7-2021

Great Bay Estuary Tier 2 Seagrass/Seaweed Monitoring Program 2021 Quality Assurance Project Plan

Kalle Matso
University of New Hampshire

Trevor Mattera
University of New Hampshire

David Burdick
University of New Hampshire

Tom Gregory
University of New Hampshire

Great Bay Estuary Tier 2 Seagrass/Seaweed Monitoring Program 2021

EPA Grant # CE99171125; RFA 21099

Quality Assurance Project Plan

July 2021

Prepared by

Kalle Matso, Piscataqua Region Estuaries Partnership, University of New Hampshire (UNH)
Trevor Mattera, Piscataqua Region Estuaries Partnership, UNH
David Burdick, Jackson Estuarine Laboratory, UNH
Tom Gregory, Jackson Estuarine Laboratory, UNH

Project Manager:	Signature / Date Kalle Matso, PREP
Project QA Officer:	Signature / Date Trevor Mattera, PREP
Lead Scientist	Signature / Date David Burdick, UNH/JEL
Field Operations Manager:	Signature / Date Tom Gregory
EPA Project Officer:	13 Aug 2021 Signature / Date Erik Beck, US EPA
EPA QA Officer:	8/13/21 Signature / Date Jessica Iverson, US EPA

A2 – Table of Contents

List of Appendices

List of Tables

List of Figures

A3 – Distribution List

Table 1 presents a list of people who will receive the approved QA Project Plan (QAPP), any QAPP revisions, and any amendments.

Table 1: QAPP Distribution List

QAPP Recipient Name	Project Role	Organization	Telephone Number and E-mail Address
Kalle Matso	Project Manager	PREP	603-781-6591 kalle.matso@unh.edu
Trevor Mattera	Project QA Officer	PREP	603-862-1310 trevor.mattera@unh.edu
David Burdick	Lead Scientist/Laboratory Manager	UNH	603-862-5129 david.burdick@unh.edu
Tom Gregory	Field Operations Manager	UNH	603-862-5136 tom.gregory@unh.edu
Erik Beck	EPA Project Officer	US EPA	617-918-1606 beck.erik@epa.gov
Jessica Iverson	EPA QA Officer	US EPA	617-918-8630 Iverson.jessica@epa.gov

A4 – Project/Task Organization

The Piscataqua Region Estuaries Partnership (PREP) is part of the U.S. Environmental Protection Agency's (EPA) National Estuary Program, which is a joint local/state/federal program established under the Clean Water Act with the goal of protecting and enhancing nationally significant estuarine resources. PREP receives its funding from the EPA and is administered by the University of New Hampshire (UNH).

The project will be conducted and managed by PREP. The Project Manager (Kalle Matso) will be responsible for coordinating all program activities. Trevor Mattera will be the QA Officer, responsible for ensuring that activities are conducted in a manner consistent with this document.

David Burdick will serve as the Lead Scientist and Laboratory Operations Manager. He will manage all science-based questions and decisions, in cooperation with the Project Manager and the QA Officer. Sample analysis will be supervised by the Laboratory Manager at the UNH Jackson Estuarine Laboratory (JEL). The Laboratory Manager will be responsible for laboratory operations, including conducting analyses according to the procedures in this QAPP, identifying any non-conformities or analytical problems, and reporting any problems to the Project Manager and Project QA Officer.

Tom Gregory will manage all field operations and be responsible for "stop/go" decisions for monitoring activities during extreme events. Tom will be responsible for resolving any logistical problems and communicating the results to other field staff.

At the end of the project, the Project QA Officer will review the results of QA/QC tests and verify that the procedures of this QAPP were completed. The Project QA Officer will be responsible for a memorandum summarizing any deviations from the procedures in the QAPP, the results of the QA/QC tests, and whether the reported data meet the data quality objectives of the project.

Funding for PREP is provided by the EPA. Therefore, the Project Manager will be accountable to the EPA Project Manager (Erik Beck) and the EPA Project QA Officer (Nora Conlon). The EPA Project Manager and EPA Project QA Officer will be responsible for approving the QAPP.

The principal user of the data from this project will be PREP for State of Our Estuaries reports. The Lead Scientist and the Project Manager will work together to prepare a report at the end of the project with all the data and the QA summary report.

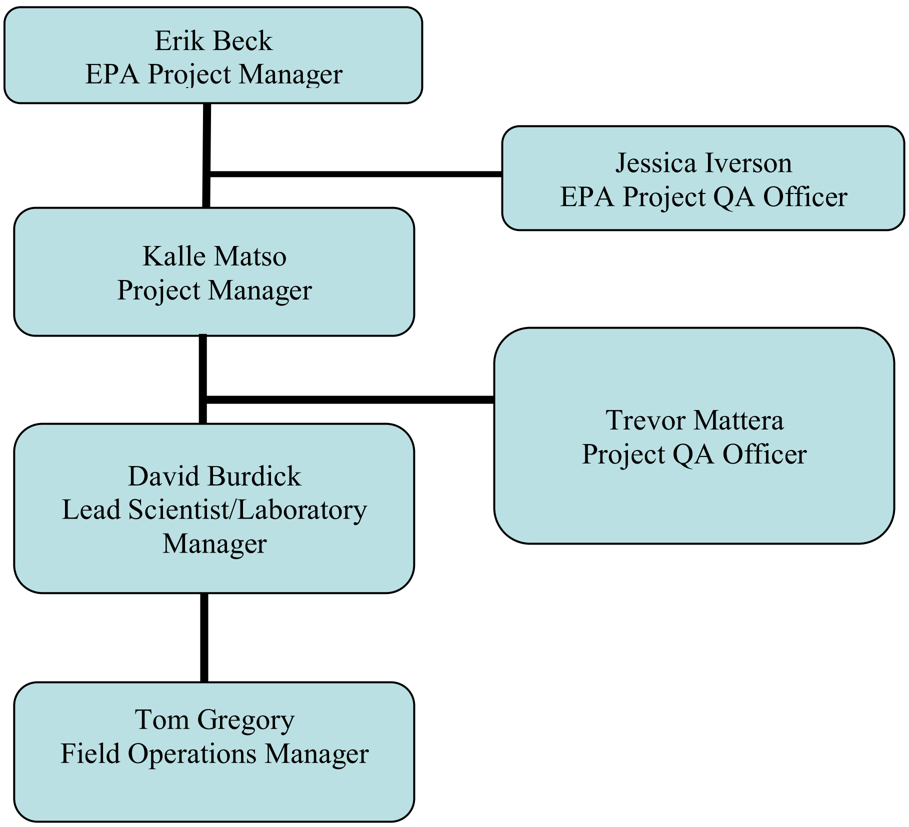

Figure 1: Project Organization

A5 – Problem Definition/Background

The long leaves of eelgrass (*Zostera marina*) slow the flow of water, encouraging suspended materials to settle, thereby promoting water clarity. Eelgrass roots stabilize sediments and both the roots and leaves take up nutrients from sediments and the water. Eelgrass provides habitat for fish and shellfish, and it produces significant amounts of organic matter for the larger food web. Because of eelgrass' importance, monitoring of this resource is critical. For many years, the distribution of eelgrass throughout the Great Bay Estuary has been monitored. It is well recognized, however, that distribution only tells part of the story of eelgrass health; it is also important to describe in greater detail how particular eelgrass meadows are changing with regard to more specific parameters, such as percent cover, canopy height, above and belowground biomass, etc.

Widgeon grass (*Ruppia maritima*), another seagrass, also exists in the Great Bay Estuary. Widgeon grass is more ephemeral than eelgrass, but some studies indicate that widgeon grass may expand in the future due to its ability to tolerate warmer and fresher waters.

Seaweeds constitute another important macrophyte covered by this QAPP. Seaweed blooms can be a sign of estuarine ecosystem impairment because they can occur in response to elevated nitrogen inputs as well as other environmental factors. In addition, seaweed can entangle and ultimately outcompete eelgrass in subtidal environments. Therefore, PREP aims to track the abundance of seaweed in the Great Bay Estuary as an indicator of estuarine health.

In 2012, Neckles et al. introduced a system of hierarchical monitoring based on three “tiers.” Tier 1 focuses on presence/absence at the scale of the entire estuary; Tier 2 uses a sample of sites throughout the estuary to characterize eelgrass health and monitor for differences between various zones of the estuary; Tier 3 (SeagrassNet) is focused on permanent transects and is used as a basis for asking specific cause-effect research questions, though limited in spatial scale.

Although this Tier 2 work is new to Great Bay Estuary, it will build in part on previous seaweed monitoring begun in 2013 (Cianciola and Burdick 2014) and continued by Burdick and partners through 2020 (e.g., Burdick et al. 2020). Previous QAPPs related to seaweed monitoring are accessible at: http://scholars.unh.edu/prep/

The objective of this project is to determine trends in eelgrass, widgeon grass and seaweed abundance and health as well as sediment characteristics at randomly selected sites throughout the Great Bay Estuary. The results will be published in PREP State of Our Estuaries reports (e.g., PREP 2017) and are of interest to the EPA, the NH Department of Environmental Services (NHDES), municipalities in the Piscataqua Region Watershed, and other partners.

Since this is the first year of the Tier 2 program, this QAPP will apply only to the year 2021. In the future, the QAPP will be amended and turned into a five-year plan.

A6 – Project/Task Description

Table 2: Project Schedule Timeline

Activity	Dates		Product	Due Date
	Anticipated Date(s) of Initiation	Anticipated Date(s) of Completion		
QAPP Preparation/Revision	1/1 (of sampling year)	4/30 (of sampling year)	QAPP document	5/1 (of sampling year)
Training	6/1 (of sampling year)	7/1 (of sampling year)	Field crews trained on SOPs	7/1 (of sampling year)
Monitoring and Sample Collection (July and August)	7/1 (of sampling year)	8/30 (of sampling year)	Samples collected for analysis	10/30 (of sampling year)

Activity	Dates		Product	Due Date
	Anticipated Date(s) of Initiation	Anticipated Date(s) of Completion		
Sample Analysis	7/1 (of sampling year)	11/30 (of sampling year)	Data on seagrass, seaweeds and sediments.	12/30 (of sampling year)
Data Quality Audit	8/1 (of sampling year)	3/1 (following sampling year)	Memorandum (see Section C2) summarizing any QAPP nonconformance	3/30 (following sampling year)
Annual Report	1/1 (following sampling year)	3/31 (following sampling year)	Final project report	5/1 (following sampling year)

A7 – Quality Objectives and Criteria

Data quality objectives for the Tier 2 monitoring program are summarized in Table 3.

Table 3: Data quality objectives, criteria, and quality control protocols for the program.

Data Quality Objective	Criteria	Protocol
Precision	Biomass measurements should be maintained to 1/100 of a gram.	Field assessment team will measure biomass with a Sartorius Balance (Type = E2000D).
Bias	Percent cover estimates should be comparable across members of the field assessment team within ±10%	Field assessment team members will "calibrate" their visual interpretations of percent cover prior and during field work by reviewing visual guides. Also, after initial sampling days, the team will review photos and assessments to better calibrate the assessment.
Spatial accuracy	GPS units should have a reported accuracy less than or equal to 2 meters.	For this project, it is understood that sites will not remain exactly the same from year to year. Rather, the goal is to sample within the error of a standard GPS to ensure that the sampling takes place within the proper 1-acre hexagon. Field teams will note and record the GPS coordinates of the location where they drop anchor. The QA Officer will double check after the fact to make sure that locations were within the designated hexagons.
Comparability	Field and laboratory data should be collected using standardized methods.	Check that protocols from the QAPP were used for field observations. The QA Officer should use filtering functions to check the field assessment team's spreadsheets for data entry errors. All percent cover values should fall into one of the categories specified in the sampling methods. A minimum of 10% of field observations should be checked against electronic spreadsheets.

Completeness	Field observations should be made for 85% of all scheduled samples listed in the sampling section at all 50 pre-determined sites: both "basic" and "enhanced."	Check field observations against site locations. Document reasons for any deviations from sampling protocol.

A8 – Special Training/Certification

The Laboratory and Field Operations Managers will organize and implement a training session for field staff. The training session will cover protocols for locating sites, placing quadrats, collecting samples and storing samples, when appropriate. The training will be based on the QAPP. Field staff will sign an attendance sheet for the training, which will be retained by the Field Operations Manager. The training will be completed before sampling begins.

Table 4: Special Personnel Training Requirements

Project Function	Description of Training	Training Provided by	Training Provided to	Location of Training Records
Quadrat Placement and Percent Cover of Seagrass and Seaweed Assessment	Using GPS to arrive at site; location of quadrat placements; review of percent cover assessment; review of sampling procedures for other parameters. (See Section B2 for more on Sampling Methods.)	Project Manager, Lead Scientist and Field Operations Manager	All field team staff	With Lead Scientist and Field Operations Manager

A9 – Documents and Records

QA Project Plan

The Project Manager will be responsible for maintaining the approved QAPP and for distributing the latest version to all parties on the distribution list in section A3. A copy of the approved plan will be posted to the PREP website (scholars.unh.edu/prep).

Field Data Sheets

The field data sheets for this project are attached as Appendix A. Field crews fill in these forms during the day and return them to the Field Operations Manager upon completion. The original forms, or scanned copies of the original forms, will be retained on file by the Field Manager.

Laboratory Data Sheets

Laboratory results from the Laboratory Manager will be stored in the form of electronic laboratory data sheets. The Project QA Officer will confirm the results of the required QC tests performed.

Reports to Management

The Project QA Officer will collaborate with the Field Operations and Laboratory Managers to produce a final report for PREP. This report will describe any deviations from the protocols established in the QAPP as well as summarize the results. The annual report will be posted to the PREP website (scholars.unh.edu/prep).

Archiving

The QAPP and final reports will be kept on file at PREP for a minimum of 10 years after the publication date of the final report. The original field data sheets, or scanned copies of the original field data sheets, will be retained by the Field Operations Manager and laboratory data sheets will be retained by the Laboratory Manager for a minimum of 5 years.

B1 – Sampling Process Design

The sampling approach calls for a portion of sites that are designated as "basic" and a portion that are designated as "enhanced," with the latter category having more sampled parameters than the former. See Table 5 for details. Based on resource constraints, it was determined that a total of 50 sites was the maximum number that was logistically feasible.

In order to ensure objectivity in determining where sites would be located, the 50 sites were generated by an outside contractor from EPA's Office of Research and Development, using a "generalized random-tessellation stratified design," which can incorporate specific needs (i.e., strata) of the study while maintaining a spatially well-balanced random sample (Neckles et al. 2012).

The EPA contractor was given the following conditions in the instructions for the site selection:

- The "sample frame"—the area from which possible locations are chosen—will be the areas that had seagrass (eelgrass, widgeon grass, or combination of both) according to the 2019 seagrass distribution report (https://scholars.unh.edu/prep/438/).

  This caveat reflects the purpose of Tier 2 sampling to monitor the condition of existing seagrass, rather than the distribution of seagrass, which is the purpose of the remote sensing and field verification approach involved with Tier 1 sampling. (Note that while the intent of this approach is to sample existing seagrass, some sites may end up being either bare or containing only seaweed. This is an acknowledged scenario in Tier 2 monitoring; "zero" percent cover and biomass for seagrass at some sites will be incorporated into the analysis of seagrass health changes over space and time.)
- The 25 basic and 25 enhanced sites should be distributed between the different hydrodynamic zones of the Great Bay Estuary, including: Great Bay, Little Bay, Piscataqua River (Upper and Lower combined) and Portsmouth Harbor, and Atlantic Coast (Bilgili et al. 2005).
- Seek overlap between 2013 seaweed/seagrass monitoring (Cianciola and Burdick 2014) and new "enhanced" sites. The reason for this is to note trends in seagrass or seaweed cover and also to provide the potential for comparing nitrogen isotope ratios between 2013 and current sampling. (Note that the 2013 sites were also generated by an outside contractor according to the tessellated hexagon design.)

As noted earlier, the hexagon approach is a recognized method for maintaining a spatially well-balanced sampling approach. For this application, each hex covers 1acre. This size was most suitable to the Great Bay Estuary given the narrow shapes of some eelgrass meadows. For each site within a hex, two "back-up" sites were provided in the event that a site turns out to be inaccessible or some other problem is encountered.

Sites will be sampled once a year in the July/August time period. Table 5 shows the number of samples that will be collected for each parameter.

Table 5: Sample Summary

Analytical Parameter	No. of Stations	Samples per Event per Site	Total # to Lab	Container Size/Type; Preservation Requirements	Comment
Seagrass Percent Cover	50	4	n/a	n/a	n/a
Seagrass Canopy Height	50	4	200	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	4 Sample Bags (one from each quadrat), each with three seagrass shoots chosen at random.) Canopy height is measured in the boat, and other parameters (see below) are assessed at the lab.
Epiphytes	50	See above	See above	Same as above	See procedure description below and Appendix C.
Seaweed Percent Cover	50	4	n/a	n/a	n/a
Seaweed Biomass	50	4	200	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	3 of the 4 bags will be processed to seaweed color (brown, red, green); 1 of the 4 bags will be processed to species.
Seagrass Biomass	25 enhanced	4	100	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	Seagrass biomass assessed 3 ways for comparison. See description below and Appendix G.
Seagrass Reproductive Capacity	25 enhanced	4	10 shoots per site	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	See description below and Appendix H.
Seagrass Tissue Analysis	25 enhanced	Uses shoots from canopy height sample	See left	See information for canopy height.	Carbon-Nitrogen Tissue Analysis performed on shoots removed in previous step. See Appendix D.
Nutrient Pollution Index (NPI)	25 enhanced	Uses shoots from canopy height sample	See left	See information for canopy height.	See description below.
Sediment Organic Matter	25 enhanced	1	25	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	See description below and Appendix E.
Sediment Grain Size	25 enhanced	4	100	1 gallon plastic bag placed in cooler; 6 hrs to lab; 7 days in lab cold storage (4ºC)	See description below and Appendix E.

A map of the stations is provided in Figure 2. Longitude and latitude for all sites are included in Appendix A.

B2 – Sampling Methods

Sample site locations are noted in Figure 2 and Appendix B. Extra sites within each hexagon are provided as back-ups in the event that the designated site is inaccessible or too deep to access at low tide via snorkeling.

Site Approach and Quadrat Placement

Boat crews will navigate to the designated coordinates using the GPS units on the boats. The accuracy of these GPS units can range from 0 to 2 meters. It is not an expectation of this sampling protocol that quadrats will be placed in the exact same spot year after year. Rather, the quadrats are meant to be a sub-sampling of a broader area.

When the crew has arrived at the correct location, the anchor is dropped. From the boat or by snorkel, the team will verify that the site is continuous (bare patches are within 1 meter of eelgrass) with at least 10% cover. At this point, the protocol will vary depending on whether snorkel or SCUBA will be used. Note that most sites are better sampled using SCUBA at high tide; visibility is generally better on higher tides, and there is also less of a chance of damaging seagrass beds with the boat propellor. However, there are some sites that can be sampled by snorkel.

For SCUBA, divers descend along the anchor line and then use the anchor as the center point of a "clock face." Quadrat A is placed 6 feet from the anchor at "12 o'clock"; Quadrats B, C and D are placed 6 feet away at 3, 6 and 9 o'clock, respectively. One diver conducts the assessments while the other divers holds bags, the camera, etc. Bags can be tied to the anchor after each quadrat is assessed so that the support diver isn't holding on to too much material. Underwater writing slates are used to communicate and to record data.

For snorkel, a stern anchor is used in addition to the bow anchor to keep the boat from changing its position. Quadrat A is placed off the bow and quadrats B, C and D are placed off the starboard, the stern and the port side, respectively. Snorkelers can hand material directly to the boat support person; assessments are communicated verbally to the boat support person, who logs the information on a waterproof data sheet.

Seagrass Percent Cover

Seagrass percent cover will be assessed according to the same methods as noted in the SeagrassNet QAPP using the same visual guides used in that document (Matso and Short 2019). Note that for both seagrass and seaweed cover, photos are taken for records and to help calibrate percent cover assessments.

Seagrass Canopy Height

Three shoots are chosen from the quadrat in a random fashion. On the boat, before being bagged and stored, the longest leaf of each shoot is measured from the youngest sprouted root and noted for canopy height.

Wasting Disease

Wasting disease (total amount of leaf tissue infected) is estimated on a percent infected scale (0-100%) for all aboveground tissue collected, for each shoot collected. (See Appendix F.)

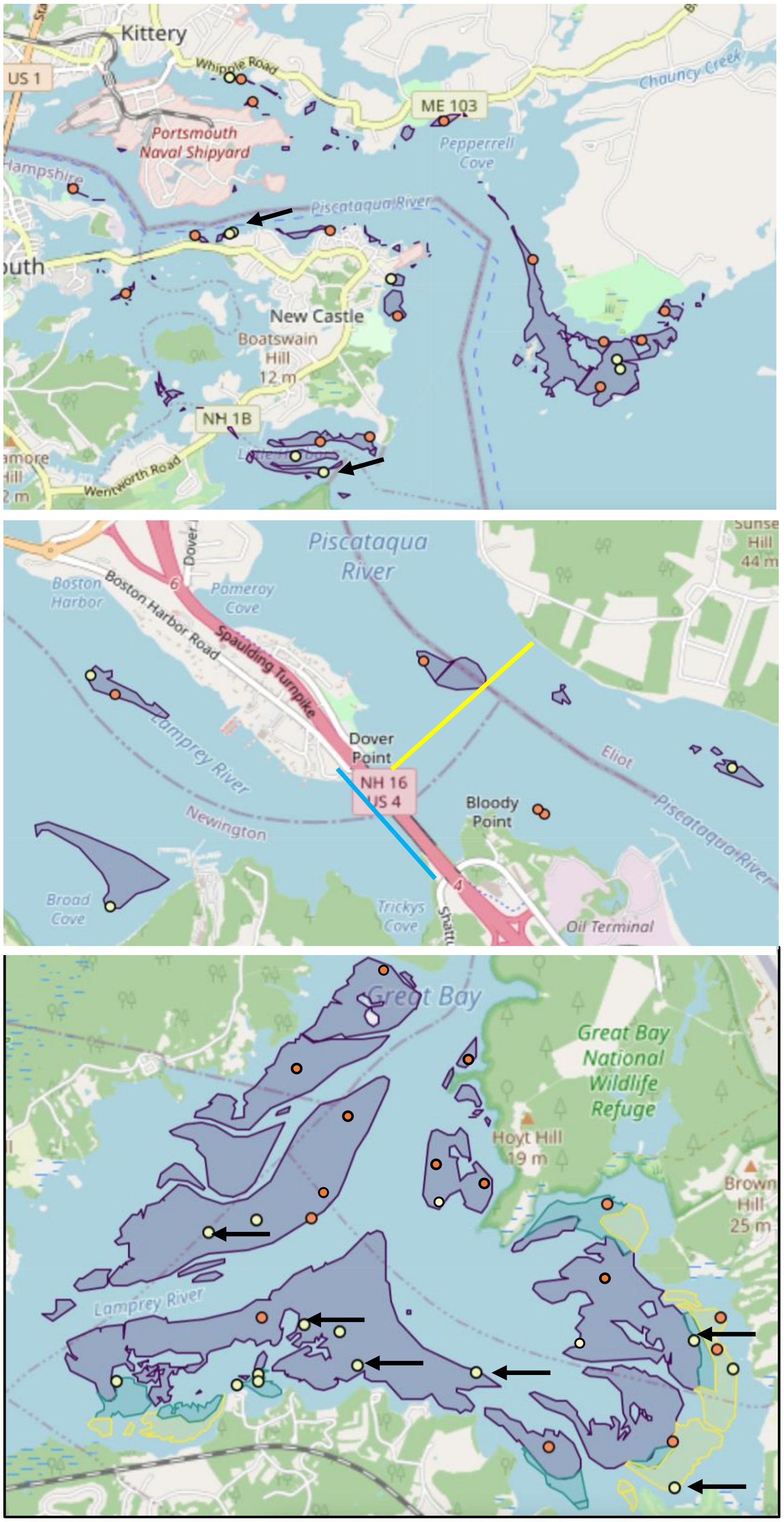

Figure 2: Approximate location of the 50+ sites throughout the estuary. 25 of the sites are denoted as "enhanced" sites, where additional parameters are analyzed. Shaded areas where seagrass was noted in 2019. Dark (purple) shading indicates eelgrass; medium (teal) shading indicates a mix of eelgrass and widgeon grass; light (green) shading indicates widgeon grass. Red (some may appear orange) dots denote "basic" sites and yellow dots denote "enhanced" sites. Black arrows point to sites that were sampled in 2013 (Cianciola and Burdick 2014). Top figure shows the Portsmouth Harbor area; Middle figure shows an area that includes Little Bay, Upper Piscataqua and Lower Piscataqua (Bilgili et al. 2005). (The yellow line separates Upper and Lower Piscataqua; the blue line separates Little Bay from Lower Piscataqua.) The lower figure shows Great Bay.

Epiphytes

Epiphyte load per shoot is derived at the lab by scraping both sides of an external and an internal leaf after Nelson (2018) and placing the epiphytes in a drying oven for 24-26 hours. Length and width of the external and internal leaves are also noted so that epiphyte load per leaf area can be calculated. (See Appendix C.)

Seaweed Percent Cover

Visual percentage cover by species within the $0.25m^2$ plots will be assessed by snorkelers. In addition, photos of plots will be obtained using stills or video (screen prints of video). Staff will visually estimate the percent of the ground covered by red, green and brown seaweed. Seaweed percent cover will be assessed according to the same methods as noted in the SeagrassNet QAPP using the same visual guides used in that document (Matso and Short 2019).

Seaweed Biomass

Sample labels are prepared prior to field visits, containing site name and date on Rite in the Rain paper. Each label is placed in a 1-gallon plastic bag. While hand-collecting will be sufficient for most samples, the field assessment team will carry a set of shears and/or a knife that can be used to cut aboveground biomass within the plot boundaries if needed. Field staff will use the field sheets to differentiate between drift and attached seaweeds. Determining the abundance of drift versus attached seaweeds has important implications for management options.

At each site, biomass of seaweed will be collected and brought to the lab for identification. For three of the four quadrats at each site, identification will focus on seaweed color category (i.e., red, green, brown); at one of the four quadrats, seaweed will be identified to species level. Dry weight will be determined on an areal basis after drying samples. Subsampling will be employed if samples are too large to determine dry weight.

Seagrass Biomass

Seagrass biomass will be assessed using the 7 cm method currently being used in the Great Bay Estuary as part of the SeagrassNet protocol (Matso and Short 2019). For this method, samplers choose a representative shoot within the quadrat and remove the shoot and at least 7 cm of rhizome material to represent the belowground biomass. Biomass for the entire quadrat is calculated by multiplying the biomass for the representative shoot by the density of the shoots. For lab procedures, see Appendix G.

Seagrass Reproductive Capacity

For each quadrat, the number of reproductive shoots will be counted and recorded on the field sheet. In addition, 10 reproductive Zostera shoots (if possible) are collected randomly in the general area around the site. If there is a depth gradient in the area, collectors will attempt to take shoots from a variety of depths. If possible, shoots will be separated in space by at least 2 m to increase the likelihood of getting different genotypes.

Back at the lab, shoots are examined to count rhipidia, spathes and viable seeds. Seeds are considered viable if they resist squeezing by forceps (i.e., they are not viable if they are soft) and if their coats are not split (Jorgensen et al. 2019). For more details on lab procedures, see Appendix H.

Seagrass Tissue Analysis

The second leaf of each shoot collected from the four quadrats will be combined to provide at least 20 mg of leaf tissue. Total carbon and nitrogen contents in blade or leaf tissues will be determined from two replicates of each sample by oxidation in a Perkin Elmer 2400 Series II CHN analyzer at the UNH Water

Quality Analysis Laboratory. See Appendix D for SOP. Tissues will be saved and stored in case it becomes relevant in the future to analyze for total phosphorus.

Nutrient Pollution Index

The Nutrient Pollution Index (NPI) will be calculated for each site using the collected shoots from each quadrat. The NPI calculation represents the ratio of leaf N content and leaf mass and requires the following measurements: sheath length; shoot height; number of leaves per shoot; leaf width; leaf dry weight and leaf carbon and nitrogen content (Lee et al. 2004). Because these measurements are either included in other procedures or very straightforward, no additional SOP is required.

Sediment Organic Matter/Grain Size

Take one sample at each site using a plastic 60cc syringe (2.5 cm diameter, 10 cm length with end removed) driven up to 10 cm into the seafloor (depth is determined by sediment compactness). These samples are placed in pre-labeled bags and homogenized and immediately placed on ice for transport to the lab. See Appendix E for SOP on processing in the lab.

B3 – Sample Handling and Custody

Biomass samples will be held in the custody of UNH JEL. Biomass samples will be held at UNH JEL at least until the Project Manager has received and reviewed the electronic data for the current year. After this time, samples that are brought to UNH JEL by the field assessment team may be given to the marine/estuarine collections of the UNH Hodgdon Herbarium or the herbarium of another institution at the discretion of UNH JEL.

B4 – Analytical Methods

At the end of each sampling season, the Science Lead will integrate the percent cover and biomass data collected in the current year into the existing regressions expressing the relationship between cover and biomass for each species. Canopy height, wasting disease, epiphytes, reproductive characteristics (# of reproductive shoots, rhipidia, seeds), seagrass tissue analysis, NPI and sediment organic matter/grain size will be analyzed to make within year to make non-statistical comparisons between sites and regions. When we have multiple years of data, we plan to test the hypothesis that none of these parameters are changing over time. This will be done using a repeated measures analysis of variance, treating year and hydrodynamic region (or distance from flushing) as fixed effects.

The Project QA Officer will screen the regression plots for outliers (see Section B5) and will remove outlying data points from the regression plots and annotate these data points where they appear in the electronic data tables.

B5 – Quality Control

The Project QA Officer will check that the data quality objectives are met using the criteria and methods from Table 3 in Section A7.

The Field Operations Manager will verify that the field crews are following the protocols correctly during the field sampling audit (see Section C1).

Databases of results will be checked for transcription errors and bad data using two methods. First, the entire data set will be printed and checked against the entries in each field or laboratory data sheet, data will be graphed to identify outliers, and the Huber Robust Fit method (K = 4) in JMP will be applied to each regression by the Science Lead. Second, the Project QA Officer will discuss outlier occurrences with the Science Lead to determine if there are outliers in the data set. The Project QA Officer/Project Manager will examine the approaches to identify outliers to determine whether these data should remain in the dataset.

B6/B7 – Instrument/Equipment Testing, Inspection, Maintenance, Calibration, and Frequency

The field assessment team will be responsible for checking the batteries in the GPS and digital camera before traveling to sampling sites each day that this equipment is in use. The GPS, camera, and a spare set of batteries will be taken into the field in a re-sealable plastic sampling bag or other watertight container. The field assessment team will also transfer photographs from the camera to a computer at the end of each sampling day to ensure that the camera has sufficient memory available to store new pictures on the next sampling day.

Calibration type and frequency for water quality instruments associated with tissue analysis are covered under the estuarine water quality QAPP at: https://scholars.unh.edu/prep/419/

For other relevant equipment requiring periodic calibrations—including balances, incubators, and drying ovens—calibration records are maintained and are traceable to the instrument. These records will be inspected by the QA Officer.

B8 – Inspection/Acceptance Requirements for Supplies and Consumables

The field assessment team will prepare field equipment for daily use, ensuring proper calibration completed, software updated, and/or power sources optimized for peak performance (i.e., charged/cycled).

B9 – Non-Direct Measurements

As noted on Page 10, most sites will be sampled by SCUBA. Ideal times will be near peak high tide, so days and times will be chosen accordingly. For a small number of sites, snorkel will be used. In those cases, peak low tide will be targeted.

Information on tides will be used to determine the dates and times at which site establishment and sampling will occur. WillyWeather forecasts at Fort Point, Dover Point, and the Squamscott River span the study area:

- Fort Point (Portsmouth Harbor) https://tides.willyweather.com/nh/rockingham-county/fort-point.html
- Dover Point https://tides.willyweather.com/nh/strafford-county/piscataqua-river--dover-point.html
- Squamscott River https://tides.willyweather.com/nh/rockingham-county/squamscott-river--railroad-bridge.html

B10 – Data Management

Field data will be recorded on standard field data sheets. Laboratory data will be transferred from laboratory data sheets to Excel spreadsheets. All laboratory data will be stored electronically in Excel spreadsheets, which will be transferred to the Project QA Officer as part of the laboratory report.

The field assessment team will provide the Project QA Officer with copies of all electronic files via an electronic data transfer system or on a flash drive within 10 business days of the completion of laboratory work for the current field season. Files will be stored in a dedicated project directory on the PREP (UNH) computer system. The Project QA Officer will be responsible for uploading the data to PREP publications website at scholars.unh.edu/prep/. Management of hardcopy data and documents is described in Section A9.

C1 – Assessments and Response Actions

In order to confirm that field sampling, field analysis, and laboratory activities are occurring as planned, the Project QA Officer, Field Operations Manager, and Laboratory Manager shall confer after the first sampling event each year to discuss the methods being employed and to review the quality assurance samples. At this time, all concerns regarding the sampling protocols and analysis techniques shall be addressed and any changes deemed necessary shall be made to ensure consistency and quality of subsequent sampling. The Project Manager will have the authority to resolve any problems encountered. Assessment frequencies and responsible personnel are shown in the following table.

Table 6: Project Assessment Table

Assessment Type	Frequency (Annual Basis)	Person Responsible for Performing Assessment	Person Responsible for Responding to Assessment Findings	Person Responsible for Monitoring Effectiveness of Corrective Actions
Field sampling audit	Once after first sampling day	Field Operations Manager	Field Operations Manager	Field Operations Manager
Field analytical audit (GPS, Camera)	Once after first sampling day	Field Operations Manager	Field Operations Manager	Field Operations Manager
Data Quality Audit	Annually	Project QA Officer	Project QA Officer	Project QA Officer

C2 – Reports to Management

The Project QA Officer will produce a QA/QC memorandum addressed to NHDES that will be part of each year's final report. Each year, the final work product will be a table containing quality assured laboratory results for each station on each date and a memorandum describing any deviations from the protocols established in the QAPP. Reports and QA/QC memos will also be shared with the EPA Project Officer. Data from the final reports will be published in PREP's State of Our Estuaries reports and uploaded to the PREP Publications website at: https://scholars.unh.edu/prep/

D1 – Data Review, Verification, and Validation

The Project QA Officer will be responsible for a memorandum to PREP summarizing any deviations from the procedures in the QAPP and the results of the QA/QC tests. The Project QA Officer will review all field data sheets and/or final computer data files for completeness and quality based on the criteria described in Section A7. The Project QA Officer will also *affirmatively* verify that the methods used for the study followed the procedures outlined in this QAPP. If questionable entries or data are encountered during the review process (see methods in Section B5), the Project QA Officer will contact the appropriate personnel to determine their validity.

D2 – Verification and Validation Procedures

The Project Manager will compare the QA memorandum against the QAPP. Any decisions made regarding the usability of the data will be left to the Project Manager; however, the Project Manager may consult with project personnel or with personnel from EPA, if necessary.

D3 – Reconciliation with User Requirements

The Project Manager will be responsible for reconciling the results from this study with the ultimate use of the data. Results that are qualified through the QA process may still be used if the limitations of the data are clearly reported to decision-makers. Data for this project are being collected as part of a long-term monitoring program. It is not possible to repeat sampling events without disrupting the time series. Therefore, the Project Manager will:

1. Review data with respect to sampling design.
2. Compare the QA memorandum with the QAPP.
3. If the data quality objectives from Section A7 are met, the user requirements have been met. If the data quality objectives have not been met, corrective action as discussed in D2 will be established by the Project Manager.

References

Bilgili A, Proel JA, Lynch DR, Smith DW and Swift MR. 2005. Estuary/ocean exchange and tidal mixing in a Gulf of Maine Estuary: A Lagrangian modeling study. *Estuarine, Coastal and Shelf Science*, **65**: 607-624.

Burdick, David M.; Moore, Gregg; Mathieson, Arthur C.; Payne, Andrew; Martin, Lara; and Peter, Chris. 2020, Seaweed Monitoring in the Great Bay Estuary: 2019 Annual Report. *PREP Reports & Publications*. 442. https://scholars.unh.edu/prep/442

Cianciola, E and Burdick, D.M., 2014. Results of 2013 Macroalgal Monitoring and Recommendations for Future Monitoring in Great Bay Estuary, New Hampshire. PREP Publications. 284. http://scholars.unh.edu/prep/284

Jorgensen MS, Labouriau R and B Olesen. 2019. Seed size and burial depth influence Zostera marina L. (eelgrass) seed survival, seedling emergence and initial seedling biomass development. PLOS One. https://doi.org/10.1371/journal.pone.0215157

Lee K, Short FT and DM Burdick. 2004. Development of a nutrient pollution indicator using the seagrass, Zostera marina, along nutrient gradients in three New England estuaries. Aquatic Botany. 78, 197 - 216.

Matso, K and Short, F. 2019. SeagrassNet Monitoring Program 2019 - 2023: Quality Assurance Project Plan. PREP Reports & Publications. 420. https://scholars.unh.edu/prep/420

Neckles, HA, Kopp BS, Peterson BJ, Pooler PS. 2012. Integrating scales of seagrass monitoring to meet conservation needs. Estuaries and Coasts. Vol. 35, No. 1, pp. 23-46. DOI 10.1007/s12237-011-9410-x

Nelson WG. 2018. An evaluation of factors controlling the abundance of epiphytes on Zostera marina along an estuarine gradient in Yaquina Bay, Oregon, USA. Aquatic Botany. 148: 53-63

PREP. 2017. State of Our Estuaries 2018. Piscataqua Region Estuaries Partnership, University of New Hampshire, Durham, NH. Published online: www.stateofourestuaries.org.

APPENDIX A

FIELD SHEET

SITE: | DATE: / / | TIME: | OBSERVERS:

CONDITIONS/COMMENTS:

Date	TIME	SUB SAMPLE	Basic or Enhanced	% cov Sgrass	% cov Sweed	CAN HT	Repro Shoot Count	Comments
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						
		A						
		B						
		C						
		D						

Appendix B

Tier 2 Site Index

SITEID	AREA	SEAGRASS STRATUM	ENHANCED?	CATEGORY	longitude	latitude
GB-GBE-1001-a	Great Bay	Eelgrass	N	Base	-70.85464075	43.05618519
GB-GBE-1001-b	Great Bay	Eelgrass	N	Base-alt	-70.85469886	43.05623786
GB-GBE-1001-c	Great Bay	Eelgrass	N	Base-alt	-70.85487771	43.05596281
GB-GBE-1002-a	Great Bay	Eelgrass	N	Base	-70.88245408	43.06541738
GB-GBE-1002-b	Great Bay	Eelgrass	N	Base-alt	-70.88257058	43.06516039
GB-GBE-1002-c	Great Bay	Eelgrass	N	Base-alt	-70.88261783	43.0652692
GB-GBE-1003-a	Great Bay	Eelgrass	Y	Base	-70.87479764	43.06435962
GB-GBE-1003-b	Great Bay	Eelgrass	Y	Base-alt	-70.8743461	43.0644788
GB-GBE-1003-c	Great Bay	Eelgrass	Y	Base-alt	-70.87488045	43.06408004
GB-GBE-1004-a	Great Bay	Eelgrass	Y	Base	-70.88297665	43.07228358
GB-GBE-1004-b	Great Bay	Eelgrass	Y	Base-alt	-70.8831125	43.07249083
GB-GBE-1004-c	Great Bay	Eelgrass	Y	Base-alt	-70.88351341	43.07246674
GB-GBE-1005-a	Great Bay	Eelgrass	N	Base	-70.87763505	43.07240838
GB-GBE-1005-b	Great Bay	Eelgrass	N	Base-alt	-70.87748183	43.07246761
GB-GBE-1005-c	Great Bay	Eelgrass	N	Base-alt	-70.87731913	43.07252852
GB-GBE-1006-a	Great Bay	Eelgrass	Y	Revisit2013	-70.87836887	43.06490776
GB-GBE-1007-a	Great Bay	Eelgrass	Y	Revisit2013	-70.86160197	43.06154549
GB-GBE-1008-a	Great Bay	Eelgrass	Y	Revisit2013	-70.84053174	43.06374631
GB-GBE-1009-a	Great Bay	Eelgrass	Y	Revisit2013	-70.87312195	43.06197667
GB-GBE-1010-a	Great Bay	Eelgrass	Y	Revisit2013	-70.88755001	43.07136008
GB-GBE-1011-a	Great Bay	Eelgrass	N	OverSamp	-70.88566102	43.06339015
GB-GBE-1011-b	Great Bay	Eelgrass	N	OverSamp-alt	-70.88574652	43.06341859
GB-GBE-1011-c	Great Bay	Eelgrass	N	OverSamp-alt	-70.88566739	43.06348295
GB-GBE-1012-a	Great Bay	Eelgrass	N	OverSamp	-70.86489947	43.07335791
GB-GBE-1012-b	Great Bay	Eelgrass	N	OverSamp-alt	-70.86490952	43.0733644
GB-GBE-1012-c	Great Bay	Eelgrass	N	OverSamp-alt	-70.86504995	43.07327887
GB-GBE-1013-a	Great Bay	Eelgrass	N	OverSamp	-70.87717325	43.07387759
GB-GBE-1013-b	Great Bay	Eelgrass	N	OverSamp-alt	-70.87704194	43.07361931
GB-GBE-1013-c	Great Bay	Eelgrass	N	OverSamp-alt	-70.87721905	43.07386707
GB-GBE-1014-a	Great Bay	Eelgrass	N	OverSamp	-70.86941374	43.08967318
GB-GBE-1014-b	Great Bay	Eelgrass	N	OverSamp-alt	-70.86876856	43.08971718
GB-GBE-1014-c	Great Bay	Eelgrass	N	OverSamp-alt	-70.86882983	43.08970125
GB-GBE-1015-a	Great Bay	Eelgrass	N	OverSamp	-70.85137921	43.06434738
GB-GBE-1015-b	Great Bay	Eelgrass	N	OverSamp-alt	-70.85122652	43.06437101
GB-GBE-1015-c	Great Bay	Eelgrass	N	OverSamp-alt	-70.85115009	43.06448903
GB-GBEW-1016-a	Great Bay	Eelgrass and Widgeon Grass	Y	Base	-70.88271227	43.06132427
GB-GBEW-1016-b	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88283701	43.06094735
GB-GBEW-1016-c	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88275464	43.06081071
GB-GBEW-1017-a	Great Bay	Eelgrass and Widgeon Grass	Y	Base	-70.88485927	43.06057912
GB-GBEW-1017-b	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88465027	43.06059526
GB-GBEW-1017-c	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88480028	43.0604728
GB-GBEW-1018-a	Great Bay	Eelgrass and Widgeon Grass	Y	Base	-70.89654005	43.06086752
GB-GBEW-1018-b	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.89614818	43.06092356
GB-GBEW-1018-c	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.89627873	43.06090351
GB-GBEW-1019-a	Great Bay	Eelgrass and Widgeon Grass	Y	Base	-70.88275464	43.06081071
GB-GBEW-1019-b	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88271227	43.06132427

GB-GBEW-1019-c	Great Bay	Eelgrass and Widgeon Grass	Y	Base-alt	-70.88283701	43.06094735
GB-GBEW-1020-a	Great Bay	Eelgrass and Widgeon Grass	N	Base	-70.84892669	43.07344126
GB-GBEW-1020-b	Great Bay	Eelgrass and Widgeon Grass	N	Base-alt	-70.84934375	43.07351549
GB-GBEW-1020-c	Great Bay	Eelgrass and Widgeon Grass	N	Base-alt	-70.84901425	43.0738022
GB-GBEW-1021-a	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp	-70.89821417	43.06010869
GB-GBEW-1021-b	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.89817368	43.06002479
GB-GBEW-1021-c	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.89774516	43.05999894
GB-GBEW-1022-a	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp	-70.8535026	43.05345302
GB-GBEW-1022-b	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.85340561	43.0533421
GB-GBEW-1022-c	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.85357437	43.05325506
GB-GBEW-1023-a	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp	-70.88117007	43.06060297
GB-GBEW-1023-b	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.88086129	43.0604144
GB-GBEW-1023-c	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.88113968	43.060384
GB-GBEW-1024-a	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp	-70.89461992	43.05993283
GB-GBEW-1024-b	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.89458913	43.05990734
GB-GBEW-1024-c	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.89486641	43.05993778
GB-GBEW-1025-a	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp	-70.85322908	43.07354202
GB-GBEW-1025-b	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.85336789	43.07354362
GB-GBEW-1025-c	Great Bay	Eelgrass and Widgeon Grass	N	OverSamp-alt	-70.85311136	43.07353297
GB-GBW-1026-a	Great Bay	Widgeon Grass	Y	Base	-70.83679744	43.06171561
GB-GBW-1026-b	Great Bay	Widgeon Grass	Y	Base-alt	-70.83675341	43.06156605
GB-GBW-1026-c	Great Bay	Widgeon Grass	Y	Base-alt	-70.83685889	43.06189617
GB-GBW-1027-a	Great Bay	Widgeon Grass	N	Base	-70.8426463	43.05655117
GB-GBW-1027-b	Great Bay	Widgeon Grass	N	Base-alt	-70.84254582	43.05680948
GB-GBW-1027-c	Great Bay	Widgeon Grass	N	Base-alt	-70.84281876	43.05635386
GB-GBW-1028-a	Great Bay	Widgeon Grass	N	Base	-70.83797607	43.06537999
GB-GBW-1028-b	Great Bay	Widgeon Grass	N	Base-alt	-70.83814176	43.06548956
GB-GBW-1028-c	Great Bay	Widgeon Grass	N	Base-alt	-70.83803196	43.06568721
GB-GBW-1029-a	Great Bay	Widgeon Grass	N	Base	-70.83830561	43.06306772
GB-GBW-1029-b	Great Bay	Widgeon Grass	N	Base-alt	-70.83785529	43.06301862
GB-GBW-1029-c	Great Bay	Widgeon Grass	N	Base-alt	-70.83798396	43.06296976
GB-GBW-1030-a	Great Bay	Widgeon Grass	Y	Revisit2013	-70.84240034	43.05336443
GB-GBW-1031-a	Great Bay	Widgeon Grass	N	OverSamp	-70.84636862	43.05457652
GB-GBW-1031-b	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84677584	43.05474595
GB-GBW-1031-c	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84650749	43.05463491
GB-GBW-1032-a	Great Bay	Widgeon Grass	N	OverSamp	-70.84138756	43.05746945
GB-GBW-1032-b	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84189449	43.05766444
GB-GBW-1032-c	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84189492	43.05768809
GB-GBW-1033-a	Great Bay	Widgeon Grass	N	OverSamp	-70.83794855	43.05756968
GB-GBW-1033-b	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.83817	43.05783037
GB-GBW-1033-c	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.8383495	43.05721932
GB-GBW-1034-a	Great Bay	Widgeon Grass	N	OverSamp	-70.8429545	43.05456743
GB-GBW-1034-b	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84268169	43.05507794
GB-GBW-1034-c	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84318877	43.0551124
GB-GBW-1035-a	Great Bay	Widgeon Grass	N	OverSamp	-70.84162486	43.06628002
GB-GBW-1035-b	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.8415247	43.06628763
GB-GBW-1035-c	Great Bay	Widgeon Grass	N	OverSamp-alt	-70.84163857	43.06629966
GB-GI-1036-a	Gerrish Island Beds	Eelgrass	N	Base	-70.69040645	43.06081375
GB-GI-1036-b	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.69078689	43.06081383

GB-GI-1036-c	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.69065197	43.06050684
GB-GI-1037-a	Gerrish Island Beds	Eelgrass	N	Base	-70.68362346	43.06671543
GB-GI-1037-b	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.68311821	43.06657644
GB-GI-1037-c	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.68324168	43.06673513
GB-GI-1038-a	Gerrish Island Beds	Eelgrass	N	Base	-70.6861175	43.06446415
GB-GI-1038-b	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.68606454	43.06453712
GB-GI-1038-c	Gerrish Island Beds	Eelgrass	N	Base-alt	-70.68633121	43.06439623
GB-GI-1039-a	Gerrish Island Beds	Eelgrass	Y	Base	-70.68820752	43.06229364
GB-GI-1039-b	Gerrish Island Beds	Eelgrass	Y	Base-alt	-70.68862716	43.06239469
GB-GI-1039-c	Gerrish Island Beds	Eelgrass	Y	Base-alt	-70.68833239	43.06230802
GB-GI-1040-a	Gerrish Island Beds	Eelgrass	Y	Base	-70.68857268	43.0629332
GB-GI-1040-b	Gerrish Island Beds	Eelgrass	Y	Base-alt	-70.6881723	43.06304525
GB-GI-1040-c	Gerrish Island Beds	Eelgrass	Y	Base-alt	-70.68795155	43.06275655
GB-GI-1041-a	Gerrish Island Beds	Eelgrass	N	OverSamp	-70.68804962	43.0620999
GB-GI-1041-b	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68745478	43.06204634
GB-GI-1041-c	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68779116	43.06229049
GB-GI-1042-a	Gerrish Island Beds	Eelgrass	N	OverSamp	-70.69190055	43.06042008
GB-GI-1042-b	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.69205987	43.06040563
GB-GI-1042-c	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.69227325	43.06036735
GB-GI-1043-a	Gerrish Island Beds	Eelgrass	N	OverSamp	-70.68940856	43.06107722
GB-GI-1043-b	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68921494	43.0610208
GB-GI-1043-c	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68916117	43.06136707
GB-GI-1044-a	Gerrish Island Beds	Eelgrass	N	OverSamp	-70.68986347	43.06166388
GB-GI-1044-b	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68995743	43.06156909
GB-GI-1044-c	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.69022467	43.06176706
GB-GI-1045-a	Gerrish Island Beds	Eelgrass	N	OverSamp	-70.68337622	43.06534318
GB-GI-1045-b	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.68346849	43.06525723
GB-GI-1045-c	Gerrish Island Beds	Eelgrass	N	OverSamp-alt	-70.6833748	43.06555682
GB-LB-1046-a	Little Bay	Eelgrass	N	Base	-70.84033124	43.12264005
GB-LB-1046-b	Little Bay	Eelgrass	N	Base-alt	-70.84024703	43.12256108
GB-LB-1046-c	Little Bay	Eelgrass	N	Base-alt	-70.84016513	43.12253391
GB-LB-1047-a	Little Bay	Eelgrass	Y	Base	-70.84164101	43.12339271
GB-LB-1047-b	Little Bay	Eelgrass	Y	Base-alt	-70.84175003	43.12333973
GB-LB-1047-c	Little Bay	Eelgrass	Y	Base-alt	-70.84174031	43.12333153
GB-LB-1048-a	Little Bay	Eelgrass	Y	Base	-70.84062645	43.11432225
GB-LB-1048-b	Little Bay	Eelgrass	Y	Base-alt	-70.84079316	43.11445995
GB-LB-1048-c	Little Bay	Eelgrass	Y	Base-alt	-70.84075329	43.11447084
GB-LB-1049-a	Little Bay	Eelgrass	N	OverSamp	-70.83860837	43.11557343
GB-LB-1049-b	Little Bay	Eelgrass	N	OverSamp-alt	-70.83801674	43.11534593
GB-LB-1049-c	Little Bay	Eelgrass	N	OverSamp-alt	-70.83845129	43.11509032
GB-LB-1050-a	Little Bay	Eelgrass	N	OverSamp	-70.84091987	43.1146778
GB-LB-1050-b	Little Bay	Eelgrass	N	OverSamp-alt	-70.84095138	43.11464914
GB-LB-1050-c	Little Bay	Eelgrass	N	OverSamp-alt	-70.84095881	43.11468833
GB-LB-1051-a	Little Bay	Eelgrass	N	OverSamp	-70.84377687	43.11733235
GB-LB-1051-b	Little Bay	Eelgrass	N	OverSamp-alt	-70.84352642	43.1172183
GB-LB-1051-c	Little Bay	Eelgrass	N	OverSamp-alt	-70.84346078	43.11718321
GB-LH-1052-a	Little Harbor/Back Channel	Eelgrass	N	Base	-70.71440888	43.0569667
GB-LH-1052-b	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.71441204	43.05694311
GB-LH-1052-c	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.71441677	43.05697098

GB-LH-1053-a	Little Harbor/Back Channel	Eelgrass	N	Base	-70.71952727	43.05659644
GB-LH-1053-b	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.71968912	43.05660328
GB-LH-1053-c	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.7190793	43.05679159
GB-LH-1054-a	Little Harbor/Back Channel	Eelgrass	N	Base	-70.73978021	43.06815928
GB-LH-1054-b	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.73995964	43.06810158
GB-LH-1054-c	Little Harbor/Back Channel	Eelgrass	N	Base-alt	-70.74032305	43.06801795
GB-LH-1055-a	Little Harbor/Back Channel	Eelgrass	Y	Base	-70.72211584	43.05546397
GB-LH-1055-b	Little Harbor/Back Channel	Eelgrass	Y	Base-alt	-70.72206553	43.05538184
GB-LH-1055-c	Little Harbor/Back Channel	Eelgrass	Y	Base-alt	-70.72224407	43.0558291
GB-LH-1056-a	Little Harbor/Back Channel	Eelgrass	Y	Revisit2013	-70.71917268	43.05425932
GB-LH-1057-a	Little Harbor/Back Channel	Eelgrass	N	OverSamp	-70.72084578	43.05419033
GB-LH-1057-b	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.72092255	43.05369826
GB-LH-1057-c	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.72075493	43.05428735
GB-LH-1058-a	Little Harbor/Back Channel	Eelgrass	N	OverSamp	-70.73369273	43.05921159
GB-LH-1058-b	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.73366517	43.05921154
GB-LH-1058-c	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.73371197	43.05922608
GB-LH-1059-a	Little Harbor/Back Channel	Eelgrass	N	OverSamp	-70.7397477	43.06733183
GB-LH-1059-b	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.73971791	43.06734933
GB-LH-1059-c	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.73922834	43.06753715
GB-LH-1060-a	Little Harbor/Back Channel	Eelgrass	N	OverSamp	-70.71898545	43.05737678
GB-LH-1060-b	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.71911716	43.05719013
GB-LH-1060-c	Little Harbor/Back Channel	Eelgrass	N	OverSamp-alt	-70.71891228	43.05729965
GB-NPR-1061-a	Lower Piscataqua River North	Eelgrass	N	Base	-70.81809318	43.11815743
GB-NPR-1061-b	Lower Piscataqua River North	Eelgrass	N	Base-alt	-70.81801837	43.11812041
GB-NPR-1061-c	Lower Piscataqua River North	Eelgrass	N	Base-alt	-70.81810558	43.118176
GB-NPR-1062-a	Lower Piscataqua River North	Eelgrass	Y	Base	-70.80788677	43.11979034
GB-NPR-1062-b	Lower Piscataqua River North	Eelgrass	Y	Base-alt	-70.80784288	43.11983854
GB-NPR-1062-c	Lower Piscataqua River North	Eelgrass	Y	Base-alt	-70.80746798	43.11976775
GB-NPR-1063-a	Lower Piscataqua River North	Eelgrass	N	Base	-70.81780409	43.1179891
GB-NPR-1063-b	Lower Piscataqua River North	Eelgrass	N	Base-alt	-70.81775083	43.11809342
GB-NPR-1063-c	Lower Piscataqua River North	Eelgrass	N	Base-alt	-70.81786616	43.11801687
GB-NPR-1064-a	Upper Piscataqua River	Eelgrass	N	Base	-70.82414622	43.12400388
GB-NPR-1064-b	Upper Piscataqua River	Eelgrass	N	Base-alt	-70.8238584	43.12399673
GB-NPR-1064-c	Upper Piscataqua River	Eelgrass	N	Base-alt	-70.82388807	43.12379643
GB-NPR-1065-a	Lower Piscataqua River North	Eelgrass	N	OverSamp	-70.81006855	43.12057069
GB-NPR-1065-b	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.81009494	43.12056537
GB-NPR-1065-c	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.81011572	43.120545
GB-NPR-1066-a	Lower Piscataqua River North	Eelgrass	N	OverSamp	-70.80676567	43.11916407
GB-NPR-1066-b	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.80654916	43.11913358
GB-NPR-1066-c	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.8064118	43.11905598
GB-NPR-1067-a	Lower Piscataqua River North	Eelgrass	N	OverSamp	-70.82275129	43.1228863
GB-NPR-1067-b	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.82281826	43.12291718
GB-NPR-1067-c	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.82308154	43.12308379
GB-NPR-1068-a	Lower Piscataqua River North	Eelgrass	N	OverSamp	-70.81694372	43.12251076
GB-NPR-1068-b	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.81696644	43.12263838
GB-NPR-1068-c	Lower Piscataqua River North	Eelgrass	N	OverSamp-alt	-70.81695294	43.12256749
GB-PH-1069-a	Portsmouth Harbor	Eelgrass	N	Base	-70.69731297	43.0708306
GB-PH-1069-b	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.69723611	43.07066949
GB-PH-1069-c	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.69725557	43.07069081

GB-PH-1070-a	Portsmouth Harbor	Eelgrass	N	Base	-70.71145188	43.0664101
GB-PH-1070-b	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.71157068	43.06639979
GB-PH-1070-c	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.71140499	43.06636334
GB-PH-1071-a	Portsmouth Harbor	Eelgrass	N	Base	-70.69006155	43.06433488
GB-PH-1071-b	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.69007517	43.06424878
GB-PH-1071-c	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.69047727	43.06423128
GB-PH-1072-a	Portsmouth Harbor	Eelgrass	Y	Base	-70.72862102	43.07292111
GB-PH-1072-b	Portsmouth Harbor	Eelgrass	Y	Base-alt	-70.72886434	43.07287713
GB-PH-1072-c	Portsmouth Harbor	Eelgrass	Y	Base-alt	-70.72846207	43.0729526
GB-PH-1073-a	Portsmouth Harbor	Eelgrass	N	Base	-70.70669581	43.08156262
GB-PH-1073-b	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.706602	43.08169042
GB-PH-1073-c	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.70668375	43.08176523
GB-PH-1074-a	Portsmouth Harbor	Eelgrass	N	Base	-70.71839425	43.07297244
GB-PH-1074-b	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.71833013	43.0729814
GB-PH-1074-c	Portsmouth Harbor	Eelgrass	N	Base-alt	-70.718334	43.07293822
GB-PH-1075-a	Portsmouth Harbor	Eelgrass	Y	Base	-70.71214167	43.06931696
GB-PH-1075-b	Portsmouth Harbor	Eelgrass	Y	Base-alt	-70.71214977	43.06935456
GB-PH-1075-c	Portsmouth Harbor	Eelgrass	Y	Base-alt	-70.71214813	43.06928942
GB-PH-1076-a	Portsmouth Harbor	Eelgrass	Y	Revisit2013	-70.72902821	43.07280202
GB-PH-1077-a	Odiorne Point Beds	Eelgrass	N	OverSamp	-70.71429571	43.05581085
GB-PH-1077-b	Odiorne Point Beds	Eelgrass	N	OverSamp-alt	-70.71413988	43.0556507
GB-PH-1077-c	Odiorne Point Beds	Eelgrass	N	OverSamp-alt	-70.71419936	43.05572659
GB-PH-1078-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.69519532	43.06275467
GB-PH-1078-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.6949525	43.06251128
GB-PH-1078-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69486512	43.06279299
GB-PH-1079-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.69687685	43.06651025
GB-PH-1079-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69697262	43.06647805
GB-PH-1079-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69669971	43.06668905
GB-PH-1080-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.72134789	43.07250728
GB-PH-1080-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.72133764	43.07246993
GB-PH-1080-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.72132769	43.0725115
GB-PH-1081-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.70970031	43.07201752
GB-PH-1081-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.70966495	43.07200091
GB-PH-1081-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.70953556	43.07209085
GB-PH-1082-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.69756913	43.06885436
GB-PH-1082-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69764193	43.06872043
GB-PH-1082-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69764344	43.06864962
GB-PH-1083-a	Portsmouth Harbor	Eelgrass	N	OverSamp	-70.69685135	43.06548049
GB-PH-1083-b	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69691033	43.06522914
GB-PH-1083-c	Portsmouth Harbor	Eelgrass	N	OverSamp-alt	-70.69673605	43.06530409
GB-SPR-1084-a	Lower Piscataqua River South	Eelgrass	N	Base	-70.72761212	43.08479796
GB-SPR-1084-b	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.72733686	43.08472189
GB-SPR-1084-c	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.72753747	43.08477216
GB-SPR-1085-a	Lower Piscataqua River South	Eelgrass	N	Base	-70.76258814	43.09187767
GB-SPR-1085-b	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.76253092	43.091848
GB-SPR-1085-c	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.76270914	43.09185846
GB-SPR-1086-a	Lower Piscataqua River South	Eelgrass	N	Base	-70.73198194	43.07245512
GB-SPR-1086-b	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.73209887	43.07237523
GB-SPR-1086-c	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.73196443	43.07236347

GB-SPR-1087-a	Lower Piscataqua River South	Eelgrass	N	Base	-70.74417731	43.0757825
GB-SPR-1087-b	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.74395644	43.07572697
GB-SPR-1087-c	Lower Piscataqua River South	Eelgrass	N	Base-alt	-70.74408351	43.07576563
GB-SPR-1088-a	Lower Piscataqua River South	Eelgrass	Y	Base	-70.73027815	43.07219422
GB-SPR-1088-b	Lower Piscataqua River South	Eelgrass	Y	Base-alt	-70.73020619	43.07229699
GB-SPR-1088-c	Lower Piscataqua River South	Eelgrass	Y	Base-alt	-70.73021574	43.07231689
GB-SPR-1089-a	Lower Piscataqua River South	Eelgrass	N	OverSamp	-70.72682518	43.08301532
GB-SPR-1089-b	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.72718473	43.08297248
GB-SPR-1089-c	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.72667279	43.08297222
GB-SPR-1090-a	Lower Piscataqua River South	Eelgrass	N	OverSamp	-70.72472548	43.08388601
GB-SPR-1090-b	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.72442853	43.08378244
GB-SPR-1090-c	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.72481741	43.08392945
GB-SPR-1091-a	Lower Piscataqua River South	Eelgrass	N	OverSamp	-70.72999547	43.07272222
GB-SPR-1091-b	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.73022426	43.07253727
GB-SPR-1091-c	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.73011639	43.07260211
GB-SPR-1092-a	Lower Piscataqua River South	Eelgrass	N	OverSamp	-70.72699027	43.08460612
GB-SPR-1092-b	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.72674442	43.08453521
GB-SPR-1092-c	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.7270463	43.08461509
GB-SPR-1093-a	Lower Piscataqua River South	Eelgrass	N	OverSamp	-70.74041286	43.07927045
GB-SPR-1093-b	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.74041325	43.07929096
GB-SPR-1093-c	Lower Piscataqua River South	Eelgrass	N	OverSamp-alt	-70.73987527	43.07922349

Appendix C Epiphytes

- At each site, the first 6 eelgrass shoots collected are used for the epiphyte analysis. Because epiphyte load varies with blade age within a shoot, to obtain an average estimate for load within a shoot, one exterior leaf (older) and one internal leaf (younger) are selected from each shoot in the laboratory. Variation among shoots within a station are not a prime focus of the current study, and the replicate observations for the 6 shoots are pooled to calculate mean values per site and date
- Epiphytes are scraped from both sides of each leaf collected, and both epiphytes and seagrass leaves were separately placed in a drying oven for 24–36 h at 60–70 °C and dry weights of the material removed are determined for each leaf.
- While the term epiphyte biomass is used to refer to dry weight (g) of the material collected, the measurements include inorganic material since this material also reduces light available to the leaf. Leaf length and width are recorded and used to calculate leaf surface area for epiphyte attachment. The term epiphyte load is used here to refer to mg epiphytes cm^{-2} of seagrass leaf surface area (area of both sides of leaf).
- Laboratory observations should include notes on relative cover and broadly described type of epiphytes present, and serve as a quality check for consistency of the weights obtained.
- Analysis Options (not mandatory)
    - Epiphyte biomass can be compared with Zostera biomass using Box and Whiskers plots, and mean biomass compared between external and internal leaves for both variables using the non-parametric Mann- Whitney U-test. Relationships between epiphyte load and seagrass biomass can then be examined with linear regressions.
    - For analysis, epiphyte load data for both external and internal leaves can be divided into estuarine hydrodynamic zones (see Bilgili et al. 2005). Mean epiphyte load can then be compared with Two -way ANOVA. Assumptions of normality (Shapiro-Wilk test) and homoscedasticity (Brown-Forsythe) are recommended.

Calculating light reduction due to epiphytes (optional)

- In the laboratory, wash freshly removed epiphytes from a single seagrass leaf into a plastic cylinder with distilled water (60 ml). Place a light source above the cylinder, should be covered with a plastic diffuser plate. Meanwhile, place a PAR sensor (e.g., 2π, air, LI-190SA) below the chamber, and measure the amount of irradiance reaching the sensor. This value is then compared to a similarly measured irradiance value obtained using a second identical cylinder containing 60 ml of distilled water without epiphytes to control for temporal variations in strength of the light source.

APPENDIX D

Seagrass C:N Ratios from Tissue Analysis

Algal blade tissues and seagrass leaf tissue samples obtained directly adjacent to biomass cores is used for C:N determination.

I. OBJECTIVE
To determine C:N (and, potentially, P) ratios from seagrass tissue.

II. METHODS

1. PREPARATION
   a) Tissue samples for C:N:P ratios must be processed within three days of collection or dried at 60 °C for long-term storage. For seagrasses, newly formed leaves (the youngest leaf in a shoot bundle) are gently scraped and rinsed in deionized water to remove algal and faunal epiphytes. Algal tissues (if being analyzed) must appear healthy and free of epiphytes and debris.

   b) These rinsed samples are dried to a constant weight at 60 °C and homogenized by grounding to a fine powder using a mortar and pestle.

2. ANALYSIS
   a) Total carbon and nitrogen contents in blade or leaf tissues will be determined from two replicates of each sample by oxidation in a Carlo Erba model EA 1109 CHN elemental analyzer. Phosphorus content is not part of this project, but samples should be frozen for future analysis. Molar C:P, C:N, and N:P ratios can then calculated for evaluation of temporal and spatial trends.

APPENDIX E

SEDIMENT ORGANIC CONTENT AND GRAIN SIZE

I. OBJECTIVE

To determine sediment organic content determined by loss on ignition and characterize distribution of sediment size classes (gravel, sand and silt/clay fractions). See Erftemeijer and Koch 2001.

II. MATERIAL & EQUIPMENT

- Drying oven
- Muffle furnace
- Aluminum weighing dishes
- Dessicator
- Mortar and pestle
- 63µ and 2 mm sieves with catch pan
- Spatula, brush
- Plastic funnel

III. METHODS

1. ORGANIC CONTENT ANALYSIS

a) Label the underside of the aluminum pan by etching the unique serial number for the sample you are processing. Any ink or pencil labels will be lost upon combustion. Weigh labeled pan to obtain pan weight.

b) Split the total sediment sample roughly in half after either homogenizing the total sediment sample by mixing in a container or splitting the sample core vertically. One half will be used for determining organic content and one will be used for determining grain size.

c) Place one half of the total sediment sample into pre-weighed aluminum pan. Pick out any large shell material using forceps.

d) Dry in oven at 60° C for 24 hours or until completely dry. Place sample in dessicator until cooled to room temperature. Reweigh the sample to obtain the dry weight.

e) Heat sample in muffle furnace at 450° C for 4 hours to determine organic matter content. Place sample in dessicator until cooled to room temperature. Reweigh sample once it is sufficiently cooled to obtain the combusted weight.

f) Loss on Ignition is calculated as % weight loss after combustion using (Erftemeijer and Koch 2001):

$$LOI = 100 * ((dry\ wt - combusted\ wt) / dry\ wt)$$

2. GRAIN SIZE ANALYSIS
   a) Place remaining half of the sediment sample into mortar and gently grind apart the aggregates using only the weight of the pestle.

   b) After the sample has been gently disaggregated, dry sieve the sample through a stack of two sieves consisting of a 2mm (#10) and a 63µ (#230) stainless steel sieve plus a catch pan. Be careful not to touch the sieve with your hand or anything that will damage the mesh. Put the disaggregated sample into the 2mm sieve on top, cover and shake with a circular motion and tap until the sand fraction has passed through the 2 mm sieve, leaving the shell and gravel material on top of the 2mm sieve. The sand fraction is left on the 63µ sieve, and the silt-clay fraction (finest) is in the catch pan.

   c) Transfer the three grain size fractions into separate pre-weighed pans. Re-dry for an hour, then weigh when cool. The weight minus the weight of the pre-weighed pan yields the weight for each fraction.

   d) The weights of the three grain size fractions: 1) gravel/shell, 2) sand, and 3) silt/clay are summed to provide the total weight and the percent of each fraction by weight is calculated.

IV. TROUBLESHOOTING/HINTS

1. Drying of sediment samples can be checked by placing them back in the drying oven and reweighing the following day

2. Dry samples must be cooled in a dessicator before weighing. Dried and combusted samples will gain moisture from the atmosphere rapidly. Also, warm samples will give erroneous readings by creating convection currents around the pan of the balance.

V. STATISTICAL ANALYSIS & DATA USAGE

1. Input pan weight, dry weight, combusted weight, and dried sediment fractions to spreadsheet. Calculate % LOI and size fractions using appropriate formulae (above).

2. Calculate mean and standard error of five sediment samples for each eelgrass.

Appendix F
SOP for Assessing Wasting Disease

- Wasting disease is assessed as a percentage of all leaf area.
- The shoot is laid out on a lab bench with all intact leaves spread out so that they can be viewed simultaneously.
- Assessors are trained in how to recognize wasting disease, both by looking at the color (e.g., black versus brown or green) as well as shape; wasting disease tends to follow the shape of the cell walls, which are rectangular.
- The assessor estimates the percentage of total leaf area that is covered with wasting disease and notes that number on the data sheet.

Appendix G
SOP for Assessing Seagrass Biomass
(adapted from the SeagrassNet Manual, 2015)

- Empty the contents of the plastic bag into a sorting tray or on a clear area.
- Remove intact shoots and all below-ground plant material.
- Separate the seagrass shoots into the different species (e.g., eelgrass or widgeon grass).
- Separate the shoots for each species into:
  Leaf - green intact leaf blades connected to a stem or rhizome, although brown/dead attached leaves should also be included.
  Stem/Sheath – these plant parts connect the leaves to the belowground plant parts.
  Root/Rhizome - the belowground plant parts, including all the rhizomes (can be whitish to brown in color and are larger than roots) and roots growing below the sediment surface.
- Scrape epiphytes from the leaves (see Appendix C for epiphytes procedure).
- Remove all sediment from the sheath, roots, and rhizomes and re-rinse in fresh water.
- Place each component for each species in separate, paper bags with an open top and clearly label each envelope with a permanent marking pen as follows:
  - Site abbreviation (XX), site number (50), transect letter (A, B or C), quadrat number (1-12) e.g. "XX50.1A01" or "XX50.1A02" or "XX50.1A03"
  - Species abbreviation - Plant part (L, S, R) e.g. Hw – L, Hw – S, Hw - R
  - Date – write month out (Jan, Feb, Mar, Apr, May, Jun, Jul, Aug, Sep, Oct, Nov, Dec). Dates will read, e.g., 27 Jan 02, 15 Aug 02, *etc*.
- Place the biomass sample paper bags in the drying oven at 50 – 60° Celsius (120° Fahrenheit) for 24 – 48 hours or until the plant material is completely dry.
- Allow to cool, take the sample from the paper bags and weigh the sample on electronic scale in grams to an accuracy of 3 decimal places. Record the information on a datasheet.
- After weighing the sample, rewrap it in the paper bag with the label and date weighed and keep stored in a plastic bag. DO NOT THROW OUT the sample until you have received confirmation that the data has been entered and saved electronically.

Appendix H
SOP for Assessing Eelgrass Reproductive Capacity

For each reproductive shoot…

- Count the number of rhipidia, which are the main "branches" of the reproductive shoot (see figure below).
- Count the number of spathes per rhipidium. (Spathes are the sheaths that protect the flowers, both male and female parts.)
- For three spathes on each shoot, count the seeds and ovaries to assess an average per spathe. Seeds should be firm. If in doubt, use forceps to squeeze the seeds. Some seeds may be shrunken and soft, indicating an aborted seed.

(These procedures are most quickly done with one person examining the shoot and calling out numbers to an assistant or "scribe.")

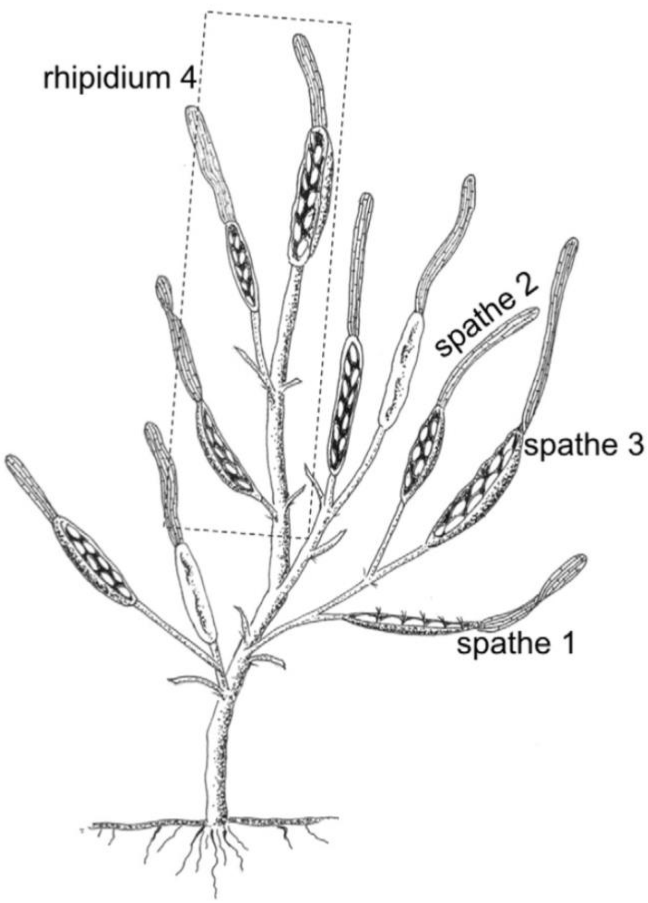

Figure 1. Stylized representation of a Zostera marina reproductive shoot with 4 rhipidia. Spathes on the second rhipidium are numbered sequentially in order of development. The youngest, tallest rhipidium (the 4th) is identified by a dashed box. (From L.J. Jackson et al. / Journal of Experimental Marine Biology and Ecology 489 (2017) 1–6.)