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*University of New Hampshire, Durham*

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THE DEVELOPMENT, VALIDATION, AND APPLICATION OF THE  
EELGRASS HEALTH INDEX

By

Nicholas Braun Anderson

Bachelor of Science, Marine and Freshwater Biology

University of New Hampshire, 2011

THESIS

Submitted to the University of New Hampshire  
In Partial Fulfillment of  
The Requirements for the Degree of

Master of Science  
in  
Natural Resources

September 2020

THE DEVELOPMENT, VALIDATION, AND APPLICATION OF THE  
EELGRASS HEALTH INDEX

By

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On August 17<sup>th</sup>, 2020

Original approval signatures are on file with the University of New Hampshire Graduate School.

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## DEDICATION

I dedicate this thesis to the Cree of Eeyou Istchee and the First Nation communities of Canada. All should respect your life close to the land and knowledge and stewardship of the natural world. I hope the work in this thesis provides new tools to monitor and assess eelgrass in James Bay and helps you continue to protect your coastal resources.

And

To my father, Gilbert Robert Anderson (1949–2013): teacher, carpenter, writer, and reader of books—he was the first one to put a fishing rod in my hands and spent endless hours with me by the river at ‘The Dam.’ I’m still hanging out below dams and “messaging about in boats.” As a parent and high school teacher, dad always said an education opens doors and creates opportunities. He was right.

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This thesis may have my name on it, but I am indebted to many people for their wise words, kindness, and support. I would like to thank my advisors, Dr. Fred Short and Dr. Catherine Ashcraft.

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## ABSTRACT

### THE DEVELOPMENT, VALIDATION, AND APPLICATION OF THE EELGRASS HEALTH INDEX

By

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University of New Hampshire

September 2020

Eelgrass (*Zostera marina* L) provides essential habitat and forage for waterbirds, fish, and other coastal marine species, nutrient and sediment capture which improves water quality, carbon storage, and wave energy buffering which reduces coastal erosion. Changes in its health can indicate other coastal ecosystem changes. Since the 1980s, eelgrass beds have declined in James Bay, Québec. The eelgrass decline coincided with a decrease in the abundance of migratory Brant and Canada geese visiting the coastal eelgrass meadows, which the geese rely on for forage during their spring and fall migrations. Geese are important species to the coastal First Nation Cree communities of Québec and are harvested by the Cree during these migration periods. The decline in eelgrass and geese threatens culturally significant hunting activities of the First Nation Cree communities of Chisasibi, Wemindji, Eastmain, and Waskaganish. Multiple hypotheses exist for the eelgrass decline but no causes have been directly linked to the loss of eelgrass. As part of a larger coastal habitat monitoring program in James Bay focused on investigating the decline of eelgrass and potential threats and stressors, we developed novel eelgrass monitoring methods suited to the large spatial area and subarctic conditions as well as the eelgrass health index, an index for assessing eelgrass health status.



This study assessed video monitoring as a potential methodology for monitoring eelgrass. Video monitoring and conventional observations were conducted side by side in the Great Bay Estuary in Maine and New Hampshire and compared. Observations for three eelgrass parameters, percent cover, shoot density, and plant height, were made during July and August 2019. Validation for each eelgrass parameter using conventional methods demonstrated that video monitoring results were consistent with results from conventional monitoring. Each of the parameters observed using both methods demonstrated a significant positive linear relationship ( $p < 0.0001$ ) with a moderate to strong goodness of fit. Cover and biomass data collected from this study and previous SeagrassNet monitoring surveys in the region were also used to develop a model to predict eelgrass biomass from percent cover. A simple linear regression using square root transformed biomass was selected as the best model to estimate biomass from cover. Based on the results, the novel video monitoring methodology is found to be a reliable alternative to conventional monitoring methods, which can improve the ability to collect comprehensive data during field monitoring under certain conditions.

The validated video monitoring methods were then applied to assess the current health of James Bay eelgrass beds. Monitoring of eelgrass percent cover, shoot density, plant height, and biomass was conducted from June to September in 2017 and 2018. Eelgrass cover, density, and height were averaged using the geometric mean equation and reference values to calculate the eelgrass health index (EHI)—a novel tool for assessing eelgrass health status. The eelgrass health index was validated using two methods: 1) biomass observations from James Bay and from five long-term SeagrassNet eelgrass monitoring sites in the United States, and 2) a survey of experiential knowledge. We found that ratings from the new EHI are consistent with accepted

metrics and can be used across North America, and with further testing, the EHI could potentially be applied to eelgrass beds throughout the northern hemisphere.

We found that eelgrass is impaired throughout eastern James Bay. Contemporary EHI ratings were compared to historic eelgrass data using a model to predict eelgrass cover from biomass. We found that in comparison to EHI ratings calculated from the historic data, contemporary eelgrass health has declined at two sites in northern Chisasibi, Attikuan and Tees Bay and persisted in similar conditions to the historic environment at Kakassituk.

## INTRODUCTION

The eelgrass beds of James Bay, Québec were once some of the most extensive in North America (Lalumière et al., 1994). The First Nation Cree of James Bay relied on the eelgrass meadows as hunting grounds. Brant and Canada geese frequented the meadows on their spring and fall migrations to feast on eelgrass and marine invertebrates, and subsequently provided the Cree good hunting on either side the of long Canadian winter. Today, geese and the goose hunt are still an essential part of Cree culture. Every coastal James Bay Cree community includes the goose in their nation's emblem and during the migration multi-week holidays occur to allow for goose hunting trips. But, a major decline in eelgrass coincided with the disappearance of the geese in the late 1990s (Castelli et al., 2015) and, today, the goose migration and hunt have diminished from what they once were. The Cree are taking actions to maintain what remains of the goose migration and its cultural significance. For example, communities have created more favorable terrestrial habitat for migrating geese (Sayles & Mulrennan, 2010) and some are traveling to hunt geese in alternative regions in southern Canada.

Seagrasses are meadow forming marine plants. They create ecosystems that provide essential forage and habitat for coastal and marine species, which support commercial fisheries and tourism (Bertelli and Unsworth, 2014; Plummer et al., 2013). Eelgrass (*Zostera marina* L), a species of widely distributed seagrass, inhabits northern temperate and arctic waters. In North America eelgrass is found from North Carolina to northern Québec on the Atlantic coast and from Baja California in Mexico to Alaska on the Pacific coast (Moore & Short, 2006).

Eelgrass has specific habitat requirements and a range of conditions can cause stress and mortality in eelgrass if they are extreme enough or persist long enough. Eelgrass inhabits coastal waters, generally growing in subtidal areas. Although eelgrass can grow in the intertidal zone, it

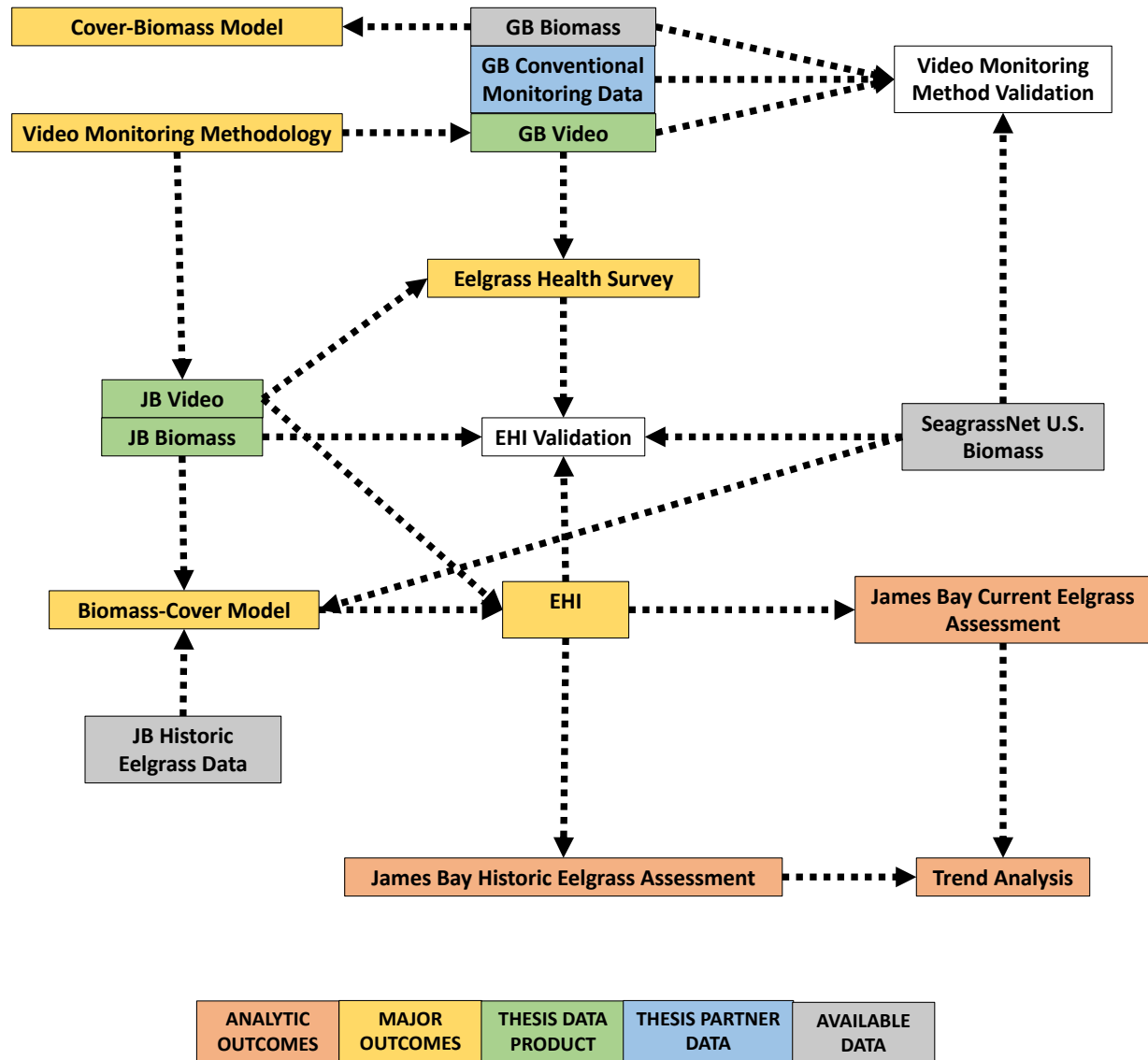
is not tolerant of long-term emersion and is susceptible to exposure and desiccation. As a plant, eelgrass needs sunlight for photosynthesis and production, which limits its depth range to shallow coastal waters. Eelgrass populations exhibit both annual and perennial growth strategies based on their location (Blok et al., 2018). In arctic waters with sea ice and polar night conditions, eelgrass has adapted to survive on reserve carbohydrate stores and with continued under-ice photosynthesis (McRoy, 1971; Olesen and Sand-Jensen, 1993). Water conditions such as turbidity and colored dissolved organic matter (CDOM) can reduce light penetration and negatively affect eelgrass beds. Shoot carbohydrates, production, and growth are directly linked to light availability and eelgrass declines when without light for extended periods of time (Bertelli and Unsworth, 2018; Ochieng et al., 2010). Eelgrass is generally tolerant of lower water temperatures (McRoy, 1971), and has an upper thermal limit of approximately 25 °C. Eelgrass as a euryhaline plant, is tolerant of a wide range of salinities (Nejrup & Pedersen, 2008). Specific eelgrass populations have different salinity tolerances: in a study on Baltic eelgrass, eelgrass performance did not change within a salinity range of 20–25 part per thousand (ppt) for populations from high (16–26 ppt) and low (6 ppt) salinity environments (Salo et al., 2014). Eelgrass production did decrease when salinities were < 15 ppt for both high- and low-salinity tolerant populations (Salo et al., 2014). Extended periods of low salinity can be lethal to eelgrass. Additional environmental stressors, such as low light and high temperatures, can also compound to negatively affect eelgrass.

Despite observations of eelgrass decline, there is no consensus about the current health of eelgrass in James Bay, whether it is continuing to decline, stable, or in recovery is unknown. And, the cause of decline is also unknown. Methods for monitoring and assessing eelgrass health are necessary to provide information into the decline of James Bay eelgrass beds.

The purpose of this thesis is to develop and validate methods for monitoring and assessing eelgrass health. Cree concerns about eelgrass decline in James Bay are a major motivation of this research, but eelgrass concerns are also important in other areas, including the Piscataqua River-Great Bay region within which the University of New Hampshire is located. James Bay is therefore an important case study to test the development of an index that can be applied to investigate eelgrass decline in James Bay, but also to assess eelgrass health more broadly. This research approach uses methods for non-destructive sampling with a focus on versatility and simplicity for use in remote areas, which are also designed to be accessible for community ecological monitoring.

Chapter 1 describes the development and validation of eelgrass video monitoring methods and a model for estimating eelgrass biomass from percent cover observations. Video monitoring was compared to conventional SeagrassNet monitoring methods in the Great Bay Estuary in southern Maine and New Hampshire. Eelgrass percent cover, shoot density, and average plant height were quantified using both methods at sites throughout the estuary. Data from this study and previous SeagrassNet monitoring was used to develop a model for predicting eelgrass biomass from percent cover observations. This model can work in conjunction with video monitoring or standard observations of eelgrass cover to provide biomass estimates where physical sampling is not possible. We found that video monitoring results are consistent with conventional monitoring results and provide a cost-effective and less intensive method for observation. A simple linear model was developed to predict eelgrass biomass from cover and a second exponential model was identified with potential for modelling biomass should additional data on high cover and high biomass sites be collected or become available.

Chapter 2 describes the design and validation of an eelgrass health index using research from James Bay as a case study. We employed the video monitoring methods described in the first chapter to observe eelgrass beds in James Bay. The health index was designed using observed variables that reflect abundance and productivity, eelgrass cover, shoot density, and plant height, and for the assessment of eelgrass threats and stressors in James Bay. The eelgrass health index was validated using two methods: biomass and an eelgrass health survey of experiential knowledge. Both validation efforts used data from James Bay and other eelgrass populations in the United States. The index was consistent with the results of both methods and effectively rated eelgrass. Results from the validation also suggest the eelgrass health index is generalizable to eelgrass populations throughout its global distribution with validation for new populations. We developed a model to predict percent cover values from biomass using James Bay biomass values and supplemental biomass data from long-term Great Bay Estuary eelgrass monitoring. We then used the biomass-cover model to estimate historic eelgrass cover in James Bay. Cover estimates were combined with their corresponding shoot density with the EHI equation and compared to present EHI values. Comparing the contemporary and historic EHI ratings across three sites show eelgrass decline between the mid 1980s and today. The decline was significant at two sites, Attikuan and Tees Bay, and not significant at Kakassituk.



**Fig 0.1** The project process model describes the data, different data sources, outcomes, and how they are connected. Major outcomes are the different models, methods, and the survey developed as part of my research. Thesis data products are data that were collected during the course of my thesis work, while thesis partner data are data collected in collaboration with independent partners working on other projects (PREP). Available data refers to data used that are accessible from peer reviewed publications or were used with permission from long-term seagrass monitoring data sets from SeagrassNet.

Novel video monitoring methods were necessary to observe eelgrass beds in the development of the eelgrass health index for James Bay, and the pathways between video monitoring, the eelgrass health index, and existing data sets used in this thesis are connected.

The process model illustrates my Master's research program and how the research steps and outcomes contribute to chapters one and two (Fig. 0.1). Research was conducted over two geographic regions (James Bay, Québec, Canada and New England, USA) and using two different monitoring methods (conventional and video), as well as with data sets from previous monitoring of Atlantic and Pacific eelgrass meadows. Both data collected during this project and from previous SeagrassNet monitoring projects were used in the analysis of the video monitoring and index methods. Both chapters relied on biomass data collected from previous monitoring in Great Bay, New Hampshire.



## CHAPTER I:

### A Comparative Study of Eelgrass Video Monitoring Methods in the Great Bay Estuary

#### **Abstract**

Monitoring subtidal seagrass ecosystems is a challenge and it requires training, resources, and time. We conducted a comparative study of eelgrass observation methods to evaluate video-monitoring as an alternative to conventional quadrat-transect observation methods for observing percent cover, shoot density, and plant height. A second method, a model for estimating biomass using percent cover, was also developed as a tool for when physical biomass sampling is not possible. We found that the novel video monitoring observations were consistent with conventional eelgrass monitoring results. Two models to predict eelgrass biomass from cover were created: the first one, a theoretical model describing the exponential relationship between biomass and cover and the second, a linear regression that can predict eelgrass biomass to 400 g DW m<sup>-2</sup>. These novel methods are complementary to standard monitoring methods and are intended to help researchers collect comprehensive data during field monitoring.

## Introduction

### *1.1 Significance of eelgrass*

In the Gulf of Maine and throughout northern coastal waters, eelgrass (*Zostera marina* L) provides numerous critical ecosystem services. As an underwater flowering plant, eelgrass forms bed or meadow ecosystems that capture nutrients and stabilize sediments, improving water quality conditions and supporting marine species and recreation (Waycott et al., 2009). Eelgrass beds also dampen wave energy from storms, limiting coastal erosion (Koch et al., 2009). Furthermore, eelgrass acts as a “bio-indicator,” or a sentinel species, predicting future changes to coastal ecosystems (Short et al., 2006). However, eelgrass populations in New Hampshire and throughout the northern hemisphere are declining as a result of pollution, coastal development, disease, and climate change (Short et al., 2006). If eelgrass beds disappear, water quality, fisheries, and coastal resiliency will be negatively affected (PREP 2018) and recent research has highlighted the need for expanded monitoring efforts in the Great Bay Estuary to assess the current status of eelgrass and potential restoration sites (Burdick et al., 2020).

### *1.2 Eelgrass monitoring*

Eelgrass monitoring provides data critical to understanding eelgrass threats, stressors, and environmental conditions. Typically, in situ indicators of eelgrass conditions are used to assess eelgrass health, such as percent cover, shoot density, and biomass (Harris et al., 2012). However, the fieldwork to collect standard data on these parameters can be time-intensive, complex, and expensive (Tyne et al., 2010). For example, transect surveys provide high resolution data essential for large scale modeling and evaluation of eelgrass ecosystems (Neckles et al., 2012). But, transect surveys often require specialized experience, such as training in underwater visual

censuses (UVC) via SCUBA, which limits who can participate in monitoring, the sites that can be observed, and the conditions under which research can be conducted (Tyne et al., 2010). In northern waters water temperatures are cold during winters and throughout much of the year and visibility after coastal storms is low, which regularly create prohibitive conditions for in situ monitoring, or limit monitoring to a seasonal endeavor. These challenging conditions necessitate creative new solutions that build on techniques already used by researchers.

### *1.3 Use of videography in eelgrass monitoring*

Underwater videography has previously been used to monitor eelgrass and to efficiently collect data over large areas (McDonald et al., 2006), but its versatility has not been fully adapted to quantify data collected from transect survey methods. Historically, videography has been used to record “conspicuous changes”, but was considered too underdeveloped to capture clear imagery to quantify seagrass characteristics at the spatial scale needed for detailed observation and analysis (Kirkman, 1996). Specifically, video monitoring has been used to assess eelgrass cover (McDonald et al., 2006; Reeves et al., 2007; Schultz et al., 2011). Video was also used to estimate shoot density as a function of cover or coarsely by ocular estimate, but density has not been directly assessed from video using objective observation methods (Schultz et al., 2011; O’Neill et al., 2011). Density has also been estimated using an observer rating system from least shoots to most shoots (0 – 25, 26 – 50, 51 – 75, 76 – 100) (Schultz et al., 2011). While other previous studies have coupled video with an existing method, side-scan sonar, to identify eelgrass beds and provide data to estimate cover and height (Lefebvre et al., 2009), the video was not used to assess other plant and population characteristics.

Underwater video cameras have improved substantially in recent decades in unit size, cost, image-quality, and ease of use (Mallet & Pelletier, 2014). As a result of these improvements, many eelgrass parameters can now be quantified through video-monitoring, which reduces the need for multiple instruments and has the potential to improve the accuracy and scope of current observation methods. This research develops a methodology to advance the use of video-monitoring and analysis in non-destructive and efficient eelgrass monitoring.

Biomass is an important metric for evaluating the health of plant communities and seagrass beds (Collins and Weaver, 1988; Carstensen et al., 2016). It requires in situ collection of eelgrass shoots through coring, which can produce high variance in the results as cores often damage plants or capture leaf material from plants outside of the core sample. Single shoot selection is another alternative, but still requires underwater collection as well as shoot density data for the final calculation.

Two previous studies developed methods for estimating seagrass biomass using cover observations. Cover-biomass relationships were developed for eelgrass beds in Denmark using depth, light, and eelgrass cover (Carstensen et al., 2016). The Danish model provides a more comprehensive representation of the ecosystem because it includes environmental factors, but the environmental data required for the model may not always be available for specific sites. A second study on United States (U.S.) Gulf coast genera (*Thalassia*, *Halodule*, *Syringodium*) excluded abiotic factors and used simple linear models to model biomass from cover (Congdon et al., 2017). Linear models are effective for modeling biomass at values  $< 300$  g DW m<sup>-2</sup>, which works for many eelgrass beds. However, linear models do not accurately model higher eelgrass biomass, which is problematic because eelgrass can reach high values in excess of 1500 g DW m<sup>-2</sup> (McRoy 1970; Olesen et al., 2015). A non-linear cover-biomass model would provide

researchers a simple method for estimating eelgrass biomass from percent cover when collection efforts fail or are impossible, as well as a method for estimating higher biomass values.

#### *1.4. Study objectives*

This study had two objectives: (1) evaluate the potential of video-monitoring as an alternative technique for eelgrass monitoring by comparing video transect surveys to conventional visual surveys and (2) develop a cover-biomass model to estimate biomass using cover observations. We compare novel video-monitoring assessments to standard in situ survey assessments of eelgrass population-level characteristics (cover, density, and height) and environmental conditions (light and temperature). We then calibrate and test an eelgrass percent cover-biomass model using biomass samples collected from field sites and associated percent cover assessed by ocular estimation and video monitoring. The development of a percent cover-biomass model for eelgrass will allow researchers to estimate biomass based only on observed percent cover, which is important at sites where eelgrass cannot or should not be harvested due to adverse conditions or restoration efforts, respectively. This model will reduce the amount of time and laboratory effort typically needed for biomass quantification, as compared to techniques that require field sample collection and processing.

## Methods

### 2.1. Monitoring sites

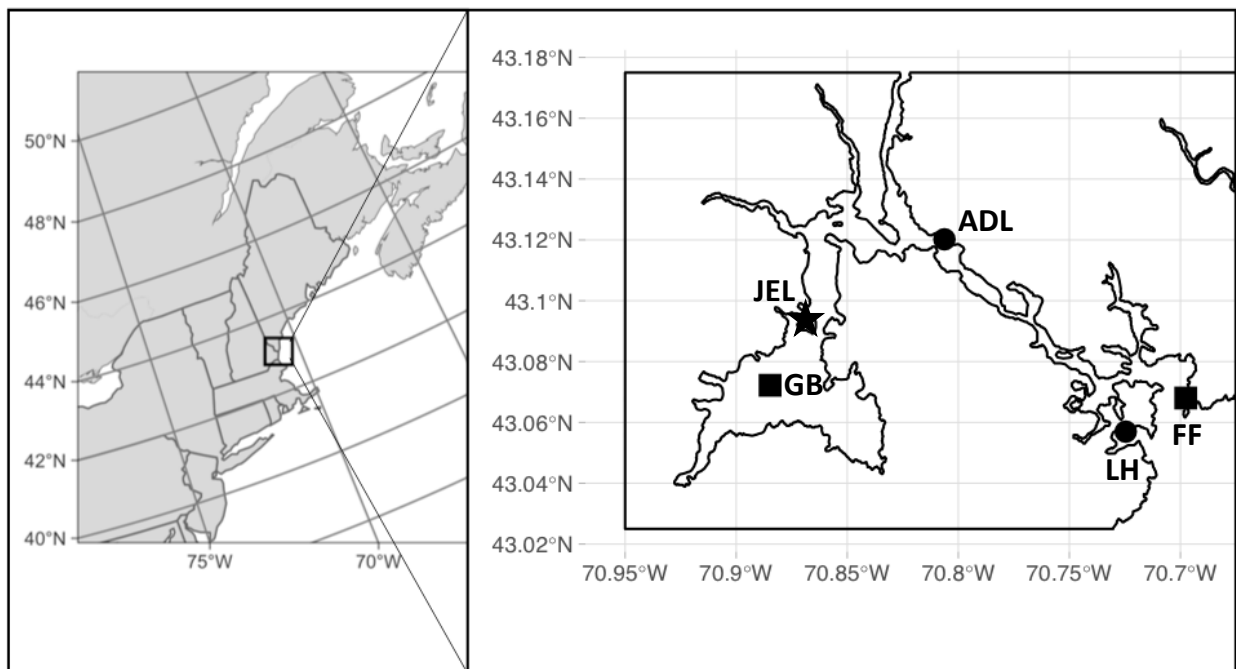
Eelgrass was observed in the Great Bay Estuary (GBE) during the peak growing season in July and August 2019. The GBE system is home to a wide range of eelgrass conditions within a compact and accessible region, and is, therefore, ideal for monitoring (Table 1.1). Four general monitoring sites were selected to represent a broad range of conditions under which eelgrass grows locally: Fort Foster (FF), Little Harbor (LH), Adlington Creek (ADL), and Great Bay (GB) (Fig. 1.1).

**Table 1.1** Survey site coordinates, observation method, and physical and environmental characteristics. Sites are reported starting with the upper estuary first and descending to the lower estuary last.

Site	Location	Estuary location	Method	Depth (m)	Clarity	Substrate
Great Bay	43.072°N, 70.884°W	Upper	Trans. (x3)	0.5	Low	Mud
Adlington Creek	43.12°N, 70.806°W	Middle	Spot	1.0	Moderate	Mud/sand
Little Harbor	43.057°N, 70.842°W	Lower	Trans. (x1)	0.75	High	Mud
Fort Foster	43.068°N, 70.697°W	Lower	Trans. (x3)	2.0	High	Sand

Eleven transect surveys were conducted across the four sites in July and August of 2019. GB and LH were surveyed twice, once in July and a month later in August, while ADL and FF were surveyed once. Survey sites had either one or three transects. Sites with more extensive eelgrass beds or a steeper depth gradient had three transects arranged along a depth gradient from shallow to deeper water to capture variation within the eelgrass bed. Three established transects were monitored at both GB and FF, while at LH only one transect was monitored. Eelgrass at the ADL site was too sparse for a transect to capture more than one quadrat per transect and was therefore observed via spot-monitoring, i.e., a quadrat was placed where eelgrass was present to guarantee the site was included in the survey and monitoring conducted. One established transect (A) in Great Bay contained no eelgrass. Sampling was therefore shifted to an adjacent site close

to the edge of a mudflat, A1, where eelgrass had recently begun to re-colonize and was only monitored once.



**Fig. 1.1** Map of the sample sites in the Great Bay Estuary. Squares denote established SeagrassNet sites and circles denote new sites. Established sites had three transects each. New sites only had one transect each.

## 2.2. Eelgrass monitoring methodologies

Monitoring surveys were conducted using a 0.25 m<sup>2</sup> PVC frame quadrat, generally along 50 m transects. Transects were established with screw anchors, a 50 m tape measure, and marker floats. For quadrat observations, video and conventional SeagrassNet monitoring were conducted along these transects at predetermined random intervals between 0 and 50 m for each site. Five to 12 quadrats were observed per transect based on eelgrass abundance. SeagrassNet observations for each quadrat were collected as close to low tide as was feasible. The same quadrats were observed by video either before or after SeagrassNet monitoring had occurred since video monitoring was less dependent on tidal depth. SeagrassNet monitoring was

conducted by wading or SCUBA and supported by motorboat. Video observations were made by wading or snorkeling, depending on the site depth, and supported by kayak or motorboat.

### *2.2.1. Conventional monitoring*

Standard SeagrassNet monitoring procedures based on the *SeagrassNet Habitat Monitoring Manual* were used as conventional methods to quantify eelgrass percent cover, plant height, and shoot density (Short et al., 2015). Eelgrass percent cover was determined in the field by ocular estimation of the eelgrass present within the PVC quadrat. Researchers conducting the monitoring had previous cover observation training based on the Seagrass Percentage Cover Guide (Short et al., 2015). Shoot density counts were made for each 0.25 m<sup>2</sup> quadrat by counting all shoots present within the 0.25 m<sup>2</sup> area or a subsection of the quadrat (0.125 m<sup>2</sup> or 0.0625 m<sup>2</sup>) depending on whether eelgrass density was high or very high. The height of five shoots from each quadrat was measured and averaged to determine mean height. If a site had very sparse eelgrass density (< 5 shoots m<sup>-2</sup>), which was observed at ADL, the shoots present were measured and averaged for a 1 m<sup>2</sup> area as eelgrass was absent from the area surrounding the observed quadrat.

### *2.2.2. Video monitoring*

Video-monitoring was conducted with a GoPro camera and white disk attached to an extension pole (Fig. 1.2) and analyzed at the University of New Hampshire's Jackson Estuarine Laboratory using the National Institute of Health's ImageJ/Fiji version 2.0.0 and Adobe Photoshop Elements 10. Video for each quadrat was recorded at two heights above the substrate: over-canopy (0.5 m–1.5 m) and under canopy (0.25 m–0.5 m). Over-canopy filming height was



adjusted based on the eelgrass canopy to be above the eelgrass bed. Under-canopy video was collected to capture the basal area of the eelgrass bed to quantify shoot density. A white disk mounted at the end of the extension pole was used as a reference (diameter = 15.2 cm) to calculate the dimensions of the video frame and for estimating eelgrass shoot height. Video was recorded at each quadrat for approximately 15–30 seconds to allow any disturbance to the sediment or eelgrass to settle.



**Fig. 1.2** Eelgrass videography set-up with camera, extension pole, and white disk (left) and a representative image from the video showing the white disc within the eelgrass bed (right).

### *Percent cover*

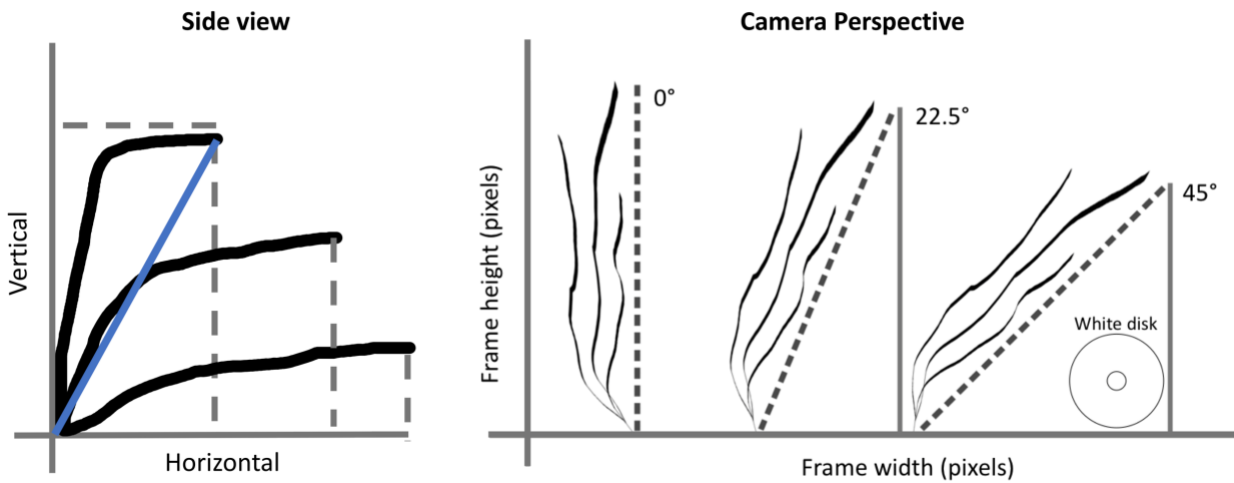
Percent cover from video frames was observed from within each quadrat using point-intercept counts (Caratti, 2006), or where the quadrat was not visible, from within a square grid approximating the same area. Images were overlaid with a semi-transparent grid in photoshop and every intercept where eelgrass leaf cover was present was marked and counted. Total marked intercepts for a quadrat observation were then summed, divided by the total possible intercepts, and multiplied by 100 to calculate the estimated cover percentage (0 -100%).

### *Shoot density*

Density was estimated by counting the number of shoots visible within a section of the frame. Video taken below the canopy was used to assess shoot density where cover or water conditions obscured the basal area and prevented shoot counts. At sites with high shoot density, a subsection of the frame within the quadrat was observed and then multiplied by its corresponding correction factor (4 or 16) to calculate the estimated shoot density  $m^{-2}$ . Individual shoots were denoted with a circle in Photoshop and counted once the subsection had been completely observed. If the basal area of eelgrass shoots in a quadrat was not apparent, then the video was reviewed as a reference to identify eelgrass shoots. The wave action and waterflow helped in the identification of shoots as motion provided more context to which leaves were associated with a specific plant.

### *Plant height*

Eelgrass plant height was estimated with the Pythagorean theorem using estimated horizontal leaf length and canopy height. The area of the image frame was calculated based on visual references: the white disk (15.2 cm diameter) or the known area of the quadrat in the image (50  $cm^2$ ). The visual reference pixel dimensions were calculated in Photoshop and Preview (v 10.0) and used to calculate the image area. Eelgrass horizontal length was estimated using these values at angle increments of  $22.5^\circ$  (Fig. 1.3). Vertical height was estimated based on marks on the extension pole (25, 50, and 100 cm) and the height of the camera. Horizontal length and vertical height were used to calculate the hypotenuse, which was included as the estimated plant height. For shoots estimated at less than 0.5 m in height or flattened close to the substrate by a strong current, estimates were often made using only the horizontal or vertical value.



**Fig. 1.3** The side view (left) and camera perspective (right) for estimating the vertical and horizontal eelgrass length from video. Eelgrass height estimates are the hypotenuse, denoted by the blue line, calculated from the vertical canopy height and observed horizontal eelgrass length values as estimated from the extension pole and image frame dimensions, respectively.

### 2.3. Environmental monitoring

Eelgrass grows under a wide range of physical conditions and two factors are critical for its abundance and productivity: light and temperature. Light availability and water temperature affect eelgrass growth and mortality (Ochieng et al., 2010; Short & Neckles, 1999). In the Great Bay Estuary light availability and water temperature are the major physical parameters affecting eelgrass distribution. To characterize these two drivers of eelgrass productivity across the monitoring sites, light (Lux) and temperature (Celsius) were recorded at each site using in situ HOBO pendant data loggers (Light/Temp, 64k/UA-002-64). Loggers were stationed near one transect at each site in areas unobscured by eelgrass cover and representative of the area. Loggers recorded light and temperature every 5 minutes. An additional sensor was deployed above water at the Jackson Estuarine Laboratory to record fully unsubmerged lux values (Fig. 1.1), which were used to standardize site lux values to a unitless value, percent light. Lux and temperature

observations were averaged for the two-week period after logger deployment in early August. Lux values between 09:00 and 15:00 were selected to represent peak daylight and all other values were excluded from the calculations. Lux values in this range with inflated values ( $50,000 < \text{Lux}$ ) or with corresponding high temperature values ( $29 < ^\circ\text{C}$ ) were also removed from the calculation as observations where data logger emersion potentially occurred. Depth was estimated for each site based on low tide measurements and field observations during eelgrass monitoring. Site clarity and substrate type were observed in the field and verified post hoc while viewing site videos.

#### *2.4. Cover-biomass model development*

During field monitoring, eelgrass shoots were collected from each transect site for biomass measurements. Shoots were separated from rhizomes at the base of the meristem and dried at  $60\ ^\circ\text{C}$  for 72 hours. After drying the leaves were weighed for each quadrat and the grams dry weight (g DW) multiplied by the shoot density (shoots  $\text{m}^{-2}$ ) for the quadrat to calculate aboveground biomass (ABG; g DW  $\text{m}^{-2}$ ). Percent cover and biomass were averaged by transect ( $N = 11$ ) and incorporated into a model to predict biomass from percent cover.

Multiple models were explored to describe the relationship between cover and biomass. Three exponential regressions—3-polynomial (P), biexponential 4P, and biexponential 5P—were fit to the data. Two other models were also explored using transformed data in an effort to normalize the data distribution: a Monod equation using log transformed biomass and a simple linear regression using square root transformed biomass. The model(s) that best represented the relationship between eelgrass biomass and cover was selected based on goodness of fit, Akaike Information Criterion (AIC) score (Akaike, 1974), and nearness to regressing through the origin,

i.e., when eelgrass is absent (0% cover) biomass is zero. Models were explored using the JMP Pro 14 'Fit Curve' platform and through Microsoft Excel and the Solver add-in.

To increase the sample size of the data used in the model, cover and biomass observations from two long-term monitoring sites in the GBE were included in the model (data was used with permission from SeagrassNet). Data was collected using conventional monitoring methods from the *SeagrassNet Habitat Monitoring Manual* for three transects at Fishing Island (FI) between April 2002 and October 2010 and from three transects in Great Bay between August 2007 and to July 2018. Biomass and cover values for these sites were also averaged by transect for each sampling effort, which produced 30 observations for Fishing Island and 68 additional observations for Great Bay.

Two outliers in the cover-biomass model with contradictory values, e.g., low cover and high biomass or high cover and low biomass, were flagged and removed from the analysis. Both outliers were from the supplementary data from previous monitoring in Great Bay. High cover and high biomass, or high leverage points, were maintained in the dataset for developing the model because these points were important for maximizing the range that the model could be used to predict biomass.

## 2.5. Data analysis

Quadrat observations were averaged by transect and the relationships between SeagrassNet- and video-monitoring for each eelgrass parameter were analyzed using simple linear regression. A two-way ANOVA was used to test the difference between methods for each transect and the interaction between methods and transects. Light and temperature means were reported by site and ANOVA was used to test if they were significantly different at the four

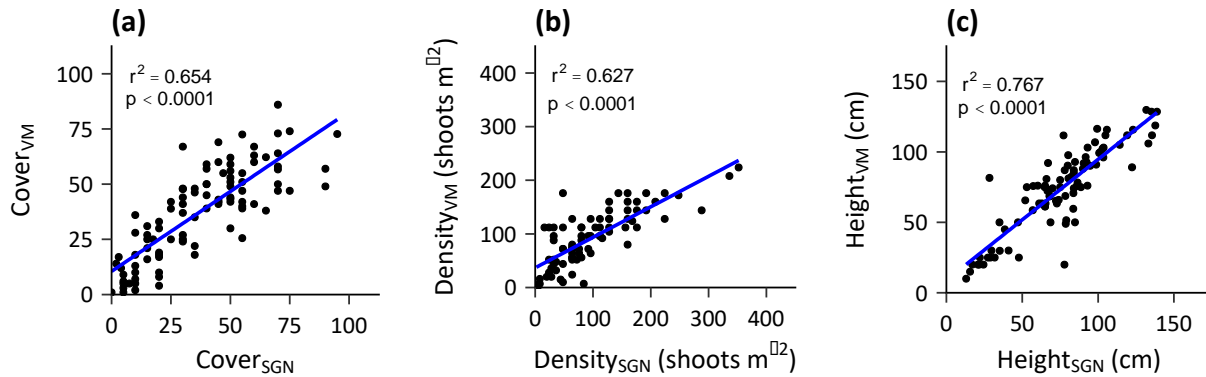
study sites to ascertain if site conditions were representative of the range of light and temperature regimes in GBE eelgrass beds. The cover-biomass relationship was analyzed using the JMP Pro 14 from the Statistical Analysis Systems Institute Inc. (SAS) 'Fit Curve' platform. The map of the region and estuary were created in R using the 'ggplot2' and 'tmap' packages and data from <https://www.naturalearthdata.com/>. Figure formatting in R was done using modified code based on AFS journal requirements (Glassic et al., 2019).

## Results

This section first describes the linear relationship between video monitoring and conventional eelgrass observation methods for percent cover, shoot density, and plant height. Next, it compares the two methods across all the monitoring transects. After that, it describes the model selection process for estimating biomass from cover and how the final model works to predict biomass.

### *3.1. Eelgrass monitoring methods linear relationships*

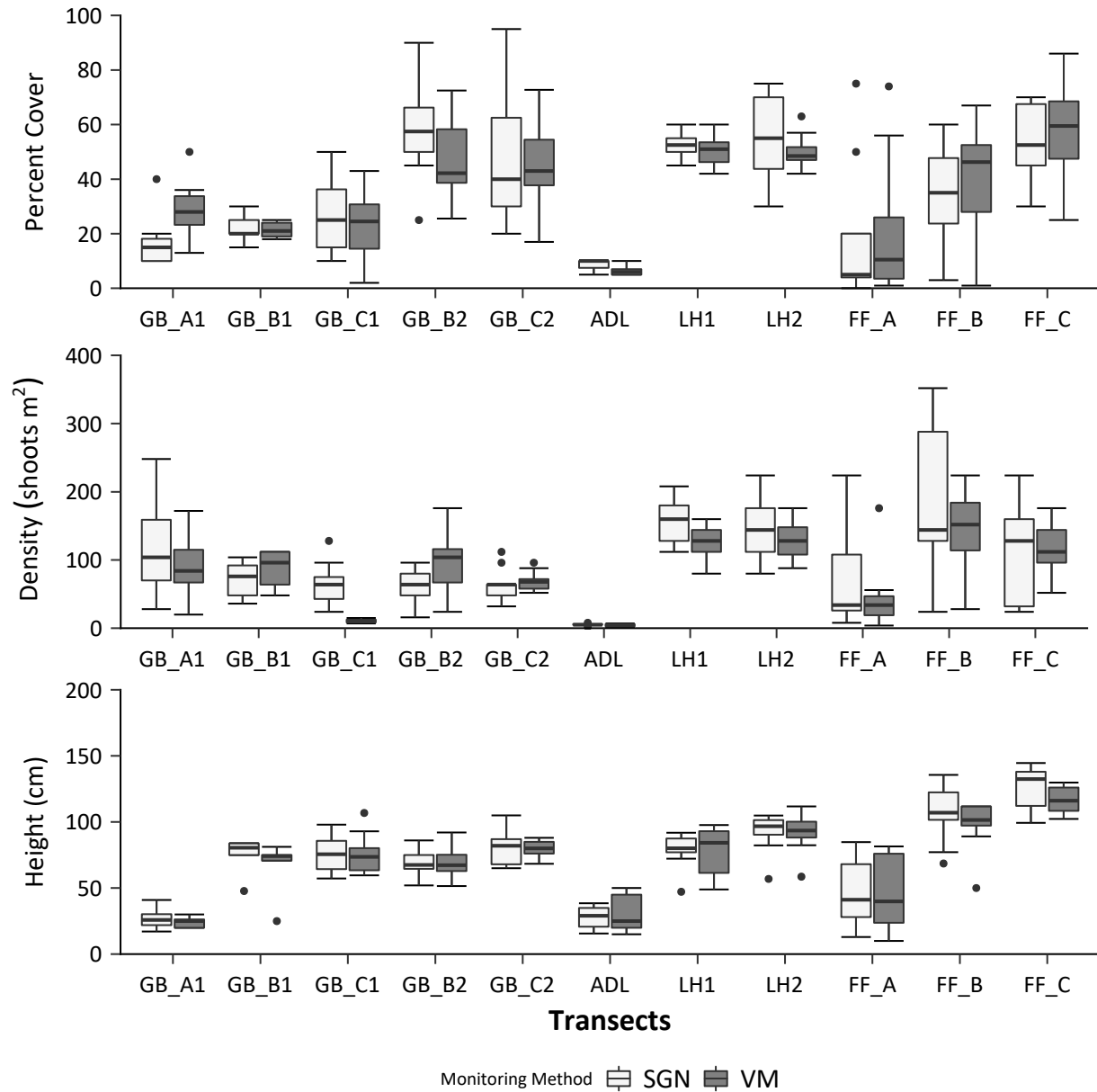
Video-monitoring observations were generally consistent with standard SeagrassNet observations. Regressions for conventional field observation and video-monitoring methods of eelgrass cover, shoot density, and plant height all demonstrated significant positive linear trends (Fig. 1.4). The relationship between the two cover observation methods was moderate but significant ( $r^2 = 0.654$ ,  $p < 0.0001$ ; Fig. 1.4a). The greatest cover observation by video-monitoring (86%) was still well below the maximum recorded using conventional methods (95%), which could be due to either underestimation by the video-monitoring or overestimation by the conventional methods. Conventional cover estimates are made using a visual assessment of eelgrass. The inherently subjective nature of this method may contribute to a less robust relationship between the conventional and video-monitoring methods. Other factors that could contribute to a less robust relationship between the two methods are changes in the physical conditions, e.g., changes in the currents or light levels.



**Fig. 1.4** Simple regressions for eelgrass parameter results for video monitoring (VM) as a function of SeagrassNet (SGN) observations for cover (a), density (b), and height (c).

Similar to cover, the relationship between the methods for observing density was moderate but significant ( $r^2 = 0.627$ ,  $p < 0.0001$ ; Fig. 1.4b). The density relationship had the most gradual slope, approaching 0.5 with the video-monitoring observations regularly underestimating density as compared to conventional shoot counts. This finding suggests that for a more accurate assessment, future studies using video-monitoring to evaluate density may need to include a correction factor to bring the values closer to a 1:1 slope. The model for height performed the best with a strong relationship between both monitoring methods ( $r^2 = 0.787$ ,  $p < 0.0001$ ; Fig. 1.4c). Height also had a slope approaching 1:1 at 0.895:1, which suggests that no correction would be necessary in future application of this model.





**Fig. 1.5** Eelgrass parameters by monitoring method and transect. Boxplots are organized first from upper (left) to lower (right) estuary location and then chronologically for sites that were sampled multiple times. Boxes represent the interquartile range and black dots represent outliers, 1.5x the interquartile range (IQR).

Video-monitoring and conventional monitoring methods produced similar results for each eelgrass parameter when comparing the observations for each transect. Eelgrass cover was not significantly different between both methods ( $p = 0.907$ ; Fig. 1.5), but it did vary

significantly across the 11 sites monitored ( $p < 0.0001$ ). Shoot density and eelgrass height followed the same pattern with significant variation across the sites ( $p < 0.0001$ ) but not between the two methods (density,  $p = 0.109$ ; height,  $p = 0.283$ ). Interactions between site and method were also not significant for any of the observed parameters (cover:  $p = 0.621$ , density:  $p = 0.368$ , height:  $p = 0.977$ ). The finding that the selected eelgrass beds vary in cover, density, and height, supports the site selection approach, which aimed for diversity across eelgrass beds. The variation between observations using the two monitoring methods was not significantly different, which supports the use of video-monitoring as an alternative technique for eelgrass monitoring.

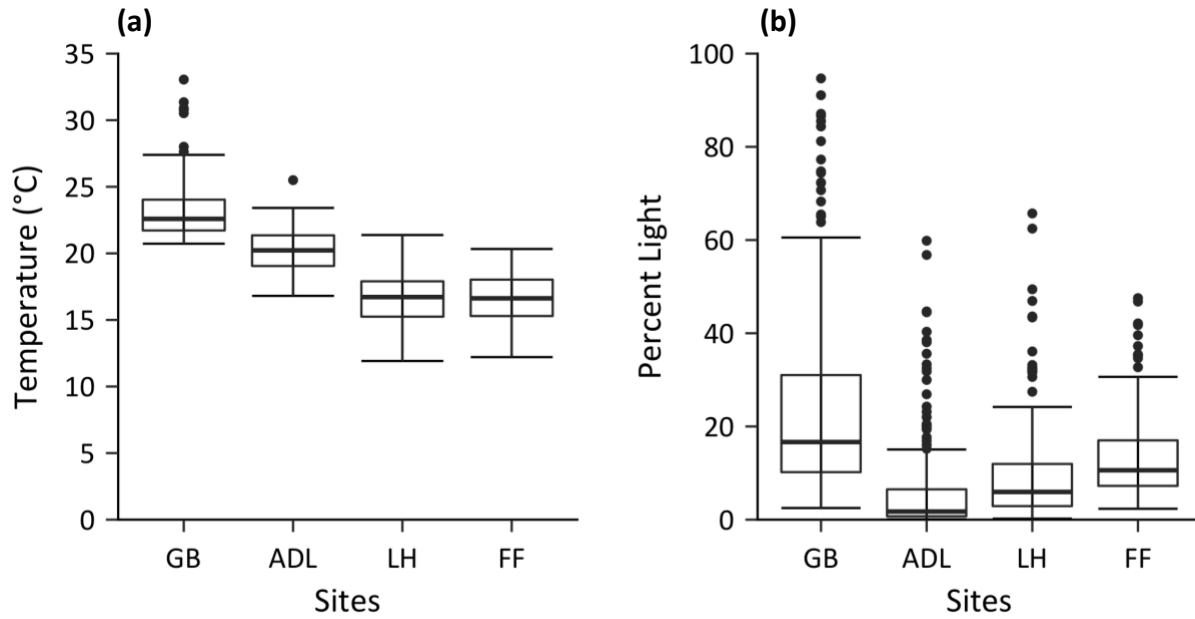
### *3.2. Environmental conditions*

The sites observed in this study had diverse light and temperature conditions, and therefore provided a range of conditions under which eelgrass grows in the region to test the video monitoring methods. Temperature throughout the estuary was relatively stable during the observation period for coastal and mid-estuary sites (Table 1.2) but varied significantly between the coastal sites and the sites in the inner river and bay ( $r^2 = 0.836$ ,  $p < 0.0001$ ; Fig. 1.6a). The lower estuary coastal sites at FF and LH had lower mean water temperatures (14.92 °C and 15.21 °C; Table 1.2, Fig. 1.6a), characteristic of areas with more oceanic influence (Table 2). The mid and upper estuary sites at ADL and GB, respectively, had higher mean water temperatures (18.54 °C and 22.25 °C; Table 1.2, Fig. 1.6a). As compared to the coastal sites, the ADL and GB sites are shallow, more protected sites with less mixing with ocean water, which could explain the temperature gradient.

**Table 1.2** Survey sites environmental conditions and average and median in parentheses eelgrass parameters. Values are reported starting with sites in the upper estuary first and descending to the lower estuary last. CI are confidence intervals for temperature and percent light.

<b>Site</b>	<b>Temp (°C)</b>	<b>95% CI</b>	<b>% Light</b>	<b>95% CI</b>
Great Bay (GB)	22.25 (22.6)	(22.15, 22.35)	17.45 (16.7)	(16.43, 18.46)
Adlington Creek (ADL)	18.54 (20.2)	(18.41, 18.66)	10.69 (1.75)	(9.409, 11.96)
Little Harbor (LH)	15.21 (16.7)	(15.17, 15.29)	13.14 (5.95)	(12.21, 14.077)
Fort Foster (FF)	14.92 (16.6)	(14.83, 15.01)	14.56 (10.6)	(13.65, 15.47)

Percent light varied between sites but did not show a trend across the estuary ( $r^2 = 0.025$ ,  $p < 0.0001$ ; Fig. 1.6b). Percent light was greatest in the upper estuary at GB (17.45 %), followed by the coastal sites at FF and LH (14.56% and 13.14%), and was most limited in the mid estuary at ADL (10.69%) (Table 1.2, Fig. 1.6b). ADL was adjacent to the main shipping channel and experienced heavy commercial and recreational traffic, so wave action from vessels and resuspension of the mud substrate likely caused the site’s low light levels. LH also had lower percent light, which may be explained by conditions similar to ADL, traffic from the adjacent marina and the location of the eelgrass bed on a mudflat. FF was also near the main channel at the mouth of the Piscataqua River and experienced high boat traffic, but with strong currents and a heavier sand substrate, resuspended sediment likely settled out of the water column quickly and did not impair light penetration. Although the GB site was in mud substrate, like ADL and LH, the limited boat traffic near the eelgrass bed and the location of the sites deeper in the bed may have led to the relatively improved light conditions. GB on average was also the shallowest site (0.5 m), so light did not have as far to penetrate as at the other sites.



**Fig. 1.6** Median temperature (a) ( $r_2 = 0.836$ ,  $p < 0.0001$ ) and percent light (b) ( $r_2 = 0.025$ ,  $p < 0.0001$ ) at monitoring sites. Boxes represent the IQR, tails are 1.5 x IQR, and black dots are outliers. Sites are plotted for Great Bay Estuary from the upper estuary (left) to the lower estuary (right).

The eelgrass bed characteristics observed varied by site (Table 1.3). LH had the highest average cover and density of the four sites. Fort Foster had the next highest density followed by Great Bay, and Adlington Creek, respectively. Fort Foster and Great Bay had similar mean cover values, with the observations from both monitoring methods overlapping in their range. Eelgrass height was greatest at Fort Foster followed by Little Harbor, Great Bay, and Adlington Creek, respectively.

**Table 1.3** Eelgrass variable mean, min, and max values by observation method and site.

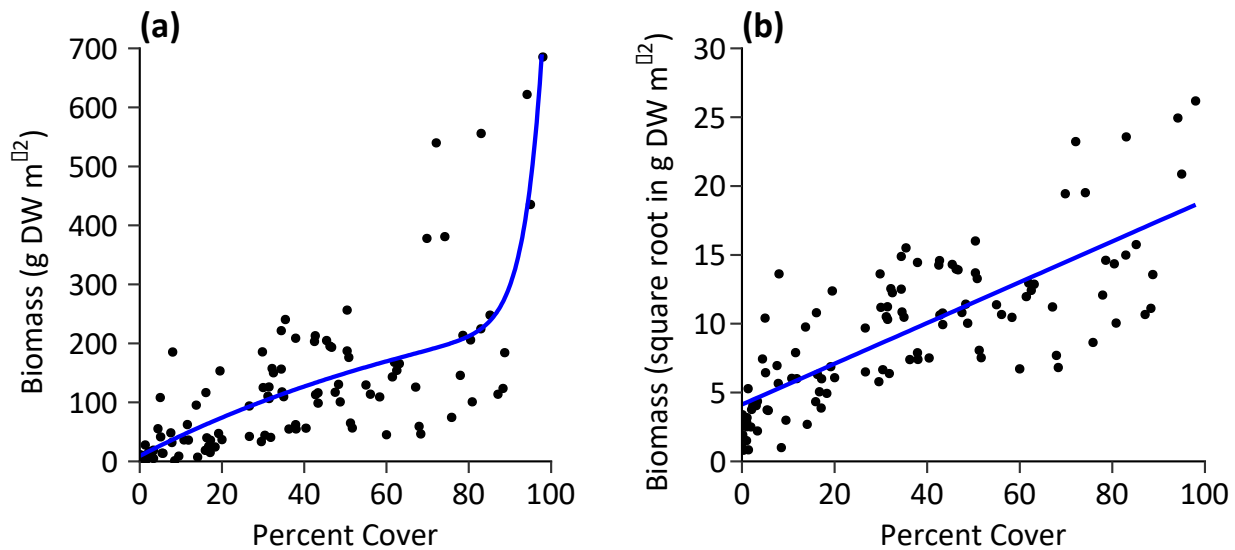
Variable data	Adlington		Fort Foster		Great Bay		Little Harbor	
	<i>SGN</i>	<i>Video</i>	<i>SGN</i>	<i>Video</i>	<i>SGN</i>	<i>Video</i>	<i>SGN</i>	<i>Video</i>
N	4.0	4.0	28.0	28.0	44.0	44.0	15.0	15.0
Mean Cover	8.1	6.5	32.9	39.1	39.0	35.0	53.7	50.6
Cover min	5.0	5.0	0.0	1.0	10.0	2.0	30.0	42.0
Cover max	10.0	10.0	75.0	86.0	100.	72.7	75.0	63.0
N	4.00	4.0	25.00	25.00	36.00	36.00	15.00	15.00
Mean Density	5.25	4.5	100.8	94.56	76.11	80.56	153.6	126.4
Density min	2.00	2.0	4.00	4.00	16.00	7.00	80.00	80.00
Density max	8.00	7.0	352.0	224.0	248.0	176.0	224.0	176.0
<i>N</i>	4.00	4	24.00	24.00	43.00	43.00	15.00	15.00
Mean Height	27.46	32.5	95.25	88.30	67.27	65.19	85.57	86.8
Height min	15.60	15.0	13.00	10.00	17.08	20.00	47.20	50.00
Height max	38.50	50.0	138.8	129.8	98.00	106.83	104.83	111.8

### 3.3. Biomass predicted from percent cover

The model to predict biomass using percent cover or cover-biomass model was created by comparing methods of linear regression (simple and exponential) and selecting one that most accurately represented the relationship between the cover (independent variable) and biomass (dependent variable) for each transect per survey. Two final models were accepted for a final comparison: a biexponential five polynomial (5P) regression (Fig. 1.7a) and a simple linear regression (Fig. 1.7b; Eq. 2). Both models had a similar fit, with the 5P model (0.619) incrementally better than the linear model (0.595). The linear model had a lower AIC score (584) than the 5P model (1275), indicating a better fit for the linear model.

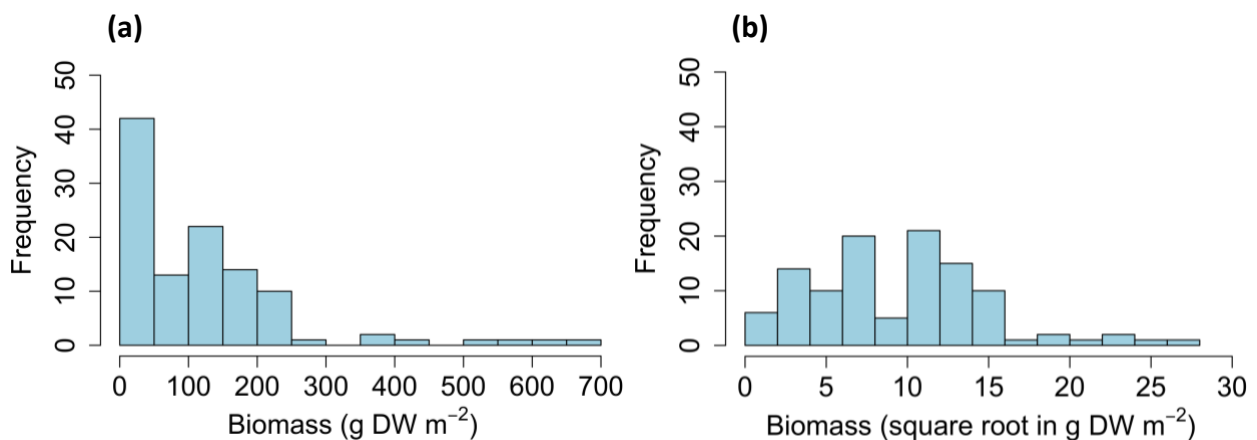
$$Biomass_{estimate} = (0.148 * Cover) + 4.129$$

(Eq. 2)



**Fig. 1.7** Biomass (mean g m<sup>-2</sup> transect<sup>-1</sup>) predicted by cover (mean transect<sup>-1</sup>) model using a biexponential five polynomial equation (a),  $r^2 = 0.619$  and a simple linear regression (b)  $r^2 = 0.595$ ,  $p < 0.0001$ .

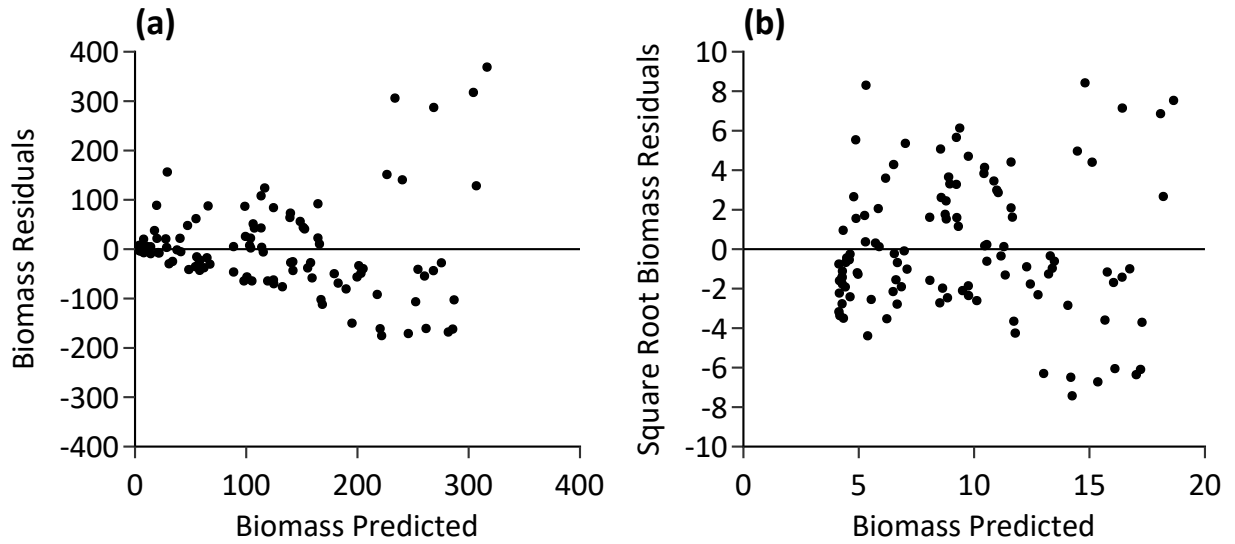
Biomass was used in its raw form for the 5P and square root transformed for the linear model. Raw biomass values were used to calculate the 5P model and not transformed in effort to preserve the natural curve and asymptotic relationship between cover and biomass (Fig 1.7a). However, the raw biomass values did not fulfill the assumption of normally distributed data with clustered residuals, a high skewness score of 2.26, and a kurtosis score of 6.26 (Fig. 1.8a; Fig. 1.9a). Other exponential models were considered for the cover-biomass model, but were found to have poorer goodness of fit in their regressions, depressed mid-range biomass values, e.g., 50% cover < 100 g DW m<sup>-2</sup> eelgrass, or a high y-intercept which did not accurately describe the relationship between cover and biomass when eelgrass was absent or at very low cover levels.



**Fig. 1.8** Biomass distribution histograms for exponential (a) and linear (b) models of biomass predicted by percent cover. The exponential model uses raw biomass data while the linear model is for the same data after square root transformation.

The linear model was square root transformed and did have a normal distribution with skewness and kurtosis values less than one and randomly distributed residuals (skewness = 0.699, kurtosis = 0.629; Fig. 1.8b; Fig. 1.9b). The cover-biomass model (Fig 1.7b) demonstrates the positive linear relationship between square root transformed biomass and cover ( $r^2 = 0.595$ ,  $p$

< 0.0001). Biomass predictions calculated using the linear model need to be squared to reflect an accurate prediction of biomass before comparison to other sites.



**Fig. 1.9** Actual by predicted and residuals plots for exponential (a) and linear (b) models of biomass predicted by percent cover.



## Discussion

### *4.1. Efficacy of video monitoring*

Videography increases opportunities for monitoring eelgrass. Monitoring sites that were previously only sampled at low tide are able to be monitored throughout the tidal cycle using video—increasing the timeframe and frequency at which sampling can occur. Year-round observations in colder waters are also possible with video monitoring. Eelgrass populations are often perennial at higher latitude sites but are generally unsafe or too difficult to sample during the winter and shoulder seasons. The inability to sample during cold weather months severely limits our knowledge of eelgrass beds in frigid or ice-covered waters (Lalumière et al., 1994; Olesen et al., 2015). Video-monitoring through the ice or from a boat in the winter may not be as targeted as observations from established transect monitoring, but it could provide data to characterize eelgrass beds from these challenging higher latitude regions.

In order to collect high-resolution observations comparable to conventional monitoring for this study, video-monitoring required in-the-water sampling. Wading or snorkeling was necessary to set up transects. At deeper water sites where SCUBA was necessary for conventional monitoring, video-monitoring was possible by snorkeling, which was much less intensive and allowed for non-SCUBA certified team members to participate. When transect sites are permanently established and well-marked, boat based monitoring would be possible and likely as effective in targeting specific sites along a transect as observed by Schultz et al. (2011). Boat-based drift transects are another way to employ video-monitoring for scouting new observation locations, rapid ecological assessment surveys, or the assessment of eelgrass beds where transects are unrealistic or not appropriate.

#### *4.2. Advantages and challenges of using video monitoring*

Videography yields a number of advantages for monitoring eelgrass. Ocular estimates for eelgrass cover have been the standard in conventional monitoring. As an approximation based on an observer viewing images of eelgrass, they can introduce subjective bias to observations (Lyons et al., 2015; Morrison, 2016). In this study, ocular estimates of eelgrass cover may have been susceptible to bias with greater cover values potentially being overestimated: overestimation by observers has been documented in eelgrass observation (Reeves et al., 2007). The point-intercept method used in this study is an accepted method of seagrass cover estimation (McDonald et al., 2006), and may reduce the effect of observer bias on observations. It also has been shown to require a similar amount of time to ocular estimates based on terrestrial vegetation studies (Godínez-Alvarez et al., 2009). Point-intercept cover counts also increase the accuracy of cover observations. Ocular estimate observations are only accurate to within a 5% level (Neckles et al., 2012); point-intercept methods, based on the total number of possible intercepts, can increase the accuracy to 1% ( $N = 100$ ) or greater based on the number of intercepts used.

Repeatability is a critical aspect of scientific research: video monitoring can increase the repeatability of eelgrass field sampling analysis (Powers & Hampton, 2019). Videography still requires field observation but inherent to the method are the saved digital files of the sample sites. This method of capturing and saving video provides other researchers the material necessary to repeat the same analysis, eliminating the need for all researchers to be present in the field at the time of observation.

This video monitoring methodology creates an eelgrass cover image library with known percent cover values that could be used to develop machine learning algorithms for automating image observation and cover estimation. Point-intercept cover estimations for this study were

done manually, but previous terrestrial vegetation studies have used alternative computer processes that may be easily translated to machine learning (Dietz & Thomas, 1996; Stojanova et al., 2010). Automation of the cover observation process could greatly accelerate the rate of observation and increase the number of images observed, potentially increasing the accuracy of the methods by increasing the sample size and also for resolution by using a greater number of vertices in the point-intercept grid counts.

Additional eelgrass parameters not tested in this study are candidates for observation using video-monitoring. 1) Leaf width measurements could be assessed by reviewing video frames or using image processing software to calculate the pixel diameter of eelgrass leaves. Where the leaves cross the reference disk the selection tool can be used to measure the pixel width of a leaf and then converted to mm using the pixel- and actual diameter of the reference disk. This would provide data necessary to calculate the leaf area index (LAI) for the quadrat. 2) Reproductive shoots are also identifiable in quadrats and easily counted to determine abundance and reproductive potential. 3) Disease presence and percent cover may be estimated from blackened leaves and used to determine the wasting index (D. M. Burdick et al., 1993). Environmental conditions can also be observed from video such as water clarity and color, substrate type, species composition, and grazing damage.

Videography has its limitations for monitoring eelgrass. Low water clarity conditions that obscure beds for visual assessment similarly limit the effectiveness of video. Deeper sites greater than 5 m in depth may require SCUBA for assessment, which reduces the minimal training and cost associated with video monitoring. Upper subtidal sites with less than 0.5 m water at low tide also make assessing eelgrass density and height challenging. The shallow water depth combined with mature plants blocks the camera from filming down through the bed and when a second

video is recorded at a lower height below the canopy, the area is often too shaded or densely vegetated for observations.

#### *4.3. Modelling biomass from cover*

In this study, the two predictive models describe different aspects of the relationship between eelgrass cover and biomass. Eelgrass cover naturally has an asymptote of 100%, which is the maximum value for this variable. Biomass does not have a defined maximum value and will continue to increase as eelgrass cover approaches its limit, so an exponential model is an intuitive fit. However, with eelgrass beds threatened and in decline worldwide it is difficult to find abundant high biomass, high cover beds to sample, which reduced the predictive range of the model. The raw biomass data we worked with was skewed towards lower values which violates the assumption of a normal distribution for regression analysis and prevents the exponential regression from being the best model available.

Previous studies have log transformed biomass to normalize the distribution and improve the model fit (Carstensen et al., 2016). We considered this strategy, but when results were back transformed the range of the model was significantly decreased to a maximum biomass prediction of 244 g DW m<sup>-2</sup>. Even with the more moderate effect of a square root transformation, the prediction range is limited in the simple linear model to 400 g DW m<sup>-2</sup>. This same predictive range limitation has also been observed in other seagrass cover-biomass models (Congdon et al., 2017) and limits the utility of these model for predicting higher biomass values. If more data for high biomass, high cover sites were available it would likely normalize the distribution for biomass and improve the exponential model; this would require a reevaluation of both models to determine which should be used for predicting eelgrass biomass. The model also has a positive

y-intercept, i.e., at zero cover the value for biomass is positive. This is improbable and may limit the accuracy of extreme low cover observations.

Eelgrass beds that are less affected by disturbance or are limited by light availability may reach maximum biomass. Generally peak biomass is a brief period during the growing season (Krause-Jensen et al., 2000), which may make these observations less common in year-round data sets. However, eelgrass beds in North America often demonstrate long periods of maximum biomass during the summer reaching average values of  $< 200 \text{ g DW m}^{-2}$  (Short et al., 1989)

Higher biomass beds ( $600 \text{ g DW m}^{-2}$  or greater), although less common in the data used in this model, were still present, and should be considered unless previous information on the bed or population justifies otherwise. Other studies of eelgrass biomass in North America and Europe suggest that a model that predicts up to  $600 \text{ g DW m}^{-2}$  will be able to predict biomass for the majority of eelgrass beds throughout its range in the Atlantic and Pacific oceans (Clausen et al., 2014). Incorporating more observations with greater biomass values ( $>300 \text{ g DW m}^{-2}$ ) could improve the normality of the distribution and the accuracy of the upper limits of this model and is an area for future research.

## **Conclusion**

The product of this study is a tested methodology for non-destructive video-monitoring of baseline eelgrass parameters. Video monitoring provided similar results compared to conventional methods for eelgrass percent cover, shoot density, and plant height and required less training and fewer participants to conduct the monitoring. Additionally, the cover-biomass model provides a framework for estimating eelgrass biomass when cores or field samples cannot be collected. Future work could focus on automating eelgrass cover analysis, observing additional eelgrass parameters, and identifying and quantifying environmental conditions from video.

## CHAPTER II:

### An Index for the Assessment of Eelgrass Health: Video Monitoring in James Bay, Québec

#### **Abstract**

Indices are a powerful method for ecological assessment and safeguarding ecosystem health. Eelgrass (*Zostera marina* L), or Sishkabash, has steeply declined throughout much of James Bay since the 1980s, threatening the traditional goose-hunting culture of the First Nation Cree communities of northern Québec, Canada. To assess eelgrass health in James Bay an eelgrass health index (EHI) was developed based on underwater video-monitoring observations. EHI ratings for sites in James Bay and the United States were validated using both biomass measurements and a survey of expert knowledge in order to demonstrate the usefulness of the EHI and engage potential users of the index. EHI ratings were consistent with both validation methods, indicating that the index is applicable to eelgrass populations beyond James Bay. Based on our application of the EHI in James Bay, data from the larger Niskamoon eelgrass study, and comparison to previous data, we find that eelgrass is impaired throughout most of eastern James Bay and persists in a relatively healthy state consistent with historic conditions only in isolated areas.

## Introduction

### 1.1 Study Motivation

It's late August in James Bay, Québec—near peak growing season for eelgrass—and, even after a whole day of searching, there is no eelgrass to be found in the bay. The Cree have asked us, Dr. Fred Short, a seagrass expert, and his team, to help understand why eelgrass has declined in traditional coastal hunting grounds. Eelgrass (*Zostera marina* L.) is a cosmopolitan species of seagrass inhabiting northern circumpolar estuaries and shallow coastal waters (Short et al. 2007). In James Bay, Québec, eelgrass beds historically covered an estimated area of 250 km<sup>2</sup>: one of the largest expanses in North America (Lalumière et al. 1994). The eelgrass beds of James Bay provide critical habitat for waterbirds and fish, such as anadromous whitefish, cisco, and sea-run brook trout, spawning habitat for Greenland cod (Ganter, 2000, Morin et al. 1991, Morin et al. 1980), and essential forage for Brant (*Branta bernicula hrota*) and Canada geese (*Branta canadensis interior*) during the spring and fall migrations (Dignard et al., 1991; Ganter, 2000).

As a Cree elder describes, once the bay was full of eelgrass. The geese that rely on the subtidal eelgrass meadows and which the coastal First Nation Cree communities of eastern James Bay revere as a food source, were everywhere during the spring and fall migration. Now, in Québec, the indigenous Cree people have observed the decline of eelgrass or Sishkabash (Short, 2008; Peloquin & Berkes; 2009; Dickey, 2015) The eelgrass is gone, hunting and fishing activities have diminished, and Cree culture is threatened. Most of the eelgrass beds we have observed throughout the summer monitoring from southern Waskaganish nation to northern Chisasibi near Hudson Bay have been sparse or fouled by algae. The eelgrass beds in only a couple isolated bays resemble the expansive historic meadows that Cree elders and trappers



describe. The applied question is important, but also raises interesting methodological challenges. How should eelgrass be assessed over the extensive Québec coastline of James Bay with only the summer months available for monitoring? How should the different eelgrass characteristics observed be translated into a meaningful value that allows for comparison over space and across time? The request from the Cree Nations of James Bay to better understand observed eelgrass decline and the associated methodological questions motivate this research.

### *1.2. Eelgrass and James Bay*

The decline of eelgrass in James Bay is not an isolated occurrence. Seagrass species are threatened worldwide by myriad issues and eelgrass as the dominant circumpolar species of seagrass at northern latitudes is no exception (Short et al., 2007). Historic eelgrass die-offs in the North Atlantic were caused by wasting disease and a marine slime mold (*Labyrinthula zosterae*) in the 1930s and along the US eastern seaboard again in the 1980s (Muehlstein et al., 1991). Habitat destruction and impairment from coastal development, dredging, and chain and anchor dragging are a persistent threats (Short and Wyllie-Echeverria, 1996), as well as water pollution and eutrophication, which reduce light penetration and limit the photosynthetic productivity of eelgrass (Orth et al., 2006). Climate change also threatens temperate and arctic seagrass species. Increasing occurrences of extreme weather and warming ocean temperatures can destroy eelgrass beds or cause sublethal stress when water temperatures approach a species' thermal limit (Short and Neckles, 1999; Niu et al., 2012). Invasive species are also a threat to seagrasses through both competition and predation (Williams, 2007).

The James Bay region has undergone major changes in the last half century, which may contribute to eelgrass decline. Hydro-development has altered the flow regimes of the La Grande

and other major rivers in the region (Déry et al., 2018; Dynesius & Nilsson, 1994). The re-location of coastal Cree communities has created concentrated urban settlements located at the mouths of major rivers like the Rupert, Eastmain, and La Grande (Niezen, 1993). Wastewater effluent from the substantial coastal development can impair nearshore water quality. Furthermore, upstream resource extraction from interior forestry and mining may negatively affect downstream coastal water quality.

Previous eelgrass studies in James Bay (Lalumière & Lemieux, 1995; Short, 2008) have suggested different causes for the eelgrass decline, but have not clearly linked the possible causes to eelgrass decline. Lalumière and Lemieux (1995) proposed that isostatic rebound of coastal lands, climate change, and wasting disease were primary causes. In contrast, Short (2008) posited that regional hydro-development caused eelgrass decline by reducing coastal water clarity and salinity and increasing water temperatures.

In the dynamic and contested system of James Bay, a rapid, accurate, and efficient method for assessing eelgrass health is needed. An ecological index to assess eelgrass health will provide a tool to help identify threats and stressors affecting eelgrass decline.

### *1.3. Ecological indices*

Environmental and biotic indices are methods for assessing ecological status (Martínez-Crego et al. 2010). The synthesis of data through an index provides accessible values or ratings for research and management practices, and for communicating ecological status to a broader audience (Ebert & Welsch 2004; Shin & Shannon, 2010). Indices are used for a broad range of purposes across ecological fields, from characterizing species diversity (Shannon and Simpson diversity indices) to analyzing satellite imagery of vegetation (NDVI). Specific to marine

environments, many indices exist for the assessment of anthropogenic effects on coastal habitats and species. While indices provide a framework for synthesizing physical, chemical and biological parameters to a standardized and representative value, they can be challenging to use, often requiring significant training, data, and time.

### *1.3.1. The structure of an index*

Seagrass indices share three key elements: careful selection of plant and environmental metrics, defined reference values, and a formula for calculating final index ratings. Seagrass indices incorporate chemical, physical and biological characteristics of seagrasses and their environment (Table 2.1). However, the focus has generally been on meadow level parameters such as areal coverage, abundance, biomass, and the number of taxa present. Index reference values are the greatest or “best” possible conditions defined for a specific parameter. These values can be hypothetical conditions, the “best” or maximal observed condition from field observations, or the mean of a sample of the highest parameter observations. Reference values used in index calculation should be based on the best available data and provide a method for standardizing values by canceling out units. Standardization of values is one method for calculating an index. More often, an equation is used to combine multiple metrics. The arithmetic mean, weighted combination rule, and Euclidean distance formulas have all been used to combine metrics into a final index rating (Table 2.1).

**Table 2.1** Previous indices designed to assess ecological quality of seagrasses and the associated metrics and equations. Nations are listed using their ISO 3166-1 alpha-3 code.

<b>Authors and year</b>	<b>Index name and location</b>	<b>Genera and species</b>	<b>Environmental characteristics included</b>	<b>Plant-specific characteristics included</b>	<b>Equation description</b>
Burdick, Short, and Wolf. 1993	Wasting Index New England, USA	1	No	Shoot dimensions, leaves per shoot, wasting disease leaf cover	Leaf area index
Lee, Short, and Burdick. 2004	Nutrient Pollution Indicator (NPI) New England, USA	1	Yes	Percent leaf N, leaf mass	Ratio of percent N to leaf mass
Lopez y Rojo et al. 2010	BiPo W. Mediterranean basin, DZA, ESP, FRA, ITA, MLT, TUN	1	Yes	Shoot leaf surface area, length, and shoot density	Arithmetic mean, reference values and weighting, 0–1 scale
Neto, Barosso, and Barría. 2013	Seagrass Quality Index (SQI) Modego Estuary, PRT	2	No	Number of taxa, bed extent, and shoot density	Combination rule equation, reference values, 0–1 scale
Irving, Tanner, and Haylard. 2013	Habitat Structure Index (HSI) Gulf St. Vincent, AUS	4	No	Area, continuity, proximity, percent cover, and taxa present	Euclidean distance, reference values and weighting, 0–1 scale, Scalar used (0.422)
García-Marín et al. 2013	ZoNI Gulf of Cadiz, ESP and PRT	1	No	Percent cover, shoot density, biomass (total, above-, and belowground), biomass ratio, leaf length, carbohydrates, and leaf nitrogen content	PCA analysis to ID C1 reference values used
Karamfilov, Berov, and Panayotidis, 2019	ZnoPI Black Sea BGR	1	No	Shoot density, biomass (total, above-, and belowground), biomass ratio, leaf length, and leaf area)	PCA analysis

### 1.3.2. Review of seagrass indices

Efficient multi-parameter eelgrass indices exist for assessing specific eelgrass conditions. The wasting index (WI) was developed solely to evaluate the extent of wasting disease in eelgrass beds (D. M. Burdick et al., 1993), and the nutrient pollution index (NPI) to assess nitrogen enrichment (Lee et al., 2004). Both indices are effective for investigating these specific conditions and for characterizing the extent of disease or eutrophication in an eelgrass bed. These indices do not characterize the broader status and health of eelgrass independent from these conditions, making them inappropriate for assessing base-level eelgrass health.

Seagrass indices have also been developed to assess environmental status using seagrass morphometrics and abundance data. BiPo, an index developed by Lopez y Rojo et al. (2010) assessed anthropogenic pressure on Neptune grass (*Posidonia oceanica*) meadow health using both environmental and population-specific parameters (Table 2.1). BiPo is an effective index for assessing *Posidonia* but not for eelgrass. It relies on environmental data (depth limit), which could create a circular feedback loop if used to assess environmental stressors, negating its utility for exploring light limitation and other stressors. Irving et al. (2013) developed the habitat structure index (HSI) using video sampling. Designed for Australian seagrasses, the HSI used taxa present and areal coverage characteristics of seagrass beds to determine ratings (Table 2.1). The HSI assigns higher ratings for multi-species beds and is not suited for assessing seagrass populations where monospecific beds are standard.

Specific to the *Zostera* genus, three indices have been created to assess dwarf eelgrass health (*Z. noltii*). The Seagrass Quality Index (SQI) incorporated taxa present, areal coverage, and density as metrics for comparison to anthropogenic indicators and stressors (Table 2.1), keeping environmental conditions independent of the index for stressor analysis (Neto et al.

(2013). Another index, ZoNI, was designed using a multiple data sets (N = 9) of population, individual, and physiological metrics (Table 2.1), (García-Marín et al. (2013)). And, ZnoPI, a third index for dwarf eelgrass similar to ZoNI, evaluated parameters by PCA analysis before calculating a rating (Karamfilov et al., 2019). Dwarf eelgrass does require its own indices, however, in translation the latter two indices are difficult to apply due to their extensive data requirements and because all three indices were designed for a different species inhabiting the upper intertidal areas of Europe (Borum et al., 2004).

As, “[the] mostly widely distributed marine angiosperm in the northern hemisphere,” an index specific to eelgrass health assessment is a broadly applicable tool (Green and Short, 2003; Krause-Jensen et al., 2005). However, none of the previously reviewed indices were designed specifically for or using North American eelgrass populations and few used methods that make monitoring accessible to a broader community. No suitable index exists to rate the health of eelgrass solely using simple nondestructive monitoring methods.

In summary, the seagrass indices previously described are limited in their efficiency and effectiveness for rating eelgrass health because they (1) were not designed specifically to assess *Z. marina*, (2) produced ratings that used the number of seagrass taxa present as a metric, (3) are too specific in their scope, (4) require extensive time and effort in sampling to collect the required data, and (5) include environmental data in their rating calculation.

#### *1.4 Study objectives*

The aim of this study was to develop an index for assessing eelgrass health using non-destructive and accessible observation and analysis. Building on previous video-monitoring, e.g., Irving et al., 2013, we developed the Eelgrass Health Index (EHI) using underwater video to

assess the status of eelgrass beds. We used percent cover, shoot density, and plant height as simple metrics to represent eelgrass extent, abundance, and productivity, to calculate the EHI. We designed the EHI to be a versatile index where two or more parameters (e.g., cover and density) could be used to calculate a rating, making it adaptable to available data. We focused on developing and testing the EHI, and also applied the index to provide an assessment of the current eelgrass conditions in our case study in eastern James Bay.

## Methods

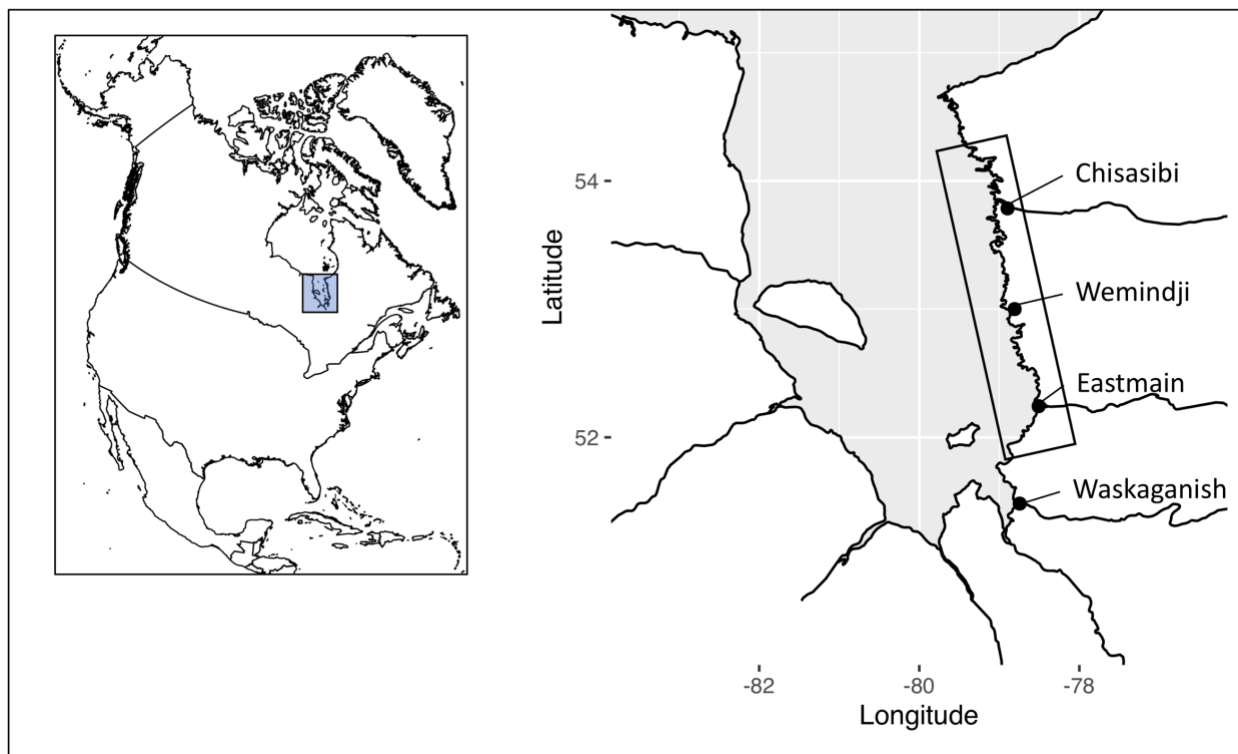
### *2.1. Study area*

James Bay forms the southernmost extent of the Hudson Bay estuary, covering a total area of 67,000 km<sup>2</sup> (El-Sabh and Koutitonsky, 1977). It is a large shallow basin (< 50 m), with high freshwater input and seasonal ice cover (Stewart & Lockhart, 2004). James Bay is bordered by Ontario to the west, Québec to the east, and Nunavut to the north. The Québec coastline runs approximately 360 km north to south and is characterized by numerous shallow bays, coastal islands, and river debouchments. Video-monitoring sites were located from N 51° 49' 31" to N 54° 17' 26" in James Bay (Fig. 2.1).

Eelgrass was monitored in the James Bay Cree nations of Chisasibi, Wemindji, Eastmain, and Waskaganish. The coastal waters of the Cree nations are divided into family hunting grounds or traplines. We generally monitored multiple sites (1–13, median = 3) within a single Cree trapline to observe the eelgrass conditions across the trapline. Sites within traplines were considered equivalent to a single transect in conventional monitoring and the still frames observed from the video, were analogous to quadrat observations. Sites were not delineated by distance but by eelgrass beds that were observed to be separated by geomorphological characteristics such as channels, islands, peninsulas, and embayments. This method did introduce some inherent subjectivity because of limited available recent mapping and ground-truthing efforts of eelgrass distribution across the region and water clarity for observation to verify the edges of eelgrass beds. Monitoring sites were selected based on Cree knowledge of historic eelgrass distribution, previous research conducted by Curtis (1975) and Lalumière (1994), and from a preliminary field assessment conducted in 2016 (F. Short, per com).



Researchers and Cree trappers, prior to monitoring, conducted a visual assessment of a site using an underwater viewing device (Aquascope). Visual assessment was used to identify the center of the bed or where the eelgrass coverage was most continuous and had the most mature plants. Video observations were taken from Cree motorized freighter canoes (Fig 2.2) during peak growing season (June–September) in 2017 and 2018, in collaboration with Cree subsistence hunters, called Trappers, and other James Bay community members.



**Fig. 2.1** Map of James Bay and the survey area in 2017 and 2018. The study area surveyed during the sampling season is the area within the black outline. Cree nations are labeled and denoted with black dots on the map.



**Fig. 2.2** Cree motorized canoe (made by Nor-West Canoe Company) used for observing eelgrass beds (Photo: N. Anderson).

## *2.2. Field videography and video observation*

Video-monitoring was conducted at multiple sites (1–13) within 24 of the 35 coastal James Bay traplines in Québec. Traplines were accessed with the permission and support of each trapline’s Tallyman (manager) and trappers. Traplines not included in the survey were omitted either because of difficulty accessing them due to their remote locations (extreme north and south regions or because of great distances from Cree nations, > 50 km) or lack of permission. Geographic coordinates for each monitoring site were recorded using an iPad synced with a GPS unit (Dual XGPS 160,  $\pm 2.5$  m accuracy) or using the GoPro camera’s built-in GPS system.

Initial measurements of depth, temperature, and salinity were recorded at each site using a YSI unit (63-25FT or EcoSense EC300A) and a handheld depth sounder (Vexilar). Water clarity was estimated using a white disk as a Secchi disk alternative and substrate type was identified from grab samples or visual observation by Aquascope. At sites with suitable water

clarity, eelgrass presence was determined by visual assessment and, at sites where water clarity was limited, eelgrass was observed from video post-field work. Sites where clarity was greatly impaired or where eelgrass was not observed and no video was collected were categorized as “unknown” for eelgrass presence.

Based on water clarity and eelgrass canopy height, a video camera (GoPro, Hero5 Black) attached to a 4 m extension pole was positioned at approximately 0.25 m, 0.5 m, or 1.0 m intervals from a bottom mounted white disk (~15.2 cm diameter) and illuminated by ambient light (Chapter 1). Video observations were recorded while motoring or drifting in a straight line at ambient speeds between 0.5 and 1 m s<sup>-1</sup> across the center of the eelgrass bed. One ‘drift-transect’ was conducted per site and observations were recorded and analyzed post-fieldwork.

### *2.3. Eelgrass video analysis*

Eelgrass characteristics were observed and quantified in the development of the EHI. Video observations were analyzed post-fieldwork and during the initial viewing of a site video, 10 unique frames representative of the center of the bed and most robust eelgrass were selected for analysis. Selected frames were copied and saved as individual still images before being saved in a composite file for analysis using image editing software (Adobe Photoshop Elements 10 and National Institute of Health’s ImageJ/Fiji version 2.0.0). If an image’s quality was poor due to low light conditions or turbidity, then adjustments were made to increase clarity using image enhancement tools such as the ‘levels,’ ‘contrast,’ and ‘color correction’ options.

Video was analyzed using eelgrass video monitoring methods (Chapter 1). Cover was estimated using the point-intercept method. Point intercept cover counts were conducted by overlaying a semi-transparent (25–50%) grid layer on the observation image in

Adobe Photoshop, and where grid vertices crossed eelgrass leaves in the frame the intercept was marked. Total marked intercepts for a site observation were then summed, divided by the total possible intercepts, and multiplied by 100 to calculate an estimated cover percentage (0–100%).

Eelgrass shoot density was estimated using two different methods: shoot counts and leaf counts. Visual counts of shoots were prioritized as an observation method since they more directly followed standard shoot observation methods by counting the number of shoots present within the frame (Short 2006) at sites where basal area was not obscured by low water clarity or high vegetative cover. For sites with high shoot density, shoots were counted from a subarea of the frame (0.5 or 0.25). The total shoots counted were then multiplied by a corresponding correction factor (2 or 4) based on the subarea sampled. Total shoots per frame were divided by the area of the frame to calculate shoot density (shoots  $m^{-2}$ ). The observation frame area was calculated using the known diameter of the white disk width in the frame and the proportional ratio of pixels for its diameter as determined using the selection tool. Using pixel height and width of the frame and this ratio equivalent metric values (m) were calculated and used to estimate the frame area. At sites where poor water clarity or canopy cover limited observation for shoot counts, leaf counts were used as an alternative method. Individual eelgrass leaves were counted where they overlapped with the upper half of the white disk visible in the frame. Total leaves counted per frame were divided by three, a conservative average for the total leaves per mature eelgrass shoot, and then divided by the white disk area (0.0182  $m^2$ ) to determine shoot density (shoots  $m^{-2}$ ). This provided an estimate for the number of plants present with the area of the disk used as the subarea sampled.

Eelgrass plant height was estimated based on the diameter of the white disk, camera distance from the substrate, and calculated frame dimensions. For shoots estimated at less than

0.5 m in height or flattened close to the substrate by a strong current, estimates were made at 5 cm increments using the known dimensions of the white disk, markings on the pole, and camera height. For shoots greater than 0.5 m, estimated canopy height and horizontal leaf length were used to calculate the hypotenuse, which was used as the estimated value for plant height. The hypotenuse was selected as a representative value accounting for the natural curve in eelgrass as it suspends in tidal and current driven marine environments.

#### *2.4. Eelgrass health index design*

Three different characteristics of eelgrass beds were included in the EHI: percent cover, shoot density, and mean plant height observations standard to eelgrass assessment. These characteristics were considered unique measures and equal representations of eelgrass status, so no weighting was applied to the variables in the equation. Reference values for each of these variables were used as a benchmark to standardize all eelgrass observations against the greatest commonly observed conditions. Reference values were defined by calculating the mean of the five highest observed values for characteristics with population-specific maximums, density and height, while percent cover always has a maximum limit of 100. Density and height values were used from across the entire study area in calculating the respective reference values.

Observations with values above the calculated reference value were removed from the analysis, so as to not produce standardized values greater than one.

EHI ratings were calculated using the geometric mean equation. The geometric mean was used to account for the range of variables that could encompass several magnitudes, e.g., shoot density: 1 to greater than 1000 shoots per m<sup>2</sup>, and for the inclusion of different but potentially correlated variables (McDonald, 2014). The EHI was calculated for each site by initially dividing

the estimated cover, density, and height of eelgrass plants observed in each frame by their respective reference values (Equation 1), then multiplying these three relative values together, and taking the cubed root of the product. EHI values for each frame per site were then averaged and this value used as the site-specific mean. From site mean values, averages were calculated by trapline and by nation to characterize eelgrass health at increasing spatial scales.

EHI ratings for each observation were averaged by site; site averages were used for trapline and nation average calculations along a south-north latitudinal gradient. Sites where eelgrass was absent were not included in the average trapline and nation EHI ratings as they would depress the overall rating as a zero value. Only observations from a 41-day period (July 23<sup>rd</sup> to September 2<sup>nd</sup>) were included in the analysis to limit the effect of the reduced-growing season on ratings across the sampling area.

$$EHI = \sqrt[N]{\frac{Cover_{obs}}{Cover_{ref}} * \frac{Density_{obs}}{Density_{ref}} * \frac{Height_{obs}}{Height_{ref}}} \quad (1)$$

Eq. 1 Site-specific eelgrass health index rating, where, cover is determined by observed cover divided by reference cover (100), observed density and height observations are divided by respective calculated reference values, and N (3) is the total number of variables being multiplied within the root equation.

## 2.5. Index validation

The EHI was validated by comparison with two methods of eelgrass assessment: biomass and an eelgrass health survey. Biomass is a well-established and broadly used method of assessing eelgrass and in general plant production, a measure of plant health (Roberts et al., 1993; Kirkman 1996). The eelgrass health survey was based on best professional judgement (BPJ) and to our knowledge has not previously been used to assess eelgrass health.

### *2.5.1. Validation using biomass*

In this study, we used aboveground biomass data collected from 18 sites in James Bay in 2018 as a comparative method of assessing the EHI. Biomass samples were collected from sites in the four Cree nations from August to the end of the sampling season in September. Samples were collected at the same time and from the same area in which eelgrass video monitoring was conducted for comparison. Eelgrass samples were collected using a garden cultivator and extension pole and stored in a cooler until processing. Additional biomass data from previous quarterly SeagrassNet sampling at established locations (N = 5) from New Hampshire (2), Massachusetts (1), and California (2) were compared to calculated corresponding EHI values. Reference values for U.S. sites were calculated using the same method from James Bay, based on the five greatest individual site maximum densities and height values for each specific site. U.S. biomass data came from multi-year monitoring projects observing three transects per location. U.S. eelgrass beds were included to represent populations separate from James Bay to assess the EHI for rating eelgrass health beyond the James Bay population (data used with permission from SeagrassNet). James Bay biomass samples were dried per standard procedure at 60 °C in a drying oven for 72 hours and then weighed using a Mettler Toledo/AB54S balance scale. Biomass and EHI observations for each quadrat and video frame were averaged by transect and site, respectively, for all locations and compared using simple linear regression.

### *2.5.2. Validation using the eelgrass health survey*

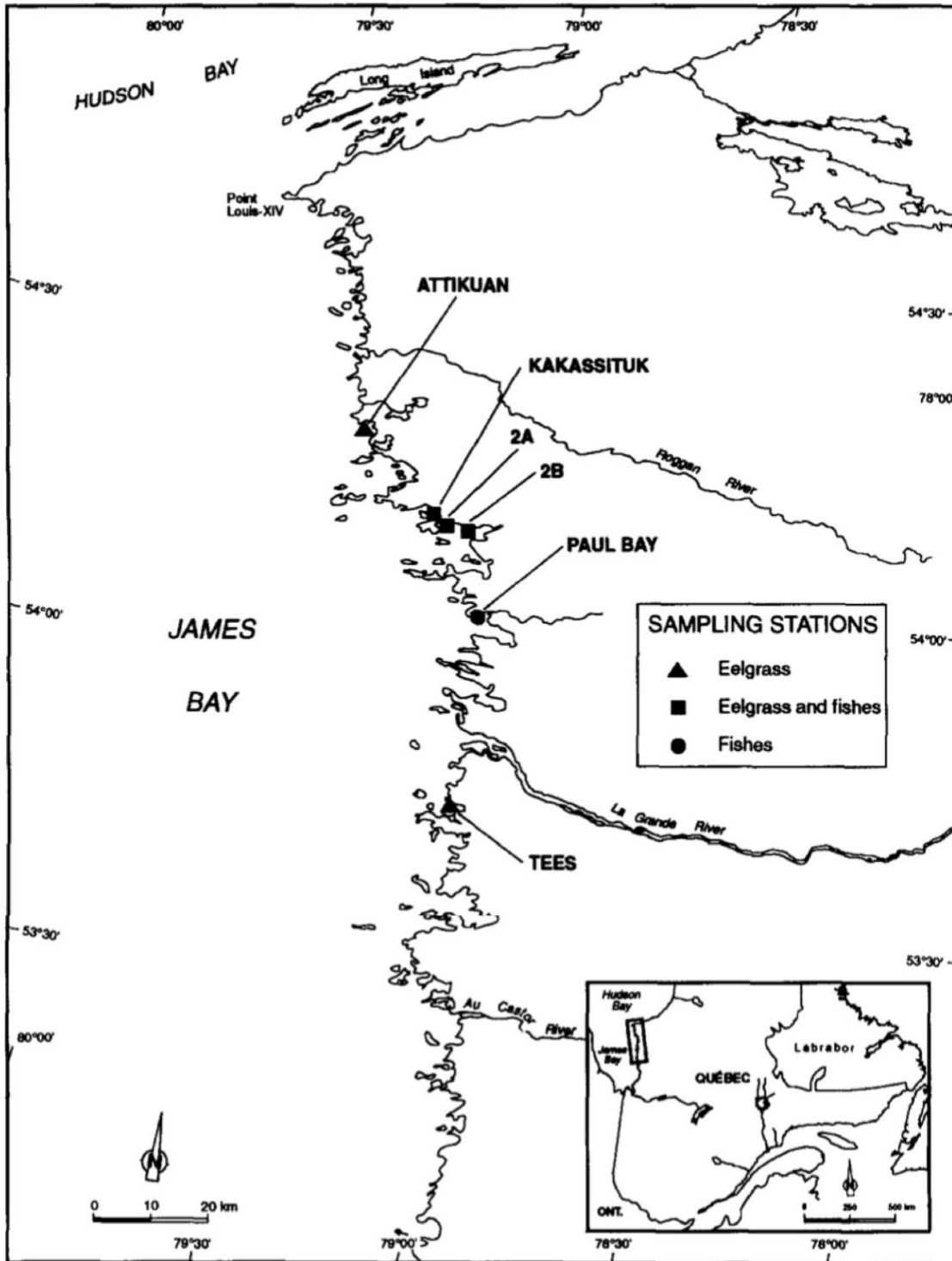
A second validation approach was piloted using an eelgrass health survey with individuals who had prior experience observing eelgrass (Anderson et al., 2020). Unfortunately, it was not possible to include Cree trappers in the survey, so a pilot survey was conducted with

researchers, resource managers, fishermen, and coastal educators from the Gulf of Maine coastal region (Appendix A). Thirty-six images were selected to represent a diverse range of eelgrass health conditions—20 from James Bay and 16 from the Great Bay Estuary (GBE; NH and ME). Images selected from outside of James Bay were included to increase the spatial and latitudinal extent of the survey by providing eelgrass and environmental conditions representative of its broader range. EHI values calculated for each image were assigned an associated Likert scale rating: 1–20 (Very Bad), 21–40 (Poor), 41–60 (Fair), 61–80 (Good), 81–100 (Excellent). Participants were asked to provide a short summary explaining the factors that influenced their rating of each image, and their responses were used to identify the contributing variables participants used in rating images. Participants also provided personal demographic information, e.g., level of education, profession, and years observing eelgrass (Appendix B). The survey was distributed to participants online using the Qualtrics Insight Platform. EHI ratings for James Bay and NH sites were normalized for analysis using the arcsine square root transformation and then compared to survey responses using regression analysis and a mixed effects model to account for the small sample size and variation between individuals.

## *2.6. Analysis of historic and current data*

Cover and height data from past research were not available for comparison. However, density and biomass values were collected at three sampling stations in Chisasibi's James Bay eelgrass beds from 1986 to the early 1991 (Fig 2.3; Lalumière et al., 1994). Shoot counts and biomass samples at each station were collected along five 20 m transects per station in the first half of August every year.





**Fig. 1.** Location of the study area and the sampling stations.

**Fig. 2.3** Historic sites sampled by Lalumière et al. (1994) between 1986 and 1991. Data collected at Attikuan, Kakassituk, and Tees Bay in this study was used model historic cover to calculate EHI ratings for comparison to contemporary EHI ratings. This figure was produced by Lalumière et al. (1994) for their study on eelgrass beds in James Bay.

Using 18 observations from the present study in James Bay and supplemental data (N = 111) from eelgrass monitoring conducted in New Hampshire, USA, we developed a model to estimate historic cover from biomass at three sites in James Bay: Attikuan, Kakassituk, and Tees Bay (Eq. 2). Raw biomass values had a right-skewed distribution, so to normalize the distribution a square root transformation was applied to biomass data prior to analysis (skewness = 0.698, kurtosis = 0.248). Using historic shoot densities and the modeled cover values derived from the historic data, we calculated EHI ratings and compared them to EHI ratings calculated using the same two variables in our present study to investigate eelgrass health over time in northern James Bay. EHI ratings were plotted through time by site and the mean of the historic observations compared to current values by percent change.

$$Cover_{estimate} = (m * biomass_{sqrt}) + b \quad (2)$$

Eq. 2. Eelgrass biomass-cover relationship modelled by simple linear regression, where:  $m$  is the slope and  $b$  is the y-intercept. Biomass was square root transformed prior to calculating estimated cover.

## 2.7. Statistical analysis

Statistical analysis was conducted using JMP Pro 14.3.0 (SAS Institute Inc.) and R. Figures were created in R and the maps made with the ‘ggplot2’, ‘maps’, ‘mapdata’, ‘RColorBrewer’, and ‘Rworldmap’ packages in R and river data was accessed from <https://www.naturalearthdata.com/>. ANOVA was used to test the difference between EHI ratings and eelgrass health survey ranking and for EHI Ratings between Cree traplines and Cree

Nations. Eelgrass health survey results were analyzed with a linear mixed effects model in R using Dr. Jonathan Lefcheck's 'piecewiseSEM' package.

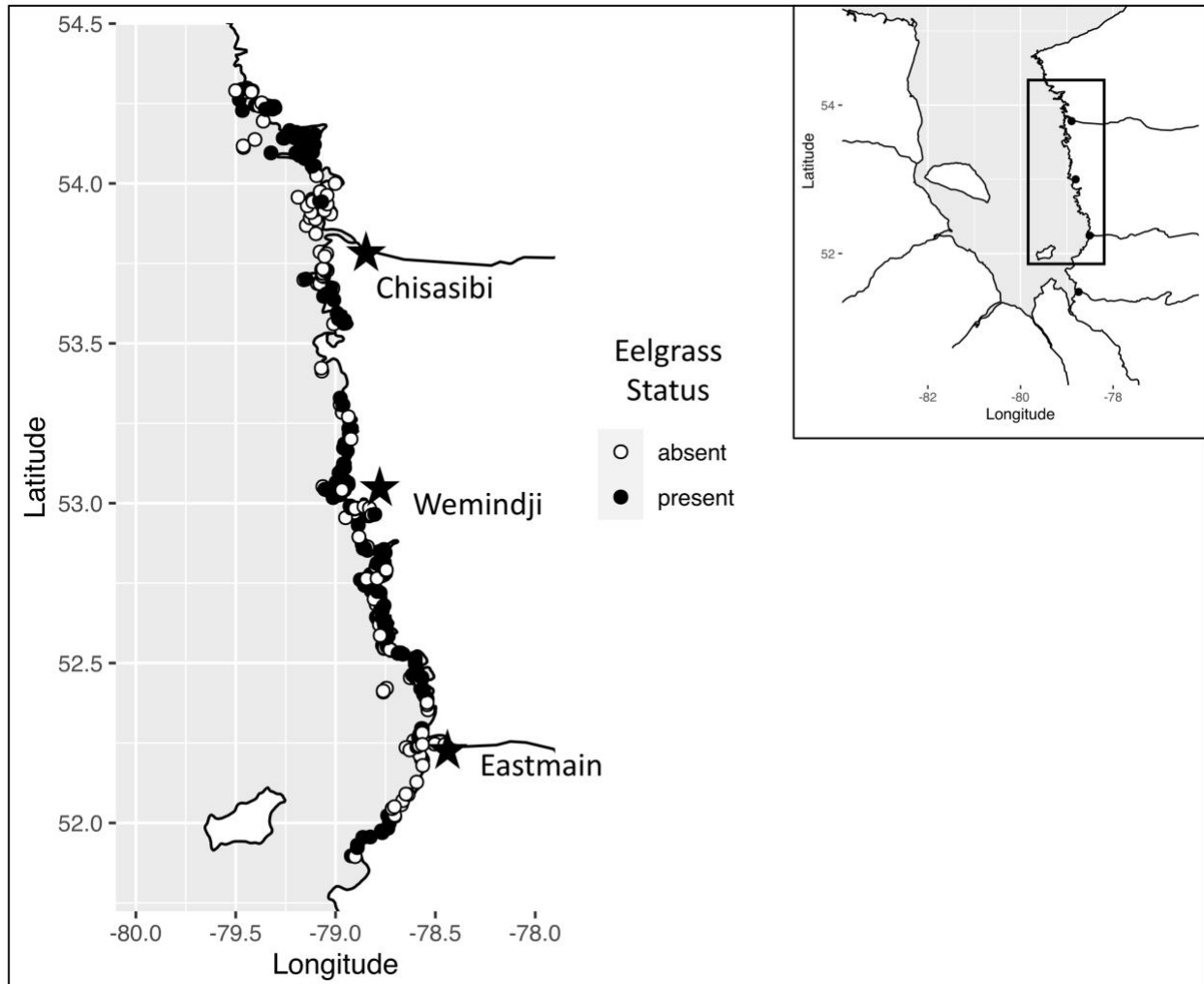
## Results

### 3.1. Overview

This section first demonstrates the validation results from both the biomass and health survey approaches. We then present index ratings for the various eelgrass observation sites. Then, the index is applied and compared to historic data to assess change in eelgrass health over time.

The EHI demonstrated a positive relationship for both biomass and the eelgrass health survey responses. Across the long-term biomass sites, EHI ratings reached maximum values at different percentages, indicating the need for EHI ratings to be standardized by site before comparison across different eelgrass populations. Overall, EHI ratings throughout James Bay skewed towards the lower end of the rating scale ( $mean = 27.2$ ) with high rated locations being rare. The EHI ratings agreed with Cree observations of eelgrass decline and the re-analysis of the Lalumière (1994) eelgrass data showed that historic sites at Attikuan and Tees Bay both had a significant decrease in EHI ratings, while at Kakassituk, EHI ratings were higher and had not significantly changed.

During the 2017 and 2018 field seasons, 371 sites were surveyed for eelgrass. Eelgrass was present at 230 sites, absent at 113 sites, and of unknown status due to poor observation conditions at 28 (Figure 2.4). Fifty-nine of the sites had video of eelgrass of acceptable quality for observation and were used to create the EHI.



**Fig. 2.4** James Bay, Canada from 50–56° N and 78–82° W. White points represent sites where eelgrass was absent; black points represent sites where eelgrass was present. Unknown sites are omitted from the map.

### 3.2. Eelgrass health index validation

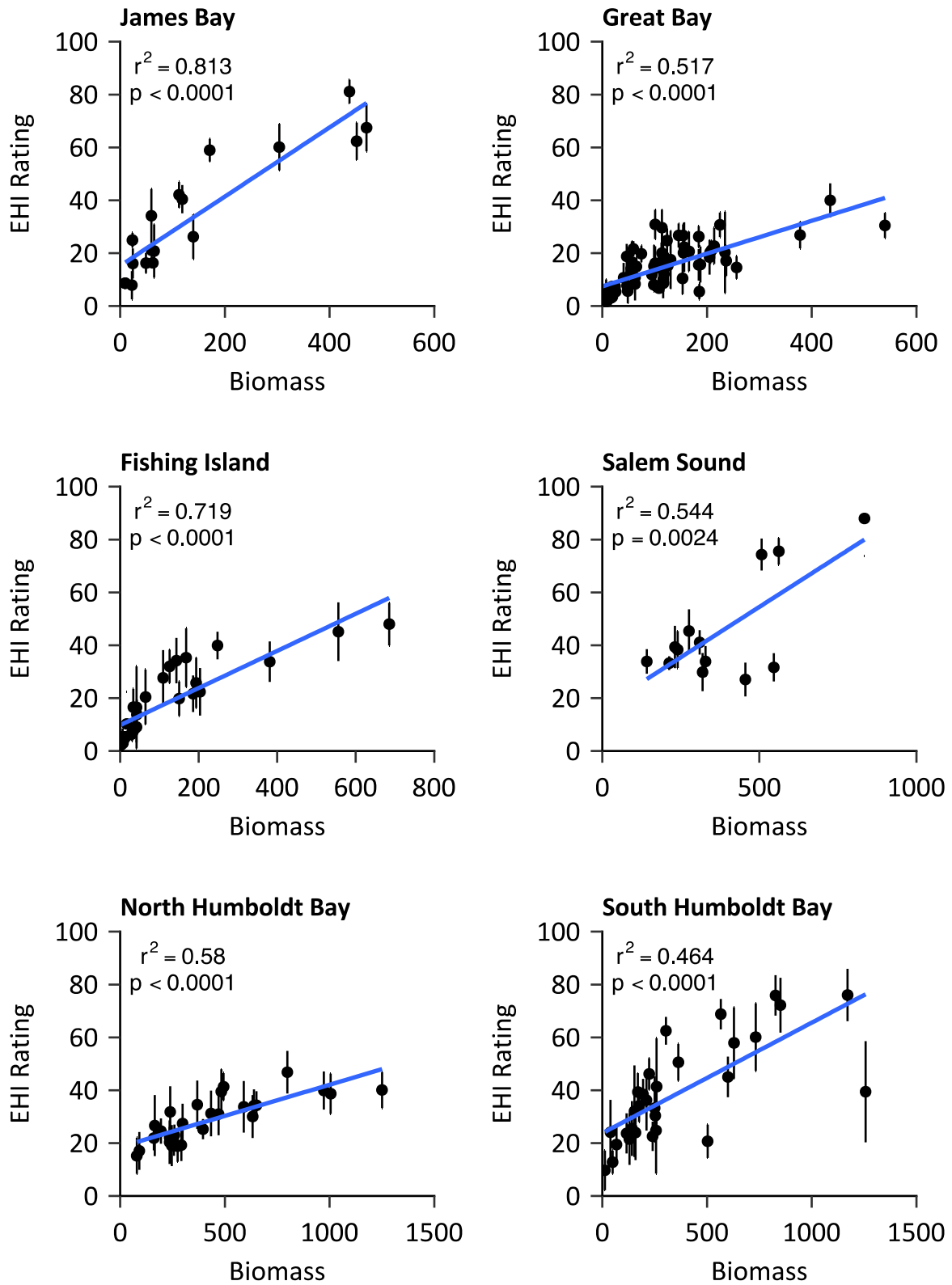
Biomass and EHI ratings were generally in agreement and followed a positive linear trend. Sites with low biomass received low EHI ratings, while sites with greater biomass received higher ratings. The EHI for James Bay demonstrated a strong positive relationship with biomass ( $r_2 = 0.813$ ,  $p < 0.0001$ ; Fig. 2.5.; Table 2.2.). This positive relationship between the EHI and biomass persisted at SeagrassNet sites in the US with a moderate to strong trend varying

by site. The lowest agreement in the model was at South Humboldt Bay, CA ( $r^2 = 0.464$ ,  $p < 0.0001$ ; Table 2.2.) and the highest at Fishing Island, ME ( $r^2 = 0.719$ ,  $p < 0.0001$ ; Table 2.2.).

**Table 2.2** Biomass site data and equations for Fig. 3.2. \*Low tide depth for James Bay is the average and S.E. of the 18 sites monitored. U.S. study areas each had three transects and each transect was considered a different site for comparison.

Site	Years	Sites	N	Low tide depth (m)	Equation
James Bay	2017–2018	18	18	*1.53 ± 0.13	0.131x + 16.26
Great Bay	2007–2018	3	65	0.5–1	0.062x + 7.44
Fishing Island	2002–2010	3	29	0.5	0.064x + 8.55
Salem Sound	2008–2012	3	13	1.0 <	0.061x + 18.24
Humboldt N	2007–2011	3	28	0	0.021x + 19.31
Humboldt S	2007–2011	3	30	0	0.038x + 23.90

Neither geographic location nor tidal depth appeared to affect the overall trend between models (Table 2.2.). However, the number of sample sites and length of sampling period may have. SeagrassNet biomass and eelgrass observations were sampled quarterly from three established transects at each site. James Bay sites were only sampled once, so every site visited was considered one transect, potentially introducing a wider range of conditions. James Bay had a higher diversity of possible conditions sampled across its sites and this may have improved the overall relationship between biomass and the EHI.



**Fig. 2.5** The relationship between biomass and the eelgrass health index ratings for James Bay and five SeagrassNet sites in the United States. Black lines represent standard error.

### *3.3. Eelgrass health survey results*

Nineteen individuals participated in the eelgrass health survey. Participants were from Maine, New Hampshire, and Massachusetts and came from a variety of academic and professional fields with a wide range of experience in eelgrass observation (Table 2.3). Approximately 50% of participants had previously observed eelgrass in the field using comparable in situ methods, wading or underwater observations. The majority (26%) of participants had 5–10 years observation experience, with 21% having less than 5 years, and all the groups with more than 10 years of experience making up the remaining 53% (Table 2.3).

Participant evaluations of eelgrass health were consistent with EHI. Eelgrass images assessed by participants as representing “good” or “excellent” eelgrass health had average EHI ratings of 60.51 and 72.99 (Table 2.4.), respectively, and were significantly greater than mean EHI ratings for other categories. In fact, all EHI means were significantly different across the rating categories (Figure 2.6). Although there were significant differences between the means, EHI ratings included in the survey only represented the lower end of the “Excellent” rating. No ratings greater than 86.8 were observed from James Bay or Great Bay for inclusion in the survey. This may have skewed the overall agreement between participant responses at higher levels and EHI ratings. It should be noted that both systems, James Bay and Great Bay, had degraded environmental conditions which stress eelgrass and are likely for the limited high EHI ratings (Short, 2008; Short, 2017, respectively).



**Table 2.3** Eelgrass health survey question and respondent characteristics (Eelgrass health survey, November 2019, N = 19). Eelgrass health question order randomized for participants.

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**Survey Questions**

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*Rating* — “How would you rate the health of the eelgrass in this frame?”  
*Rationale* — “Why did you select this rating? If you would like, please use a specific value (1 - 100) to rate the eelgrass and include that here. (optional)”

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**Participant characteristics**

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*Field* — “What is your profession?”  
 Academia/Research (63%)  
 Education (11%)  
 Management (16%)  
 Other (11%)

*Education* — “What is the highest level of school you have completed or degree you received?”  
 GED (5%)  
 Bachelor’s (11%)  
 Master’s (42%)  
 PhD (42%)

*Experience* — “How long have you been working with and/or observing eelgrass?”  
 Less than 5 years (21%)  
 5–10 years (26%)  
 10–20 years (21%)  
 20–30 years (21%)  
 More than 30 years (11%)

*Capacity* — “In what capacity do you have experience with eelgrass? (Check all that apply)”  
 Research (40%)  
 Management (26%)  
 Education (20%)  
 Recreation (11%)  
 Commercial (3%)

*Location* — “Where do you most often observe eelgrass?”  
 Maine (15%)  
 New Hampshire (55%)  
 Massachusetts (25%)

*Observation type* — “How have you observed eelgrass? (Check all that apply)”  
 By boat/above water (35%)  
 Underwater (25%)  
 Wading (23%)  
 Laboratory (15%)  
 Other (2%)

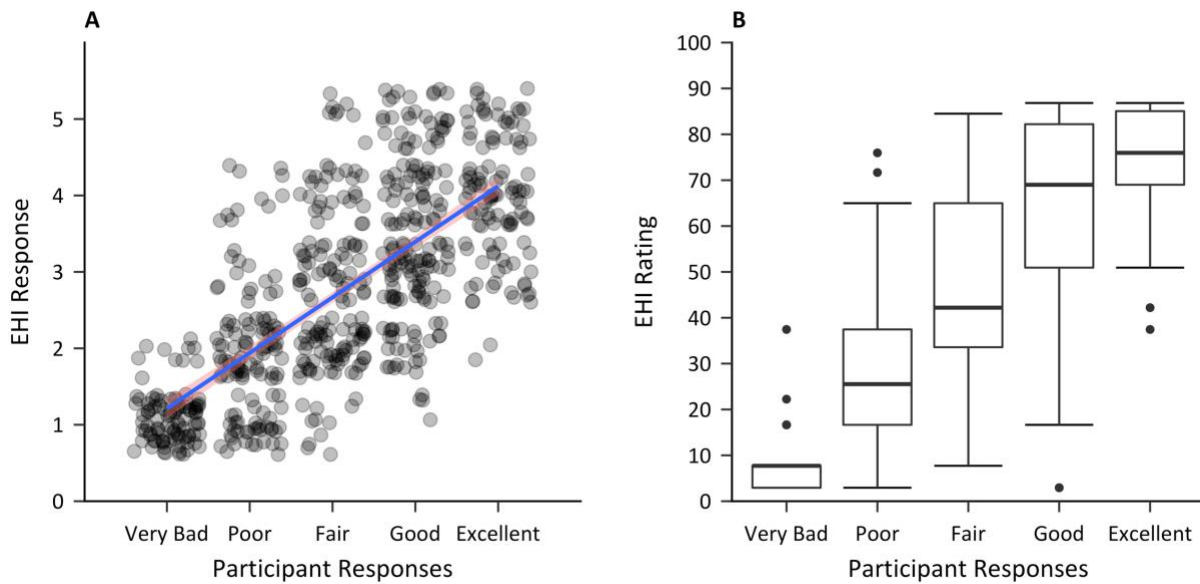
*Population Status* — “In this population, how would you describe the current eelgrass health status?”  
 Good (32%)  
 Fair (47%)  
 Poor (21%)

*Change* — “In this population, how would you describe eelgrass conditions as they have changed over the last 5 years?”  
 Improving (25%)  
 No Change (50%)  
 Declining (25%)

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**Table 2.4** Eelgrass health survey results with the number of responses by category, mean EHI level for all survey participants, standard error, and confidence intervals.

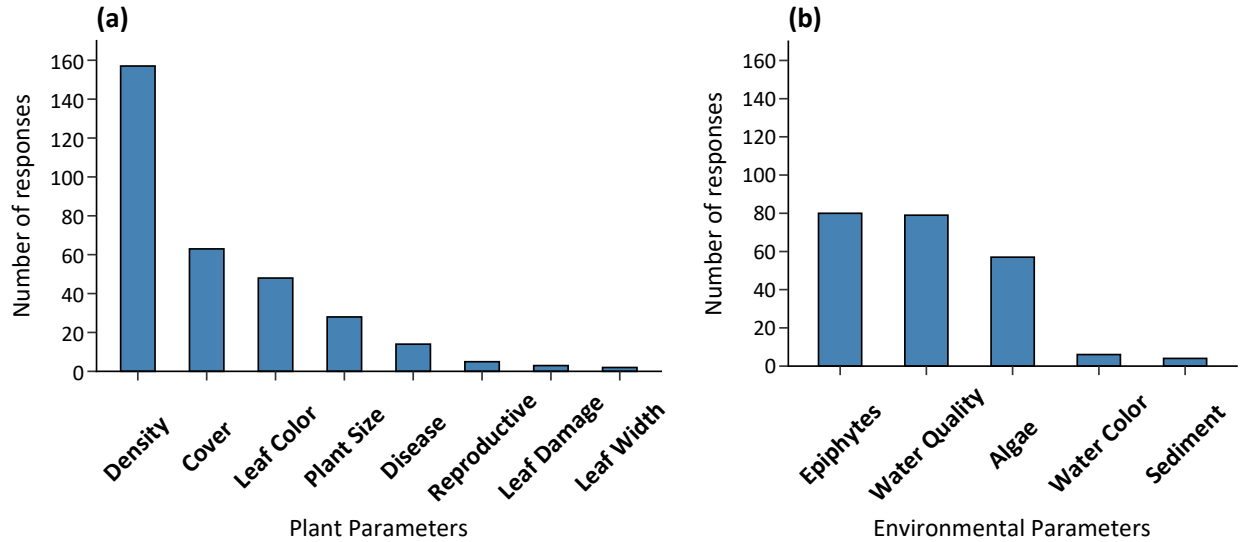
Level	Number	Mean	Std Error	Lower 95%	Upper 95%
Excellent	109	72.99	1.54	69.97	76.00
Good	169	60.51	1.23	58.09	62.93
Fair	148	43.96	1.32	41.38	46.55
Poor	127	28.13	1.42	25.34	30.92
Very Bad	98	11.62	1.62	8.44	14.80



**Fig. 2.6** Linear regression analysis of the eelgrass health index ratings and survey participant responses. Jitter was added to A to avoid overlapping of points. A) Index ratings are converted to values from 1 to 5, equivalent to intervals on the 1 to 100 index scale. Adjusted  $r^2 = 0.5959$ ,  $p < 0.0001$ . B) For the boxplot of responses, black dots indicate outliers (1.5 x IQR). Conditional  $r^2$  is approximately 0.70, accounting for fixed and random effects in the model.

In this study the choice of variables used to calculate the EHI was supported by the qualitative responses of the eelgrass health survey (Fig. 2.6). For eelgrass variables identified by survey participants, density had the greatest number of responses ( $N = 157$ ), cover the second most ( $N = 63$ ), and “plant.size” the fourth greatest number ( $N = 28$ ) (Fig. 2.6). Leaf color was ranked third of these four variables, but it was not included in the index because it can be affected by other variables such as disease, epiphytes, and water conditions. Leaf color was

likely selected by participants because blackening of leaves can be an indicator of necrotic tissue caused by wasting disease, a pathogen that negatively affects eelgrass and can cause mortality.



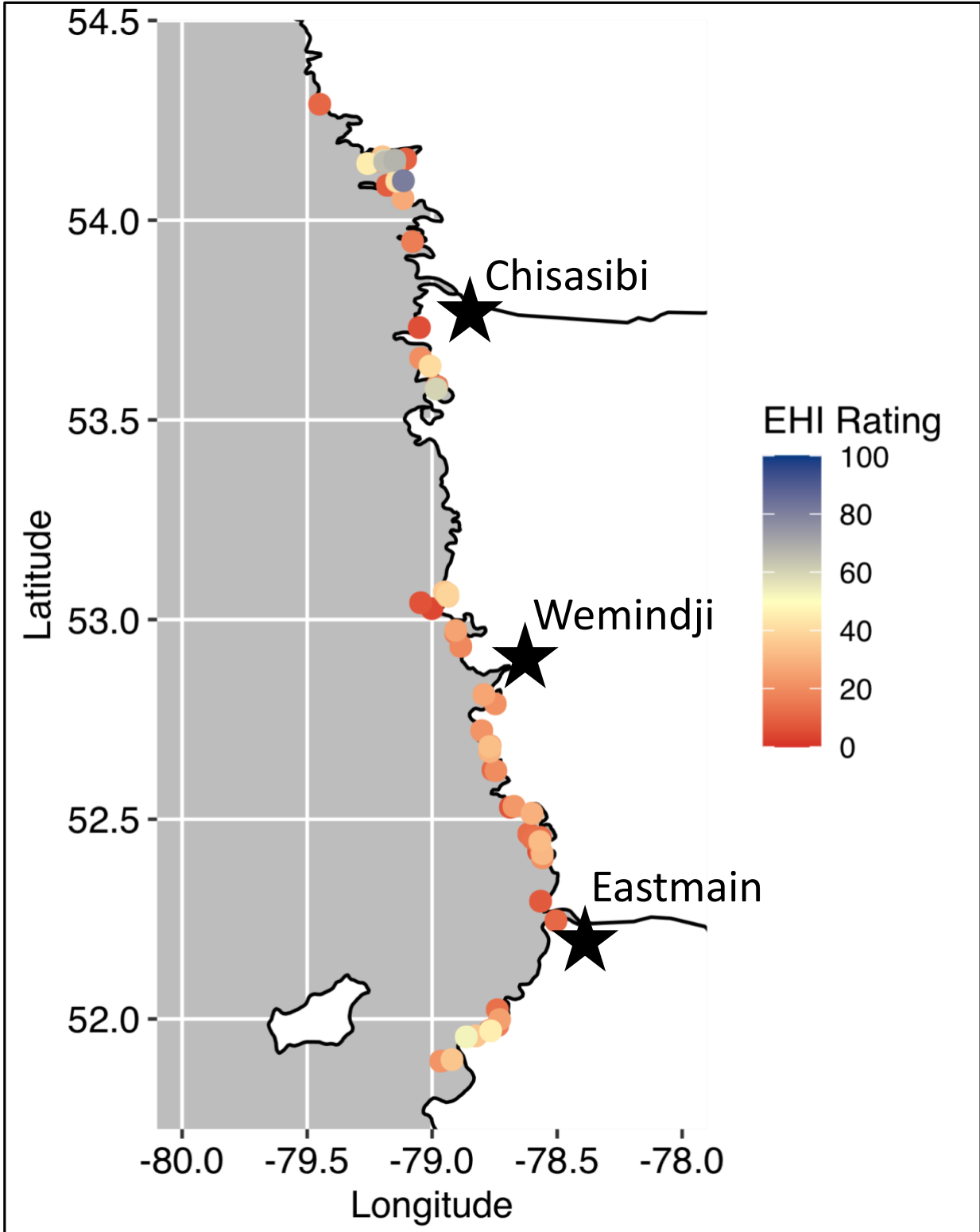
**Fig. 2.7** Eelgrass health survey qualitative responses. (a) Plant characteristics and the number of participant responses that cited them. (b) Environmental characteristics and the number of participant responses that cited them.

The participant responses also provided insight into what the responses may have been like if the Cree were surveyed. For example, a fisherman with extensive experience in New England, commented in the survey that they had never seen eelgrass in excellent condition (Anderson et al., 2020). This response mirrors observations by Cree trappers and community members in James Bay about declining eelgrass health (Peloquin & Berkes, 2009b).

In comparison to eelgrass observed today, Cree elders remember a much more healthy baseline from four decades ago where eelgrass was up to 2.5 m in height and beds were so dense they fouled outboard motors (Anderson, per com; Lalumière et al., 1994). The context provided by the fisherman and reflected in the Cree trappers' observations are important for calibrating contemporary eelgrass health assessment, where eelgrass that appears healthy today is considered impaired relative to historic conditions.

### 3.4. James Bay eelgrass health index

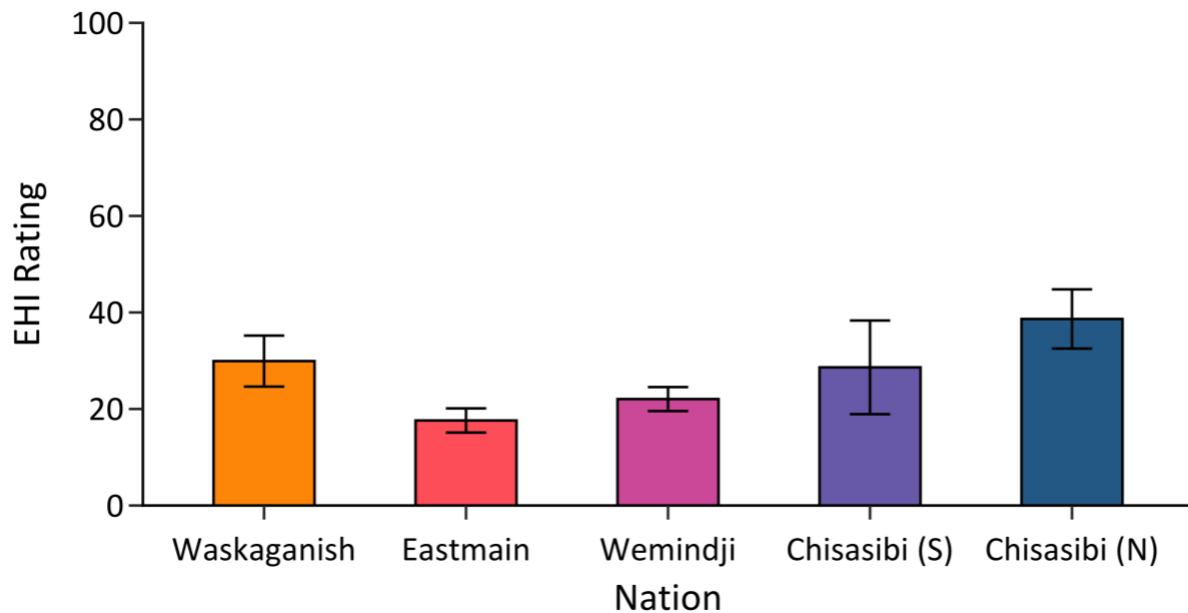
Eelgrass health conditions varied widely throughout eastern James Bay (Figs. 2.8, 2.9, 2.10). Sites with high EHI conditions were rare with a bay-wide mean  $26.9 \pm 2.29$  S.E and significant differences between EHI ratings and the different Cree nations. ( $r_2 = 0.21$ ,  $p = 0.0128$ ; Fig 2.9). For traplines, the relationship was close to the 0.05 threshold with  $p = 0.0873$ ,  $r_2 = 0.37$ , but would not be considered significant (Fig 2.10). Sites surveyed north of the La Grande River (CH04 in Chisasibi (N)) had the highest mean EHI ratings of the four Nations' coastal territories ( $38.6 \pm 4.19$  S.E.; Table 2.5). The general trend by nation was decreasing EHI ratings from north to south, with the median rating in Chisasibi (S) with an average of  $28.6 \pm 7.26$  S.E and Eastmain rating the lowest with an average of  $17.6 \pm 4.50$  S.E. The exception to the trend was the southernmost nation of Waskaganish, which had the second highest EHI rating for the five nations at  $29.9 \pm 5.74$  SE (Fig. 2.9; Table 2.5). Percent cover and height values followed the same north-south decreasing trend with Waskaganish continuing as an exception. Shoot density followed the same trend with one exception, Waskaganish had the highest density of the Cree Nations (shoots  $m^{-2}$ ),  $252.1 \pm 49.1$  S.E. followed by Chisasibi (N),  $214.5 \pm 35.9$  S.E. Overall, no nation received a mean EHI rating higher than 38.6 with a mean rating of 26.9.



**Fig. 2.8** Eastern coastal James Bay in Québec. EHI locations and ratings from 2017 and 2018 are marked: red points correspond with lower EHI ratings, yellow with mid-range ratings, and blue with higher EHI ratings.

**Table 2.5** Mean and standard error for observed percent cover, plant height (cm), and shoot density (shoots m<sup>-2</sup>), and mean EHI rating and standard error calculated for the Cree Nations of James Bay.

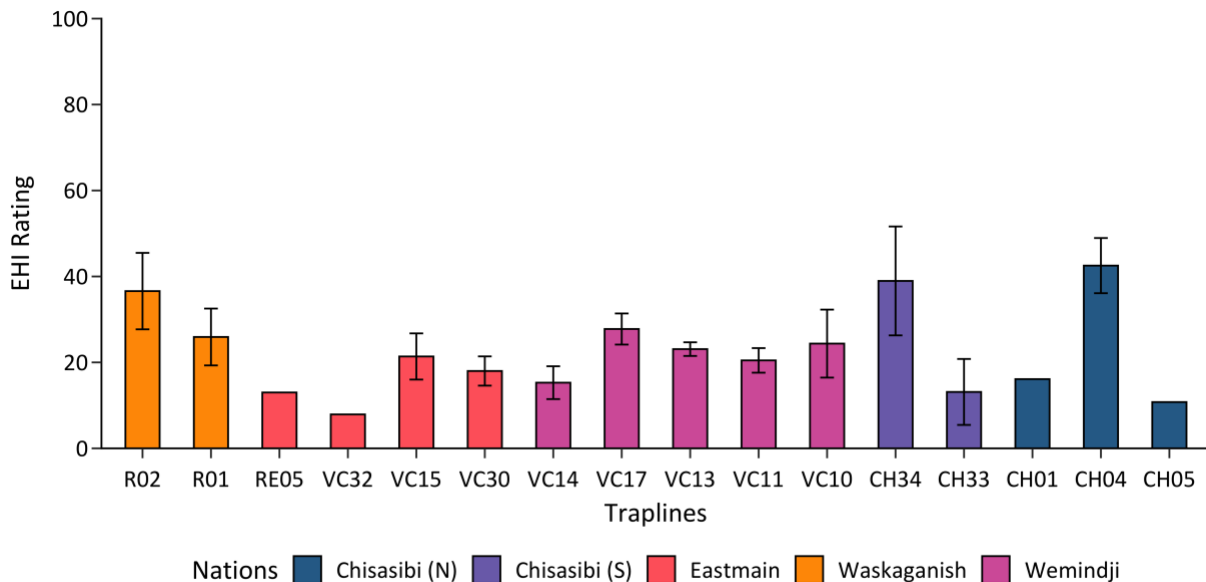
Nation	N	Percent Cover	Plant Height	Shoot Density	EHI Rating
Chisasibi (N)	15	48.6 ± 7.31	66.9 ± 9.57	214.5 ± 42.4	38.6 ± 6.14
Chisasibi (S)	5	34.4 ± 13.6	47.3 ± 12.2	206.6 ± 71.9	28.6 ± 9.70
Wemindji	18	23.9 ± 3.20	37.8 ± 2.89	157.2 ± 27.1	22.1 ± 2.48
Eastmain	13	16.4 ± 3.14	33.9 ± 3.80	135.8 ± 30.3	17.6 ± 2.51
Waskaganish	8	28.6 ± 5.94	49.7 ± 3.97	252.1 ± 59.1	29.9 ± 5.28



**Fig. 2.9** Mean eelgrass health rating by Cree nation. Bars show calculated ratings based on cover, density, and height, ( $r^2 = 0.242$ ,  $p < 0.0048$ ) and are oriented South to North from left to right. Error bars represent one standard error. N and S in x-axis labels indicate north and south of the La Grande River.

**Table 2.6** Mean and standard error for observed percent cover, plant height (cm), and shoot density (shoots m<sup>-2</sup>), and mean EHI rating and standard error calculated for the 19 traplines surveyed in James Bay where eelgrass was present. *p-values*: percent cover = 0.0002, plant height < 0.0001, shoot density = 0.1502, and EHI = 0.002.

Trapline	Nation	N	Percent Cover	Plant Height	Shoot Density	EHI Rating
R02	Waskaganish	3	36.6 ± 9.43	53.9 ± 6.96	288.3 ± 90.4	36.6 ± 8.9
R01	Waskaganish	5	23.7 ± 7.53	47.1 ± 5.03	230.4 ± 83.7	25.9 ± 6.62
RE05	Eastmain	1	13.3	47	39.9	13
VC32	Eastmain	1	4.6	41.7	28.4	7.9
VC30	Eastmain	8	16.03 ± 4.29	26.8 ± 3.04	171.3 ± 42.94	18 ± 3.4
VC15	Eastmain	3	22.5 ± 6.41	46.1 ± 10.4	108.9 ± 33	21.4 ± 5.37
VC14	Wemindji	4	16.3 ± 5.36	29.6 ± 2.29	97.4 ± 39.8	15.3 ± 3.82
VC17	Wemindji	3	26.6 ± 3.99	40.8 ± 5.25	216.7 ± 58	25.9 ± 3.19
VC13	Wemindji	3	25.7 ± 2.93	37.3 ± 5.93	156.8 ± 37.7	23.1 ± 1.59
VC11	Wemindji	3	22.2 ± 2.39	39.8 ± 8.6	112.3 ± 19.1	20 ± 2.08
VC10	Wemindji	5	27.5 ± 10.3	35.3 ± 7.4	191.2 ± 73.8	24.4 ± 7.9
CH34	Chisasibi (S)	3	44.4 ± 20.9	63.4 ± 11.75	289.8 ± 85.8	39 ± 12.7
CH33	Chisasibi (S)	2	19.3 ± 12.4	23.2 ± 9.88	81.9 ± 61	13.1 ± 7.58
CH01	Chisasibi (N)	1	10.94	44.6	82.7	16.1
CH04	Chisasibi (N)	13	54.03 ± 7.33	71.4 ± 10.5	239 ± 45.1	42.5 ± 6.42
CH05	Chisasibi (N)	1	16.3	31.3	26.5	10.8



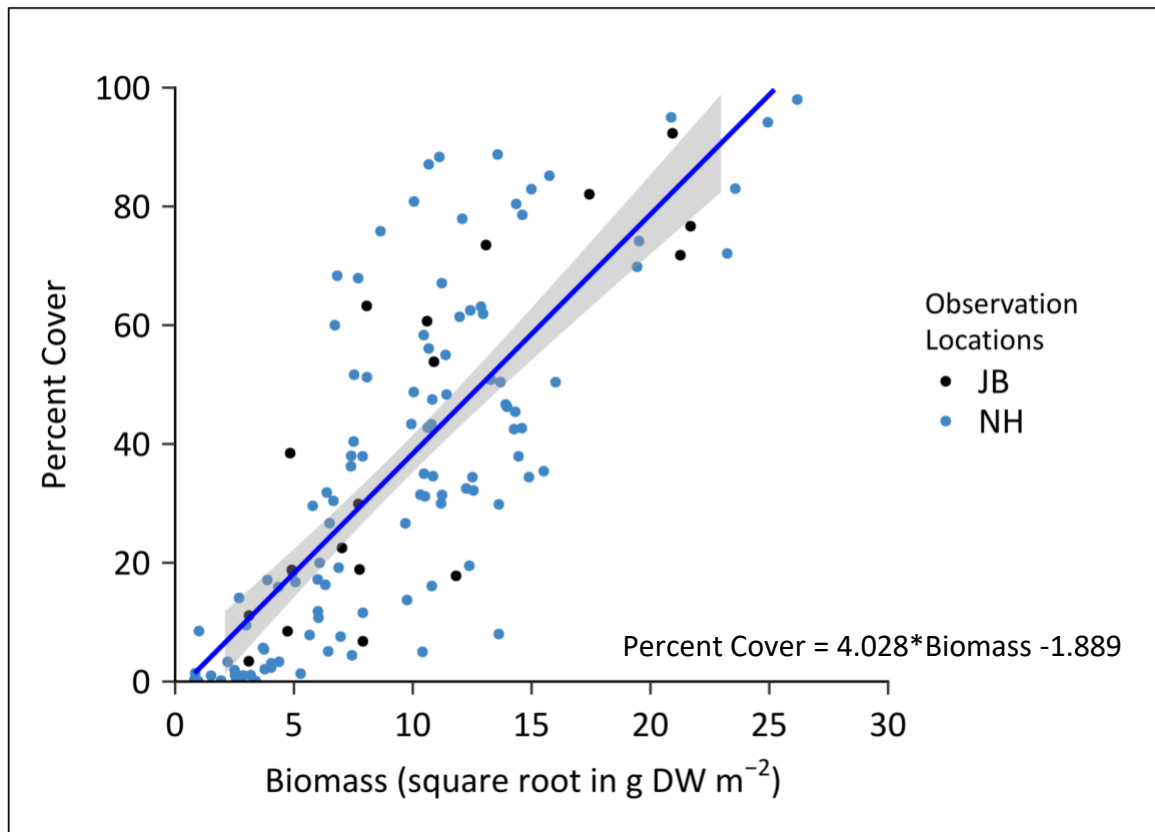
**Fig. 2.10** Eelgrass health rating by trapline. Bars show calculated rating based on cover, density, and height; ( $r^2 = 0.42$ ,  $p = 0.002$ ). Bars are oriented South to North from left to right. Error bars represent standard error.

Eelgrass beds in eastern James Bay—as determined by the EHI—are in an impaired state. The majority of eelgrass beds in coastal James Bay received low EHI ratings with the highest ratings found in isolated locations most often at the northern and southern extremes of the monitoring sites. These findings suggest no latitudinal trend or negative influence of warmer water temperatures in more southern regions or throughout the eelgrass range in James Bay, but further analysis is necessary to determine the effect of latitude on regional water temperatures. Higher rated EHI sites were often more distant from rivers with major hydro-development and Cree communities. One example, the Bay of Many Islands in the Cree community of Chisasibi was one of the northern-most traplines (CH04) sampled and had the highest EHI ratings observed in this study.

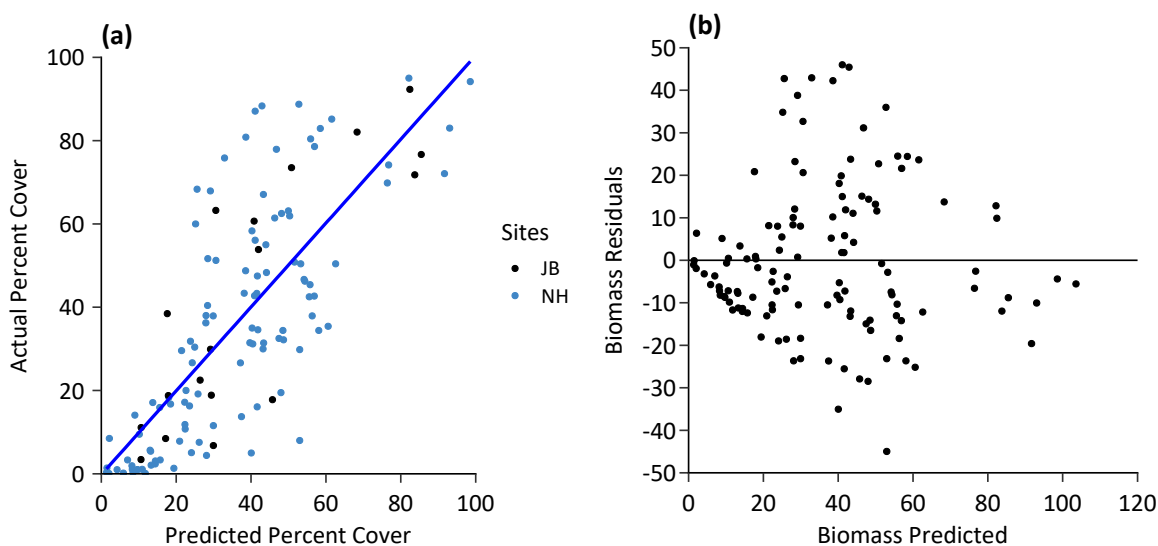
### *3.5. Past and present James Bay eelgrass density*

Historic eelgrass cover was modeled using biomass data collected between 1986 and 1991 from northern coastal traplines in Chisasibi (Lalumière et al., 1994). The linear model developed had a moderately strong fit that was significant ( $r^2 = 0.612$ ,  $p < 0.0001$ ; Fig. 2.11) and randomly distributed residuals (Fig. 2.12). The data in the model for both James Bay and New Hampshire occurred in a similar range, indicating that the Great Bay data was appropriate for inclusion in the model (Fig 2.11).





**Fig. 2.11** The relationship between eelgrass cover and biomass ( $r^2 = 0.612$ ,  $p < 0.0001$ ). The model is based on eelgrass observations from New Hampshire (NH) and James Bay (JB). NE observations are represented with blue dots; black dots represent observations from JB.



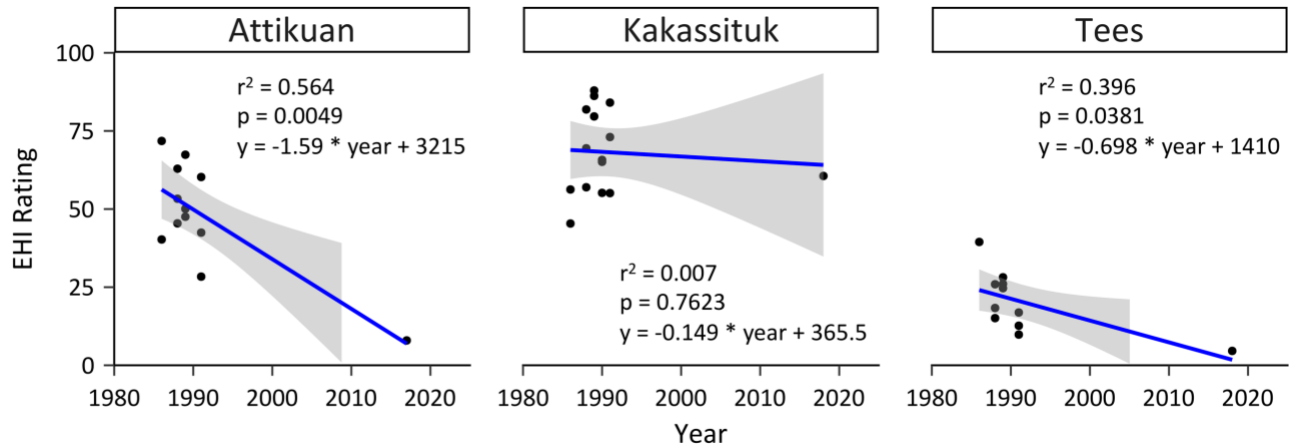
**Fig. 2.12** Actual by predicted (a) and residuals plots (b) for James Bay and New England. In a black dots denote sites from James Bay and blue dots, sites from New England while in b black dots represent all sites. James Bay ( $r^2 = 0.70$ ,  $p < 0.0001$ ), New Hampshire ( $r^2 = 0.60$   $p < 0.0001$ ).

EHI ratings were calculated for the historic monitoring sites using the modelled cover values and Lalumière study density observations. Present study EHI values were recalculated for these three sites using only percent cover and shoot density observations to be consistent with the data used to calculate the historic EHI ratings. Historic sampling was conducted at three depth ranges, 0.5, 1, and 1.5 m per site with 5 or 10 replicates per site (Lalumière et al., 1994). Historic EHI ratings were calculated for each replicate, then averaged by depth. All depth ranges were included in the model.

**Table 2.7** Historic and recent EHI ratings and calculated percent change in EHI ratings using Lalumière et al. (1986–1991) observations averaged across all depths (0.5, 1.0, and 1.5 m) for the historic data and present study observations (2017–2018).

	<b>Attikuan</b>	<b>CI</b>	<b>Kakassituk</b>	<b>CI</b>	<b>Tees Bay</b>	<b>CI</b>
Historic	51.64	8.76	68.64	7.98	21.71	6.22
Present	8.00	-	61.0	-	5.00	-
Percent Change	- 84.51	-	-11.13	-	-76.97	-

Historically, the highest EHI rated beds were found at Kakassituk ( $\bar{x} = 68.6$ ) followed by Attikuan ( $\bar{x} = 51.6$ ) and with the lowest at Tees Bay ( $\bar{x} = 21.7$ ; Table 2.7). These same sites observed in 2017 and 2018 by video-monitoring, had lower ratings with the mean EHI of 8.00, 61.0, and 5.00 for Attikuan, Kakassituk, and Tees bays, respectively (Table 2.7). Historic and present EHI ratings at Kakassituk were not significantly different, but at both Attikuan ( $r_2 = 0.559$ ,  $p = 0.005$ ) and Tees ( $r_2 = 0.391$ ,  $p = 0.04$ ) the decrease in EHI rating was statistically significant (Figure 2.13). Attikuan had the greatest negative percent change with an 84.5% decrease in its EHI rating (Table 2.7). Tees Bay had the next greatest change with a 77.0% decline in its rating. Tees Bay’s historic average EHI was already low ( $\bar{x} = 21.7$ ; Table 2.7).



**Fig. 2.13** Comparison between Lalumière et al. data from 1986–1991 and 2017–2018 EHI ratings. The grey cone represents the confidence interval for the fit of the line.

### 3.6 Study Limitations

This study aimed to monitor a range of sites across a large region of the Québec coast of James Bay. The short time period for monitoring limited the number of sites and data available for inclusion in the EHI. Most sites could only be sampled once during the study. Sites that were sampled early in the growing season could not be included in the analysis since immature eelgrass bed EHI ratings could introduce seasonal growth condition patterns to the index. Turbid and low light water conditions also prevented some sites from being monitored using videography. Due to these sampling limitations, this study’s application of the EHI provides a snapshot of eelgrass health conditions at selected sites during the peak growing season but cannot assess how eelgrass health changed at individual sites over the growing season.

Although the validation results suggest the EHI may be applicable to eelgrass outside of North America, further testing is necessary throughout the global distribution of eelgrass to determine the geographic range of the EHI’s applicability. Specific reference values need to be defined for sites being monitored over time to account for natural changes during the growing season and between different eelgrass populations (Irving et al., 2013).

The assessment of eelgrass health over time is limited by the availability of historic data on eelgrass beds in James Bay. Data are available from only a single academic paper describing conditions from 1986 to 1991 (Lalumière et al., 1994). Although monitoring on that project continued until 2000, the results were not available (Lalumière and Lemieux, 2002; Dickey, 2015). The comparison and assessment of eelgrass health over time would be strengthened with additional time series data and could be possible if the data were available.

## Discussion

### *4.1. EHI design and validation*

The eelgrass health index, as biotic indices based on a sentinel species, is an easy to use and interpret method for assessing eelgrass health (Martínez-Crego et al., 2010). EHI observation methods and equations were designed to be accessible for community-based monitoring and research in James Bay Cree Nations. The variables used in the EHI were consistent with variables selected for use by other indices and were based on standard eelgrass observations. The ability of the EHI's equation to be adjusted to available variables also expands the EHI's applicability to previous eelgrass monitoring, allowing for comparison with and integration with existing data sets.

Other seagrass indices focused on testing individual variables for inclusion in an index and the ability of the index to produce stratified ratings, i.e. levels (Karamfilov et al., 2019; Lopez y Royo et al., 2010; Neto et al., 2013). We did not assign levels as per the European Water Framework Directive (WFD). Final validation was field application to anthropogenic stressors or to monitoring site quality, e.g., marine protected areas to commercial harbors.

The EHI is can readily be adapted to the European Water Framework Directive (WFD) for inclusion in ecology quality status metric (EQS), a measure of ecosystem overall health (Foden & Brazier, 2007). Our index uses a percentage scale for ease of communication, which can be modified to the WFD 0–1 scale. Categorical levels of eelgrass conditions already exist from the eelgrass health survey and translate directly to the new scale. As the index was designed as tool for comparison to environmental conditions, it can also easily be compared to anthropogenic effects as is one of the purposes of WFD indices.

Biomass and the eelgrass health survey were complementary approaches for validating the EHI, which, together, provide a more robust validation of the EHI than a single test (Fazey et al., 2006). The biomass values provide quantitative measurements and the survey provides both quantitative and qualitative results. The survey is based on experiential knowledge and the responses therefore provide a validation based on the observed state in the world, as well as context for understanding how contemporary conditions relate to historic conditions. In addition, survey participants' comments about their selected ratings provide insight into eelgrass variables that can be considered for inclusion in future iterations of the EHI and environmental conditions for testing as potential threats and stressors.

The comparison of the EHI to both biomass and the eelgrass health survey had considerable variation in their results. This may be because variables included in the EHI are related to biomass, e.g., more shoots and larger plants are correlated with greater biomass. Yet, biomass and EHI ratings are not the same and eelgrass bed conditions can have contrasting conditions. For example, new growth eelgrass beds may have high shoot density but low overall biomass because the new shoots are smaller and not mature. The variation in the results from the survey were likely due to the subjective nature of different observer's interpretation of eelgrass health by relying on a preexisting idea of eelgrass health or including environmental factors independent of eelgrass in the rating.

#### *4.2. Considerations for application of the EHI*

The EHI could be used to compare eelgrass conditions over the growing season. To use the EHI to assess seasonal changes at a site, reference values would need to be calculated for each sampling event to account for seasonal changes in the eelgrass bed, e.g., lower height or

cover at the beginning of the growing season before the bed has naturally matured. Routine monitoring at a site and calculation of EHI values could be compared through time and to investigate the interannual and long-term health conditions of an eelgrass bed. Repeat sampling would also provide reference values for eelgrass during different growth states over a season. EHI ratings coupled with areal data may also be used as a metric for determining eelgrass donor beds for restoration. Careful selection of eelgrass donor beds is important to prevent spreading disease and invasive species (Katwijk et al., 2009). Furthermore, the EHI could be used to monitor eelgrass health after establishment at sites where eelgrass was previously absent and for comparison to applications of the site selection model in eelgrass restoration (Short et al., 2002)

One consideration for future use is that the EHI requires data from two or more eelgrass variables, e.g., cover and density, to calculate an index rating, and the index cannot be calculated with only one variable. And, EHI ratings between sites are only comparable when the same variables are used to calculate the index. For example, if two variables are represented in the data for one site and three are available for another, then the index has to be re-calculated based on the fewest available parameters in order to compare the two sites.

Across the long-term biomass sites, EHI ratings plateaued at different percentages, indicating the need for EHI ratings to be standardized by site before comparison across different eelgrass populations (Fig. 2.4). Similar to the reference values used in the index, the greatest rating could be used to standardize other ratings. It is critical that this calibration be done with caution: in stressed or impaired populations (as in Great Bay and James Bay), a site's "best" condition may still be rated as poor or fair and should not be standardized to qualify as an excellent rating.

### *4.3. The EHI and James Bay*

The EHI focuses on characteristics specific to eelgrass and final calculated index rating independent of environmental variables. One potential application of the index is using it to identify stressors affecting eelgrass decline in James Bay. The EHI also does not directly provide information on causative stressors but it could be used to analyze the effect of physical and environmental conditions such as the ones cited by participants in the eelgrass health survey or other non-visible physical conditions such as salinity, light availability, and temperature. Salinity is of particular interest as hydro-development has significantly changed river discharge volumes and seasonal flow regimes throughout the major watersheds of James Bay (Berkes, 1989). Eelgrass populations vary in their low salinity tolerance (Salo, 2014), and in James Bay, changes to salinity and its effect on eelgrass are poorly understood and an area to apply the EHI.

The decline in James Bay eelgrass over time is clearly illustrated by comparing the historic to current EHI ratings. Most studies document the decline starting in the late 1990s, but without available data from before 1986 and after 1991 to calculate additional EHI ratings and fill in the time gaps, it has been impossible to determine if eelgrass beds are still in decline, static, or improving in health (Short, 2008; Dickey, 2015). This is a conservative estimate since the density reference values for each time period were independently calculated. To standardize the calculation, reference values would only be used from the pooled best observations if similar in value or from the best period observed; in this case, the historic observations. Historic densities were greater, so this would further decrease the current EHI ratings.

The decrease in EHI ratings at Attikuan and in Tees Bay over the last 30 years may have been caused by freshwater discharge from the mouth of the La Grande River. Hydro-development on the La Grande and regional James Bay rivers and the corresponding alterations



to the river flow that increased discharge and altered seasonal flow regimes are potentially having a negative effect on eelgrass at these sites. Attikuan to the north and Tees Bay immediately south of the mouth of the La Grande River are within the river's freshwater plume. Increased freshwater input reduces salinity and has previously been demonstrated to cause stress to eelgrass and in severe instances, mortality (Salo et al., 2014); at these two sites it may have dropped low enough for sustained periods to negatively affect eelgrass productivity and abundance. The third site, Kakassituk, is a more protected bay partially blocked off by a peninsula and numerous islands; these natural features may act to shelter Kakassituk from the discharge from the La Grande allowing eelgrass shoot density to maintain levels similar to 30 years ago. The low EHI ratings as reported here are not unique to Attikuan and Tees Bay and were found widely throughout James Bay in the ratings calculated for this study. Other effects from the broad regional hydro-development and from other factors are likely affecting the coast-wide impairment of eelgrass beds and are critical areas for current research to understanding eelgrass threats and stressors in James Bay (Short et al., 2019).

## Conclusion

The EHI is a powerful tool designed to directly evaluate eelgrass health for comparative analysis of environmental conditions to identify threats and stressors. The validation of the EHI using eelgrass biomass from James Bay, Atlantic, and Pacific eelgrass beds demonstrates its applicability to different eelgrass populations in North America. The eelgrass health survey was a second validation and a framework for including experiential knowledge in ecological assessments.

In our application of the EHI to James Bay, we found that eelgrass generally received low ratings, indicating an impaired condition throughout coastal waters in James Bay, Québec. Only a few sites had EHI ratings greater than 50 and these sites generally were in remote or sheltered areas. In the investigation of three historic sites near the mouth of the La Grande River in Chisasibi, the sites with the most direct exposure to the river's plume had a significant decrease in EHI ratings and eelgrass health since historic monitoring efforts 30 years ago. The one site that was more protected did not have significant decrease in its EHI rating as compared to historic levels.

Eelgrass as a widely distributed species of seagrass in the northern hemisphere has a broad applicability for coastal ecological assessment. Applying the EHI to eelgrass beds in other countries could help to determine how effective the index is throughout the global range of eelgrass. Individual characteristics of eelgrass still provide important information on eelgrass abundance and productivity; the EHI builds on these metrics and provides a more complete assessment of eelgrass health for use in coastal management, conservation, and restoration.

The EHI was designed for community-based research with a focus on making the methods accessible to both researchers and non-scientists. These methods are intended to make monitoring more feasible for all involved in coastal research and stewardship.

## FINAL REFLECTIONS

My thesis presents a practical methodology to increase opportunities for monitoring and assessing eelgrass health. The assessment approach responds to an explicit question from the Cree to contribute to informing stewardship of James Bay's coastal eelgrass beds but is intended to be relevant to eelgrass beds globally. In summary, I found:

- Video monitoring is a simple and effective alternative to conventional eelgrass monitoring to observe eelgrass cover, density, and height.
- The EHI is consistent with both biomass and experiential knowledge assessments of eelgrass and can therefore be used as a tool for assessing eelgrass health conditions in temperate and subarctic North American eelgrass populations and, with further validation, be considered for assessing eelgrass in other parts of the world.
- James Bay eelgrass beds, as assessed by the EHI rating, are in an impaired state (mean coast wide EHI = 26.9). Sites with the highest EHI ratings were in isolated areas throughout the study area with high values in Chisasibi (N) (81.1), Chisasibi (S) (60.1), and Waskaganish (52.7).
- The health of James Bay eelgrass beds has declined over the past four decades. Only one of three sites did not have a significant decrease in its EHI rating as compared to historic EHI ratings.

- Eelgrass biomass (square root transformed) can be predicted with moderate accuracy using percent cover and a simple linear regression. The relationship between eelgrass cover and biomass (untransformed) appears exponential, but further investigation is necessary. Eelgrass percent cover can be also predicted with moderate accuracy using biomass (square root transformed) by reversing and supplementing the variables used in the eelgrass cover-biomass model.

### *Next Steps*

A comparative analysis using the EHI is currently in progress to investigate the cause(s) of eelgrass decline in James Bay (Short et al. 2019). Environmental conditions that regulate eelgrass productivity, light, temperature, salinity, and water color, are being compared to EHI ratings. Results from this analysis could be used to determine which of these physical variables are causing eelgrass stress and decline. Work is also underway to make the data and methods from this study available to Cree communities. Data are being prepared and hosted by the University of New Hampshire scholar's repository (<https://scholars.unh.edu/>), while the EHI and associated video monitoring methods are being described in standard operating procedures for use by the Cree and communities interested in monitoring coastal eelgrass beds. The new community-based Chisasibi Eeyou Research and Restoration Institute (CERRI) was recently launched in Chisasibi. The focus of their research is on coastal health and the EHI is planned as a tool that they can use for assessment.

Some potential variables for inclusion in EHI calculations were identified in the eelgrass health survey. While we selected variables for the EHI based on ease of observation and baseline eelgrass monitoring, previous studies have focused more on the selection process to determine

which variables were most important (García-marín et al., 2013). Testing the addition of new variables to calculate EHI ratings would also likely increase the representative power of the index, but each iteration would have to be re-validated.

There is local interest in New Hampshire and Maine in applying the EHI to on-going eelgrass monitoring and assessment. The EHI may provide a coastal science partnership (PREP) a new method of communicating eelgrass status in Great Bay Estuary. Future work with local partners may provide opportunities to test the EHI against environmental conditions.

### *Working with the Cree*

Over the course of my master's degree, I learned many things both inside and outside of the classroom from northern Cree communities and trappers and as a university graduate student. These are my own observations and do not reflect those of the Cree or of anyone else involved in this research.

Working with local communities was exciting and challenging. I experienced enthusiasm for our project and a desire to see results, but with only a limited time in Québec, it was challenging to establish a community-based monitoring program. I imagine this is true in other engaged research efforts. Older Cree trappers who observed the decline in eelgrass were invested in the eelgrass and coastal monitoring project, seeing the potential to prevent further decline in the coastal eelgrass beds and the geese, waterfowl, and fishes that rely on them, and were open to sharing their knowledge of the historic conditions of eelgrass beds. They also were intimately familiar with the coastal waters and it quickly became apparent that we should follow their lead in where we monitored eelgrass. Cree youth, many of who are more technologically savvy than I am, did not have this firsthand knowledge of the eelgrass decline, but were learning it from the community elders and trappers we worked with. It is my sincere hope that the coastal Cree communities continue to share knowledge across generations and that the methods developed in my thesis will help them to continue to observe and assess eelgrass in James Bay.

## *Graduate school*

Here are a few key take-aways from my experience as a student at the University of New Hampshire. 1) If you want to be an ecologist take statistics classes, the more the better. I took the minimum required and I regret it. The same is true for coding. Coding is not necessary for research, but it can streamline analysis and interpretation as you advance in it. It is also listed as a preferred skill by many employers in the sciences. 2) “Research takes priority to classes.” I heard this from multiple peers and the real truth I found was one my advisor described perfectly: that as a student, scientist, or professor, you are always chasing balance, or “juggling”. Balance does not really exist in academic environments, instead it is more of a state of perpetually taking care of the most pressing work. Ideally, classes support research and vice versa, but I believe this tension exists at most universities. 3) Work at a university’s writing center. It is service for your fellow students and it makes you think deeply about your own writing and writing process—something all scientists should embrace. I was fortunate to work at the Connor’s Writing Center at UNH, and it was rich experience with an incredible community that I would not trade for anything.

There were also harder lessons: when bad things happen, and they always do, look for opportunities and focus your efforts. Our project funding was cut before our third planned field season. This was the motivation for me to apply for and receive departmental support for academic achievement, co-write my first successful grant proposal for NH Sea Grant funding, and, most importantly, foster new partnerships. This experience and the successes that came out have made my master’s education a much richer and more fulfilling experience and the research that resulted from it is also better from the experience.



There are many more things I have learned, but the final one is that this and all theses are the product of many people pulling together to work towards a common goal. Without collaboration, this research is not possible. And, of the all the models I explored on this project, collaboration is the one I am certain will stay with me.

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<https://doi.org/10.1016/j.jembe.2007.05.032>

## APPENDICES

### **Appendix A: IRB Approval Letter**

# University of New Hampshire

Research Integrity Services, Service Building  
51 College Road, Durham, NH 03824-3585  
Fax: 603-862-3564

18-Oct-2019

Anderson, Nicholas B.  
NREN, James Hall  
College Road  
Durham, NH 03824

**IRB #:** 8004

**Study:** Eelgrass Health Survey

**Study Approval Date:** 21-Mar-2019

**Modification Approval Date:** 17-Oct-2019

**Modification:** Change in study title, new survey group, online only

The Institutional Review Board for the Protection of Human Subjects in Research (IRB) has reviewed and approved your modification to this study, as indicated above. Further changes in your study must be submitted to the IRB for review and approval prior to implementation.

Researchers who conduct studies involving human subjects have responsibilities as outlined in the document, Responsibilities of Directors of Research Studies Involving Human Subjects. This document is available at <http://unh.edu/research/irb-application-resources> or from me.

Note: IRB approval is separate from UNH Purchasing approval of any proposed methods of paying study participants. Before making any payments to study participants, researchers should consult with their BSC or UNH Purchasing to ensure they are complying with institutional requirements. If such institutional requirements are not consistent with the confidentiality or anonymity assurances in the IRB-approved protocol and consent documents, the researcher may need to request a modification from the IRB.

If you have questions or concerns about your study or this approval, please feel free to contact Melissa McGee at 603-862-2005 or [melissa.mcgee@unh.edu](mailto:melissa.mcgee@unh.edu). Please refer to the IRB # above in all correspondence related to this study.

For the IRB,



Julie F. Simpson  
Director

cc: File  
Ashcraft, Catherine

## **Appendix B: Eelgrass Health Survey**

# Eelgrass Health Survey and Results

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# Eelgrass Health Survey and Results

Nicholas B. Anderson, University of New Hampshire (corresponding author)

Catherine M. Ashcraft, University of New Hampshire

Dante T. Torio, University of New Hampshire

Frederick T. Short, University of New Hampshire (emeritus)

## Eelgrass Health Survey Introduction

Researchers at the University of New Hampshire designed, tested, and conducted an eelgrass health survey, which aimed:

- To increase the accuracy of research results. Survey respondents provided health ratings based on images of eelgrass beds, which were used to calibrate and validate a novel visual health index to assess eelgrass health using video monitoring,
- To build confidence in the new visual health index among potential future users by incorporating experiential knowledge from individuals familiar with eelgrass beds.
- To identify the plant-specific and environmental characteristics survey respondents consider important for assessing eelgrass health.

The University of New Hampshire Institutional Review Board for the Protection of Human Subjects in Research approved this study (IRB #: 8004; Study approval date: 3/21/2019; Modification approval date: 10/17/19). The survey was conducted online using Qualtrics during October and November 2019.

Researchers recruited individuals with prior experience observing eelgrass beds to participate in the survey. Recruitment aimed to survey participants with diverse backgrounds and, therefore, diverse experiences with eelgrass beds. Nineteen individuals completed the survey. Participant backgrounds included coastal researchers, resource managers, educators, and fishermen. Their level of experience ranged from less than five to more than 30 years. Most respondents had earned a graduate degree. Respondents reported most often observing eelgrass beds in Maine, New Hampshire, and Massachusetts

Survey participants were presented with images. The order in which images were presented was randomly rotated. Respondents were asked to select one of five eelgrass health ratings: “Very Bad”, “Poor”, “Fair”, “Good”, and “Excellent”. Respondents could also provide a rationale for their selections. Images used in the survey came from sites in James Bay, Québec, Canada, and the Great Bay and Piscataqua River estuaries in New Hampshire and Maine, U.S.A. Images were chosen to represent a broad range of eelgrass health conditions. Prior to use in the survey, the survey researchers rated all images using the eelgrass health index (range: 1 – 100) and standardized to the scale used in the survey (Very Bad: 1–20, Poor: 21–40, Fair: 41-60, Good: 61-80, Excellent: 81-100).

This document aims to make the survey and complete data openly available to anyone interested in the results or who wants to build on this research. Consistent with the approved IRB protocol, survey data were de-identified and are presented in an aggregated format to protect the identity of individual respondents. The data set includes:

Part 1:

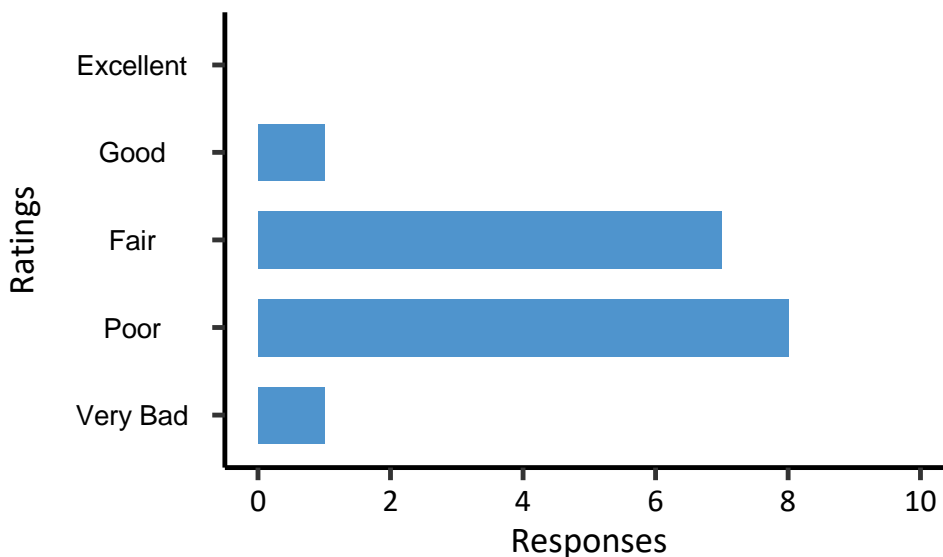
- Demographic data and general background data about survey respondents

Part 2:

- The eelgrass images presented to survey respondents
- Respondents' eelgrass health ratings for each image (y-axis in the sample bar plot below) and the number of survey respondents who selected each rating (x-axis in the sample bar plot below). Where respondents provided a rationale for their selection of specific ratings, their written responses are included on the page following the corresponding data plot. In order to protect respondents' confidentiality, respondents' comments are presented in aggregate and their order randomly rotated across survey images.
- Researchers' rating of each image calculated using the eelgrass health index, presented in two formats:
  - The value of the Eelgrass Health Index rating (EHI) is indicated at the top of each image ("EHI=42.2" in the sample data plot below).
  - The standardized rating of the EHI is indicated as a gray shaded box around the title of the appropriate rating on the y-axis (see gray shading around "Fair" in the sample plot below)

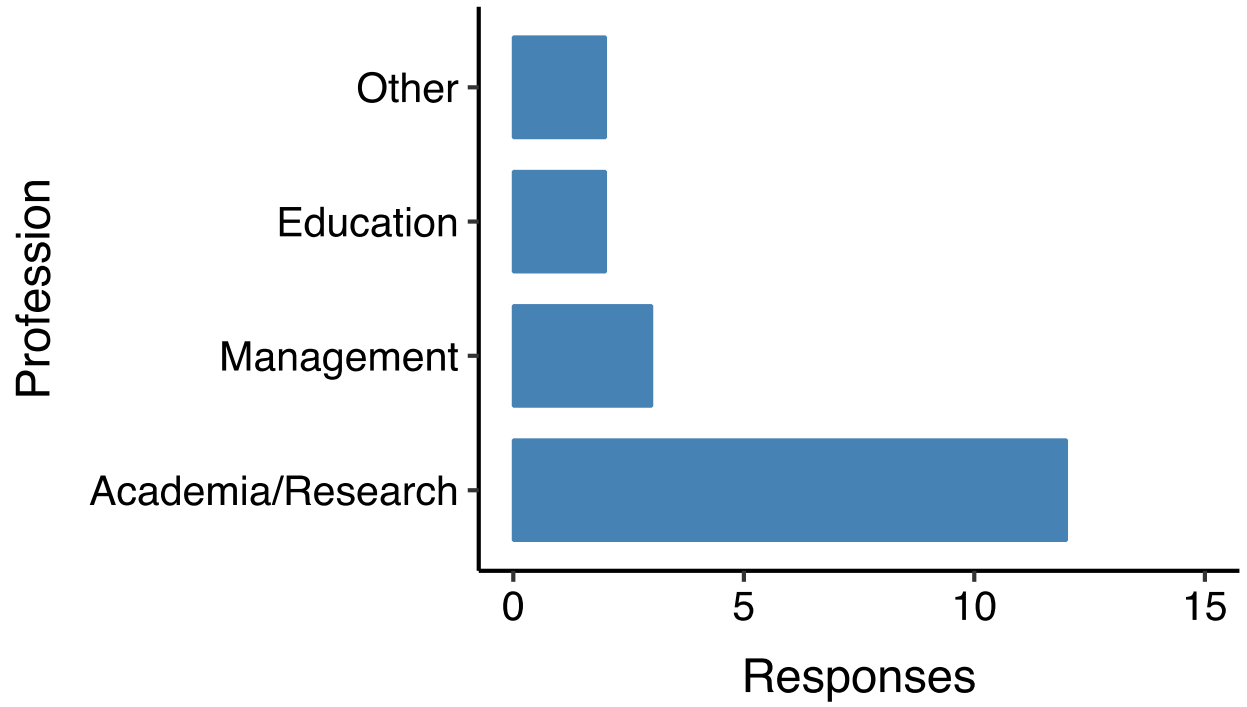
**Sample Bar Plot**

EHI: 22.3

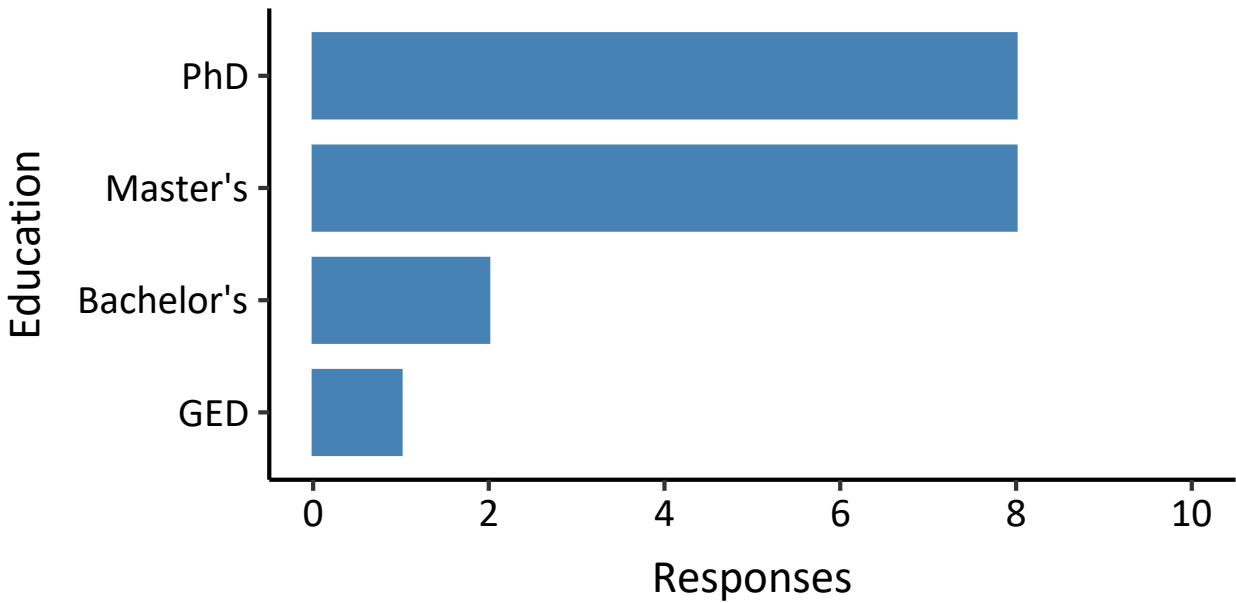


**Part I: Background Questions**

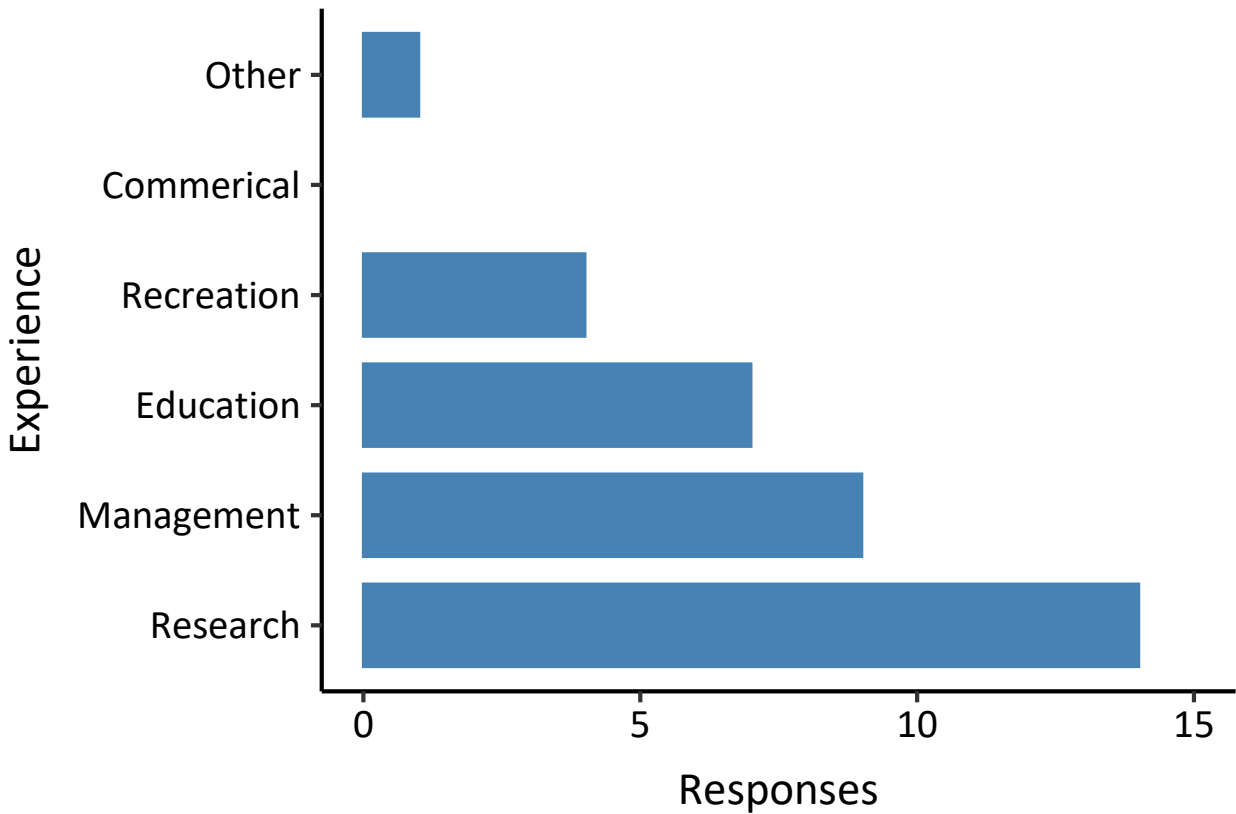
**Q2 - What is Your Profession?**



**Q3 - What is the highest level of school you have completed or degree you have received?**



**Q4 - In what capacity do you have experience with eelgrass? (Check all that apply)**

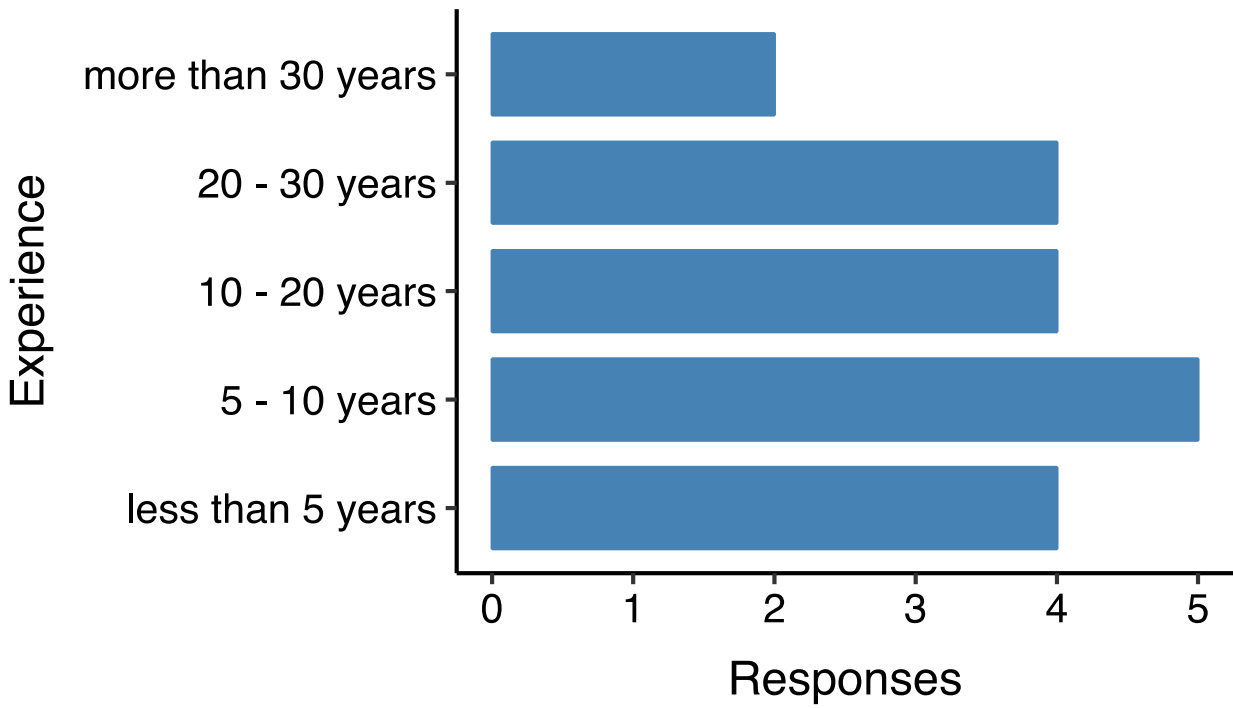


**Q5 - Please explain why you selected other for experience.**

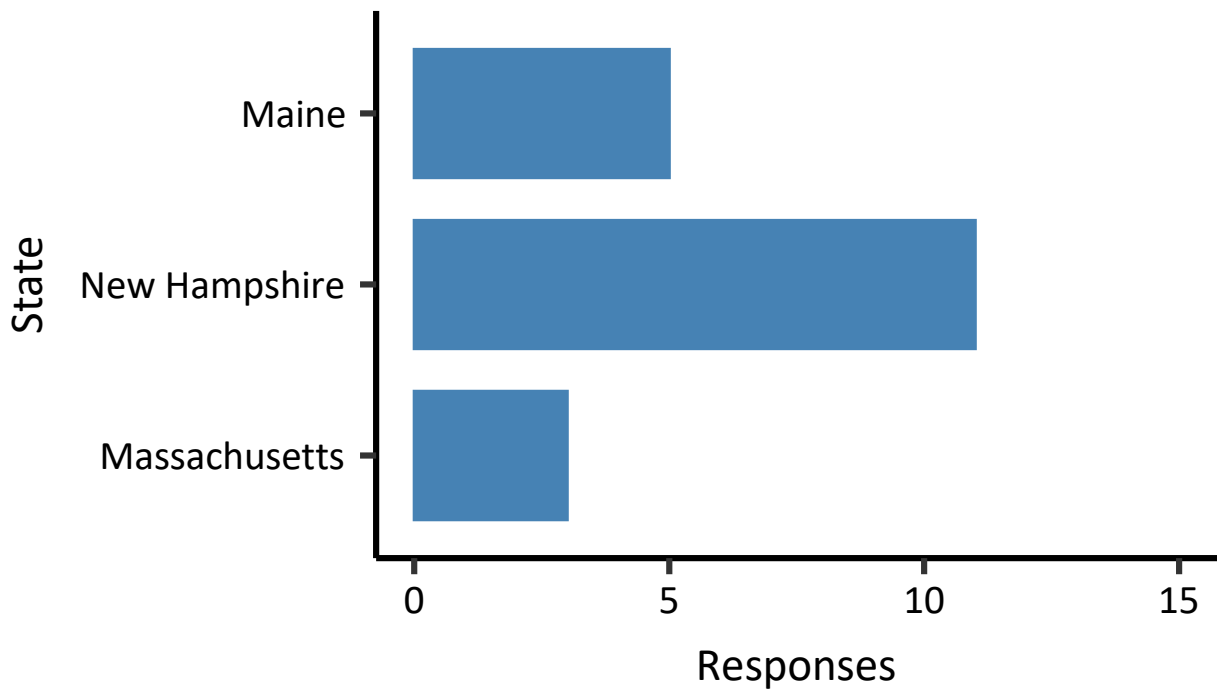
- No responses.



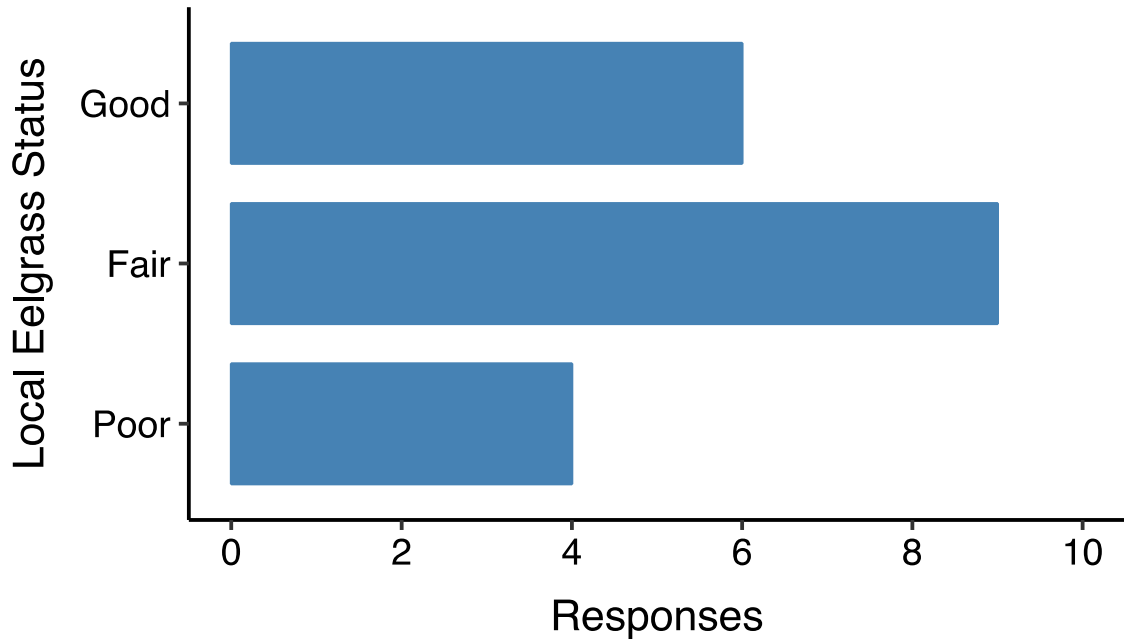
**Q6 - How long have you been working with and/or observing eelgrass?**



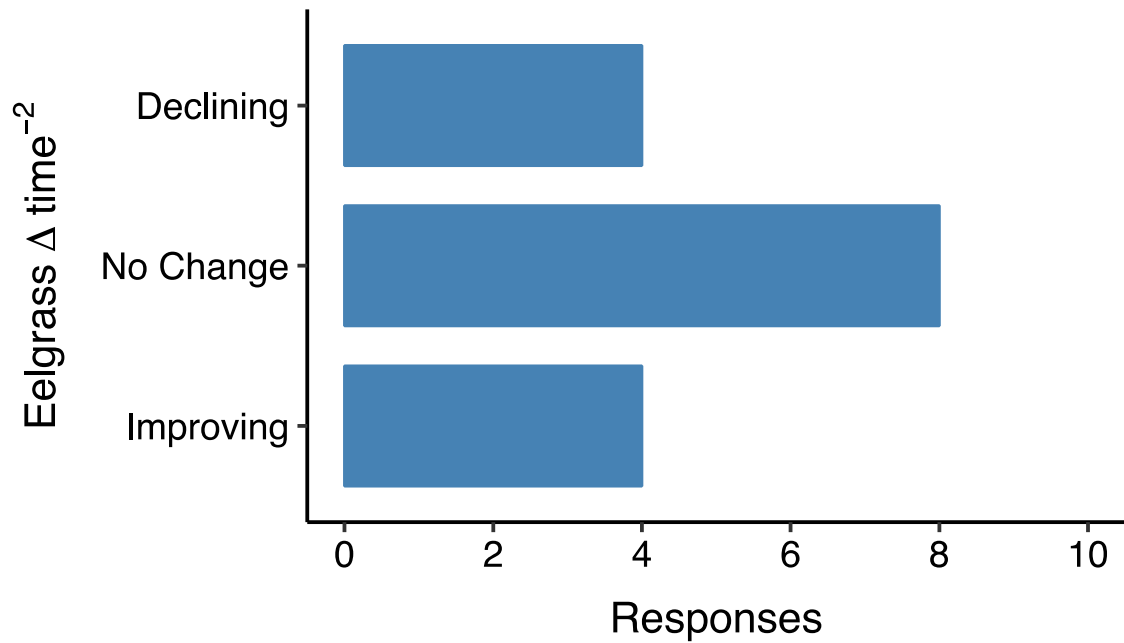
**Q7 - Where do you most often observe eelgrass?**



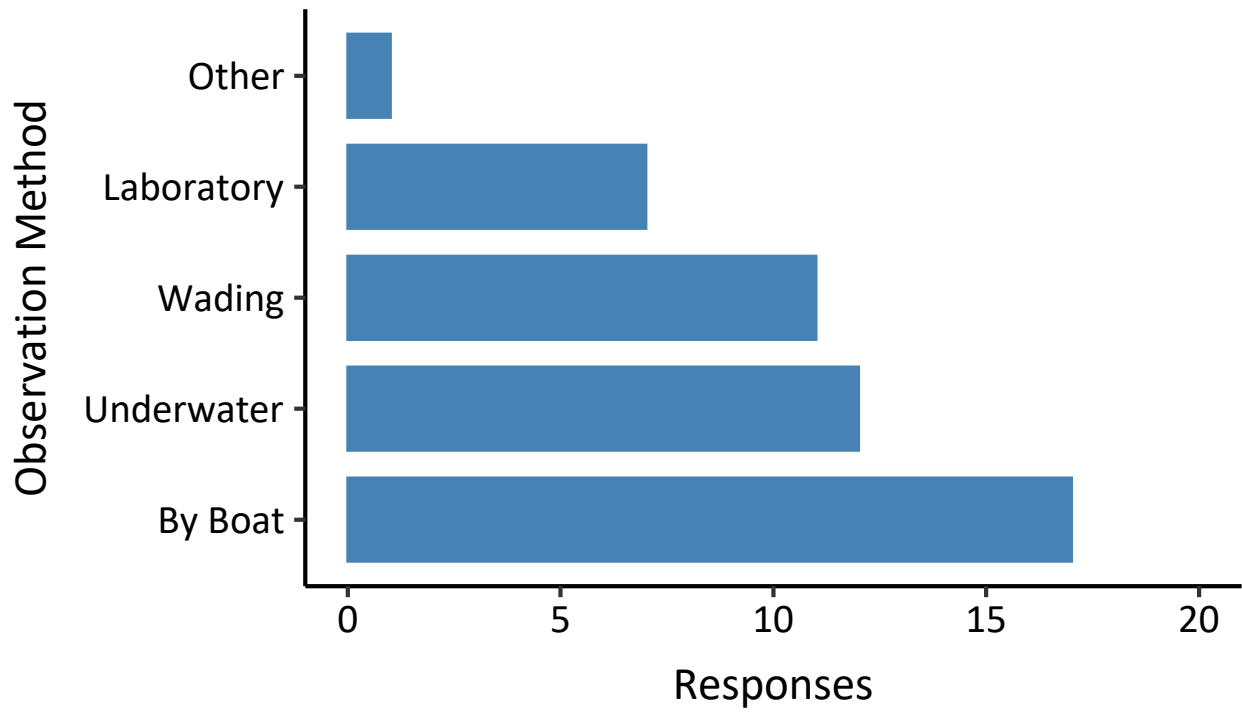
**Q8 - In this population, how would you describe the current eelgrass health status?**



**Q9 - In this population, how would you describe eelgrass conditions as they have changed over the last 5 years.**



**Q10 - How have you observed eelgrass? (Check all that apply)**



**Q11 - If 'Other' please explain**

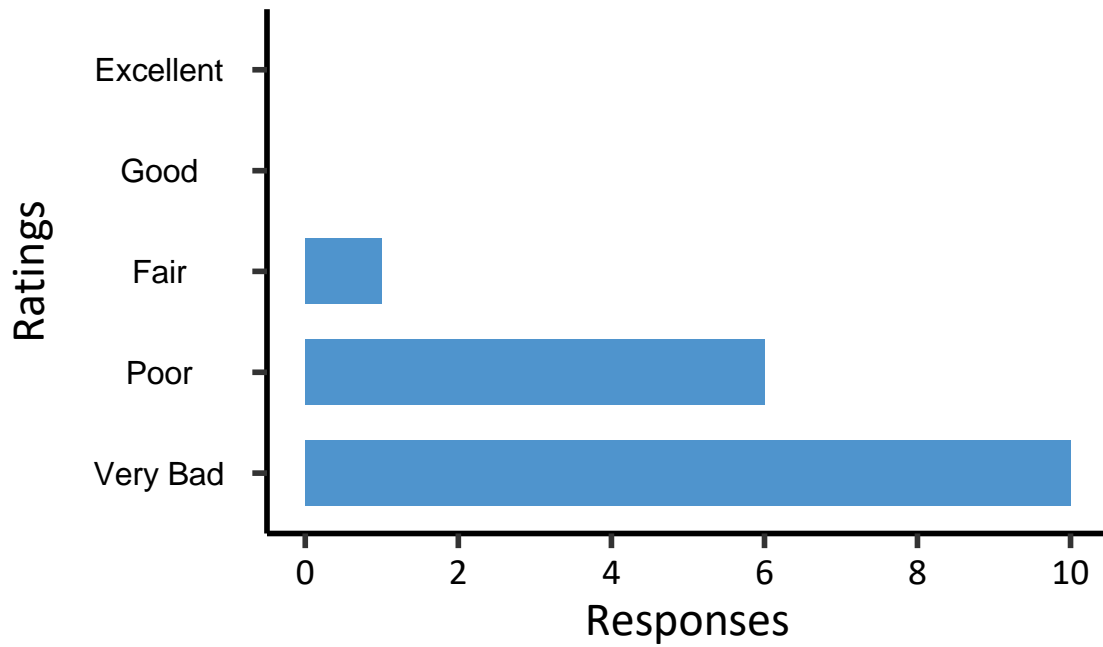
- From Shore
- Underwater camera from boat
- Review Reports of Others

**Part II: Eelgrass Health Survey**



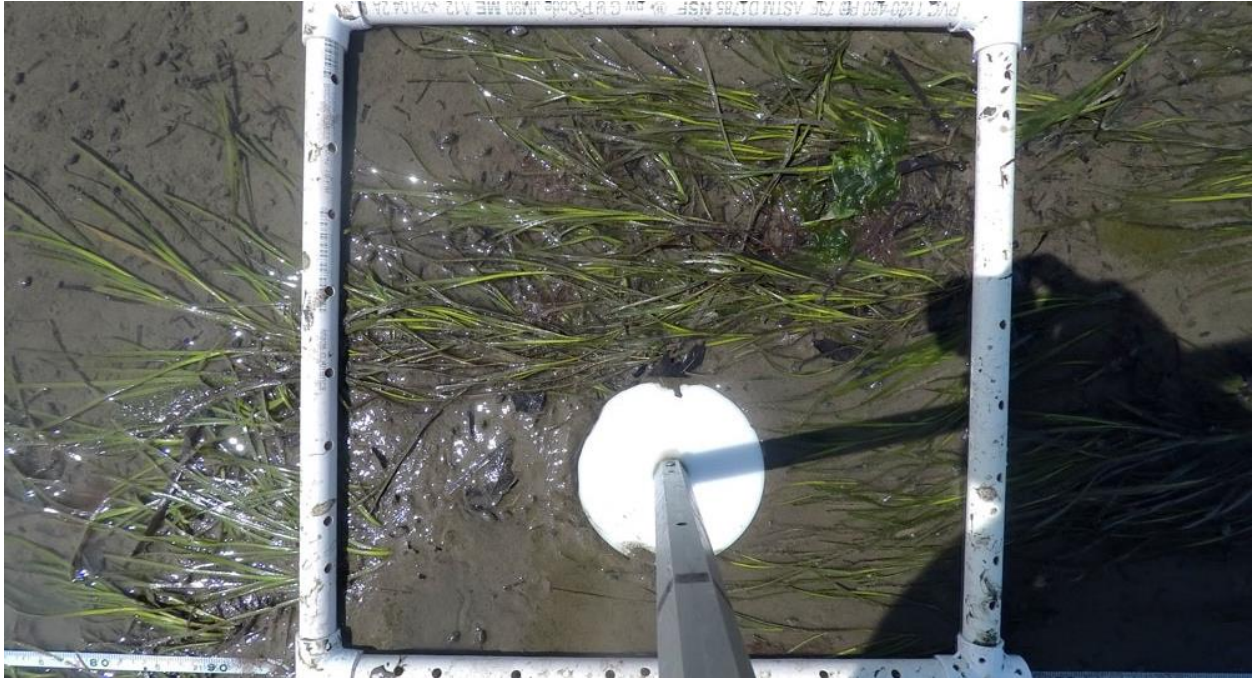
**Q1.1 - How would you rate the health of the eelgrass in this frame?**

**EHI: 7.70**



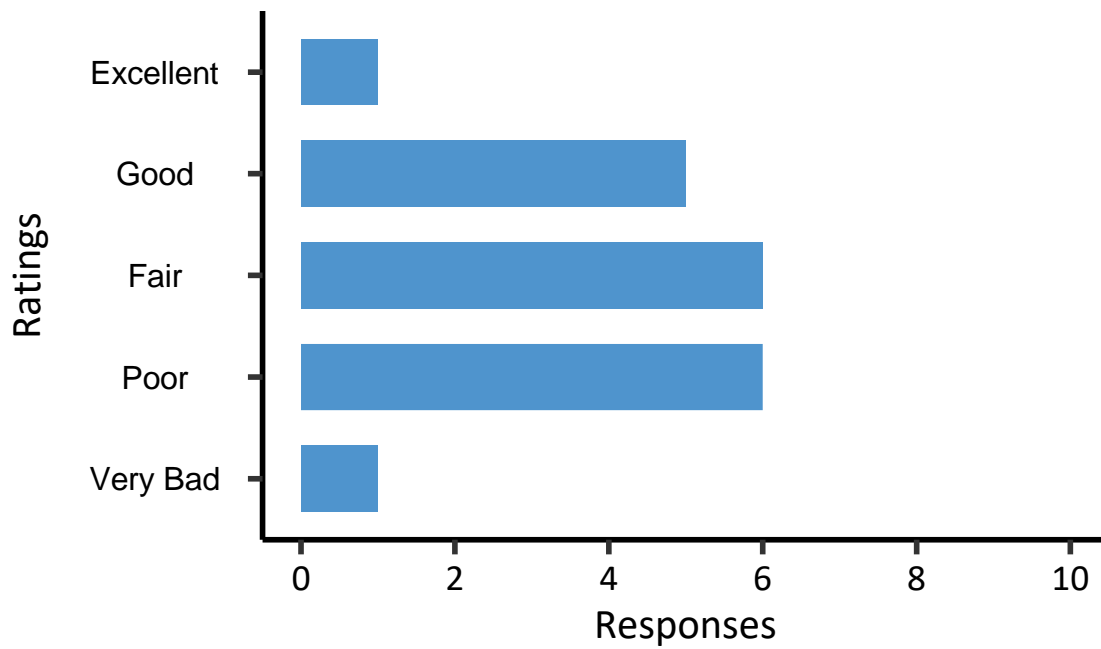
**Q1.2 - Why did you select this rating? If you would like use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- small shoots, sparse density
- That's an eelgrass bed? Those poor little shoots...
- low cover, poor water clarity
- Low clarity and no eelgrass
- Low density and percent cover, sediment accumulated on above ground biomass
- poor image?
- about 10% cover with poor WQ
- borderline very bad/poor low shoot density, poor water clarity
- Qualitative Visual Assessment
- sparse blades and cloudy water
- small, sparse cover, etc.



Q2.1 - How would you rate the health of the eelgrass in this frame?

EHI: 42.2



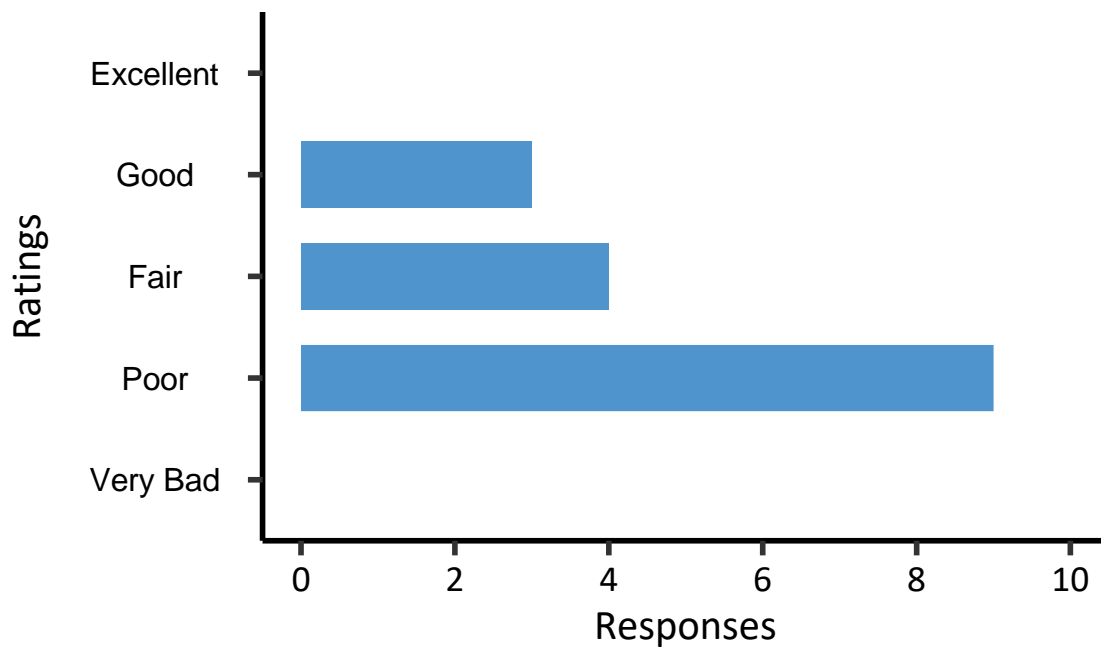
**Q2.2 - Why did you select this rating? If you would like use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- presence of some macroalgae, moderate cover eelgrass
- moderate density, low epiphyte
- Qualitative Visual Assessment
- Sparse but some healthy looking shoots
- sparse coverage and some competition from ulva
- smaller plants, patchy coverage
- This bed is on the upper end of the Poor category (maybe low end of Fair?). Plants look healthy but small. In addition, they must be fully exposed at low tide which probably doesn't help their survival.
- About 50% cover with drift algae observed
- Plants are green and look healthy, intertidal meadows are always more sparse due to the physical stress of where they exist and the potential exposure to geese and other grazers
- poor cover, some coating, small



Q3.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 27.8





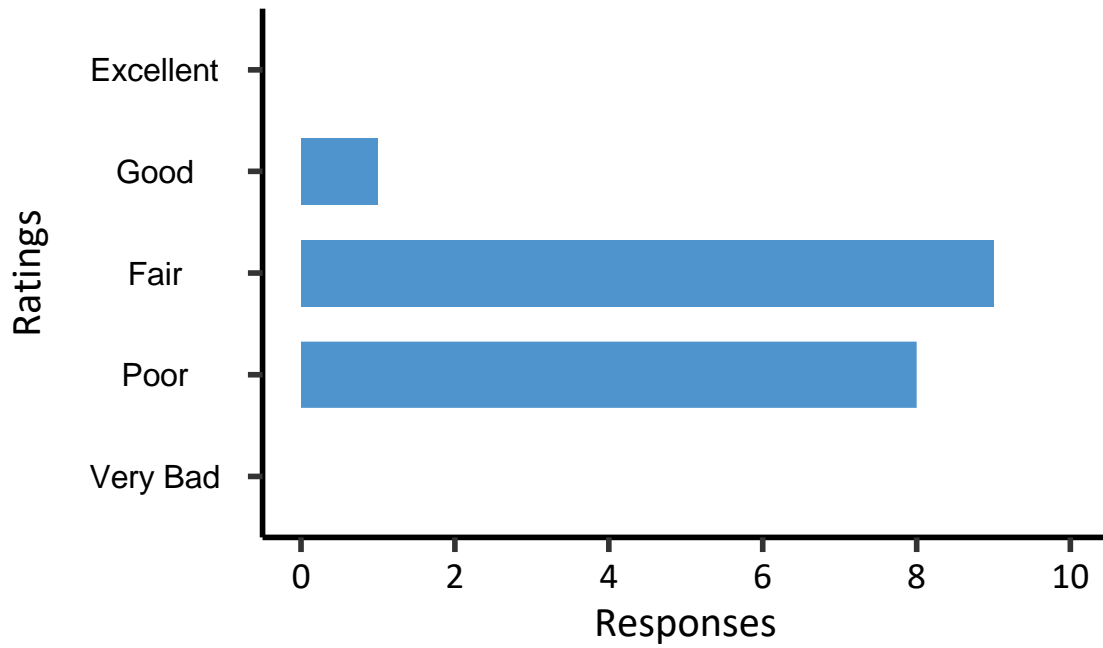
**Q3.2 - Why did you select this rating? If you would like use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- borderline poor/fair, water clarity looks poor, but shoot density could be in the fair range
- Qualitative Visual Assessment
- maybe hard to tell with bad water clarity
- hard to tell - looks very sparse
- can't tell, poor image
- Water quality is poor but about 60% cover of plants
- moderate density, poor water clarity
- Thicker than previous picture
- plants look ok despite poor water clarity



Q4.1 - How would you rate the health of the eelgrass in this frame?

EHI: 37.5



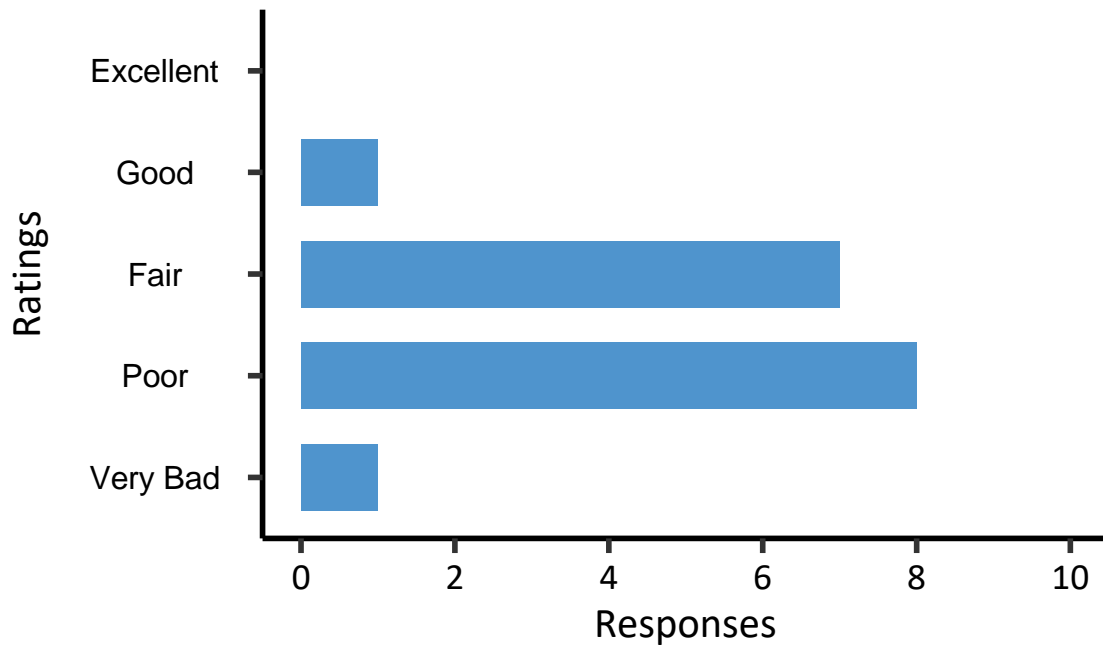
**Q4.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- low shoot density, limited water clarity and the evidence of wasting disease prevalent on some leaves
- water is cloudy; some epiphytes are noticeable; and some wasting disease is evident
- Ugh! 25% cover but poor WQ and algae level unknown
- densey vegetated with many reproductive shoots
- Qualitative Visual Assessment
- Moderate water column turbidity, moderately lengthy shoots with fouling
- Decent bed density



Q5.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 22.3



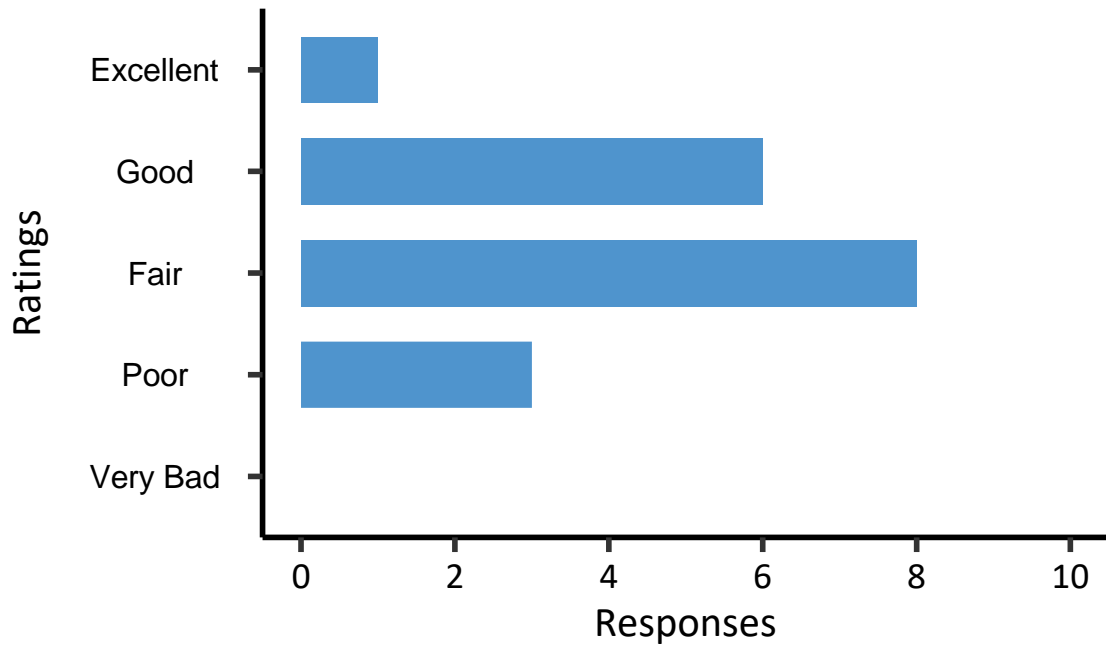
**Q5.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Low clarity but moderate density
- Few shoots rooted in quadrat
- Really hard to tell from the picture. Plants seem tall but can't get a real indication of density.
- Hard to tell - looks sparse
- Qualitative Visual Assessment
- Hard to tell, but water column turbidity and presence of fouling community combined with lower density would indicate a less healthy bed
- borderline fair/poor; water clarity is poor, but shoot density looks fair, under the premise that water clarity may reflect short term conditions while shoot density integrates conditions over time, putting this in the fair category
- I do not see much algae or epiphytes, but it is not very dense
- moderate density, little algae , poor water clarity
- can't tell, poor image
- About 50% plant cover but lousy WQ



**Q6.1 - How would you rate the health of the eelgrass in this frame?**

**EHI: 65.0**



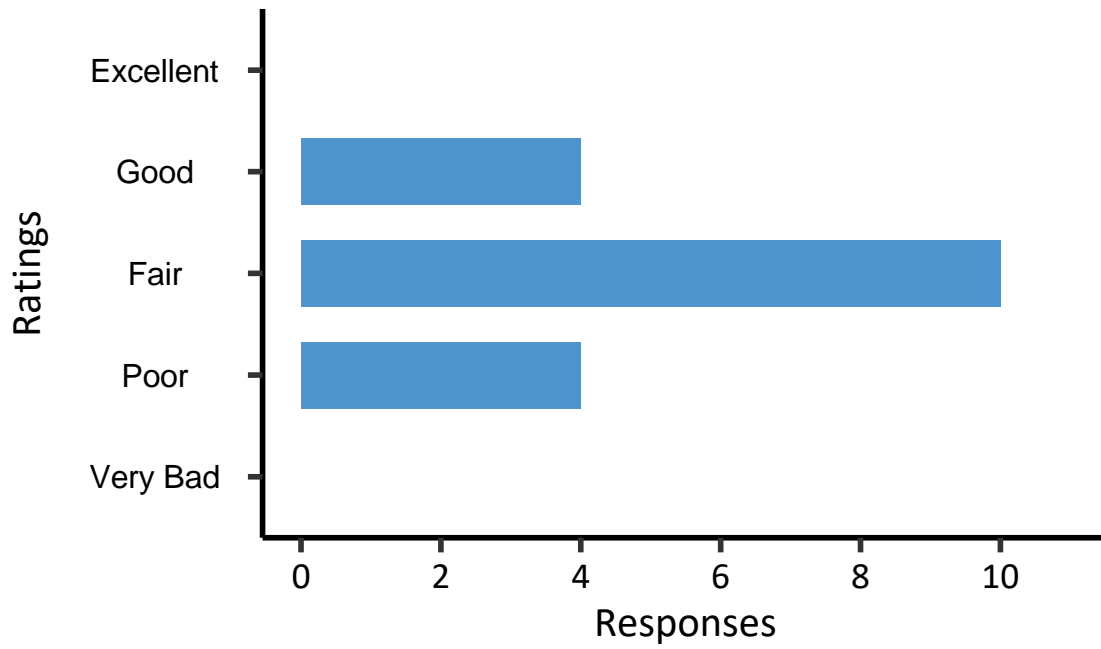
**Q6.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- high density, low epiphytes
- Good density, plants look ok
- Qualitative Visual Assessment
- lots of epiphytes
- wasting disease?, epiphytes
- Similar to previous but poorer water quality but at the same time good coverage by plants
- unhealthy
- very similar to meadows I see on Martha's Vineyard, where by the late summer/early fall they begin to get covered with epiphytes, but somehow the meadows persist



Q7.1 - How would you rate the health of the eelgrass in this frame?

EHI: 56.1





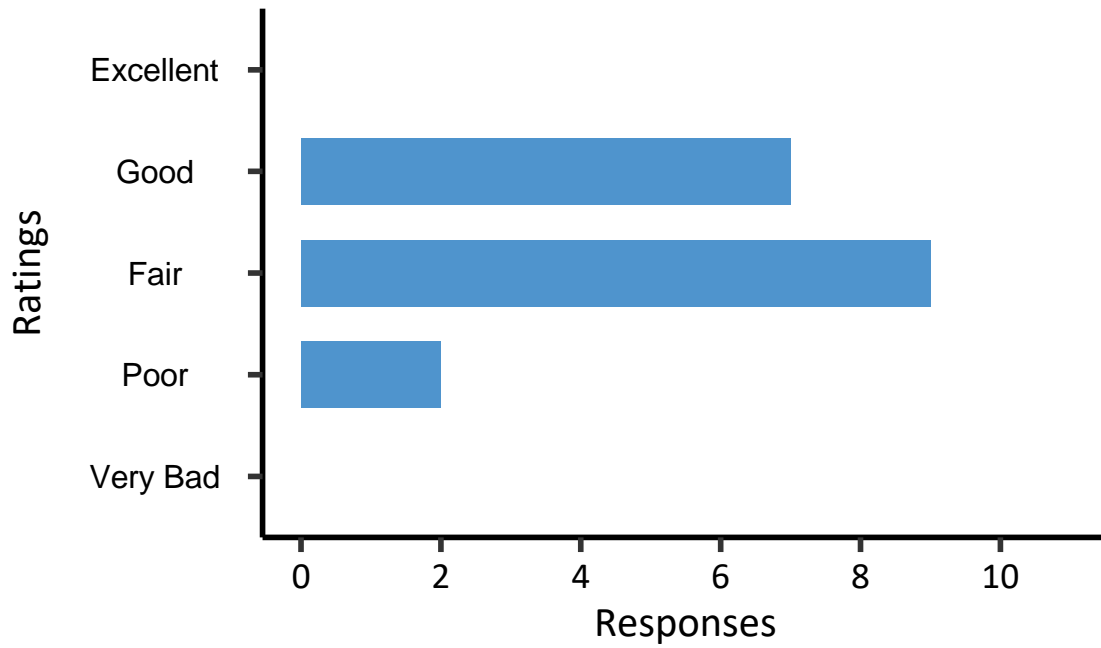
**Q7.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- denser, but looks like it does have growth on the blades
- looks unhealthy
- borderline good to excellent, high shoot density, but plenty of epiphytes, would be useful to know the time of year when photos are taken as each meadow will look different at various parts of the growing season
- high cover, moderate to high epiphytes
- high density, high epiphytes
- Epiphytes on leaves and graying water color are questionable
- Qualitative Visual Assessment
- dense plants (80 % cover) with many old, epiphytized leaves
- dense but epiphytes
- high density, high epiphyte
- epiphytes



Q8.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 33.6



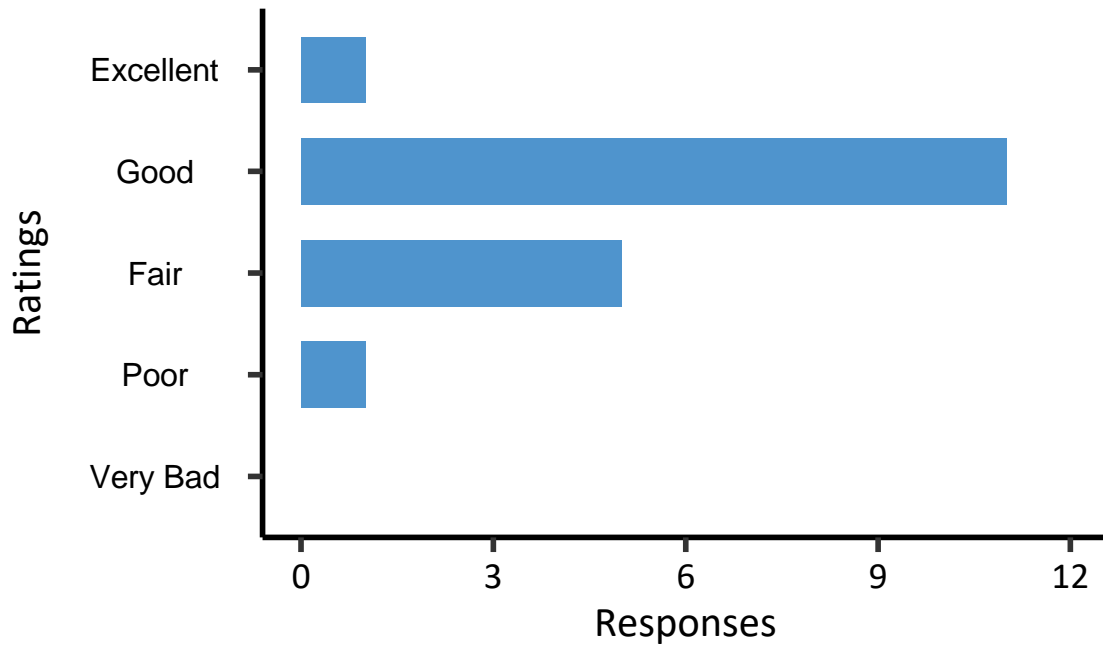
**Q8.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- thin blades, water quality could be light limiting, looks like there's some algal cover at sediment level
- High density but clarity ok
- Dense plants but algae and smothering evident
- based on the %cover, color of the vegetation and the hint of epiphytes
- Qualitative Visual Assessment
- high cover, moderate epiphytes
- Plants look ok, I think that's macroalgae



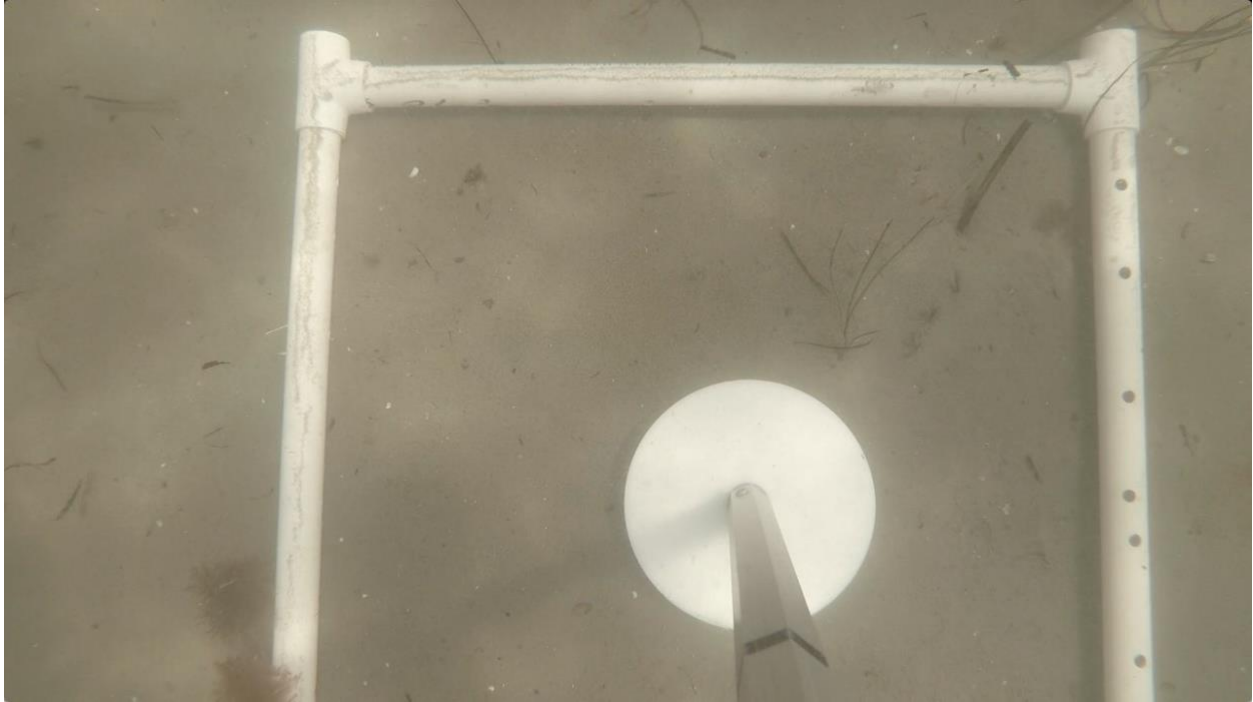
**Q9.1 - How would you rate the health of the eelgrass in this frame?**

**EHI: 50.9**



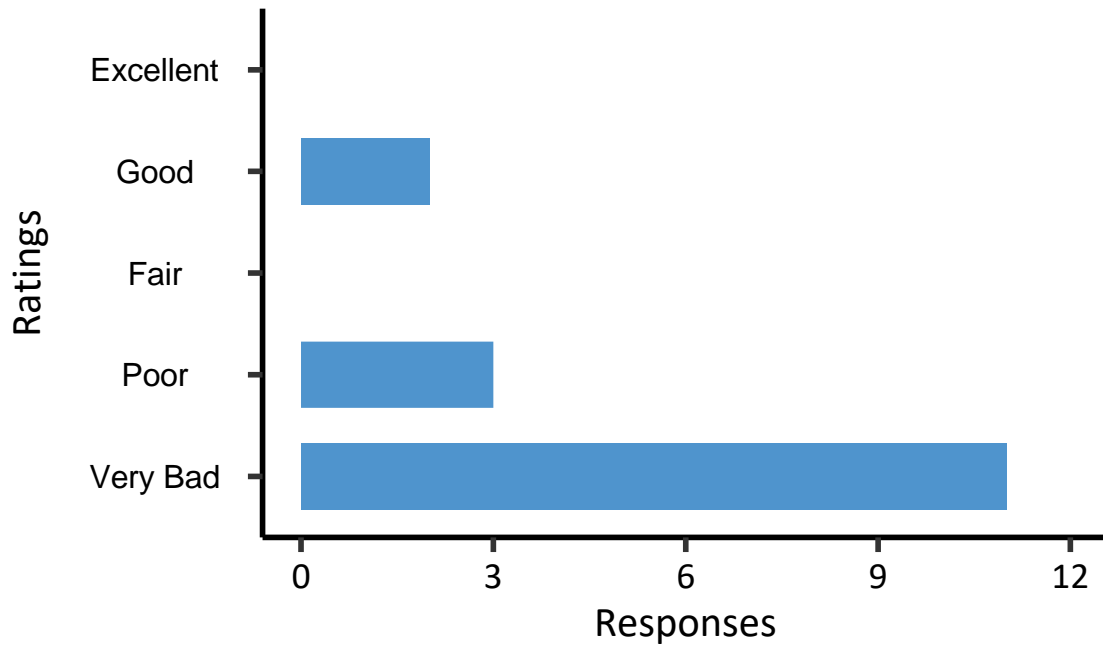
**Q9.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Qualitative Visual Assessment
- shoot density are water clarity are good, despite the presence of epiphytes
- Good density, lot of algae
- Vacillating between good and fair. great eelgrass cover and tall stems but WQ not great and leaves covered by epiphytes; some but not all shoots are reproductive
- dense, high epiphytes
- no disease, long blades, moderate density, but some epiphytes
- Relatively high aboveground biomass and potentially shoot density--hard to say if minor-moderate fouling is present
- Fairly dense moderate clarity
- lots of epiphytes
- dense



Q10.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 2.94



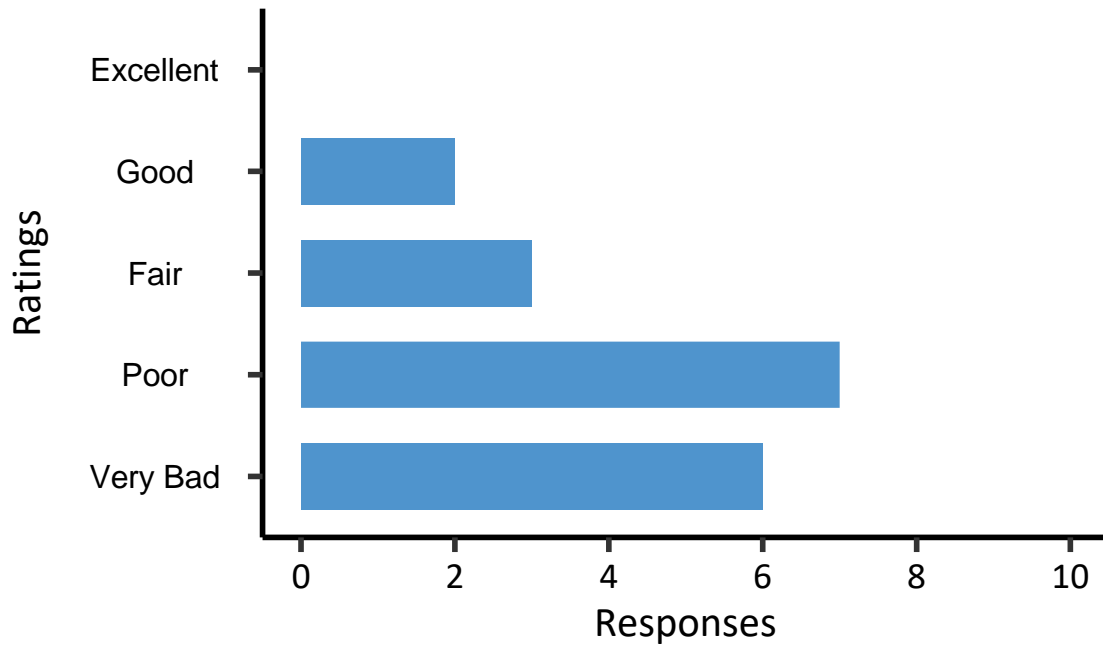
**Q10.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- find this one impossible to rate accurately -- newly colonizing seedling?
- One shoot and high water clarity
- Really hard to tell without context as to where in the bed this occurs. Could well be located at the shallow edge.
- Just bad
- 1-2% ZM but good WQ and little algae
- Very little eelgrass visible.
- Rating strictly based on shoot density, but that being said without knowing the context of this photo it is difficult to judge. Water clarity looks good and there may be many reasons why eelgrass is currently not present. Perhaps this shoot is the first to colonize this area
- Qualitative Visual Assessment
- what eelgrass?
- Low everything
- only one seedling in frame, chance of survival is minimal
- No grass?



Q11.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 16.7





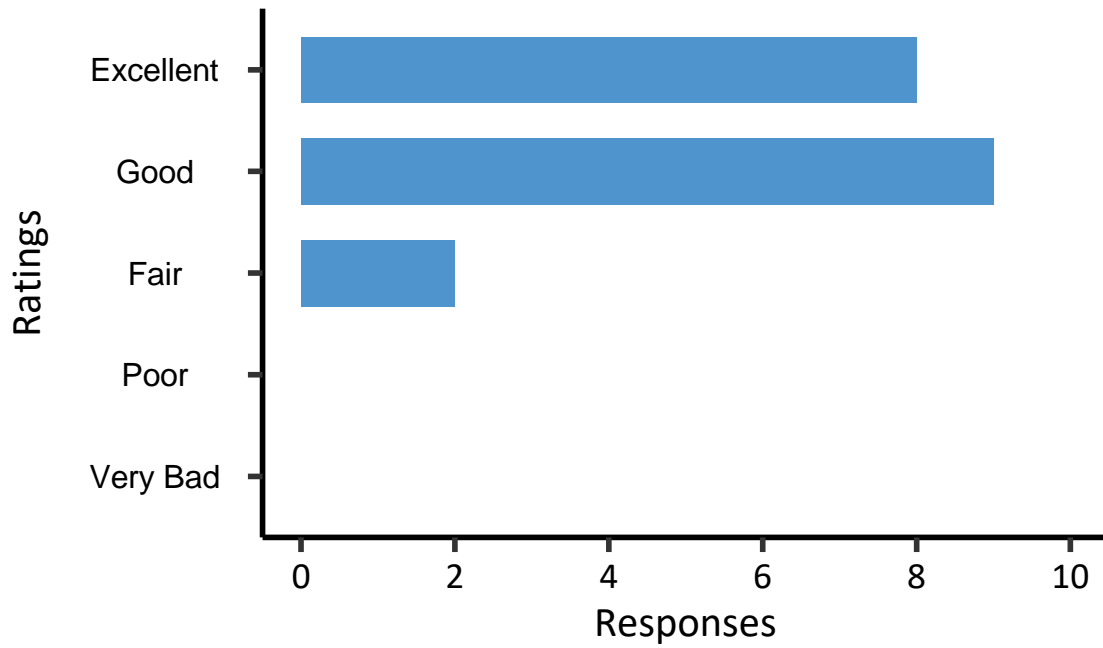
**Q11.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Few healthy looking plants, macro algae
- green leave, low epiphytes, sparse density
- Qualitative Visual Assessment
- low levels of plants cover (15%) but looking healthy; major problems of drift macro algae
- Eelgrass is alive however there is a lot of algae
- Eelgrass appears healthy but sparse. Hard to tell if dark tissue within quad is necrotic eelgrass or macroalgae.
- borderline poor to fair, fair water clarity, but low shoot density, plus the presence of drift algae that may hinder eelgrass growth



Q12.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 77.4



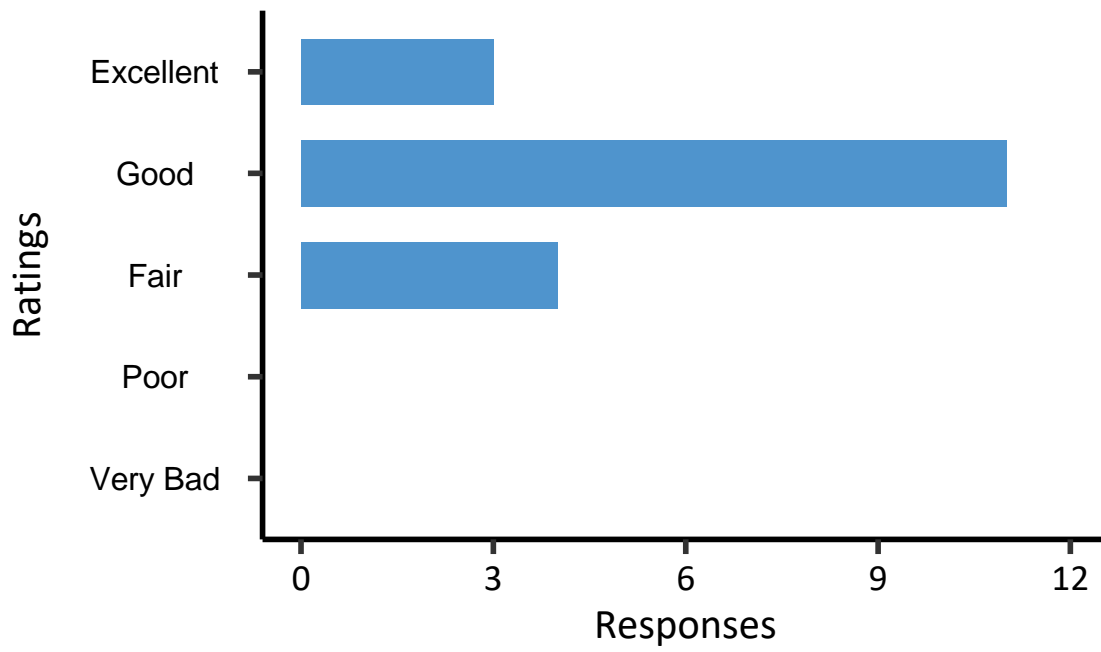
**Q12.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- borderline good to excellent, high shoot density, water clarity a bit compromised, perhaps some evidence of disease
- high density, few epiphytes, tall growth
- high cover, low to moderate epiphytes
- Great density and healthy looking leaves
- within the frame, quite dense and clean blades
- Good clarity and high density
- dense plants within quadrat, minor macro algae
- Qualitative Visual Assessment
- Plants are very dense and seemingly tall. Blades are wide. There are reproductive shoots!
- some loss or disturbance



Q13.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 59.9



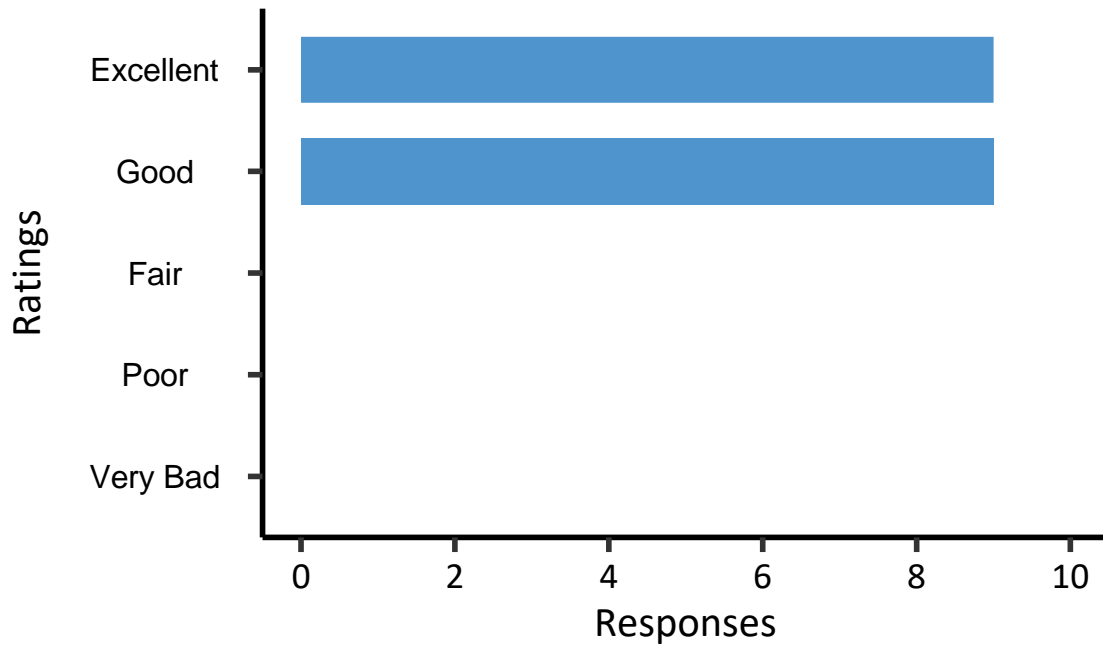
**Q13.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Qualitative Visual Assessment
- borderline excellent to good, plants look healthy, shoot density is good, though might be lower than previous picture due to the high number of reproductive shoots
- Good density and clarity
- some bare spots
- Good density, healthy looking plants
- About 60% cover but algae mixed in the bed



Q14.1 - How would you rate the health of the eelgrass in this frame?

EHI: 86.8



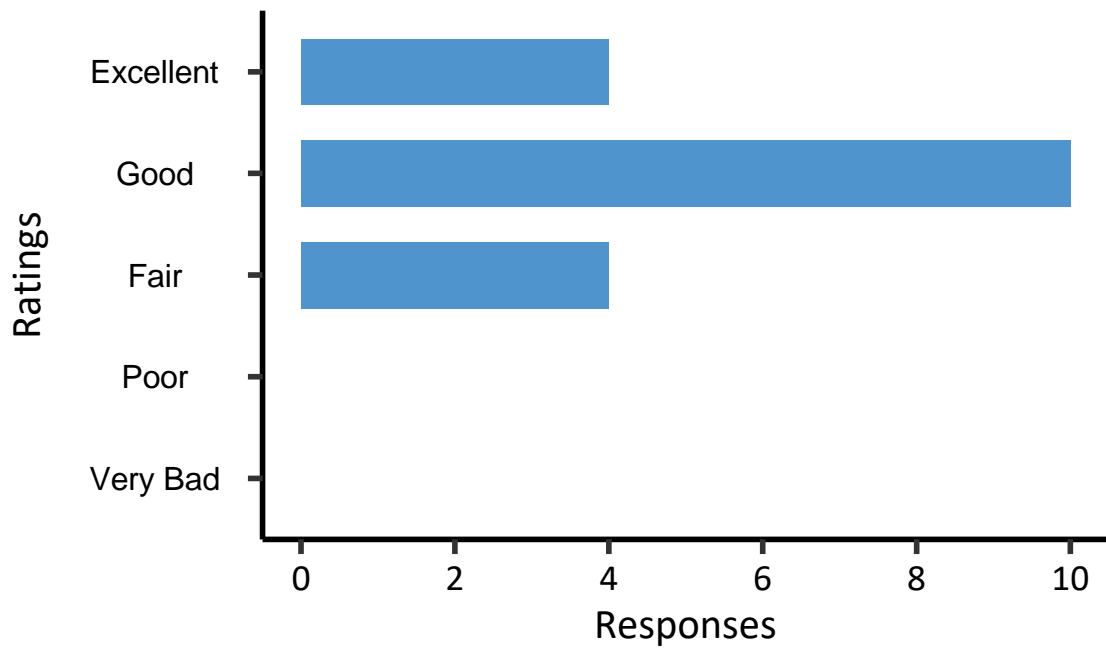
**Q14.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Blades seem longer and more dense, but that could just be a function of longer blades making it look like there are more plants, when the density is actually similar.
- dense, unfouled shoots, relatively high percent cover
- dense plants
- great water clarity, healthy long thick green leaves
- green leaves, dense, no algae
- Qualitative Visual Assessment
- dense, long leaves in good health
- Great density, healthy looking plants
- Water clarity could be better but overall good density and clarity



Q15.1 - How would you rate the health of the eelgrass in this frame?

EHI: 84.5





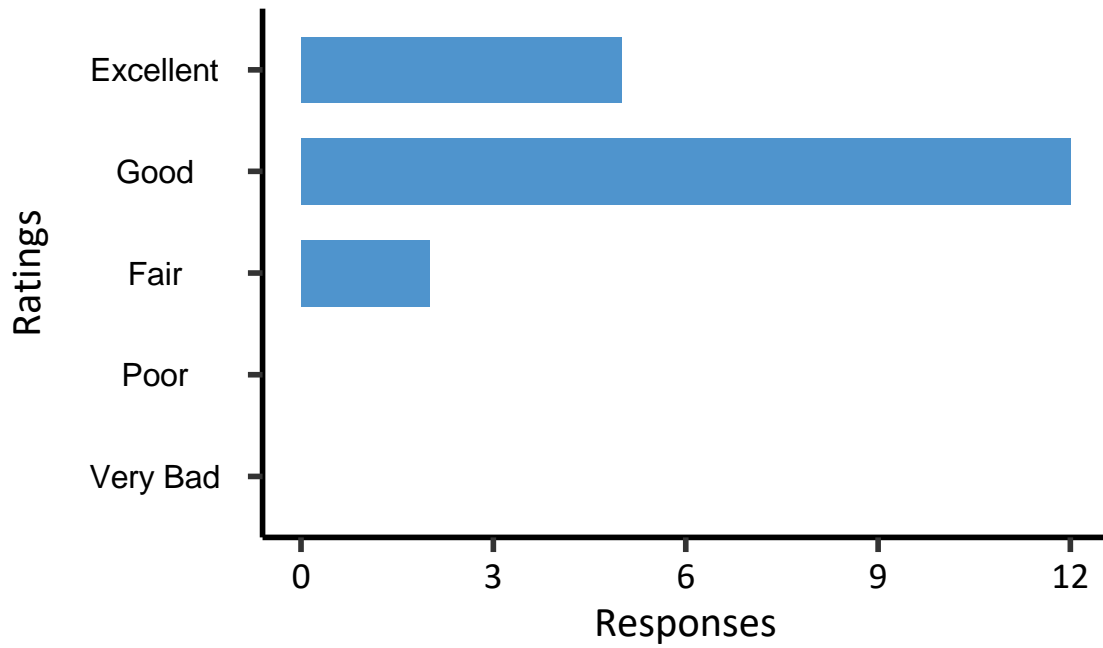
**Q15.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- About 60% cover but drift algae visible
- green leaves, dense, little algae
- would have been excellent but for the bare spots
- A "high Good" - dense and healthy looking plants
- have never seen excellent.
- good water clarity, healthy looking plants
- Qualitative Visual Assessment
- borderline excellent/good, water clarity is better than prior picture, plants look healthy and shoot density is good



Q16.1 - How would you rate the health of the eelgrass in this frame?

EHI: 69.0



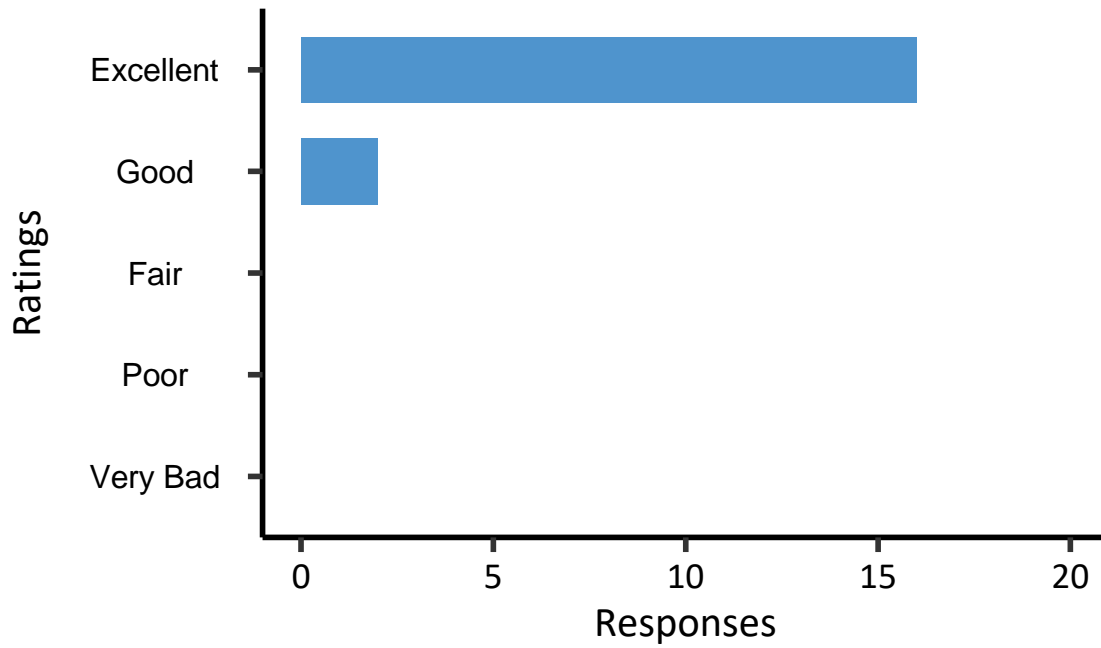
**Q16.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- This bed seems to be on the lower end of the Excellent spectrum. Eelgrass is a decent height although there is definitely some bare space in the quadrat. Grass isn't covered with epiphytes or algae.
- Some bare ground visible, otherwise would have rated it excellent
- Qualitative Visual Assessment
- borderline good to excellent, shoot density is pretty good and the shoots are a healthy green color
- about 30% of bottom is visible through the blades
- Shoots look healthy and green high density
- Excellent density and healthy-looking plants



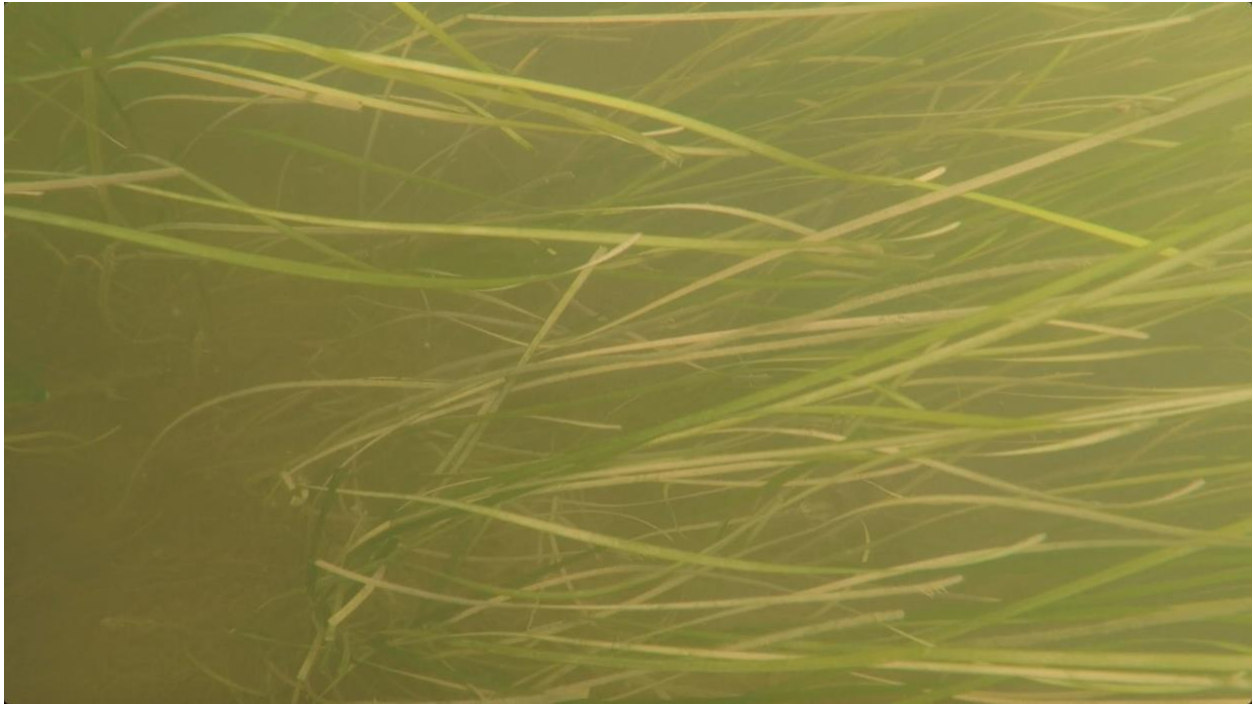
Q17.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 85.1



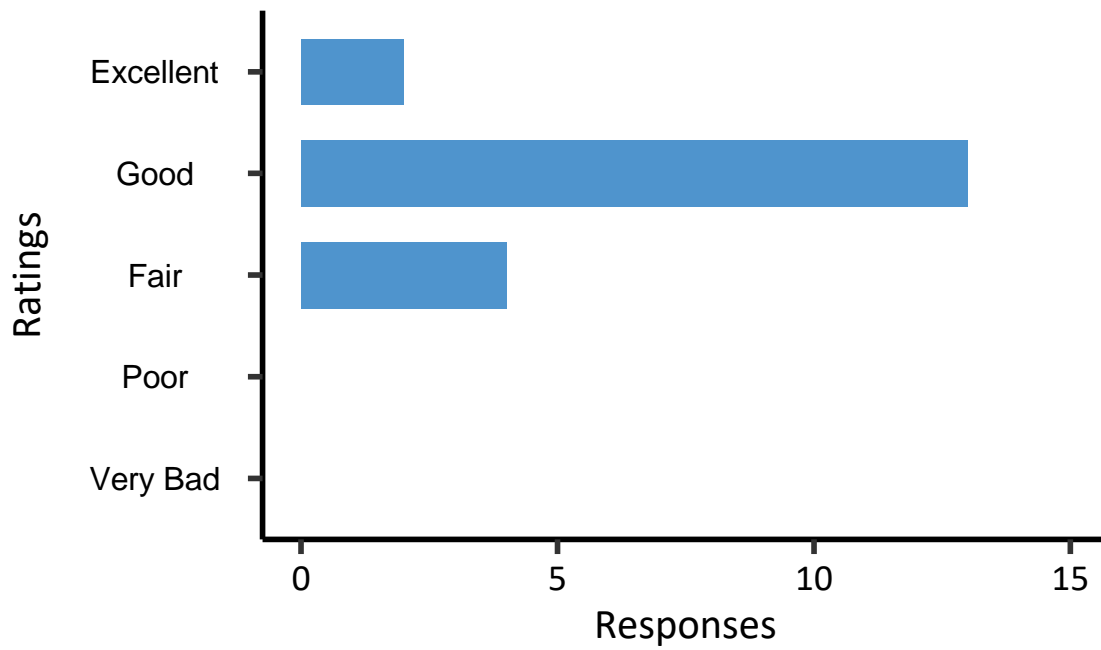
**Q17.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- long, dense, clean
- really healthy lush thick meadow, no evidence of disease or epiphytes on leaves
- Qualitative Visual Assessment
- Great density, healthy looking plants
- Dense bed water clarity not bad
- same comments as for prior photo, with even higher percent cover
- appears to be healthy and very dense eelgrass, though it could be the angle of the photo and the length of the blades.
- I cannot even discern the quadrat
- Thickest picture yet
- bright green leaves, no macro algae



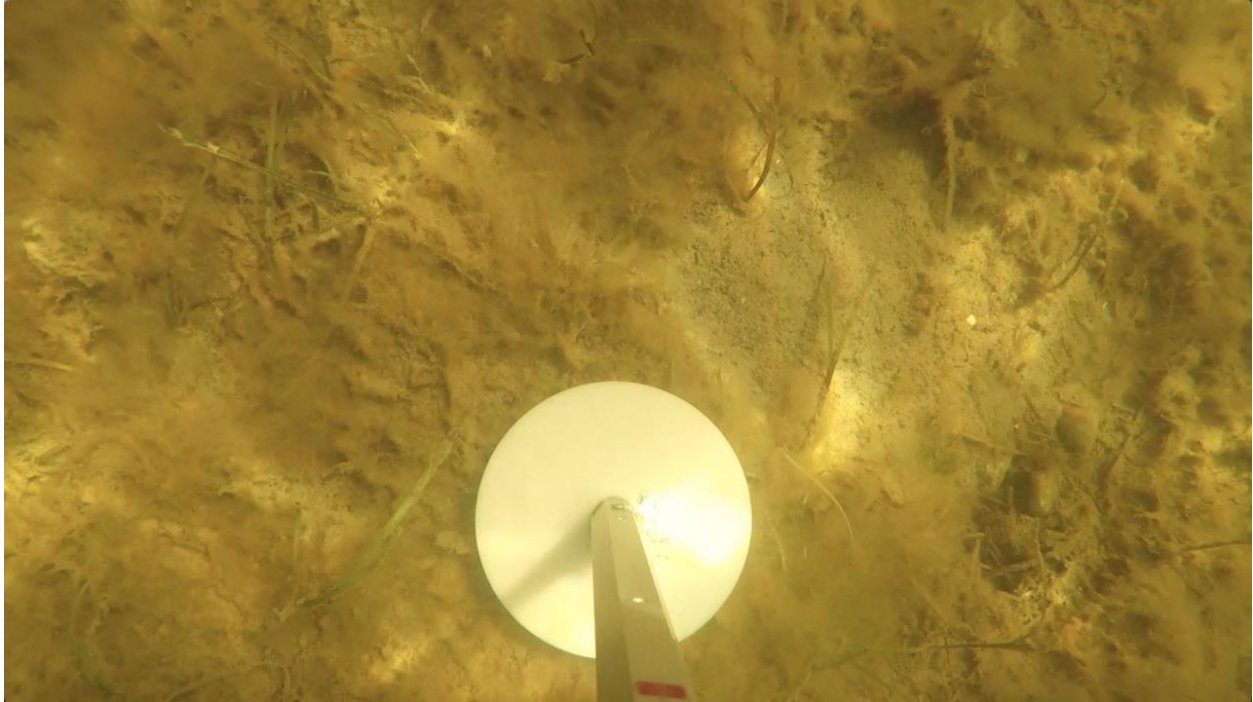
Q18.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 82.2



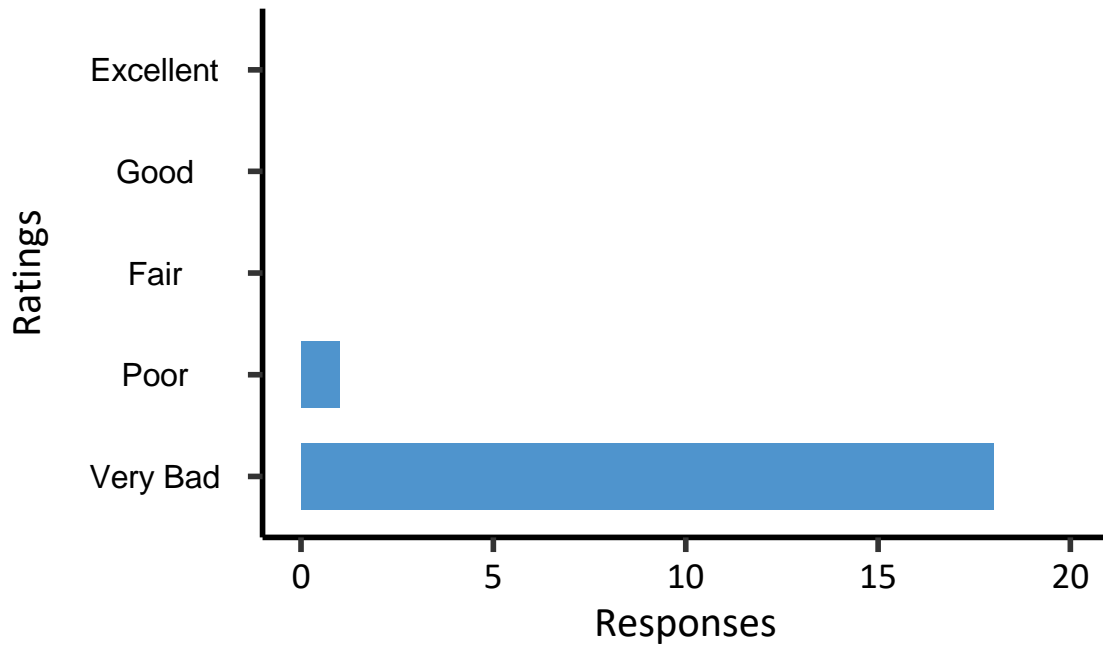
**Q18.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- dense and bright green blades, no macro algae
- The plants here are tall which puts this in the Good category. Many of the blades look muddy and broken though.
- Qualitative Visual Assessment
- Water quality not optimal
- Pretty dense looking, healthy looking plants
- Large amount of aboveground biomass with lengthy shoots and possible epiphytic or sediment cover demonstrates stress.
- not much green
- shoot density not quite excellent and some yellowing of shoots, could be taken late in growing season
- bare spot ; leaves whitefish brown with epiphytes
- clean blades, large plants



Q19.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 5.18





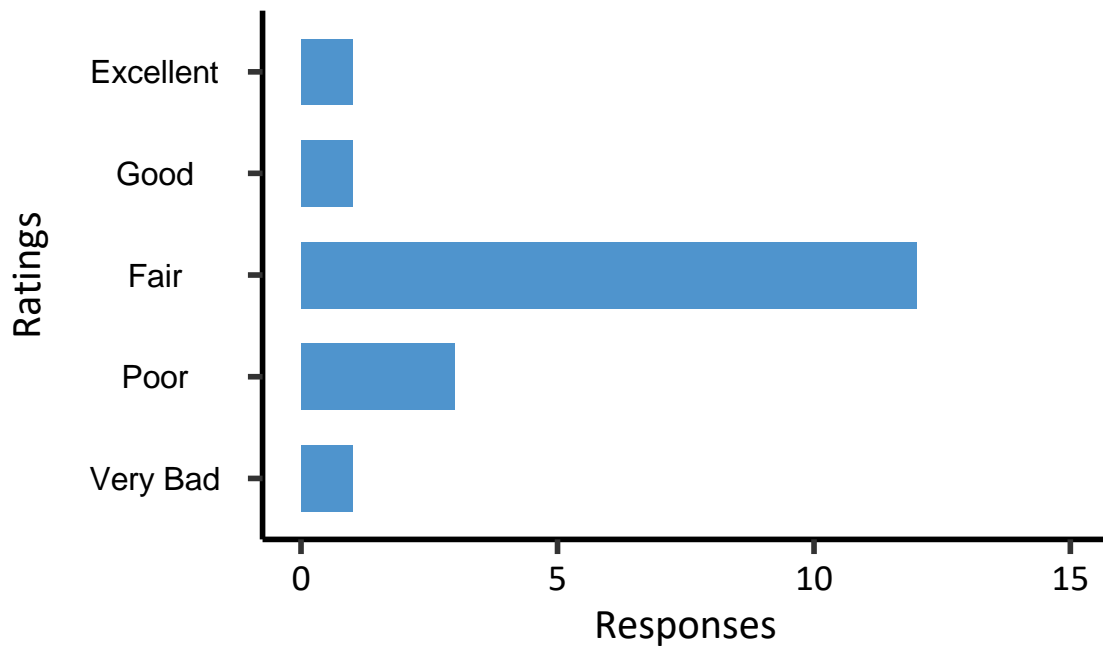
**Q19.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- About %5 cover and smothered by algae; yuck
- extensive epiphytes, algal growth some evidence of disease
- heavy epiphytes, very low density
- Lots of algae and low density
- very few shoots, high macro algae cover
- Comments same as for prior photo.
- heaps of epiphytes or algae
- just a couple of struggling shoots
- Qualitative Visual Assessment



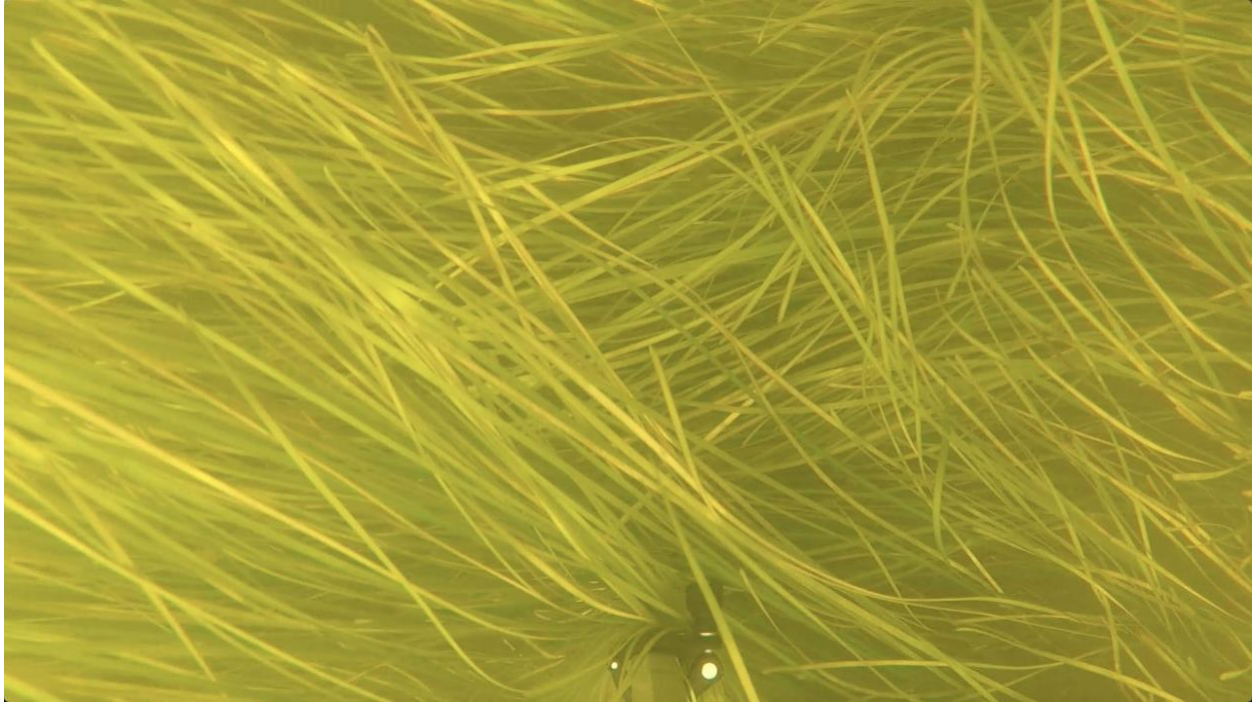
Q20.1 - How would you rate the health of the eelgrass in this frame?

EHI: 39.8



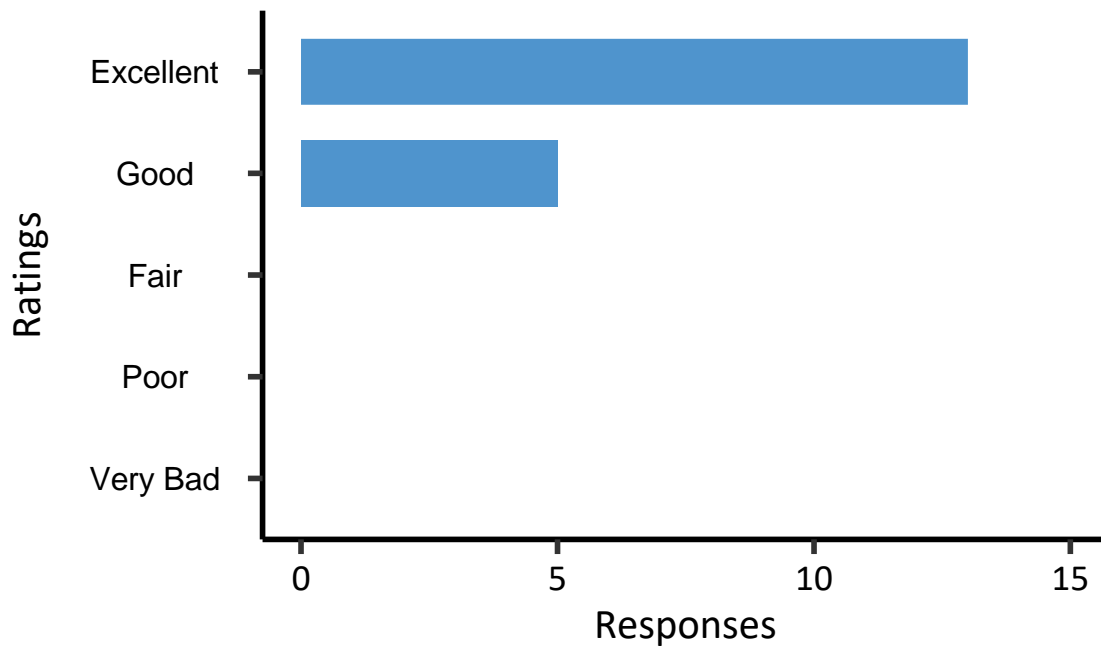
**Q20.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- high end of fair to low end of good, shoot density in this particular spot is fair, but water clarity and the shoots themselves look good
- Qualitative Visual Assessment
- sparse bed
- green leaves, moderate density, low epiphytes
- blades look healthy, but sparse cover
- slightly more bare spots and cloudier water
- Under 50% cover with some macro algae present
- Low clarity and density



Q21.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 79.6



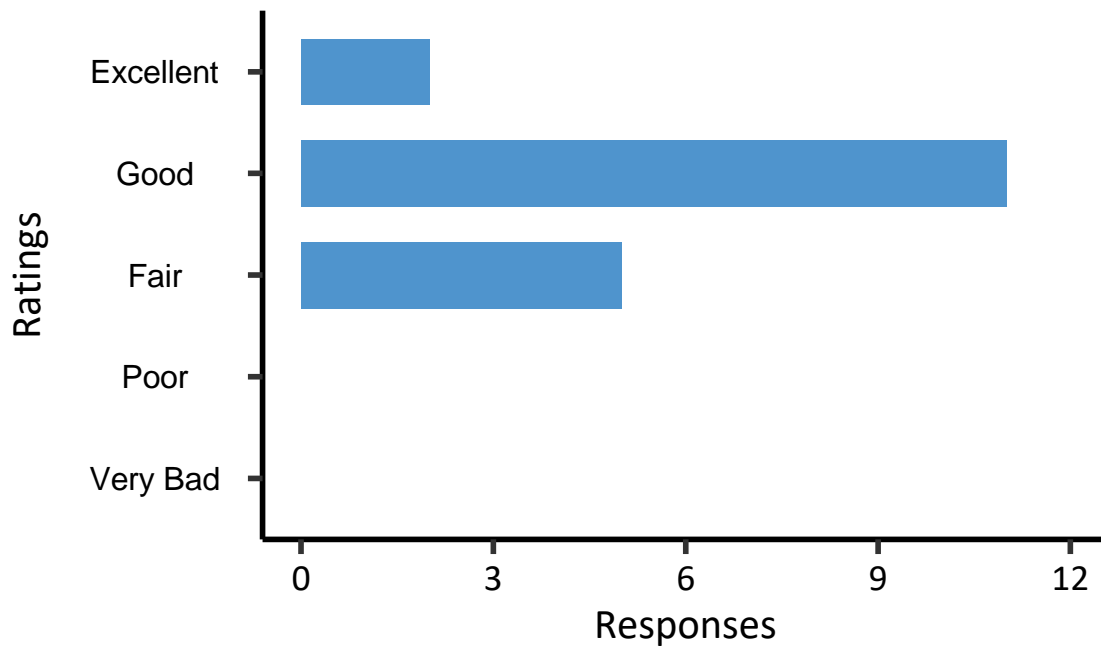
**Q21.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Plants look really healthy and shoots are long but yellowish tint to water may be affecting health
- nice thick meadow with good water clarity
- Dense plants with little to no epiphytes or macro algae
- maybe good to excellent, high density low epiphytes blades look healthy
- Great density, healthy looking plants
- high good but not excellent due to bleached blades
- Qualitative Visual Assessment



Q22.1 - How would you rate the health of the eelgrass in this frame?

EHI: 63.6



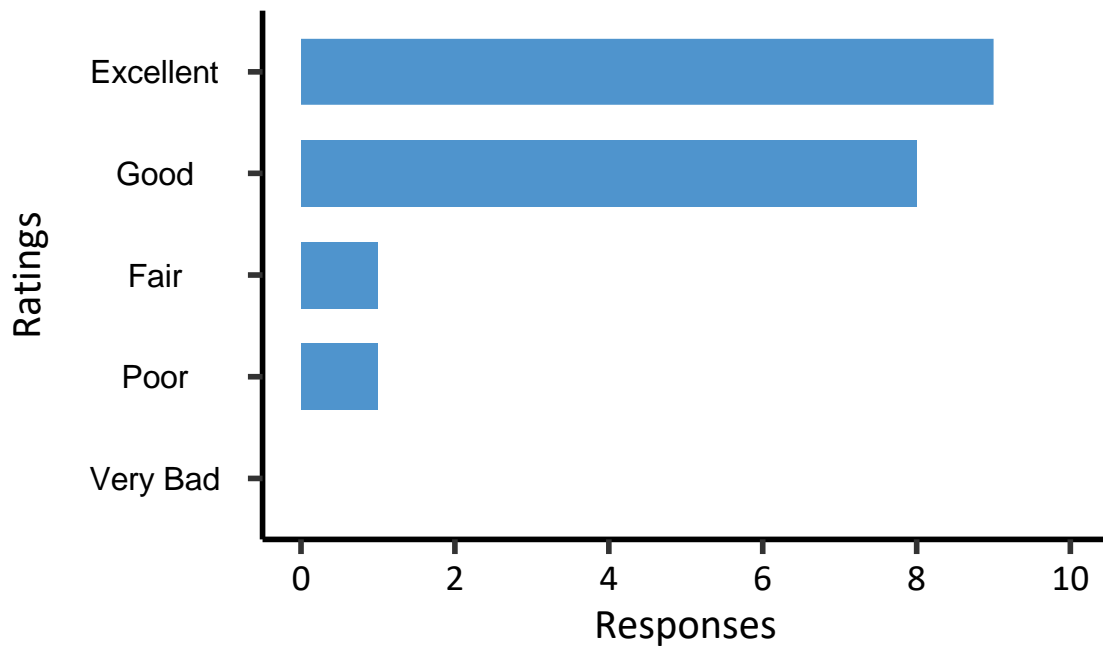
**Q22.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Yellow water color not great for eelgrass
- Qualitative Visual Assessment
- wasting disease?
- I don't see the frame; looks to be about 40-50% cover
- green eaves, high density, low epiphytes
- Plants look healthy and thick, water clarity seems good
- good density and healthy looking plants
- I've never seen excellent



Q23.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 71.7





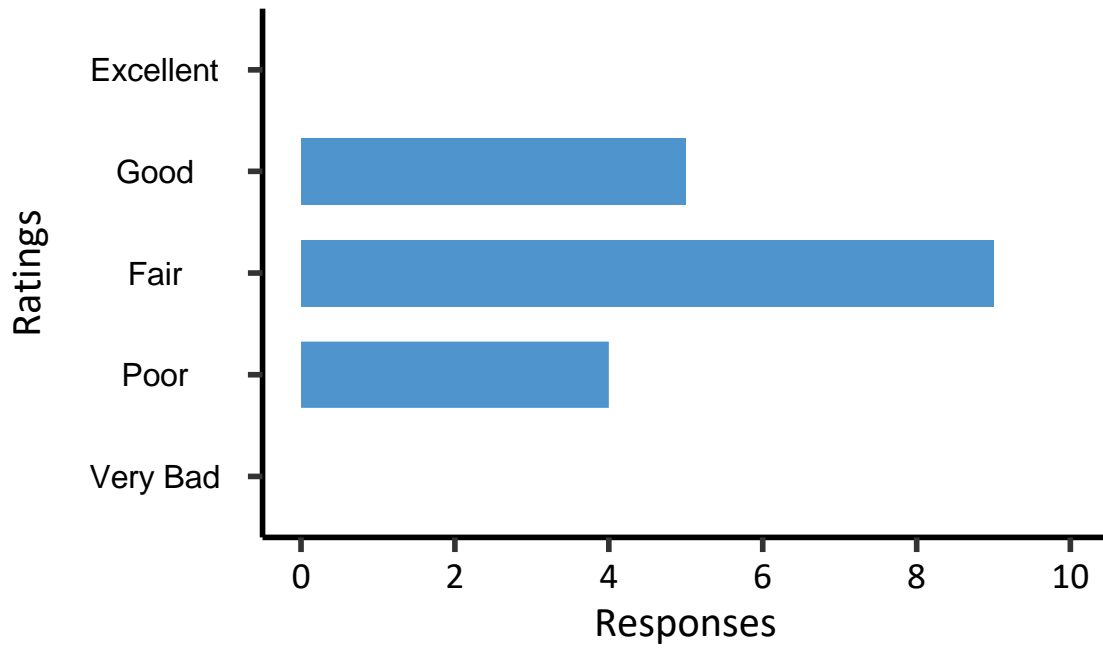
**Q23.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- high fair, low good
- dense grass, though water is a bit cloudy and some epiphytes are evident
- Lower end of good, high density but high red algae or other epiphytes
- some wasting disease or leaves in poor shape
- Water is yellow not sure of health of plants without seeing them
- Qualitative Visual Assessment
- borderline excellent/good, hard to infer a scale from the photo, but vegetation looks thick, could be later in the growing season so the yellowing of the leaves
- This bed is on the low end of the Excellent spectrum (maybe high end of Good?) simply because the plants/canopy seem dense. It's difficult to truly assess without seeing more.
- high cover, moderate epiphytes, wondering about water clarity though
- Great density, plants don't look as healthy as some of the others
- Thick cover and minor macro algae



Q24.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 47.8



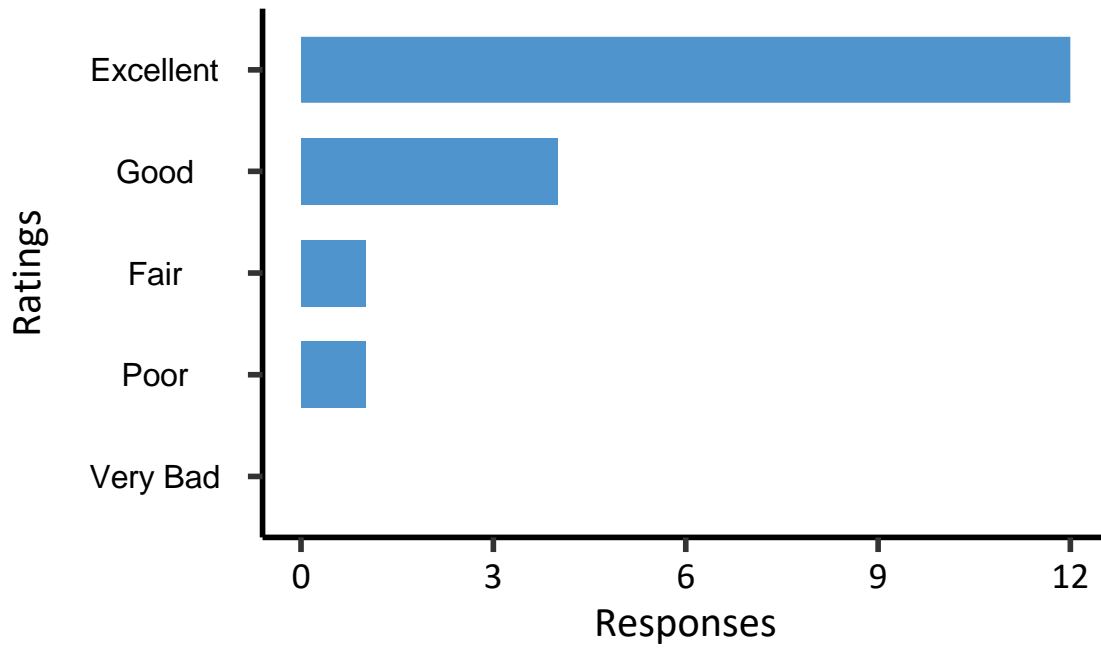
**Q24.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Good density, maybe some kind of epiphyte?
- smaller plants, some epiphytization, lower water clarity/ light limitation
- Good plant cover but epiphytes and poor WQ
- borderline fair to good, shoot density is good, some evidence of epiphytes and water clarity is a bit cloudy, could be late season photo
- green leaves, dense, some algae
- Qualitative Visual Assessment
- High epiphytes
- More epiphytes are visible and water seems less clear, so rating it worse than the previous photo.
- Moderate density but fouled (hydroids, tunicates?), with lower water clarity than prior image
- Plants look ok but water quality not great



Q25.1 - How would you rate the health of the eelgrass in this frame?

EHI: 76.0



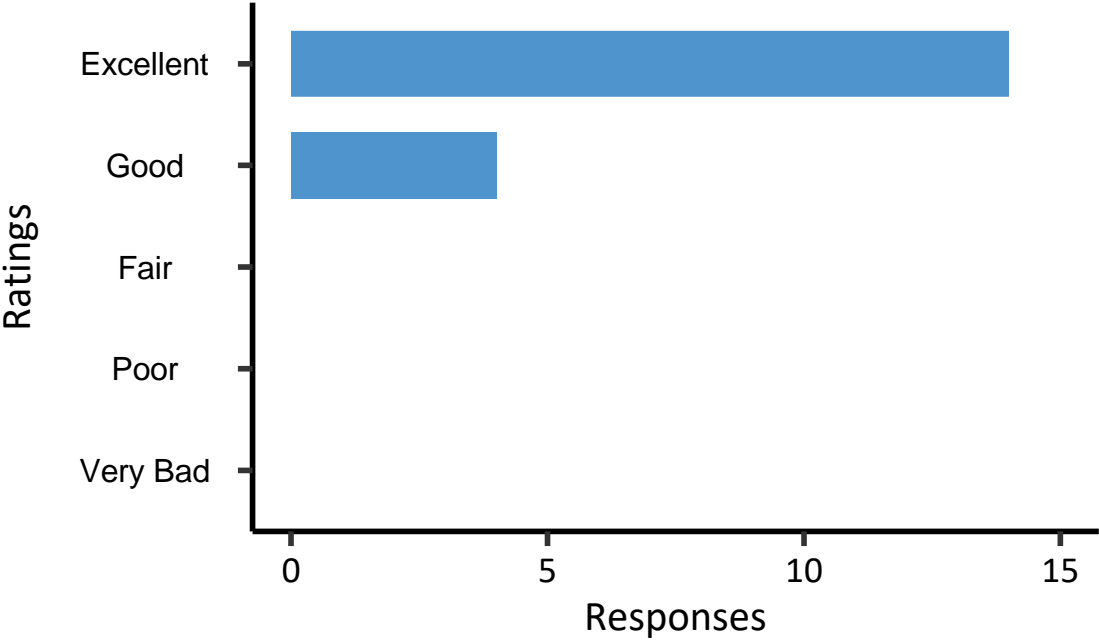
**Q25.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Hard to know if yellowing of tissue is an artifact of the photo, but generally, high density and cover with minimal fouling demonstrates a healthier bed
- Great density
- upper end of good
- seems very dense but some of the grass seems as though it's been grazed on
- dense, clean plants
- Qualitative Visual Assessment
- high shoot density and shoot color is good, no epiphytes or disease
- dense, no algal or growth on blades



Q26.1 - How would you rate the health of the eelgrass in this frame?

EHI: 63.9



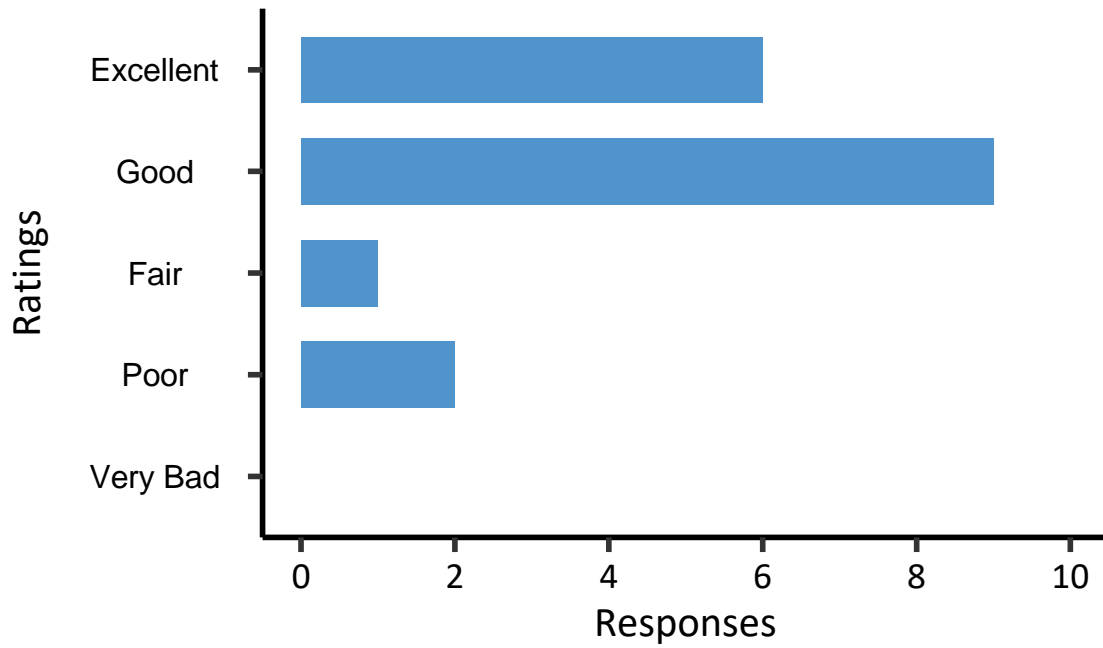
**Q26.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- dense, long healthy looking leaves
- lush meadow with great water clarity
- bright green, very dense, no algae, high water clarity
- luxuriant, but watch out for jellyfish!
- Great density, healthy looking plants
- high cover; hard to discern but maybe calcareous epiphytes
- Same comments as for prior photo
- Qualitative Visual Assessment
- Long thick, dense vegetation



Q27.1 - How would you rate the health of the eelgrass in this frame?

EHI: 42.8





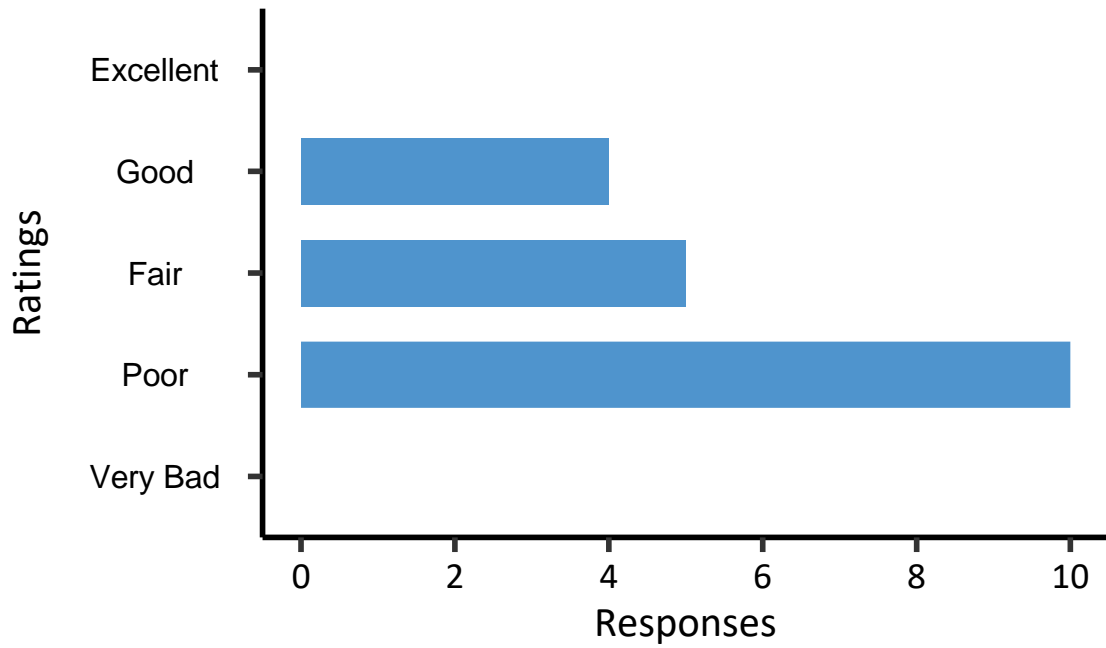
**Q27.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Good clarity shoots look short and lower density but ok
- Thin
- borderline excellent to good, shoot density is good, while shoot/leave color and water clarity are excellent
- Short grasses 60% cover with some macroalgae but good WQ
- Good density and coverage, lack of macroalgae
- Qualitative Visual Assessment
- similar to first photo, good percent cover and clean plants, though density and canopy height could be higher
- green leaves, dense, little algae



Q28.1 - How would you rate the health of the eelgrass in this frame?

EHI: 25.4



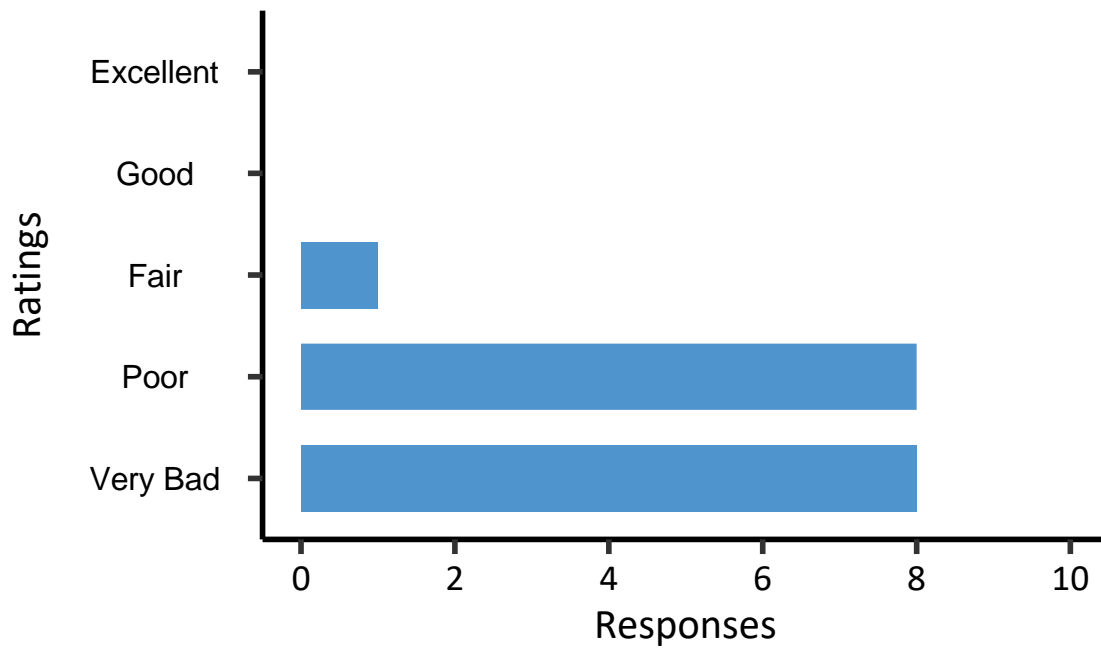
**Q28.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- densely vegetated with reduced water clarity
- water clarity is fair and shoot density is fair, hard to see the actual health of the individual shoots
- About 25% cover but poor WQ and either dead Zm or live red algae on bottom
- moderate density and height
- A "low fair" - Ok density, looks like macroalgae
- Bed falls in the upper end of Poor (maybe low end Fair?). Plant density is low and blades look small and muddy.
- no sign of wasting disease, moderate density and blade length
- Low clarity density low to moderate
- Qualitative Visual Assessment



**Q29.1 - How would you rate the health of the eelgrass in this frame?**

**EHI: 7.77**



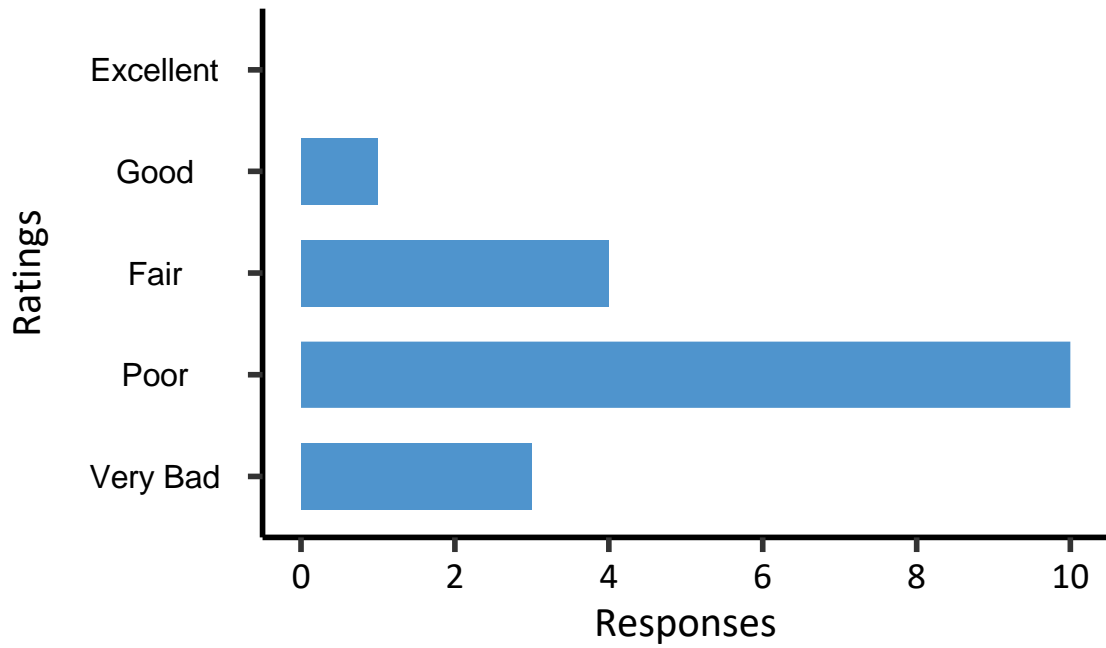
**Q29.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Low density and clarity
- few blades, poor water clarity
- hard to judge, but water clarity is poor and shoot density seems low, but what I can see of the eelgrass looks free of epiphytes and disease
- about 10% plant cover with poor WQ
- can't tell, poor image
- Little too turbid to tell, but looks like a decent plant there... probably poor coverage/density
- Qualitative Visual Assessment - Hard too see
- hard to say. Water is very cloudy. Not a lot of grass visible.
- sparsely vegetated
- low cover, very poor water clarity



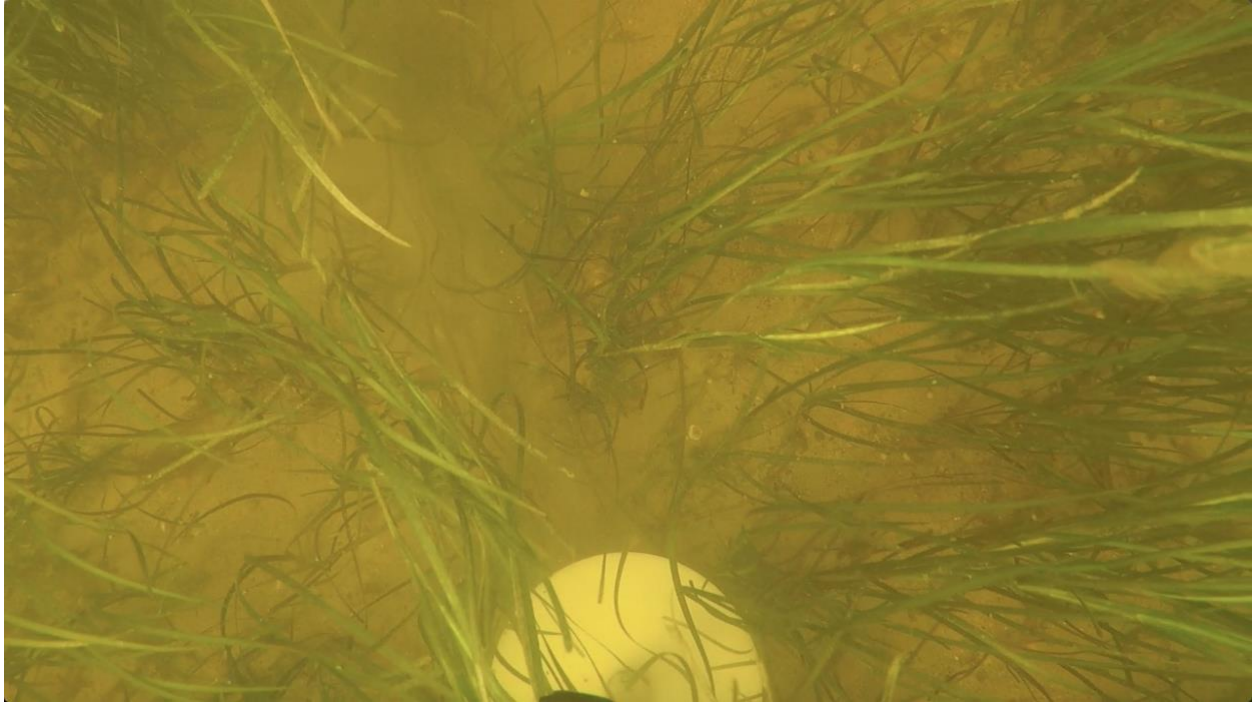
Q30.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 33.0



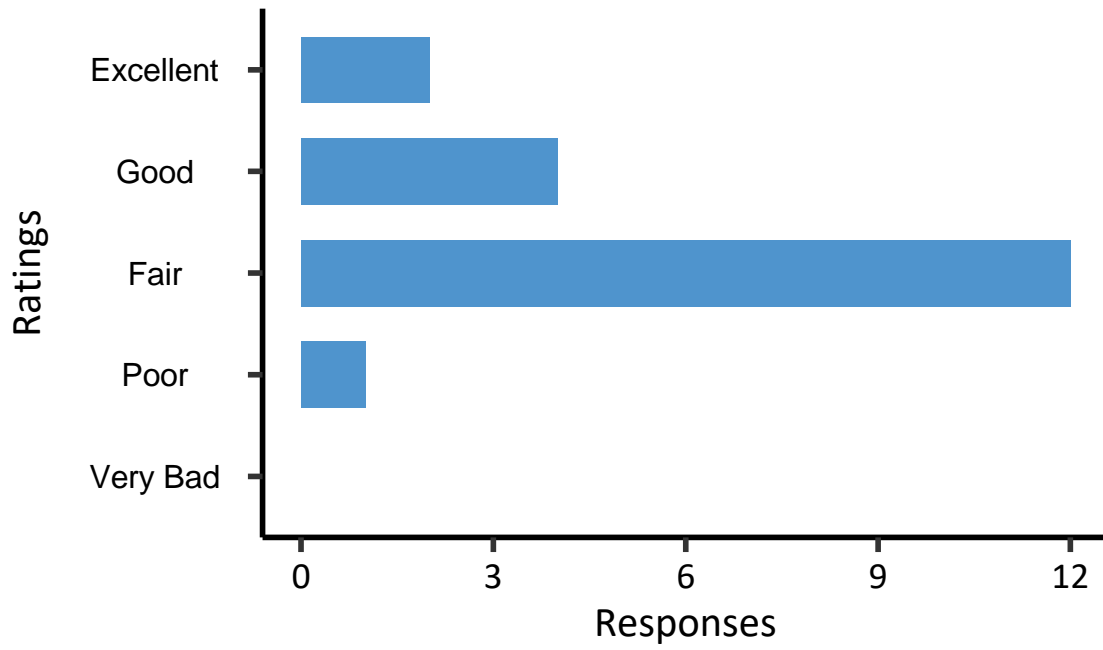
**Q30.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- I am not sure, given I do not know the size of the frame. I rated it fair as it seems the shoot density is not terribly high, also I believe I can see epiphytes
- Poor density
- About 25% cover by plants but poor WQ and some epiphytes and drift algae
- moderately dense
- Qualitative Visual Assessment
- limited water clarity and low shoot density



Q31.1 - *How would you rate the health of the eelgrass in this frame?*

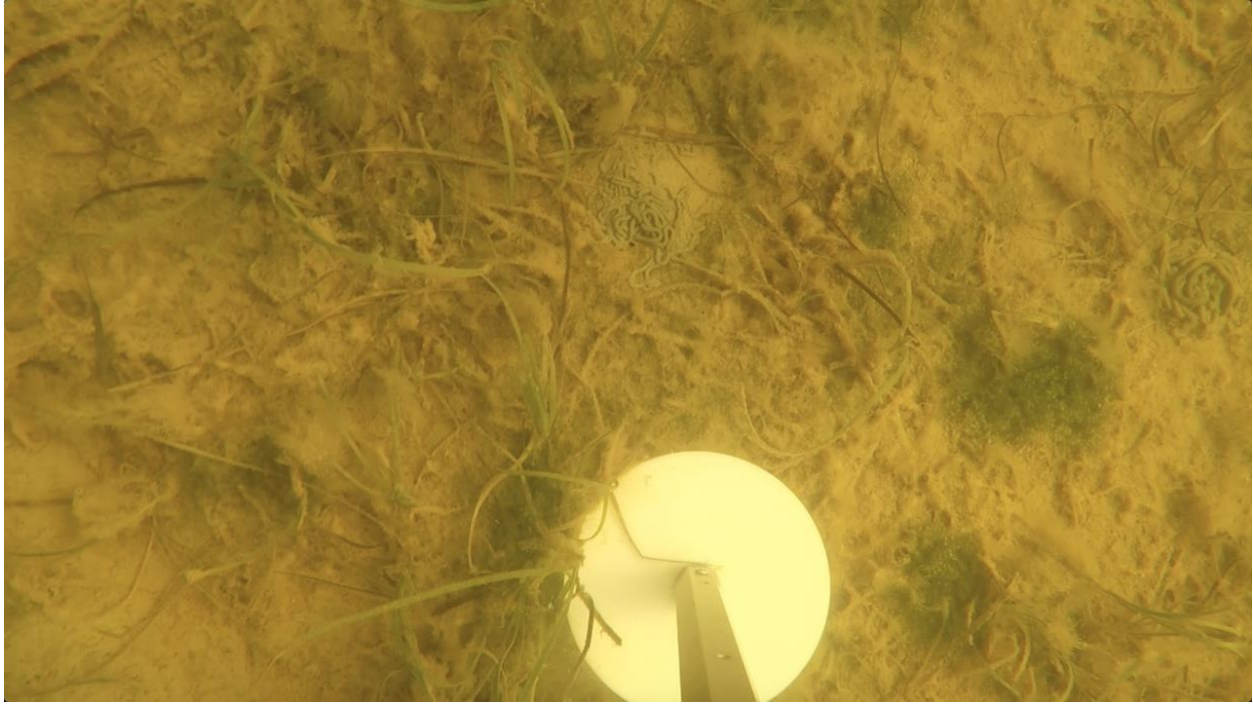
EHI: 49.6





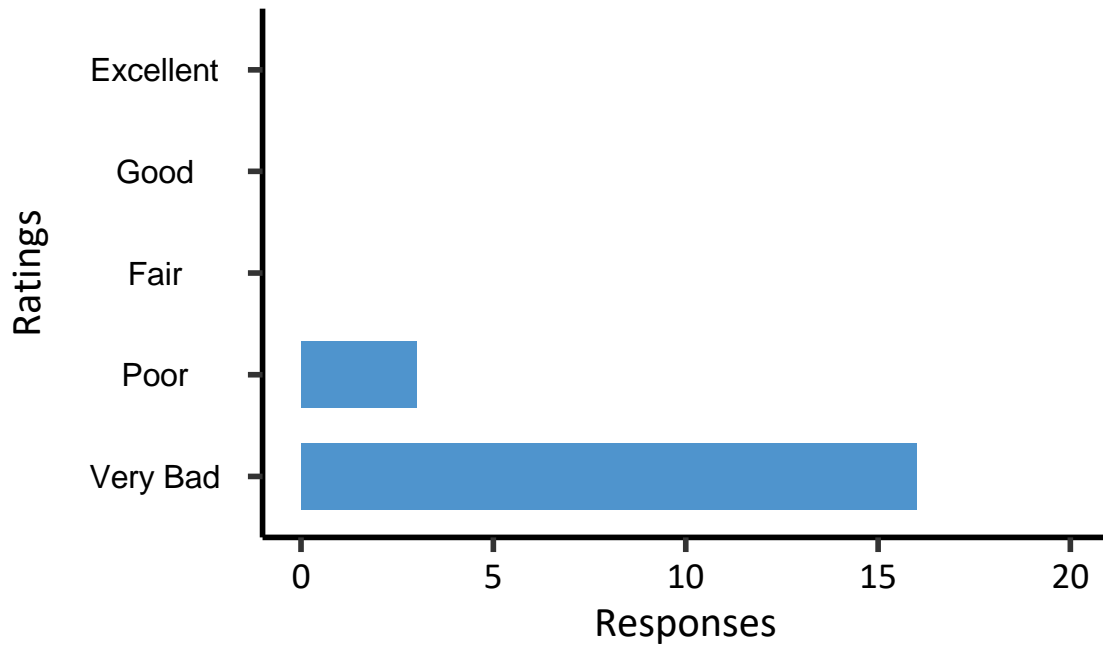
**Q31.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- 60% cover with clear water and some algae
- This bed is on the low end of the Excellent (or high end Good) spectrum. Density is pretty good and blades look green and healthy
- Qualitative Visual Assessment
- Eelgrass density is moderate but water is slightly yellow
- High percent cover, but could be more dense. Also, blades are clean without a lot of epiphytes.
- This is borderline fair to good, shoot density is low, but hard to tell the scale without quadrat. Some evidence of epiphytes, but plants look healthy green. Inferring this is Julyish due to presence of reproductive shoots
- green leaves, dense, little algae
- Looks quite sparse but not smothered by seaweed



Q32.1 - How would you rate the health of the eelgrass in this frame?

EHI: 9.89



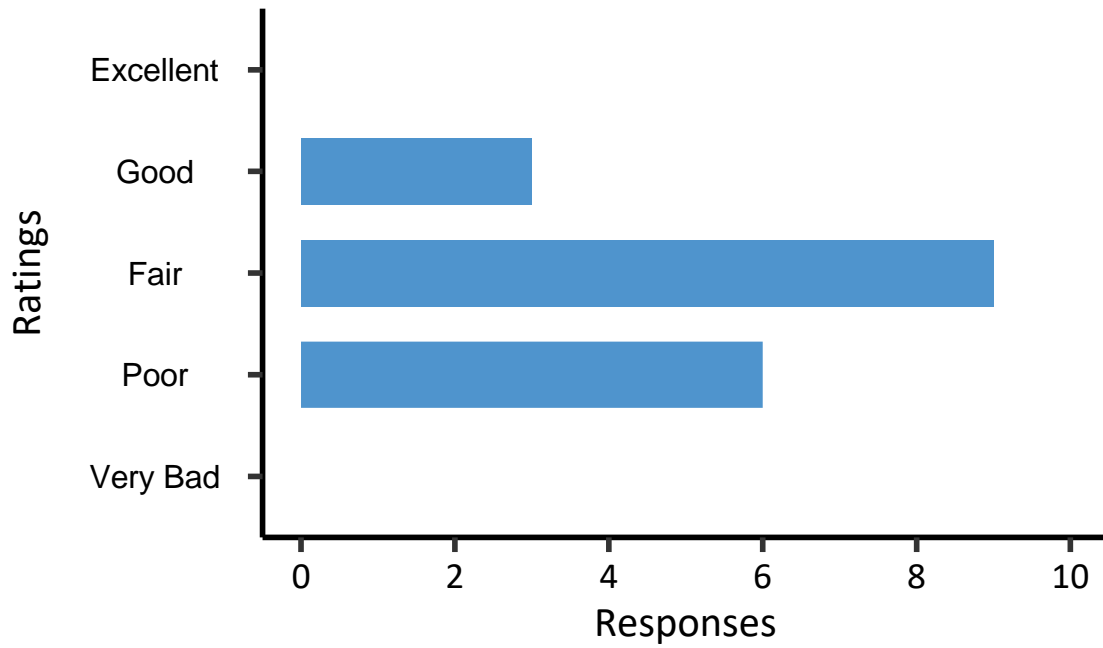
**Q32.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Low density with shoots lacking buoyancy. Benthic surface appears enriched.
- epiphytes and algae dominated
- About 20% cover but smothered by algae and chlorobium patches indicating reduced sulfur is being released and metabolized at sediment surface
- low shoot density and the presence of algae and epiphytes
- Dead and silted
- Just a couple of struggling shoots among the macroalgae
- There's not much eelgrass and what is there is covered in mud, algae, and epiphytes.
- Qualitative Visual Assessment
- Everything is dead



Q33.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 28.0



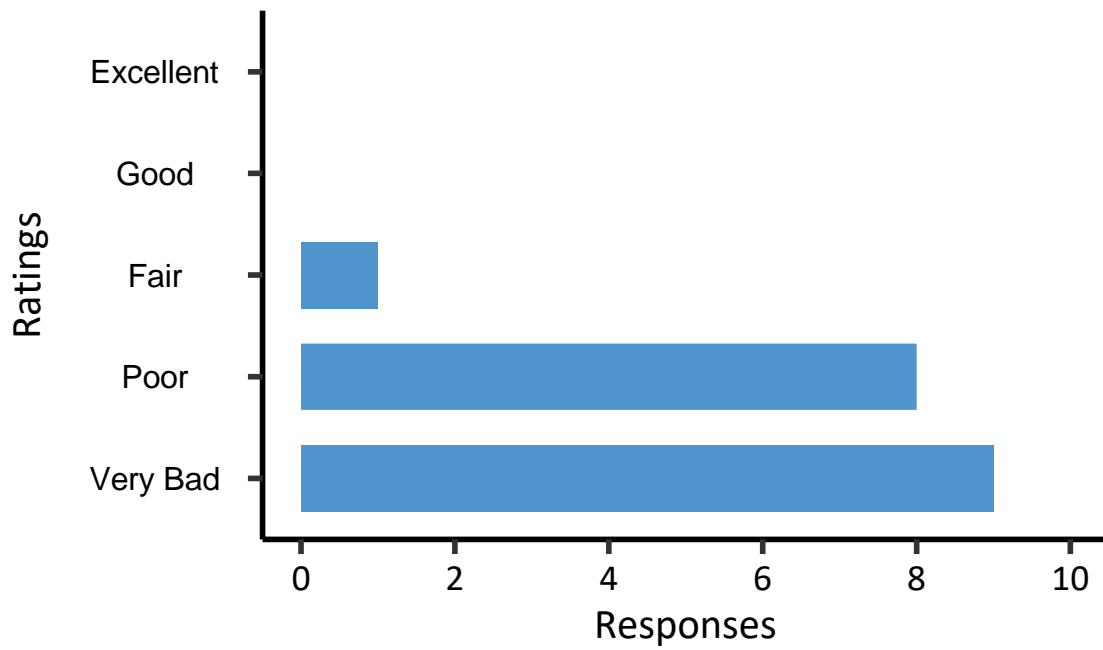
**Q33.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- fair to good, shoot density is a bit sparse to be good, but shoot color and water clarity seem good
- decent density, plants look ok
- Qualitative Visual Assessment
- low density, physical damage
- About 50% cover of vegetative plants some algae present



Q34.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 12.9



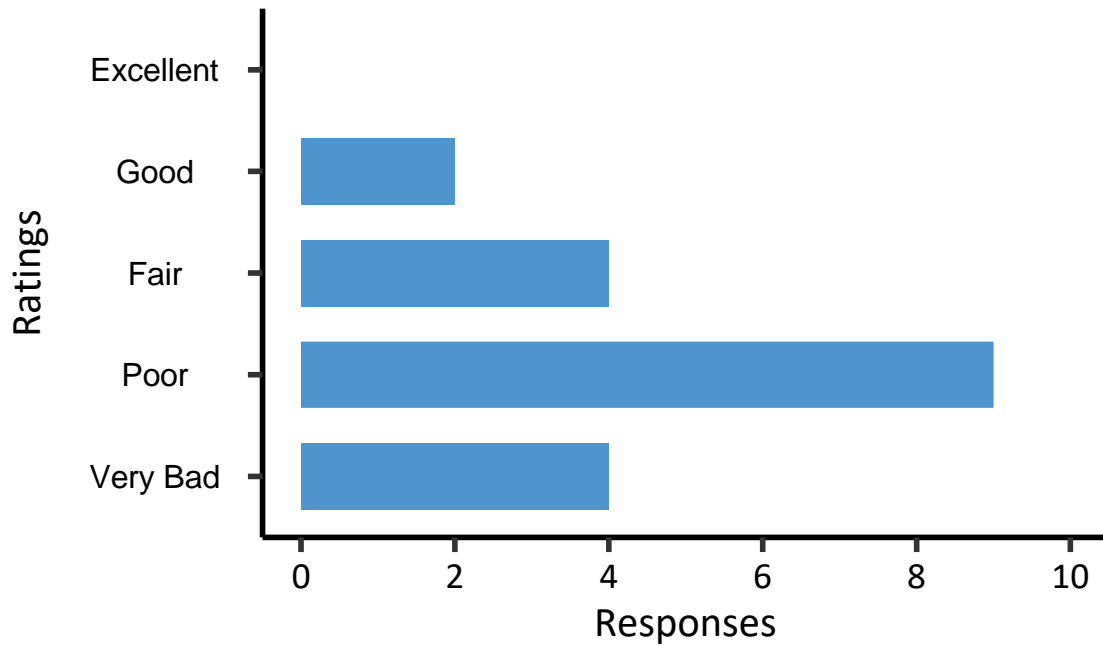
**Q34.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Lots of algae smothering plants
- Highly epiphytized
- extensive epiphytic growth and evidence of wasting disease
- looks like 10% cover and covered by nasty algae
- dense epiphytes
- Whew... macroalgae
- Clearly enriched environment.
- algal/epiphyte growth
- Qualitative Visual Assessment
- eelgrass present, though covered in algae



Q35.1 - *How would you rate the health of the eelgrass in this frame?*

EHI: 31.9





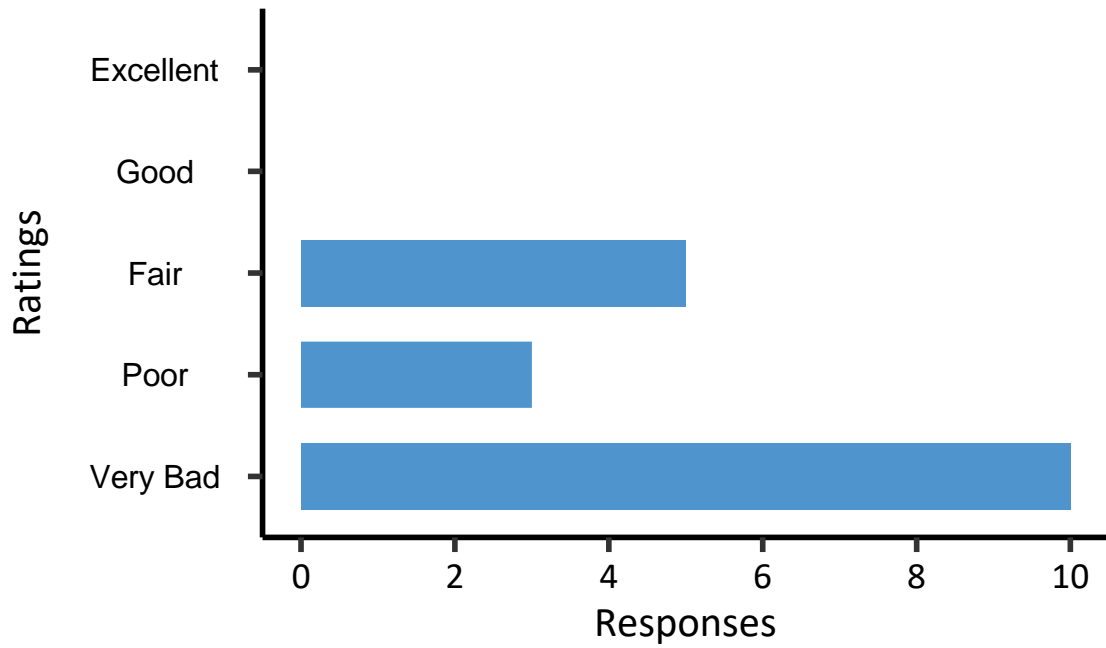
**Q35.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- 25% cover and clean plants but poor WQ
- Low density and poor water quality
- low cover, poor water clarity
- Qualitative Visual Assessment
- low shoot density and more limited water clarity
- A bit too turbid, but looks like low density bed
- Hard to tell from image quality, but greater density and cover than in prior photo, though fouling of shoots (sediment, bryozoans?) still evident
- This bed is on the low end of the Poor spectrum. The turbidity makes the grass hard to see but there are definitely a number of plants there which is good. Blades look covered in mud and epiphytes though.
- not a lot of coverage, the plants in frame look healthy, poor water clarity



Q36.1 - How would you rate the health of the eelgrass in this frame?

EHI: 16.9



**Q36.2 - Why did you select this rating? If you would like to use a specific value (1 - 100) to rate the eelgrass please include that here. (optional)**

- Qualitative Visual Assessment
- About 15% cover and enshrouded by macroalgae
- low shoot density, colored water and some epiphytes
- sparsely vegetated and reduced water clarity.
- Very sparse
- Bad density & coverage, hard to tell - but looks like poor biomass
- Same as last reponse
- very low density

## Appendix C: Eelgrass Video Monitoring SOP (In Progress)

STANDARD OPERATING PROCEDURE  
FOR VIDEO MONITORING OF EELGRASS  
FOR ENVIRONMENTAL AND PHYSICAL  
CONDITIONS

SOP 0.1  
Revision 0.1

November 6, 2019  
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### POINT OF CONTACT:

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### I. OBJECTIVE

In preparation for assessing eelgrass, a procedure is outlined to observe plants and collect limited physical data from an eelgrass bed using videography.

Overview: Video-monitoring protocol for eelgrass beds is described for collecting and recording field data.

### II. MATERIALS AND EQUIPMENT

- Boat (appropriate for sampling needs and exposure) and safety equipment
- Wading or snorkeling gear (if appropriate)
- Eelgrass sampling station map
- GPS unit
- Field notebook and pencils
- YSI Unit
- Video monitoring unit (extension pole, white disk, camera, etc...)
- Marker buoy, line (10 m), and weight
- Depth finder
- iPad
- Aquascope

### III. METHODS

#### A. Video Collection

1. Navigate to the observation site. Sites are determined based on local or expert knowledge of the region's eelgrass distribution, previous research observations, established monitoring site, or observed presence of eelgrass while in the field.
2. At each site, observers locate eelgrass beds and mark using float, line, and weight. Once marked motor into the wind and/or prevailing current towards the edge of the eelgrass bed. Anchor here and record water temperature, salinity, pH, depth, water clarity, water-color, and other environmental readings using an iPad or notebook (see SOP XYZ). Begin a monitoring track for the transect after starting the tracker synced between the iPad and GPS.
3. After preliminary conditions are recorded, prepare the monitoring pole and video camera for observation. Set the camera at the appropriate interval based on water clarity and canopy height. Intervals of 0.25 m from 0.25 to 1.5 m are possible. If conditions are uncertain, start with 0.5 m above the white disk for the first recording. Turn the camera on and state the date, location, observation distance (1 m, 0.5 m, or 0.25 m), and other important information.
4. Extend the camera, pole, and Secchi disk down to the bottom of the eelgrass bed. Raise the pole off bottom enough so the end is not buried and pull the anchor and drift for approximately 50 m. If the current or wind is negligible or opposite the desired direction, motoring or paddling can be used as propulsion.
5. After completing the observation, retrieve and turn off the camera, stop the GPS, and return to the marker buoy. Set the camera at other appropriate distance for the site and repeat the process for a second observation. An observation at 0.25 m is recommended for highly turbid or high-density sites.
6. Follow the same procedure starting at Step 1 for other sites both along the edge of the eelgrass bed, further into it, and at the opposite edge as it approaches the shore.
7. After the trip, download all of the video data to a research computer and upload a copy to the backup hard drive for analysis. Once two sets of the file are saved, clear the SD cards memory, recharge batteries, and clean camera in preparation for the next trip.

#### IV. TROUBLE SHOOTING / HINTS

1. During video monitoring, taking still photos of the observation site, the monitoring

process, and the eelgrass bed are encouraged. Having additional cameras available while the observation camera is in use is a good practice.

2. Substrate can be determined by using the video camera at 0.25 m above the Secchi disk. While anchored turn on the camera and extend the pole down to the bottom of the bay. Gently push the end of it into the sediment and rotate several times. Retrieve the camera and turn it off. If water quality is decent, it should be possible to visual assess the sediment. If not, the sound ranging from coarse chatter (stones) to a fine abrading (sand), provides the researcher with a general idea of grain size and sediment type.

## V. STATISTICAL ANALYSIS AND DATA USAGE

## VI. REFERENCES

## Appendix D: Eelgrass Video Analysis SOP (In Progress)

STANDARD OPERATING PROCEDURE  
FOR VIDEO MONITORING OF EELGRASS  
FOR ENVIRONMENTAL AND PHYSICAL  
CONDITIONS

SOP 0.1  
Revision 0.1

November 6, 2019  
Page 1 of 4

### POINT OF CONTACT:

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## II. OBJECTIVE

Using video-observations of eelgrass beds, a procedure is outlined to quantify eelgrass and habitat characteristics, build an eelgrass observation library, and conduct analysis using the eelgrass health index.

Overview: Video-observation protocol for eelgrass beds is described for quantifying data from eelgrass media.

## II. MATERIALS AND EQUIPMENT

- MP4 observation files from eelgrass sites
- Computer with graphics editor software (E.g. Adobe Photoshop, GIMP, Inkscape)
- Terminal application (Freeware mainline terminal application)
- ExifTool software
- FIJI or ImageJ (Freeware NIH image-analysis software)
- Back-up harddrive

## III. METHODS

### A. Observation Preparation

1. Extract Time and Location Data. Using the computer's Terminal program and ExifTool (by Dr. Phil Harvey) – media data extraction software – date, time, and geographic location can be extracted from .mp4 or image files from site observations. Launch terminal and install ExifTool (see ExifTool website). To extract file data,

type:

“exiftool -ee -GPSDateStamp -GPSTimeStamp -GPSLatitude -GPSLongitude -n /file > site.txt”

2. Create Metadata File. In place of /file, drag the video file being observed. Title ‘site.txt’ using an appropriate name with the date, area, and site, e.g.  
072018\_CH04\_ESP02
3. Organize and Record Metadata. ‘Site.txt’ files are saved to the Documents folder. Move file to site-specific folder and then open the newly extracted .txt file and record site date, time, and location into the observation spreadsheet.

## B. Cover

1. Frame Selection and Capture. Open and review the .mp4 file for an eelgrass observation site. Identify either an appropriate time interval or specific times for 10 unique non-overlapping frames. For each selected frame, pause the video and take a screen capture of the frame. Save all 10 frames to a site-specific folder, e.g. “CH04\_ESP02.”
2. Observation File Setup. In the site folder, create a new blank file using a graphics editing program (Photoshop, GIMP, etc.) with the same dimensions as the video frames. Name this file the same as the folder.
3. Grid Design & Creation. In the first file, create a grid either manually using the line tool or via the program’s preferences. Grid lines should be spaced vertically and horizontally at the same intervals. Ten vertical and 10 horizontal bisecting lines provide a grid with 100 possible points and is an acceptable starting value.
4. Cover Observation. Overlay the grid on the image, create a new layer, and at each intersecting point identify if eelgrass is present or absent. If present, mark using a brush tool or equivalent. If absent, leave blank.
6. Percent Cover Calculation. After surveying the entire grid, count the total observed points and divide by the total possible points, e.g. a 10 x 10 grid has 100 possible points. Multiple this value by 100 to determine the percent cover.
7. Record Percent Cover. Record this value for percent cover on the observation spreadsheet and repeat for the remaining frames for the site.



### C. Shoot Density

1. Frame Area Calculation. Frame area for each observation is based on observation distance (0.25 m, 0.5 m, 1.0 m). Area is calculated using the pixel diameter of the white disk, the known disk's actual diameter (15.2 cm), and the pixel dimensions of the frame. Using this ratio and the known pixel dimensions of an image an area value for the image is calculated (**Equation Below**).

$$\frac{15.2 \text{ cm}}{280 \text{ Pixels}} = \frac{Y \text{ cm}}{1280 \text{ pixels}} * \frac{Z \text{ cm}}{720 \text{ pixels}}$$

$$\frac{1280 \text{ pixels}}{280 \text{ Pixels}} = \frac{Y \text{ cm}}{15.2 \text{ cm}} \quad \text{and} \quad \frac{720 \text{ cm}}{280 \text{ cm}} = \frac{Z \text{ cm}}{15.2 \text{ pixels}}$$

### D. Plant Height

#### **Description in Progress**

#### IV. TROUBLE SHOOTING / HINTS

3. During video monitoring, taking still photos of the observation site, the monitoring process, and the eelgrass bed are encouraged. Having extra cameras available, while the video camera is in use is a good practice.
4. Substrate can be determined by using the video camera at 0.25 m above the Secchi disk. While anchored turn on the camera and extend the pole down to the bottom of the bay. Gently push the end of it into the sediment and rotate several times. Retrieve the camera and turn it off. If water quality is decent, it should be possible to visual assess the sediment. If not, the sound ranging from coarse chatter (stones) to a fine abrading (sand), provides the researcher with a general idea of grain size and sediment type.

#### V. STATISTICAL ANALYSIS AND DATA USAGE

#### VI. REFERENCES

Phil Harvey's Exiftool