroalgae (removed), high macroalgae (roughly double control) & a disturbed treatment	6 months	Treatment w/macroalgae removed (low macroalgae) had increased eelgrass cover, increased benthic oxygen & increased fish abundance and diversity	High macroalgal biomass ~125 g/m2 wet wt.-4 treatments
California	Everett, 1991	~6 kg ww /m2 adverse effects after 2 months and six months	6 months	Clams and shrimp abundance increased in plots where macroalgae was removed.	863 g dw/m2 removal experiment
Scotland	Bolam et al., 2000	~1 kg ww/m2 species specific effects after 6 and 20 weeks	20 weeks	Surface deposit feeders negatively affected, subsurface feeders positively affected after 6 weeks effects persisted through 20 weeks.	131 g dw/m2 - single treatment level- one time algal applic.
England	Jones & Pinn, 2006	adverse effects >70% cover	not recorded	Species diversity declined when % cover increased from 5 - 70% in one month	Correlative field study- low cover did not always=high diversity
Sweden	Pihl et al., 1995	Some negative effects with 1% cover, greatest effects >30% cover	not recorded	Crabs negatively affected by moderate and high % cover	Correlative field study- 1 day sampling events
Baltic Sea	Lauringson & Kotta, 2006	No clear relationship with mat depth and infaunal abundance	not recorded	Herbivores more prominent within mats; detritivores more prominent in sediment	Correlative field study- subtidal
Italy	Bona, 2006	0.7 kg ww/m2 and >70% cover	not recorded	Loss of Stage III benthic colonization by filter feeders.	~90 g dw/m2 - use SPI camera for correlative field study
California	Green, Sutula & Fong, 2014	identified 110-120 g dw/m2 at 4 weeks as benchmark for adverse effects	10 weeks	Reduced diversity and abundance of surface deposit feeders.	Manipulative field experiment with 5 treatment levels and biweekly monitoring of duration

Appendix 2

Macroalgal Harvesting Methods Summary

Location	Dominant Macroalgae	Harvest Biomass	Objective	Harvest method	Cost	Cost/kg N	Reference
Orbetello Lagoon, Tuscany Italy	*Chaetomorpha linum*	3,000,000 kg	prevent anoxia	Mechanical weed harvester	$5,700,000	$7.92	Lenzi, 1992
Venice Lagoon, Italy	*Ulva rigida*	392,000 kg	prevent anoxia	Mechanical weed harvester	NA	NA	Curiel, Rismondo, Bellemo, & Marzocchi 2004
Brittany, France	*Chlorophyta*	**90,000 m^3	prevent anoxia	Front end loaders	$3,600,000	$0.22	Morand & Briand, 1996
Prince Edward Island, Canada	*Ulva lactuca*	29,000 kg	prevent anoxia	Mechanical weed harvester	$47,076	$58.55	Crane & Ramsay, 2012^
Peel Inlet Western Australia	*Cladophora & Ulva*	**13,000 m^3	prevent anoxia & aesthetics	Front end loader with mesh bucket	$161,000	$58.49	Atkins Deeley, & McAlpine, 1993
Harwich, MA	*Ulva lactuca*	NA	algal blooms	Hand harvested	NA	NA	Harwich, 2008
Delaware	*Ulva & Gracilaria*	NA	prevent anoxia	Mechanical weed harvester	NA	NA	Mike Bott, DNREC, 2019
Jamaica Bay, NY	*Ulva lactuca*	1.9 m^3 in 90 minutes	harvest demonstration	Skimmer boat *	$180,000	NA	NYCDEP, 2012
Lake George, NY#	*Eurasian Milfoil*	47 kg/hour	invasives removal	DASH	$15,800/ha	$0.57	Eichler, 1993
Lake Ellwood, MI	*Eurasian Milfoil*	17 - 70 kg/hr.	invasives removal	DASH	NA	NA	Waters, 2014

^Estimated costs to remove enough macroalgae to prevent anoxic events would be $2,000,000 -$3,400,000 for 3 estuaries.
Cellina, et al.(2002) estimated that mechanical weed harvesters cost approximately 100,000 Euros/year.
*Skimmer boats are comparable to mechanical weed harvesters.
DASH-Diver Assisted Suction Harvest (includes venture pumps capable of handling roots and sediment).
#Costs for Lake George reflect labor costs only.
** For harvest biomass reported in m3, table assumes an estimated weight/m3 of 180 kg, based on Crane & Ramsay, 2012.

Appendix 3

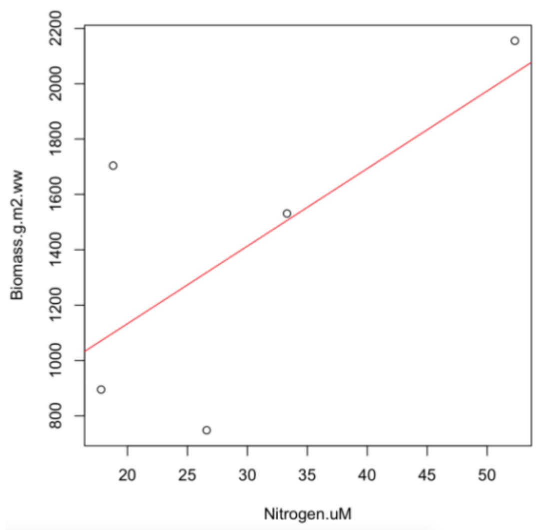
Childs River Macroalgal Biomass Data

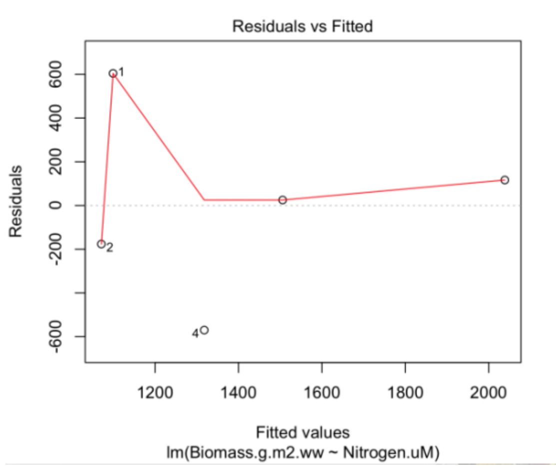
Sample	date	grams/m2 wet wt.	Mean wet wt. g/m2	% dry wt.	dry wt. g/m2	Approximate Macroalgal diversity
CR-QB-1	5/28/19	1440	1704	0.13	222	75% Gracilaria, 25% Ulva
CR-QB-2		2360				
CR-QB-3		1760				
CR-QB-4		1240				
CR-QB-5		1720				
CR-QB-1	6/4/19	1216	895	0.34	304	81% Gracilaria, 18% Ulva, trace Cladophora
CR-QB-2		944				
CR-QB-3		924				
CR-QB-4		580				
CR-QB-5		812				
CR-QB-1	6/14/19	974.08	1531	0.29	444	53% Gracilaria, 47% Ulva
CR-QB-2		1714				
CR-QB-3		1842				
CR-QB-4		998				
CR-QB-5		2127.6				
CR-QB-1	6/27/19	493.2	748	0.30	224	80 Gracilaria, 19% Ulva, trace Cladophora
CR-QB-2		725.2				
CR-QB-3		712.8				
CR-QB-4		733.6				
CR-QB-5		1073.2				
CR-QB-1	7/15/19	3072	2155	0.32	690	50% Gracilaria, 35% Ulva, 15% Cladophora
CR-QB-2		2148				
CR-QB-3		2632				
CR-QB-4		1324				
CR-QB-5		1600				

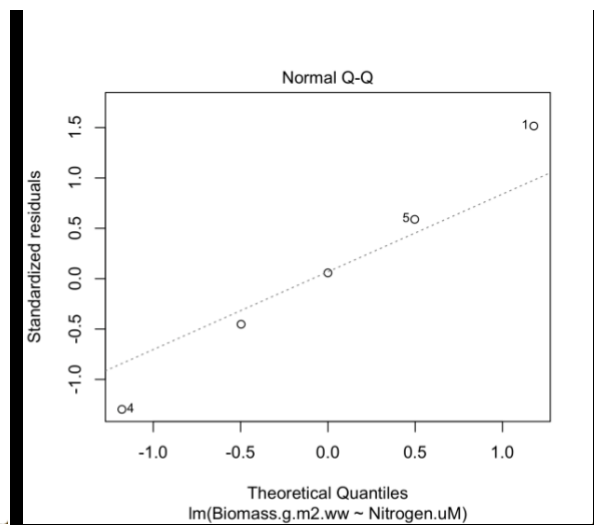
Appendix 4

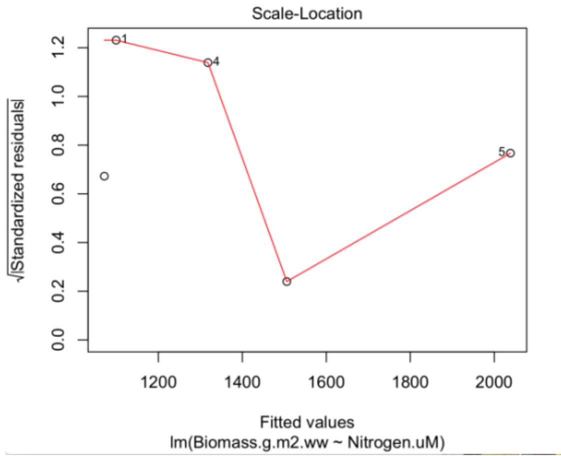
Regression Plots, Residuals and Syntax for Regression Analyses

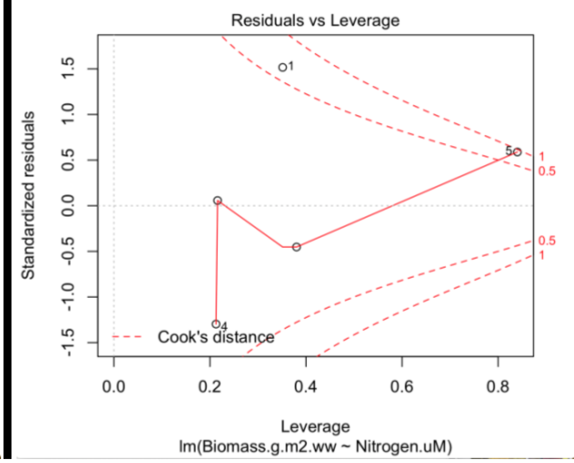
Macroalgal biomass wet weight vs. water nitrogen concentration:





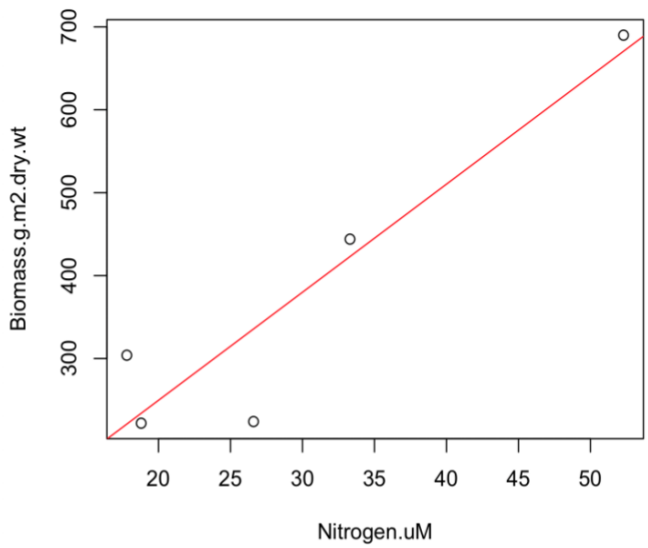


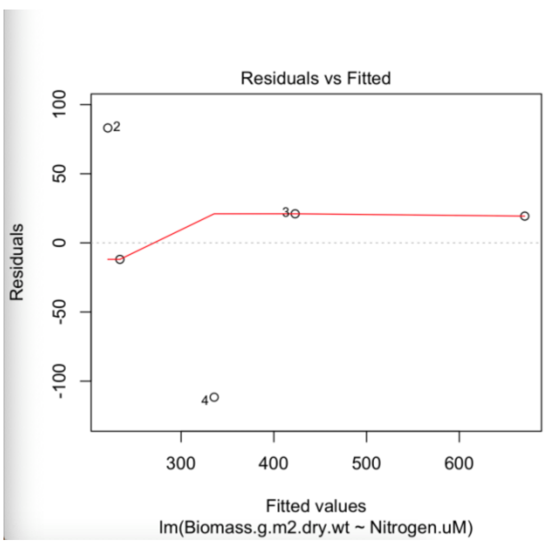


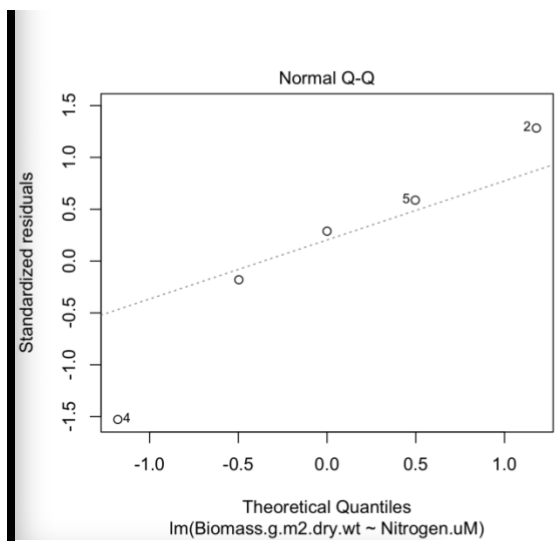


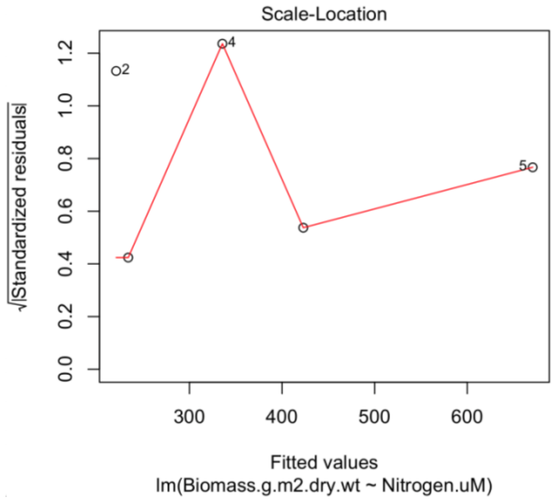

```
Call:
lm(formula = Biomass.g.m2.ww ~ Nitrogen.uM, data = biomassww)

Residuals:
      1       2       3       4       5 
 604.63 -176.34   25.17 -570.02  116.56 

Coefficients:
            Estimate Std. Error t value Pr(>|t|)
(Intercept)   572.37     568.09   1.008    0.388
Nitrogen.uM    28.03      17.58   1.595    0.209

Residual standard error: 495.2 on 3 degrees of freedom
Multiple R-squared:  0.4588, Adjusted R-squared:  0.2784 
F-statistic: 2.543 on 1 and 3 DF,  p-value: 0.2091
```

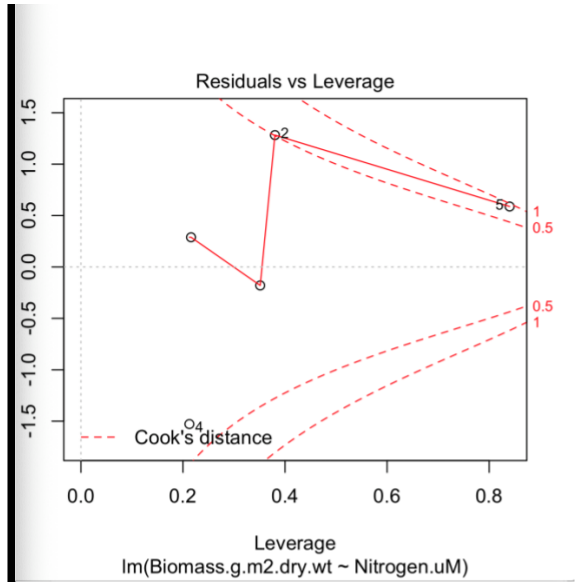
Macroalgal biomass dry weight vs. water nitrogen concentration:












```
Call:
lm(formula = Biomass.g.m2.dry.wt ~ Nitrogen.uM, data = biomassdw)

Residuals:
      1       2       3       4       5 
 -11.92   83.12   21.05 -111.60   19.36 

Coefficients:
            Estimate Std. Error t value Pr(>|t|)  
(Intercept)  -11.169     94.415  -0.118    0.913  
Nitrogen.uM   13.037      2.921   4.462    0.021 *
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 82.31 on 3 degrees of freedom
Multiple R-squared:  0.8691, Adjusted R-squared:  0.8254 
F-statistic: 19.91 on 1 and 3 DF,  p-value: 0.02096
```

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