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### Benthic Habitat Mapping in Wellfleet Harbor and Vicinity



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Cover Image: R/V Marindin at the dock in Wellfleet Harbor. Photograph by T. Smith

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## Executive Summary

Wellfleet Harbor is one of four embayments in which maps of marine benthic habitats were developed as a part of a larger study for the National Park Service (Borrelli et al., 2019). This report documents the methods of data collection, processing and analyses necessary to produce those maps in Wellfleet Harbor and surrounding areas. With additional funding from the Town of Wellfleet and the non-profit Shellfish Promotion and Tasting Incorporated (SPAT Inc.), we were able to expand the study area beyond the boundaries of the Cape Cod National Seashore (CCNS) and include most of Wellfleet Harbor, Duck Harbor, and parts of Billingsgate Shoal in Cape Cod Bay. All areas mapped for this study were within the Wellfleet Harbor Areas of Critical Environmental Concern (ACEC), established in 1989.

The Center developed benthic habitat maps using the US Coastal and Marine Ecological Classification Standard (CMECS) to comprise a baseline inventory of geological and biological data. As a national standard, CMECS was designed to classify coastal and marine habitats throughout U.S. waters, and thus allowing other investigators to use the same methods to compare results through time and across multiple disciplines as well as in different coastal systems.

This study and associated data comprise a critical baseline record of biological and physical characteristics of Wellfleet Harbor and surrounding areas. As described throughout, the classification and mapping approach employed for this analysis is only one of many possible treatments of the data. There is an opportunity to explore the data collected during this study to better understand the importance of biotic habitat characteristics, such as macroalgal canopies and eelgrass beds, overlain on substrate composition. The results and maps from this study will be useful to guide future studies of coastal resources in Wellfleet Harbor.

Vessel-based acoustic surveys ( $n = 43$ ) were conducted in Wellfleet Harbor and Cape Cod Bay in 2015 and 2016. This yielded  $57.15 \text{ km}^2$  of sidescan imagery and  $35.32 \text{ km}^2$  of co-located bathymetric data, with a mean depth of 3.98 m, and maximum depth of 12.09 m. Benthic sampling was conducted in August and September 2015, samples were collected at 28 stations resulting in 84 sieved and preserved biological samples, 28 sediment samples, 28 water column profiles, and photographic and video data for each station. A total of 35 km of seismic reflection profiles (sub-bottom) were collected in 2015, resulting in the identification of seismic reflection facies (i.e., sediment units characterized by depositional environment) of the seafloor sub-surface, including estuarine channels, glacial lake floor deposits and marine mud.

### Key Findings:

- For this study, ecosystem-based mapping was prioritized - focusing on mapping embayments rather than along arbitrary delineations (such as CCNS boundaries). This system-wide approach increased the explanatory power of the benthic habitat maps, the significance of the statistics calculated and the robustness of the baseline data which can be used for future monitoring and management of Wellfleet Harbor. Further efforts can be made to increase awareness, participation, and funding by engaging local and regional entities.

- Vessel-based acoustic mapping was greatly enhanced using phase-measuring sidescan sonar (PMSS), which is ideal for shallow water (less than 10 m) mapping. Co-located bathymetry and sidescan proved to be particularly useful for identifying eelgrass and other submerged aquatic vegetation, changes on the seafloor such as bedform migration and both natural and anthropogenic structures on the seafloor. Dual frequency high resolution sidescan produced exceptional imagery useful for many applications.
- Seismic reflection profiling (sub-bottom) worked well in the shallow water environment of Wellfleet Harbor. Five seismic facies were identified in Wellfleet Harbor, including glacial lake floor deposits, and facies interpreted as bedforms representing the extension of the barrier spit down the western side of Wellfleet Harbor onto Billingsgate Shoal. Marine mud (facies 'M'), was found to be ubiquitous throughout much of the harbor, (up to 10 m thick) and was interpreted to be estuarine mud deposited in low energy basins. Additional seismic surveys would provide managers with important information for future dredge projects and management of resources within the harbor.
- Since a great proportion of samples were taken in physically dynamic environments it is not surprising that characteristics of the substrate (i.e., grain size metrics) were the best variables for explaining patterns in benthic communities, versus factors such as depth and sediment organic content. Overall, we could explain 55.9% of species distribution based on geoform, % gravel and skewness. Of the 9838 individuals comprising 98 species we found one individual of a cryptogenic species: *Polycirrus eximius* (a polychaeta worm) at station 16. A cryptogenic species is a species whose origins are unknown; meaning it may be either a native species or an introduced species but clear evidence for either origin is absent.
- Our models suggested temperature to be an important factor explaining species distribution in and around Wellfleet Harbor. However, water temperature is an ever-changing variable and depends on weather, climate and season. Since benthic grab samples were collected over five weeks and strong tidal flow brings in cooler water, modelled biotopes including temperature would have been considered temporal biotopes that can potentially change on a day to day basis and are therefore not mappable.

## Introduction

Managers recognize the growing importance of marine benthic habitat mapping for the development of sustainable, ecosystem-based management with the goal to responsibly utilize marine resources and to inform the design and management of protected marine areas (Borrelli et al., 2019). Benthic habitat maps provide baseline biological and geophysical data that allow scientists and managers to understand the distribution of biotic (living) and abiotic (non-living) resources on the seafloor (Shumchenia and King, 2010). Increasing anthropogenic pressures on the marine environment such as the construction of coastal infrastructure, dredging, bottom fishing and nutrient loading, has amplified the need for tools in which to better monitor marine ecosystems.

Wellfleet Harbor is known for its abundant natural resources which include a \$6.3 million commercial shellfish industry, commercial and recreational fishing and recreational activities such as boating, kayaking and swimming (<http://www.wellfleet-ma.gov/>). Managers and stakeholders have long been committed to the preservation of Wellfleet's resources. The Cape Cod National Seashore was established in 1961 and encompasses 61% of the town's total area (Figure 1). In 1989, the Wellfleet Harbor Areas of Critical Environmental Concern (ACEC) was established to protect the marine resources of the watershed (<https://www.mass.gov/service-details/wellfleet-harbor-acec>). The results of this study will serve as an important tool towards ecosystem-based management of Wellfleet Harbor and surrounding waters.

Vessel-based acoustic surveys were conducted in Wellfleet Harbor and Cape Cod Bay in 2015 and 2016. Biological surveys included grab samples for microinvertebrate and sediment analysis, and the collection of water column profiles and habitat data. These data were used along with the geophysical data collected by the acoustic surveys to develop benthic habitat maps for Wellfleet Harbor using the Coastal and Marine Ecological Classification Standard (CMECS) framework. Seismic Reflection Sub-Bottom Profiling (below the surface of the seafloor) was conducted in order to provide additional surficial and stratigraphic information for the characterization and analysis of basin evolution.

Development of surficial "benthic habitat" maps was a focal component of this work. It is important to note that there are many analysis options for integrating multiple data streams to create "habitat maps" for a range of purposes (Brown et al., 2011), and this study presents one option – a multivariate classification and regression tree approach to predict benthic biotopes – described in detail below. The maps were developed using CMECS, the national standard for classifying these types of data. The NOAA Integrated Ocean and Coastal Mapping Program's unofficial slogan "Map once, use many times" is particularly pertinent to this study (as well as the other three studies in this project) (A. Chappell, pers. comm.). The data collected for this study are vast and can be analyzed and mapped in numerous ways to explore and learn more about coastal processes, physical-biological linkages, benthic ecology, and other phenomena of interest to managers and stakeholders.

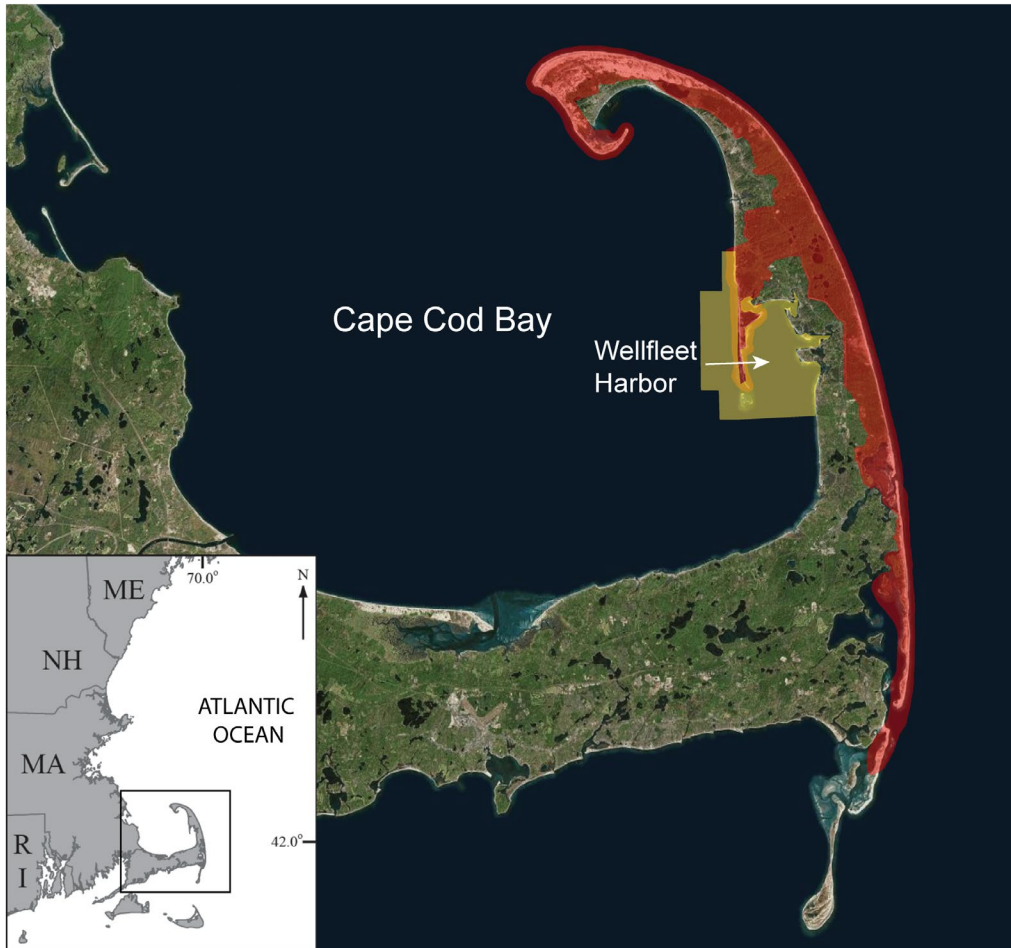


Figure 1. Locus map of The Wellfleet Harbor study area (yellow). Red area (orange in mapped areas) is the Cape Cod National Seashore boundary. All boundaries are approximate.

### **The Formation of Cape Cod and Wellfleet Harbor**

The morphology of Cape Cod is primarily the result of late Pleistocene glacial deposition by the retreat of the Laurentide ice sheet, and coastal processes in response to climate change and subsequent sea level rise (Uchupi et al., 1996). The last glacial maximum (LGM), when the ice sheet had reached its maximum extent, occurred in New England approximately 28,000 – 26,000 years before present (yr BP), (Peltier and Fairbanks, 2006; Balco and Schaefer, 2006; Balco et al., 2009). Locally, the maximum extent of the ice sheet is marked by end moraines on Nantucket and Martha’s Vineyard (Figure 2). End moraines are depositional features consisting of unconsolidated debris that accumulated at the ice front. The ice at the LGM in New England was approximately 500 m thick, and the sea level was approximately 120 meters below present sea level (Oldale and Barlow, 1986; Uchupi et al., 1996; Peltier and Fairbanks, 2006) Within a few thousand years after the ice sheet reached its maximum extent, glacial retreat began, and by 21,000 yr BP the ice occupied what is now Buzzards Bay and Cape Cod Bay (Ridge, 2003; Balco et al., 2009).



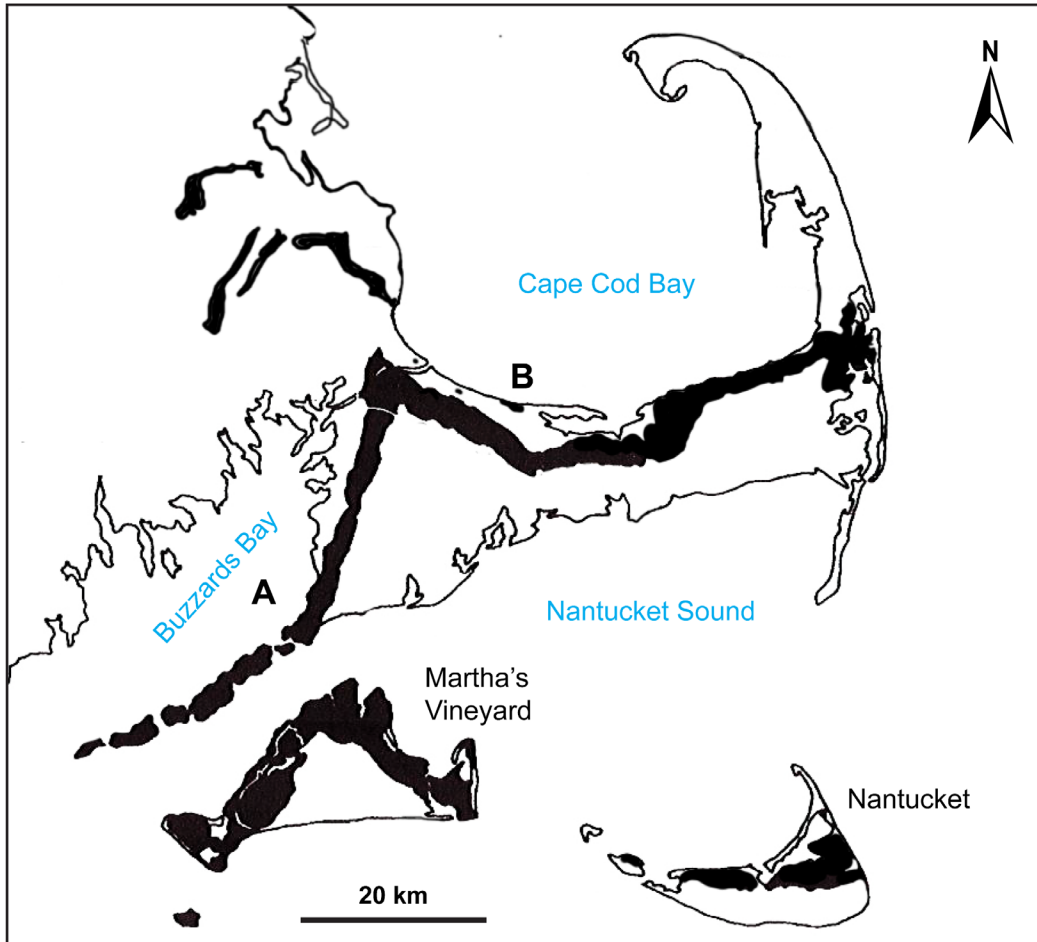


Figure 2. End moraines (in black) in Cape Cod and southern Massachusetts. A. Buzzards Bay end moraine. B. Sandwich moraine. Moraines on Nantucket and Martha's Vineyard mark the extent of the Laurentide Ice Sheet at the LGM in New England. Adapted from Schafer and Hartshorn (1965) with addition from Sirkin (1982).

The ice sheet consisted of three lobes controlled by the topography of underlying bedrock (Figure 3-A). The Narraganset Bay – Buzzards Bay lobe occupied the Narraganset Basin, and retreated generally westward. The position of the ice margin at this time is marked by the Elizabeth Islands, and the Buzzards Bay end moraines (Figure 2). The Cape Cod Bay lobe occupied Nantucket Sound and Cape Cod Bay. The South Channel lobe was located east of the Cape Cod Bay lobe, occupying the Great South Channel in the Gulf of Maine (Figure 3-A) (Oldale and Barlow, 1986). Climate fluctuations caused temporary re-advances, overriding former deposits and thrusting sediment and debris in front of the ice margin. The resulting landform is known as a 'tectonic end moraine'. The Sandwich end moraine formed in this manner as described by Oldale and O'Hara (1984). The lobes retreated at different rates, allowing glacial lakes to develop in front of the ice margin (Figure 3-B). Glacial Lake Cape Cod Bay was dammed to the north by the Cape Cod Bay lobe and to the east by the South Channel lobe (Figure 3-B) (Oldale and Barlow, 1986; Uchupi et al., 1996).

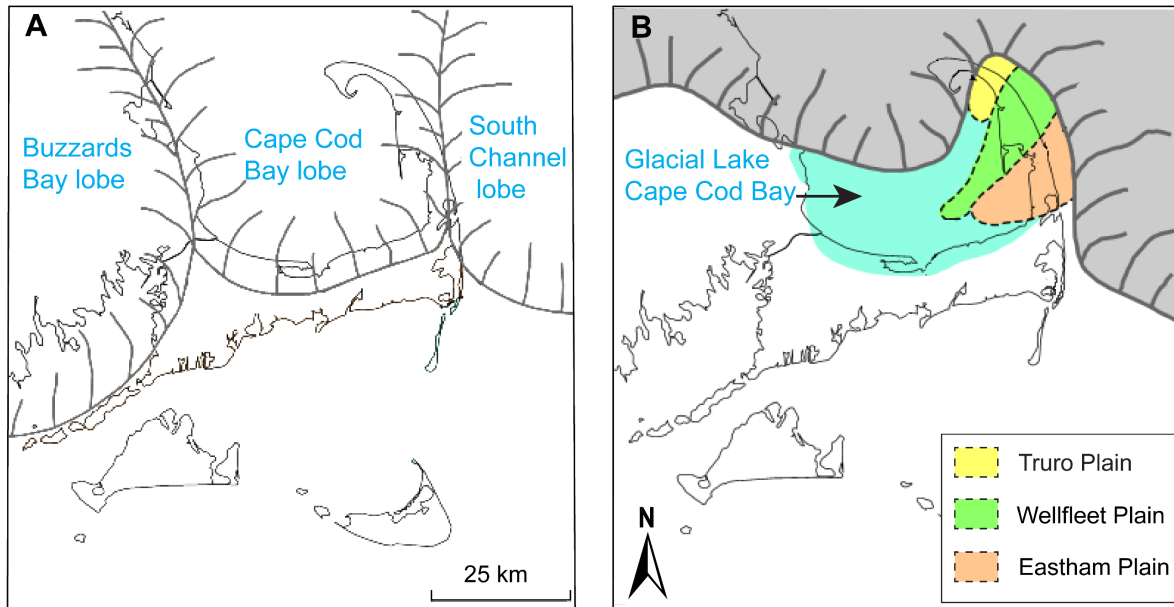


Figure 3. A. The three lobes of the Laurentide ice sheet in the Cape Cod Region at approximately 21,000 BP) modified from Oldale and Barlow, (1986). B. Glacial Lake Cape Cod Bay and the formation of the plains of lower Cape Cod, at approximately 18,000 BP, (Poppe et al, 2007, Balco et al., 2009), modified from Oldale and Barlow (1986).

Shortly after 21,000 yr B.P., the Cape Cod Bay lobe had retreated into the center of Cape Cod Bay, as the South Channel lobe retreated both northward and eastward. The ice margin is thought to have been located about 3 to 7 km east of the present coastline of lower Cape Cod at this time (Uchupi et al., 1996). The northward retreat of the Cape Cod Bay lobe was interrupted by a minor re-advance, deforming former meltwater deposits in front of it to form a moraine underlying Billingsgate Shoal (Oldale and Ohara, 1984; Uchupi et al., 1996). Glacial meltwater streams formed the expansive and relatively flat plains of Truro, Wellfleet and Eastham (Figure 3-B). Near the ice margin, sediment was deposited over and around stagnant ice. When the ice melted back, these deposits collapsed, forming a surface of irregular topography with steeply sloping topographic highs and lows (Oldale and Barlow, 1986).

Wellfleet Harbor occupies a part of a large ice block depression that formed as stagnant ice, possibly from a sub-lobe of the South Channel lobe, melted (Oldale and Barlow, 1986). Sediment deposited in holes or depressions in the ice formed Griffin Island, Great Island and Great Beach Hill.

Approximately 16,000 yr BP, the Cape Cod Bay and South Channel lobes had retreated north into Massachusetts Bay, and meltwater deposition from the South Channel lobe formed Stellwagen Bank. By about 14,000 yr BP northeastern Massachusetts was ice free (Uchupi et al., 1996).

The relative sea level rise rate had averaged about 6 m/1,000 yr until approximately 6,000 yr BP, when the rate slowed to about 2 m/1,000 yr until the sea reached its present level approximately 1,000 yr BP. By 6,000 yr BP Stellwagen Bank, and much of the Billingsgate Shoal moraine were submerged. Most of the coarse sediment derived from erosion of the west side of the Outer Cape was

transported southward to the submerging Billingsgate Shoal moraine (Uchupi et al., 1996). As the rate of sea level rise began to decrease 6,000 yr BP, southeastern Cape Cod began to take on its characteristic morphology (Davis 1895, Johnson 1925, Uchupi et al., 1996). Between 6,000-4,000 yr BP, barrier spits began to develop and subsequently small embayments were formed. Coastal salt marshes throughout New England began to form 4,000 yr BP (Redfield and Rubin 1962, Redfield 1972, Roman et al., 2000).

### **Wellfleet Harbor**

A barrier spit extending from north to south partially encloses the harbor from Cape Cod Bay (Figure 1). The spit is composed of islands connected by sediment entrained and deposited in the direction of net sediment transport, from north to south. A nodal point exists in the region just north of Griffin Island and has persisted since the 1930s (Figure 4) (Giese, et al., 2014). A nodal point is an area where the net direction of alongshore sediment transport diverges. Net sediment transport north of the nodal point travels in a northerly direction and the inverse is true to the south. The Cape Cod Bay shoreline in this area is somewhat protected from the predominant winter winds (NW) and the position of the nodal point is in part related to this sheltering. The shoreline to the north of the nodal point is protected from the NW winds by the Provincetown Hook and as such the SW winds move material to the north. The SW winds are buffered somewhat by Billingsgate Shoal, a large subaqueous depositional feature. The entrance to the harbor is large and not tidally restrictive and as such, little tidal lag is seen between Wellfleet Harbor and immediately adjacent areas in Cape Cod Bay. The mean tidal range is 3.05 m with a spring tidal range of 3.54 m (<http://tidesandcurrents.noaa.gov>).

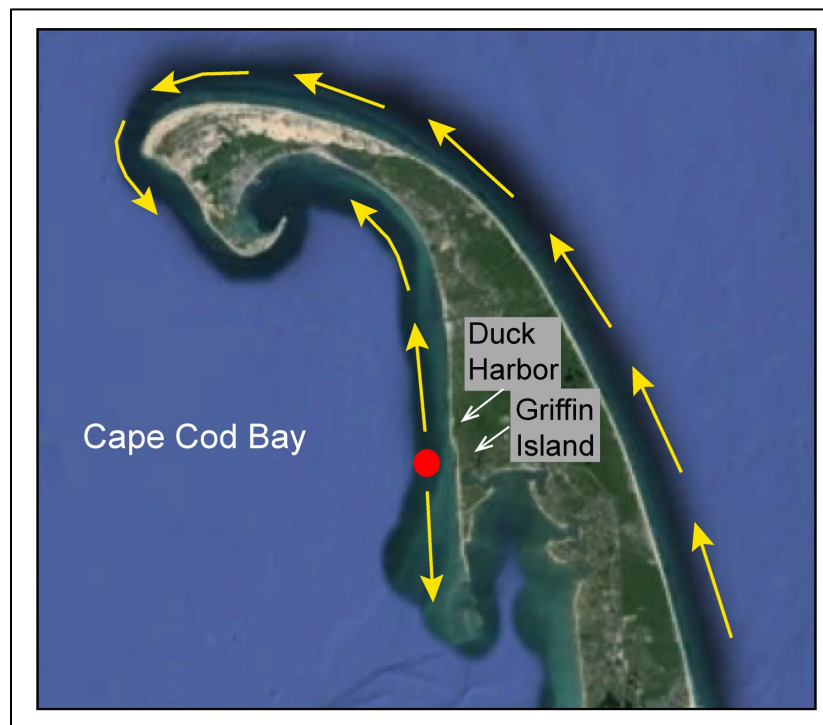


Figure 4. Sediment transport for lower Cape Cod and the approximate location of nodal point (red dot) along the shoreline between Duck Harbor and Griffin Island, after Giese et al., 2014.

## **Benthic Habitat Maps**

The purpose of this work was to integrate the physical and biological characteristics of benthic marine habitats from data obtained by CCS into a series of map products that describe the CMECS Geofom, Substrate, and Biotic Components. CMECS itself is “data agnostic” (FGDC 2012), meaning that as a classification scheme, it does not prescribe a particular method, set of methods, or analysis techniques. Indeed, this is a strength of CMECS, and one that allows the user to separate this type of project into three distinct steps: data collection, analysis, and classification.

There are three recognized approaches for integrating benthic physical and biological data into habitat maps (Brown et al., 2011). The first approach, “abiotic surrogacy”, does not truly integrate physical and biological data but assumes that physical environmental patterns correspond to biological patterns. The abiotic surrogate approach is applied at broad scales and is used to define benthic landscapes from remotely sensed data, often with little or no ground-truthing. For example, Dunn and Halpin (2009) modeled seafloor rugosity from low-resolution (90m) bathymetry data as a proxy for high biodiversity.

The second and most common approach, known as “assemble first, predict later”, can be used to develop single-species maps or assemblage maps based on observed physical and biological characteristics using a classification scheme as a guide (Brown et al., 2011). With this approach, physical and biological datasets are each analyzed separately, i.e., geologic characteristics are delineated from acoustic and grain size data, then biological characteristics are identified from analysis of grab samples or underwater photography. Maps are constructed by overlaying the occurrence of biological characteristics with the geologic characteristics and determining the correlation between datasets. The degree of correlation between geologic and biological characteristics is used as justification for assigning habitat units from the chosen classification scheme and extrapolating those habitat units across the study area into places where ground-truthing data were not collected. However, benthic infauna often overlap sediment transitions or boundaries and equating substrate with benthic assemblage type will lead to inaccurate maps (Diaz, Solan et al., 2004, Stevens and Connolly 2004, Shumchenia and King. 2010). This underscores the need for the third approach.

The “predict first, assemble later” approach, described by Brown et al., (2011) as more sophisticated and objective than the previous two, and noted that more recent studies are beginning to use this strategy. The concept underlying this approach is that the physical and biological data together are used to inform the development of map units – species or assemblages are modeled as functions of multiple physical variables. In the case of single species mapping, this approach is known as habitat suitability modeling (e.g., (Howell et al., 2016). Applied to species assemblages or communities (e.g., Degraer et al., 2008), this approach identifies physical variables that explain the most variance in benthic community structure, then uses those variables to create predictive habitat maps.

Since this study sought to represent ecologically meaningful physical-biological linkages and develop full-coverage habitat maps in a rapid and reproducible manner, we chose to implement the third analysis strategy: “predict first, assemble later”. Further, this analytical approach has been previously employed to identify and predict CMECS Biotopes in a shallow soft-sediment environment in Northeastern United States (Shumchenia and King. 2010), (McHenry et al., 2017). Distance based linear models were run to identify the driving physical variables responsible for the variation in benthic community structure in Wellfleet. Additionally, the thresholds of each significant variable were

calculated to determine different levels or gradients of variables influencing species community. This information was used to develop full-coverage maps showing benthic assemblages and their determining physical drivers. The resulting maps, therefore, contain units that correspond to CMECS Biotopes, “a combination of abiotic habitat and associated species” (FGDC 2012).

## Methods

### Vessel-based Acoustic Surveys

Two research vessels were used to collect acoustic data, the R/V Marindin and the R/V Portnoy (Figure 5). The R/V Marindin is a 1995 Eastern® I/O that has been modified for all-weather, shallow-water operations. It has a retractable bow mount with power hoist to raise and lower the sonar for safe operation and ease of deployment/retrieval. The bow mount eliminates most of the noise from the vessel and engine thus improving the quality of the acoustic data. This vessel combines an adequate beam (2.54 m) that yields stability at low survey speeds, a shallow draft (0.61 m) for safe operation in nearshore waters, and a modified V-hull for optimal transit time.

The R/V Portnoy is a custom-built, ultra-shallow draft pontoon boat designed specifically to survey in low energy embayments around 1 m deep (Borrelli et al., 2016) and was used extensively in 2016. Although it was designed for mapping Herring River, it performed well in the larger embayment. A 2-kW Honda™ gas-powered generator is used to power all electronics for the hydrographic surveys onboard both vessels.

To conduct high-resolution, vessel-based acoustic surveys an Edgetech 6205, a dual-frequency, phase-measuring sidescan sonar, was used. Its operating frequencies are 550 and 1600 kHz for sidescan sonar imagery and 550 kHz for bathymetry. The sidescan sonar range resolution is 1 cm, at 550 kHz, and 0.6 cm at 1600 kHz. The bathymetric range and vertical resolution are both 1 cm. The effective bathymetric swath width is 6-8 times the height of the sonar over the bottom (Edgetech, 2014). In addition a Teledyne TSS DMS-05 Motion Reference Unit mounted on the sonar was used to collect data on heave, pitch, and roll, measuring heave to 5 cm and roll and pitch to 0.05° (Teledyne TSS 2006). A HemisphereGPS® V110 vector sensor was used to measure heading. Two differential GPS receivers spaced 2 m apart yielded heading accuracies of <0.10° RMS (HemisphereGPS 2009).

Bathymetric data comprises the vertical and horizontal positions of each sounding: x and y being the horizontal coordinates (latitude and longitude) and z representing the raw depth from the sonar to the seafloor. The raw depth is then corrected to a vertical datum (a reference point, such as mean sea level), and tide corrections using Real-Time-Kinematic GPS (RTK-GPS). A Trimble® R8 GNSS receiver utilizing Real-Time-Kinematic GPS (RTK-GPS) was used for positioning and tide correction for vessel-based surveys.



Figure 5. Top: The R/V Portnoy, a custom-built pontoon. An Edgetech 6205 phase measuring sidescan sonar is custom mounted at the bow on both vessels. Bottom: The R/V Marindin.

To collect RTK-GPS data, CCS uses a proprietary Virtual Reference Station network (KeyNetGPS) that provides virtual base stations via cellphone. This negates the need to setup a terrestrial base station or post-process the GPS data, thus reducing costs, streamlining the field effort, and maximizing vessel-based survey time.

CCS undertook a rigorous analysis of this system beginning in 2012 to quantify the accuracy of this network (Mague and Borrelli in prep). Twenty-nine (29) National Geodetic Survey (NGS) and Massachusetts Department of Transportation (MassDOT) survey control points, were measured and compared with published state plane coordinate values relating to the Massachusetts Coordinate System, Mainland Zone (horizontal: NAD83; vertical NAVD88). The overall uncertainty analysis for these data yielded an average error of 0.008 m in the horizontal (H) and 0.006 m in the vertical (V). A Root Mean Squared Error (RMSE) of 0.0280 m (H) and 0.0247 m (V) and a National Standard for Spatial Data Accuracy (95%) of 0.0484 m (H) and 0.0483 m (V).

Edgetech's Discover Bathymetric® software was used to monitor all incoming data streams and control settings for onboard acoustic instruments to optimize data quality for at-sea conditions. Survey planning was performed using Hypack Survey® for line planning, coverage mapping and helmsman navigation. Both Discover Bathymetric® and Hypack's Hysweep® were used to collect data with the final raw output in JSF and HSX file formats respectively.



The JSF files were imported into SonarWiz® where a combination of automated and manual data processing was undertaken, including bottom tracking, slant range correction, offset entry and gain setting adjustments. After appropriate processing of each data file, mosaics were generated, which were then exported in a raster format (Geotiffs).

Post-processing of bathymetric data was performed using CARIS HIPS®. Raw data (HSX) files were converted to CARIS proprietary format using vessel configuration files developed from vessel offsets, and device information. RTK-GPS tide corrections were applied in the conversion process. Sound velocity corrections were applied using measurements collected in-situ by an internal sound velocimeter located in the sonar housing and water column profiles obtained from casts performed for each survey using a Castaway® CTD. Calibration tests were performed in the field in order to determine motion and timing offsets (roll, pitch, yaw and latency). Those offsets were recorded in the vessel file and applied when the survey lines were merged. Total Propagated Uncertainty (TPU) was computed using device manufacturer specifications recorded in the vessel configuration file. Filters were applied to remove depth outliers and “noise” typically found in the outer regions of the swath. Where necessary, area editors were used to remove spurious soundings.

## **Benthic Sampling**

### ***Field work***

To determine the biological and physical structure of the benthic habitats, field surveys were conducted for invertebrate and sediment characterization, water column structure, and video imagery. To effectively characterize each study location, benthic survey stations were determined for each system using a randomized tessellation stratified design. To provide balanced spatial coverage across the systems and statistical power of randomization, a tessellated hexagon grid is overlain onto the study area, and random points are selected within each hexagon.

All samples were collected aboard the R/V Marindin using a Young modified Van-Veen grab sampler (4 L) (Figure 6). A Castaway® CTD was used for the collection of water column data, (temperature and salinity), and a Trimble® R8 Global Positioning System (GPS) to obtain horizontal and vertical coordinates for each sample. A GoPro Hero 3™ was attached to the Van-Veen grab and high-resolution video was collected for each sample to aid in bottom characterization and documentation. The video was of sufficient resolution that screenshots could be obtained for imagery related needs. At each station, three biological replicates were taken. Each waypoint recorded was labeled by date, system name, station name and replicate number. All GPS data points were downloaded to a .csv file and imported into ArcGIS for subsequent mapping.

### ***Biological Samples***

At each benthic survey station, four replicate grab samples were collected from the seafloor, three biological replicate grab samples and one sediment grab sample using a Young modified Van Veen grab sampler (Figure 6). The anchor line was let out approximately 1 meter between each grab sample replicate to ensure that no previously disturbed areas were resampled. The Van Veen grab samples a surface area of 0.04 m<sup>2</sup> to a depth of 0.1 m below the seafloor for a total volume of 0.004 m<sup>3</sup> (4 liters). The Van Veen grab is well-suited for sand- to mud-sized samples ( $\leq 2$  mm) but does not function as well in areas with coarser grain sizes. A successful sample was attained when the two

scoops of the Van Veen grab were fully closed, at least 2 liters of material were sampled, and the surface of the sample was level-indicating the Van Veen grab did not sample the seafloor at an angle. The sampler has viewing plates allowing the user to view the top of the sample without disturbance for verification or further sub-sampling. When unsuccessful sampling was encountered due to rocks or shells interfering with the jaws closing, four attempts were made to sample with the Van Veen before the station was rejected. In this case, the nearest next randomly selected station replaced the original station. For each grab sample, a photograph of the substrate surface was taken upon the sampler returning to the research vessel, the depth of oxygenated soil was measured using a ruler, and a note was made of any biological structures on the surface including shells, worm tubes, algae/eelgrass, etc.

The contents of the Van Veen were then emptied into a bucket; a low energy wash of seawater was used to rinse any substrate adhering to the Van Veen into the bucket. The contents of the bucket were then sieved through a 1 mm mesh to retain organisms, detritus, and substrate greater than 1mm in size. A low energy wash of seawater and gentle manual agitation was used to sieve the sample to reduce damage to biological specimens. Any large bivalves, crabs, or vertebrates (fish) were measured, counted and identified (or photographed for later identification) before being returned to the water. Larger, mobile organisms collected by this method are considered ancillary data, as benthic grab sample gear cannot provide quantifiable estimates of their abundance or density. The material retained on the sieve was transferred to a fine mesh bag and brought back to the lab for preservation in 80% ethanol. Water column data was collected using a Castaway® CTD. One cast to the seafloor was conducted at each station to collect conductivity, temperature and depth (CTD) data after the boat had been anchored at the station, but before collecting the grab samples. The CTD has a built-in GPS which records latitude and longitude at the start and end of the cast. The time of cast was recorded, and the depth indicated by the CTD was verified against the R/V's depth sounder reading to increase the confidence that the CTD had reached the seafloor.

### ***Sediment Samples***

In addition to the three biological samples taken at each station, a fourth sample was used to document the grain size of the sediment. This sample was taken between the second and third biological replicate to ensure that the sediment sample was generally representative of the substrate sampled by the biological replicates. For each sediment grab sample, a photograph of the substrate surface was taken. The surface sediment was transferred to a 100ml Whirl-Pak®, and later dried at the lab for future analysis.



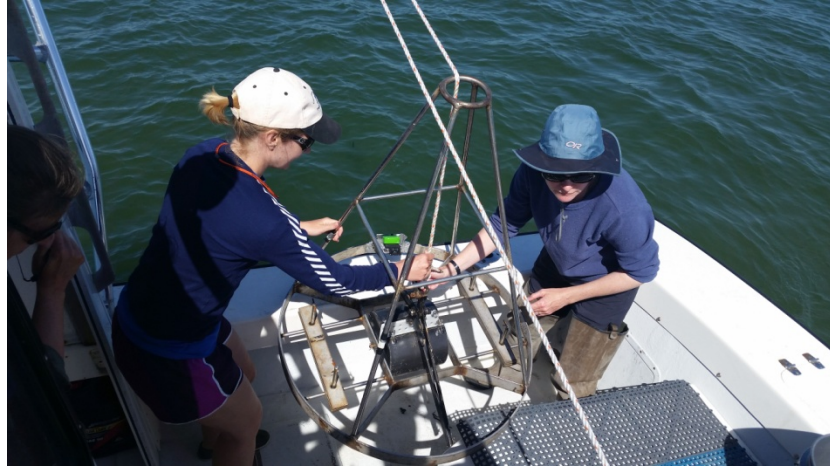


Figure 6. Young-Modified Van Veen grab sampler. C.G. Kennedy and Dr. M. Tyrrell Pictured onboard the R/V Marindin.

### **Invertebrate sample processing**

To determine the benthic invertebrates found in each biological grab sample, the contents of each grab were transferred to triple-labeled glass jars and preserved with 80% ethanol with Rose Bengal to dye invertebrates. Date of field sampling, preservation, processing and identification were all recorded on sample tracking data sheets as well as any notes about samples.

To sort out or “pick” the invertebrates from the substrate, the preservative was drained from the sample and disposed of according to CCS/NPS hazardous waste management plan. The sample was gently spread out into a large white plastic trays and water was added. The sample was visually inspected, and all invertebrates were picked out of the sample and sorted into general categories as discerned by the unaided eye (i.e., worms, shellfish, amphipods etc.). All personnel and volunteers were trained by the project biologist on proper picking technique and on general visual cues to find invertebrates. Quality control for each sample was performed by the project biologist by double checking each portion of each sample to ensure that all invertebrates had been found. Specimens were then immediately identified or preserved in 70% ethanol in 20 ml glass scintillation vials.

Specimens were identified by the project biologist using a dissecting microscope. All initial and final identifications, counts, and any notes were recorded on the identification data sheet. Specimens were identified to species level when possible or to genus, family or order depending on the difficulty of identification. All identified specimens were counted. Pictures of representative specimen of each species were taken using a digital microscope camera. Unidentified specimens were photographed using the same methods and were preserved to be identified/verified at a later time.

### **Sediment sample processing and analysis**

To characterize the sediment substrate of the benthic habitat for each sample location, the frozen sediment samples were processed for sediment grain size analysis and organic matter content. The sediment samples were thawed, and the excess overlying water was removed using a syringe, being careful not to disturb sediments.

### Organic matter content by loss on ignition (LOI)

To determine organic matter content of sediments for each sample, 20-30 grams of sediment were placed on pre-weighed aluminum trays, and the wet weight of the sample was recorded before being placed in a drying oven at 105°C for 24 hours. Dried samples were removed from the oven and placed in a desiccator. Each sample was weighed, and the dry weight was recorded. After recording the initial dry weight, all samples were broken using either a clean spatula for sandy samples or a clean mortar and pestle to grind finer samples. After a sample was ground, it was re-dried and reweighed to account for any lost material. To determine the proportion of organic matter, the homogenized samples were placed in a muffle furnace at 550°C for four hours. After ignition, the samples were re-weighed, and the percent organic matter as loss on ignition was determined by the following calculation.

$$\text{LOI (\%)} = (M_{\text{dry}} - M_{\text{dish}}) - (M_{\text{ignite}} - M_{\text{dish}}) / (M_{\text{dry}} - M_{\text{dish}}) * 100$$

$M_{\text{dry}}$  is the weight of the dried sample (at 105° C) plus the aluminum dish

$M_{\text{ignite}}$  is the weight of the ignited sample (at 550° C) plus the aluminum dish

$M_{\text{dish}}$  is the weight of the aluminum dish

LOI data were then corrected for salt content by using salinity data from CTD casts.

### Grain size analysis

Percentages of each of the size fractions for each sample were calculated from grain size data measured by the following methods:

#### *Grain size analysis gravel fraction (> 2mm)*

For those samples with larger (gravel) sized grains, the fraction of sediment with a grain size greater than 2 mm (gravel) was measured by sieving. The sample was sieved in a 2mm sieve and the fraction of sediment retained by the sieve was weighed. Shells then were manually removed and weighed.

The percentage > 2mm grain size fraction was calculated as.

$$> 2\text{mm (\%)} = (M_{>2\text{mm}} - M_{\text{shell}}) / (M_{\text{total}} - M_{\text{shell}})$$

$M_{>2\text{mm}}$  = weight of ignited >2mm substrate (including shells) plus the aluminum dish

$M_{\text{shell}}$  = weight of shells manually removed from >2mm substrate plus aluminum dish

$M_{\text{total}}$  = total weight of sample plus aluminum dish

#### *Grain size analysis for sand and fine fractions (< 2mm)*

Grain-size analysis of grains < 2 mm in size was conducted using a Beckman-Coulter LS 13 320 Laser Diffraction Particle Size Analyzer at the Woods Hole Oceanographic Institute's Coastal Systems Laboratory. Sediment samples were thawed and wet sieved to remove all particles > 2 mm. To remove any organic content that could interfere with the particle analyzer, samples were pre-treated with hydrogen peroxide by placing 5-10 grams of sediment sample into a clean, labeled 50 ml

centrifuge tube and adding 1 ml of 30% hydrogen peroxide. The sample was then capped, gently shaken and uncapped to allow for reaction to occur. Hydrogen peroxide was added in 1 ml increments to the sample until no reaction (no bubbling or foaming) was observed (up to 10 ml of hydrogen peroxide per sample). Once the reaction was complete, the tube with sample was filled with deionized (DI) water and allowed to sit overnight to ensure that any remaining hydrogen peroxide was removed. The samples were then centrifuged at 2,200 rpm for six minutes and the water was decanted. Samples were stored in a refrigerator until analysis in the particle analyzer.

Samples were individually run on the particle analyzer according to manufacturer protocols. Prior to loading into analyzer, each sample was vortexed for 10 seconds to evenly mix the sample and a small amount of sample was placed in the Beckman Coulter plastic tube using a spatula. The tube and spatula were carefully rinsed with DI water between running samples. All grain size results were saved to .csv files. All data were reported using Wentworth grain size thresholds and classes (Folk 1974).

### **Seismic Reflection Profiling**

This study utilized seismic reflection profiling, a method of imaging the subsurface using pulses of acoustic energy (sound waves) propagated into the sediment (Figure 7). Sound waves reflect from the boundaries between materials with different acoustic impedances (i.e., a constant related to the propagation of sound waves in an acoustic medium), allowing sedimentary layers with different bulk densities to be discerned. The highest contrast in acoustic impedance occurs between the seafloor and the adjacent water column, especially in areas of particularly hard or dense surficial sediment. Sound waves propagated into the sediment reflect back to the towed instrument and transmit up the cable to the processing software. In areas of high contrast in acoustic impedance, the sound waves can ‘bounce’ between the seafloor and instrument, producing an echo of the seafloor. This echo appears on the seismic reflection profile at multiple ranges of the water depth (i.e., a water depth of 5 m, multiples would occur at 10 meters, 15 meters and so on). These echoes are known as “multiples”.

Profiles were collected using an EdgeTech, SB-216S Full-Spectrum sub-bottom profiler, operated at a frequency sweep of 2 - 10 kHz, producing a vertical resolution of <15 cm (Edgetech 1998). Towfish height was maintained 1 m below the surface of the water, towed at a speed of < 1.5 m/s. Spatial location was embedded into the sub-bottom files using the serial NMEA 0813 (National Marine Electronics Association) output of a Trimble R8 RTK- GPS with reported accuracies discussed above. Depth to reflectors was calculated using an acoustic velocity of 1,500 m/s in both water and sediment.

Actual penetration of the seismic signal and resolution of adjacent layers depends on the frequency and power of the seismic system and the nature of the subsurface sediment. High-frequency chirp systems provide high resolution; however, they have a more limited penetration below the seafloor. Lower frequency seismic systems offer more penetration below the seafloor but offer less resolution of layers. This study utilized a high-frequency seismic reflection profiler, with a sweep from 2-10 kHz. Penetration is typically greater in lower density (often finer-grained) sediment; however, the

presence of naturally occurring gas (e.g., methane) scatters the seismic signal and obscures the geology beneath.

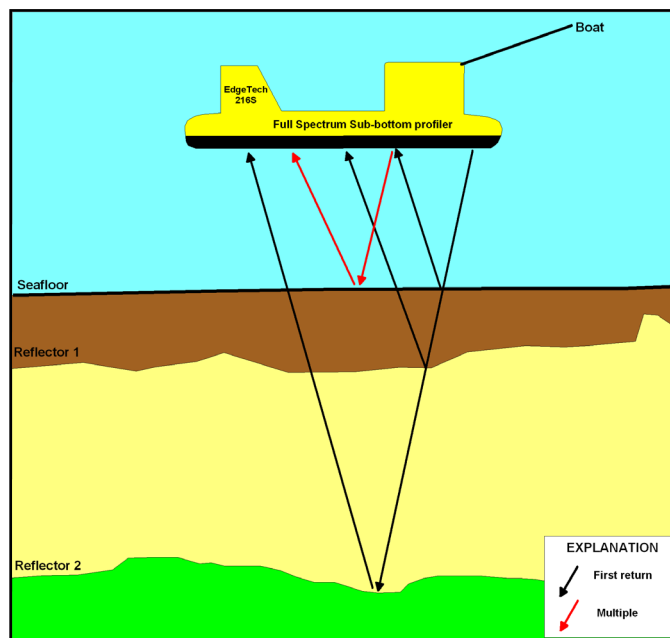


Figure 7. Schematic view of the seismic reflection profiler used in this study.

SonarWiz™ was used to process the seismic reflection profiles. On all files, the seafloor was manually digitized, allowing the images to be accurately corrected for time varied gain and contrast to maximize visibility of internal reflectors or sediment layers. Time varied gain accounts for the inherent differences in intensity between returning signals with depth in profile. The seismic reflection profiles are displayed in an inverse medium yellow-orange known as a Klein color scheme, named for the color of analog paper records produced by that company's wet-paper recordings in the 1970s, and we believe that the inverse Klein scheme allows us to better see detail on the digital records than traditional gray-scale images. Interpreting seismic reflection profiles is done by identifying seismic facies. Seismic facies are sedimentary packages, distinguishable from adjacent units based on internal characteristics, (i.e., the intensity, spacing, continuity, and internal geometry of seismic reflectors), external geomorphic form, and stratigraphic relationship to other units (Roksandic 1976, Vail et al., 1977).

### **Benthic marine habitat mapping framework**

Each dataset used in the production of the final data products can be mapped and interpreted separately. However, studies show that abiotic factors such as grain size, sediment organic content, and geomorphology can (partially) explain the variance observed in benthic community composition. The reverse can also be the case when certain biotic assemblages influence the physical composition and/or structure of the environment (e.g., shellfish beds, tube worm mats, eelgrass beds). For this study we focused on these physical-biological relationships which are critical to effective resource management, aimed to characterize them and create representative benthic habitat maps by using CMECS.

The physical data (grain size and acoustic data) were used to classify the CMECS Geoform Component and Substrate Component. Data collected on species abundance and diversity were used to classify the CMECS Biotic Component. Finally, we used statistical approaches to identify physical variables that explained the highest proportion of the variance in the benthic community data and classified the results into CMECS biotopes. Because the biotopes reported here are based on a single set of observations for each area and are therefore not predictable (repeat measurements are necessary for predictions), we refer to these results as “preliminary biotopes”. Preliminary biotopes give us a sense for which physical variables are influencing or driving benthic community composition in each study area.

### ***Physical characteristics***

#### CMECS Geoforms

The CMECS Geoform Component describes the major geomorphic and structural characteristics of the coast and seafloor, but is not intended to be a geological classification per se (FGDC 2012). Rather, the Geoform Component describes aspects of the physical environment that are relevant to, and drivers of, benthic community composition and distribution (FGDC 2012). At the scale of the data collected (i.e., 1-meter resolution swath bathymetry and 0.5-meter backscatter imagery), Level 1 and Level 2 Geoforms are readily described. Level 1 Geoforms are generally larger than 1 km<sup>2</sup>, whereas Level 2 Geoforms are generally smaller. An example of a Level 1 Geoform is a “Basin”; an example of a Level 2 Geoform is “Ripples” (e.g., bedforms). Level 1 and Level 2 Geoforms were delineated by classifying several metrics derived from the bathymetry grid for each area using the Benthic Terrain Modeler (BTM) Toolbox in ArcGIS Desktop (Wright et al., 2012). We selected this method based on its rapidity and reproducibility. Using the bathymetry grid as an input, the slope, fine-scale bathymetric position index (BPI), and broad-scale BPI were calculated.

The slope for each cell was calculated as the maximum rate of change from the cell to its neighbor using the BTM Toolbox. The output was a continuous raster.

BPI is a focal mean calculation where a cell’s elevation is compared to surrounding cells within a user-defined area. BPI is greater than zero where ridges or crests exist and less than zero where depressions or valleys exist. BPI is calculated using the following equation,

$$BPI < scalefactor > = int \left( (bathy - focalmean(bathy, annulus, irad, orad)) + 0.5 \right)$$

Where *scalefactor* = outer radius in map units, *irad* = inner radius of annulus in cells, *orad* = outer radius of annulus in cells, and *bathy* = bathymetric grid.

Given that the input bathymetry grid had a resolution of 1 m, search radii were chosen that ensured that the algorithm would detect features <1km<sup>2</sup> in size (i.e., between the expected size of CMECS Level 1 and 2 Geoforms). Broad-scale BPI was calculated using an inner radius = 25 m and an outer radius = 250 m. Fine-scale BPI was calculated using an inner radius = 5 m and an outer radius = 25 m. These search radii, therefore, could detect features from 5 meters across to 250 meters across. Using the BTM Toolbox, the BPI grids were standardized by subtracting the mean, dividing by the standard deviation, and multiplying by 100.

To distinguish geomorphological features based on Broad- and Fine-scale BPI values, slope, and depth, the classification dictionary in the BTM Toolbox (Table 1) was developed for this study. To distinguish between “Flat” (0 - < 5°) and “Sloping/Steeply Sloping” (>5°) areas, the CMECS Slope Modifier was used. The CMECS “Shallow Infralittoral, 0-5 meters” Benthic Depth Zone modifier was found to be insufficient for describing relevant Level 1 and Level 2 Geoform types in the CCNS study areas. Therefore, habitats within flat areas were further distinguished by applying depth thresholds of 1 and 3 meters, which were described as customized CMECS Benthic Depth Zone Modifiers.

Table 1. Classification dictionary developed in the Benthic Terrain Modeler (BTM) toolbox for Cape Cod National Seashore. BPI values are standardized and multiplied by 100 (i.e., dimensionless).

Geoform	Broad BPI	Fine BPI	Slope (°)	Depth (m)
Basins and channels	< -100			
Flats <1m	-100 – 100	<100	0-5	<1
Flats between 1-3m	-100 – 100	<100	0-5	1-3
Flats >3m	-100 – 100	<100	0-5	>3
Bedforms and shallow slopes >5°	-100 – 100	<100	>5	<3.5
Margins and deeper slopes >5°	-100 – 100	<100	>5	>3.5
Platforms	-100 – 100	>100		
Banks	> 100			

### CMECS Substrate

The CMECS Substrate Component is a characterization of the composition and particle size of the surface layers of the substrate (FGDC 2012). Substrates represent the non-living components that support, intersperse, or overlay the living components of the seafloor environment (FGDC 2012). The CMECS Substrate Component uses Wentworth grain size thresholds and classes (Folk 1974).

To classify Substrate Subgroups at each sampling point, percentages of gravel, sand, silt, and clay fractions of each sample were used. Substrate Subgroups are the finest classification level in the Substrate Component and include units such as “medium sand”, “very fine sand”, and “silt”. Classification was performed using SEDCLASS software (Poppe et al., 2003), and then the relevant Substrate Groups, Subclasses, and Classes for each sample were identified.

To develop a continuous map of substrate types for each area, the median grain size at each sampling point was interpolated using *spline with barriers*. A relatively simple kernel smoothing method was employed, which interpolated median grain size within each study area, bounded by a polygon of that area. Interpolation provides an objective, repeatable, and rapid way to estimate median grain size across each study area without an extensive field sampling effort. While interpolation will introduce uncertainty into the final products, the tradeoff for full coverage (i.e., reproducible maps) is worthwhile. Importantly, the median grain size metric was expected to yield a different classification result than the classification derived from the station specific weight percentages of gravel, sand, silt, and clay. The resulting median grain size surfaces were then classified by CMECS Substrate Subgroup units.

## ***Biological characteristics***

### CMECS Biotic Component

The CMECS Biotic Component deals with the classification of organisms in both the water column and on the seafloor; here we deal only with organisms on the seafloor (i.e., CMECS Biotic Setting = Benthic Biota). We can further narrow our scope of classification to the Biotic Class “Faunal bed” since all of the observations provided by this project were from sediment grab samples. Faunal beds are highly dependent on substrate type and include two Subclasses: “Attached fauna” and “Soft sediment fauna”. The next two hierarchical levels are Biotic Groups and Biotic Communities. We defined Biotic Communities based on species dominance, then described the appropriate Biotic Group and Class for each Community.

Biotic Communities were defined by cluster analysis of the benthic invertebrate species data in PRIMER (Clarke and Gorley 2015). First, the species-sites matrix was reduced to include only those species contributing to the top 95% of the total observed abundance. To verify that this new species-abundance matrix was representative of the benthic community in each area, the correlation coefficient between matrices based on the original and top 95% of total observed abundances were calculated. A Pearson correlation resulted in statistically significant similarity (0.9916) between 100% and 95% abundances. As a result, the 95% abundance matrices were found to be representative of the dataset. Using the top 95% dataset, the mean abundance was calculated for each species across all three replicate samples at each site. Then, the data were fourth root transformed to reduce the influence of highly abundant species and a dissimilarity matrix was calculated using the Bray-Curtis index of dissimilarity. PRIMER’s SIMPORF and Cluster methods were employed, to determine the optimal number of clusters. If the same species was dominant in more than one cluster, they were classified as the same CMECS Biotic Community (FGDC 2012).

### Preliminary biotopes

To more fully examine the relationships between physical variables and benthic community composition, distance based lineal modelling (DistLM) was conducted using the PERMANOVA+ extension on PRIMER (PRIMER-E v7, Plymouth). The model analyses the relationship between a multivariate dataset (benthic community dataset), as described by a resemblance matrix (Bray Curtis dissimilarity) and a set of one or more predictor variables (sediment characteristics) using distance-based redundancy analysis (dbRDA) (Figure 8). The routine allows for sediment characteristics to be considered individually or grouped together in specific sets and obtains p-values testing the null hypothesis (no relationship) using the appropriate permutation methods (Clarke and Warwick 2001). DistLM does a partition of variation according to a regression or multiple regression model and can be used to analyze models containing a mixture of categorical and continuous variables.

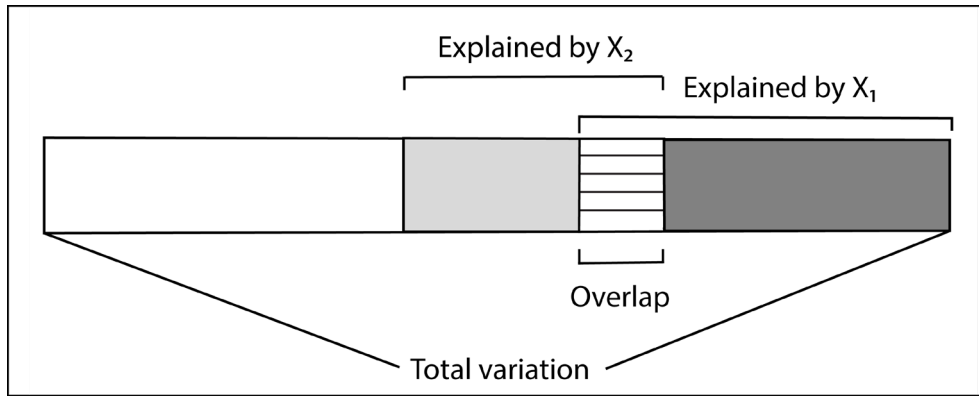


Figure 8. Conceptual diagram of regression as a partitioning of the total variation into portions that are explained by the predictor variables ( $X_1$  and  $X_2$ ), a portion that can be explained by both variables (overlap) and a portion that is left unexplained. (Clarke and Warwick 2001)

The predictor variables used for this analysis were 10 sediment characteristics listed in table 2. Grain size metrics were chosen in particular, because they were consistently associated with benthic invertebrate sampling stations. Defining biotopes using only sediment variables allowed for retention of the maximum number of stations examined with DistLM and thus classifying biotopes in the most robust way.

Table 2. Sediment grain size characteristics used to run distance based linear models.

Grain size metric	Grain size metric
% clay	Mode
% silt	Skewness
% sand	Sorting
% gravel	Kurtosis
% organic content (LOI)	Median
Mean	

Indicator species were determined for the most influential characteristics when possible by using LINKTREE. LINKTREE identifies thresholds in each of the variables (e.g., geoforms or grain size metrics) that correspond to occurrences of different benthic assemblages. The benthic assemblages corresponding to these thresholds were used to determine indicator species for the underlying variables.

An indicator species is defined as frequently associated with certain environmental conditions or characteristics (e.g., Geoform: basins and channels) while being not often associated with any other environmental condition or characteristic (e.g., any other Geoform). Indicator species were calculated according to (Dufrene and Legendre 1997):

$$IndVal_{ij} = A_{ij} * B_{ij}$$



Where  $A_{ij}$  is the proportion of the individuals of species  $i$  that are present in biotope  $j$  and  $B_{ij}$  is the proportion of stations in biotope  $j$  that contain species  $i$ .

The indicator species values range from 0 (poor indicator) to 1 (perfect indicator). PRIMER's RELATE function, based on a Pearson Correlation, was used to determine the significance level of the indicator species. Only indicator species with a  $p$ -value  $< 0.05\%$  were reported.

To develop a continuous map of biotope types for Wellfleet Harbor and surrounding areas, we interpolated the categorical biotope classifications as presence or absence at each station, per biotope. Biotopes were interpolated between each station using spline with barriers. The biotope interpolations were then merged by the *Mosaic to New Raster* function in ArcGIS using the maximum value to determine overlapping biotope. Although the interpolations introduce uncertainty between stations, the continuous map allows for future comparisons.

## Results

### Vessel-Based Acoustic Surveys

Vessel based acoustic surveys were conducted in Wellfleet Harbor and surrounding waters in 2015 (08 August – 12 December), and 2016 (12 July – 07 September). Areas mapped outside of the harbor include Duck Harbor and Billingsgate Shoal in Cape Cod Bay, Herring River, Loagy Bay, Silver Spring Harbor and Hatches Creek (Figure 9). Southern parts of Wellfleet Harbor were also included (Figure 1). With the exception of a few areas in Cape Cod Bay, the area mapped is a shallow water environment ( $< 10$  m), with a mean depth of 3.98 m across more than 57.15 km<sup>2</sup> (Table 3).

Differences in sidescan and bathymetric coverage of processed data occur because of how the data are collected. Sidescan coverage is a function of range from the port and starboard transducers, which is set by the surveyor. In order to acquire a 100 m swath of sidescan, the operator sets the port and starboard ranges to 50 m. Conversely, bathymetric data coverage is a function of water depth. For this study a 6:1 - 8:1 ratio of swath width to water depth was typical. For example, mapping in 3 m of water yielded an 18 - 24 m swath of the seafloor.

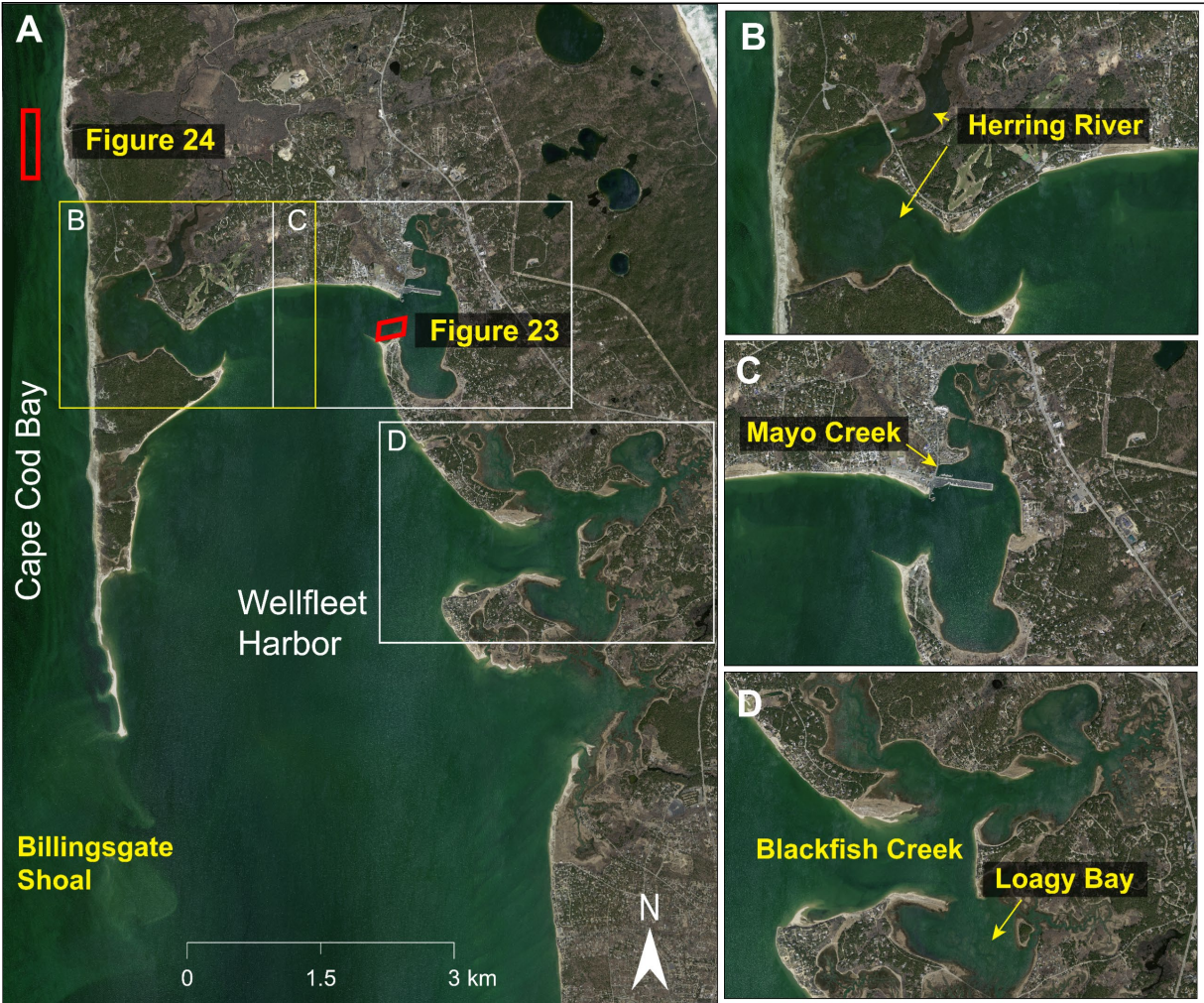


Figure 9. Wellfleet Harbor and surrounding embayments. A. The study area showing the eastern edge of Billingsgate Shoal (bottom right), and locations of eelgrass beds (Figure 24), and aquaculture (Figure 23). B. The mouth of Herring River. C. “Inner Harbor” as referred to by this study, and Mayo Creek. D: Blackfish Creek and Loagy Bay.

Table 3. Results of vessel-based acoustic surveys from two field seasons (2015-2016). Values for sidescan and bathymetry represent processed data used for the Benthic Habitat Maps and not the total amount of raw data collected.

Embayment	Survey days	Survey lines	Survey Line Length (km)	Area Mapped SSS (km <sup>2</sup> )	Area Mapped Bathy (km <sup>2</sup> )	Mean depth (m)	Max depth (m)
Wellfleet Harbor	43	1464	1533	57.15	35.32	3.98	12.09

The use of sidescan imagery in benthic habitat mapping is critical for describing material properties that cannot be determined from the ‘topography’ of the seafloor (as measured by bathymetric data), (McGonigle et al., 2014). Sidescan imagery is particularly useful for identifying submerged aquatic vegetation (SAV) such as eelgrass. For this study, sidescan imagery was used to identify and map a total of 19.3 hectares of eelgrass habitat at a minimum mapping unit of 100 m<sup>2</sup> (Figure 10). Although smaller patches of eelgrass may be present in areas within the harbor, the designation of eelgrass

habitat was not given if the mapped area was smaller than 100 m<sup>2</sup> (0.01 hectares, (ha)). The extent of eelgrass habitat mapped for this study was limited to the range of the sonar and the boundaries of the study area. The most extensive eelgrass beds were mapped outside of the harbor in Cape Cod Bay near Duck Harbor and on Billingsgate Shoal west of Jeremy Point. Eelgrass beds within these areas appear to extend beyond the mapped area but were not within the boundaries of the study area. Eelgrass habitat was not designated within Wellfleet Harbor (Figure 10).

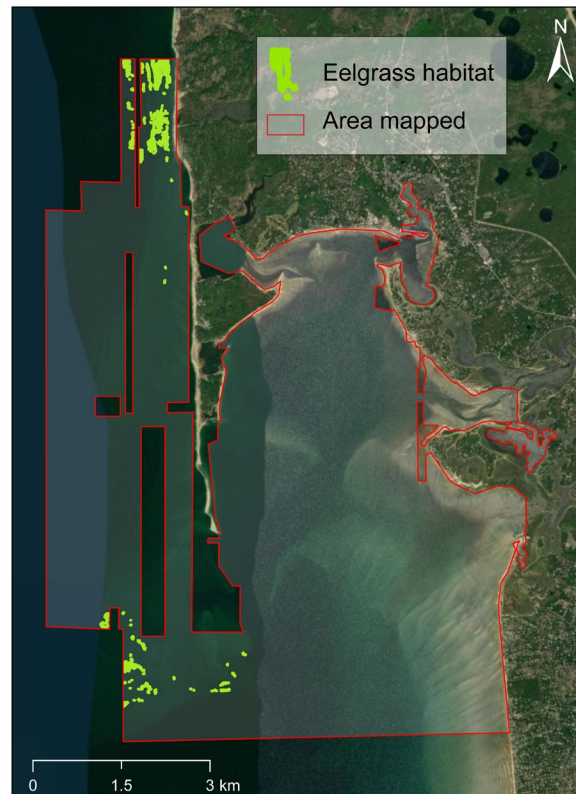


Figure 10. Eelgrass habitat mapped in 2015 (19.3 ha). Eelgrass habitat that extended beyond the range of the acoustic surveys that could not be identified in the sidescan imagery were not included.

Sidescan imagery was used to map aquaculture infrastructure within Wellfleet Harbor. Oyster cultch is composed of mostly sea clam and oyster shell that have been cleaned and placed in the harbor by the Town of Wellfleet as a substrate, or habitat in which to encourage the collection and propagation of wild oysters. Submerged infrastructure used by the commercial shellfish industry primarily to grow oysters are prevalent in the harbor. For example, a total of 12.25 ha of aquaculture infrastructure were mapped in the north, near the entrance to the inner harbor, and Blackfish Creek.

### Seismic Reflection Profiling

A total of 35 km of seismic reflection profiles were collected within the survey area (Figure 11). Overall, the coarse (sand or gravel) surface sediment, natural gas in the subsurface or the presence of dense beds of benthic aquatic vegetation limited penetration of the seismic signal in portions of the study area. The seven most common seismic facies are summarized below. Individual reflectors



representing depositional layers within the facies described below could be further identified and described with additional mapping and/or sediment coring.

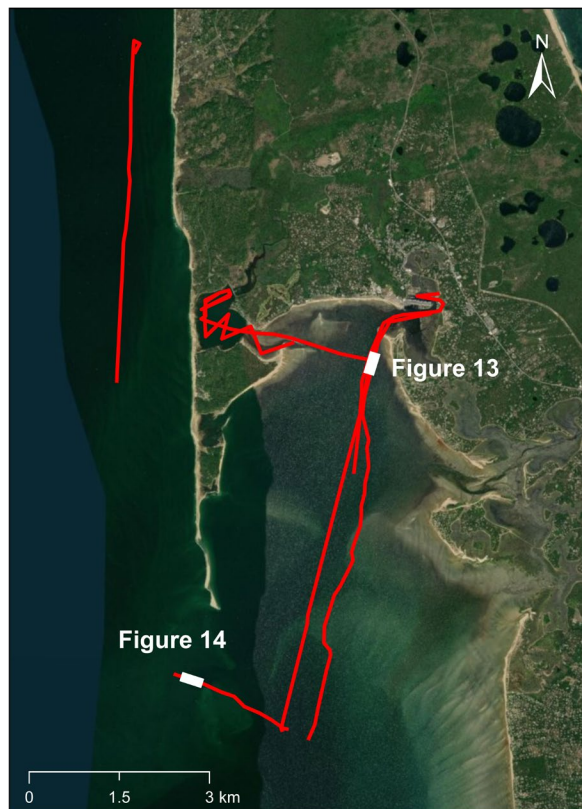


Figure 11. Locus map showing the extent of sub-bottom seismic reflection profiles collected for Wellfleet Harbor (red lines) and location of figures 12 and 13.

### ***Seismic Facies Identified***

#### **Facies GLF: Glacial Lakefloor**

Facies GLF is characterized by parallel, laterally continuous reflectors that drape underlying topography. This facies is interpreted to have been deposited in a glacial lakefloor depositional environment. While the sediment is composed of laminated silt and clay (interpreted as likely varve sequences), individual seismic reflectors represent groupings of sedimentary couplets rather than individual varves. This facies was identified only in limited ‘glimpses’ where the seismic penetration was sufficient.

#### **Facies Glu: Glacial deposits – undifferentiated**

Facies Glu is identified by a strong reflector, often with a hummocky, collapsed topography. This facies was identified at various depths ranging from deposits that crop out at the seafloor to the limit of seismic penetration. Seismic penetration in this facies is often limited, due to the sediment size (sand to boulders). This facies is interpreted to represent (often collapsed) glacial stratified deposits of the Harwich Plain, Nauset Heights, Wellfleet Plain and Eastham Plain deposits of Oldale and Barlow (1986).

### Facies E: Estuarine Channel

Facies E is identified by a basal reflector that truncates underlying units as an erosional unconformity with a concave, channel like morphology, often filled with parallel, laminated reflectors (Figure 12). This unit is interpreted to represent post-glacial fluvial, spring sapping or tidal channels modified or formed during Holocene marine transgression.

### Facies M: Marine mud

Facies M is ubiquitous throughout much of the harbor, occurring as an acoustically transparent layer that drapes the underlying units up to 10 m thick (Figure 12). This facies is interpreted to be estuarine mud deposited in low-energy basins. Occasionally, in low-energy basins, the lower half of the unit shows a slightly darker seismic return, either with a distinct seismic reflector or with a gradual increase in seismic intensity.

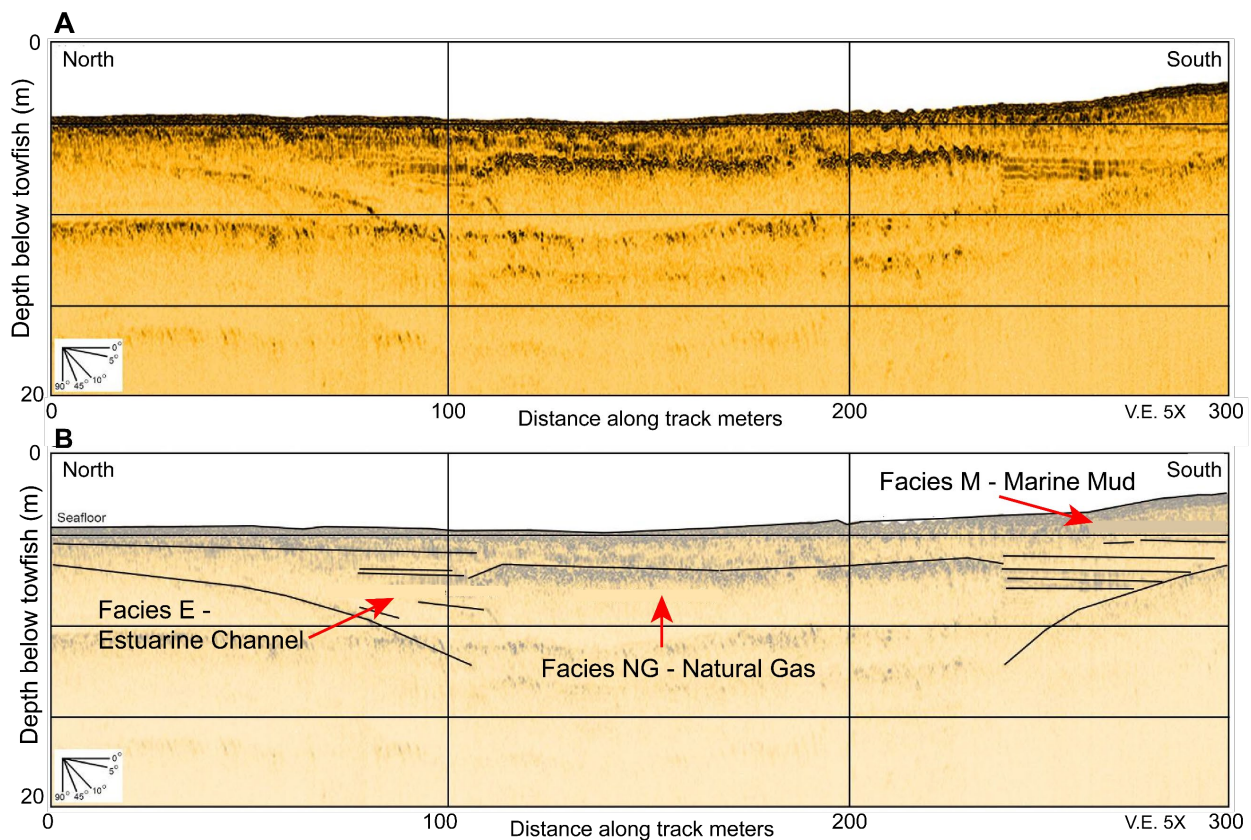


Figure 12. A. Sub-bottom seismic reflection profile from Wellfleet Harbor. B. Interpreted seismic reflection profile from Wellfleet Harbor showing facies E, M and NG. For profile locations, see fig. 11.

### Facies PR: Prograding Reflectors

Identified along the western end of Wellfleet Harbor adjacent to Billingsgate Island, this facies is characterized by dipping reflectors, oriented towards the east, although it is likely that these are viewed at an apparent dip rather than the true dip direction (Figure 13). The apparent dip angle (5-7°) bearing 115° correspond to a true dip of 14-15° bearing 180°. This facies is interpreted to represent prograding reflectors, formed as the barrier spit extended down the western side of Wellfleet Harbor.

While dipping reflectors have been interpreted elsewhere as forming on the delta slope of the North Truro deposits prograding into Glacial Lake Cape Cod (Borrelli et al., 2014), the apparent dip direction here (east), indicates deposition of these reflectors as a prograding barrier spit is more likely.

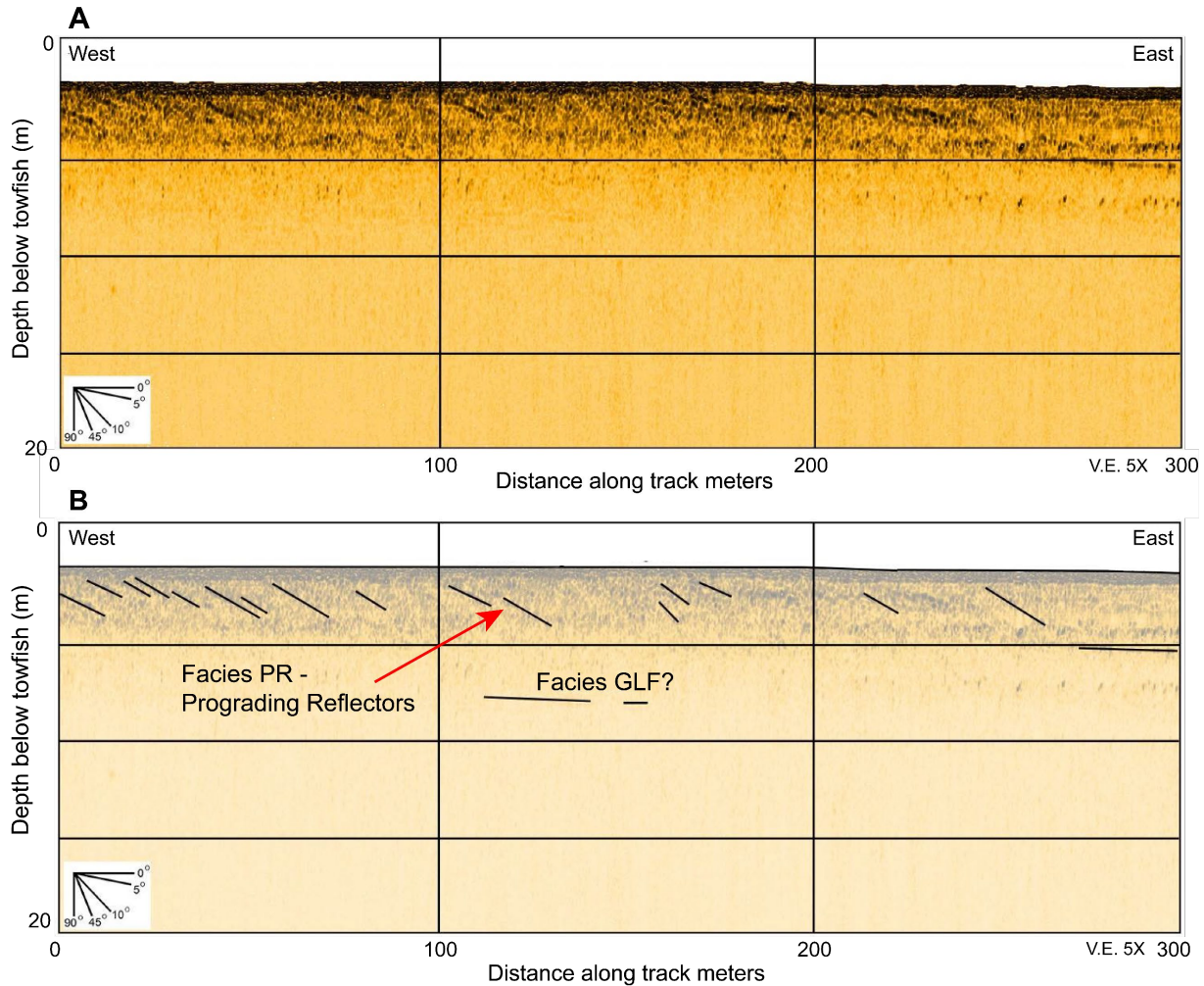


Figure 13. A. Sub-bottom seismic reflection profile from Wellfleet Harbor. B. Interpreted seismic reflection profile from Wellfleet Harbor showing facies PR and possibly facies GLF. For profile locations, see fig. 11.

### Benthic Sampling

Between August 12<sup>th</sup> and September 1<sup>st</sup>, 2015, 28 stations within Wellfleet Harbor and its surrounding areas were sampled in triplicate, resulting in a total of 84 sieved and preserved biological samples (Figure 14). In addition, sediment samples (n=28), conductivity and water temperature, and video data were collected at each station. (Table 4).



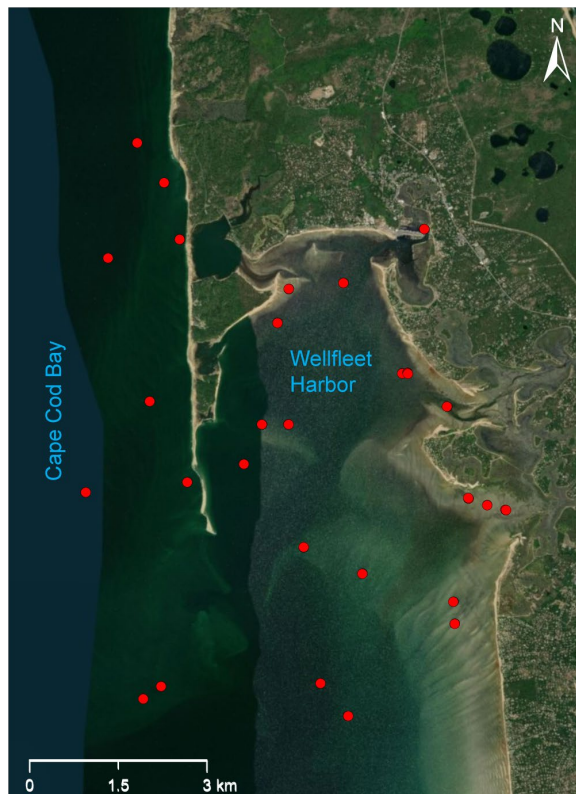


Figure 14. Locations of the 28 Benthic samplings stations in Wellfleet Harbor and vicinity.

Table 4. Benthic mapping instruments, data products, and number of sites sampled by the Center for Coastal Studies.

<b>Instrument</b>	<b>Data</b>	<b>Resolution</b>	<b>Number of sites</b>
Phase-measuring sidescan sonar	Bathymetry grid	1.0 meter	N/A
	Sidescan sonar mosaic	0.5 meter	N/A
CTD Profiler	Water column data	N/A	28
Young-modified Van Veen grab sampler	Sediment particle size and distribution metrics	N/A	28
	Sediment organic content	N/A	28
	Benthic infauna abundance	N/A	28

## Benthic habitat mapping

### *Physical characteristics*

#### CMECS Geoforms

CMECS Geoforms were analyzed using the Benthic Terrain Modeler (BTM) extension in ArcGIS10.3 from the bathymetric data collected during the acoustic survey. Slope, broad and fine scale BPI as well as the bathymetric data were utilized to classify the structures throughout Wellfleet Harbor. The BTM analysis and classification delineated eight distinct units that were classified as CMECS Geoforms (Figure 15).

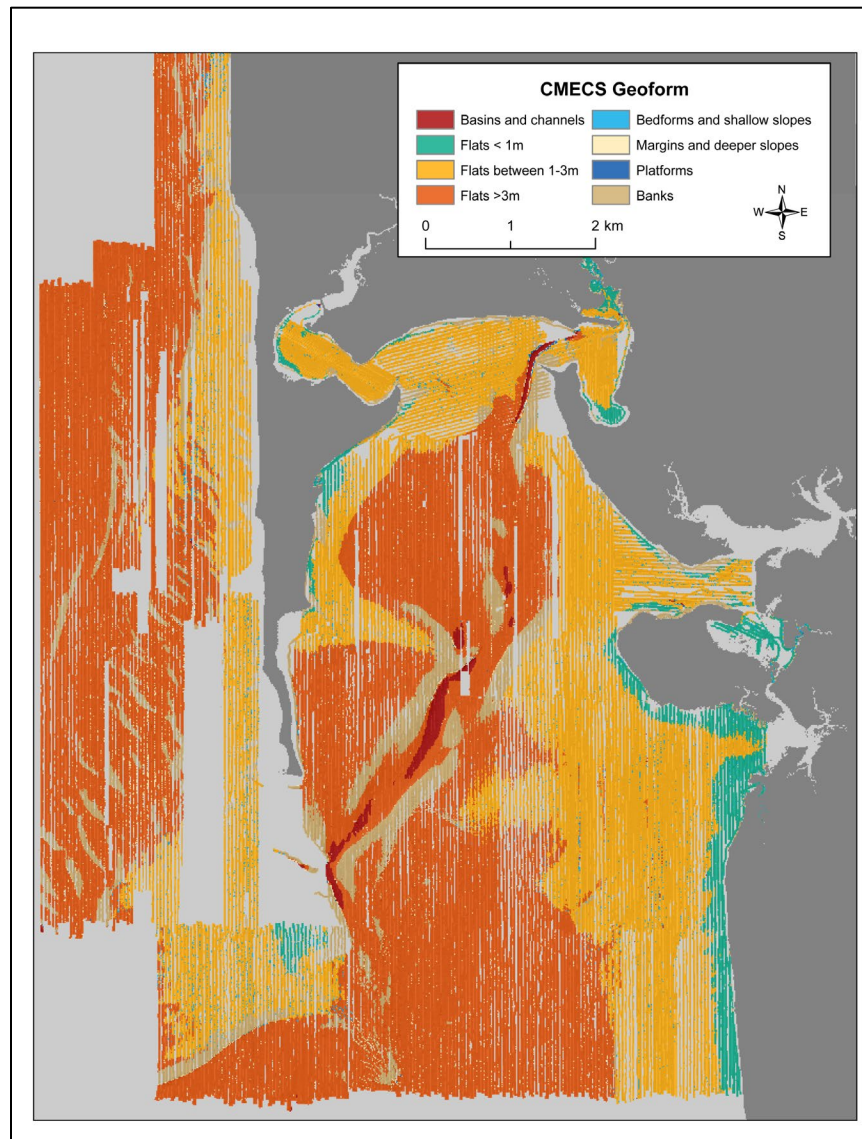


Figure 15. CMECS Geoforms for Wellfleet Harbor.



### CMECS Substrate

For the CMECS Substrate Group and Subgroup classification median grain size (U50) was interpolated between stations using the spline with barriers tool in ArcGIS10.3. Interpolated maps show the majority of sediment throughout the system to be coarse sand (500-1000  $\mu\text{m}$ ) or very coarse sand (1000-2000  $\mu\text{m}$ ) while three stations were found to be coarse silt (32-62  $\mu\text{m}$ ) (Figure 16, Appendix D, Table D-1).

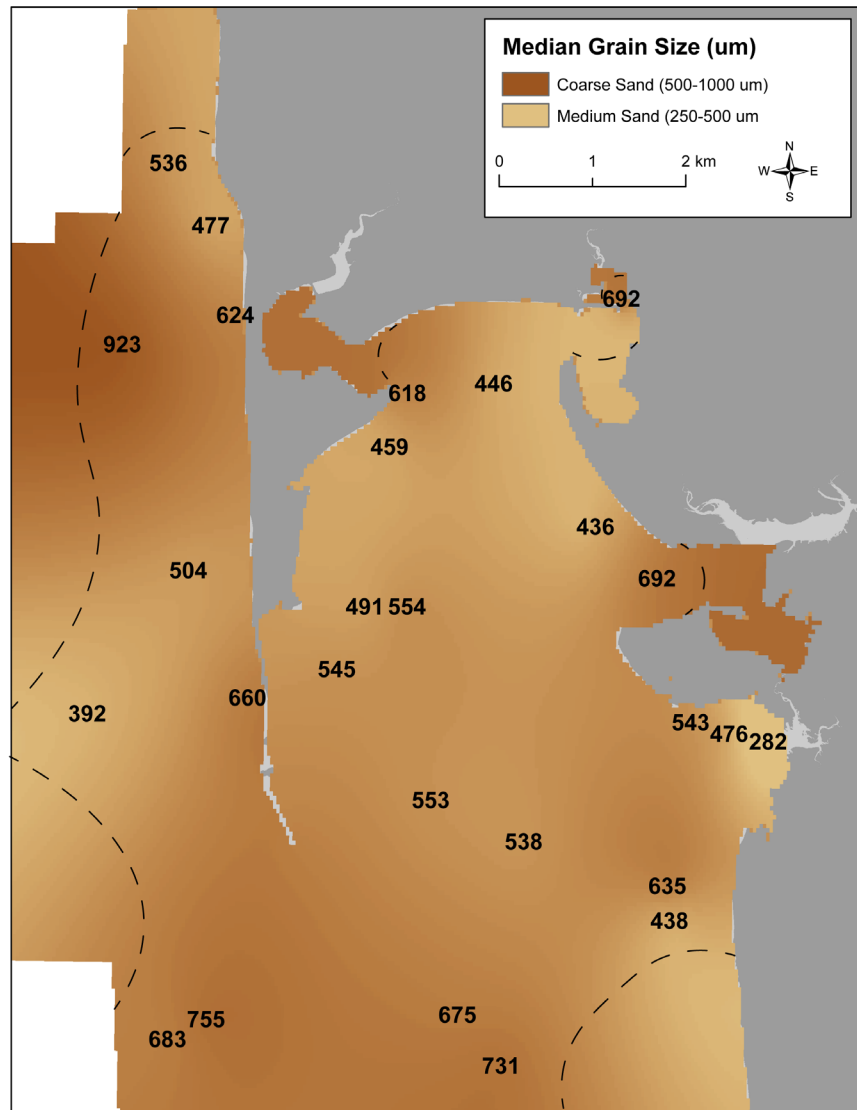


Figure 16. Median grain size in microns (interpolated) for Wellfleet Harbor. Data points are labeled by median grain size. Dashed lines indicate areas where interpolation is beyond observed data points.

### ***Biological characteristics***

#### CMECS Biotic Component

Out of 98 species and 9838 individuals (Appendix C) in Wellfleet Harbor and its surrounding areas, 31 invertebrate species comprised 95% of all individuals. We found one individual of *Polycirrus*

*eximius* (a polychaet worm) that is considered a cryptogenic species in Massachusetts (Figure 19). Cryptogenic means the origin of the species are unknown. In ecology, a cryptogenic species may be either a native species or an introduced species, clear evidence for either origin being absent. PRIMER's SIMPORD and Cluster analysis indicated that the optimal number of clusters is 6 (Figure 17). Classifying each significant cluster into CMECS Biotic Communities based on species dominance yielded 4 Biotic Communities (Table 5).

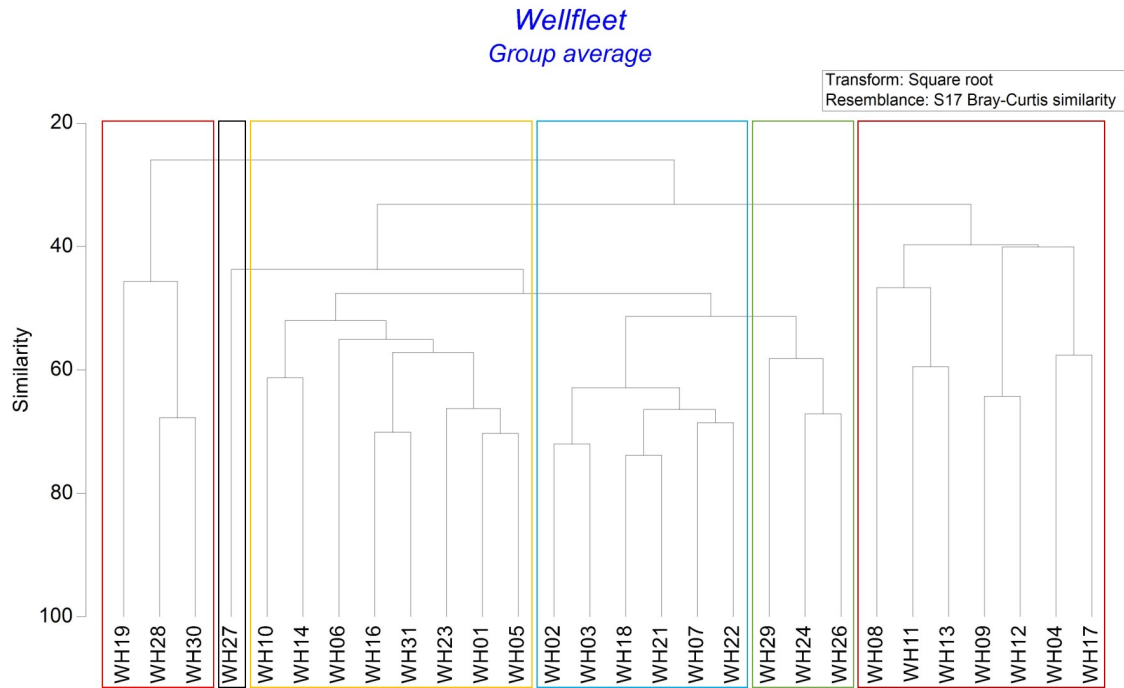


Figure 17. PRIMER's cluster analysis based on species composition at each station. Colored boxes indicate clusters

Table 5. Calculated Clusters for Wellfleet Harbor with most abundant species and biotic component classification according to CMECS.

Wellfleet Cluster	Dominant species	CMECS Biotic Community	CMECS Biotic Group	CMECS Biotic Subclass
Cluster 1	<i>Spisula solidissima</i>	Spisula bed	Clam bed	Soft sediment fauna
Cluster 2	Nephtyidae	Nephtys bed	Larger deep burrowing fauna	Soft sediment fauna
Cluster 3	Nephtyidae	Nephtys bed	Larger deep burrowing fauna	Soft sediment fauna
Cluster 4	<i>Crepidula fornicata</i>	Attached Crepidula	Sessile gastropods	Attached fauna
Cluster 5	<i>Crepidula fornicata</i>	Attached Crepidula	Sessile gastropods	Attached fauna
Cluster 6	Glyceridae/Goniadidae	Glycera bed	Larger deep burrowing fauna	Soft sediment fauna

**Preliminary biotopes**

Results of PRIMER’s DistLM show that a total 55.9% of the species distribution in Wellfleet Harbor can be explained by factors collected for other CMECS components with Geoforms (21.4 %), % gravel (9.6%) and skewness (6.9%) explaining the majority. Skewness measures the degree to which a cumulative curve approaches symmetry. Symmetrical curves have a skewness equal to 0; those with a larger proportion of fine material are positively skewed and those with a large proportion of coarse material are negatively skewed. Temperature was another very influential factor in determining preliminary biotopes.

LINKTREE showed one split for Geoform separating stations into “flats <1m” and other geoforms, thus creating the first biotope. Five splits for skewness created another 3 biotopes and splits for % gravel and a further separation of geoforms created the remaining 5 biotopes (Table 5 and Figure 18).

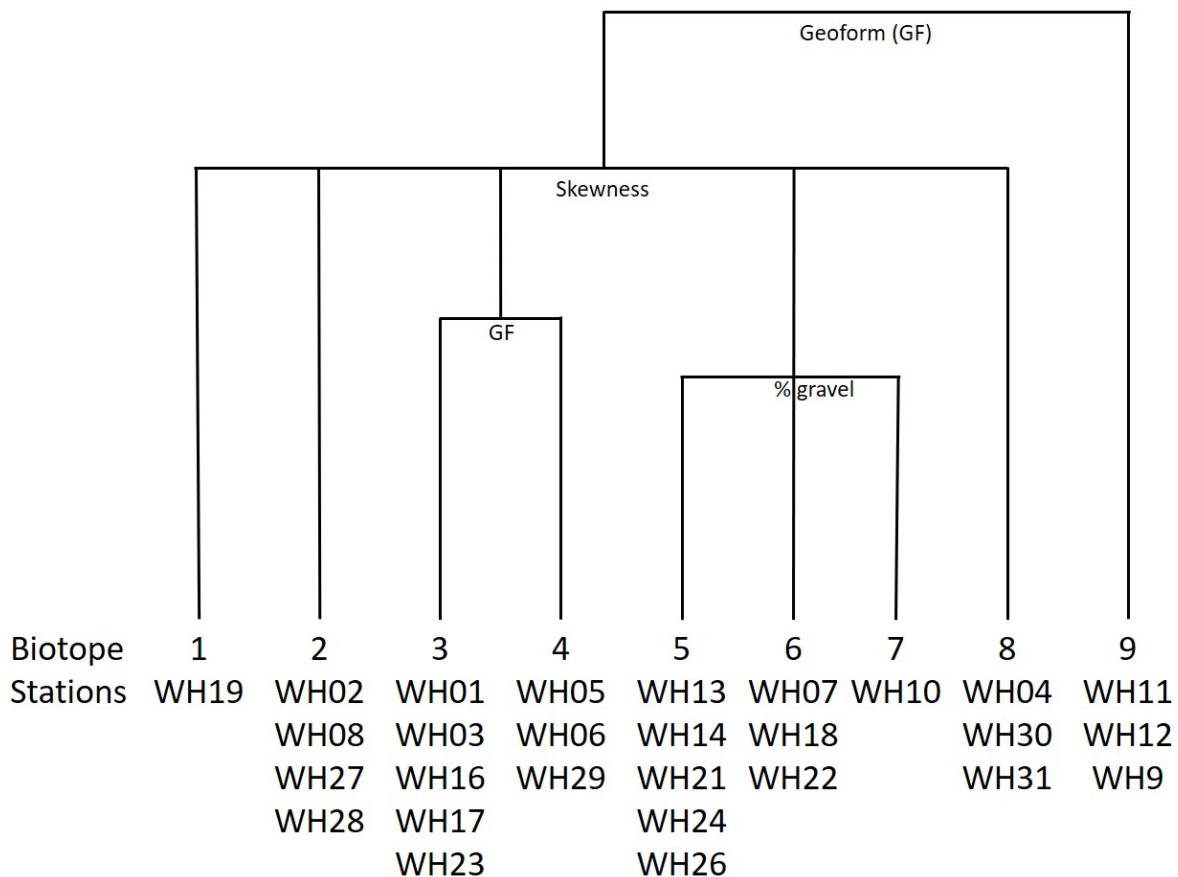


Figure 18. Cluster diagram showing 6 optimal biotopes based on geoform, sediment characteristics and the abundance of species that accounted for 95% of the total abundance in Wellfleet Harbor.



Figure 19. Picture of *Polycirrus exemius*, a polychate or bristle worm found in Wellfleet Harbor is a type of Spaghetti mouth worm. This species of worm can grow up to 1 inch in length (not including the tentacles) and is considered a cryptogenic species in Massachusetts.

Table 6. Calculated indicator species for each biotope with indicator species value (IndVal) form 0 (bad indicator) to 1 (good indicator). Only indicator species with a p-value < 0.05 are reported here.

<b>Biotope</b>	<b>Description</b>	<b>Indicator Species</b>	<b>IndVal</b>
1	Other Geoforms with very high proportions of coarse material (99% gravel and sand)	<i>Spisula solidissima</i>	0.77
4	Flats >3m	<i>Ameritella agilis</i>	0.39
5	Other geoforms with 0 – 3.2% gravel	<i>Crepidula fornicata</i>	0.29
6	Other geoforms with 3.3 - 10% gravel	<i>Crepidula fornicata</i>	0.29
7	Other geoforms with symmetrical grain size distribution and 100% sand	Glyceridae	0.41

Indicator species were found to be significant (significance <0.05%) for biotopes 1, 4, 5, 6 and 7 (Table 6). Although Nephtyidae, *C. fornicata*, *M. mercenarian* and *T. obsoleta* were present in biotopes, 2, 3,

8 and 9 respectively, no significant correlation between species and biotope could be determined. *Spisula solidissima* (Figure 20B), the Atlantic surf clam was indicative (IndVal: 0.77) of biotope 1 (other geoforms with very high proportions of coarse material; 99% gravel and sand), *Ameritella agilis*, the Northern dwarf-tellin indicated biotope 4 (Figure 20C) (IndVal: 0.39; Flats >3m), *Crepidula fornicata*, the common slipper shell or slipper limpet, was indicative of biotopes 5 and 6 (Figure 20A) (IndVal: 0.29 at both). Biotope characteristics of biotopes 5 and 6 were very similar and when combined yielded a higher indicator value and correlation between biotope and indicator species (new IndVal: 0.35). Glyceridae, the polychaeta family of blood worms, were indicative of biotope 7 (Figure 20D) (IndVal: 0.41; other geoforms with symmetrical grain size distribution and 100% sand).

Categorical biotopes were interpolated between using the spline with barriers tool in ArcGIS 10.3. Interpolated maps display a reproducible representation of the biotic communities throughout Wellfleet Harbor and its surrounding areas (Figure 21).

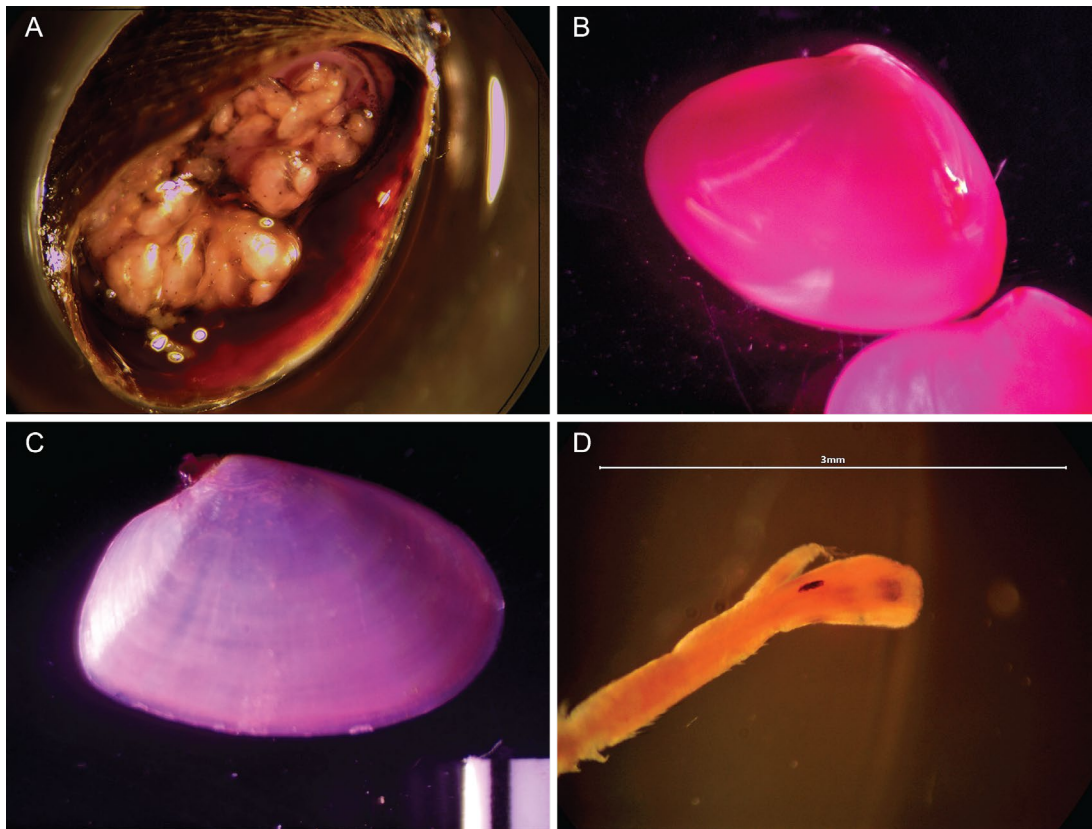


Figure 20. Indicator species of each biotope. A. Limpet or slipper shell (*C. fornicata*, photographed from below, 40mm), was indicative of biotopes 5 and 6. B. Atlantic surf clam (*S. solidissima*, 4mm) was indicative of biotope 1. C. Northern dwarf-tellin (*A. agilis*, 7mm) indicates biotope 4. D. Bloodworm (belonging to the polychaet family Glyceridae) indicated biotope 7.



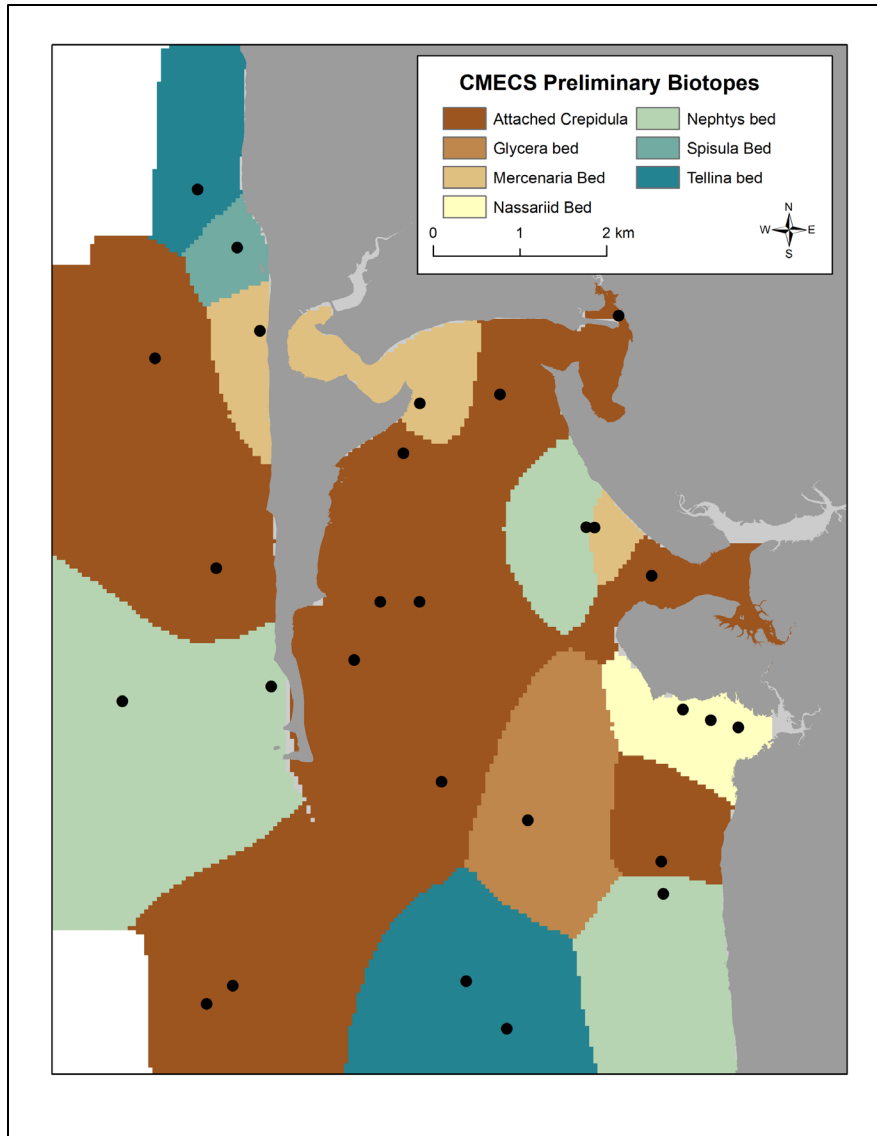


Figure 21. Interpolated map of CMECS biotopes in Wellfleet Harbor based on PRIMER's LINKTREE and distance based linear models. Black dots represent station locations.

## Discussion

### Vessel-based Acoustic Surveys

Acoustic surveys are essential for the planning and implementation of benthic habitat mapping. Sidescan imagery is used to identify characteristics such as the presence of submerged aquatic vegetation (e.g., eelgrass, macro algae), geomorphology of the seafloor, changes in the substrate such as grainsize, and the presence of anthropogenic impacts to the environment such as submerged structures (e.g., aquaculture, marine debris) and seafloor disturbance from activities such as recreational boating, channel dredging for navigation, or certain types of fishing. Bathymetry data

provides information about the depth, slope and rugosity (roughness) of the seafloor, and the magnitude and dimensions of submerged structures – all of which are factors that could influence benthic invertebrate species diversity and abundance. Analyses of these data are not limited to the CMECS models, as they provide a baseline for future studies as well as useful ancillary data that tangentially relate to the ecosystem state.

An example of using sidescan imagery and co-located bathymetry for the identification and analysis of seafloor characteristics is shown in Figure 22. Bathymetry data are typically displayed using a color range in which ‘cool’ colors represent deep water (relative to the range of depths in a given data set), and ‘warm’ colors represent shallow depths. In this figure, the bathymetry shows an abrupt change in elevation (black arrow). Co-located sidescan mosaic (right panel) shows a sharp change in the substrate (white arrow) coinciding with a change in elevation shown in the bathymetry. In this image, the darker color represents finer grained sediment (e.g., silt, fine sand), that tends to absorb acoustic energy, as opposed to sandy tidal flats that reflect acoustic energy back to the sonar sensors. This change in substrate is indicative of an abrupt transition from a high energy environment to a lower energy environment.

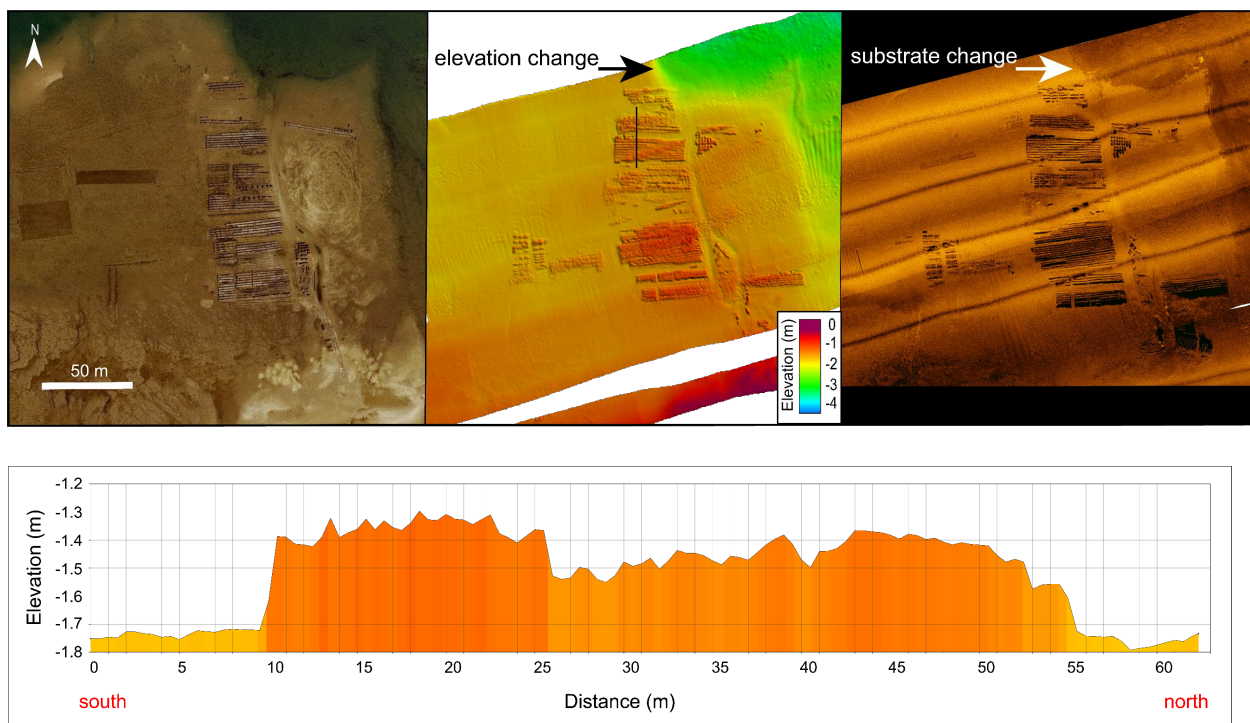


Figure 22. Oyster cultivation grants in Wellfleet Harbor. Left: 2014 aerial imagery. Center: 2016 Bathymetric image. Colors represent the elevation from the seafloor (depth) in NAVD 88 m. Right: 2016 co-located sidescan mosaic. Bottom: Profile of cultivation racks drawn from the bathymetry, the black line in the center panel shows location of the profile. See Figure 9 for location.

### **Identification of Eelgrass Habitat**

Sidescan imagery is ideal for the identification of eelgrass habitat. Detection is typically straightforward - eelgrass beds can be identified and targeted from the sidescan data in real-time

while conducting surveys. Sidescan imagery can later be used to compute metrics such as spatial heterogeneity (patchiness) and percent coverage. The co-location of bathymetric data and sidescan imagery allows for more rigorous analysis with the addition of position and elevation data derived from the bathymetry (Figure 23). While tidal currents can reduce the apparent height of the eelgrass, analysis of the co-located bathymetric and sidescan data can still provide a first-order approximation of above ground biomass (e.g., volume). Using bathymetric data without sidescan imagery makes identifying eelgrass difficult without other corroborative evidence such as underwater video, imagery, etc.

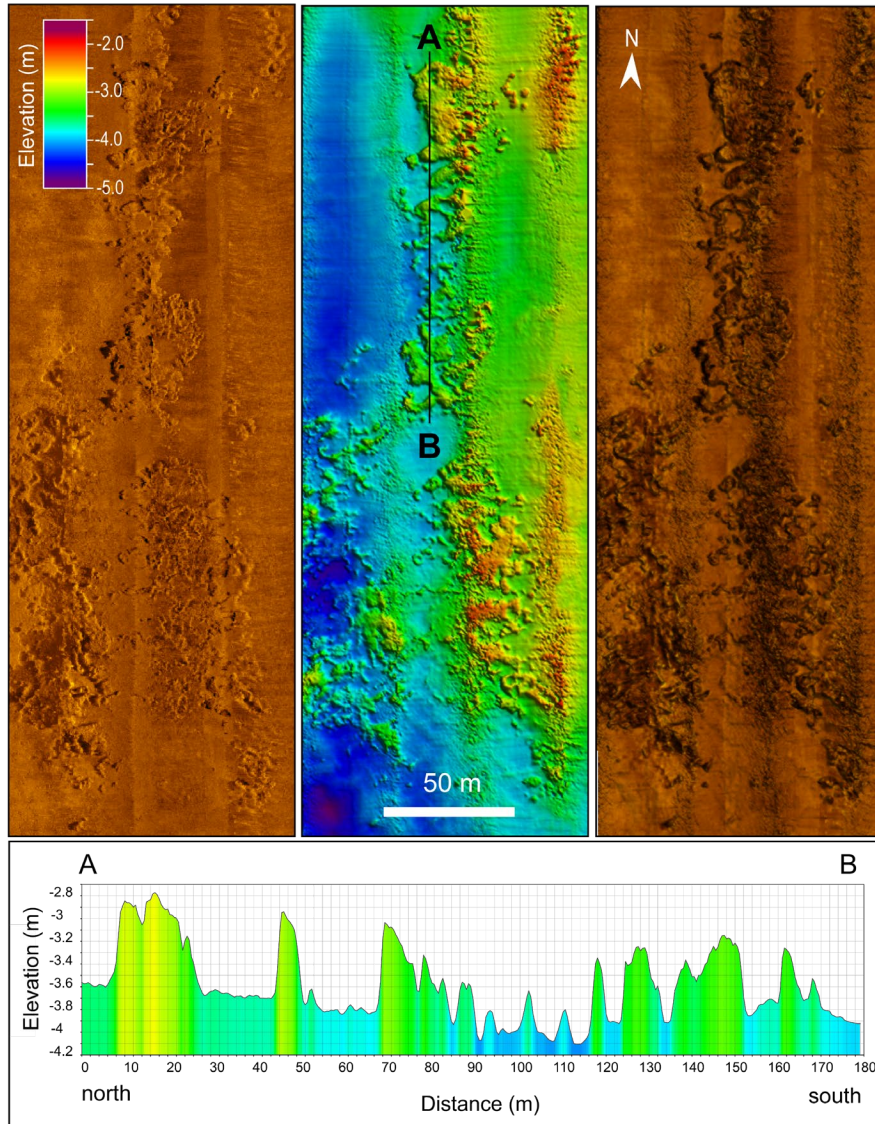


Figure 23. Eelgrass habitat near Duck Harbor. Left: Sidescan sonar mosaic. Center: Swath bathymetry over the same area. Right: Co-located sidescan mosaic draped over bathymetric data. Bottom: Profile of eelgrass beds generated from the bathymetry data. Color bar (upper left) represents depth based on NAVD 88 m vertical datum. See figure 9 for location.



### **Mapping Anthropogenic Features**

Although not always utilized in Benthic Habitat mapping, a category for anthropogenic Geoform features exists within the CMECS modeling framework. Mapping anthropogenic features are potentially useful tools for managers as they provide spatially explicit digital models of habitat evolution. For example, acoustic maps that include cultch habitat in Wellfleet Harbor provide useful information for the Town's oyster cultch program, and oyster shell recycling programs (Figure 24).

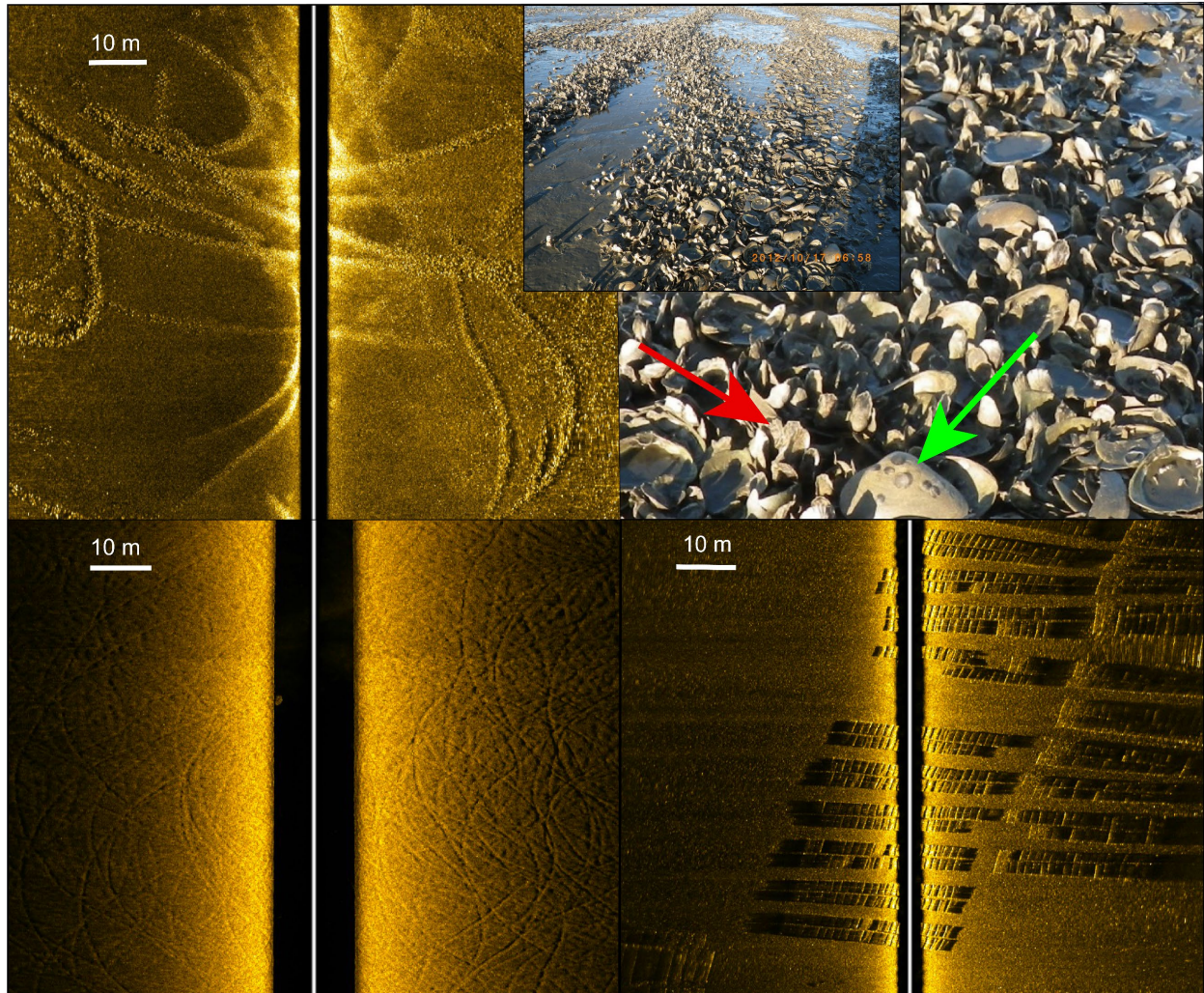


Figure 24. Examples of anthropogenic geoform features. Top left: Side scan imagery showing cultch habitat deposited in Wellfleet Harbor. Top right: Cultch habitat at low tide (inset); red arrow – oysters growing in the cultch, green arrow – oyster spat growing on sea clam shells (far right). Bottom left: Dredge tracks from shellfish harvesting. Bottom right: Aquaculture farming in Wellfleet Harbor. Photographs were taken in 2012 by CCS for the Town of Wellfleet Oyster Propagation Program. Sidescan was mapped for this project in 2015 and 2016.



Wellfleet Harbor has many human-altered stretches of intertidal and sub-tidal seafloor. Sidescan sonar can capture images of these areas and allow managers to better understand the state of the alterations as well as ongoing natural processes. In Figure 25, the state of the aquaculture equipment in the center of the image is clear. Missing or damaged equipment could be determined by an investigation of these images, which is very valuable for sub-tidal grants. These data also allow for managers and other scientists to assess the state of the seafloor for future studies that are related to, or independent from this mapping project.

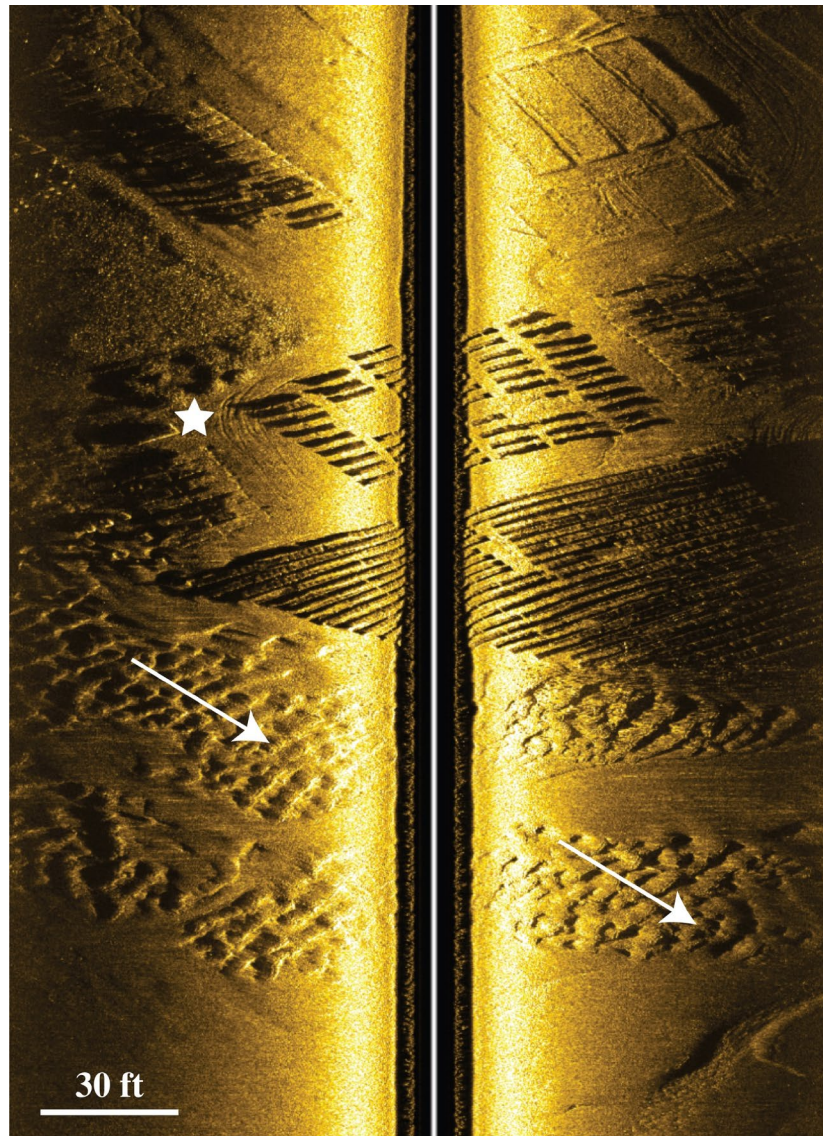


Figure 25. Raw Sidescan imagery of seafloor between Indian Neck and the entrance to Loagy Bay. Black strip in the middle will be removed during processing. Arrows indicate direction of sediment transport and star denotes areas where tire tracks are present to demonstrate resolution of data.

These data sets are snapshots in time in a very dynamic system, some say that ‘the maps are obsolete before the boat gets back to the dock’. This may be true, however, the state of the seafloor for this particular time period has been captured and recorded for future use. These data can serve as a

baseline inventory against which change can be measured, which is critically important given the speed with which change is occurring in these systems from natural and human-induced causes.

### ***Acoustic Survey Planning***

Operating boats in the nearshore is inherently hazardous to personnel and equipment. The design of the survey platform and type of acoustic instruments is critical for reducing the exposure to these hazards. In 2016 the R/V Portnoy was used exclusively in Wellfleet Harbor to map areas that were too shallow to map with the R/V Marindin. The shallow draft and exceptional visibility of navigation hazards onboard the R/V Portnoy greatly improved survey efficiency and data quality in very shallow waters. This is demonstrated by comparing Pleasant Bay which was mapped using the R/V Marindin for the larger NPS project in 2014, and Wellfleet Harbor which was mapped in 2015-2016 using both the Marindin and the Portnoy. In Pleasant Bay, shoals less than 1 meter in depth (NAVD 88), were not mapped, whereas in Wellfleet, the Portnoy was able to map most shoals less than 1 m (Figure 26).

Another challenge in survey planning when working in tidally restricted embayments is the short survey window. Surveys should be done in daylight hours and often can only be efficiently accessed 1-2 hours before and after high tide. Careful survey planning can optimize these times and deeper areas can be mapped outside of this window, but this adds additional survey days, and mobilization and de-mobilization costs. The R/V Portnoy's shallow draft platform, custom designed for optimal maneuverability and ease of deployment, not only increased the area mapped, but also extended the length of the survey days in these tidally restricted areas. However, one caveat is that the Portnoy is not seaworthy in wave dominated areas outside of the harbor, such as Billingsgate shoal near Jeremy Point, which were too shallow to map using the Marindin.

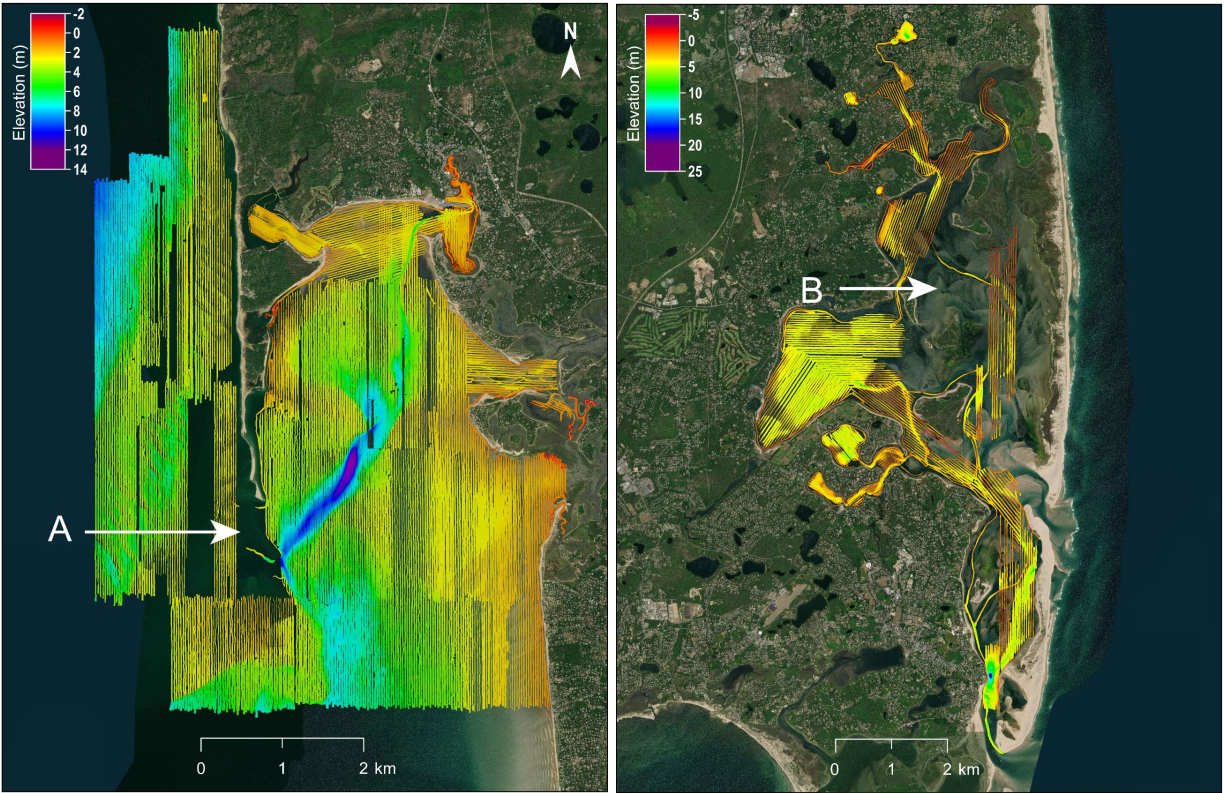


Figure 26. Bathymetric maps of Wellfleet Harbor (left) and Pleasant Bay (right). Colors represent the elevation from the seafloor in NAVD 88 m. Orange and red represent shoals less than 1 m. A. Billingsgate Shoal south of Jeremy Point could not be safely mapped by the R/V Marindin or the R/V Portnoy. B. Shoals less than 1 m that could not be mapped using the R/V Marindin.

When surveying with a phase-measuring sidescan sonar a choice must be made to prioritize the collection of bathymetric data or backscatter imagery. If, for example, the survey planner intends to collect sidescan sonar data at 200% overlap with a 50 m range setting (100 m swath), lines would be spaced at approximately 40 m, accounting for vessel drifting, etc. However, in 3 m of water a bathymetric swath of approximately 18-24 m could be expected to leave a 16-22 m swath of seafloor with no bathymetric data. If 100% bathymetric coverage (Figure 27) was sought at the same 3 m water depth survey lines would need to be spaced at approximately 20 m apart. This would yield an unnecessary degree of backscatter imagery overlap of 500%, if set at 50 m range.



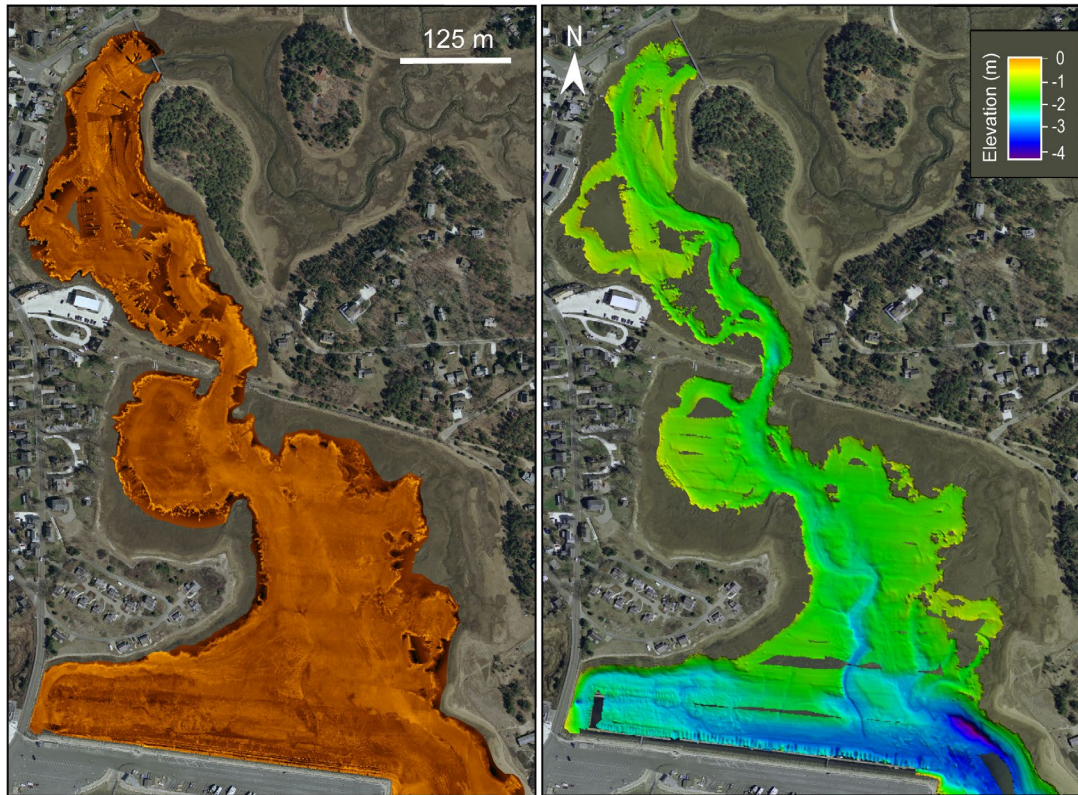


Figure 27. Inner Harbor north of the pier. Left: Sidescan mosaic from data collected in 2016. Right: Co-located bathymetric data. This area was mapped using 20 m line spacing in order to achieve 100% bathymetry.

Additional problems for the hydrographer working in small coastal embayments are the quick turns required at the ends of tightly spaced survey transects and in generally navigating these areas. The performance of some science grade motion sensors or gyrocompasses that measure the heave, pitch and roll of the vessel is greatly improved if the vessel travels in a roughly straight line for 30-45 seconds in order to re-calibrate or ‘settle’ after turning before data are recorded. A routine maneuver in the open ocean becomes difficult if not impossible in small coastal embayments.

### **Sub-bottom Seismic Reflection Profiling** ***Gaseous Sediment***

A distinct seismic facies (Facies NG) referred to as a ‘gas wipeout’, is produced by the scattering of the seismic signal by gas bubbles within the sediment. This gas is usually methane in estuarine and lagoon sediment and is common in the subsurface of other estuaries and coastal lagoons (Schubel 1974, Claypool and Kvenvolden 1983, Ussler et al., 2003). The ‘wipeout’ produced by the gas does not allow the thickness of the underlying reflectors below the gas to be measured. Gas was not widespread in Wellfleet Harbor; it was limited to small pockets identified in incised channels (facies E, Figure 12) and small pockets of gas within facies M. Areas of Wellfleet with shellfish beds or

dense benthic aquatic vegetation limited penetration of the seismic signal in other areas likely to contain natural gas.

Gas was found typically 0.5 to 1.0 m below the seafloor, suggesting it was being produced in situ by the decomposition of organic material within the modern marine sediment. Gas can occur deeper and can be produced as buried marsh sediment (peat) or older marine or lacustrine deposits decay. These deeper areas of gas were identified in a large estuarine channel in Wellfleet Harbor (Figure 12) and in other areas where facies E occurred. These incised, filled channel deposits, and would make excellent candidates for future coring studies. Buried marsh likely occurs in other parts of the study areas, however in many likely areas (tidal creeks and channels) either seismic penetration was limited, or data was not collected.

While methane can be released to the atmosphere from the sediment, no evidence of pockmarks (Kelley et al., 1994, Rogers et al., 2006) or gas seeps were observed on either side-scan sonar or seismic reflection profiles. This suggests that while methane is being produced in situ, it is not actively being released. Disturbance of this sediment (i.e., dredging) could release some methane to the atmosphere, although the actual volume of gas in this sediment cannot be determined from the seismic profiles.

#### ***Thickness of surface habitats***

The thickness of the surface sediment (usually Facies M) was calculated by subtracting the elevation of the basal reflector of these deposits from the elevation of the seafloor. Interpretation of sediment thickness requires sufficient seismic penetration to laterally trace a seismic reflector marking the base of the marine sediment, so thickness measurements are limited to areas where the seismic signal could penetrate consistently. The velocity of the soundwaves was assumed to be 1500 m/s in all calculations. Measurements of the thickness of marine sediment were limited to the center of the harbor and portions of Lower Herring River. The absolute thickness ranged from 0 to 8 m, with an average thickness of 2.5 m.

#### **Benthic Habitat Mapping**

CMECS Geoform component, reliant on the acoustic survey, identified eight CMECS Geoforms in Wellfleet Harbor (Figure 15): Basins and Channels, Platforms, Banks, Flats < 1m, Flats 1-3m Flats > 3m, Bedforms and shallow slopes >5° and margins and deeper slopes > 5°. The random sampling design used for benthic invertebrate surveys did not encompass all geoforms due to the limited number (and size) identified by the benthic terrain modeler. Further discussion of scale and geoform classification is required for CMECS to fully incorporate this as a meaningful mapping component across all systems.

CMECS Substrate components (Figure 16), particularly median grain size, shows a uniform area of medium and coarse sand in Wellfleet Harbor. The steady wave action and tidal currents in and around Wellfleet Harbor create a largely homogeneous substrate. Locations in low energy environments (e.g., Mayo Creek, Herring River) were not sampled.

The cluster analysis of benthic invertebrate (98 species and 9838 individuals) abundances generated 6 clusters, two of which had the same dominant species. CMECS geoform and substrate components explained 55.9% of species distribution in Wellfleet Harbor. The top three explanatory variables were geoform, % gravel and skewness (Figure 18). Indicator species for Wellfleet Harbor (Table 6) overlap with CMECS Biotic Communities but are not necessarily conspicuous members of the CMECS biotic components catalogue.

CMECS geoform component explained 21% of species distribution, indicating that the geologic nature of Wellfleet Harbor and, to a lesser extent, its sediment characteristics, are influential characteristic in determining diversity and abundance in and around Wellfleet Harbor. CMECS water column components, particularly temperature, also contributed to the explanation of species distribution in Wellfleet Harbor. We already know that water characteristics greatly influence benthic habitat quality (Howes et al., 2006) and factors such as dissolved oxygen likely play a role in driving the composition of benthic communities. However, temperature depends on factors such as weather, tide and time of year. Sampling in Wellfleet Harbor spanned several weeks during which temperatures likely changed. Thus, using temperature to explain species distribution, would result in seasonal biotopes, which are unstable and difficult to recreate. More research, across multiple seasons and years, needs to be conducted in order to establish a sound water column component in general and a reliable temperature baseline in particular.

Four significant indicator species could be determined (*Gemma gemma*, *Odontosyllis fulgurans*, *Molgula manhattensis* and *Chironomidae* larvae), which were also the most dominant species in five of the six clusters in the benthic community cluster analysis, suggesting that they play an important role in the overall composition of benthic communities in Wellfleet Harbor.

### **Data Analysis and Mapping Approach**

The approaches used for data analysis and classification for this study were chosen based on previous work in similar environments (Shumchenia and King, 2010) with the broad goal to delineate ecologically meaningful map units rapidly and reproducibly, and create maps using CMECS as a common language. The choice of analysis and classification approach was adapted to what the desired map products were. The raw data collected, analyzed, and classified in this project can be used to address multiple questions and provides a baseline for future ecological monitoring.

### **Mapping the CMECS Geoform Component**

The non-bathymetric physical variables within CMECS Geoforms (e.g., percent sand, percent gravel and percent organic content) show us that Geoforms are not compositionally distinct, instead they often overlap and could not be separated by grain size metrics. We did not calculate indicator species for CMECS Geoforms as the randomly determined sampling stations fell into only 4 of the described 8 Geoform categories, thus yielding non-significant results when analyzing for indicator species.

### **Mapping the CMECS Substrate Component**

The substrate classification based on weight percentages of gravel, sand, silt, and clay was sometimes different than the classified interpolation of median grain size. Neither classification is wrong, but the differences in these two approaches demonstrate the complexity in decision-making required for

benthic habitat mapping. The full coverage median grain size map (Figure 16) is built on the assumption that sediments conform to gradients in the study areas (i.e., there are no stark boundaries in sediment type). However, sources of error in the interpolation process, and sampling spacing that does not match the scale of environmental heterogeneity should both be considered as they influence the accuracy of the resulting substrate maps. In spite of these drawbacks, we opted to interpolate sediment characteristics instead of hand-drawing boundaries from aerial photography, bathymetry, and/or sidescan backscatter. Although those methods have been traditionally used by experienced coastal geologists, we assumed that the results would not be repeatable, nor would the knowledge and experience required to interpret such data be easily transferred to new staff, students, or other analysts.

### **Mapping the CMECS Biotic Component**

For this study, dominance was used as the metric to define Biotic Communities, and Indicator Species Value (IndVal: a measure of the specificity and fidelity of a species) as the metric to define Biotopes. Using dominance is recommended in the CMECS Technical Guidance Document (FGDC 2012), but can cause classification confusion when a single species may be dominant in several statistically distinct assemblages, as was the case in Wellfleet Harbor (*Crepidula fornicata* and Nephthyidae, Table B-1). One option to address this problem is to use a secondary- and/or tertiary-dominant species to define a CMECS Co-Occurring element. Another option is to use a different metric to describe Biotic Component units altogether however, this is not recommended in the CMECS Technical Guidance document.

This study used the Indicator Species Value for species in each calculated biotope because it is a rapid and reproducible way to add biological information to a map based on sediment variables. The Indicator Species Value determines which species are both abundant in a certain biotope *and* rarely found in other biotopes (i.e., have high biotope fidelity). A permutation of the data provides a measure of significance. The association between a biotope and an indicator species can be used to predict species presence, given the presence of the biotope, or vice versa. In this way, indicator species can be valuable in repetitive mapping or habitat monitoring. Crucially, indicator species may not be dominants, but if they are identified in subsequent surveys, their presence may be used to infer a particular biotope type. It is important to note that indicator species only meet the statistical criteria described above and do not necessarily have a unique ecological role, particular susceptibility to stressors, or other special characteristics. However, any of the previous statements could be true for any of the indicator species identified in this study, but those associations were not explored, tested, or verified within this study.

To determine biotopes in Wellfleet Harbor sediment variables, geoforms and infaunal abundance were examined as part of the biotope analysis. A total of 55.9% of species distribution could be explained by PRIMER's distance based linear model (DistLM). The three most important drivers in terms of species distribution, explaining a total of 37.9%, were geoform, % gravel and skewness with the remaining seven sediment characteristics explaining the other 18%. LINKTREE analysis showed a classification tree with eight splits, resulting in 9 biotopes.



Water column parameters (salinity and temperature) were included in the DistLM analysis as it is known that water quality greatly influences benthic habitat quality (Howes, Kelley et al., 2006) likely plays a role in driving the composition of benthic communities. However, water quality, especially temperature, is influenced by many factors such as tides, weather and time of the year, which have not been investigated in the course of this project. Water temperatures were likely changing at a faster pace than we were able to collect samples in Wellfleet Harbor (five weeks in August and September 2015) and while temperature data initially appeared to be an important factor in determining biotopes, we decided to exclude it from our analysis. The resulting biotopes would have been temporal and unstable and in terms of reproducibility and observability in nature. Additionally, temperature biotopes were likely an artifact of the sampling regime. More research, across multiple seasons and years, needs to be conducted in order to establish and integrate a sound water column component baseline.

Previous work using CMECS and similar statistical analyses, could explain 21-68.9% of the variance in benthic assemblage structure (Shumchenia and King, 2010, McHenry et al., 2017). These studies place the results of this work in context and indicate that the results reported here are well within the range of expectations for the methods used. Regardless, the explanatory power of this project could be increased by introducing a stratified random sampling regime based on acoustic imagery (in order to encompass all geoforms mapped) and collecting benthic invertebrate samples on a faster scale to avoid naturally occurring seasonal changes in species distribution, abundance and diversity.

### ***Sources of Uncertainty***

There are many sources of uncertainty to consider in “snapshot” surveys of the environment. First, the uncertainty in the representativeness of the observations themselves – do the measured parameters deviate over time, and on a regular basis? It is extremely likely that there is temporal variation, but that type of variability cannot be assessed with the data collected for this study. The data collected for this study provide a baseline from which future variability could be measured and assessed.

Second, there are sources of uncertainty in the selected analysis methods, including mapping. In order to map the data, assumptions were made that the mean infaunal abundances at each station or the median grain size were appropriate representatives of the datasets. A potentially large source of uncertainty in the maps produced for this study is the interpolation procedure. The tradeoff of certainty for full coverage and reproducibility was considered to be worthwhile. The alternatives to this approach would be either point-based maps (not full coverage), or manually drawn boundaries inferred from aerial photography, bathymetry, and/or backscatter imagery with assigned classifications based on a summary statistic (not reproducible).

## Summary:

- For this study, system-based mapping was prioritized - focusing on mapping embayments rather than along arbitrary delineations (such as CCNS boundaries). This system-wide approach not only provided comprehensive benthic habitat maps, but also robust baseline data for monitoring and management of Wellfleet Harbor. Additional outside support from the town of Wellfleet and other non-profits groups allowed entire systems to be mapped well outside park boundaries.
- Vessel-based acoustic mapping was greatly enhanced using phase-measuring sidescan sonar (PMSS), which is ideal for shallow water (less than 10 m) mapping. Co-located bathymetry and sidescan proved to be particularly useful for identifying eelgrass and other submerged aquatic vegetation, changes on the seafloor such as bedform migration and for identifying both natural and anthropogenic structures on the seafloor. Dual frequency high resolution sidescan produced exceptional imagery useful for many applications.
- Customized mapping platforms across two vessels made it possible to broaden the extent of ‘mappable’ areas in Wellfleet Harbor. The bow mount and configuration of the sonar and ancillary sensors was designed to be easily transported (i.e., switched back and forth between vessels), set up and broken down. This configuration reduced deployment time, optimized space on the vessel and helped to reduce uncertainty associated with sensor offsets and human - induced measurement errors. Shallow draft and maneuverability of both vessels were important for the navigation of varying depths and sea conditions.
- The high resolution sidescan mosaics are valuable for supervised delineation of eelgrass, aquaculture habitat and potentially other bottom features such as boulders and marine debris. The mosaics have the potential to verify surficial habitat patterns and adding fine-scale detail to benthic habitat maps. However, sidescan mosaics did not lend themselves to rapid or reproducible automated interpretation in the context of the Geoform, Substrate, or Biotic Components.
- Seismic reflection profiling (sub-bottom) worked well in the shallow water environment of Wellfleet Harbor. Five seismic facies were identified in Wellfleet Harbor, including glacial lake floor deposits, and facies interpreted as bedforms representing the extension of the barrier spit down the western side of Wellfleet Harbor onto Billingsgate Shoal. Marine mud (facies ‘M’), was found to be ubiquitous throughout much of the Harbor, (up to 10 m thick) and was interpreted to be estuarine mud deposited in low energy basins. Additional seismic surveys would provide managers with important information for future dredge projects and management of resources within the harbor.
- Since a great proportion of samples were taken in physically dynamic environments it is not surprising that characteristics of the substrate (i.e., grain size metrics) were the best variables for explaining patterns in benthic communities, versus factors such as depth and sediment organic content.

- We could explain 55.9% of species distribution based on geoform, % gravel and skewness. Of the 9838 individuals comprising 98 species we found one individual of a cryptogenic species: *Polycirrus eximius* (a polychaeta worm) at station 16. A cryptogenic species is a species whose origins are unknown; meaning it may be either a native species or an introduced species but clear evidence for either origin is absent.
- Our models suggested temperature to be an important factor explaining species distribution in and around Wellfleet Harbor. However, water temperature is an ever-changing variable and depends on weather, climate and season. Since benthic grab samples were collected over five weeks and strong tidal flow brings in cooler water, modelled biotopes including temperature would have been considered temporal biotopes that can potentially change on a day to day basis and are therefore not mappable.
- This study and associated data comprise a critical baseline record of biological and physical characteristics of Wellfleet Harbor and surrounding areas. As described throughout, the classification and mapping approach employed for this analysis is only one of many possible treatments of the data. There is an opportunity to explore the data collected during this study to better understand the importance of biotic habitat characteristics, such as macroalgal canopies and eelgrass beds, overlain on substrate composition. Future work might include an examination within system and among system differences. The results and maps from this study will be useful to guide future studies of coastal resources in Wellfleet Harbor.

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# Appendix A



Figure A-1. Sidescan Mosaic of Wellfleet Harbor and vicinity



## Appendix B

Table B-1. CMECS Biotic Component classifications for Wellfleet Harbor Stations 1-31.

Station	Component	Biotic Setting	Biotic Class	Biotic Subclass	Biotic group	Biotic Community	Most abundant species
WH01	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH02	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH03	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH04	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH05	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH06	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH07	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH08	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH09	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH10	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH11	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH12	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH13	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH14	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH16	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH17	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Glycera bed	Glyceridae/Goniadidae
WH18	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH19	Biotic	Benthic/Attached Biota	Faunal Bed	Soft Sediment Fauna	Clam Bed	Spisula Bed	Spisula solidissima
WH21	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH22	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH23	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH24	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH26	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH27	Biotic	Benthic/Attached Biota	Faunal Bed	Soft sediment fauna	Larger deep-burrowing fauna	Nephtys bed	Nephtyidae
WH28	Biotic	Benthic/Attached Biota	Faunal Bed	Soft Sediment Fauna	Clam Bed	Spisula Bed	Spisula solidissima
WH29	Biotic	Benthic/Attached Biota	Faunal Bed	Attached Fauna	Sessile Gastropod	Attached Crepidula	Crepidula fornicata
WH30	Biotic	Benthic/Attached Biota	Faunal Bed	Soft Sediment Fauna	Clam Bed	Spisula Bed	Spisula solidissima

WH31 | Biotic | Benthic/Attached Biota | Faunal Bed | Soft sediment fauna | Larger deep-burrowing fauna | Nephtys bed | Nephtyidae

Table B-2. Substrate component classifications for Wellfleet Harbor stations 1 -31.

Station	Component	Substrate Origin	Substrate Class	Substrate Subclass	Substrate Group
WH01	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH02	Substrate	Geologic Substrate	Unconsolidated mineral substrate	sand	Medium Sand
WH03	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH04	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Sand	Coarse Sand
WH05	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Gravelly	Gravelly Muddy Sand
WH06	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH07	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Gravelly	Gravelly Sand
WH08	Substrate	Geologic Substrate	Unconsolidated mineral substrate	sand	Medium Sand
WH09	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH10	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Sand	Coarse Sand
WH11	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH12	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH13	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH14	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH16	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH17	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH18	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH19	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Gravelly	Gravelly Muddy Sand
WH21	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH22	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH23	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH24	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH26	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH27	Substrate	Geologic Substrate	Unconsolidated mineral substrate	sand	Medium Sand
WH28	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
WH29	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand



<b>WH30</b>	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand
<b>WH31</b>	Substrate	Geologic Substrate	Unconsolidated mineral substrate	Slightly gravelly	Slightly Gravelly Sand

Table B-3. Geoform component classifications for Wellfleet Harbor stations 1 -31.

<b>Station</b>	<b>Component</b>	<b>Tectonic Setting Subcomponent</b>	<b>Physiographic Setting Subcomponent</b>	<b>Geoform Level 1</b>	<b>Geoform Level 2</b>
<b>WH01</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Banks
<b>WH09</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats <1m
<b>WH11</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats <1m
<b>WH12</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats <1m
<b>WH05</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH06</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH10</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH21</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH22</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH24</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH26</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH27</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH29</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats >3m
<b>WH02</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH03</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH04</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH07</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH08</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH13</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH14</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH16</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH17</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH18</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH19</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m

<b>WH23</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH28</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH30</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m
<b>WH31</b>	Geoform	Passive Continental Margin	Lagoonal Estuary	Barrier Flat	Flats between 1-3m

## Appendix C

Table C-1. List of species present in Wellfleet Harbor.

Species	Kingdom	Phylum	Class	Order	Family	Genus
<b>Acteocina canaliculata</b>	Animalia	Mollusca	Gastropoda	Cephalaspidea	Tornatinidae	Acteocina
<b>Ampelisca sp</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Ampeliscidae	Ampelisca
<b>Ampharetidae</b>	Animalia	Annelida	Polychaeta	Terebellida	Ampharetidae	
<b>Anachis sp</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Columbellidae	Anachis
<b>Anadara transversa</b>	Animalia	Mollusca	Bivalvia	Arcida	Arcidae	Anadara
<b>Anomia sp</b>	Animalia	Mollusca	Bivalvia	Pectinida	Anomiidae	Anomia
<b>Aoridae</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Aoridae	
<b>Arabella iricolor</b>	Animalia	Annelida	Polychaeta	Eunicida	Oeononidae	Arabella
<b>Argopecten irradians</b>	Animalia	Mollusca	Bivalvia	Pectinida	Pectinidae	Argopecten
<b>Aricidea sp</b>	Animalia	Annelida	Polychaeta	Scolecida	Paraonidae	Aricidea
<b>Astyris lunata</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Columbellidae	Astyris
<b>Batea catharinensis</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Bateidae	Batea
<b>Bittium alternatum</b>	Animalia	Mollusca	Gastropoda	Caenogastropoda	Cerithiidae	Bittium
<b>Cancer</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Canceridae	Cancer
<b>Capitellidae</b>	Animalia	Annelida	Polychaeta		Capitellidae	
<b>Caprellidae</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Caprellidae	
<b>Chiridotea coeca</b>	Animalia	Arthropoda	Malacostraca	Isopoda	Chaetiliidae	Chiridotea
<b>Cirratulidae</b>	Animalia	Annelida	Polychaeta	Terebellida	Cirratulidae	
<b>Clymenella torquata</b>	Animalia	Annelida	Polychaeta		Maldanidae	Clymenella
<b>Caryocorbula contracta</b>	Animalia	Mollusca	Bivalvia	Myida	Corbulidae	Caryocorbula
<b>Corophium spp</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Corophiidae	Corophium
<b>Crangon septemspinosa</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Crangonidae	Crangon
<b>Crepidula fornicata</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Calyptraeidae	Crepidula
<b>Ergaea walshi</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Calyptraeidae	Ergaea
<b>Cumacea</b>	Animalia	Arthropoda	Malacostraca	Cumacea		
<b>Diopatra cuprea</b>	Animalia	Annelida	Polychaeta	Eunicida	Onuphidae	Diopatra
<b>Dyspanopeus sayi</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Panopeidae	Dyspanopeus



<b>Edotia triloba</b>	Animalia	Arthropoda	Malacostraca	Isopoda	Idoteidae	Edotia
<b>Elasmopus levis</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Maeridae	Elasmopus
<b>Ensis leei</b>	Animalia	Mollusca	Bivalvia	Adapedonta	Pharidae	Ensis
<b>Erichthonius punctatus</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Ischyroceridae	Erichthonius
<b>Eupleura caudata</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Muricidae	Eupleura
<b>Euspira heros</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Naticidae	Euspira
<b>Gemma gemma</b>	Animalia	Mollusca	Bivalvia	Venerida	Veneridae	Gemma
<b>Glycera americana</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Glyceridae	Glycera
<b>Haminoea solitaria</b>	Animalia	Mollusca	Gastropoda	Cephalaspidea	Haminoeidae	Haminoea
<b>Harmothoe imbricata</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Polynoidae	Harmothoe
<b>Haustoriidae sp</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Haustoriidae	
<b>Hexapanopeus angustifrons</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Panopeidae	Hexapanopeus
<b>Holothuroidea</b>	Animalia	Echinodermata	Holothuroidea			
<b>Lacuna vincta</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Littorinidae	Lacuna
<b>Leptosynapta</b>	Animalia	Echinodermata	Holothuroidea	Apodida	Synaptidae	Leptosynapta
<b>Lumbrineridae</b>	Animalia	Annelida	Polychaeta	Eunicida	Lumbrineridae	
<b>Lyonsia hyalina</b>	Animalia	Mollusca	Bivalvia		Lyonsiidae	Lyonsia
<b>Lysianopsis alba</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Lysianassidae	Lysianopsis
<b>Majidae</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Majidae	
<b>Maldanidae</b>	Animalia	Annelida	Polychaeta		Maldanidae	
<b>Melita nitida</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Melitidae	Melita
<b>Mercenaria mercenaria</b>	Animalia	Mollusca	Bivalvia	Venerida	Veneridae	Mercenaria
<b>Microdeutopus anomalus</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Aoridae	Microdeutopus
<b>Mitrella lunata</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Columbellidae	Astysis
<b>Mulinia lateralis</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Muricidae	Mulinia
<b>Muricidae</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Muricidae	
<b>Tritia obsoleta</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Nassariidae	Tritia
<b>Tritia trivittata</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Nassariidae	Tritia
<b>Naticidae sp</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Naticidae	
<b>Nephtys bucera</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Nephtyidae	Nephtys

<b>Nereididae</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Nereididae	
<b>Ocypodidae</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Ocypodidae	
<b>Oligochaeta</b>	Animalia	Annelida	Clitellata			
<b>Onuphidae</b>	Animalia	Annelida	Polychaeta	Eunicida	Onuphidae	
<b>Ophelia spp</b>	Animalia	Annelida	Polychaeta		Opheliidae	Ophelia
<b>Orbiniidae</b>	Animalia	Annelida	Polychaeta		Orbiniidae	
<b>Ovalipes ocellatus</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Ovalipidae	Ovalipes
<b>Oxyurostylis smithi</b>	Animalia	Arthropoda	Malacostraca	Cumacea	Diastylidae	Oxyurostylis
<b>Pagurus longicarpus</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Paguridae	Pagurus
<b>Pagurus sp</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Paguridae	Pagurus
<b>Palaemon</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Palaemonidae	Palaemon
<b>Pandora gouldiana</b>	Animalia	Mollusca	Bivalvia		Pandoridae	Pandora
<b>Panopeus herbstii</b>	Animalia	Arthropoda	Malacostraca	Decapoda	Panopeidae	Panopeus
<b>Paraonidae spp</b>	Animalia	Annelida	Polychaeta		Paraonidae	
<b>Pectinaria gouldii</b>	Animalia	Annelida	Polychaeta	Terebellida	Pectinariidae	Pectinaria
<b>Phoxocephalidae spp</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Phoxocephalidae	
<b>Phyllodocidae</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Phyllodocidae	
<b>Polychaeta</b>	Animalia	Annelida	Polychaeta			
<b>Polycirrus eximius</b>	Animalia	Annelida	Polychaeta	Terebellida	Terebellidae	Polycirrus
<b>Polydora cornuta</b>	Animalia	Annelida	Polychaeta	Spionida	Spionidae	Polydora
<b>Polygordius spp</b>	Animalia	Annelida	Polychaeta		Polygordiidae	Polygordius
<b>Polyplacophora</b>	Animalia	Mollusca	Polyplacophora			
<b>Rissoidae</b>	Animalia	Mollusca	Gastropoda	Littorinimorpha	Rissoidae	
<b>Sabellidae</b>	Animalia	Annelida	Polychaeta	Sabellida	Sabellidae	
<b>Parougia caeca</b>	Animalia	Annelida	Polychaeta	Eunicida	Dorvilleidae	Parougia
<b>Sigalionidae</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Sigalionidae	
<b>Sipuncula</b>	Animalia	Sipuncula				
<b>Solemya velum</b>	Animalia	Mollusca	Bivalvia	Solemyida	Solemyidae	Solemya
<b>Sphaeroma quadridentatum</b>	Animalia	Arthropoda	Malacostraca	Isopoda	Sphaeromatidae	Sphaeroma
<b>Spiochaetopterus oculatus</b>	Animalia	Annelida	Polychaeta		Chaetopteridae	Spiochaetopterus
<b>Spionidae</b>	Animalia	Annelida	Polychaeta	Spionida	Spionidae	

<b>Spisula solidissima</b>	Animalia	Mollusca	Bivalvia	Venerida	Mactridae	Spisula
<b>Streblospio benedicti</b>	Animalia	Annelida	Polychaeta	Spionida	Spionidae	Streblospio
<b>Syllidae</b>	Animalia	Annelida	Polychaeta	Phyllodocida	Syllidae	
<b>Ameritella agilis</b>	Animalia	Mollusca	Bivalvia	Cardiida	Tellinidae	Ameritella
<b>Terebellidae</b>	Animalia	Annelida	Polychaeta	Terebellida	Terebellidae	
<b>Testudinalia testudinalis</b>	Animalia	Mollusca	Gastropoda		Lottiidae	Testudinalia
<b>Unciola sp</b>	Animalia	Arthropoda	Malacostraca	Amphipoda	Unciolidae	Unciola
<b>Anemonia</b>	Animalia	Cnidaria	Anthozoa	Actinaria	Actiniidae	Anemonia
<b>Tanaididae</b>	Animalia	Arthropoda	Malacostraca	Tanaidacea	Tanaididae	
<b>Urosalpinx cinerea</b>	Animalia	Mollusca	Gastropoda	Neogastropoda	Muricidae	Urosalpinx

## Appendix D

Table D-1. Grain size analysis across benthic invertebrate stations (LOI – percent organic matter as loss on ignition).

Name	Year	Station	Latitude	Longitude	Gravel_%	Sand_%	Silt_%	Clay_%	LOI %
2015_WH1-S	2015	1	41.90059	-70.05179	3.446	95.887	0.533	0.135	0.621
2015_WH2-S	2015	2	41.908547	-70.028738	0.000	98.667	1.091	0.242	0.715
2015_WH3-S	2015	3	41.915995	-70.054301	0.218	97.560	1.964	0.257	0.752
2015_WH4-S	2015	4	41.921201	-70.052075	0.000	100.000	0.000	0.000	0.459
2015_WH5-S	2015	5	41.856312	-70.038901	11.756	87.692	0.472	0.079	0.630
2015_WH6-S	2015	6	41.861219	-70.044639	1.524	97.877	0.502	0.098	0.454
2015_WH7-S	2015	7	41.873886	-70.017753	6.613	92.628	0.647	0.112	0.612
2015_WH8-S	2015	8	41.870536	-70.017416	0.000	98.526	1.325	0.148	1.408
2015_WH9-S	2015	9	41.88971	-70.01496	0.306	98.257	1.265	0.172	0.787
2015_WH10-S	2015	10	41.878009	-70.036357	-0.121	100.121	0.000	0.000	1.138
2015_WH11-S	2015	11	41.888641	-70.011076	0.035	99.187	0.625	0.154	0.675
2015_WH12-S	2015	12	41.88792	-70.00723	0.205	97.690	1.826	0.278	3.981
2015_WH13-S	2015	13	41.860463	-70.077095	2.961	95.850	1.075	0.114	0.000
2015_WH14-S	2015	14	41.858516	-70.080713	3.111	95.773	1.008	0.108	0.615
2015_WH16-S	2015	16	41.90358	-70.01953	0.492	98.854	0.528	0.126	0.624
2015_WH17-S	2015	17	41.930578	-70.024588	1.950	96.938	0.939	0.173	0.641
2015_WH18-S	2015	18	41.92222	-70.040948	3.958	91.854	3.832	0.356	1.702
2015_WH19-S	2015	19	41.937139	-70.07777	9.765	89.618	0.505	0.113	0.554
2015_WH21-S	2015	21	41.894458	-70.060797	0.416	97.893	1.461	0.230	1.455
2015_WH22-S	2015	22	41.88192	-70.04841	3.276	95.793	0.930	0.000	0.697
2015_WH23-S	2015	23	41.900523	-70.057228	0.397	98.775	0.684	0.144	0.442
2015_WH24-S	2015	24	41.903813	-70.080153	1.374	97.922	0.579	0.125	0.448
2015_WH26-S	2015	26	41.925555	-70.089017	1.455	98.545	0.000	0.000	0.300
2015_WH27-S	2015	27	41.889868	-70.092973	0.000	98.182	1.651	0.166	0.644
2015_WH28-S	2015	28	41.891584	-70.072288	2.176	97.252	0.477	0.095	0.381
2015_WH29-S	2015	29	41.94315	-70.083357	2.251	96.537	1.089	0.123	0.795
2015_WH30-S	2015	30	41.92854	-70.07448	2.835	97.165	0.000	0.000	0.471

2015_WH31-S	2015	31	41.9085	-70.02757	0.178	99.822	0.000	0.000	0.653
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## Appendix E

Table E-1 Water column data for Wellfleet Harbor stations 1 - 31 (averages).

Station	Latitude	Longitude	Temperature C	Sound Vel. m <sup>-1</sup>	Depth m	Salinity ppt
WH01	41.90058	-70.05179	25.29	1530.61	1.96	30.78
WH02	41.90870	-70.02882	21.97	1522.16	1.65	30.77
WH03	41.91606	-70.05430	22.64	1524.26	2.10	31.06
WH04	41.92123	-70.05214	22.58	1523.80	1.21	30.82
WH05	41.85641	-70.03888	24.34	1528.52	3.15	31.00
WH06	41.86140	-70.04463	24.41	1528.71	3.30	30.99
WH07	41.87421	-70.01766	21.50	1521.35	1.35	31.17
WH08	41.87052	-70.01739	25.44	1531.25	1.50	31.05
WH09	41.88972	-70.01499	24.88	1530.11	1.21	31.27
WH10	41.87799	-70.03638	25.80	1532.06	1.64	31.01
WH11	41.88869	-70.01106	24.26	1528.58	1.06	31.26
WH12	41.88801	-70.00730	24.67	1529.68	1.04	31.34
WH13	41.86053	-70.07695	19.61	1516.19	1.07	31.24
WH14	41.85838	-70.08071	19.96	1517.12	1.49	31.19
WH16	41.90352	-70.01944	24.93	1530.29	2.24	31.31
WH17	41.93055	-70.02450	25.00	1529.38	1.95	30.30
WH18	41.92228	-70.04097	22.38	1523.21	1.79	30.74
WH19	41.93718	-70.07783	21.44	1521.06	1.50	31.08
WH21	41.89444	-70.06079	25.10	1530.08	1.80	30.72
WH22	41.88193	-70.04836	23.73	1526.82	2.25	30.86
WH23	41.90055	-70.05720	25.36	1530.74	1.49	30.75
WH24	41.90378	-70.08011	21.59	1521.41	2.69	31.01
WH26	41.92561	-70.08903	22.01	1522.73	3.45	31.17
WH27	41.88981	-70.09290	20.46	1518.54	2.10	31.19
WH28	41.89161	-70.07220	22.02	1522.51	1.35	30.98
WH29	41.94311	-70.08339	22.38	1523.61	3.44	31.09
WH30	41.92847	-70.07428	22.74	1524.56	1.50	31.10



<b>WH31</b>	41.90790	-70.02461	24.59	1529.38	2.25	31.24
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