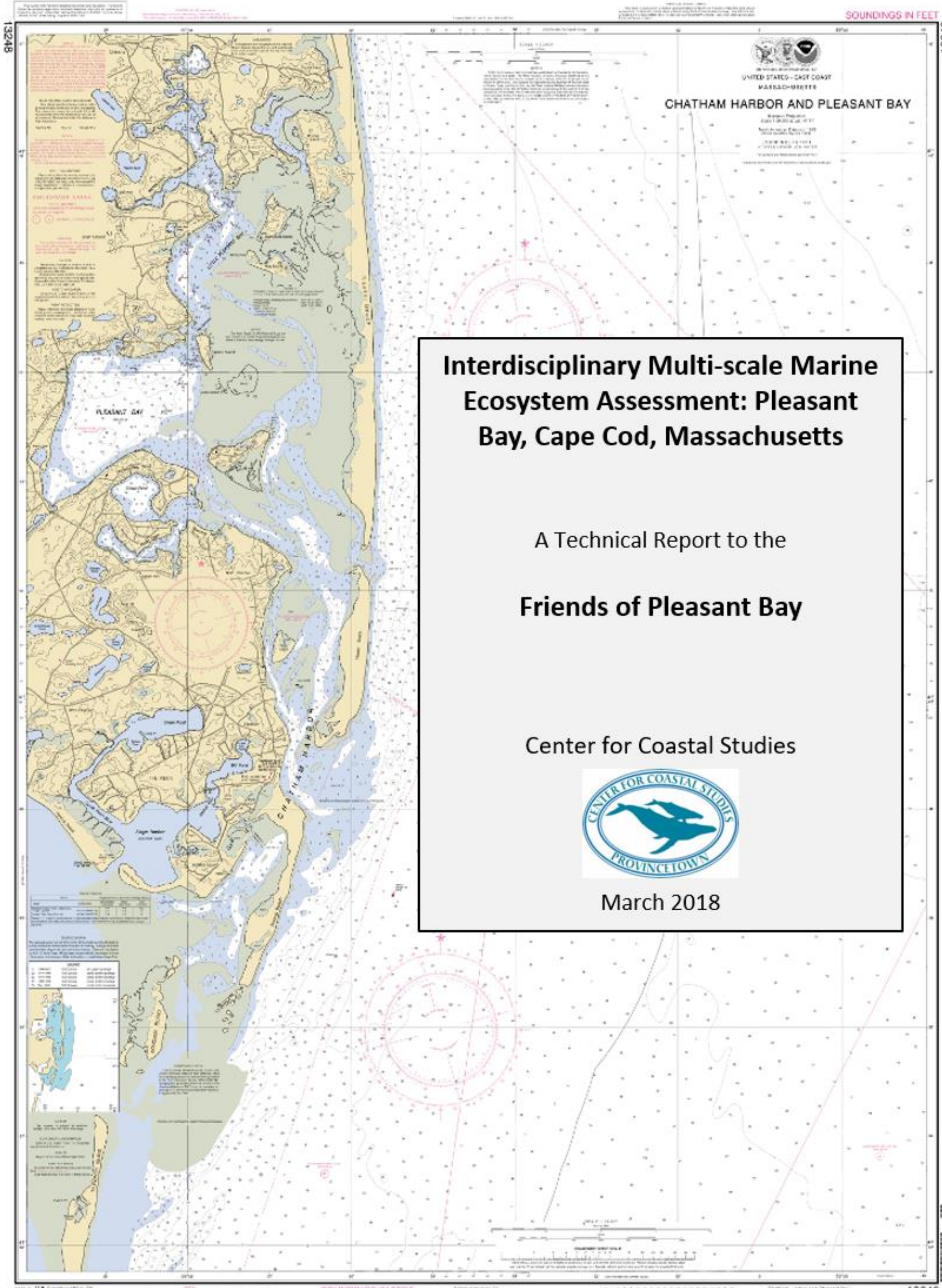

Interdisciplinary Multi-scale Marine Ecosystem Assessment: Pleasant Bay, Cape Cod, Massachusetts

A Technical Report to the

Friends of Pleasant Bay

Center for Coastal Studies

March 2018

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Interdisciplinary Multi-scale Marine Ecosystem Assessment: Pleasant Bay, Cape Cod, Massachusetts

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Summary

Summary: Pleasant Bay Environmental Assessment

Pleasant Bay is a coastal embayment that is part of the Nauset Beach/Monomoy Island - barrier spit-barrier island system. The Bay's watershed spans the towns of Orleans, Brewster, Harwich and Chatham. It is a highly valued regional resource, designated by the state and recognized by the surrounding towns as an Area of Critical Environmental Concern.

The Friends of Pleasant Bay (FOPB) funded the Center for Coastal Studies (CCS) to conduct an environmental assessment of Pleasant Bay between 2014 and 2017. The goal of this assessment was to create an important dataset of baseline information assessing the present status of the natural resources of Pleasant Bay that can be used to develop a long-term habitat monitoring program. Additional support was provided by the US National Park Service (NPS), Cape Cod National Seashore (CCNS), the International Fund for Animal Welfare (IFAW) and many volunteers.

The Pleasant Bay Environmental Assessment:

- Developed high-resolution benthic habitat maps, integrating data collected through acoustic mapping of the Bay, seismic reflection profiling, sediment coring, bottom grab samples and videos to type sediment and identify the micro-invertebrates by sediment type.
- Determined the distribution and relative abundance of individual species of shellfish and finfish using a variety of capture methods.
- Described the seasonal distribution for gray and harbor seals in Pleasant Bay during 2014 and 2015 based on aerial surveys
- Provided additional information on the diet of gray and harbor seals in Pleasant Bay through scat content analysis
- Provided an initial representation of the interrelationships among the Bay's biological and physical features.

The following Technical Report is presented in four chapters:

(1) A Benthic habitat map for Pleasant Bay, Cape Cod, Massachusetts

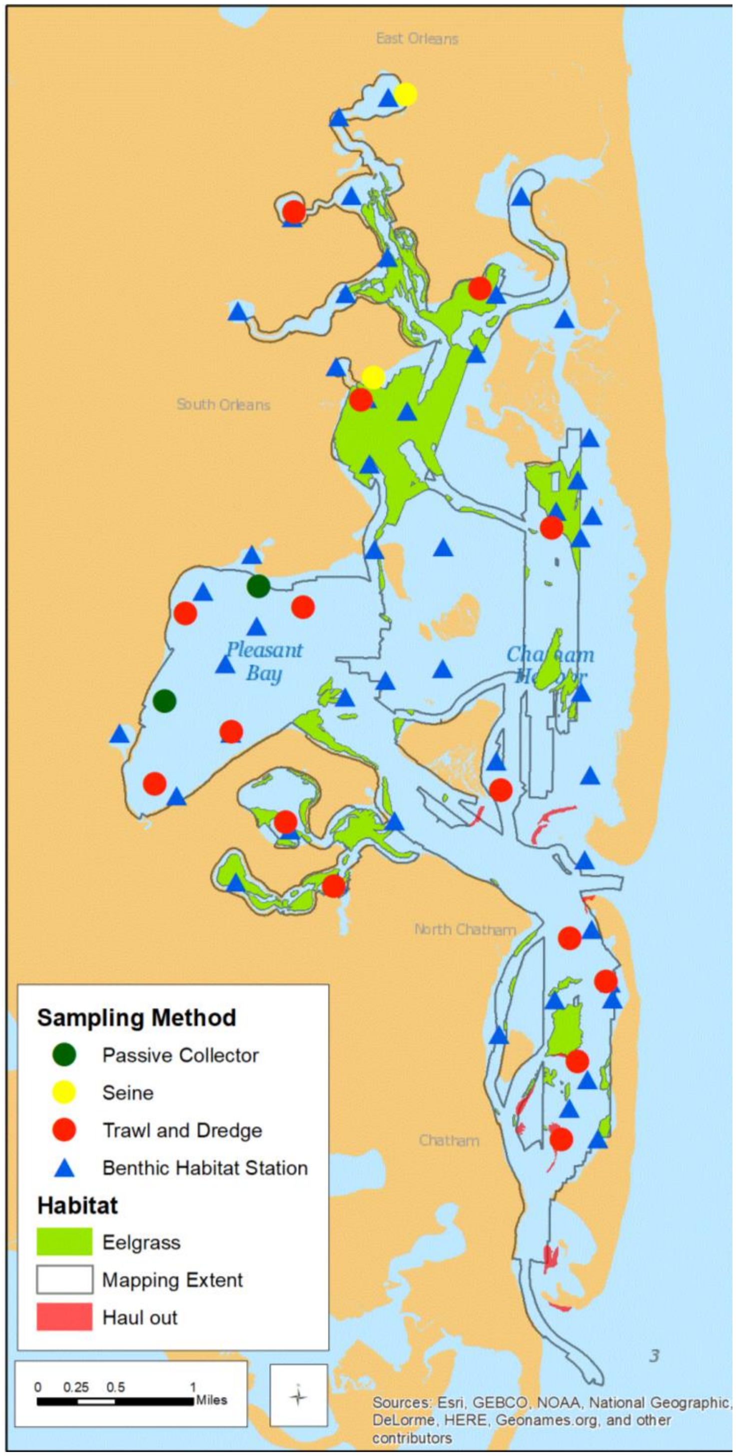
(2) Fisheries investigations in Pleasant Bay, Cape Cod, Massachusetts

(3) The seasonal distribution, counts and prey of harbor seals (*Phoca vitulina vitulina)* and gray seals (*Halichoerus grypus atlantica*) in Pleasant Bay, Cape Cod, Massachusetts

(4) Integrating habitats and their constituents of Pleasant Bay, Cape Cod, Massachusetts

Chapter 1: “A Benthic Habitat Map for Pleasant Bay” discusses the methods used to develop benthic habitat maps in Pleasant Bay. The collection, interpretation and synthesis of acoustic, vessel-based surveys; benthic grab samples and analysis; sub-bottom profiling; and sediment coring, was undertaken to provide insights into current conditions within the bay and to guide future studies. All of these data and maps were developed using the Coastal and Marine Ecological Classification Standard, or CMECS, the federally-mandated system for producing such maps.

Sidescan sonar imagery covering 16.82 km^2 was collected, and within that area, 6.78 km^2 of co-located bathymetric data were collected with a mean depth of 3.20 m. from July-December 2014. A total of 192 bottom grab samples were collected to sample macroinvertebrates and sediments along with other habitat data at 48 locations within Pleasant Bay, 15 of which were selected to overlap with benthic stations sampled by the Massachusetts Estuaries Project (MEP) study conducted in 2003. The acoustic surveys and subsequent map production identified natural geological processes, as well as human-induced impacts on the seafloor. Using CMECS, eight sedimentary features and, their associated micro-invertebrate communities and indicator species were identified within the bay. Based on species abundance and distribution twelve distinct biotic communities were also identified. There were 150 micro-invertebrate species found but only 32 of these comprised the top 95% of all individuals in the benthic communities.



Chapter 2: “Fisheries Investigations in Pleasant Bay” consists of results from an inventory of shellfish and finfish in Pleasant Bay, with a focus on commercially and recreationally important species. Systematic and opportunistic fish and invertebrate

sampling was conducted in Pleasant Bay from July 2014 through October 2017. This comprehensive inventory indicated that Pleasant Bay is home to a diverse assemblage of marine animals, many of which utilize the Bay as spawning or nursery habitat.

Sampling methods included trawls (90 tows), beach seining (15 hauls), dredging (102 tows), ventless lobster traps (6 sets), and gillnets (3 sets). Passive collectors targeting juvenile lobster were set and recovered in summer 2014, and additional trawl, seine, and passive collector sampling efforts targeting tropical fishes occurred on an opportunistic basis from August to October 2016 and 2017. Where practical, sampling efforts were conducted using similar methods and gears compared to previous studies conducted in the same area, particularly the 1965-66 Massachusetts Division of Marine Fisheries (MADMF) assessment. Intertidal and subtidal survey effort (trawl, dredge and beach seining) was distributed relatively evenly over the year, although there were gaps due to fall and winter weather conditions. The overall species community and seasonal abundance of most species was broadly similar to that observed in the 1965-66 MADMF study. There were several species collected in this study that were not observed by Fiske et al. (1967) and vice versa.

Chapter 3: "The Seasonal distribution, counts and prey of harbor seals (*Phoca vitulina vitulina*) and gray seals (*Halichoerus grypus atlantica*) in Pleasant Bay" summarizes the distribution and counts, and prey of harbor seals (*Phoca vitulina vitulina*) and gray seals (*Halichoerus grpyus atlantica*) in Pleasant Bay. Monthly aerials surveys were conducted in 2014 and 2015. Scat samples were collected at established harbor and gray seal haul-outs monthly from January 2016 through March 2017. Prey was identified using otoliths and other hard part remains recovered through scat processing. Percent frequency of occurrence (%FO) was calculated for all prey items identified.

Nine haul-out sites were identified that were used throughout the study period (species dependent). In 2015, as gray seal numbers increased inside the Bay, their distribution shifted north to include a series of developing tidal sand bars west of Nauset Beach and southeast of Strong Island. The diets of both harbor and gray seals were largely dominated by sand lance. Hard parts of herring and cod species were present 33% in winter scat of harbor seals. Longfin squid were the second most abundant prey item in gray seal scat.

Chapter 4: "Integrating habitats and their constituents of Pleasant Bay" discusses the integration of data collected in the benthic habitat, fisheries independent, and seal surveys as described in the previous chapters. This chapter outlines and utilizes non-parametric statistics and presents links between the benthic invertebrate communities, and the shellfish and finfish communities. This chapter also examines links between fish distributions and seal diet observed from scat analysis. The analysis presented identifies linkages throughout the habitats of Pleasant Bay.

The statistical methodologies employed identified species that are driving community differences are indicative of different habitat types, i.e. sand, eelgrass, mud, gravel as these groups have various community compositions with different proportions of micro-invertebrates. These distinct benthic community types are linked to the distribution of shellfish within the Bay. In examining shellfish species caught in the trawls and dredges, the analyses revealed that their distribution was significant when compared to both distance from inlet (describing a gradient of open ocean species to estuarine associated species) and across different benthic invertebrate community assemblages.

Further examination of fish communities indicates that the species driving fish community trends adjacent to seal haul outs are the same species that are present in seal diet analysis. The fisheries independent surveys and the seal diet surveys indicate that seals are potentially utilizing the resources in Pleasant Bay and the hard parts analysis indicates that they are consuming what is seasonally abundant within the system.

CHAPTER 1
A benthic habitat map for Pleasant Bay, Cape Cod, Massachusetts

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Abstract

Cape Cod National Seashore was chosen as one of four contemporaneous studies in coastal National Parks along the Atlantic coast to develop methods to produce benthic habitat maps. With support from the Friends of Pleasant Bay the entirety of Pleasant Bay and Chatham Harbor was mapped as much of the Bay is outside of Seashore boundaries. This chapter discusses the methods of data collection, processing and analysis for the production of the benthic habitat maps. Data from a phase-measuring sidescan sonar, bottom grab samples, seismic reflection profiling, and sediment coring were all used to develop submerged marine habitat maps using the Coastal and Marine Ecological Classification Standard (CMECS). One of the motivations for the large study was to provide managers with a baseline inventory of benthic habitats against which to measure future change resulting from natural and anthropogenic phenomena.

Vessel-based acoustic surveys (n = 16) were conducted in Pleasant Bay from 14 July - 04 December 2014. Sidescan sonar imagery covering 16.82 km^2 was collected, and within that area, 6.78 km^2 of co-located bathymetric data were collected with a mean depth of 3.20 m. A total of 192 bottom grab samples were collected to sample macroinvertebrates and sediments along with other water column and habitat data at 48 locations within Pleasant Bay. These data were used along with the geophysical and coring data to develop final habitat maps using the CMECS framework.

1.1. Introduction

The benthic habitat mapping project presented here was part of a larger study within Cape Cod National Seashore, which itself was one of four contemporaneous studies conducted by the US National Park Service (NPS). The other associated studies took place in Fire Island National Seashore, Gateway National Recreation Area and Assateague Island National Seashore. The larger NPS project was designed to create a baseline inventory of existing marine (or benthic) habitats in coastal parks. These baseline data can also be used to measure future natural and anthropogenic change in these environments. The impetus for these studies was the realization after Hurricane Sandy, that very few, if any benthic habitat maps existed in coastal parks and managing resources without a map of those resources can result in a disjointed and piecemeal approach.

Hurricane Sandy was late-season hurricane that made landfall three times, first in Jamaica, then Cuba and finally in the United States. The hurricane had weakened, and maximum sustained winds had decreased to 70 knots, several hours before landfall near Brigantine, New Jersey. There were 147 deaths associated with the storm and over $50 billion in damage (Blake et al., 2013). Coastal managers and other stakeholders needed to assess the impact of the storm on both human uses and ecological resources. Ecological assessments can be particularly challenging in marine settings due to the difficulty of accessing submerged resources, and can be further complicated if no pre-existing maps of those resources are available. Most of the 85 coastal parks in the U.S. have little or no data with regards to benthic habitats in spite of the recognized role these data play in understanding and appropriately managing these resources (Curdts and Cross, 2013). The ecological value of submerged habitats, the presumed ecological impact of a powerful storm, and the lack of rigorous pre-storm resource maps were the driving forces for the larger project.

Development of 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, Brown et al. 2012), 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 the Coastal and Marine Ecological Classification Systems (CMECS), the national standard for these types of data products. 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.

The data collection methods used in each of the four studies were largely similar to enable comparison of results from park to park and to guide future investigators seeking to update the inventories and conduct ancillary research based on these data. The data set for the larger NPS project did not include 15 additional sites funded through the Friends of Pleasant Bay (FOPB) and therefore the maps within this report are different from the NPS report (Borrelli et al., 2018) as the additional locations provided more detail to be represented.

Vessel-based acoustic data used to map the seafloor combined with benthic grab samples were central to the four studies. These surficial data provide a snapshot of existing conditions both physically and biologically as well as provide a baseline from which to measure future changes in precise locations and throughout several embayments. Surficial data were augmented in Pleasant Bay with Seismic Reflection (Sub-Bottom) Profiling to provide the context for sediment cores that together, enable characterization and analysis of basin evolution.

1.1.1. Study Area

The Laurentide ice sheet advanced into southern New England approximately 25,000 years Before Present (BP) and was at its maximum southern extent at 20,000 years BP (Uchupi et al., 1996). The glacial ice in southern New England during this time was approximately 500 m thick. From 20,000-18,000 years BP, as temperatures increased and the ice-receded, the bulk of the sediment that comprises Cape Cod was deposited (Uchupi et al., 1996). As the rate of sea level rise began to decrease 6,000 years BP, southeastern Cape Cod began to take on its characteristic morphology (Davis, 1895; Johnson, 1925; Uchupi et al., 1996). Between 6,000-4,000 years BP barrier spits began to develop and subsequently small embayments were formed. Coastal salt marshes throughout New England began to form 4,000 years BP (Redfield and Rubin, 1962; Redfield, 1972; Roman et al., 2000). In working with science staff from Cape Cod National Seashore it was determined that the majority of the mapping conducted for this study was to be done in and around these embayments.

Eroding glacial bluffs are the primary source of material for the 'outer cape' littoral cell. A littoral cell is a stretch of coast that includes all the sources, transport paths and sinks of sediment (Komar, 1998).

A nodal point currently exists approximately in the vicinity of Nauset Beach, immediately south of Nauset Inlet, and from there the direction of net sediment transport diverges to either the north or south (Giese et al., 2011).

This littoral cell is approximately 90 km long and stretches from Long Point in Provincetown to Monomoy Island in Chatham. The entirety of the shoreline is owned by either Cape Cod National Seashore or the Monomoy National Wildlife Refuge, and may constitute the largest natural littoral cell in the lower 48 states. The predominant winds in the study area are from the southwest (SW) in the summer and northwest (NW) in the winter. The primary drivers of change along the coast are extratropical storms, known locally as nor'easters, as the dominant winds associated with these storms come from the northeast.

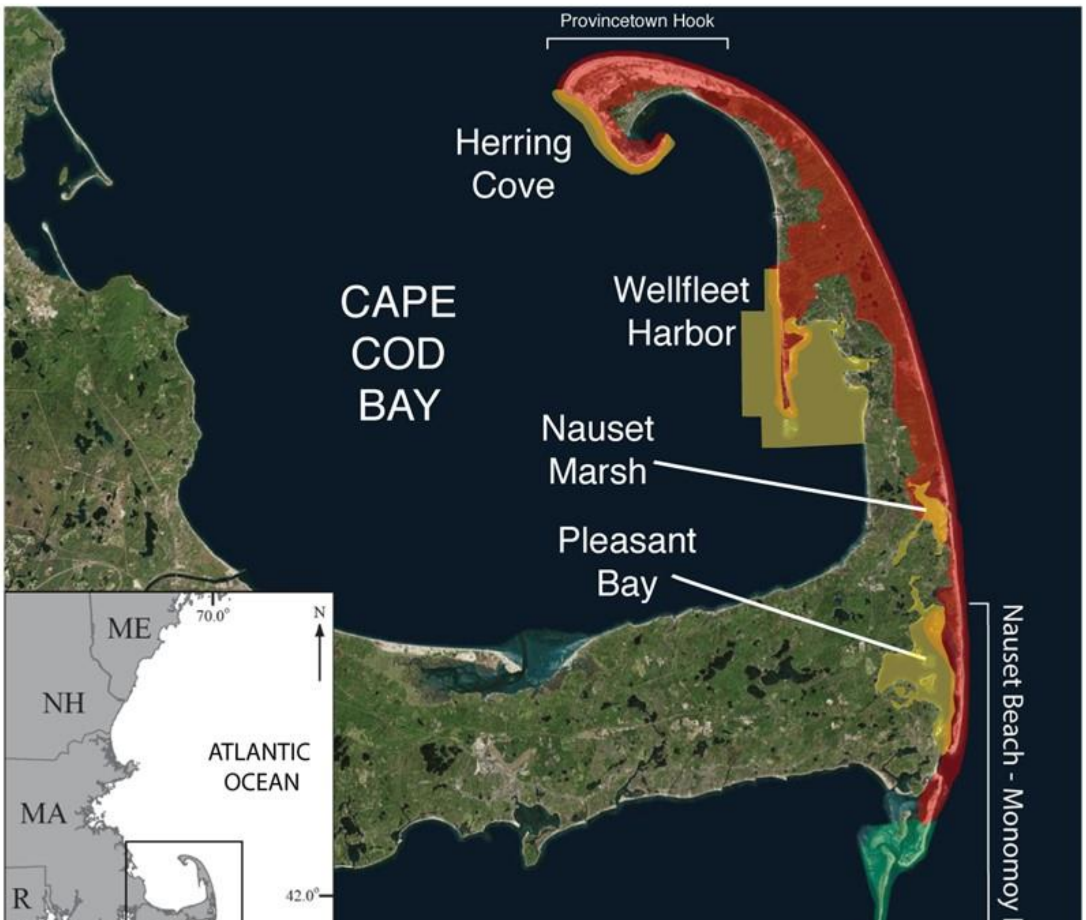

Figure 1.1. Locus map of Cape Cod National Seashore and the four survey sites, including coastal embayments and boundary of Monomoy National Wildlife nearshore waters. Red area (orange in mapped areas) is the park boundary, Green area is surrounding Monomoy Island

Pleasant Bay is a coastal lagoon[1] that is part of the Nauset Beach/Monomoy Island - barrier spit-barrier island system (figure 1.1). The westernmost area of the embayment, locally known as ‘Big Bay’, is an ice-block basin or ‘kettle hole’, as are many of the sub-embayments within Pleasant Bay (figure 1.2). The formation of these features occurs during de-glaciation as the retreating ice sheet breaks up and blocks of ice are deposited and buried by sediments transported by waters from the melting glacier.

Two tidal inlets currently provide the embayment with direct tidal exchange to the Atlantic Ocean. The system has a 150-year cycle of inlet formation, migration, and new inlet formation with a period of tide-dominated inlet development followed by a wave-dominated inlet migration phase (Giese, 1988; Giese et al., 2009). In 2007 a new inlet formed updrift of the exiting inlet which formed in 1987. The direction of net sediment transport is from north to south (Giese, et al, 2011). It was expected that after the 2007 inlet formed in a more hydrodynamically efficient position the 1987 inlet would close. Almost immediately after the 1987 inlet formed it became the primary inlet and by 1991-1992 it was the only inlet. Conversely, since the 2007 inlet formed the system has remained a two-inlet system and has become less stable, which is atypical for this system (Borrelli, et al., 2011). However, recent work suggests that the 2007 inlet may be capturing more of the tidal prism (Legare and Giese, 2016) and could soon become the primary inlet.

The present day Nauset Beach barrier spit that partially encloses Pleasant Bay is a direct result of incoming waves eroding the coastal bluffs to the north and entraining that material into the longshore sediment transport system in a predominantly southerly direction. The Nauset Beach barrier spit migrates landward primarily as a result of erosion on the open ocean shoreline and deposition on the backbarrier shoreline during overwash events (Leatherman, 1979).

The tidal range just offshore of Nauset Beach is 2.0 m, in Chatham Harbor the tidal range is 1.4 m with a spring tidal range of 1.6 m (http://tidesandcurrents.noaa.gov). The significant wave height outside the bay is 1.5 m (http://www.ndbc.noaa.gov). This is a mixed-energy, wave-dominated coast according to the classification developed by (Hayes, 1979). Pleasant Bay is 5 km across at its widest point with mean and maximum depths of 2.0 m and 6.0 m, respectively (Howes et al., 2006).

1.1.2. Submerged Habitat Maps

The purpose of this work was to integrate the physical and biological characteristics of submerged marine habitats from data obtained by CCS into a series of map products that describe the CMECS Geoform, Substrate, and Biotic Components. CMECS itself is “data agnostic” (FGDC 2012), meaning

[1] Pleasant Bay is historically referred to as an estuary. However, the primary characteristic of an estuary is an embayment where salt water and freshwater (typically from a river) mix. It is in this area of mixing where estuarine processes occur and dominate. There is little freshwater input to Pleasant Bay, and very few areas where estuarine characteristics prevail, therefore from a physical, biological and hydrological standpoint it would more rightly be characterized as a lagoon and is referred to as such in the technical report.

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.

Data collection was guided by the standards established for the broader NPS project. Between 2014 and 2016, the Center for Coastal Studies (CCS) conducted mapping surveys in and around CCNS to characterize submerged marine habitats. At each study site, CCS collected vessel-based acoustic data and bottom grab samples, and produced several data products useful for characterizing the physical and biological elements of submerged marine habitats. These methods and resulting data are discussed more in the next section. Data analysis involves the integration of these various data streams into sets of habitat maps. 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. For example, Zajac et al. (2000) mapped seafloor geologic units in Long Island Sound from backscatter imagery and sediment grain size; since infaunal assemblages showed a high degree of variation within these geologic units, they mapped significantly different infaunal assemblages as points overlaid on top of the geologic map. This result underscores the need for the third and final approach, known as "predict first, assemble later". Benthic infauna often overlap sediment transitions or boundaries and equating substrate with benthic assemblage type will lead to inaccurate maps (Diaz et al., 2004, Hewitt et al., 2004, Stevens and Connolly 2004, Shumchenia and King 2010). Brown et al. (2011) described the "predict first, assemble later" approach 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 which physical variables explain the most variance in benthic community structure, then uses those variables to create predictive habitat maps.

Figure 1.2. The study site: Pleasant Bay (PB). Aerial photograph from 2014

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 "predict first, assemble later" analysis strategy. Further, this analytical approach has been employed to identify and predict CMECS Biotopes in a shallow soft-sediment environment in the Northeast U.S. (Shumchenia and King, 2010). Recently it has been used to predict species assemblages, biomass, and diversity on the Maine coastal shelf (McHenry et al. 2017). To identify which physical variables were responsible for the most variation in benthic community structure at each study site within CCNS, multivariate regression trees (MRT) created with LINKTREE from the field data were analyzed. LINKTREE also identified thresholds in each of the driving physical variables that correspond to occurrences of different benthic assemblages. This information was used to develop full-coverage maps of the driving physical variables, classified using the thresholds identified by LINKTREE that correspond to those differences in benthic assemblages. The resulting maps, therefore, contained units that correspond to CMECS Biotopes, "a combination of abiotic habitat and associated species" (FGDC, 2012).

The detailed methods used to classify the data products in this study are described in the next section. However, it is important to note here that the classification scheme chosen for this study and the wider NPS project—CMECS—is a national standard. As such, CMECS was designed to classify coastal and marine habitats throughout U.S. waters, and its ability to offer classification precision at local, ecologically-relevant scales is still evolving. In fact, it was the intention of using the lessons learned from these four studies to improve CMECS (M. Finkbeiner, pers. comm.). Shumchenia and King (2010) found that the thematic resolution of the CMECS Substrate Component was not adequate to differentiate biotopes identified in Greenwich Bay, RI. In other words, the data collection and analysis methods used for any study may be robust and designed to capture the environmental and biological variability at a particular location, but classifying those data using CMECS may obscure important habitat patterns. Regardless, as a national standard, CMECS has significant value as a common language for the inventory and/or comparison of habitat data collected by disparate and varied programs within U.S. regions, coasts, and the nation as a whole.

Given the process described above to develop data collection, analysis, and classification approaches, there are a few caveats or limitations to consider. First, the four study areas within CCNS are each varied and diverse environments, and increased sampling effort and alternative data analysis is usually required to characterize heterogeneous habitats. However, a limited number of ground-truth (e.g., sediment properties, infauna) samples were allocated to each study area in order to ensure adequate coverage of each individual study area. Second, and relatedly, ground-truth locations were chosen prior to the interpretation of the bathymetry and data. Therefore, although this study design attempted to distribute samples among all the possible estimated geologic habitat types, this may not have been successful in practice. Lastly, water quality is likely an important factor influencing biotic elements of submerged marine habitats in coastal estuaries, and likely CCNS embayments as well. An earlier study in PB detected low dissolved oxygen and stressed benthic communities in areas of high organic loading (Howes et al., 2006). Water quality variables could be important drivers of benthic community structure in CCNS, but this study was unable to test those associations given the limited temporal duration of the data collection.

1.2. Methods

1.2.1. Vessel-based Acoustic Surveys

The research vessel R/V Marindin was used to collect acoustic data (figure 1.3). The R/V Marindin is a 1995 Eastern® I/O with a 300 HP Mercrusier™ engine. The vessel 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 vessel also has a diver ladder and a davit. The requisite safety equipment onboard includes radar, depth-sounder and GPS and compass for navigation.

A suite of instruments is required to conduct high-resolution, vessel-based acoustic surveys. The Edgetech 6205 is a dual-frequency, phase-measuring sidescan sonar and was used for all surveys. Its operating frequencies are 550 and 1600 kHz for backscatter imagery and 550 kHz for bathymetry. The sidescan sonar range resolution is 1 cm, and the horizontal beamwidth is 0.5 degrees at 550 kHz. The corresponding quantities at 1600 kHz are 0.6 cm and 0.2 deg. The bathymetric range and vertical resolution are both 1 cm. Use of chirp signals and correlation processing has enabled the stated range resolutions. The respective bandwidths at 550 and 1600 kHz are 67 and 145 kHz (Edgetech, 2014). The effective bathymetric swath width is 6-8 times the height of the sonar over the bottom. A Teledyne TSS DMS-05 Motion Reference Unit mounted on the sonar collects 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 is used to measure heading. Two differential GPS receivers spaced 2 m apart yield heading accuracies of <0.10° RMS (HemisphereGPS, 2009). A Trimble® R8 GNSS receiver utilizing Real-Time-Kinematic GPS (RTK-GPS) is used for positioning and tide correction for vessel-based surveys.

CCS subscribes to a proprietary Virtual Reference Station network (KeyNetGPS) that provides virtual base stations via cellphone from southern Maine to Virginia. This allows CCS to collect RTK-GPS without the need to setup a terrestrial base station or post-process the GPS data, thereby reducing mobilization and demobilization 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, with published state plane coordinate values relating to the Massachusetts Coordinate System, Mainland Zone (horizontal: NAD83; vertical NAVD88), were occupied. Control points were distributed over a wide geographic area up to 50 km away from CCS.

Figure 1.3. The R/V Marindin, a modified Eastern™, 8.2 m LOA, with a 2.6 m beam and a 0.61 m draft. The Edgetech 6205 is pictured on the custom-designed and built mounting gear

Multiple observation sessions, or occupations, were conducted at each control point with occupations of 1 second, 90 second, and 900 second. To minimize potential initialization error, the unit was shut down at the end of each session and re-initialized prior to the beginning of the next session. The results of each session (i.e., each 1 s, 90 s, and 900 s occupation) were averaged to obtain final x, y, and z values to further evaluate the accuracy of short-term occupation. Survey results from each station for each respective time period were then compared with published NGS and MassDOT values and the differences (error) used to assess and quantify uncertainty. Significantly, there was little difference between the error obtained for the 1 s, 90 s, and 900 s occupations. 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® 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 as Geotiffs.

Post-processing of bathymetric data was performed using CARIS HIPS®. Raw HSX files were converted to CARIS HDCS 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 YSI Castaway® CTD. Patch tests were performed 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. Real-time uncertainty data collected in Discover Bathymetric and stored in JSF format were not supported in CARIS, and therefore not utilized at the time of processing. However, Total Propagated Uncertainty (TPU) was computed using device manufacturer specifications recorded in the vessel file. Select filters were applied to the bathymetric data in order to remove noise in the far-field regions and depth outliers. When necessary, area editors were used to manually remove spurious soundings.

Other outliers and spurious soundings in interim bathymetric surfaces were identified in Fledermaus® and ESRI® ArcGIS v10.x. The utilization of multiple software packages to visualize errors and potential spurious soundings is ideal, as each software may visually represent the surface differently. Interim surfaces were displayed with high vertical exaggerations in ArcGIS and Fledermaus and inconstancies in the surface were identified via manual rotation and 'fly-throughs' around the surface as well as using the "Slope" tool in ArcGIS v10.3 to identify potentially problematic changes in slope. Each of these potential errors was compared to co-registered sidescan sonar mosaics and individual sonar lines in SonarWiz® v.5. If evidence of a genuine reflector was identified in the backscatter imagery, it was not removed, if the object could not be found and was determined to be an artifact of the survey and/or water conditions, then those soundings were removed. ESRI® ArcGIS v.10.x was used for visualization, two-dimensional and three-dimensional spatial analysis and to produce maps and figures.

Final surfaces were created from the processed sounding data using the CUBE (Combined Uncertainty Bathymetry Estimator) algorithm (Calder, et al., 2006), and were exported to multiple formats including Geotiffs, Bathymetric Attribute Grids (BAGs) and ASCII files.

1.2.2. Benthic Sampling

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 Pleasant Bay, benthic survey stations were determined 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.

In the field, random stations were located using a Trimble® R8, when on station, the boat was anchored before samples were collected. All samples were collected aboard the R/V Marindin and the Trimble® R8 was used to record boat position for each sample location. Each waypoint recorded was labeled by date, system name, station name and sample number. All GPS data points were downloaded to a .csv file and uploaded into ArcGIS for subsequent mapping.

1.2.2.1. Field work

Biological Samples

At each benthic survey station, four replicate grab samples were collected from the seafloor, three biological replicate grab samples and onesediment grab sample using a Young-Modified Van Veen grab sampler (figure 1.4). A GoPro™ Hero 3 was attached to the sampler, and high-resolution video was collected for each sample to aid in bottom characterization and documentation. The video was of sufficient resolution that still grabs could be obtained for imagery related needs. 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^2 to a depth of 0.1 m below the seafloor for a total volume of 0.004 m^3 (4 liters). This instrument is well-suited for sand- to mud-sized samples (≤2 mm) but does not sample well in areas with coarser grain sizes. A successful sample was attained when the two scoops of the Van Veen were fully closed, at least 2 liters of material were sampled, and the surface of the sample was level (i.e. the Van Veen did not sample the seafloor at an angle). 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.

Figure 1. 4. Young-Modified Van Veen grab sampler. C.G. Kennedy and Dr. M. Tyrrell Pictured onboard the R/V Marindin

For each grab sample, a photograph of the substrate surface was taken upon the sampler returning to the ship (the tops of the Van Veen open to facilitate inspection and analysis of the bottom grab sample), 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. Previous studies in the similar habitats have shown few additional organisms were retained onto a 0.5 mm mesh sieve (Fox et al. 2009) and pilot testing in 2014 revealed the same result (Fox pers. obs.). 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 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 70% ethanol until processing and analysis. Any seawater used for sieving or rinsing the sample was first pumped into large buckets and visually inspected before being used to reduce the chance of pelagic animals accidentally being introduced into the sample.

Water column data was collected using a YSI 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 replicate samples taken at each station, a fourth sample was taken to characterize the sediment. This sample was taken between the first and second biological replicate to ensure that the sediment sample was generally representative of the substrate sampled for the biological replicates. The surface sediment was transferred to a 100ml Whirl-Pak® using a stainless-steel spoon, stored on ice, and later frozen at the lab for future analysis.

1.2.2.2. Sample Processing

Biological Samples

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 (final concentration approximately 70%) and Rose Bengal to dye invertebrates. To process the samples, the ethanol was drained from the sample andthe sample was gently rinsed to remove any remaining preservative and then spread out into a large white plastic pan to which water was added. The sample was visually inspected, and all invertebrates were picked and sorted into general categories as could be discerned by the unaided eye (i.e. worms, shellfish, amphipods etc.). All personnel were trained by the project biologist on proper picking technique and on general visual cues to recognize invertebrates. Each sample was double-checked by a different person to ensure that all invertebrates had been found. Sorted specimens were stored for future identification. All invertebrates for each sample were preserved in 70% ethanol and stored for further identification and enumeration.

Invertebrate specimens were identified by the project biologist or trained personnel using dissecting microscopes. Specimens were identified to species when possible or to genus, families, or orders depending on the difficulty of identification, and enumerated. A voucher sample was labeled and recorded as a representative example of a particular species. All identified specimens were counted. Pictures were taken of voucher specimen, anatomical features of various specimen and for later identification and/or confirmation when necessary, using a digital microscope camera.

Sediment Samples

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 the sample. After the 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.

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

M_{dry} is the weight of the dried sample (at 105° C) plus the aluminum dish
M_{ignite} is the weight of the ignited sample (at 550° C) plus the aluminum dish
M_{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 by dividing the weight of the > 2mm fraction by the sample's total dry weight as follows:

Calculation of Percent Gravel (> 2mm):

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

$M_{>2mm}$ = weight of ignited >2mm substrate (including shells) plus the aluminum dish
M_{shell} = weight of shells manually removed from >2mm substrate plus aluminum dish
M_{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. After sitting overnight, the samples were centrifuged at 2,200 rpm for six minutes and the water was decanted two times. Prepared 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).

1.2.3. 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 1.5). Sound waves reflect from the boundaries between materials with different acoustic impedances, 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' (figure 1.5).

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 <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 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.

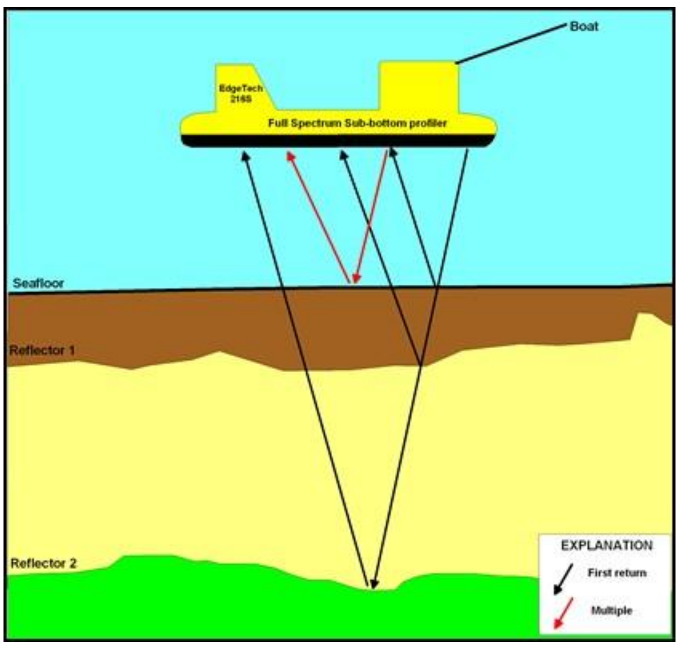

Figure 1. 5. Schematic view of the seismic reflection profiler used in this study

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 1970's, 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).

1.2.4. Sediment Core Sampling

Sampling locations were selected based on results from the seismic reflection profiling surveys. We selected locations with relatively thick sequences of fine-grained marine mud (figure 1.6). We utilized Kullenberg piston gravity cores to sample the undisturbed upper sediment and a Livingstone square rod piston corer to sample to refusal. We interpreted refusal as the ravinement surface known to be present below the Holocene unit. Composite core records were produced utilizing the upper Kullenberg and lower Livingstone sections, correlated by stratigraphy and physical properties.

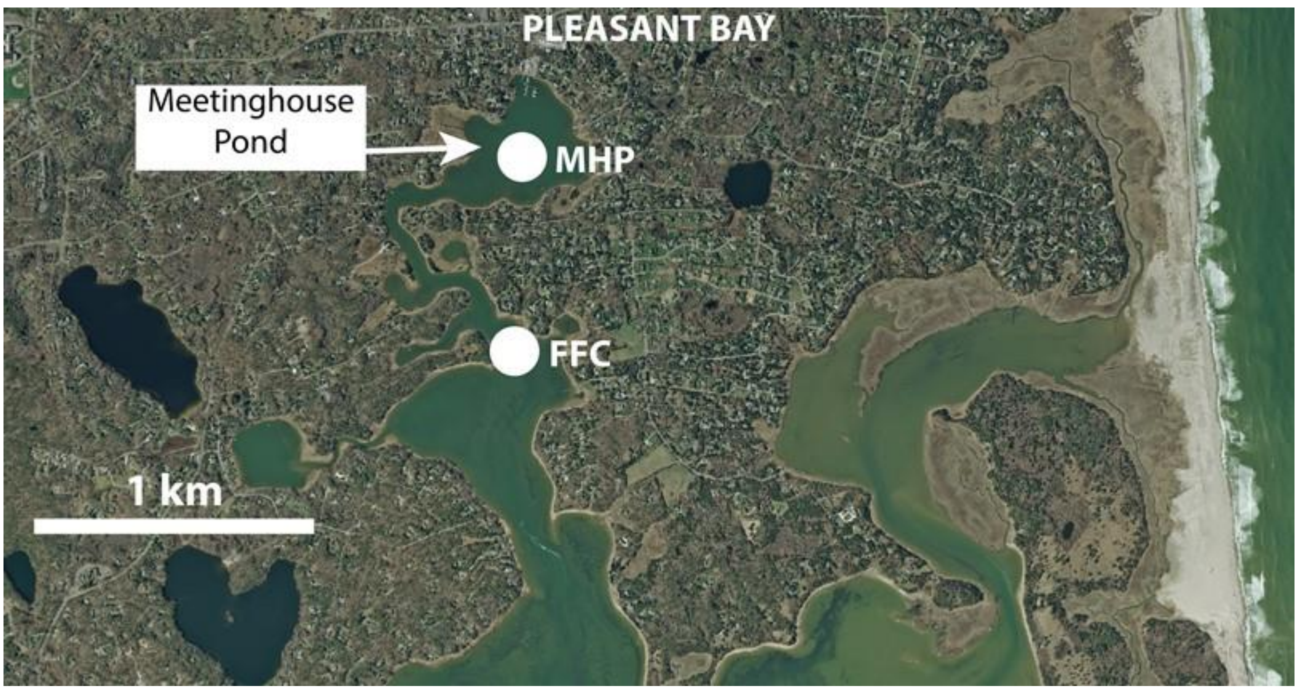

Figure 1. 6. Locus map of coring locations (white circles) sampled in this study. Meetinghouse Pond (MHP) and Frost Fish Cove (FFC) within northern Pleasant Bay

Cores were stored at 4°C after recovery. For analysis and subsampling cores were split horizontally, imaged, and lithologies were described. Volume magnetic susceptibility (κ) was measured with a Bartington MS2E high resolution sensor. Measurements were taken at 1cm resolution down the split face of one half each core, while zeroing the sensor between each measurement, the other half of the core was stored for possible future analysis. Wet and dry bulk densities were determined by massing 1 cm^3 subsamples both wet and after freeze drying.

A continuous flow elemental analyzer/stable isotope ratio mass spectrometer (EA/IRMS) was used to quantify stable carbon ($\delta^{13}C$), nitrogen ($\delta^{15}N$), and sulfur ($\delta^{34}S$) isotope ratios and elemental concentrations for discreet samples from composite cores. Two rounds of analyses were conducted in order to avoid complications of N analysis associated with acidification techniques to remove inorganic carbon necessary for $\delta^{13}C$ analyses (Brodie et al., 2011). For $\delta^{15}N$ and $\delta^{34}S$ analyses dry homogenized sediment was weighed into tin capsules. The $\delta^{13}C$ samples (dry homogenized sediment) were acidified through fumigation for six hours in silver capsules (Harris et al., 2001). After drying, samples and silver capsules were wrapped in tin capsules, and 3 times the sample weight of tungsten trioxide was added to assist in sample combustion. Standard reference samples (USGS-40, USGS-41, IAEA S-2, and IAEA S-3) were measured for calibration, and isotopic composition of all samples were expressed in standard format relative to Vienna Pee Dee Belemnite (VPDB), atmospheric air (AIR), and Vienna Cafion Diablo Troilite (VCDT), respectively. Ratios of OC/N and OC/S were calculated from element concentration data.

1.2.5. Submerged marine habitat mapping framework

This section describes the data analysis and data classification steps that were used to develop submerged marine habitat maps from the data described above. The data were analyzed to identify relationships among surficial acoustic data products, sediment characteristics, and biological data that could be used to predict biotopes, or combinations of abiotic habitat and associated species. Because the biotopes identified in this study are based on a single set of observations for each study area, these results are referenced as “preliminary biotopes”. Preliminary biotopes give us an indication of which physical variables are influencing, or driving benthic community composition in each study area. The goal of the data classification was to translate the acoustic data, sediment characteristics, benthic sampling data, and biotope data products into maps representing the CMECS Geoform, Substrate, and Biotic Components.

In the following subsections, are descriptions of how the results of the data analysis were fed directly into the CMECS classifications. There are numerous analysis methodologies available to segment acoustic data, sediment characteristics, and biological data into classifiable units (see Brown et al. 2011 for a review). The approaches and methods used for this study represent only one set of options, chosen because of their previous application in similar environments (e.g., Shumchenia and King, 2010).

1.2.5.1. 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^2, whereas Level 2

Geoforms are generally smaller. Level 1 and Level 2 Geoforms were delineated by classifying several metrics derived from the bathymetry grid 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,

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

Where *scalefactor* = out 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 $<1km^2$ 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 and an outer radius = 250. Fine-scale BPI was calculated using an inner radius = 5 and an outer radius = 25. 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 1.1) 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. 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 1.1).

Table 1.1. 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 the median grain size at each sampling point was interpolated using interpolation with barriers. A relatively simple kernel smoothing method was employed, which interpolated median grain size, bounded by a polygon of that area. The model bandwidth of 4000 m was adjusted for each variable to minimize local root mean square error. 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. To display uncertainty in the interpolated maps, the standard error of each interpolation was plotted in the same units as the interpolated variable (microns).

Summary of physical characteristics

Summary statistics for all physical characteristics were calculated, including backscatter imagery, grain size, and organic content within each CMECS Geoform.

1.2.5.2. Biological Characteristics

CMECS Biotic Component

The CMECS Biotic Component deals with the classification of organisms in both the water column and on the seafloor; in this study, only organisms on the seafloor were included (i.e., CMECS Biotic Setting = Benthic Biota). The scope of classification was refined to the Biotic Class "Faunal bed," since all of the observations 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. The Biotic Communities for this study were defined based on dominance at each sample location (as per CMECS Technical Guidance Document 2014), and then described by the appropriate Biotic Group and Class for each Community.

Biotic Communities were defined by cluster analysis of the benthic infauna 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 krCluster method was 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 (CMECS Technical Guidance Document 2014).

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 1.7). 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.

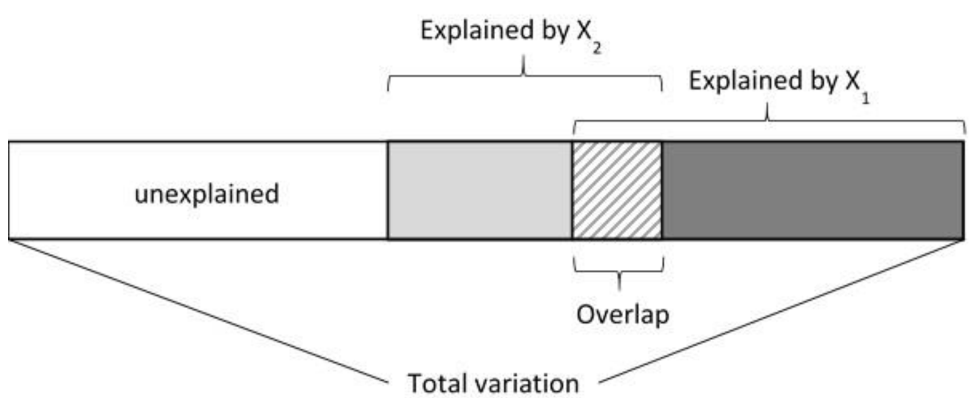

Figure 1.7. Conceptual diagram of regression as a partitioning of the total variation into portions that are explained by the predictor variables (X_1 and X_2), a portion that can be explained by both variables (overlap) and a portion that is left unexplained. (Clarke and Warwick, 2001)

The predictor variables used for this analysis were 11 sediment characteristics listed in table 1.2. Grain size metrics were chosen in particular, because they were consistently associated with benthic invertebrate sampling stations. However, kurtosis was excluded from the model as it represents a purely statistical feature and has no consequences on describing actual sediment samples collected 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.

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.

Table 1.2. Grain size metrics used in biotope analysis in Pleasant Bay

Grain size metrics
% clay
% silt
% sand
% gravel
Organic content (% weight)
Mean
Median
Mode
Standard deviation
Skewness
(Kurtosis)

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 Aij is the proportion of the individuals of species i that are present in biotope j and Bij 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 significance level < 5% were reported.

1.3. Results

1.3.1. Vessel-based Acoustic Surveys

In Pleasant Bay and Chatham Harbor over 16.82 km^2 were mapped over 16 vessel-based surveys in 2014 (14 July - 04 December) with 844 survey lines with a total length of 439 km (table 1.3). The mean depth was 3.2 m with a maximum depth of 20.02 m. Traditional sidescan sonars are set by the operator to capture a portion of the ensonified seafloor, as a function of range from the port and starboard transducers. The default setting for all four projects was 50 m range, yielding a 100 m swath for backscatter imagery. These settings could change if operators were in water depths where a 100 m swath was not possible (shallow waters) or advisable (deeper waters) or if other survey conditions warranted. Conversely, bathymetric data is a function of water depth and therefore the area of seafloor mapped will vary depending on depth within the survey area. For this study a 6:1 - 8:1 ratio of swath width to water depth was typical. For example, in 3 m of water an 18 - 24 m swath of the seafloor was mapped. In 3 m of water the range for the backscatter imagery would be set to 50 m yielding a 100 m swath. Bathymetric data (6.78 km^2) collected within Pleasant Bay covered 40.0 % of the total area mapped as defined by the backscatter imagery (16.82 km^2) (Appendix A). All the backscatter imagery was collected with a minimum of 200% overlap and bathymetric coverage was incidental unless 100% bathymetric coverage was requested by park staff or deemed necessary by investigators. The areas mapped for bathymetry and backscatter imagery are derived from the final surfaces or mosaics, not individual survey lines and/or swaths.

Table 1.3 Results of vessel-based surveys from 2014 field season in Pleasant Bay.

Survey days	Survey lines	Survey Line Length (km)	Area Mapped SSS (km^2)	Area Mapped Bathy (km^2)	Mean depth (m)	Max depth (m)
16	844	439	16.82	6.78	3.20	20.02

1.3.2. Benthic Sampling

Between June 24th and August 1st, 2014, forty-eight stations within Pleasant Bay were sampled resulting in a total of 144 sieved and preserved biological samples (three replicates per station), 48 sediment samples, 48 water column profiles, photographic and video data at each station (table 1.4). During initial site selection 33 locations were chosen, and later 15 stations were added when the project was enlarged. The 15 stations were selected to overlap with benthic stations that were sampled by the Massachusetts Estuaries Project study conducted in 2003 (Howes et al., 2006) these stations were not included in the statistical analysis as at the data was not available to us at the time of writing of this report.

Table 1.4 Benthic mapping instruments, data products, and number of sites sampled by the Center for Coastal Studies in Pleasant Bay.

Instrument	Data	Resolution	Samples
Phase-measuring sidescan sonar	Bathymetry grid	1.0 m	-
	Sidescan sonar mosaic	0.5 m	-
Young-modified Van Veen grab sampler	Sediment particle size and distribution metrics	-	48
	Sediment organic content	-	48
	Benthic infauna abundance	-	33

1.3.3. Seismic Reflection Profiling

A total of 42 km of seismic reflection profiles were collected within Pleasant Bay (figure 1.8). Overall, the coarse (sand or gravel) surface sediment, natural gas in the subsurface or the presence of dense beds of submerged aquatic vegetation limited penetration of the seismic signal in portions of the study areas. 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 1.8. Locus map showing the extent of sub-bottom seismic reflection profiles in Pleasant Bay collected for this study (red lines).

1.3.3.1. Seismic Facies Identified

Facies GIM: Glacial Ice Marginal

Facies GIM is characterized by extremely hummocky, chaotic internal and external seismic reflectors with highly variable, often steep topographic relief (figure 1.9). This facies is interpreted to represent highly collapsed coarse grained (sand and gravel) glacial stratified deposits. This unit was mapped under portions of Chatham Harbor.

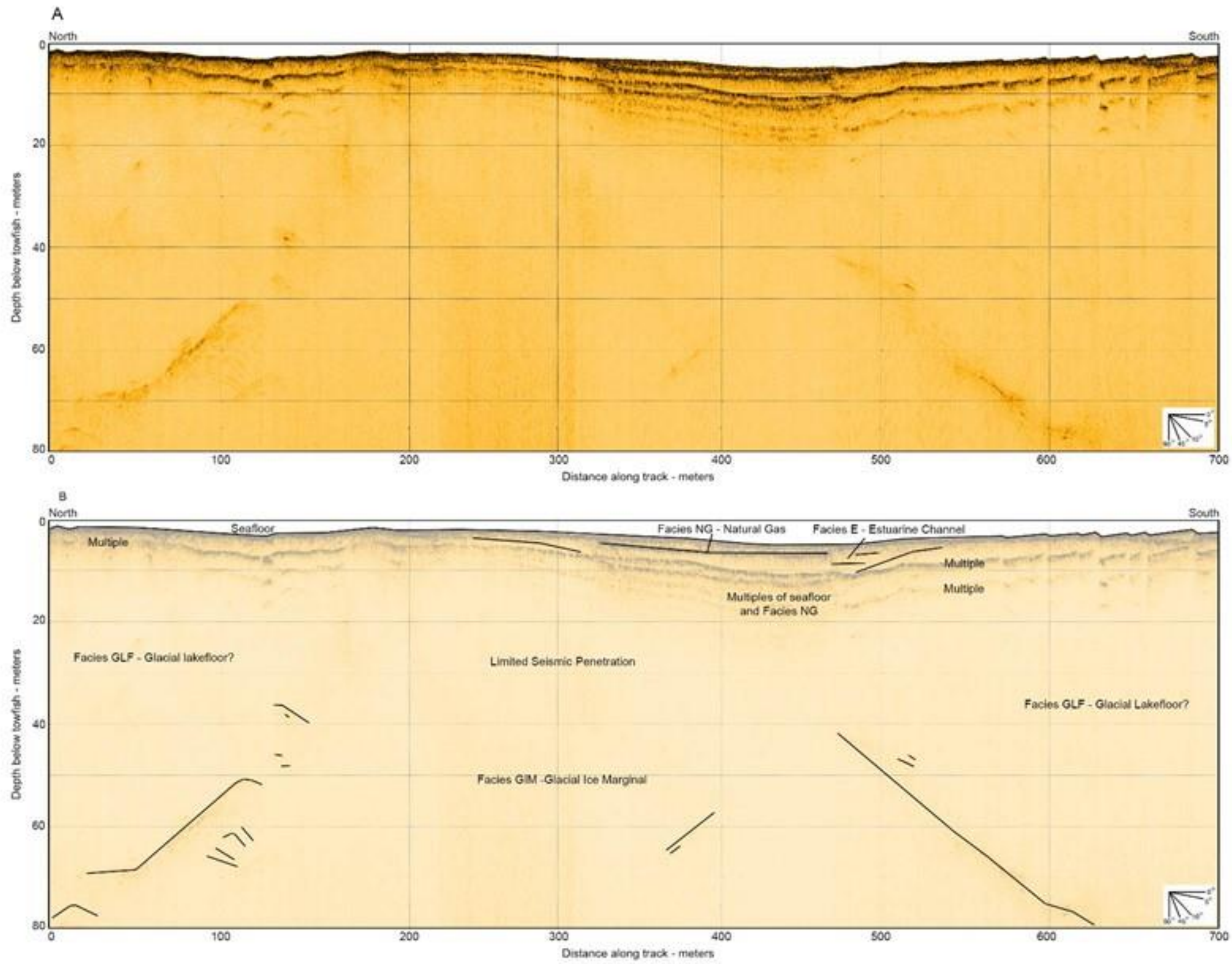

Figure 1.9. A: Sub-bottom seismic reflection profile from Chatham Harbor. B: Interpreted seismic reflection profile from Chatham Harbor showing facies NG, E, GIM and possible facies GLF

Facies GLF: Glacial Lakefloor

Facies GLF is characterized by parallel, laterally continuous reflectors that drape underlying topography (figure 1.9). 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. This facies was identified in Crows Pond, Ryders Cove and Round Cove.

Facies Glu: Glacial deposits – undifferentiated

Facies Glu is identified by a strong reflector, often with a hummocky, collapsed topography (figures 1.10 and 1.12). 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).

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 1.9). This unit is interpreted to represent post-glacial fluvial, spring sapping or tidal channels modified or formed during Holocene marine transgression.

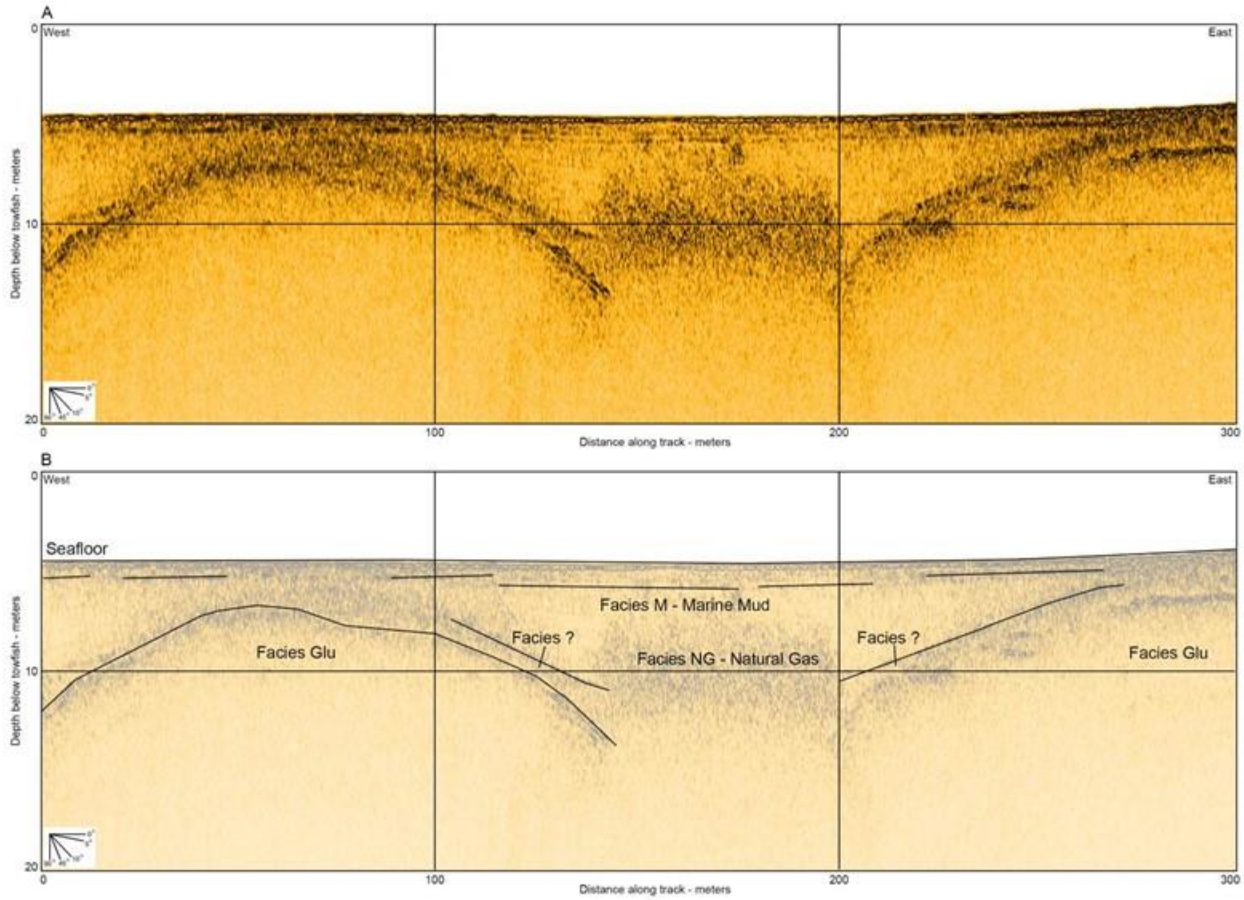

Figure 1.10. A: Sub-bottom seismic reflection profile from Crows Pond, Pleasant Bay. B: Interpreted seismic reflection profile from Crows Pond, Pleasant Bay showing facies NG, M and Glu. It remains unclear what the reflectors at the base of facies M (and coinciding with facies NG) represent.

Facies IC: Tidal Inlet/Channel deposits

Facies IC was mapped in portions of Chatham Harbor, and is characterized by a hard surface reflector, usually with obvious tidal bedforms (dunes) on the surface (figure 1.11). Seismic penetration was limited in this sandy depositional environment, however where penetrated it was 2 - 5 m thick, and appears to overlie stratified glacial deposits.

Facies NG: Natural Gas

Facies NG has a distinct seismic signature, with a dark, opaque upper seismic reflector that typically has a convex up reflection that obscures or 'wipes out' the underlying seismic record (figures 1.9, 1.10, 1.12). This facies is interpreted to represent gas bubbles in the sediment, and the gas is likely buried methane formed from decayed organic matter. The source of the organic matter is probably a combination of freshwater and saltwater marsh peat, and organic-rich (Holocene aged) marine sediment.

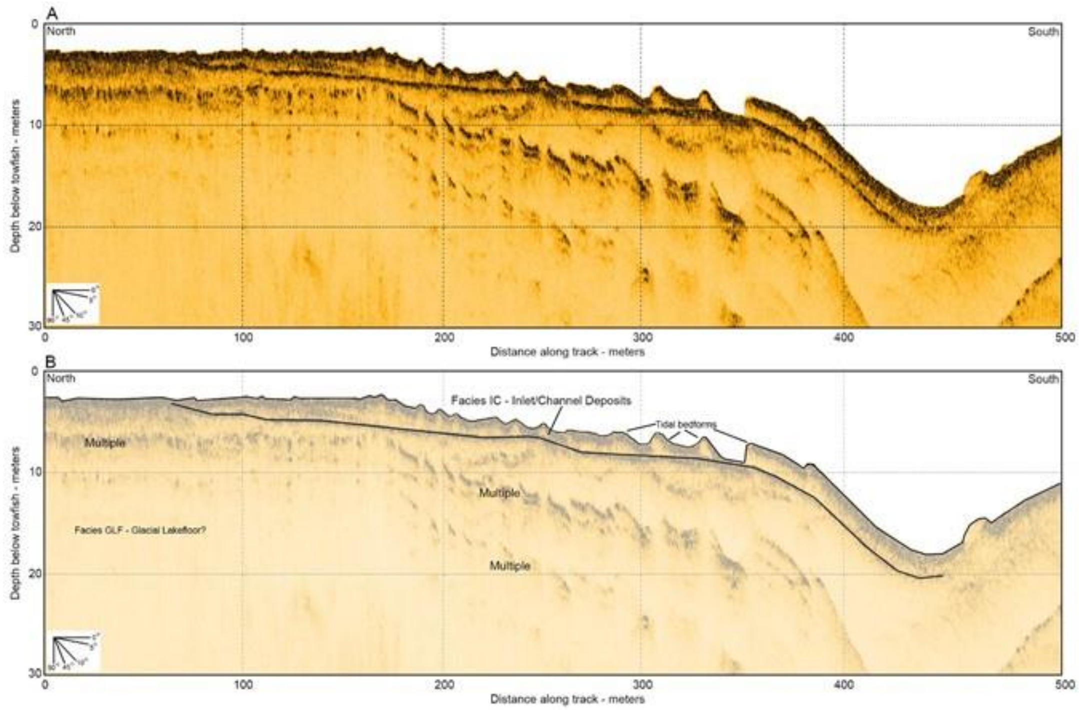

Figure 1.11. A. Sub-bottom seismic reflection profile from Chatham Harbor. B. Interpreted seismic reflection profile from Chatham Harbor showing facies IC

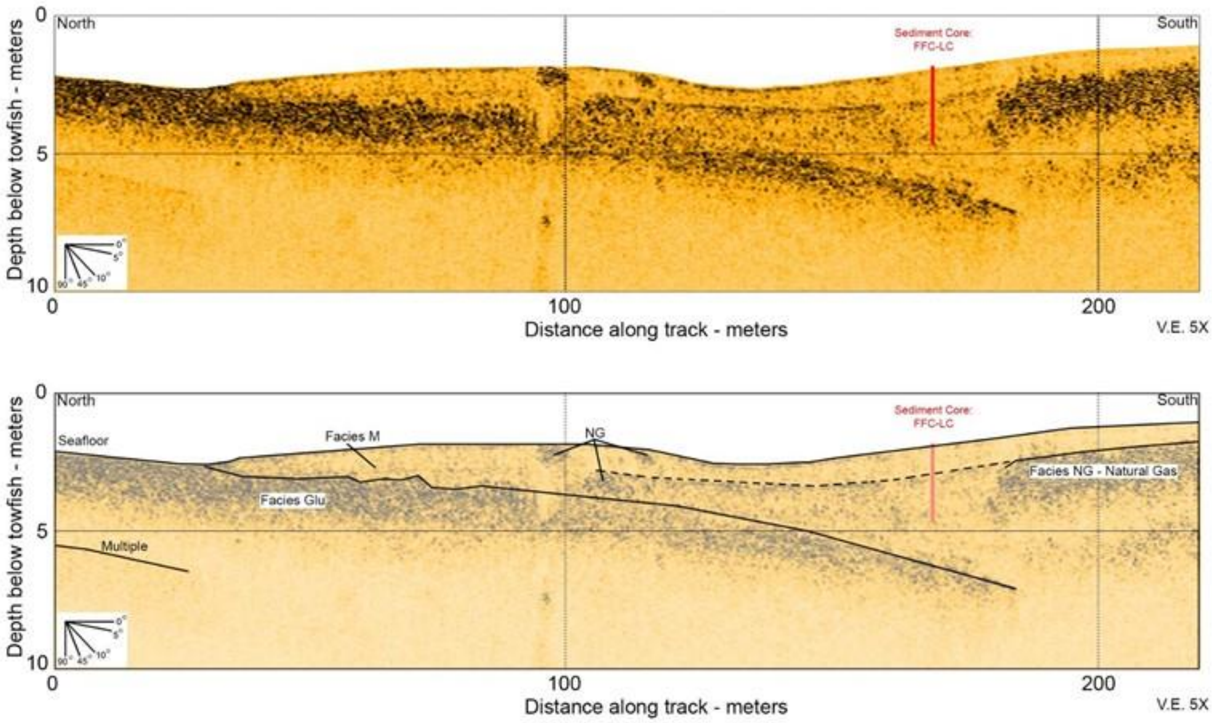

Figure 1.12. A. Sub-bottom seismic reflection profile from Frost Fish Cove, Pleasant Bay. The red line in both images represents the location and depth of sediment core FFC-LC (Love, et al., 2015). B. Interpreted seismic reflection profile from Frost Fish Cove, Pleasant Bay showing facies M, NG and Glu. Note the dashed line within Facies M coinciding with the depth of Facies NG and an increase in grainsize from silt above 82 cm to very fine sand below 82 cm (See figure 1.13)

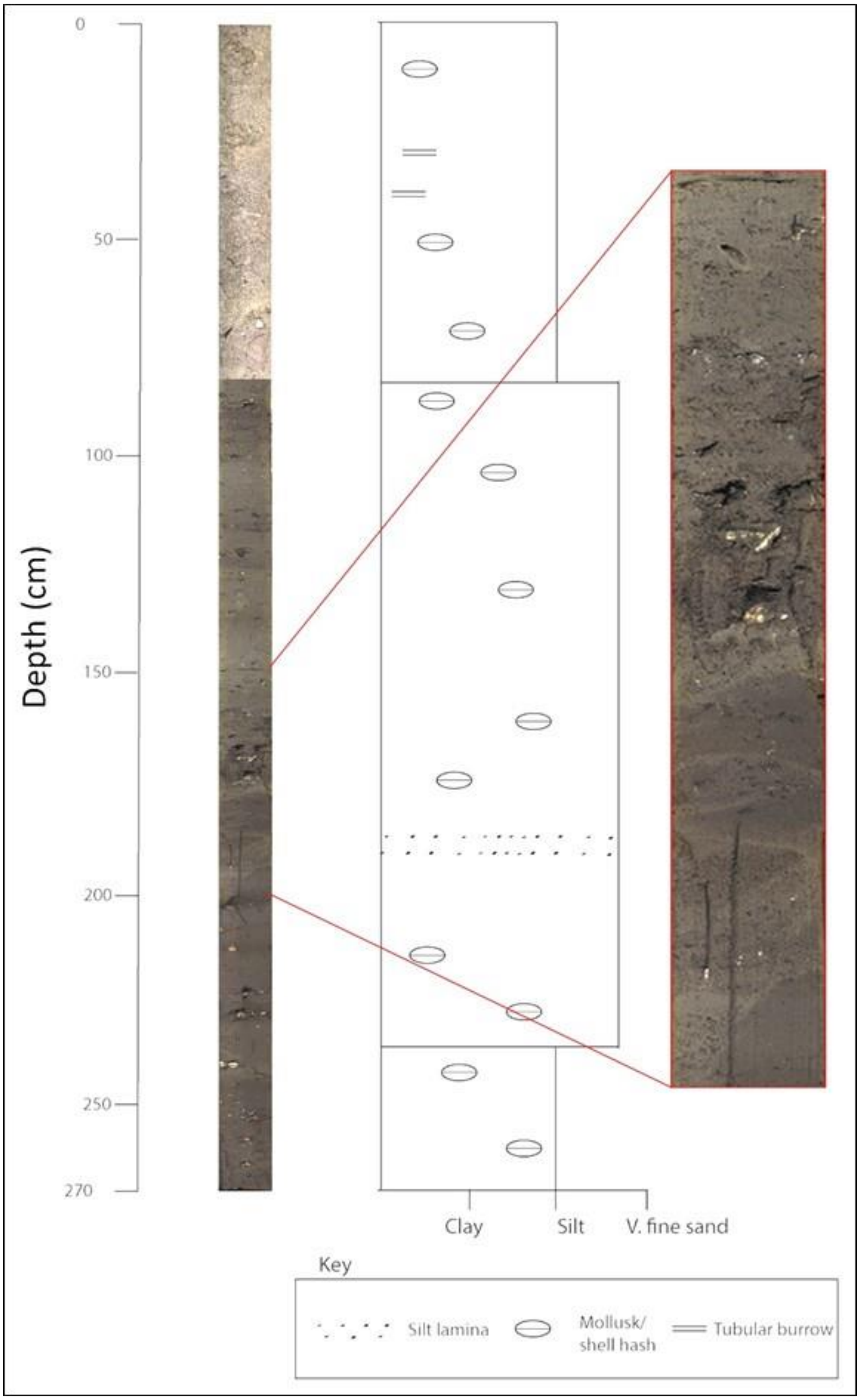

Figure 1.13. Core image and grainsize from the core from Frost Fish Cove (FFC). Image and data from Love et al., (2015). Note the discontinuity at 82 cm, and transition from silt to very fine sand, back to silt at 240 cm. The discontinuity at 82 cm coincides with the depth of natural gas and a slightly darker seismic reflector (figure 1.12). This relationship and implications on the depositional history bears further investigation

1.3.4. Sediment Core Sampling

Total sediment thickness of Holocene-aged sediment was found to be 2.84 m at Frost Fish Cove (figure 1.14), and 1.03 m at Meeting House Pond. Due to the short record recovered at Meeting House Pond, we opted to focus analyses on the Frost Fish Cove record for Pleasant Bay (table 1.5).

Table 1.5. Sediment core metadata. Kullenberg cores (KC) are gravity driven piston cores designed to sample the surface sediments while Livingstone cores (LC) are square rod piston cores utilized to gain maximum sediment penetration (to ravinement surface).

Core ID	Core Location	Water Depth (ft)	Sediment Acquisition (cm)
MHP KC1 Sec 1-2	41°46.778'N, 069°58.066'W	16.6	93
MHP LC1 Sec 1-2	41°46.772'N, 069°58.071'W	16.0	103
FFC KC1 Sec 1-2	41°46.408'N, 069°57.935'W	5.0	133
FFC LC1 Sec 1-3	41°46.411'N, 069°57.934'W	6.7	284

Composite chronologies were calculated for Frost Fish Cove (FFC) based on chronostratigraphic constraints. Since each core had a quality sediment/water interface, we assigned an age of 2014 CE (-64 cal BP) to the top of each core. The base of the anthropogenic magnetic susceptibility signal was assigned a date of 1880 CE (70 cal BP) (Santschi et al., 1984). Seven radiocarbon dates were obtained from the two locations (table 1.6). Dates were calibrated with IntCal13 (bulk sediment) or Marine13 (carbonate) (Reimer et al., 2013) and an age model was calculated using the Bacon Bayesian statistical program (Blaauw and Christen, 2011).

Table 1.6. Uncorrected radiocarbon ages from Frost Fish Cove (FFC). Age models with corrected radiocarbon ages and additional chronologic control are discussed below. All analyses conducted at DirectAMS.

Laboratory Code	Core location/depth (cm)	Material	Uncorrected Radiocarbon age (BP)	$\delta^{13}C$ (per mil)
D-AMS 015296	FFC/254	*M. mercenaria*	1775±29	0.4
D-AMS 015297	FFC/191	*L. littorea*	1558±24	-1.7
D-AMS 016304	FFC/227	*L. littorea*	1434±20	-3.0

1.3.4.1. Sediment Accumulation

The Frost Fish Cove age model is presented in figure 1.14. A substantial increase in sedimentation rate is noted in the upper portion of the record, interpreted here to represent anthropogenic sediment focusing to the depocenter. The calculated median basal age of the sediment is 1458 cal BP (492 CE). Since the core was taken to refusal, the base of the core is likely the base of the Holocene marine unit, directly overlying the ravinement surface. This interpretation suggests that marine waters transgressed to the Frost Fish Cove location within Pleasant Bay by 1458 cal BP.

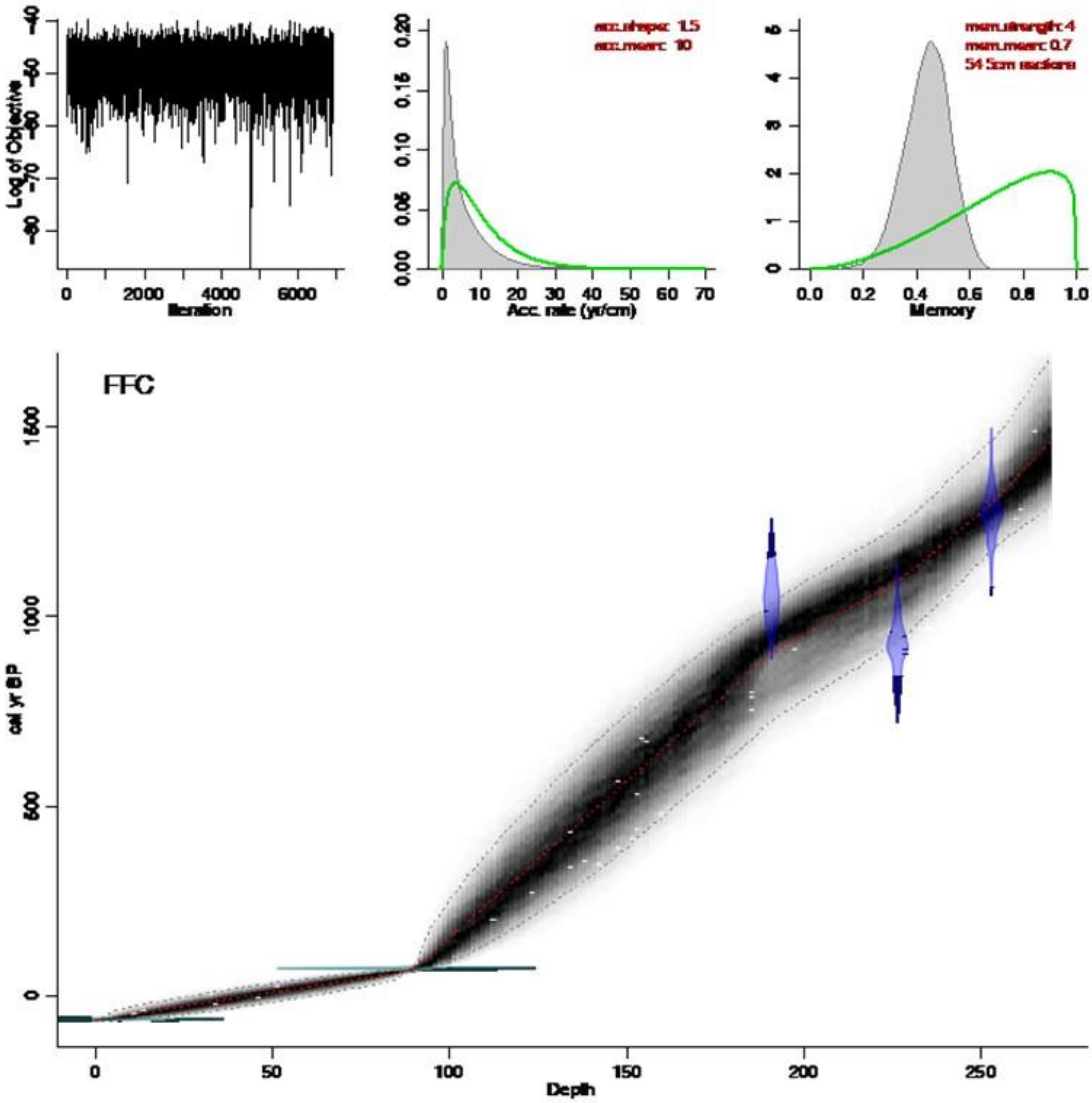

Figure 1.14. Sediment depth: age model for composite Frost Fish Cove (FFC) sediment record. Green controls represent the sediment surface (-64 cal BP) and the base of the anthropogenic magnetic susceptibility peak (70 cal BP). Blue controls are from radiocarbon dates calibrated with the IntCal13 and Marine13 calibration datasets (Reimer et al., 2013)

1.3.4.2. Sediment Stratigraphy

Sediments of the Frost Fish Cove sediment core (figure 1.14) contain abundant *L. littorea* and *M. mercenaria* shells throughout, and vertical burrowing is evident at depths from 30-40 cm (table 1.6). The upper 82 cm contain a dark brown silty mud that displays a sulfuric odor. A transition to a lighter brown, sandy mud occurs below to 230 cm. A gradual fining of grain size occurs here, and the lowest sediments are a massive silt. At 88 cm in depth, a significant (T-test: $p<0.0001$) rise in volume magnetic susceptibility occurs, marking the onset of the anthropogenic zone. Within this zone, volume magnetic susceptibility values average 5.77 *10-5 + 3.25 *10-5 SI, while preceding values average 0.88*10-5 SI. As in all locations in this study, consistently low susceptibility is indicative of the presence of diamagnetic organic content of the sediment, with little terrigenous input. Wet and dry bulk densities retain a consistent average of 1.32 g/cm^3 and 0.67 g/cm^3, however greater low frequency variability is observed below 154 cm.

1.3.4.3. Chemical Stratigraphy

At Frost Fish Cove proxies for organic production, %OC and $\delta^{13}C$, both display low frequency variability below depths of 180 cm. Averages here are 4.46‰ + 1.00 and -18.60 + 2.17‰, respectively. Values for these proxies from the sediment-water interface to 180 cm exhibit lower variability within a similar range in average values of 2.71 + 0.63‰ and -18.72 + 1.54‰. C/N ratios plotted against $\delta^{13}C$ (figure 1.15) identify marine phytoplankton as the dominant source of organic material. Influence of C3 and/or C4 land plants may be evident in slightly lower $\delta^{13}C$ values in this location. Values for $\delta^{15}N$ in Frost Fish Cove fall between 4-6‰, with no appreciable variation within the anthropogenic zone, and likely indicate that marine phytoplankton are a stable and dominant source of nitrogen. Total accumulation of both organic carbon and nitrogen increase in close proximity to the anthropogenic zone, driven by an overall increase in sedimentation. C/S ratios here express low variability throughout the core, although a noticeable, rapid decrease in sulfur in shallow sediments may be evidence of the introduction of more freshwater to the basin.

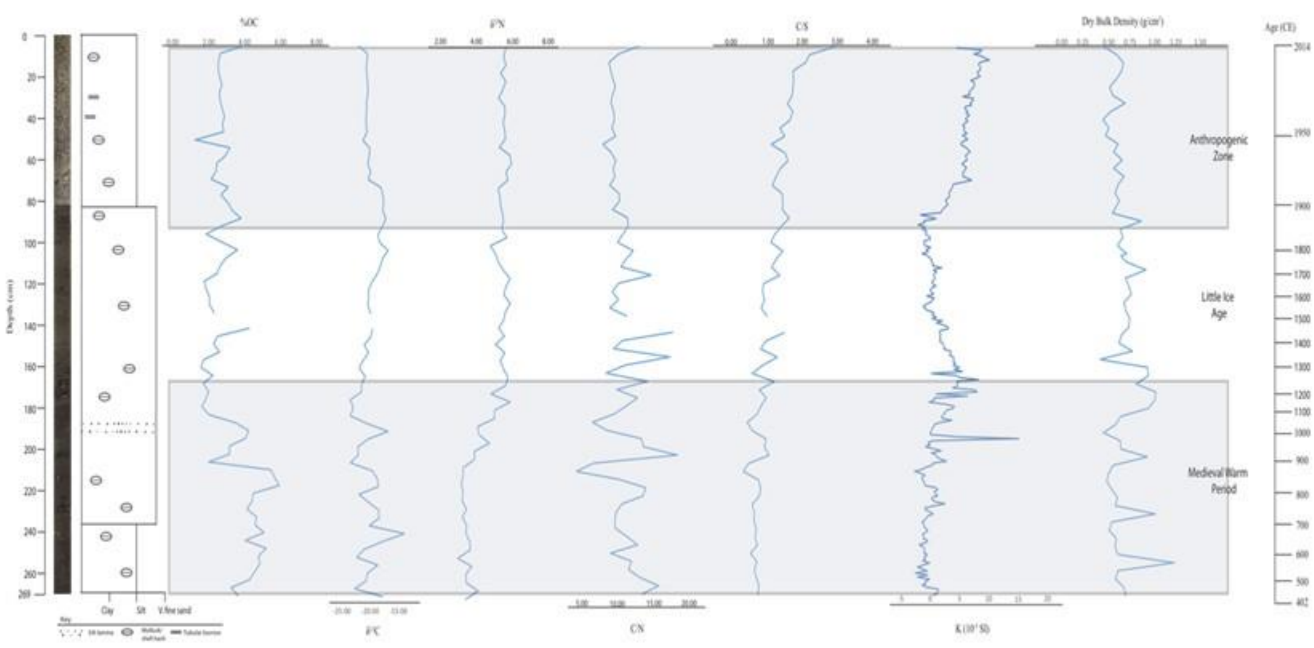

Figure 1.15. Late Holocene stratigraphy preserved at Frost Fish Cove, Pleasant Bay, Cape Cod

1.3.5. Submerged habitat mapping

1.3.5.1. Physical Characteristics

CMECS Geoforms

The distribution of CMECS Geoforms is shown below (figure 1.16). Indicator species were found for platforms, deeper flats and banks (table 1.7, figure 1.17).

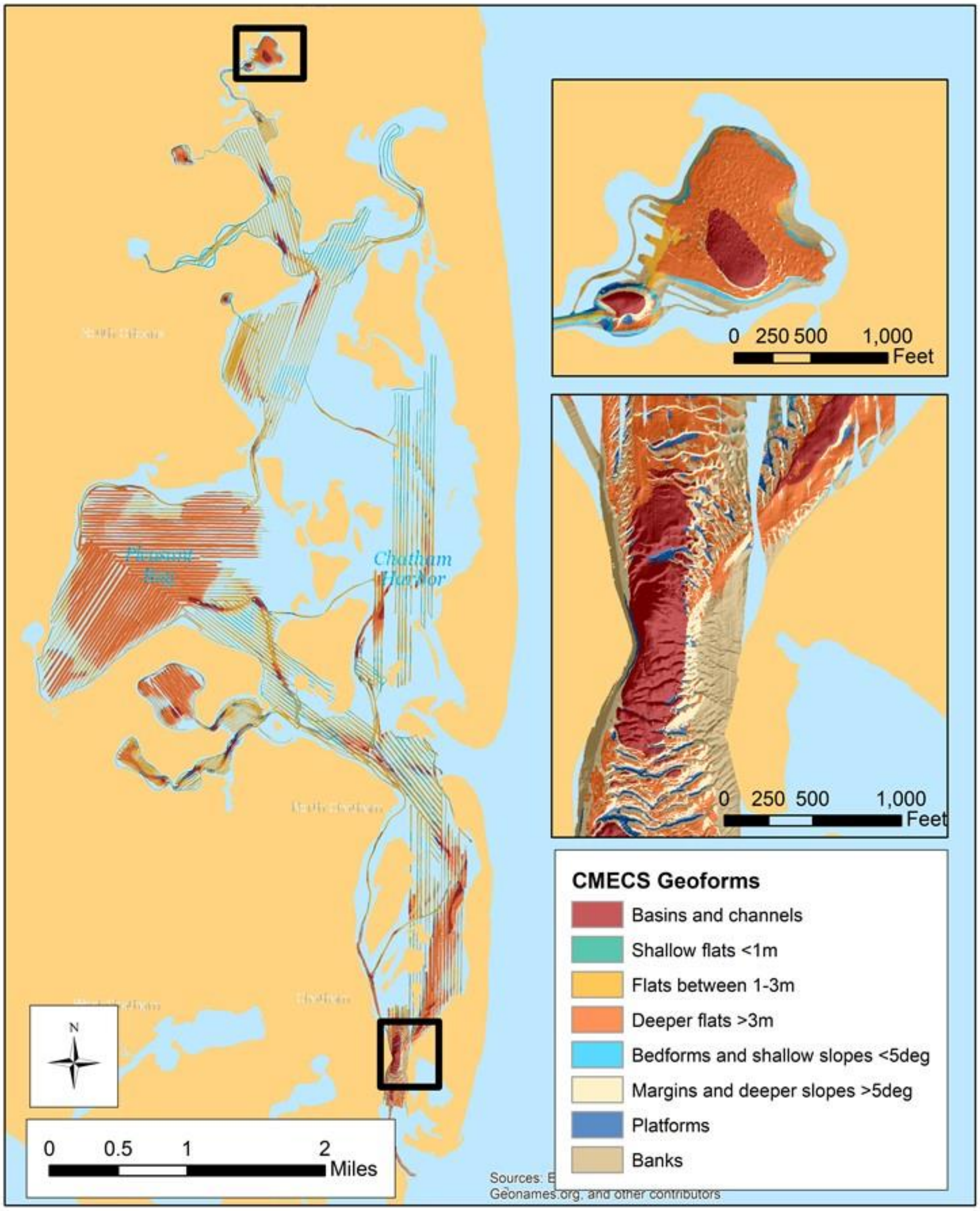

Figure 1.16. CMECS Geoforms for Pleasant Bay.

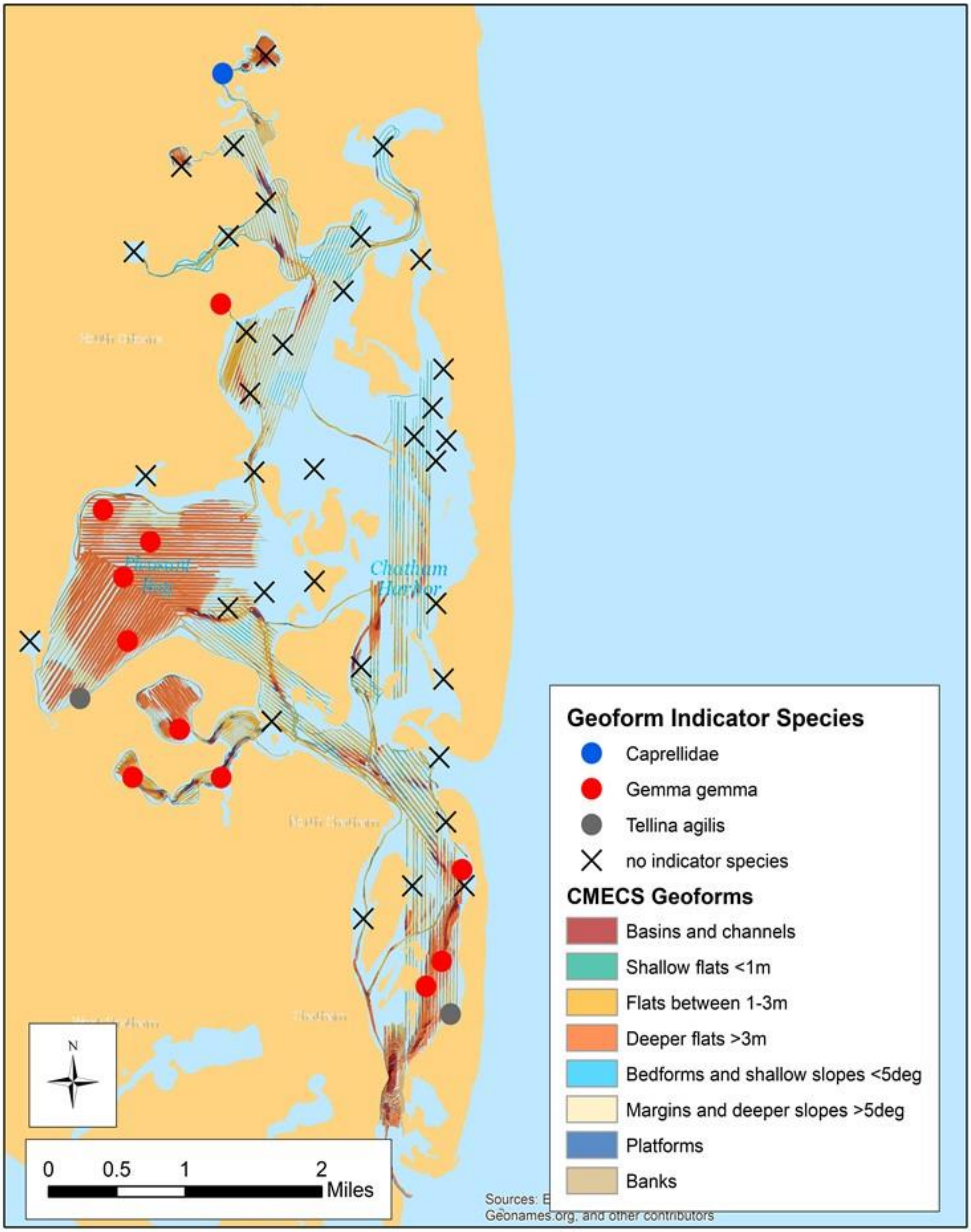

Figure 1.17. Significant indicator species for CMECS Geoforms in Pleasant Bay.

Table 1.7. Significant indicator species for Pleasant Bay CMECS Geoforms.

Geoform	Species	IndVal	Significance level (%)
Platforms	Caprellidae	0.287	4.2
Deeper flats (>3m)	*Gemma gemma*	0.363	4.3
Banks	*Tellina agilis*	0.260	4.2

CMECS Substrate

The CMECS Substrate Group and Subgroup classification as well as interpolations of median grain size for Pleasant Bay are shown below (figure 1.18). Interpolations (models) of median grain size within each area are shown below for PB (figure 1.19).

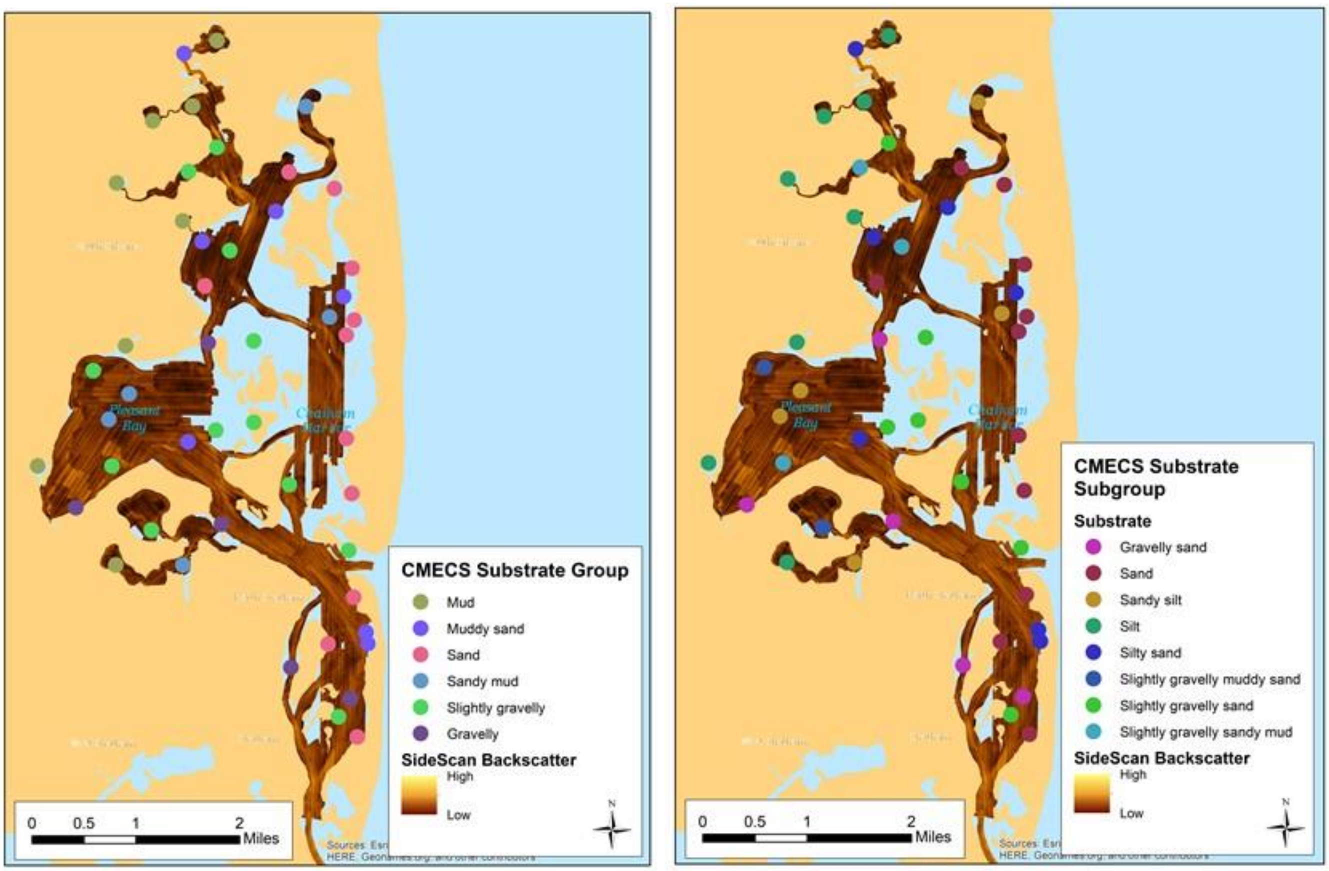

Figure 1.18. Left: CMECS Substrate Group for Pleasant Bay. Right: CMECS Substrate Subgroup for Pleasant Bay.

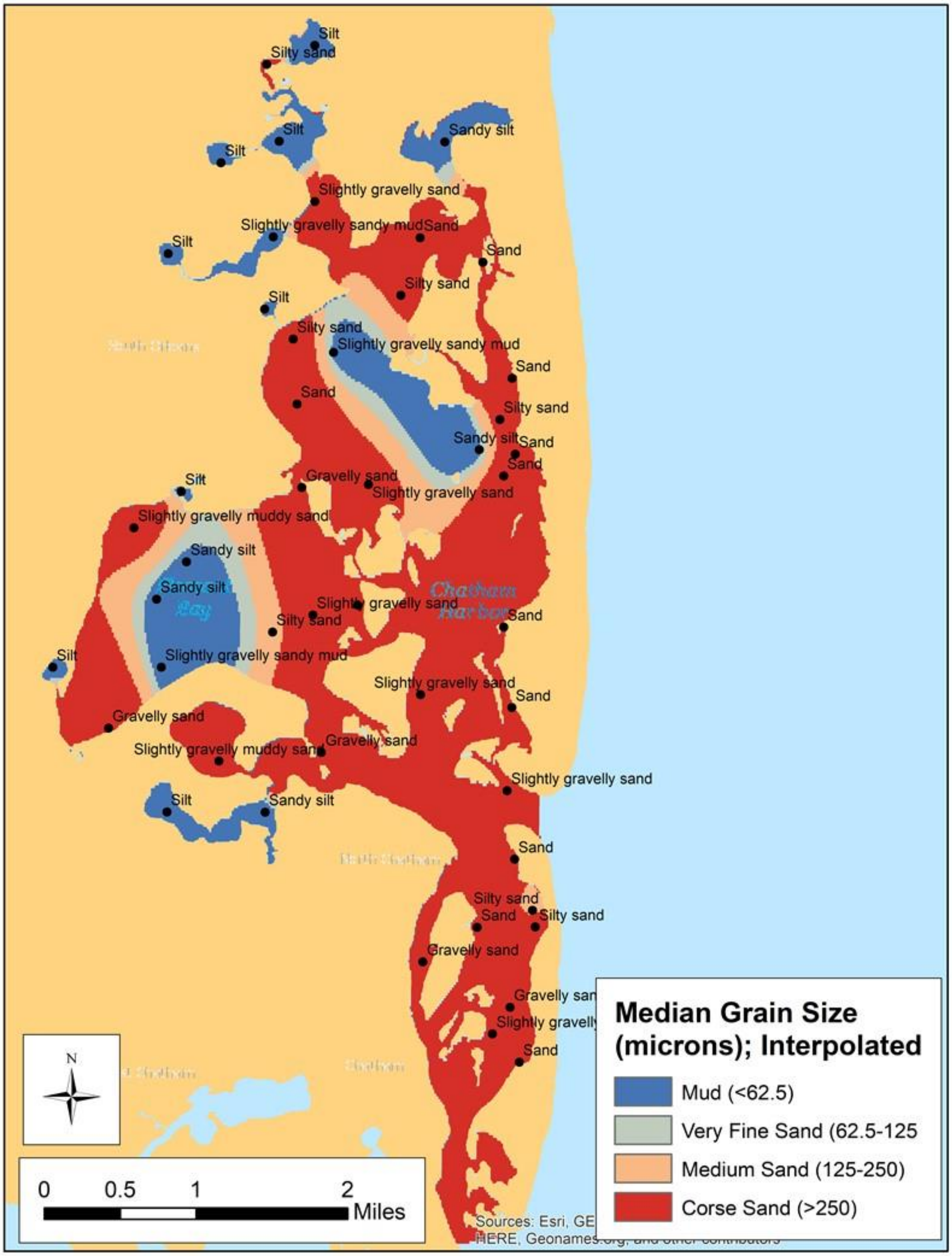

Figure 1.19. Median grain size in microns (interpolated) for Pleasant Bay. Predicted standard error for the interpolation is shown top right. Data points are labeled by CMECS Substrate Subgroup derived from the classification of weight percentages of gravel, sand, silt and clay

Summary of physical characteristics

Figure 1.20 summarizes selected characteristics of the physical variables within each CMECS Geoform in Pleasant Bay. 13 stations could not be assigned a CMECS geoform as they were inaccessible by boat and therefore are not included in this graph and could not be mapped.

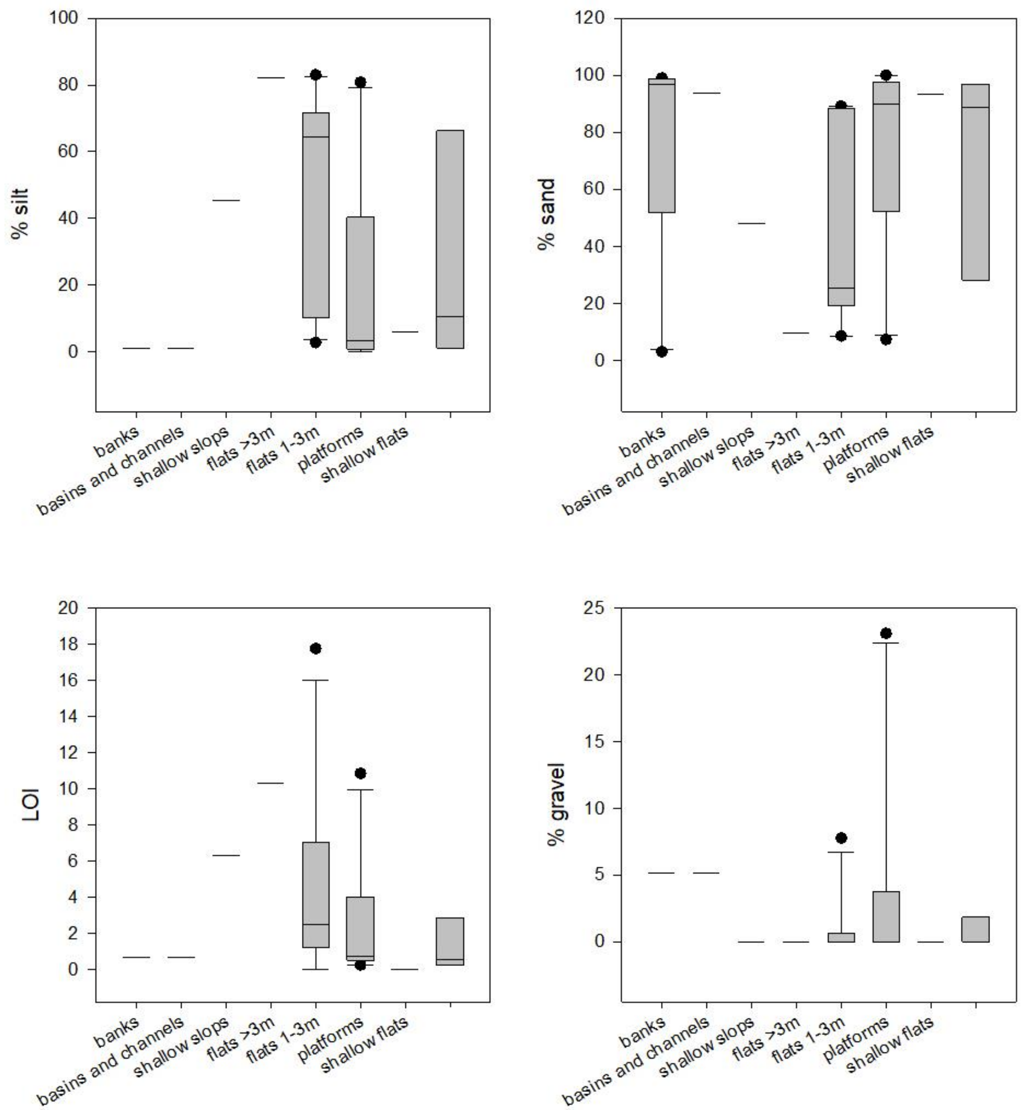

Figure 1.20. Box plots of physical variables in CMECS Geoforms for Pleasant Bay. The black bar in each box represents the median, and each bar is bounded by the lower and upper quartiles. The vertical size of the box is the interquartile range, or the general “spread” of the data. The top whisker denotes the maximum value or the third quartile plus 1.5 times the interquartile range, whichever is smaller. The bottom whisker denotes the minimum value or the first quartile plus 1.5 times the interquartile range, whichever is larger. Outliers are shown with black circles, and are defined as any value above and below 1.5 times the interquartile range.

1.3.5.2. Biological Characteristics

CMECS Biotic Component

In Pleasant Bay, 32 invertebrate species comprised the top 95% of all individuals. Different sets of benthic assemblages were found in each area. At PB, 31 infaunal species comprised the top 95% of all individuals (148 species total, 6 of which could not be identified further than order or class). PRIMER's krCluster indicated that the optimal number of clusters is 17 (figure 1.21). Classifying each significant cluster into CMECS Biotic Communities based on dominance yielded 12 Biotic Communities (table 1.8). A map of Pleasant Bay sampling locations coded according to CMECS Biotic Group are shown in figure 1.22.

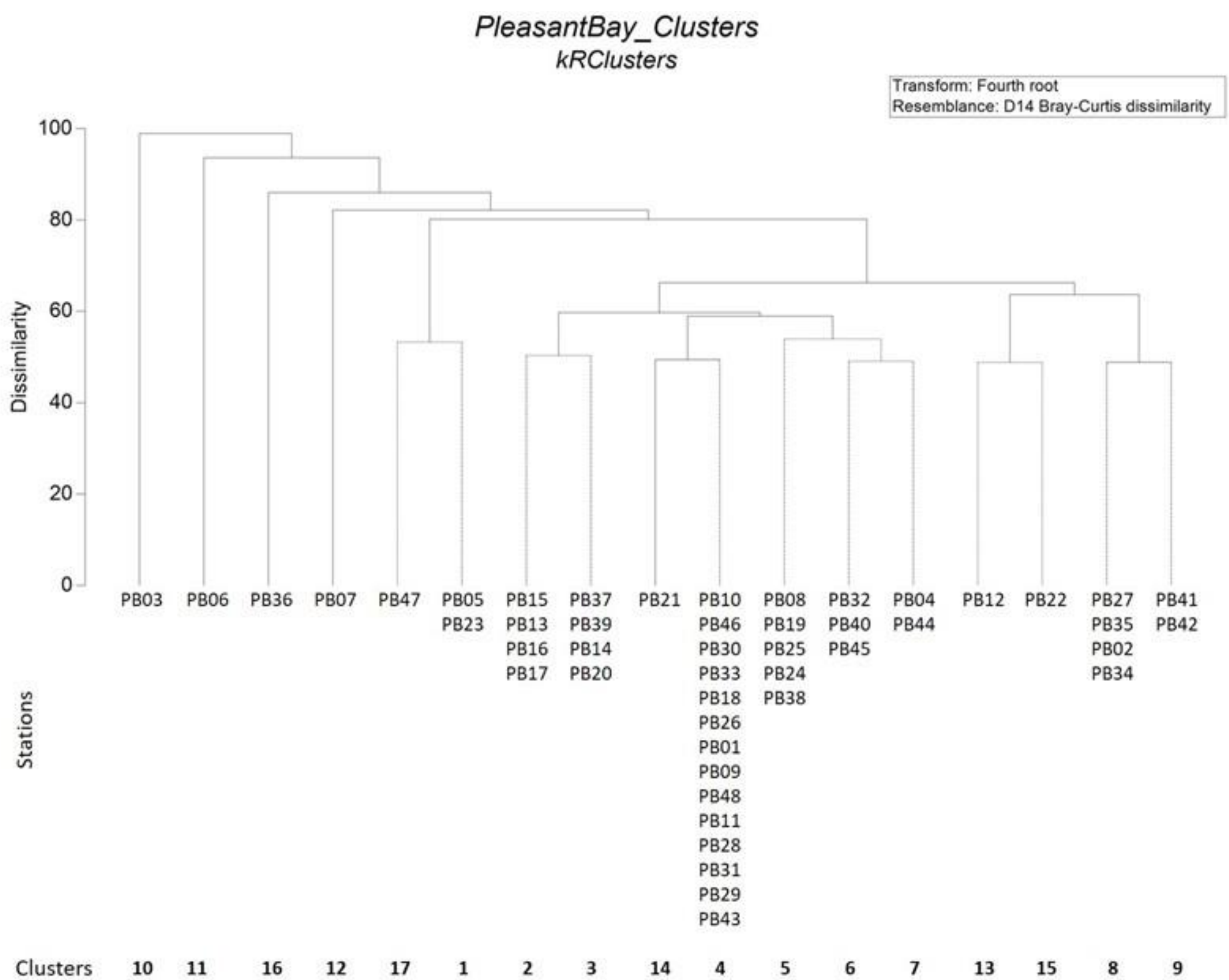

Figure 1.21. Cluster diagram showing 17 optimal clusters based on the abundance of species that accounted for 95% of the total abundance in Pleasant Bay.

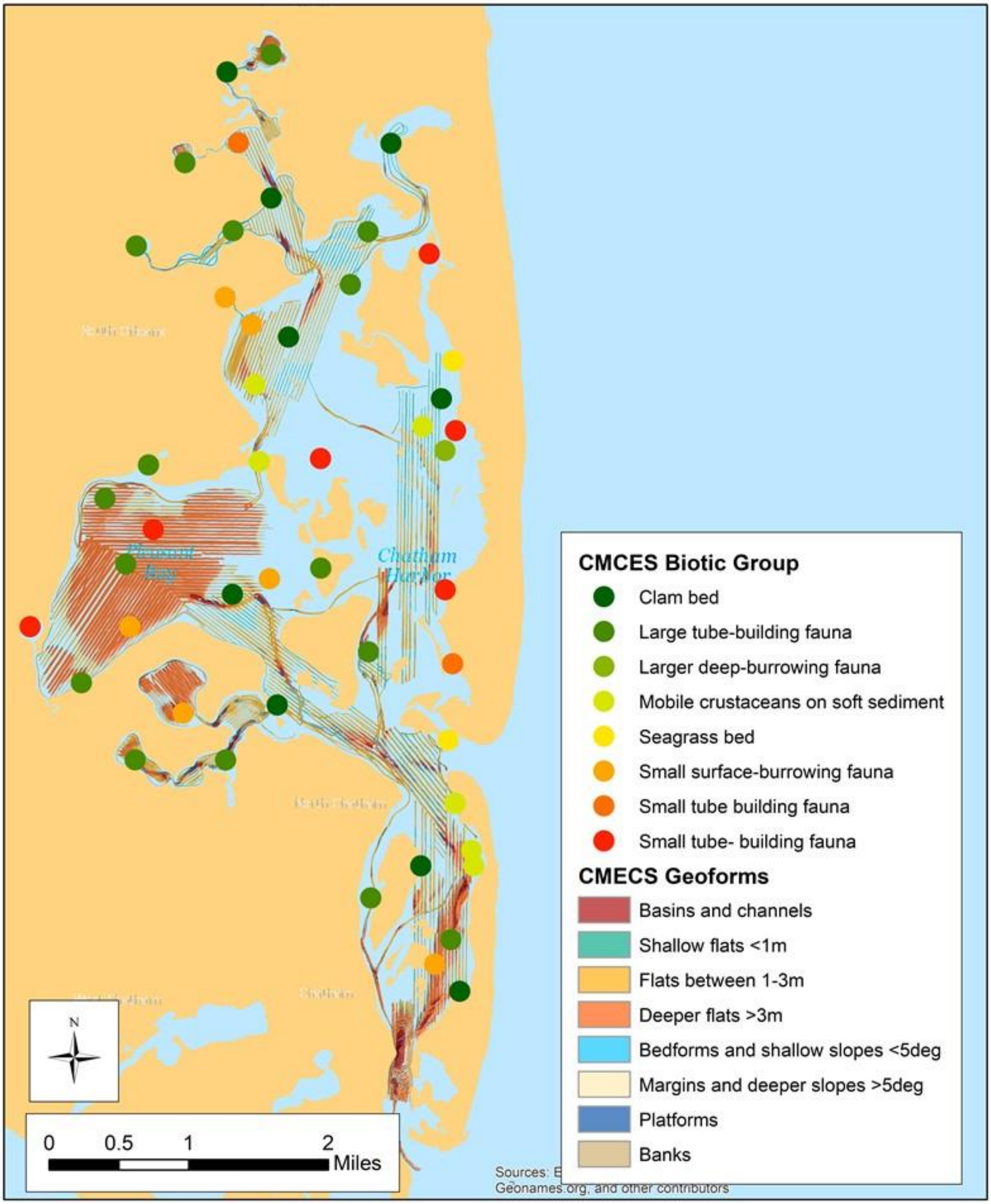

Figure 1.22. Pleasant Bay sampling locations coded according to CMECS biotic group

Table 1.8. CMECS Biotic Component classification for Pleasant Bay according to the cluster analysis.

Pleasant Bay cluster	Dominant species	CMECS Biotic Community	CMECS Biotic Group	CMECS Biotic Subclass
Cluster 1	*Ampelisca* spp	Ampelisca bed	Large tube-building fauna	Soft sediment fauna
Cluster 2	*Acteocina canaliculata*	Acteocina bed	Small surface-burrowing fauna	Soft sediment fauna
Cluster 3	Caprellidae*	Caprellid bed	Mobile crustaceans on soft sediment	Soft sediment fauna
Cluster 4	*Ampelisca* spp	Ampelisca bed	Large tube-building fauna	Soft sediment fauna
Cluster 5	*Streblospio benedicti*	Streblospio bed	Small tube- building fauna	Soft sediment fauna
Cluster 6	*Gemma gemma*	Gemma bed	Clam bed	Soft sediment fauna
Cluster 7	Cirratulidae*	Terebellid bed	Small tube building fauna	Soft sediment fauna
Cluster 8	*Gemma gemma*	Gemma bed	Clam bed	Soft sediment fauna
Cluster 9	*Gemma gemma*	Gemma bed	Clam bed	Soft sediment fauna
Cluster 10	*Nephtys* spp	Nephtys bed	Larger deep-burrowing fauna	Soft sediment fauna
Cluster 11	*Dexiospira spirillum*	*Zostera marina* - Herbaceous Vegetation	Seagrass bed	Aquatic Vascular Vegetation
Cluster 12	Capitellidae*	Capitellid bed	Small surface-burrowing fauna	Soft sediment fauna
Cluster 13	Spionidae*	Spionidae bed	Small tube- building fauna	Soft sediment fauna
Cluster 14	Caprellidae*	Caprellid bed	Mobile crustaceans on soft sediment	Soft sediment fauna
Cluster 15	*Tellina agilis*	Tellina bed	Small surface-burrowing fauna	Soft sediment fauna
Cluster 16	Haustoriidae*	Haustoriid bed	Mobile crustaceans on soft sediment	Soft sediment fauna
Cluster 17	*Idotea balthica*	*Zostera marina* - Herbaceous Vegetation	Seagrass bed	Aquatic Vascular Vegetation

**family was the lowest identifiable taxonomic level*

1.3.5.3. Preliminary Biotopes

Results of PRIMER's DistLM show that 3 variables explain the species distribution in Pleasant Bay. A total of 22.18% of the variation was explained by skewness (11.11%, p=0.001), SD or standard deviation (7.5%, p=0.001) and % clay (3.58%, p=0.015). Skewness is the asymmetry in a statistical distribution, causing the curve to appear skewed to the left or to the right and defines the extent to which a distribution differs from a normal distribution. Left or negative skewness indicates less coarse sediment than the average Pleasant Bay sediment while right or positive skewness indicates more coarse sediment than the average Pleasant Bay sediment. Standard deviation indicates sorting of a sample. Low values indicate well sorted samples while high values indicate poorly sorted samples. % clay is the percentage of sediment between 0.004 – 0.001mm grain size (figure 1.23).

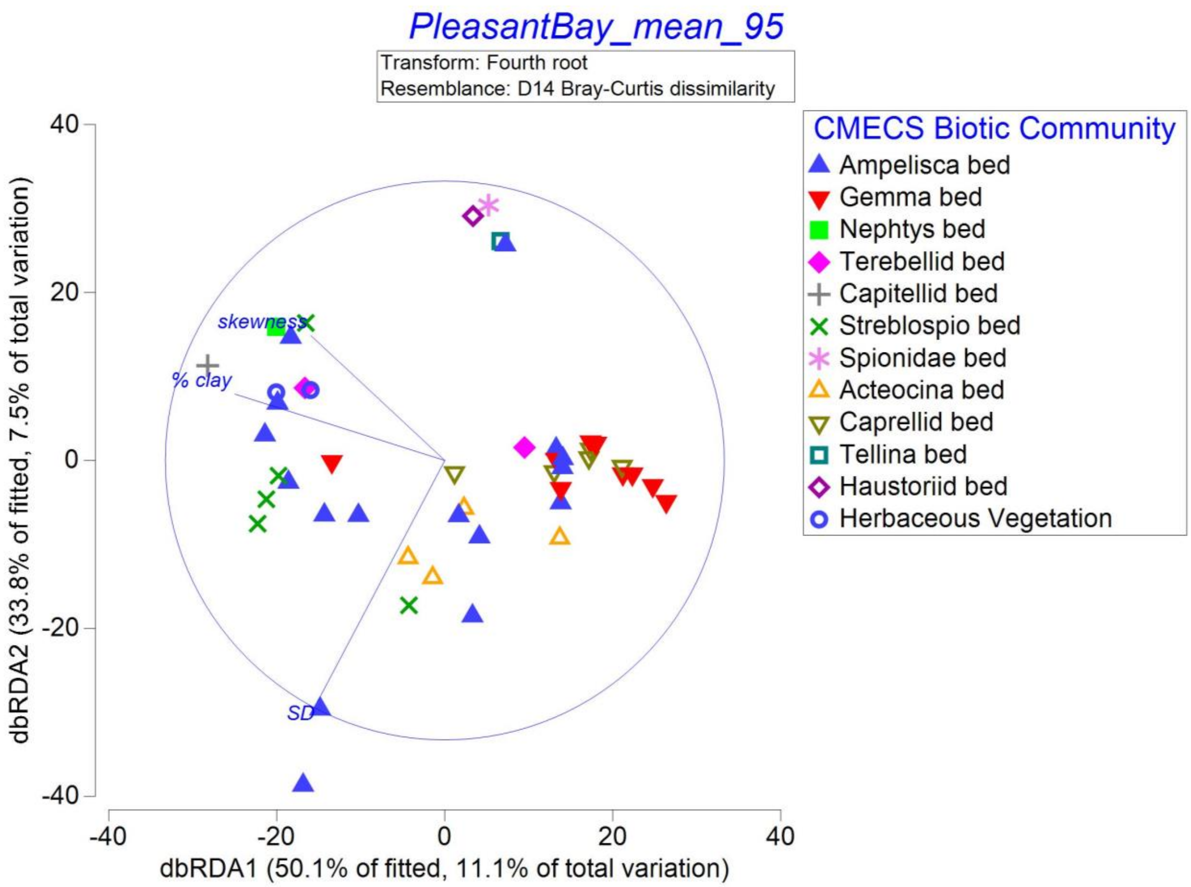

Figure 1.23. Results of the DistLM analysis using Biotic Communities and grain size metrics in a Non-metric multidimensional scaling plot. Axes are dimensionless, distance of symbols represents their relationship. Symbols correspond to biotic communities based on cluster analysis and abundance (table 1.8).

LINKTREE showed a split for skewness between 0.155 and 0.174, for standard deviation between 1.45 and 1.75 and for % clay between 11.9 and 16.7. The skewness split separates station 36 (cluster 16) from all other stations. This can be traced back to the fact that the sediment at this station consisted of 100% sand, with clay, silt and gravel being absent. Haustoridae are the dominant species in cluster

16. However, this family of Annelida is also present at other stations, therefore it does not meet the requirements of an indicator species (definition: species/group that is frequently associated with certain environmental conditions or characteristics while being not often associated with any other environmental condition or characteristic).

Indicator species were found to be significant, albeit poorly correlated, for standard deviation and % clay. *Tellina agilis* (IndVal: 0.003) was indicative of standard deviation < 1.45 and *Ampelisca* sp. (IndVal: 0.314) was indicative of standard deviation >1.45. *Ampelisca sp.* was also indicative of low percentages of clay (< 11.9%; IndVal: 0.314) while Capitellidae were indicative of high percentages of clay (>11.9%; IndVal: 0.001). The indicator values (IndVal) for *Tellina agilis* and Capitellidae in low SD sediments and high % clay respectively, are too low to be considered valuable. *Ampelisca sp.* is indicative of high standard deviation indicating poorly sorted sediments, as well as areas of low percentages of clay.

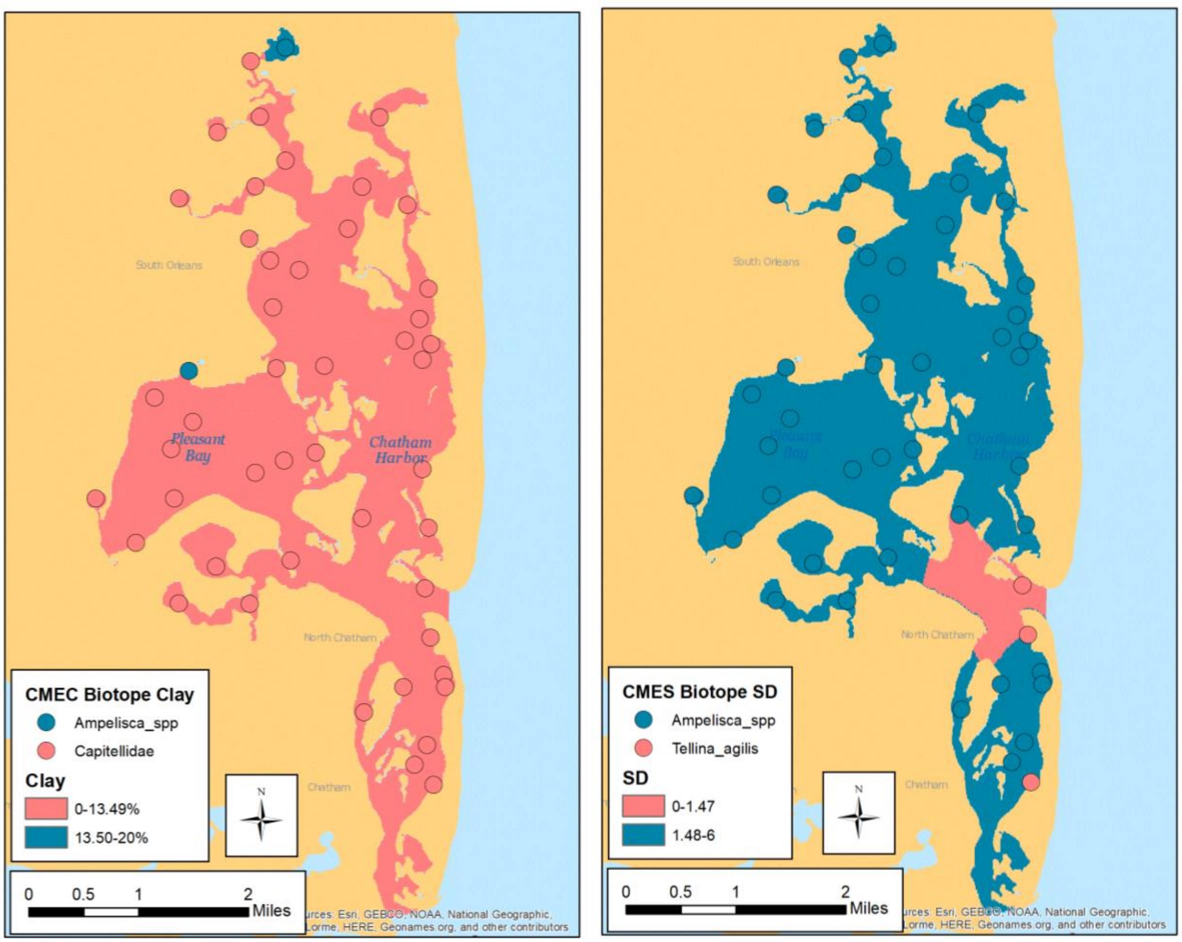

Figure 1.24. CMECS Biotopes map for Pleasant Bay classified by the sediment characteristics % clay showing *Ampelisca sp*. on high clay and Capitellidae on low clay (left) and standard deviation showing *Ampelisca sp* on low SD and *Tellina agilis* on high SD (right)

1.4. Discussion

1.4.1. Vessel- based Acoustic Surveys

Maps, although static, provide essential information that adds to our understanding of dynamic coastal environments. Rather than the thought that 'the map is obsolete before the boat gets back to the dock', we believe these types of surveys should be thought of in a "map once, use many times" framework. For example, a collection of static maps produced for this project were able to document ongoing, short-term coastal processes. Sediment transported into an area near the deepest basin in Pleasant Bay ('Big Bay') provided eelgrass with a shallow water environment where it was able to grow (figure 1.25). Less than 2 km away an existing eelgrass bed was being buried by the natural movement of sediment into the area (figure 1.25). Both these areas are proximal to the tidal inlet that formed in 2007. This inlet formation increased tidal currents and tidal ranges (Adams and Giese, 2008) as well as tidal flushing and improved water quality. However, this increase in tidal currents has also led to more energetic sediment transport in the area – leading to eelgrass habitat creation in one case and eelgrass burial in another. Without the data from this study the burial/loss of eelgrass might have been incorrectly linked to another natural or anthropogenic cause. These data therefore provide important context that may be of use to other researchers and resource managers.

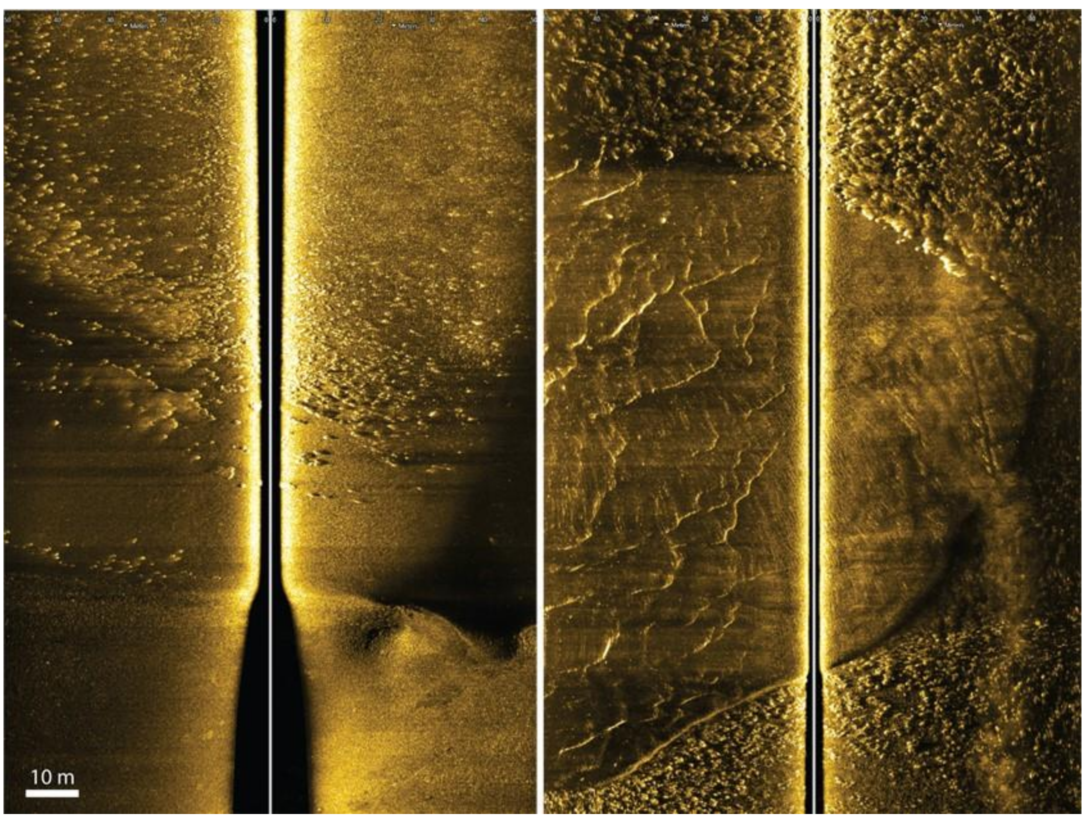

Figure 1.25. Raw sidescan sonar images from Pleasant Bay. Left: An area in Pleasant Bay where sediment being transported into a deeper basin is providing habitat for eelgrass to grow in. Right: An area within the same embayment where the natural movement is sediment is burying eelgrass beds. If these data did not capture this 'snapshot' in time the loss of eelgrass may been wrongly attributed to other phenomena, whether natural or anthropogenic.

The co-location of the bathymetric and backscatter imagery allows for analysis that would otherwise be problematic or much more time-consuming and with a higher level of uncertainty. Sidescan sonar imagery is ideal for mapping eelgrass. Eelgrass is typically straightforward to identify using sidescan data, and metrics like spatial heterogeneity (patchiness) and percent coverage can be determined. Above-ground biomass is another metric that can be determined from interpreting the co-located bathymetric data. While tidal currents can reduce the apparent height of the eelgrass, it can still provide a first-order approximation of above ground biomass (e.g., volume). Using bathymetric data without backscatter imagery makes identifying eelgrass difficult without other corroborative evidence such as underwater video, imagery, etc.

Eelgrass was identified in 290.21 ha in Pleasant Bay. A polygon was created identifying eelgrass at 1:5,000 scale and a minimum mapping unit of 100 m^2 (0.01 hectares). Eelgrass was identified from backscatter imagery in both the mosaic and individual lines exported from SonarWiz v5, thus eelgrass was only identified in areas that had sidescan imagery (figure 1.26). Eelgrass was included here if it could be identified in patches 100 m^2 or larger and within 100 m^2 of each other.

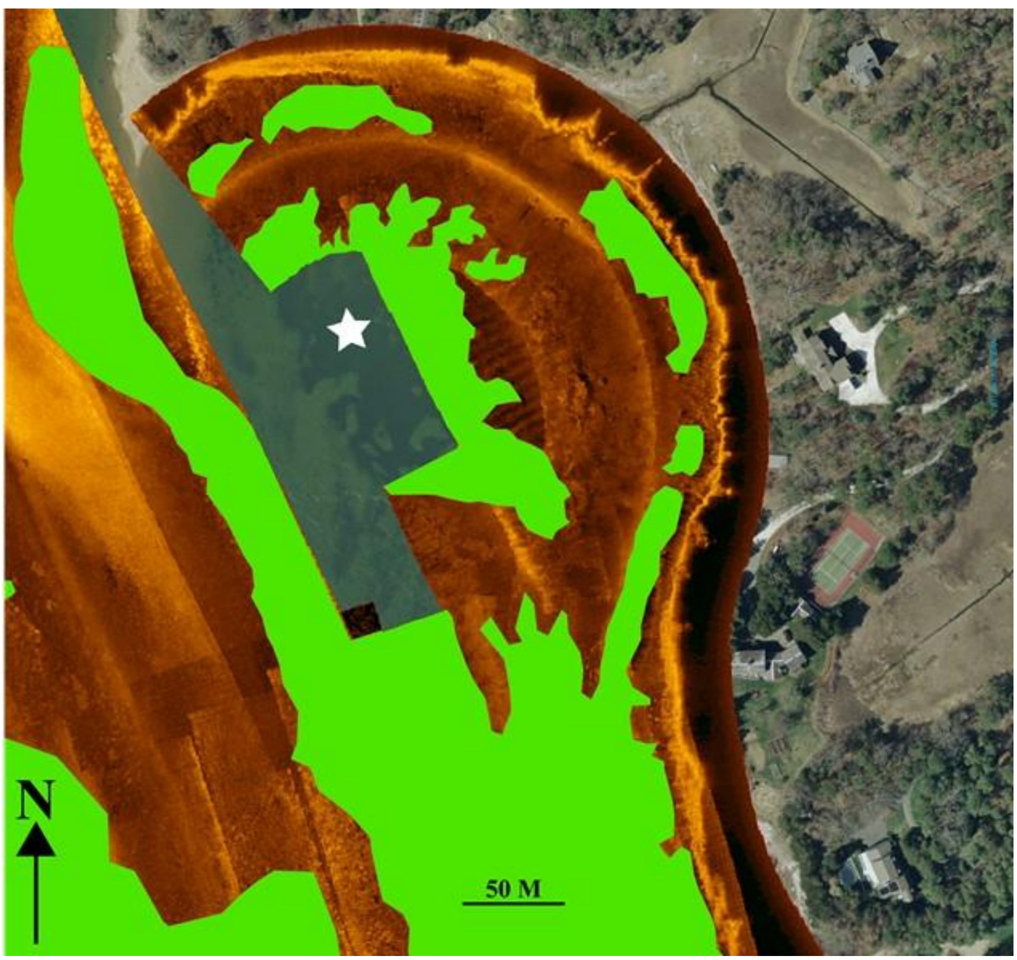

Figure 1.26. Example of eelgrass mapping near Barley Neck. Only areas with sidescan sonar coverage were included. Green polygons are areas where eelgrass is present in patches >100 m^2. The white star is in an area of likely eelgrass beds but was not included as no sidescan imagery was collected in that location.

Collecting acoustic data also provides useful ancillary data that tangentially relate to ecosystem state. Chains attached to mooring blocks make circular patterns in the seafloor as the surface buoy is moved along the surface of (figure 1.27). Mooring chain-scour is another example of a process that is detectable and quantifiable in acoustic datasets. Quantitative spatial analysis of these data could further document the type of habitat disturbed and the area over which the disturbance has occurred. The volume of material removed by the chain-scouring could also be calculated using the bathymetric data. This data could be useful for understanding and quantifying potential impacts of proposed new or expanded mooring fields within or adjacent to coastal parks.

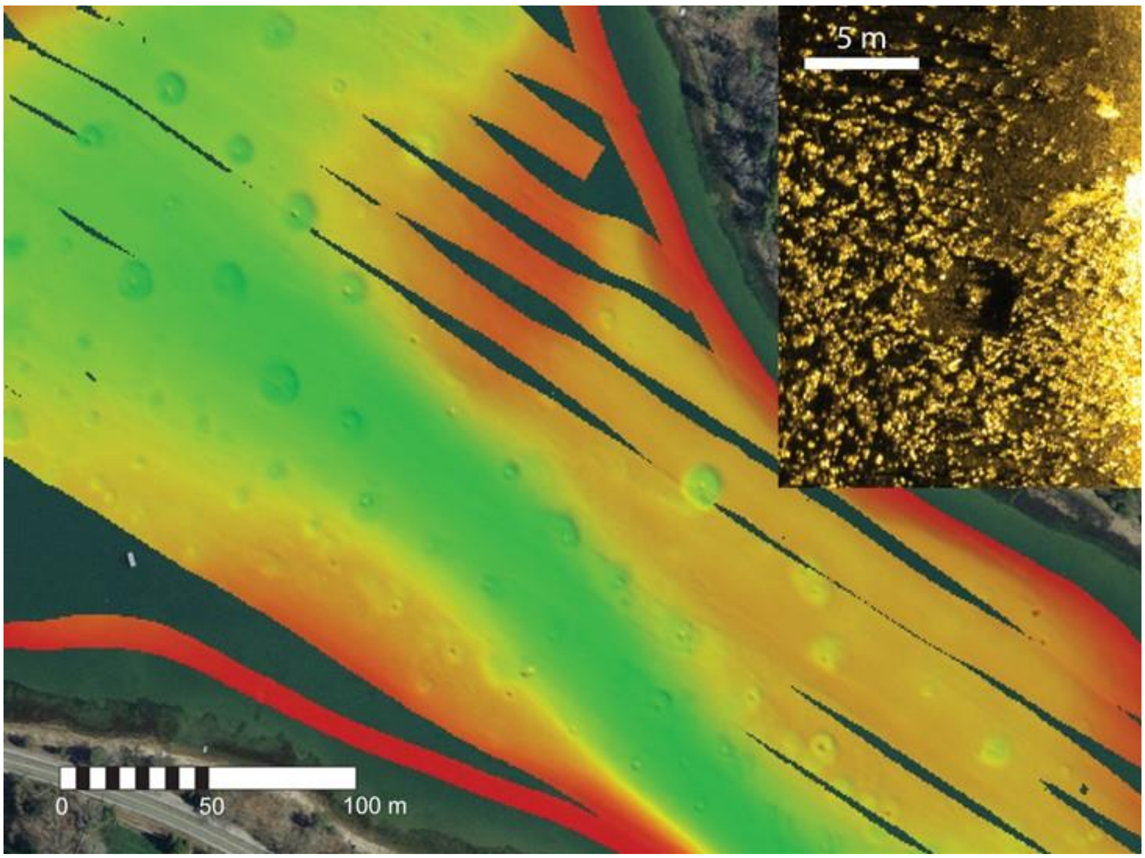

Figure 1.27. Swath bathymetry and backscatter imagery in a mooring field in Crows Pond. The circular patterns seen in the bathymetry are caused by the chains attached to the mooring block. The inset is backscatter imagery in the mooring field where the chain has removed eelgrass or prevented it from growing.

Marine debris is an ever-increasing problem in the world's oceans and for coastal areas in particular (Jeffery, et al., 2016). Acoustic data allow for a first order quantification of the impacts of recreational and commercial usage (Havens, et al., 2008). In Pleasant Bay coir (coconut fiber) logs were stacked and secured to the base of an eroding coastal bluff. Coir logs are often installed at the base of an eroding bank or bluff to attenuate wave energy that would otherwise erode the coastal feature. It has been shown that this method of erosion control often redirects wave energy to areas in front of the bluff, lowering the elevation of the area and undermining the installment or to adjacent properties thereby initiating or accelerating erosion there. During a storm event the coir logs were eroded from the bluff and were scattered along the seafloor. These elongate objects (~3m) were noticed on the seafloor in the backscatter imagery (figure 1.28). Using an underwater video camera, they were identified as coir logs. At low tide investigators revisited the site and unsuccessfully attempted to remove the logs as they represented a hazard to navigation. The locations of these objects were given to the local Harbormaster's Office.

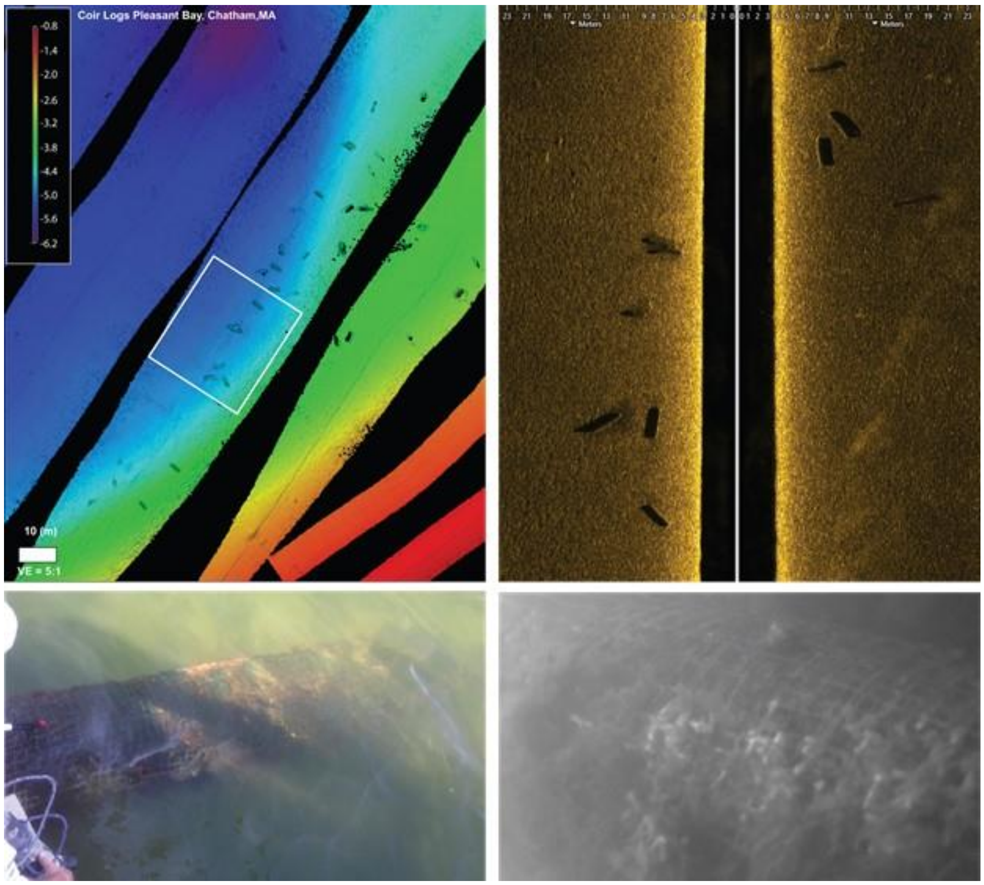

Figure 1.28. Erosion control coir logs on the seafloor near Nickerson's Neck. Upper Left: Swath bathymetry of the area. Upper Right: Raw backscatter imagery of coir logs. Lower left: Photograph taken from boat of coir log at low tide. Lower Right: Screengrab from underwater video taken of coir log at high tide.

Another obstacle 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. Again, the shallow-draft platform extended the length of the survey days in these tidally-restricted areas.

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. However, if 100% bathymetric coverage 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. One could reduce the set range, but if standardization of sidescan sonar data is called for this may not be an option.

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.

1.4.2. Sub-bottom Seismic Reflection Profiling

1.4.2.1. Gaseous Sediment

A distinct seismic facies (Facies NG) which obscures underlying reflectors was mapped through many of the deeper, low-energy basins in Pleasant Bay (figure 1.10). This seismic facies is referred to as a ‘gas wipeout’, 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 gas is common in the subsurface of other estuaries and coastal lagoons (Claypool and Kvenvolden, 1983; Schubel, 1974; 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. The presence of gas, coupled with the nearly acoustically transparent sediment overlying the gas wipeout indicates that these deeper basins are a significant depositional sink for fine-grained (silt and clay) sized organic sediment.

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 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.

Pleasant Bay/Chatham Harbor

Gaseous sediment was identified in Pleasant Bay, portions of Round Cove, and the central basin in Meeting House Pond encompassing a total area of 2.4 km^2. Gaseous sediment was most extensive in ‘Big Bay’, where it was limited to portions of the basin where water depths were > 4 m (figure 1.29).

Shallower areas, including along the margins of the basins, the shallow sill separating the basins of Pleasant Bay east of Round Cove and the slight topographic highs along the southern shore of 'Big Bay'.

Figure 1.29. Map of Pleasant Bay showing the extent of gaseous sediment. 2014 USGS Digital Orthophotograph Basemap (*www.massgis.gov*). North is towards the top of the page.

1.4.2.2. Thickness of Surface Habitat

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.

While much of the deeper portions of Pleasant Bay were obscured by the presence of natural gas, the thickness of Facies M (marine mud) was measured along the edges of the main basin as well as portions of Crows Pond and 'Little Bay'. Thickness ranged from 0 m (glacial deposits cropping out at the seafloor) to 11 m (figure 1.10). The thickest deposits were mapped along the northern edge of Crows

Pond. The acoustically transparent layers (with some stratification visible near the surface drape the hummocky, collapsed glacial surface. It is unclear here if the sediment is all marine mud, or if some of the sediment in these kettles represents older (freshwater) deposits. Facies M in Pleasant Bay generally increases in thickness towards the center of the basin, however the absolute thickness of facies M could not be determined in areas obscured by natural gas or areas where seismic penetration was limited. The thickness of inlet channel deposits were measured on seismic lines within Chatham Harbor, ranging from 0 – 5.6 m thick (average thickness 2.2 m) (figure 1.30).

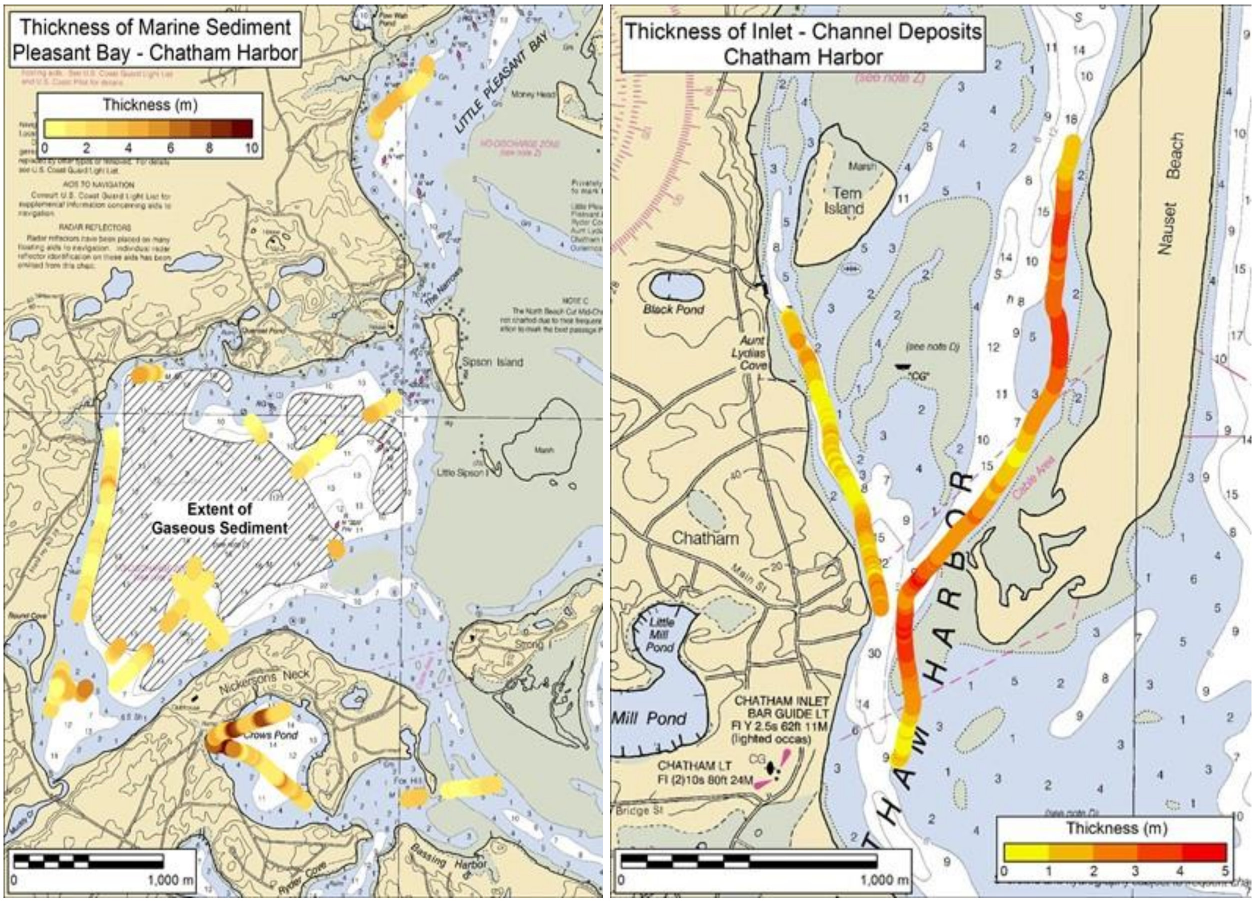

Figure 1.30. Left: Interpreted thickness of marine sediment (Facies M) in Pleasant Bay and adjacent areas. The base of facies M was obscured by the presence of natural gas in the deeper portions of Pleasant Bay. Hatched area shows the extent of gaseous sediment in Pleasant Bay. Right: Interpreted thickness of inlet channel deposits (Facies IC) in Chatham Harbor. The undulating pattern in the thickness is the result of tidal bedforms on the seafloor. North is towards the top of the page Chatham Harbor.

1.4.3. Sediment Core Sampling

1.4.3.1. Past Climate Considerations

Sediment cores were collected in Pleasant Bay, Nauset Marsh and Wellfleet Harbor for the larger NPS study. The bulk of the discussion herein will center around the cores taken from Pleasant Bay, however due to the historical contemporaneous formation and evolution of Nauset Marsh and Pleasant Bay the core taken in Town Cove (OTC) in Nauset Marsh will also be discussed. For more information regarding the cores in Nauset Marsh and Wellfleet Harbor please see (Borrelli, et al., (2018).

Patterns in physical stratigraphy and proxy data link the two coring sites, including a time-correlated volume magnetic susceptibility peak. In OTC sediments this property increases in value from the base up to a depth of 279 cm, at an age modeled year of 1233 CE. Subsequently, susceptibility trends downward, before rising again at the anthropogenic zone. A markedly similar sequence appears in FFC sediments, with susceptibility increasing from the core base to 160 cm (1247CE). The discrepancy in ages between FFC and OTC falls within the minimum error value of 24 years for the relevant radiocarbon dated samples. Viewing volume magnetic susceptibility as a proxy for large scale climatic patterns lends insight into these time correlated sequences. Jones and Mann (2004) have compiled climatic data for the North Atlantic for the past two millennia, and these data indicate the regional occurrence of the Medieval Warm Period from 950 to 1200CE. While complications exist in placing regions in the context of a climatic period recognized worldwide, it may be broadly stated that this period would have given rise to conditions favoring increased weathering of glaciated sediments, and thus enriched magnetically susceptible material sediment deposits. In the sediment records presented here, this rise in susceptibility and subsequent decreasing trend correlate well to climate models for the region. Following the interval for the Medieval Warm Period in our record, low volume magnetic susceptibility coincides with the Little Ice Age spanning 1450 to 1850 CE (Jones and Mann, 2004), wherein climate conditions would have favored a depletion of weathered material in sediment deposits. The anthropogenic zone denoted in FFC, marked most precisely by an increase in volume magnetic susceptibility, provides the uppermost horizon describing alternating trends in this proxy through both core locations. Thus, three distinct, time-constrained packages of sediment material may be identified.

1.4.3.2. Anthropogenic Effects

Identification of an anthropogenic zone in each core location was achieved through correlation of physical characteristics and chemical proxies, with a sharp increase in volume magnetic susceptibility most precisely marking the onset of this zone. Both OTC and FFC experience significant increases in sedimentation rate as a result of anthropogenic activity, likely due in part to increased runoff. The freshening effect of this runoff is evident in the increasing trends of C/S in both OTC and FFC towards the top of each core. Increased fluxes of nitrogen rich runoff and groundwater to the sites has been documented (Carmichael et al., 2004), however this signal is not immediately evident in the sediment record. No variation in organic production trends, marked by $\delta^{13}C$ and %OC values, appears to occur within the anthropogenic zone, while $\delta^{15}N$ values likewise show no appreciable change following the introduction of anthropogenic influences. While production and eutrophication signals are not prevalent, it is worth noting the rapid increase in mass accumulation of organic material and nitrogen that coincides with an increased sedimentation rate. In light of a limited response to anthropogenic activity, it may be that OTC and FFC are effectively flushing out much of the culturally derived nutrients. The relatively short residency times of both environments supports this assumption, as do the chemical proxies present in OTC and FFC sediment cores.

1.4.3.3. Controls on Production

As the record of anthropogenic effects may be limited in the core locations, identifying alternative drivers of phytoplankton production may lend insight into the environmental dynamics at play. Prior to the anthropogenic zone a shift to finer grained sediment occurs in both locations. Within these periods of fine sedimentation in OTC, a relationship between %OC, $\delta^{13}C$, and dry bulk density is seen to occur occasionally, wherein an increase in dry bulk density correlates to a decrease in %OC. This may easily be explained by a dilution effect, making analysis of %OC potentially misleading as a proxy for organic production. Contemporaneous with the dilution effect, however, is a decrease in $\delta^{13}C$. In some instances, an inverse relationship is noted between bulk density and phytoplankton production. With phytoplankton producing a bulk of the organic carbon in these environments, this relationship may represent a periodic control on production by way of turbidity. Suspended sediments have been shown to act as a control on phytoplankton production in estuaries by limiting the photic zone (Cloern, 1987). The conditions present in OTC during periods of lowered organic production are consistent with proxy indicators of a limited photic zone, with an increase in suspended fine material limiting light penetration. A transition to fine grained sedimentation, occurring in 1901 CE in OTC, may be the result of increased tidal energy being introduced into landward back-barrier environments as sea levels have risen, allowing the progradation of flood tidal delta material further into the back barrier. Further, the tidal energy introduced may contribute to bank erosion and the reworking of glacial sediments. This pattern of sedimentation and resulting influence on organic production preceded the onset of the anthropogenic zone in OTC, while in FFC the transition to finer sediment deposition occurred in step with the onset of anthropogenic influence. Considering both the influx of nutrient rich runoff and groundwater seepage with site specific dynamics of sedimentation and photic zone limitation offers a more complex view of the drivers of production in OTC, and may be beneficial to further study of ecology in these locations.

1.4.4. Submerged Marine Habitat Mapping

1.4.4.1. Key Findings

- The results reported for this study represent one way to map and classify submerged marine habitats with the available data
- Submerged habitat maps are the result of a series of data collection and analysis decisions that provide important context for the interpretation of those maps and classified data products
- The Benthic Terrain Modeler toolbox provided a rapid, objective, and repeatable method for mapping and classifying CMECS Geoforms from bathymetry data
- Mapping Geoforms and Geoform Indicator Species provided a rapid, objective, and repeatable approach for integrating geological and biological information
- Point-based substrate maps are extremely reproducible but lack full coverage. Interpolated substrate maps provide full coverage, but require choosing a single grain size metric on which to

base the CMECS Substrate classification, and they may suffer inaccuracies stemming from sampling density or interpolation technique

- Using dominance as the metric to classify the CMECS Biotic Component is easily reproducible but can cause confusion if a single species dominates multiple statistically distinct assemblages. Indicator species analysis provides an alternative equally reproducible method to identifying species that are both abundant and have high fidelity in a particular biotope. Biotope Indicator Species' presence in future sampling may be used to infer the presence of a particular submerged Biotope
- Distance based linear modelling (DistLM) produced simplistic results with generally low predictive power for PB.
- The high resolution sidescan backscatter mosaics did not lend themselves to rapid or reproducible automated interpretation in the context of the Geoform, Substrate, or Biotic Components, but are valuable for supervised delineation of eelgrass and other bottom features, and potentially for verifying surficial habitat patterns and adding fine-scale detail to submerged habitat maps.

1.4.4.2. Data Analysis and Classification 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 could—and in many cases, certainly should—be made with consideration of what the desired map products should be, what scale(s) of map products would be useful, and how the map products will be used. For example, if the purpose of the study was to map the ecological characteristics of eelgrass beds, a sampling strategy and analysis method that compared sediment characteristics and benthic assemblages within and outside eelgrass beds could be implemented. Similarly, if the goal of the study was to determine the ecological effects of non-storm coastal geologic processes, biological sampling would have occurred multiple times a year, and the map products could be produced to represent biological stability or resilience. The data collected, analyzed, and classified in this study could be used to address any of these questions, and others, as well as to provide a baseline for future ecological monitoring in CCNS with further analysis.

Mapping the CMECS Geoform Component

The Benthic Terrain Modeler toolbox and bathymetric position indices were useful tools to map CMECS Geoforms because they were rapid, objective, and reproducible. The classification dictionary developed for this study can be applied repeatedly to new bathymetric datasets from CCNS to quickly produce updated maps of Geoforms, to examine the effects of coastal change, for example. To examine habitats at a finer spatial scale, the search radii that were used to define broad- and fine-scale BPIs certainly could be adjusted based on the desired minimum mapping unit for a particular study or map.

It is clear from the summaries of non-bathymetric physical variables within Geoforms (percent sand, percent gravel, percent organic content, and backscatter imagery) that some Geoforms are not compositionally distinct, whereas others are quite different. For example, “Flats 1-3 m” and “Flats >3 m” within PB had distinct percentages of gravel, whereas the ranges of percent organic content in Herring Cove between “Flats >3m” and “Banks” were overlapping (figure 1.16). We did not perform tests to detect significant differences in the non-bathymetric physical characteristics of Geoforms as part of this study. The determination of what variables (e.g., backscatter and sediment characteristics) are responsible for compositional differences among Geoform types could be a topic for future study, paired with additional sediment samples, and keeping in mind that some Geoforms (e.g., Banks and Platforms) could be expected to have similar sediment composition but differ in their geomorphological (bathymetric) position.

This study used the Indicator Species Value for species in each Geoform because it is a rapid and reproducible way to add biological information to a geologic map. The Indicator Species Value determines which species are both abundant in a certain Geoform *and* rarely found in other Geoforms (i.e., have high Geoform fidelity). A permutation of the data provides a measure of significance. The association between a Geoform and an indicator species can be used to predict species presence, given the presence of the Geoform, or vice versa. In this way, indicator species can be valuable in repetitive mapping or habitat monitoring.

Mapping the CMECS Substrate Component

Maps showing the CMECS Substrate Groups and Subgroups at discrete points were perhaps the most straightforward maps developed in this study (figures 1.18). These maps are clear and easily reproduced because the CMECS Substrate Component classification structure is based on the Wentworth classification used by geologists to categorize marine sediments. The USGS SEDCLASS software completes the Wentworth classification from the weight percentages of gravel, sand, silt, and clay in each sample, which can then be directly cross-walked to the CMECS Subgroup level.

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 submerged habitat mapping. The full coverage median grain size maps are also built on the assumption that sediments conform to gradients in the study areas (i.e., there are no stark boundaries in sediment type). The gradient model could be more often true than a discrete-boundary model, but there are certainly observed examples of the latter. For example, there may be a discrete boundary between a new patch of eelgrass that traps fine-grained sediments within a larger sandy flood-tidal delta deposit. In addition, sources of error in the interpolation process, and sampling spacing that does not match the scale of environmental heterogeneity should both be considered to 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. The maps in this report are meant to represent the results of objective mapping decisions necessary for consistently characterizing shallow submerged environments. 77

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. There are benefits and limitations to each approach. Using dominance is recommended in the CMECS Technical Guidance Document (2014-2), but can cause classification confusion when a single species may be dominant in several statistically distinct assemblages, as was the case in Pleasant Bay (table 1.8). 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. This is not recommended in the CMECS Technical Guidance document, but is worth considering. As described above, 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). 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 in this analysis.

Within individual study areas, only sediment variables and infaunal abundance were examined as part of the biotope analysis, whereas in the CCNS-wide analysis, acoustic, sediment, and infaunal abundances were all integrated into biotopes. Overall, the limited number and scope of the physical variables were likely responsible for the simplistic biotope results. In most of these CCNS-wide analyses, only one or two physical variables were found to explain the highest proportion of the variance in benthic assemblages, and the total variance explained was not very high (between 12.6-55.1%). In contrast, previous work using this method, where the resulting classification tree had 7 branches derived from 3 variables that explained 68.9% of the variance in benthic assemblage structure (Shumchenia and King, 2010). In coastal Maine, abiotic variables explained between 37-59% of the variance in benthic assemblages (McHenry et al. 2017). These two previous studies place the results of this work in context, and indicate that the results reported here are not outside the range of expectations for the methods used. Furthermore, acoustic and sediment variables were examined only at the sample-scale for this study, but future work that considers those variables at some larger patch size surrounding each sampling point could result in stronger abiotic-biotic associations. A patch summary might better represent the environment that benthic assemblages experience, and therefore might yield better explanatory and predictive power.

The benthic habitat mapping study conducted in Pleasant Bay for the Cape Cod National Seashore included 33 stations spread throughout the Bay. Additional funding enabled us to add 15 stations to the already existing dataset. The 15 stations coincided with previously studied areas (Howes et at

2006), however, none of these data were available at the time of writing this report. As expected, including these 15 stations in our statistical analysis changed the results significantly. Not only did the overall number of individuals identified change, but also species abundances and dominance in certain areas. Previously, statistics identified 10 biotic communities, now the results show 17. More biotic communities generally point towards more diversity, however, when looking at the dominant species, some biotic communities overlapped. Even though the additional 15 stations resulted in an additional 15 species identified in Pleasant Bay, the significance of the statistical analysis identifying indicator species for specific habitats (including sediment characteristics and sediment formations on the seafloor) decreased.

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 detailed examination of the distribution patterns of each dataset was beyond the scope of this study, but could be used to determine the most appropriate metric for each collected dataset. 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). For this analysis, maps of standard error for each interpolation were produced, so that users can interpret areas of the map that may have higher uncertainty than others. In general, interpolation is challenging in oddly-shaped water bodies. Barrier interpolation is essential in these water bodies to avoid extremely poor interpolation results, but any physically isolated parts of the embayments can be expected to have locally higher standard error. The standard error of interpolated maps can be lowered by increasing sampling point density.

1.4.4.3. Habitat Maps for Pleasant Bay

CMECS Geoforms in PB were related to the distance from the mouth of the bay. There are several small depositional basins in the interior parts of PB, as well as more exposed and physically dynamic areas with channels and bedforms. PB sites encompassed a wide range of CMECS Substrate Groups (i.e., from mud to gravelly) and Subgroups (i.e., from silt to gravelly sand).

The cluster analysis of the benthic infauna abundances generated many clusters, but several of these had the same dominant species (table 1.8). In addition, sediment type only differentiated benthic communities broadly, i.e., the LINKTREE had two branches differentiating between organisms living in coarse versus fine sediments. The fine sediments branch split again between poorly sorted and well sorted sediments. Furthermore, the sediment variables did not explain much of the variance in the benthic infauna species data. These results suggest that other biological and physical factors may be structuring benthic communities in PB. Possible examples include biotic interactions such as competition and predation, as well as dominant benthic vegetation type or water quality. It has been shown that water quality influences benthic habitat quality in PB (Howes et al., 2006), and factors such as dissolved oxygen likely play an important role in driving the composition of benthic communities in the system.

When the physical variables were forced to predict the biology, the predicted biotopes for PB made intuitive sense – *Ampelisca spp*. is common species in these environments (Hale et al., 2017; Gosner 1978). The first major split at skewness was expected and significant, but only explained a very small area of Pleasant Bay. Sediment at station 36 (Cluster 16) was composed of 100% sand and therefore coarser overall than any other station/cluster, thus causing a splitting apart of one single station and preventing the determination of an indicator species. Haustoridae are the dominant species in cluster 16. However, this family of Annelida is also present at other stations, therefore it does not meet the requirements of an indicator species (definition: species/group that is frequently associated with certain environmental conditions or characteristics while being not often associated with any other environmental condition or characteristic). The next split separated the remaining 47 stations into areas with poorly sorted sediment and areas with well sorted sediment. A third split (% clay) made ecological sense, however it was of low correlation as well as low significance.

Three significant indicator species could be determined (*Ampelisca sp*, *Tellina agilis* and Capitellidae), which were also the most dominant species in several clusters in the benthic community cluster analysis, suggesting that they play an important role in the overall composition of benthic communities in PB. The biotope maps (figure 1.24) predicted large parts of Pleasant Bay to contain low amounts of clay and be well sorted and the individual sites were rarely misclassified (i.e., the interpolation of % clay and sortedness was robust). Geoform indicator species for PB were not conspicuous members of CMECS Biotic Communities or Biotopes. *Gemma gemma* were indicative of Basins and Channels, *Tellina agilis* indicated banks, and Caprellidae indicated platforms. None of these associations is odd, but they could be useful in developing further studies to examine the underlying causes of physical-biological relationships.

1.5. Conclusions

This study focused largely on mapping embayments rather than along arbitrary delineations. Ecosystem-based mapping should be the priority rather than mapping within seashore boundaries. In this instance it was made possible by the additional support from the Friends of Pleasant Bay. It is an example demonstrating that sustained efforts can be made to increase awareness, participation, and funding by engaging local and seasonal residents and other stakeholder groups.

At early meetings related to the larger NPS project the four mapping teams, regional and relevant park staff were critical to improving the quality of this project as well as maximizing future comparative work within and between these four subject parks. It is of great value logistically, financially and with regards to final product quality to coordinate these multiple site projects, and we believe that this method is a vast improvement over independent mapping projects. Further, the FOPB and future investigators will benefit from these baseline data with which to study system evolution from myriad scientific and policy disciplines.

Every effort should be made—if not insisted upon in future studies—to coordinate sonar surveys and benthic grab sampling in order for the former to guide the latter. For example, the late spring and early summer sonar surveys could be completed with the aim of immediately producing a reconnaissance sidescan mosaic and geologic habitat interpretation to guide benthic grab samples starting in July through September. While we were able to test some physical-biological relationships here, their statistical associations may not be as strong as if we had structured the sediment and benthic community sampling in this way.

The seismic reflection profiling—though conducted at a reconnaissance scale—provided a third dimension to the mapping which may not have directly be incorporated into final map products but may be a valuable addition to CMECS in the future. A more systematic mapping approach would refine the extent and distribution of seismic facies and sediment thickness and improve the understanding regarding the development and geologic processes in the study area. These data also provide valuable guidance for the sediment coring work. Additional sediment cores in the areas of gaseous sediment could help to quantify the volume of gas in these areas. Cores in areas of 'deeper' gas could provide insight into the formation and source of gas in these estuarine environments.

Sediment coring coupled with mapping thick deposits of facies M, which show an increase in seismic intensity with depth, could be enhanced. These areas could provide insight into changes in depositional processes as these areas transitioned from upland or freshwater environments to lagoon environments. Studies of sediment deposition, measured using a combination of sediment traps to examine modern sedimentation rates and well-dated sediment cores in the low-energy areas could quantify the volume of 'blue' carbon being sequestered in these low-energy depositional sinks.

The depositional history recorded in sediment cores from back barrier depocenters in Cape Cod provides insight into the shifting ecological, climatic, and sea level conditions over the past ~1500 years. Age-depth constrained physical and chemical characteristics of these sediments have allowed the identification of three distinct stratigraphic packages, reflecting climatic shifts from the Medieval Warm Period to the Little Ice Age, and the onset of an anthropogenic zone. Elemental and isotopic analyses have defined ecological conditions and primary organic production sources through time, and demonstrate how these have adjusted in response to rising sea level and anthropogenic influence. Marine phytoplankton have dominated organic carbon input through the depositional history presented here, while influence of terrestrial C3 and C4 plants in Frost Fish Cove has decreased with sea level transgression. Contemporaneous response to rising sea level in both backbarrier systems, described in terms of time-constrained inundation around 1080 CE, demonstrate the influence of rapid transgression and its importance in shaping coastal environments. Along with increased delivery of nutrients by anthropogenic activity, organic production appears to be influenced by an increase in the reach of tidal energy and the resulting rise in fine suspended sediments. Study of sediment cores from locations in the backbarrier system of Cape Cod provide a model for the use of proxy data in creating paleoenvironmental histories for coastal regions, and for the application of proxy data in aiding analysis of continuing coastal response to rising sea levels.

Predictive maps of substrate type and biotope were highly dependent on the robustness of the geospatial interpolations of the driving physical variables. In some cases, these interpolations likely had areas of high error due to the number of samples and their distribution. These results should be interpreted with these caveats in mind.

Since a greater 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. Acoustic variables were not identified as important explanatory factors for the variance in the CCNS-wide physical-biological data, suggesting that though these data are important to develop maps of eelgrass, shellfish, human-induced impacts and sediment transport, they may not be well-suited as rapid-assessment indicator of the benthic communities within CMECS at present.

Multivariate regression trees (LINKTREE) in general did not explain the majority of the variance in the data, but relative error values for Herring Cove were well within the typical range for these types of analyses. Canonical ordination techniques (e.g., redundancy analysis or canonical correspondence analysis) may offer more promise in helping to characterize physical-biological relationships. These alternate methods combine multiple regression with ordination, and thus allow more flexibility in the range and number of physical variables that correspond to important biological patterns.

Despite the limitations of the field survey design, considerable local detail exists in the dataset that will be revealed in future hypothesis testing and statistical analyses. This study and associated data comprise a critical baseline record of biological and physical characteristics within Pleasant Bay. 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. As described throughout, the classification and mapping approach employed for this analysis is only one of many possible treatments of the data. 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 Pleasant Bay.

As the first such study of many to follow, especially considering the fact that two sampling events were combined here (33 stations for CCNS, 15 stations for Friends of Pleasant Bay), the lessons learned are already apparent. Conducting this study in Pleasant Bay, and indeed Wellfleet Harbor, Nauset Marsh and Herring Cove (as part of CCNS), underlined the importance of creating maps using acoustics before choosing benthic invertebrate sampling stations. By doing this, specific areas that might be small in size, but important for species diversity (e.g. eelgrass beds), can be purposefully included via random stratified sampling design. This has already been successfully implemented in two other projects. Lab protocols on invertebrate picking and identification have also been slightly changed and have streamlined the process in the two previously mentioned studies.

Acknowledgments

The authors would like to thank Captain Ted Lucas, who piloted both research vessels for all the surveys in this study, for his steady patience, capable helmsmanship, and invaluable insights that improved our shallow water survey techniques. We thank Lev Gerstle for completing the long and arduous benthic invertebrate processing and identification, along with the many volunteers and paid interns including: Ta-Bin Brown, Paul and Helen Gigley, Jean Horner, Kathy and Ali LaBella, Ian Kinahan, Heather Shea, Jim Woodworth and Melissa Yow; also, Violet Doucette, Natasha Drotar, Tim Famulare, Maddie Genco, and Jack Olson. From Cape Cod National Seashore, we thank Dr. Megan Tyrrell for critical early guidance and review of our acoustic survey plans and bottom sampling techniques onboard the Research Vessel. We thank Mark Adams for invaluable input throughout the project, Dr. Jason Taylor for many fruitful conversations about park concerns, and finally George Price, park superintendent, for allowing us access to park laboratories and his very busy and dedicated staff, who spent many hours helping make this project immeasurably better.Blak

The Research Vessel (R/V) Marindin was named after Henry L. Marindin, a Topographer with the U.S. Coast and Geodetic Survey. Between 1887 and 1889 Marindin completed a detailed survey of the outer Cape from Chatham to Provincetown, during which he collected 190 profiles along 56 km of shoreline in what is now the Cape Cod National Seashore. Surveying from the top of the coastal bluffs out to ~10 m of water depth, Marindin's goal was to develop a foundational dataset for concerns of the day and needs of the future:

> *Among the objects aimed at one was to obtain an accurate mold of the exposed shore of Cape Cod for present use in determining the amount of waste or fill since previous surveys – where the surveys are sufficiently detailed for this purpose – but the more direct object was to provide a base for future comparisons, which will be of value to geologists and others who study the changes in the coast-line. (Marindin, H.L. 1889)*

As its namesake was one of the first investigators to realize the importance of mapping the land/sea interface as one system, this customized, shallow-draft boat used to map that area between the marine and terrestrial environments and thus linking them was commissioned in his honor in 2012.

Note: Some text and figures from this chapter are taken verbatim, without accreditation, from a larger study Borrelli, et al, (2018), which itself was an amalgamation of other reports including Shumchenia (2016), Oakley (2016), and Hubeny and Love (2016). As those authors are also co-authors of the larger report (Borrelli, et al., 2018) citations for each usage would have been redundant and significantly reduced the readability of this chapter.

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CHAPTER 2
Fisheries investigations in Pleasant Bay, Cape Cod, Massachusetts

Nichols O.C., B.J. Legare, T. Famulare, E. Sgarlat, and T. Lucas

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Abstract

Pleasant Bay is a coastal lagoon system featuring diverse habitats that support a variety of commercially, recreationally, and ecologically important marine species. The 2013 Pleasant Bay Resource Management Plan (PBRMP) noted the changes in shellfish and finfish abundance, species composition and fishing activity within the Bay since a 1965-66 study conducted by the Massachusetts Division of Marine Fisheries. Based on the recommendations of the PBRMP, we conducted an inventory of shellfish and finfish in the Bay, with a focus on commercially and recreationally important species. Intertidal and subtidal fish and invertebrate sampling was conducted in Pleasant Bay from June 2015 through June 2016. A survey for postlarval lobsters was conducted in 2014 and opportunistic sampling was conducted from July 2015 through October 2017. Where practical, sampling efforts were conducted using similar methods and gears to previous studies in the same area or more recent studies in the wider region. Intertidal and subtidal survey effort (trawl, n = 90 tows; dredge, n = 102 tows; seine, n = 15 hauls) was distributed relatively evenly over the year, although there were gaps due to fall and winter weather conditions. The overall species community and seasonal abundance of most species was broadly similar to that observed in the 1965-66 MADMF study. This study included more sampling methods and greater spatial coverage than the previous study, and documented greater species diversity. Fish community composition and seasonal patterns of abundance during this study were broadly similar to those observed during other recent studies along the eastern shore of Cape Cod. This comprehensive inventory indicated that Pleasant Bay is home to a diverse assemblage of marine animals, many of which utilize the Bay as spawning or nursery habitat.

Differences in species diversity and relative abundance were observed between this study and the 1965-66 MADMF study, as well as between years during this study. Long-term monitoring is necessary to place our observations in a broader context.

2.1. Introduction

Pleasant Bay is a coastal lagoon system featuring diverse habitats and separated from the North Atlantic Ocean by a barrier beach. The Bay is surrounded by ca. 69 km of coastline, and its watershed includes the towns of Orleans, Chatham, Harwich and Brewster. The 2013 Pleasant Bay Resource Management Plan (PBRMP) noted the changes in shellfish and finfish abundance, species composition and fishing activity within the Bay since a 1965-66 study conducted by the Massachusetts Division of Marine Fisheries (MADMF; Fiske et. al., 1967). At the time of the Fiske et al. (1967) study, winter flounder (*Pseudopleuronectes americanus*) supported a sizable commercial fishery in Pleasant Bay, harvested by small trawlers. As noted in the PBRMP, the species' abundance has since declined in the Bay due to unknown causes and no longer supports a fishery. Pleasant Bay is a known habitat for the American eel (*Anguilla rostrata*) and at one time supported a small-scale harvest of the species (Fiske et al., 1967). There are multiple anadromous fish runs around the perimeter of the Bay, frequented by migrating herring – the runs are monitored by multiple agencies and were not sampled during this study. The Bay has become increasingly known for abundances of striped bass (*Morone saxatilis*) and bluefish (*Pomatomus saltatrix*), which are often targeted by recreational fisheries. Fiske et al. (1967) noted the abundance of forage species such as sand lance (*Ammodytes spp.*) and silversides (*Menidia menidia*), as well as longfin inshore squid (*Doryteuthis pealeii*).

The Bay historically supported a variety of shell-fisheries, most notably targeting quahogs (*Mercenaria mercenaria*), bay scallops (*Argopecten irradians*), and soft shell clams (*Mya arenaria*; Fiske et al., 1967). According to the PBRMP, abundances of the above and other shellfish species have changed over the last few decades, as have the associated fisheries. The results of dedicated shellfish surveys are reported on in Nichols and Grieco (2018; see Appendix 2). In the past several decades, American lobsters (*Homarus americanus*) have sometimes reached commercially harvestable abundances in Pleasant Bay[2], and fall observations of juvenile lobsters (~3-4 cm total length) in Pleasant Bay mooring areas over the past decade[3] have led to the hypothesis that Pleasant Bay may be an important nursery habitat for lobsters.

Among the recommendations of the comprehensive MADMF study was that it be repeated in ten years; to date, no efforts have been made to repeat this study. A recommendation in the Fisheries Management section of the PBRMP was to conduct research on the status of Pleasant Bay's fisheries habitat, specifically to develop and conduct a long term monitoring program of the Bay's finfish and shellfish habitat. Following the recommendations of the PBRMP and supported by the Friends of Pleasant Bay, we conducted an inventory of shellfish and finfish in the Bay, with a focus on commercially and recreationally important species.

2.2. Methods

Intertidal and subtidal fish and invertebrate sampling was conducted in Pleasant Bay from June 2015 through June 2016, and some additional trawl and seine sampling effort targeting tropical fishes occurred from August through October in 2016 and 2017 (M. O'Neill, Gulf Stream Orphan Project). Where practical, sampling efforts were conducted using similar methods and gears to previous studies in the same area (e.g. Fiske *et al*. 1967) or more recent studies in the wider region (e.g. Chase *et al*. 2002). Subtidal sampling stations were chosen as a subset of 15 sites chosen at random for benthic habitat sampling (Borrelli *et al.*, 2018). The distribution of stations (figure 2.1) was chosen based on a combination of accessibility (depth) and preliminary data on habitat type as determined from benthic sampling. All fish and most macroinvertebrates were identified to the lowest practical taxon. In the interest of efficiency, small species that would not likely be consistently captured by sampling gears (e.g. small amphipods and isopods < 2 cm) were not consistently documented. Sand and grass shrimp (*Crangon* and *Palaemonetes* spp.) were noted but not counted. The appropriate size measurement was taken (Total Length [TL], Fork Length [FL], Carapace Length/Width [CL/CW], Mantle Length [ML], etc.) for all fishes and most invertebrates. Large catches were subsampled for size data. Priority was given to species of commercial or recreational importance. With the exception of organisms that were

[2] K. Martin, F/V Time Bandit, personal communication
[3] C. Beggs, Ames Marine, personal communication

preserved because they could not be readily identified in the field using the appropriate key (e.g. Gosner, 1978; Robins *et al*., 1986), all specimens were released alive.

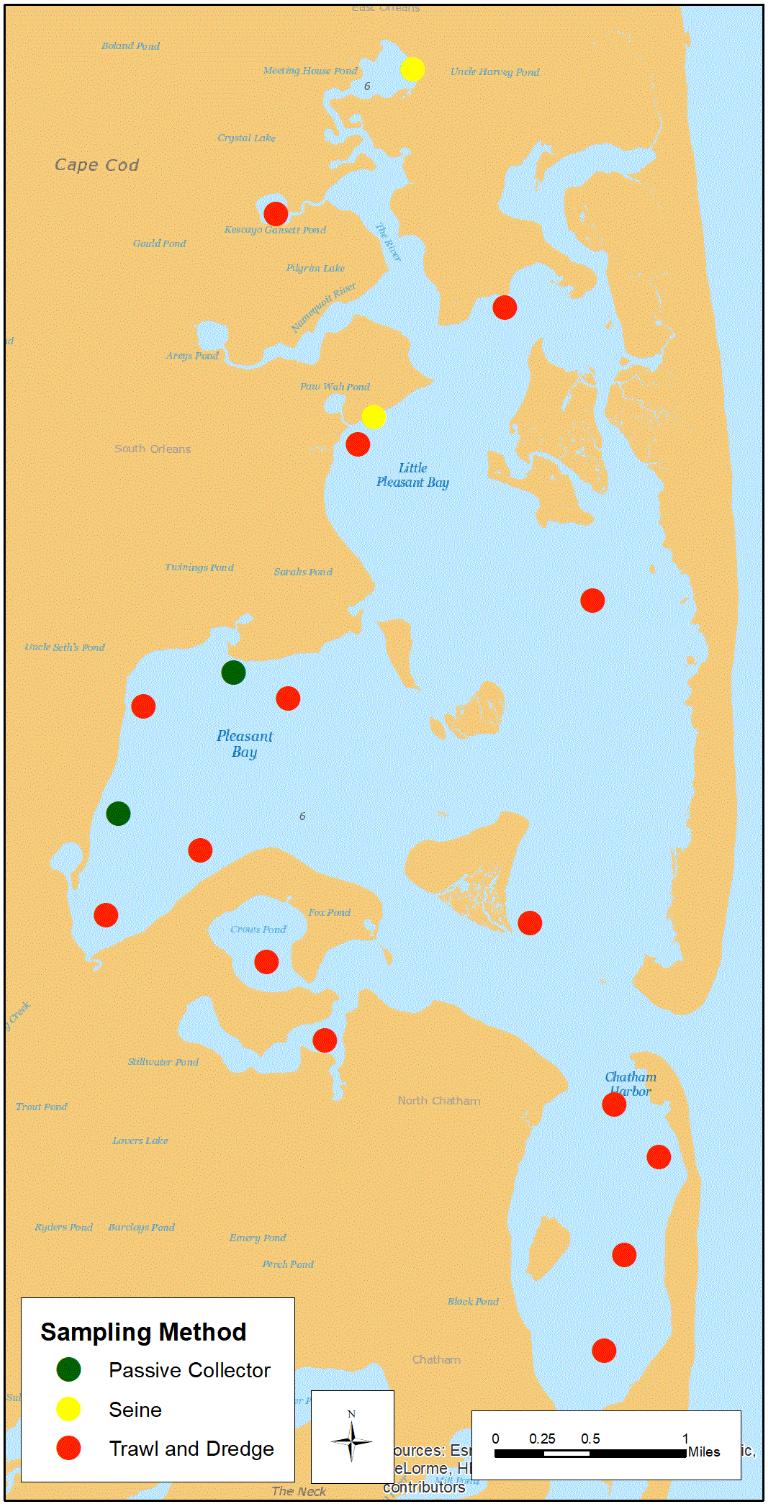

Figure 2.1. Sampling stations in Pleasant Bay.

2.2.1. Seine Sampling

Contingent upon weather and ice cover, intertidal surveys were conducted approximately every two months (bimonthly) from June 2015 to June 2016, consisting of two days of beach seine sampling at two stations sampled in a previous study in 1965-66 (Fiske *et al*. 1967; figure 2.1). Intertidal sampling was conducted using a 50’ (15.2 m) beach seine with a 1.2 m depth, 1.2 m square bag, and 4.8 mm knotless mesh, following the standardized methods of Chase *et al*. (2002).

2.2.2. Trawl and Dredge Sampling

Contingent upon weather and ice cover, subtidal surveys were conducted approximately every two months from June 2015 to June 2016 at a subset of 15 sites previously selected at random during 2014 benthic habitat sampling (Borrelli *et al*., 2018; figure 2.1), consisting of three consecutive days over which trawl and dredge sampling were conducted. Subtidal surveys were conducted on board R/V *Shackleton*, a 20’ (~7 m) center-console v-hull vessel with a 110 horsepower (hp) outboard engine (figure 2.2). Sampling was conducted with a small trawl net and a commercial bay scallop dredge.

Figure 2.2. R/V *Shackleton*, used for subtidal surveys. Note scallop dredge on culling board.

The net was identical to that used by Chase et al. (2002), a 30’ (9.1 m) sweep Wilcox shrimp trawl with a 8.2 m head rope, 3.8 cm stretched mesh in the wings and cod end, and a 6.4 mm knotless mesh cod-end liner. The net was attached to 81 x 41 cm oak doors with steel runners by 1.5 m rope legs. Tow lines were set at ca. 4:1 scope and adjusted for depth. The bay scallop dredge was a standard commercial design consisting of a 26” (66 cm) wide lightweight frame with a 4’ (122 cm) sweep chain, and a catch bag made of 2” (5 cm) steel rings and 1.5” (3.5 cm) square mesh. The tow line was set at ca. 4:1 scope and adjusted for depth.

Both the trawl net and scallop dredge were deployed in a standardized manner, with consistent tow times and speeds (trawl: 5 minutes at 2 knots, dredge: 3 minutes at 3 knots). Tow start and end locations and depths were recorded using a Garmin 76 GPS and the boat’s sounder (Faria Instruments DS1002 dual-temperature depth sounder). A duplicate tow was conducted immediately adjacent to the location of the first, in the opposite direction. Seawater and air temperatures were recorded at the beginning of each tow using the sounder.

2.2.3. Ventless Lobster Traps

Ventless traps identical to those used by MADMF and other regional agencies for lobster surveys (Courchene and Stokesbury, 2011) were set at selected trawl/dredge stations. Traps were based on standard commercial lobster traps made from 2.5 cm mesh with 12.7 cm diameter entrance rings, one parlor and one kitchen, but without the standard escape vent required on commercial traps to allow escape of smaller animals. Ventless traps were set in strings of 5, spaced ca. 30 m apart, each with a single buoy, weak link, and line. Bait consisted primarily of frozen alewife (*Alosa pseudoharengus*) donated by MADMF from their herring sampling program, although other bait was used (sea herring, bonito, etc.) as necessary. Traps were soaked for 3-5 days depending on weather conditions.

2.2.4. Gillnet Sampling

In order to sample larger, highly mobile fish species such as striped bass and bluefish, a 200’ (61 m) long, 6’ (1.8 m) deep #6 monofilament experimental gillnet was set on 3 occasions at depths < 5 m adjacent to a trawl/dredge station, consisting of four 50’ (15 m) panels with square mesh sizes increasing at 0.5” (1.3 cm) increments from 2” (5.1 cm) to 3.5” (8.9). The gillnet was strung from 0.5” foamcore float line and 30# leadcore lead line and set at the bottom using trawl anchors. The ends were marked with single buoys attached to the float line. Due to the presence of seals and seabirds in the area, soak times were relatively short and the gear was visually monitored for protected species presence several times over the course of each set.

2.2.5. Passive Collectors

Twenty passive postlarval collectors (Wahle et al., 2009) were filled with 4-10” cobble and deployed in two locations (10 at each site; figure 2.1) in Pleasant Bay from a mooring barge on 18 July 2014. Collectors were marked with a single bullet buoy and deployed in approximately 10 feet of water near mooring fields where juvenile lobsters had been previously observed. Collectors were retrieved on 25 October 2014 using the same mooring barge. The cobble was carefully removed from each collector and all fish and invertebrates were identified to the lowest practical taxon and counted. Lobsters were measured to the nearest 0.5 mm carapace length (CL).

2.2.6. Opportunistic Sampling

Several means of opportunistic data collection were conducted during this study, including minnow trap sampling at selected docks around Pleasant Bay, additional passive collector, trawl and seine sampling targeting tropical fishes for the Gulf Stream Orphan Project in summer and early fall 2016-2017, seine sampling for the BioBlitz event conducted by Pleasant Bay Community Boating, and observations of recreational fishing activity.

2.3. Results

Intertidal and subtidal survey effort was distributed relatively evenly over the year, although there were gaps due to fall and winter weather conditions (table 2.1). Multi-day trawl and dredge surveys sometimes spanned multiple calendar months.

Table 2.1. Monthly effort (number of tows/hauls) by gear type, June 2015-June 2016

Gear Type	Total	Jun 2015	Jul 2015	Aug 2015	Sep 2015	Oct 2015	Nov 2015	Dec 2015	Jan 2016	Feb 2016	Mar 2016	Apr 2016	May 2016	Jun 2016
dredge	102	7	19	6	12		22	6			6	12		12
trawl	90	2	17	6	11		18	6			6	11		13
seine	15	2	1	1		2		1		2	2		2	2

2.3.1. Seine Sampling

When possible given weather and tide, both seine stations were sampled bimonthly (table 2.2). The most commonly captured organisms in the seine were the mummichog (*Fundulus heteroclitus*, n = 4,361), Atlantic silverside (*Menidia menidia*, n = 2,435), striped killifish (*F. majalis*, n = 1,659), and fourspine stickleback (*Apeltes quadracus*, n = 789). These four species accounted for 95% of the total catch (table 2.2). Sand and grass shrimp (*Crangon* and *Palaemonetes* spp.) were noted but not consistently counted.

Table 2.2. Organisms captured by seine sampling, June 2015-June 2016.

Common Name	Total	Jun 2015	Jul 2015	Aug 2015	Sep 2015	Oct 2015	Nov 2015	Dec 2015	Jan 2016	Feb 2016	Mar 2016	Apr 2016	May 2016	Jun 2016
Alewife	33													33
American eel	1			1										
Atlantic needlefish	1			1										
Atlantic silverside	2435	43	23	187		1919		36		2	129		44	52
Atlantic tomcod	2							2						
Bluefish	1													1
Fourspine stickleback	789	110	35	8		208		17		21	52		180	158
Green crab	19	13	2			1								3
Long-clawed hermit crab	2	2												
Menhaden	161			151		10								
Mud crab	1			1										
Mud snail	81			81										
Mummichog	4361	48	47	818		511		1174		4	2		1042	715
Northern pipefish	12					2							3	7
Pollock	2										2			
Sand lance	1												1	
Sheepshead	20			4		11		5						
Striped killifish	1659		18	481		150		996		1	8		5	
Threespine stickleback	6					4				1	1			
Total Organisms	9587													
# hauls	15	2	1	1		2		1		2	2		2	2

2.3.2. Trawl and Dredge Sampling

The most commonly captured organisms captured by the trawl were fourspine sticklebacks (n = 2,132), young-of-the-year (YoY) sea herring (*Clupea harengus*, n = 172), and Atlantic silversides (n = 111) and the rock crab (*Cancer irroratus*, n = 356). These four species accounted for 75% of the total catch (table 2.3).

Table 2.3. Organisms captured by trawl sampling, June 2015-June 2016.

Common Name	Total	Jun 2015	Jul 2015	Aug 2015	Sep 2015	Oct 2015	Nov 2015	Dec 2015	Jan 2016	Feb 2016	Mar 2016	Apr 2016	May 2016	Jun 2016
American eel	20	2									1	17		
American lobster	1				1									
Atlantic menhaden	2		1				1							
Atlantic moonfish	2				2									
Atlantic silverside	111				69		42							
Atlantic tomcod	5						1	1				2		1
Bay scallop	55				52		8							
Bittium	9			1	3		4					1		
Blue crab	15	4			10									1
Blue mussel	43		0	23			20							0
Bubble shell	1				1									
Common periwinkle	27		19	2	1		5							
Cunner	63				35		23	3			2			
Fourspine stickleback	2132	9	34		1343		675	2			29	35		5
Green crab	63		16	22			12	6			2	4		1
Grubby	9			1			7					1		
Gulf Stream flounder	3											1		2
Hermit crab	10		3	2	3		2							
Horseshoe crab	16		8	3	4									1
Lady crab	8		2	4	1									1
Longfin squid	64		16	6	8									34
Lumpfish	7						5	2						
Mud crab	23		13		3		1				3	1		2
Mummichog	73				73									
Northern pipefish	30		2		12			2				10		4
Oyster drill	42	2	7	1	18		8					1		5
Pollock	2			2										
Red hake	36		3	2			26	3						2
Rock crab	356		83	188	38		14	13			1	6		13
Rock gunnel	4		1	1										2
Sand lance	73			2	26		44							1
Sculpin	2						2							
Scup	16				16									
Sea herring	172						1					171		
Sea star	59		17	8	17		8	1				3		5
Seaboard goby	6						6							
Spider crab	38	2	12	7	1						2	6		8
Spotted hake	1		1											
Striped killifish	1				1									
Tautog	1				1									
Threespine stickleback	2											2		
Transverse ark	2						2							
Winter flounder	60	2	9	8	4		11	3			2	7		14
Total Organisms	3670													
# tows	90	2	17	6	11		18	6			6	11		13

Table 2.4. Organisms captured by dredge sampling, June 2015-June 2016.

Common Name	Total	Jun 2015	Jul 2015	Aug 2015	Sep 2015	Oct 2015	Nov 2015	Dec 2015	Jan 2016	Feb 2016	Mar 2016	Apr 2016	May 2016	Jun 2016
American eel	2	1										1		
Bay scallop	36	3			10		18	1				4		
Bittium	7		2	3							1			1
Blood ark	2		2											
Blue crab	7	3			2		1							1
Blue mussel	13	1	0	12	0		0							
Bubble shell	28	28												
Channeled whelk	5	1	2				1					1		
Common periwinkle	27		24				2					1		
Cunner	6						3	3						
Dog whelk	3	3												
Eastern oyster	2											2		
Fourspine stickleback	181	12	14		7		110	2			1	33		2
Green crab	13		1	2			7				1	1		1
Grubby	3						2	1						
Hermit crab	25	5	6	7	1		3	2				1		
Horseshoe crab	13	1	2	1	4							1		4
Knobbed whelk	20	3	9	1	2		1					1		3
Lady crab	3			1	2									
Mud crab	34	4	9		4		12					2		3
Mummichog	4						4							
Northern pipefish	6				1		1				1	3		
Oyster drill	64	16	7		12		18				1	5		5
Quahog	2						1							1
Red hake	1						1							
Rock crab	256	6	59	62	35		54	7			5	13		15
Rock gunnel	4		1				1	2						
Sea robin	1													1
Sea star	234	1	51	13	122		20	3			6	12		6
Seaboard goby	31	2	1		4		20	1				2		1
Spider crab	51	9	18	2							4	10		8
Striped killifish	1											1		
Surf clam	1										1			0
Transverse ark	8		6		1							1		
Winter flounder	2		1				1							
Total Organisms	1096													
# hauls	102	7	19	6	12		22	6			6	12		12

The high relative abundance of fourspine sticklebacks was biased upwards by one tow with very high abundance (that was not repeated) in eelgrass habitat in September 2015, although it would still have been the dominant species with that sample removed from the data.

The most commonly captured organisms captured by the dredge were the rock crab (n = 256), sea stars (*Asterias sp.*, n = 234), the oyster drill (*Urosalpix cinerea*, n = 64), and the fourspine stickleback (n = 181). These four species accounted for 66% of the total catch (table 2.4).

More dredge samples were collected than trawl samples due to the tendency of the trawl to become clogged with eelgrass or marine algae in some areas. Species diversity of trawl samples was greater than seine and dredge catches (tables 2.2-4). Sand and grass shrimp (*Crangon* and *Palaemonetes* spp.) were noted but not consistently counted.

2.3.3. Ventless Lobster Traps

Two strings of five ventless lobster traps were set and hauled during six subtidal surveys. Soak times ranged from 2-3 days. The most abundant species captured in the traps were the rock crab and the spider crab (*Libinia* sp.). Only one lobster was captured (8.6 cm CL), on 2 July 2015. See Appendix 1 for a full list of species captured in ventless lobster traps.

2.3.4. Gillnet Sampling

The experimental gillnet was set three times (2/27 July and 6 October 2015). Soak times ranged from 3.5 to 5.5 hours. No fish were captured. The only animals captured in the net were crabs, which proved difficult to remove without damage. Due to the destructive effects of the net on non-target species, the lack of fish catch, and the presence of seals in the area, no further gillnet sets were attempted. See Appendix 1 for a full list of invertebrates captured during gillnet sampling.

2.3.5. Passive Collectors

A total of 26 subadult lobsters were recovered from the passive collectors (table 2.5), all but one of which could be defined as, "early benthic phase" (EBP, ≤ 40 mm CL), after Wahle and Steneck (1991). Lengths ranged from 8 to 46 mm CL. Eleven YoY (≤ 13 mm CL[4]) postlarval lobsters were found (table 2.5). Total subadult lobster density averaged 2.4 organisms per square meter (orgs./m^2), while mean YoY density was 1.0 orgs./m^2 (table 2.5).

Table 2.5. Subadult American lobster (*Homarus americanus*) counts and densities (organisms per square meter), all specimens and young-of-the-year (YoY) by site (SD = Standard Deviation).

	All subadults			Young-of-the-Year (YoY)		
Site	Total count	Density (orgs./m^2)	SD	Total count	Density (orgs./m^2)	SD
Town Line	10	1.8	1.5	4	0.7	0.9
Quanset	16	2.9	1.8	7	1.3	1.5
Both	26	2.4	1.7	11	1.0	1.2

Several other species of invertebrate were documented, as well as several fish species (Appendix 1). Of note were juveniles of two species of tropical fish, the spotfin butterflyfish (*Chaetodon ocellatus*) and snowy grouper (*Hyporthodus niveatus*).

[4] EBP lobsters ≤ 13 mm CL are considered to be YoY in southern New England; D. Perry, MADMF, personal communication

2.3.6. Opportunistic Sampling

Several species not otherwise detected by other means were documented via opportunistic means and are noted in Appendix 1. Small ‘schoolie’ striped bass were collected during beach seining at Pleasant Bay Community Boating on 25 May 2017 during the BioBlitz, and recreational anglers were observed catching them in the River. Seine sampling in summer 2016 targeting tropical species captured two specimens of the white mullet (*Mugil curema*). Trawl surveys in summer and early fall 2017 targeting tropical species sampled a subset of the 2015-16 trawl stations and captured Atlantic mackerel (*Scomber scombrus*), butterfish (*Peprilus triacanthus*), bay anchovy (*Anchoa mitchilli*), windowpane (*Scopthalmus aquosus*) and mantis shrimp (*Squilla empusa*), none of which were recorded during 2015-16 sampling. Passive collectors targeting tropical fish in summer 2016 and 2017 captured black sea bass (*Centropristis striata*), which had not been caught in other gears. Minnow traps captured Asian shore crabs (*Hemigrapsus sanguineus*), which were not captured in other gears.

2.3.7. Seasonal Relative Abundance and Size Composition of Select Species

Atlantic silversides were among the most numerically dominant fishes captured during seine and trawl sampling (tables 2.2-3). Since silversides were consistently captured in seine sampling, the species was an ideal candidate for examining seasonal trends in abundance and size. Greatest relative abundance (number of fish per haul) observed by seine sampling occurred in October 2015 (figure 2.3). Size of silversides ranged from 3.5 to 12.5 cm FL and increased over the year from July-December to January-June (figure 2.4).

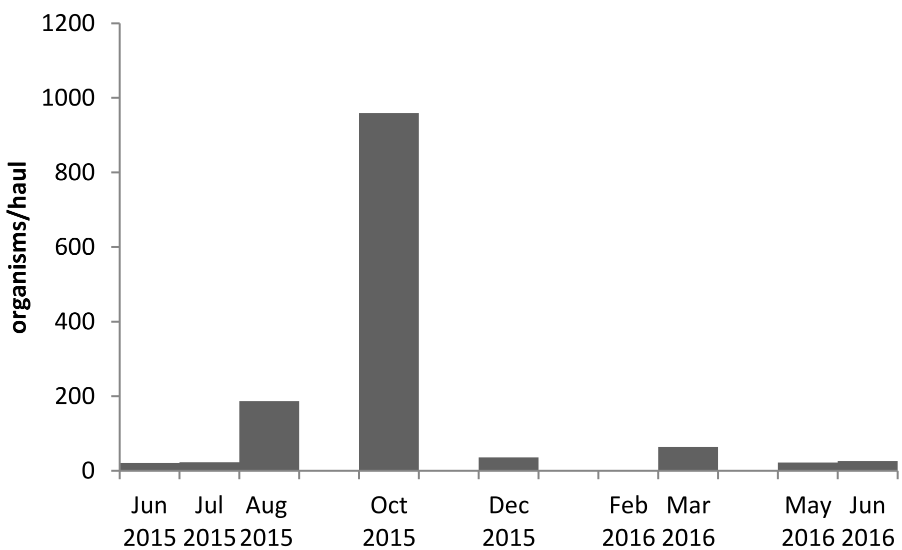

Figure 2.3. Seasonal relative abundance (organisms/haul) of Atlantic silversides (*Menidia menidia*) captured by seine sampling, June 2015-June 2016.

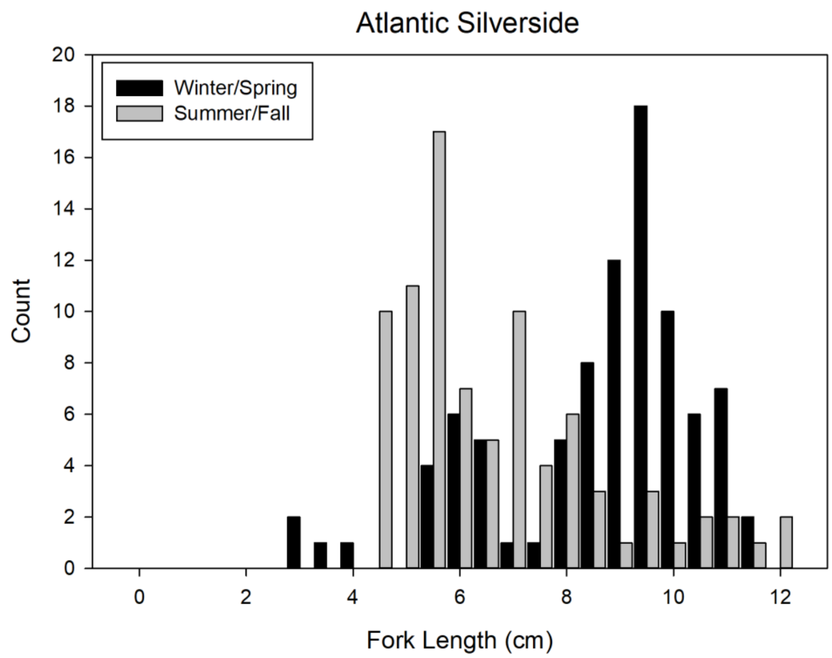

Figure 2.4. Seasonal length frequency of Atlantic silversides (*Menidia menidia*) captured by seine sampling, June 2015-June 2016. For the purposes of this figure, winter/spring are defined as January-June, and summer/fall are defined as July-December.

Winter flounder were consistently captured during trawl sampling (table 2.3). Greatest relative abundance (number of fish per tow) observed by trawl sampling occurred in August 2015 (figure 2.5). Size of winter flounder ranged from 9 to 19 cm TL (figure 2.6).

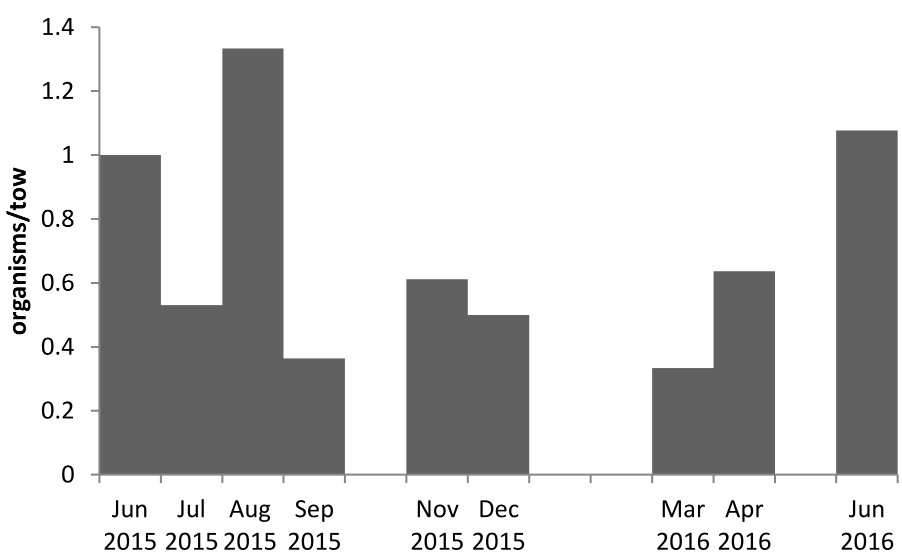

Figure 2.5. Seasonal relative abundance (organisms/tow) of winter flounder (*Pseudopleuronectes americanus*) captured by trawl sampling, June 2015-June 2016.

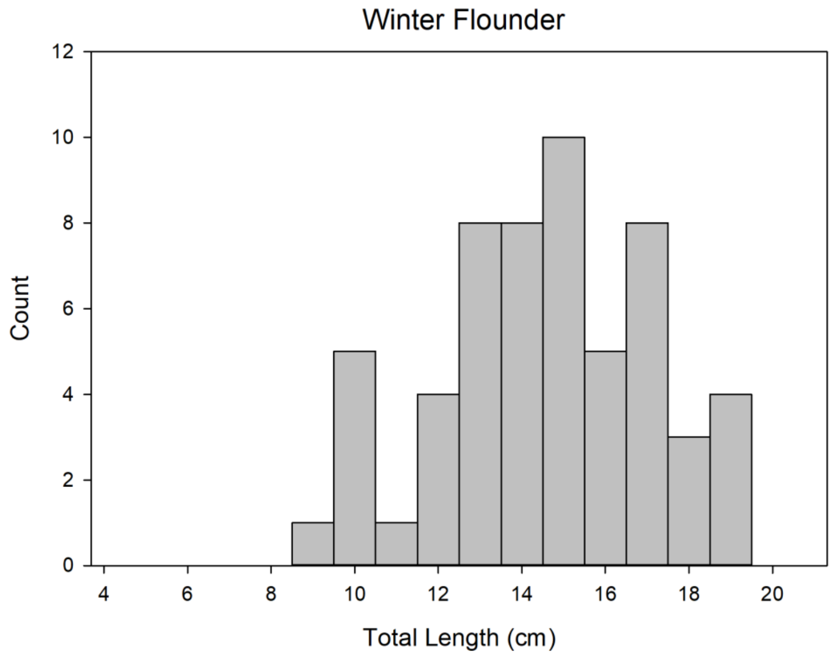

Figure 2.6. Length frequency of winter flounder (*Pseudopleuronectes americanus*) captured in trawls, June 2015-June 2016.

Longfin squid were captured seasonally during trawl sampling (table 2.3). Greatest relative abundance (number of squid per tow) observed by trawl sampling occurred in June 2016 (figure 2.7). Size of squid ranged from 2 to 19 cm mantle length (ML) (figure 2.8). Squid egg masses were captured in the trawl in July and September 2015.

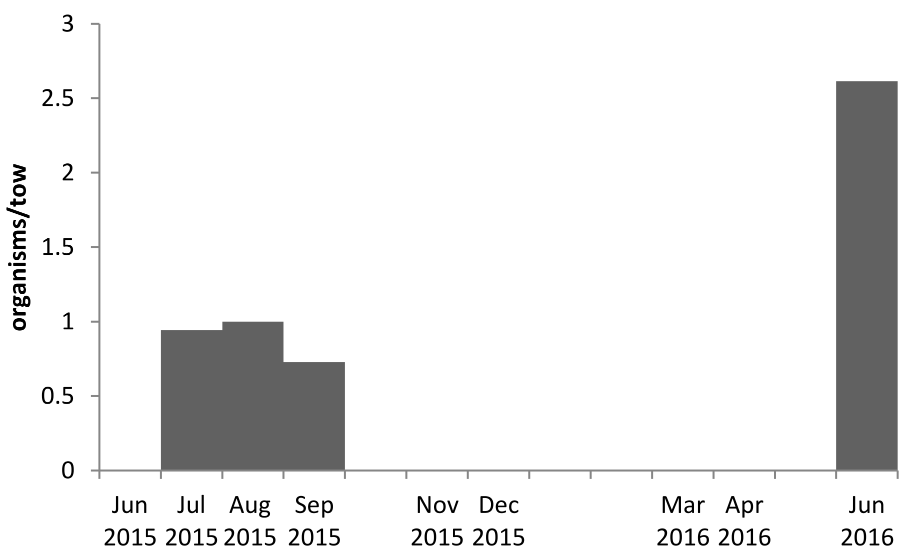

Figure 2.7. Seasonal relative abundance (organisms/tow) of longfin squid (*Doryteuthis pealeii*) captured by trawl sampling, June 2015-June 2016.

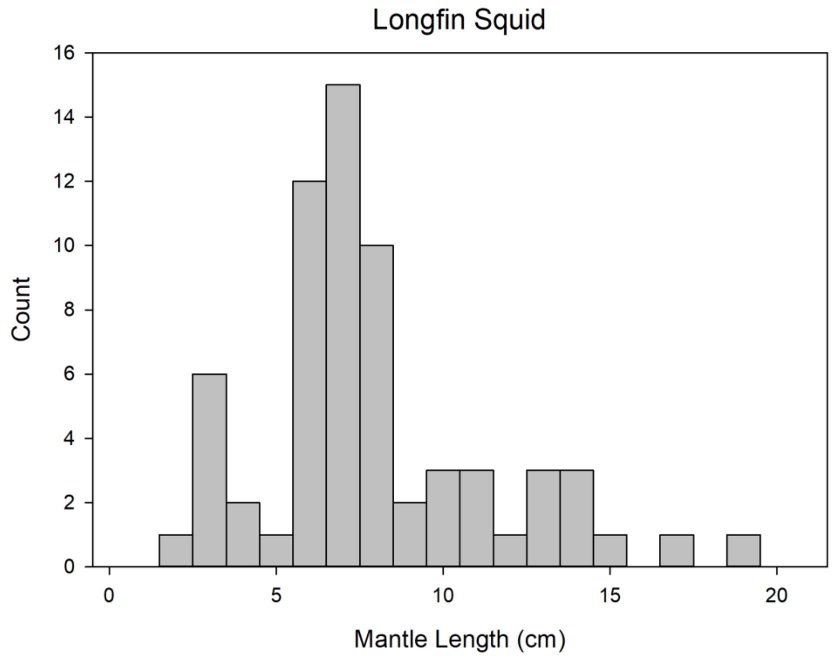

Figure 2.8. Length frequency of longfin squid (*Doryteuthis pealeii*) captured in trawls, June 2015-June 2016.

2.4. Discussion

This comprehensive inventory indicates that Pleasant Bay is home to a diverse assemblage of marine animals. Spatial relationships between species communities and the diverse habitats of the Bay are discussed by Legare *et al.* (2018). The overall species community and seasonal abundance of most species was broadly similar to that observed in the 1965-66 MADMF study. There were several species collected in this study that were not observed by Fiske *et al.* (1967) and vice versa (Appendix 1). This study included more sampling methods and greater spatial coverage than the previous study, and documented greater species diversity. Differences between the two studies are likely largely due to the variety of methods used in the present study and differences in spatiotemporal resolution between the two studies. The two seine stations sampled routinely during this study (figure 2.1) were also sampled monthly by Fiske *et al.* (1967); species diversity (Appendix 1) and seasonal trends in abundance for most species were relatively similar between the two studies. While peak abundance of some species occurred 1-2 months later in the year in 2015 than in 1965 (e.g. Atlantic silverside; table 2.2, figure 2.3), caution must be exercised in interpreting apparent differences between two years of data collected 50 years apart. Many of the trawl sites sampled in 1965 (Fiske *et al.*, 1967) are no longer accessible due to the dynamic seafloor environment of Pleasant Bay. Fish community composition and seasonal patterns of abundance during this study were broadly similar to those observed during other recent studies along the eastern shore of Cape Cod. A 1985-87 survey of Nauset Marsh (Able *et al.*, 2002) and a 2007-08 study of the surf zone off Coast Guard Beach in Truro (Estes, 2013) documented peak species abundance and diversity in summer and early fall, similar to observations during this study (tables 2.2-4).

Fourspine sticklebacks and some of the other dominant fish species (e.g. *Fundulus spp.*) sampled are ubiquitous in coastal systems in the region and are frequently among the most abundant (Fiske *et al*., 1967; Able *et al*., 2002). Forage species (e.g. sand lance, silversides, sea herring, and longfin squid) were well-represented in trawl surveys (table 2.3) as was observed in the Fiske *et al.* (1967) study. Winter flounder were among the most abundant species at stations sampled in 1965-66 (Fiske *et al*., 1967), but were relatively less abundant in the present study. Highly mobile fish species (e.g. striped bass, bluefish) were likely underrepresented during this study due to the sampling gears used. The collection of 'schoolie' juvenile striped bass during opportunistic beach seining and observations of anglers catching them suggests that they are present in the Bay but that a different capture method should be used to sample them. A standardized rod-and-reel survey would be ideal.

Although several species of bivalve mollusk were captured during the systematic surveys presented in this chapter, these surveys were not intended to provide abundance estimates for most species – see Nichols and Grieco (2018 Appendix 2) for results of dedicated shellfish surveys. It is notable that two of the four most abundant organisms captured in Bay-wide dredge sampling were specialist shellfish predators (sea stars and oyster drills). Bay scallops occurred in relatively low abundance. Two other commercially and recreationally important species, horseshoe crabs and blue crabs, were not abundant relative to some other invertebrates but this may be in part be a product of their depth distribution relative to the subtidal trawl and dredge surveys. The abundance of rock crabs is noteworthy – while not currently commercially valuable in local waters, there is an expanding interest in harvesting the species in Maine and marketing them as 'peekytoe' crabs.

Many fish and invertebrate species were found in the Bay in juvenile stages, but rarely at larger sizes (e.g. winter flounder, American lobster). All of the winter flounder captured in trawls were < 20 cm TL (figure 2.6) and were most likely ≤ 1 year old based on age-length relationships for the species in the Georges Bank region (Penttila *et al*., 1989). Juvenile winter flounder relative abundance was greatest in August 2015, an opposite pattern from that observed by Fiske *et al.* (1967), who documented two peaks in abundance in April and October 1965 (all sizes). While it is possible that the larger winter flounder up to 41 cm TL observed in the Bay by Fiske *et al.* (1967) avoided the sampling gear used in this study, the consistency of our methods with their earlier work renders this possibility unlikely. It is unclear what has caused the decline in abundance of winter flounder, particularly large fish in spawning condition, in Pleasant Bay. Fishermen reported declining catches to Fiske *et al.* (1967), and it is possible that winter flounder have changed their historical pattern of habitat use and are spawning offshore as has been observed in other estuarine habitats in the region (Decelles and Cadrin, 2010).

The YoY lobster density estimates observed in this study are higher than those recently observed elsewhere in Massachusetts using the same technique as part of the coast-wide American Lobster Settlement Index[5] (Wahle *et al.*, 2013). However, caution must be used in comparing these results, as the settlement collectors are primarily used to sample cobble habitats thought to be the primary settlement habitat for lobsters (Wahle and Steneck, 1991; Wahle *et al.*, 2013). Despite the use of ventless traps, their deployment at randomly selected stations may have caused lobsters to be

[5] https://umaine.edu/wahlelab/american-lobster-settlement-index-alsi/american-lobster-settlement-index/

underrepresented in the trap catches. The occurrence of larger juvenile lobsters in the collectors indicates the persistence of multiple cohorts in Pleasant Bay, as lobsters of this small size are unlikely to move in and out of the Bay. Targeted sampling of adult lobsters in the Bay based on local fishing community knowledge may provide further insights regarding residence of lobsters and the potential to support a fishery.

While the findings of this study may be affected by sampling bias for some species, it is apparent that the Bay is a nursery habitat for many species, including several commercially and recreationally important species. Spring presence of YoY sea herring was noted in the Bay by Fiske *et al.* (1967) as well as in Nauset Marsh to the north (Able *et al.*, 2002). Longfin squid were present in summer months (figure 2.7) and are among the species that apparently spawn in the Bay, as evidenced by the seasonal presence of egg masses and small juveniles, although based on size-at-maturity analyses (Hatfield and Cadrin, 2002), it is most likely that only the largest squid captured in trawls (figure 2.8) were sexually mature. The seasonal pattern in size of Atlantic silversides captured by seine (figure 2.4) was consistent with growth of this short-lived annual species through its entire lifecycle (Conover and Ross, 1982), although variation in relative abundance (figure 2.3) indicates that they may move in and out of the Bay seasonally. An understanding of the relationship between the occurrence of juvenile and adult organisms in Pleasant Bay and the waters east of Cape Cod is necessary to understand the potential contribution of nursery habitat in Pleasant Bay to sustaining populations inside and outside of the Bay.

This study employed a variety of sampling gears, each of which yielded different results even when deployed at the same station. While the greatest species diversity was observed in trawls, dredges towed at the same stations revealed species that may not have been caught by the trawl at all, or greater relative abundances of cryptobenthic species such as the seaboard goby (*Gobiosoma ginsburgi*, tables 2.3-2.4; Nichols and Van Tassell, in prep). Passive collectors captured fish species that were not otherwise represented in other gears (e.g. juvenile spotfin butterflyfish, snowy grouper, black sea bass). The incorporation of a third year of opportunistic trawl survey data added several new species to our inventory, including pelagic species such as mackerel and butterfish that had not previously been reported in the Bay. Given observed shifts in species communities in other estuarine habitats (e.g. Collie et al., 2008; Howell and Auster, 2012), long-term monitoring is necessary to place these observations in a broader context. The presence of tropical fishes in the study area warrants further investigation – this work has begun in Pleasant Bay as an offshoot of this study, in collaboration with the New England Aquarium and the Gulf Stream Orphan Project (M. O'Neill, Principal Investigator).

2.5. Conclusions/Recommendations

This comprehensive inventory indicated that Pleasant Bay is home to a diverse assemblage of marine animals. The standardized, replicable methods employed during this study established baseline data on distribution and relative abundance of a wide variety of animals, including those of commercial, recreational, and ecological importance.

Differences in species diversity and relative abundance were observed between this study and the 1965-66 MADMF study, as well as between years during this study. Caution must be exercised in interpreting differences between two years of data collected 50 years apart. Long-term monitoring is necessary to place these observations in a broader context. This could be accomplished via surveying a subset of trawl/dredge stations annually or every few years, in conjunction with planned educational activities on board a "Floating Classroom" currently under construction.

Trawl and dredge sampling at randomly selected sites captured a diverse array of fishes and macro-invertebrates in a standardized manner and provided quantitative indices of relative abundance. Targeted rather than random trap sampling for lobster may provide a better understanding of the use of the Bay by adult lobsters. Diver-based suction sampling is an alternative to the use of benthic collectors for YoY lobsters, and may be more applicable for future quantitative sampling of habitats in Pleasant Bay, although benthic collectors are a useful tool for initial identification of settlement habitat (Wahle *et al.*, 2013). A standardized rod-and-reel survey would be ideal for sampling large, highly mobile fish species such as striped bass and bluefish.

Pleasant Bay is spawning and nursery habitat for a variety of marine animals. An understanding of the relationship between the occurrence of juvenile and adult organisms in Pleasant Bay and the waters east of Cape Cod is necessary to understand the potential contribution of nursery habitat in Pleasant Bay to sustaining populations inside and outside of the Bay. This could be accomplished with tagging and telemetry studies to assess fine-scale patterns of habitat use and movements in and out of Pleasant Bay.

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CHAPTER 3

The seasonal distribution, counts and prey of harbor seals (*Phoca vitulina vitulina*) and gray seals (*Halichoerus grypus atlantica*) in Pleasant Bay, Cape Cod, Massachusetts

Sette L. M., C. Accardo, K. McKenna, K. Patchett, K. Rose, B. W. Sharp, G. T. Waring

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Abstract

Both harbor and gray seals regularly haul-out on tidal sand bars inside Pleasant Bay (PB). To begin to understand their distribution, numbers, and role in the ecosystem, monthly aerial surveys were flown in 2014 and 2015 and scat sampling at haul-outs were conducted monthly from January of 2016 through March of 2017. In 2014, harbor seals were observed inside PB on haul-outs from January through May. In 2015, the aerial team was unable to fly in January due to inclement weather. Flights resumed in February, and harbor seals were present February through May and again in December. The maximum daily count for harbor seals inside PB was 936 in February 2014 and 753 in March 2015. Gray seals were present in PB June through November 2014 and June through December 2015. The highest daily count for gray seals was 1276 in June 2014 and 2379 in August 2015. Harbor and gray seals both utilized tidal sand bars in Chatham Harbor for hauling out in both years. However, in 2015, as gray seal numbers increased inside PB, gray seal distribution shifted north to include a series of developing tidal sand bars west of Nauset Beach and southeast of Strong Island. Due to the increasing number of gray seals and the dynamic nature of the PB system, continued monitoring is recommended.

The diet of harbor and gray seals in southeastern Massachusetts inshore waters is not well studied. To build on previous research, we collected scat samples monthly inside Pleasant Bay. Prey was estimated based on the recovery and identification of fish otoliths and other hard parts (squid beaks, denticles from skate spp., crustacean carapaces, shells, bones and teeth) using frequency of occurrence (FO) analysis. For both harbor and gray seals sand lance *(Ammodytes* spp.*)* was recovered most frequently, FO 93% winter, 83% spring in harbor seal samples and 67% in spring and 89% fall in gray seal samples. The remaining prey species recovered for harbor seals were herring spp. (Clupeidae) 33% and cod spp. (Gadidae) 17% in winter and spring, blue mussel *(Mytilus edulis)* 13% and ocean pout *(Zoarces americanus)7* % were recovered in winter only. Gray seal prey beyond sand lance in summer and fall included longfin squid *(Doryteuthis pealeii)* 28% and 26%, blue mussel *(Mytilus edulis)* 28% and 7%, skate spp. (Radidae) 19% and 15%, Cod spp.(Gadidae) 7% and 9%, snail spp. (Gatrapoda) 3% and 5%, crustacean spp. (Crustacea) 4% & 5%, and flounder spp. (Plueronectidae) 5% in summer only. A minimum of 5 prey taxa were identified for harbor seals and 8 prey taxa for gray seals.

Key words: Aerial survey, harbor seal, *(Phoca vitulina vitulina)*, gray seal, *(Halichoerus grypus atlantica*), distribution, counts, diet, hard part analysis, Pleasant Bay (PB).

3.1. Introduction

3.1.1. Seal Species and Study Area

Harbor seals are distributed in the North Atlantic and North Pacific oceans. The sub-population found in the northwest Atlantic is *Phoca vitulina vitulina* (King, 1980). Harbor seals are present in southeastern Massachusetts in fall, winter and spring and move north for pupping and breeding in May (Schneider and

Payne, 1983; Waring *et al.,* 2006). The most current abundance estimate for U.S. waters for harbor seals is 75,834 (CV=0.15). This estimate is based on aerial surveys completed in 2012 (Waring *et al.* 2015).

Gray seals *(Halichoerus grypus atlantica)* are year round residents in southeastern Massachusetts, including breeding colonies on Muskeget and Monomoy Islands, and distributed only in the North Atlantic and Baltic Sea (King, 1983). There is no current abundance estimate for gray seals in U.S. waters at this time (2016 NOAA SARS).

Pleasant Bay, including Chatham Harbor, is known to be an important area for invertebrates, fish, birds and marine animals to congregate for food and refuge (Fiske *et al.* 1967). The barrier beaches to the east and the islands that border and exist within the boundary of PB provide a natural buffer to wave energy and winds that come off the North Atlantic. The active shoaling and resulting tidal sand bars that develop provide ideal habitat for both harbor and gray seals to haul-out. The most persistent haul-outs were identified prior to and during this study and are included here.

Figure 3.1. Gray seal (top) and harbor seal (bottom) at Chatham Harbor haul-out in October, 2016

3.1.2. Aerial Survey of Pleasant Bay

For over 30 years harbor and gray seal aerial survey efforts in southeastern Massachusetts have focused on winter and spring distribution and abundance of harbor seals outside pupping and breeding season, as well as gray seal pup counts in winter in Nantucket Sound (Knapp & Winn,1978 ; Kraus, 1980; Schneider et al., 1980; Prescott, 1982; Payne & Schneider, 1983; Rosenfeld et al., 1988; Payne & Selzer, 1989; Early et al., 1995; Rough, 1995; Barlas, 1999; NEFSC, 2005-2011; deHart, 2002; Wood Lafond, 2009; Waring et al., 2015a; Johnston *et al.* 2017).

Barlas (1999) conducted surveys from October 1998 through June1999 to monitor harbor seal and gray seal distribution in southern New England. Harbor seals were observed on haul-outs in Chatham Harbor from

December 1998 through April, 1999. The highest daily count of harbor seals in Chatham Harbor was 2389 individuals in February of 1999. Gray seals on the other hand, were only observed on two occasions, in January and March 1999, with two seals present at one haul-out, on each day. These observations represent the first insights in distribution and counts for both harbor and gray seals inside PB (based on the investigator's knowledge).

3.1.3. Seal diet analysis

Previous studies on harbor and gray seal diet in Southern New England suggest that both species forage on a wide variety of pelagic and benthic species which are seasonally available (Payne & Seltzer, 1989; Ferland, 1999; Williams, 1999; Waring *et al.* 2000; Wood, 2001; Ampella, 2009; Wenzel *et al.* 2017). The goal of this study is to describe the prey species recovered and identified from harbor and gray seal scats collected at haul-outs inside Pleasant Bay.

3.2. Methods

3.2.1. Aerial Survey of Pleasant Bay

Aerial surveys were flown monthly under NOAA Permit No. 17670 in a Cessna Skymaster (models 337; N2697S), a twin-engine high wing aircraft. Surveys were conducted at a standard altitude of approximately 230 meters (750 feet) and a ground speed of approximately 100 knots, using methodology developed by CeTAP (Scott and Gilbert 1982, CeTAP 1982). The survey team consisted of a pilot, co-pilot and one or two observers positioned in the rear seats. One observer photographed using a 7D Canon with a fixed 300mm lens. Settings on the camera had to be adjusted for light and wind conditions. However, the team preferred to use a maximum of 400 ISO to minimize pixilation, a shutter speed greater than or equal to 1/1000 of a second to minimize blurred images, and an F stop of 8 or greater for increased depth of field. Whenever a second observer was on board, they assisted with data collection. If only one observer was available, site data was recorded on a voice recorder. Surveys were scheduled on days with good visibility, no precipitation and with winds below 25 knots. To capture the greatest number of seals hauled out, surveys were flown within two hours of low tide (dependent on weather and light). The observers scanned the area to determine where the haul-outs were set up and then circled the area to photograph. Any opportunistic sightings of whales, sea turtles or sharks were also noted. Track line data was recorded in Logger 2000 and entered in a format compatible with CCS and Northeast Fisheries Science Center (NEFSC) databases. For daily minimum counts and speciation, standard image processing software was used to stitch together images in order to create an image of an entire haul-out site (Josephson *et al.,* 2015). Counts were done by manually marking all identified seals in the images. Counts by image, species, site, date, and general age class were entered into an Oracle database. Other data collected and recorded from the imagery included evidence of entanglement or other human interactions, any brands, tags or distinctive natural markings, and occasional comments on behavior observed. NEFSC (Josephson *et al.,* 2015) completed the final counts. The counts do not include seals observed in the water, therefore, the counts should be considered the minimum number present on any given survey date (Josephson *et al.,* 2015).

3.2.2. Seal diet analysis

In 2016, a total of 25 harbor seal and 63 gray seal scats were collected at haul-out sites inside PB (figure 3.2). Monthly boat based surveys were conducted inside PB at established haul-outs identified by CCS in previously conducted aerial surveys (2014 and 2015) (figure 3.2). When new haul-outs were observed and were accessible for speciation and landing, samples were collected. Collections were timed with low tide (Payne and Selzer, 1989; Wood, 2001; Ampella, 2009; Raposa, 2009) to allow for the greatest number of seals present at a given site, and to encounter more available scats for collection. Samples were collected only at sites where minimal species overlap was observed. Once a haul-out site was selected and species present were determined, the team landed adjacent to the group of seals and slowly flushed the animals. Scat collection was completed using a slotted scoop that allowed the collector to minimize bycatch of sand and other materials that could potentially contaminate or bias the scat sample. Each individual sample was placed inside a sterile plastic freezer bag, labeled with the date, location and species and stored in at -20^{o} C^{0} until processing at CCS. Scat collection data sheets were completed for each monthly collection and reported to the NEFSC.

Individual scat samples were processed at the CCS wet lab in Provincetown, MA. Scats were first thawed and then gently washed with warm water and soap in a stack of graduated sieves of 2.0 mm, 1.0 mm and 0.5 mm mesh size (Wood, 2001; Ampella, 2009). The remaining hard parts were left to air dry and then stored in sterile, airtight containers. Diet was estimated based on the recovery and identification of hard parts: sagittal fish otoliths, cephalopod beaks, dermal denticles from skate species, crustacean carapaces, shells, bones and teeth. Prey species recovered from the scats were identified based on Harkonen (1986) and Campana (2004). Prey remains were identified to lowest prey taxon possible. Fred Wenzel, NEFSC, provided additional help with identification of cephalopod beaks and skate species. Including dermal denticles from skates, bones and teeth from fish, can help increase the species and number of prey recovered during analysis (Bowen, et al., 2002). Frequency of occurrence (FO) analysis was used to estimate the percentage of scats containing each prey taxa (Lance *et al.*, 2001).

Figure 3.2. CCS and IFAW staff collecting scat samples at Chatham Harbor haul-out (top left), harbor seal scat (top right), stack of sieves for scat processing and hard part recovery (bottom left), and sand lance otoliths with ocean pout jaw and teeth recovered from harbor seal scat sample (bottom right)

3.3. Results

3.3.1. Aerial Survey

Harbor seals were present in winter and spring from January through May of 2014 and February through May of 2015 (figure 3.3). The plane was unable to survey during January 2015 optimal tide cycles due to poor weather conditions, therefore, no data was collected. The maximum counts for harbor seals were 936 in February 2014 and 753 in March 2015. The total number of haul-outs and locations changed between years for both species. Harbor seals occupied one haul-out site (A) in Chatham Harbor in 2014 and two haul-outs sites, (A & B) inside Chatham Harbor in 2015 (figure 3.3).

Gray seals were present June through November in 2014 and June through December in 2015 (figure 3.3). The maximum daily counts for gray seals were 1276 in June 2014 and 2379 in August 2015 (figure 3.3). Two haul-out sites (A & B) inside Chatham Harbor were occupied by gray seals in 2014 (figure 3. 4). In 2015, gray seal numbers increased and individuals were observed on haul-out A, and on haul-out G, a tidal sand bar west of Nauset Beach and southeast of Strong Island (figures 3.3 & 3.4).

No opportunistic sightings of whales, sea turtles or sharks were recorded during the aerial surveys of Pleasant Bay in 2014 and 2015 or during boat based surveys for scat collections in 2016.

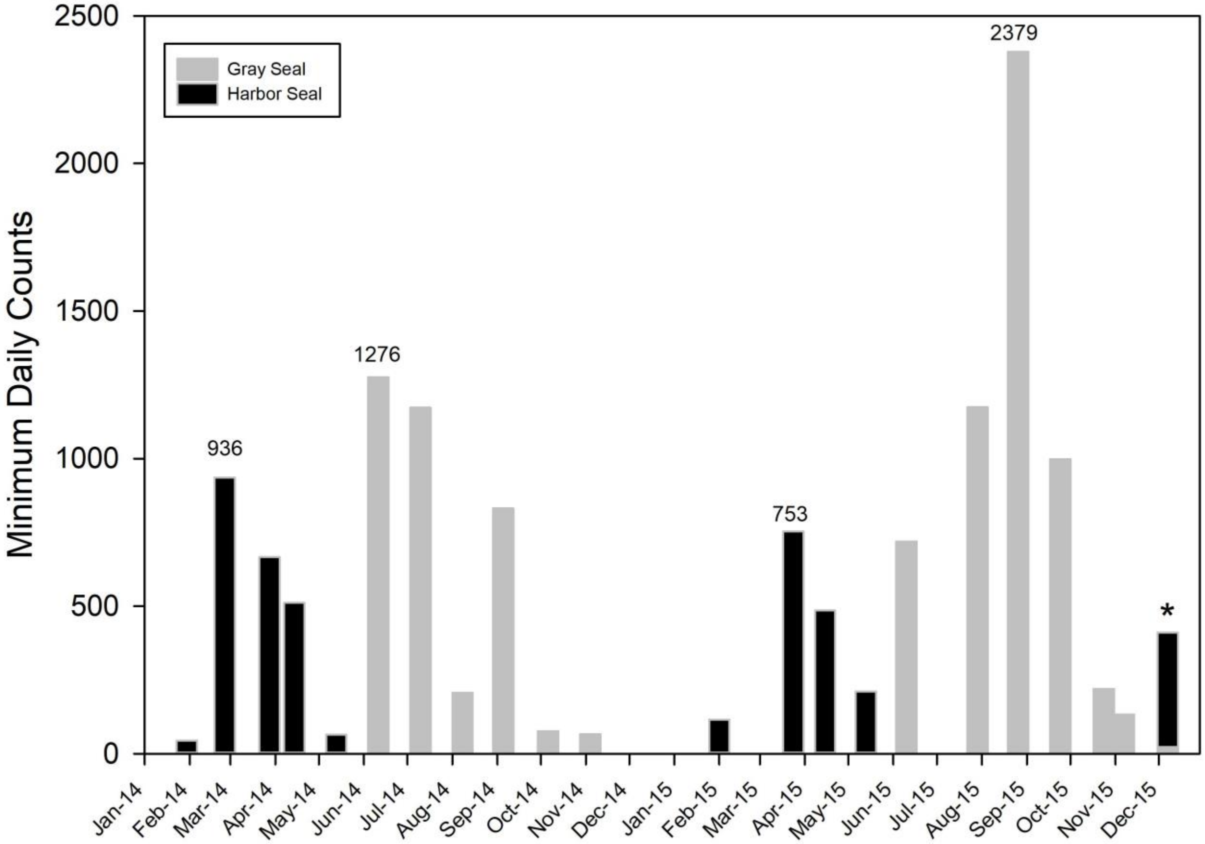

Figure 3.3. Minimum daily counts for harbor seals and gray seals at haul-outs inside Pleasant Bay (Josephson *et al.*, 2017). Asterisk (*) indicates months in which harbor and gray seals overlap.

Figure 3.4. Distribution of existing harbor seal and gray seal haul-out sites inside Pleasant Bay.

Figure 3.5. Harbor seal haul-out site "G" inside Chatham Harbor in winter 2015.

3.3.2. Seal diet analysis

In total, 25 harbor seal scat samples were collected for winter and spring, with 21 containing identifiable hard parts; four taxa of fish and one mussel species were identified (table 3.1). Frequency of occurrence (FO) was used to calculate the presence of prey recovered from the total number of scats collected (Lance *et al.*, 2001).

Sand lance was the most frequently recovered prey for harbor seals with 93% of winter scat samples and 83% of spring scat samples containing hard parts. Herring spp. and cod spp. were the second most common prey items with 33% of winter scat samples and 17% of spring scat samples containing hard parts of these fish families. Blue mussel shells and ocean pout were only found in winter scat samples (13% and 7% respectively) (table 3.1, figure 3.6).

Table 3.1. Sample data and prey species identified from harbor seal (*Phoca vitulina vitulina*) scats (N=25) from Pleasant Bay, Massachusetts. Prey types are listed in decreasing order of frequency of occurrence (FO). The seasons are represented as follows: winter (December - February); spring (March - May); summer (June - August) and fall (September -November).

	Winter	Spring	Summer	Fall
Total # of scat samples collected	17	8	0	0
Scat containing >1 identifiable pry	15	6	0	0
Scat containing no identifiable prey	1	1	0	0
Scat containing no prey	1	1	0	0
Identifiable Prey (%FO)				
Sand lance (*Ammodytes spp*.)	93%	83%		
Herring spp. (Clupeidae)	33%	17%		
Cod spp. (Gadidae)	33%	17%		
Mussel spp. (Mytilidae)	13%	0%		
Ocean pout (*Zoarces americanus*)	7%	0%		

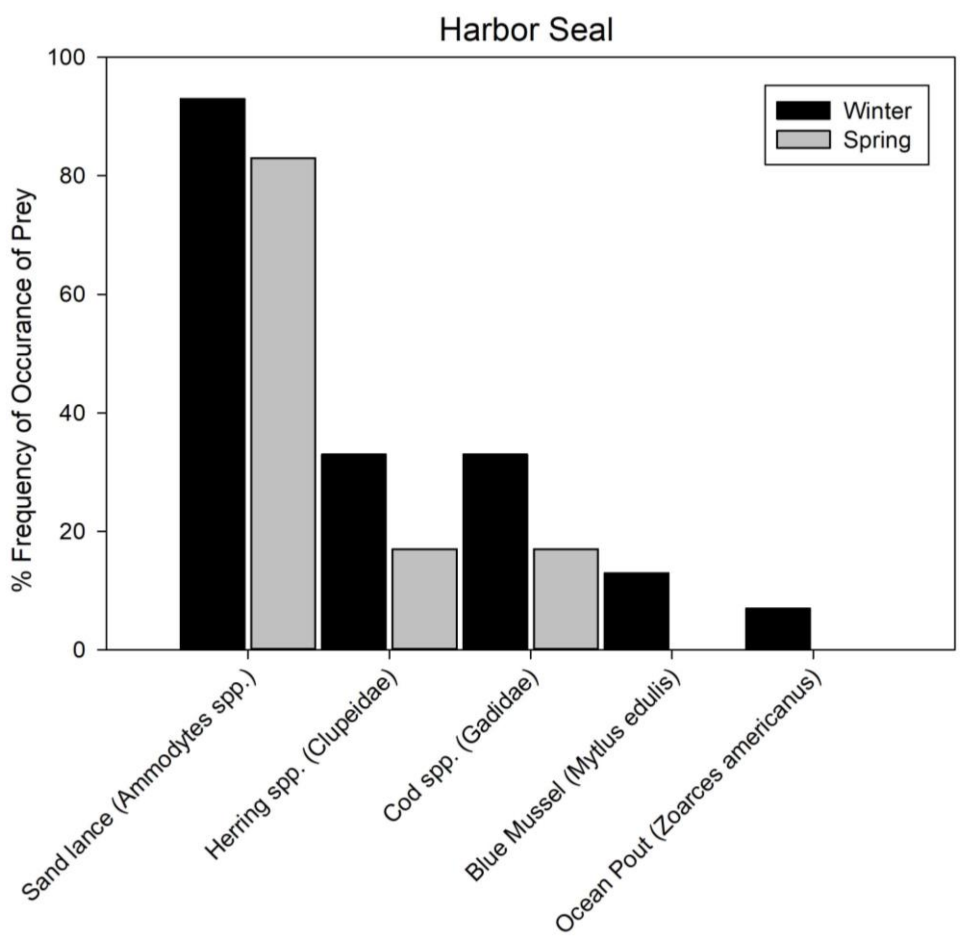

Figure 3.6. Estimated % Frequency of Occurrence of prey taxa in harbor seal scat samples.

A total of 63 grey seal scat samples were collected in summer and fall, with 48 containing identifiable prey (table 3.2). Four fish taxa, one squid species, as well as mussel, snail, and crustacean species were identified. Like harbor seal scats, sand lance was also the most frequently recovered prey species in gray seals, with 67% of summer scat samples and 89% of fall scat samples containing hard parts of this fish species. Longfin squid beaks were recovered (summer: 28%, fall: 26%) followed by blue mussel shells (summer: 28%, fall: 7%), skate spp. (summer: 19%, fall: 15%) and cod spp. (summer: 7%, fall: 9%). Fragments of snail spp. and crustacean spp. were found in 3% and 4% of summer samples and 5% of fall samples respectively. Hard parts of flounder spp. were only found in summer samples (5%) (table 3.2; figure 3.7). Blue mussels, gastropods and crustaceans were included in the analysis, however, it is not known if these are secondary prey items (prey of species that were consumed) for harbor and gray seals or targeted prey species in the seals diet.

Table 3.2. Sample data and prey species identified from gray seal (*Halichoerus grypus atlantica*) scats (N=63) from Pleasant Bay, Massachusetts in 2016. Prey types are listed in decreasing order of frequency of occurrence (FO). The seasons are represented as follows: Winter (December through February); spring (March through May); summer (June through August) and fall (September through November).

	Winter	**Spring**	**Summer**	**Fall**
Total # of scat samples collected	0	0	32	31
Scat containing >1 identifiable pry	0	0	21	27
Scat containing no identifiable prey	0	0	3	2
Scat containing no prey	0	0	8	2
Identifiable Prey (%FO)				
Sand lance (*Ammodytes spp*.)			67%	89%
Logfin Squid (*Doryteuthis pealeii*)			28%	28%
Shellfish (Bivalvia)			28%	7%
Skate (Rajidae)			19%	15%
Cod (Gadidae)			7%	9%
Snails (Gastropoda)			3%	5%
Crabs (Crustaceans)			4%	5%
Flat fish (Pleuronetidae)			5%	0%

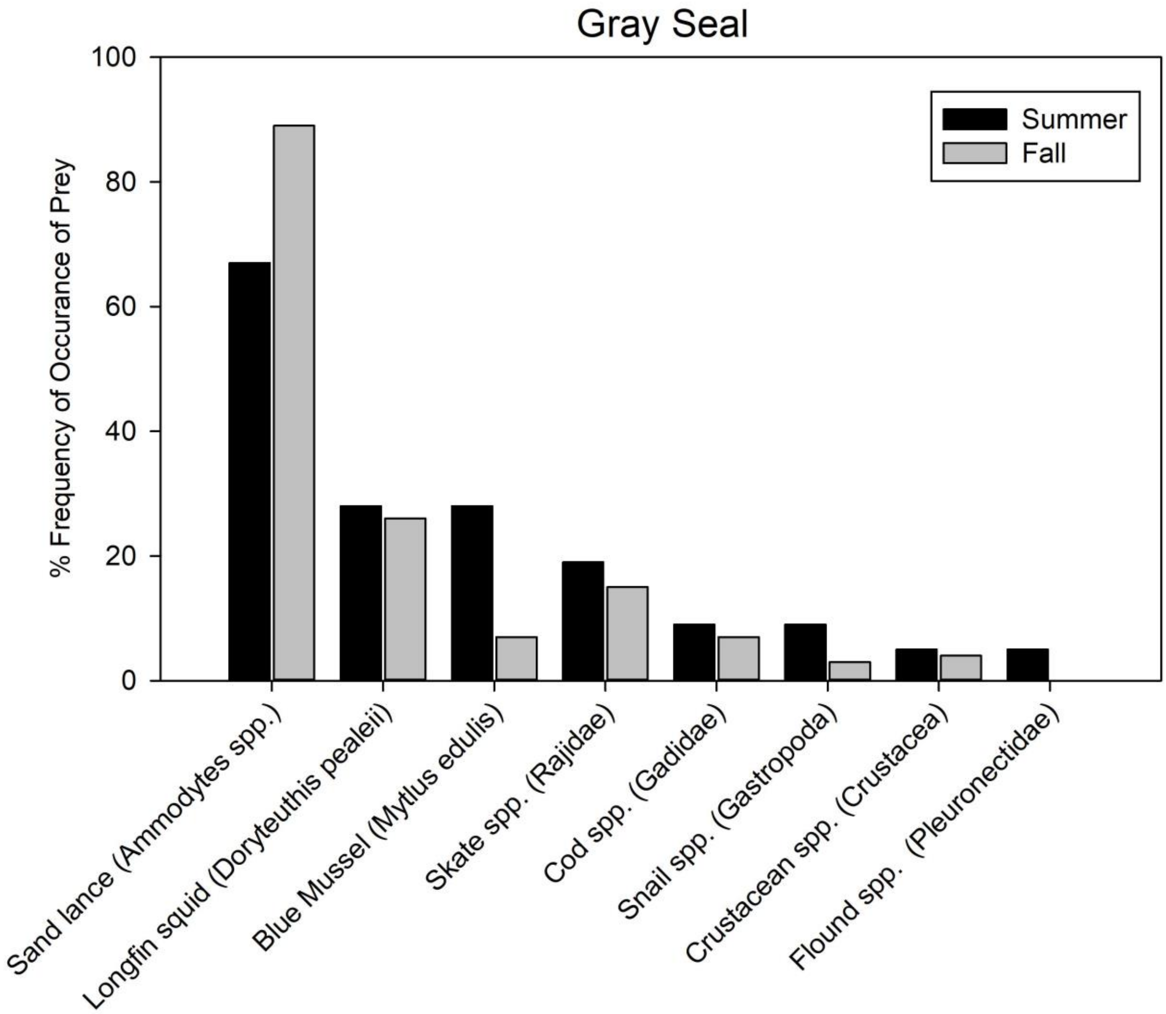

Figure 3.7. Estimated Frequency of Occurrence of prey in gray seal scat samples.

3.4. Discussion

3.4.1. Aerial Survey

Surveys showed that harbor seals inhabited PB seasonally in winter and spring, while gray seals utilize PB in summer and fall. Both species overlapped with each other only in December of 2015 (figure 3.3). We expected to observe more overlap in fall based on previous aerial work (Barlas,1999; NEFSC, 2005-2011) and boat based observations completed by CCS and NEFSC between 2007 and 2013 (Sette, unpublished data). However, activities in the area prior to surveys (e.g. shellfish harvesters, vessel traffic, seal watches or large rafts of sea ducks taking flight have been known to flush entire haul-outs and skewed the survey results. Previous studies have shown that disturbances such as these can influence haul-out behavior, especially in harbor seals, and can result in under representation of species (Bartholomew, 1949; Newby, 1973; Paulbitski, 1975, Allen et al., 1984 and Sette, *unpublished data*). The seasonal movements of harbor seals are consistent with the behavior of seals in southeastern Massachusetts waters. Inside Pleasant Bay, harbor seals were present December through May and absent June through November, during pupping and breeding season. This was shown by Waring (2001), who tagged 12 seals in Chatham Harbor in March 2001 and showed that 75% of the seals tagged moved north, off the coast of Maine, during pupping, breeding and molting season.

The absence of gray seals in winter and spring is consistent with the pupping and breeding behavior for this species in the Northwest Atlantic area (Allen, 1888; Hannah; 1998; Rough, 1995, 2000; Wood Lafond, 2009). Gray seals dispersed into PB in June, post molting, and left PB in January, to move on to pupping and breeding colonies in the US or Canada (Hanna, 1998; Rough, 1995,2000; Wood LaFond, 2009). Historically, before gray seals began to recolonize Massachusetts waters in the 1980s, the most populated seasonal harbor seal haul-out was on South Monomoy Island in winter (Kraus, 1980; Payne & Selzer, 1989). Kraus (1980) and Payne & Selzer (1989) did not observe harbor seals inside PB during aerial surveys completed in 1980 and between 1983 -1987. Rough (1995) began observing gray seals recolonizing Muskeget Island and South Monomoy Island in the mid-1980's during the pupping, breeding and molting season (Rough, 1995,2000). When Barlas (1999) completed the 1998-1999 survey work, gray seals most likely had displaced harbor seals off South Monomoy. During the time period between 1987, when Payne and Selzer (1989) flew surveys, and 1999, when Barlas (1999) completed the Southern New England aerial census, both harbor and gray seals were observed starting to colonize PB. However, the transition was not captured until the Barlas (1999) flights.

The observed 2015 northward shift of gray seals that coincided with their increasing numbers of gray seals in PB, is of particular interest. The storm events in July 2007 triggered flooding and a subsequent break-through of the north-south barrier beaches, creating a new inlet. Since then, shoaling inside the break has been changing rapidly, creating tidal sand bars between Nauset Beach and Strong Island that are ideal haul-out habitat for seals. On August 26th, 2015, seals occupied the western edge of the sandbars west of Nauset Beach (figure 3.8) and their increase in numbers observed during the August survey was surprising. The highest count for 2014 was 1276 in June. The August 2015 count increased 83% to 2379. The gray seal population in United States waters is believed to be increasing (2016 NOAA SARS). Therefore, it may be that seal numbers will continue to rise inside PB and their distribution inside PB could expand. In summer of 2016 during scat collections, the tidal sand bars "C", "D" and "E", (figure 3.4) were used consistently during the maximum low tides cycles by gray seals. In the fall, beginning in October, site "F" on the north end of North Beach Island was occupied and at site I on the south end of North Beach Island was used as well (figure 3.4).

In the winter of 2017, CCS and IFAW were surveying PB for harbor seal haul-outs and discovered them hauled out on site "G" (figure 3.4). During drone flights in June of 2017, while CCS assisted Dr. Michael Moore and IFAW with gray seal entanglement detection inside PB the expansiveness of the haul-out was noted (figure 3.9). It may very well be that the northern shoals become the dominant haul-outs for both species.

Figure 3.8. Gray seal haul-out site "G" on tidal sand bars west of Nauset Beach and southeast of Strong Island in summer of 2015.

Figure 3.9. UAS image of gray seal haul-out "G" west of Nauset Beach in June of 2017. The Image was taken by Dr. Michael Moore under NOAA Permit.18786. The drone was flown in support of IFAW's seal disentanglement efforts inside Pleasant Bay.

3.4.2. Seal diet analysis

Both harbor and gray seals take advantage of seasonally abundant prey species found in southern New England waters (Payne and Selzer, 1989; Ferland, 1999; Ampella, 2009). The predominance of sand lance in the diet of both species of seals is consistent with previous studies on diet conducted in New England (Payne and Selzer, 1989; Ferland, 1999; Ampella, 2009; Wenzel, 2017) and with the distribution of the species in Massachusetts waters (Bigelow and Shroeder, 1953; Nelson *et al,* 2004). Additionally, sand lance are most likely consumed whole by seals, thereby increasing the likelihood of recovering the sagittal otoliths (Bowen 2000; Arim and Naya, 2003). Although herring and ocean pout, both exhibiting a broad seasonal distribution in southern New England waters (Bigelow and Schroeder, 1953; APCC Report, 2016), were detected in harbor seal samples in this study, it is highly probable they are under-represented due to the delicate nature of their otoliths and bones (Bowen 2000; Arim and Naya, 2003).

Sand lance were the dominant prey recovered for gray seals in both summer and fall followed by longfin squid. Blue mussels, were recovered in summer and fall as well. Ampella (2009) recovered blue mussels in her three year study, but they were recovered infrequently >1%. Due to the distribution of mussel beds inside PB (see chapter one), and the high frequency of recovery, 28% in summer and 7% in fall, the mussels may be an important prey species for seals utilizing PB. Skate was present in both summer and fall in gray seal samples as well. Skates are bottom feeders and have a diverse diet that includes crustaceans, fish, squids, mussels, clams, oysters, worms, snails and even other skates (Bigelow and Schroeder, 1953). The mussels, marine snails and crustaceans that were recovered in this study could have been consumed by skates (secondary prey). However, that cannot be determined through hard part analysis alone. Skate and cod species were detected less frequently in gray seal samples, 7% summer and 9% fall. Whereas, in harbor seals samples, cod otoliths were recovered at 33% in winter and 17% in spring. In Ampella (2009) Cod spp. was recovered at >2% FO in gray seal samples.

On several scat surveys in 2016 inside and outside of Pleasant Bay, we observed individual gray seals eating spiny dogfish *(Squalus acanthias)* at the surface and pursuing large schools of menhaden (*Brevoortia tyrannus*) and Atlantic mackerel *(Clupea harengus)*. However, these species were not detected during scat analyses. Spiny dogfish and menhaden hard parts are rarely recovered in scat analysis. Ampella (2009) recovered >1% FO spiny dogfish and Atlantic mackerel out of 252 scats and never recovered menhaden. However, Toth (2017) recovered menhaden otoliths from harbor seal scats collected between 1996 and 2011 at haul-outs in a southern New Jersey estuary in winter and spring. Future diet studies should consider using decreased sieve mesh size to help aid in the recovery of smaller otoliths such as those found in menhaden.

While results are not available for this report, sub samples of gray seal scat collected in Pleasant Bay were provided to Keith Hernandez, a graduate student of Dr. Mike Polito at Lousiana State University for prey DNA analysis to be completed in 2018/2019. We anticipate the results from Hernandez to reflect these species and additional prey that might have been under represented in this study by employing prey DNA methodologies.

Micro Debris

Studies underway around the globe indicate that micro debris pollution could have serious impacts on marine species (Goldsworthy *et al.,* 1997; Eriksson & Burton, 2003; Bravo Rebolledo *et al.*, 2013; Kuhn *et al.* 2015; Rochman *et al.,* 2015; Ryan *et al.,* 2016; Nelms *et al.*, 2018). During the processing of scat for this study, special care was taken to minimize contamination of the samples. In four samples, single micro debris particles were detected during the separation of hard parts from soft materials. The inorganic debris was collected, cleaned and dried in sterile glass containers. Samples were run through the Nicolet iS5 FTIR spectrometer and the absorbance spectrum of the sample was matched to the corresponding non-organic spectrum in the extensive Thermo Scientific Plastic library. The first micro debris particle processed was recovered from a harbor seal scat collected in Chatham Harbor in 2016. The debris was identified as cellophane (figure 3.10; C. Hudak and Sette, 2017, *unpublished data*). Of the 88 processed seal scats (harbor seal and gray seal sample totals combined), 4% (n=4) contained single particles of micro debris. Based on these preliminary results, diet investigations using scat content for prey identification should be processed using protocols that are currently being developed to aid in the detection of marine debris, including sampling locally abundant prey fish present in PB (Bravo *et al.,* 2013 ; Nelms *et al.*, 2018).

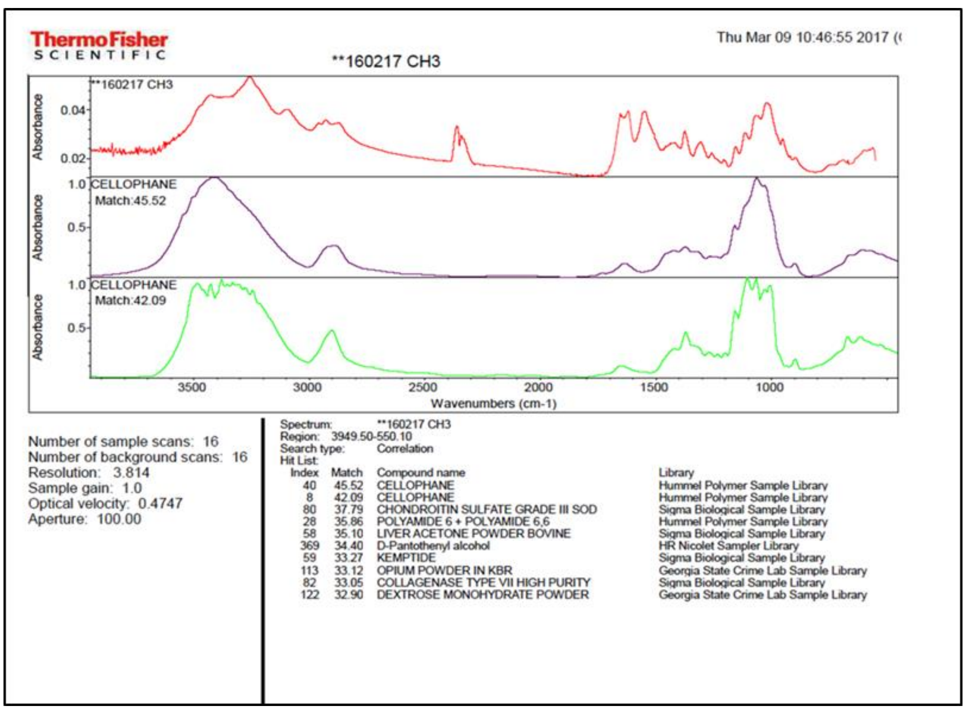

Figure 3.10. The spectrograph report of the micro debris particle (cellophane) that was recovered from a harbor seal scat sample collected on February 17, 2016 at haul-out “A” inside Pleasant Bay.

3.5. Recommendations

3.5.1. Aerial Surveys

With the increase of gray seals inside PB in summer of 2015 and the shift in distribution for both species, CCS recommends more frequent monitoring. Aerial work from planes although efficient for large areas, is costly for more contained system like Pleasant Bay. Unmanned aerial systems (UAS) teams are now available at the Northeast Fisheries Science Center and the Center for Coastal Studies. With proper permitting, UAS surveys could be used to do more frequent monitoring of seal populations within Pleasant Bay at a significantly lower cost.

There is an opportunity to coordinate with local seal watch vendors and the new Pleasant Bay Community Boating (PBCB) Floating Classroom to monitor changes in the local seal populations. Citizen science programs for water quality are in place in many towns across the Cape, including Pleasant Bay. These types of data collection platforms could complement the research underway currently in Pleasant Bay and detect changes that might otherwise be missed with less frequent formal monitoring.

Figure 3.11. Opportunistic sighting of harbor seals hauled out on rocks off the northeast side of Eastward Ho Gulf Course on January 11, 2018. Photo credit: CCS volunteer Andrea Spence

3.5.2. Seal diet analysis

For this study scat content analysis was used to capture important prey species in harbor and gray seal diet. However, based on prior studies (Bowen, 2000; Ampella, 2009) and field work and lab observations completed for this study, scat analysis alone can miss prey species with fragile otoliths, like Atlantic herring, or soft bodied prey or partially consumed prey like squid or spiny dogfish. For example, during the rinsing process of scat we observed otolith breakage, specifically in herring species. Likewise, in our field work we observed seals eating spiny dogfish during scat collections and know they are locally abundant. However, hard parts (eg. spines) were not recovered in any of our samples. New molecular methods for diet estimation using prey DNA have been developed that are able to increase the number of prey species identified in scat that hard part analysis alone would likely miss (Tollit *et al.*, 2009). In the future, we would recommend scat collection and prey DNA analysis to better estimate seal diet at haul-outs inside Pleasant Bay.

Acknowledgments

The authors wish to thank the following individuals for all that they contributed to this work. The aerial surveys would never have happened without the work (final counts and vessel support) of Beth Josephson at the Northeast Fisheries Science Center (NEFSC). Dr. Fred Wenzel from NEFSC provided guidance for otolith, squid beak and bone identification and contributed ideas about past diet work and future directions for research. Dr. Andrea Bogomolni contributed ideas and field support for both aerial and diet work. Dr. Charles "Stormy" Mayo at CCS helped coordinate seal flights using the expertise of the CCS right whale aerial team members Corey Accardo, Brigid McKenna, Jess Thompson and Christy Hudak. Pilot Joe Chronic and his team from the New England Specialized Aviation Services, Trevor Laue and Bob Bisbee provided a safe and effective flight platform with the motto of "Getting it done"! CCS's Catherine Macort helped with just about everything including making sure everyone was safe. The scat collections would not have been possible without the vessel and team support from the International Fund for Animal Welfare (IFAW) staff and volunteers. Dr. Michael Moore shared amazing UAS images with us for this report. Captain Ted Lucas helped out immensely in February and March of 2017 for added harbor seal scat surveys. CCS volunteers, David Moulton and Elizabeth Bradfield assisted with field work for the diet study by land and by sea. This project was funded by the Friends of Pleasant Bay, the Mandel/Rodis Fund, the Island Foundation and continued support from CCS.

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CHAPTER 4
Integrating habitat and their constituents of Pleasant Bay

Legare B.J, O.C. Nichols, L.M. Sette, A. Mittermayr and M. Borrelli

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Abstract

Pleasant Bay is a dynamic and complex coastal system that continues to experience natural and anthropogenic changes. Ecosystem based management requires large data sets including biotic and abiotic descriptors. Collecting data to describe a system involves data that is spatially and temporally variable and are often difficult to link and answer questions that are important to managers. Here we describe methods to link data of benthic habitat and micro-faunal communities to that from fisheries independent sampling. We discover that fish and shellfish communities vary and are linked to both habitat, micro-faunal communities as well as abiotic factors. This indicates that collecting habitat and micro-faunal community data is important in monitoring and understanding long term trends. Furthermore, fish communities, seal distribution and diet have connections and overlap. The links and connections between habitat, micro-faunal communities, fish, shellfish and seals present a story of a connected ecosystem.

4.1. Introduction

Shallow water coastal systems of New England are complex and productive ecosystems. Pleasant Bay, Massachusetts is a coastal lagoon with extensive eelgrass, shellfish, and salt marsh habitat and is subject to large but predictable variations in inlet formation and shifting access to the open ocean (Borrelli et al., 2018). Habitats are an ecological or environmental area inhabited by a particular species or group of species (ICES 2006). A habitat is made up of both physical (grain size, temperature, light, salinity) and biotic factors (food availability, presence of predators) and can be created by ecosystem engineers (eelgrass, oysters, tube forming worms) (ICES 2006). Adequately describing and defining habitats is challenging as they change temporally and have highly mixed compositions as gradient of each other (Legare and Mace 2016). Thus, data collected within the same area varies based on season as well as mapping resolution and the habitat being quantified. Therefore, ecosystem based management requires spatial and temporal data sets that encompass a variety of biotic and abiotic factors (Schumchenia and King 2010). Establishment of a baseline and its assessment is the first step in understanding an ecosystem (Cogan et al 2009). The second is to explore the connection of the inhabitants, fishes, shellfishes and predators, to the resources available (Cogan et al 2009).

An ecosystem wide habitat assessment was conducted between 2014-2016 by the Center for Coastal Studies (CCS) to create an extensive dataset for Pleasant Bay and the habitats therein (Borrelli et al., 2018, Nichols et al., 2018 Sette et al., 2018). Initial surveys included bathymetry and acoustic mapping of the seafloor, an inventory of habitat extent and creating a foundational data set of species abundances and auxiliary (physical) data. Based on these initial results, sampling was conducted in order to quantify the benthic micro-invertebrate communities and classify the characteristics of the habitats they live in. This micro-invertebrate survey included three data sets: invertebrate community composition, grain size analysis and video assessment of the benthic community. From these surveys, a habitat and resource inventory was created to understand the trends in ecological and commercially important species.

Fisheries independent surveys are common methods to understanding shellfish and fish community assemblages. Trawl and dredge surveys are often conducted to identify specific components and untangle seasonal and spatial trends of a system. Although many gears are species specific, fisheries independent sampling can provide valuable insights into the ecosystem as a whole if sampling is conducted throughout the season.

The re-emergence of two seal species within Pleasant Bay established two important resident predator populations. The gray and harbor seal occupy portions of Pleasant Bay year round, with minimal residential overlap (Sette et al., 2018). This residency creates an important top-down pressure on the food web as they utilize the resources within and adjacent to Pleasant Bay. In coordination with the habitat assessment and fisheries independent sampling, surveys of seal abundance, distribution and diet were conducted allowing for a comprehensive ecosystem picture to unfold.

Advances in analytical techniques have made it possible to examine habitats and their constituents as an interactive community and draw conclusions to describe the community assemblages and health based on data sets easily repeatable for long term monitoring.

Here we describe the integration of multiple datasets: habitat surveys, benthic micro-invertebrates and acoustic mapping, which together describe the trends and distribution of shellfish and finfish communities. This allows the discussion of trends in predator abundance (seal) and establishes the framework for long term monitoring.

4.2. Methods

4.2.1. Vessel Based Acoustic Mapping

A suite of instruments is required to conduct high-resolution, vessel-based acoustic surveys. The Edgetech 6205 is a dual-frequency, phase-measuring sidescan sonar and was used for all surveys. Its operating frequencies are 550 and 1600 kHz for backscatter imagery and 550 kHz for bathymetry. The effective bathymetric swath width is 6-8 times the height of the sonar over the bottom. A Teledyne TSS DMS-05 Motion Reference Unit mounted on the sonar collects data on heave, pitch, and roll. A HemisphereGPS® V110 vector sensor is used to measure heading. A Trimble® R8 GNSS receiver utilizing Real-Time-Kinematic GPS (RTK-GPS) is used for positioning and tide correction for vessel-based surveys.

Edgetech's Discover Bathymetric® 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 as Geotiffs.

Post-processing of bathymetric data was performed using CARIS HIPS®. Raw HSX files were converted to CARIS HDCS 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 YSI Castaway® CTD. Patch tests were performed to determine motion and timing offsets (roll, pitch, yaw and latency).

4.2.2. Benthic micro-invertebrates

To determine the biological and physical structure of the benthic habitats, field surveys and video imagery were conducted for invertebrate and sediment characterization. To effectively characterize each study location, benthic survey stations were chosen by using a randomized tessellation stratified design. In the field, random stations were located using a Trimble® R8 GPS, and the boat was anchored before samples were collected (figure 4.2).

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. A GoPro™ Hero 3 was attached to the sampler and high-resolution video was collected for each sample to aid in bottom characterization and documentation. The video was of sufficient resolution for still grabs and qualitative habitat analysis (figure 4.1) The contents of the Van Veen were then emptied into a bucket; a low energy wash was done and the contents were sieved through a 1 mm mesh to retain organisms, detritus, and substrate greater than 1mm in size. Any large bivalves, crabs, or vertebrates (fish) were measured, counted and identified (or photographed for later identification) before being returned to the water. The material retained on the sieve was transferred to a fine mesh bag and brought back to the lab for preservation in 70% ethanol until processing and analysis.

The sample to characterize the sediment was taken between the first and second biological replicates to ensure that the sediment sample was generally representative of the substrate sampled for the biological replicates. The surface sediment was transferred to a 100ml Whirl-Pak® using a stainless-steel spoon, stored on ice, and later frozen at the lab for future analysis.

4.2.1. 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 (final concentration approximately 70%) and Rose Bengal to dye invertebrates. The sample was visually inspected, and all invertebrates were picked and sorted into general categories as could be discerned by the unaided eye (i.e. worms, shellfish, amphipods etc.). Invertebrate specimens were identified by the project biologist or trained personnel using dissecting microscopes. Specimens were identified to species when possible or to genus, families, or orders depending on the difficulty of identification, and enumerated. A voucher sample was labeled and recorded as a representative example of a particular species. All identified specimens were counted. Pictures were taken of voucher specimen, anatomical features of various specimen and for later identification and/or confirmation when necessary, using a digital microscope camera.

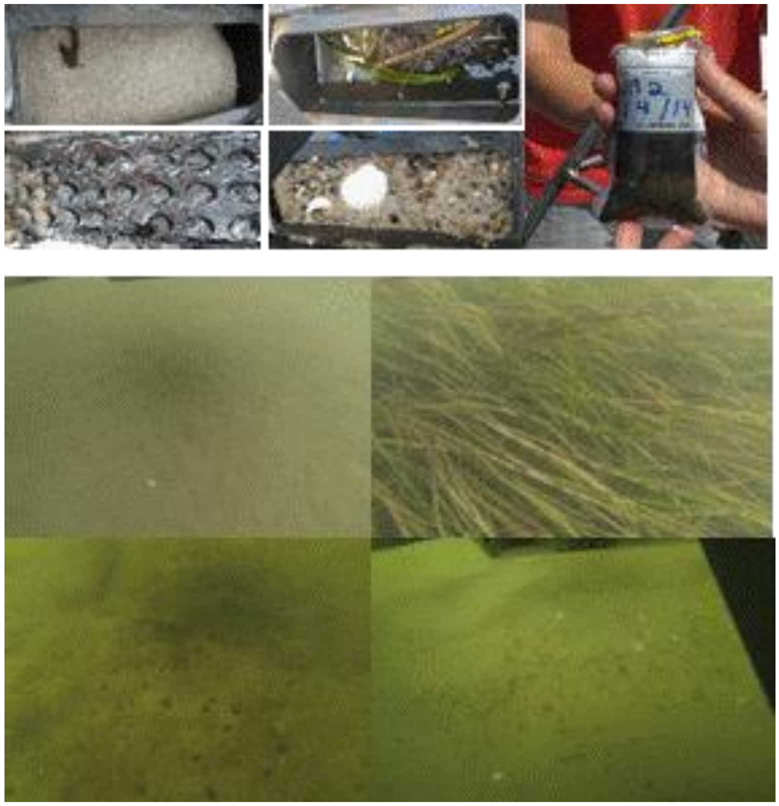

Figure 4.1. Grain size sampling with Sand, Eelgrass, Mud and Mixed Shell substrate (top) and GoPro Imagery of habitat for qualitative habitat analysis.

4.2.2. Sediment samples processing and analysis

To characterize the sediment substrate of the benthic habitat for each sample location (figure 4.2), 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.

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 and then placed in in a desiccator. Each sample was weighed, and the dry weight was recorded. After recording the initial dry weight, all samples were ground and then 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 (LOI) was determined.

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 Institution's Coastal Systems Laboratory. All data were reported using Wentworth grain size thresholds and classes (Folk, 1974).

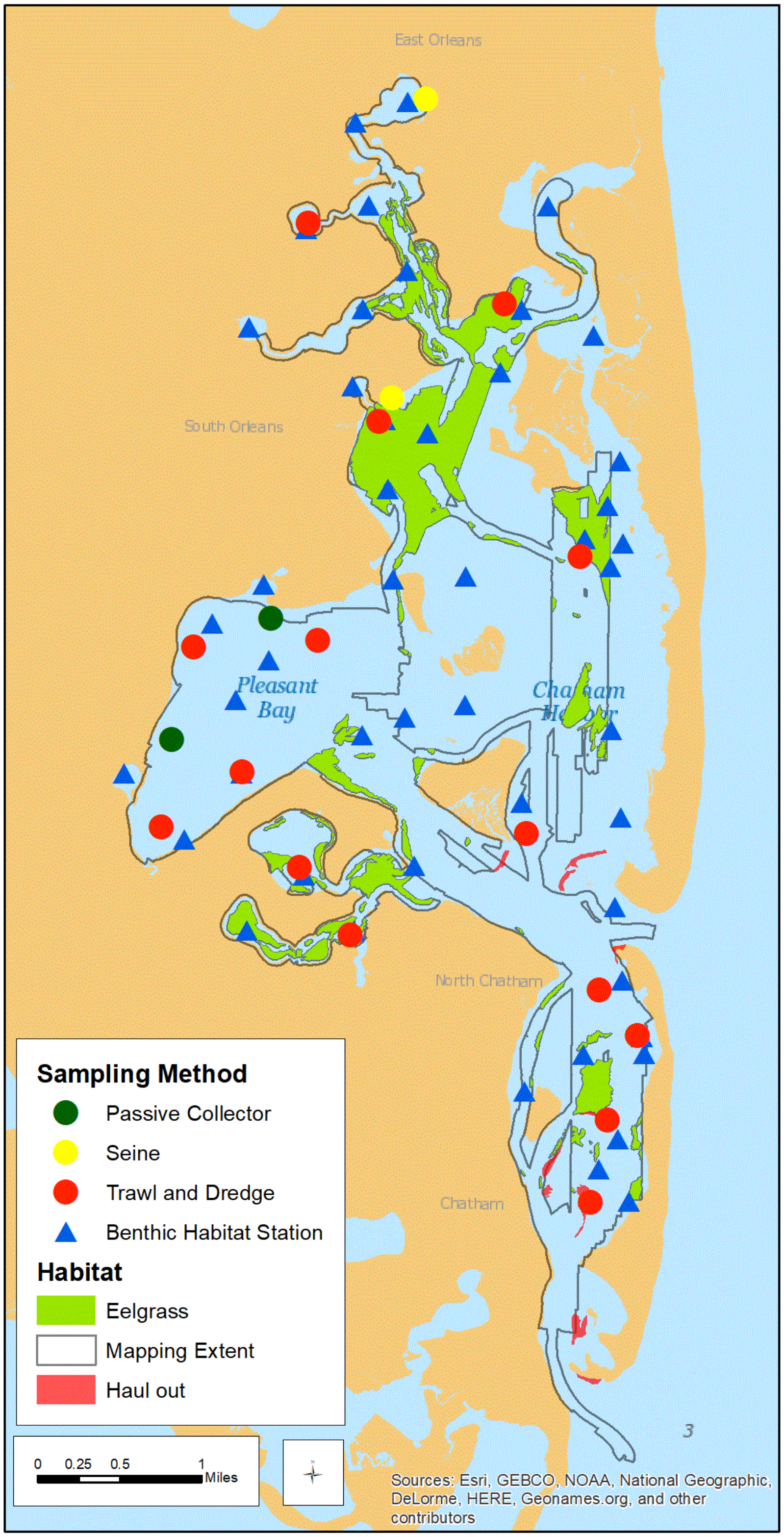

Figure 4.2. Location of dredge, trawl and benthic invertebrate samples across Pleasant Bay. Seal haul outs and eelgrass quantified from acoustic mapping noted.

4.2.3. Still pictures and qualitative habitat analysis

To quantify habitat type, screen stills were used to qualitatively identify cover and constituents of the substrate (figure 4.1). A scale of 0-4 was used to quantify benthic cover of sand, mud, shell, cobble/pebble, algae and eelgrass where, #0 equals 0% coverage, #1 equals 1-25% coverage, #2 equals 26-50% coverage, #3 equals 51-75%, and #4 equals76 -100% coverage. This was calculated for all four replicates then averaged together for each station. The use of a categorical numbering system reduced error due to visibility, visual interpretation, and angle and distance of camera toward the substrate (figure 4.1).

4.2.4. Dredge and trawl survey

Fisheries independent fish and invertebrate sampling was conducted in Pleasant Bay from June 2015 through June 2016. Where practical, sampling efforts were conducted using similar methods and gears to previous studies in the same area (e.g. Fiske et al., 1967) or more recent studies in the region (e.g. Chase et al., 2002). Subtidal sampling stations were chosen as a subset of sites chosen at random for benthic habitat sampling. All fish and most invertebrates were identified to the lowest practical taxon. In the interest of efficiency, small species that would not likely be consistently captured by sampling gears (e.g. small amphipods and isopods < 2 cm) were not consistently documented. The appropriate size measurement was taken (Total Length [TL], Fork Length [FL], Carapace Length/Width [CL/CW], Mantle Length [ML], etc.) for all fishes and most invertebrates. Sampling was conducted with a small trawl net and a commercial bay scallop dredge (see Chapter 3 of this report).

Both the trawl net and scallop dredge were deployed in a standardized manner, with consistent tow times and speeds (trawl: 5 minutes at 2 knots, dredge: 3 minutes at 3 knots). Tow start and end locations and depths were recorded using a Garmin 76 GPS and the boat's depth sounder (Faria Instruments DS1002 dual-temperature depth sounder). A duplicate tow was conducted immediately adjacent to the location of the first, in the opposite direction. Seawater and air temperature was recorded at the beginning of each tow using the sounder.

4.2.5. Seal survey

Aerial surveys were conducted at low tide monthly in 2014 and 2015 to determine the distribution and abundance of seals throughout Pleasant Bay. These surveys were conducted at an altitude of 200 m at a speed of 100 knots. The survey team photographed using a 7D Canon with a 100 - 400mm zoom lens to conduct photo records along a track lines along Pleasant Bay. The aerial survey created a spatial distribution of the residential seal populations and relative counts and abundances of seals present (figure 4.2).

To understand the connection of seals and the local resources, hard parts analysis was conducted on scat samples collected monthly at the haul out sites. The samples were collected using a cat litter scoop and was rinsed in between each samples in the seawater. The scats were stored in sterile freezer bags and stored in a minus 20 freezer until processing. Samples were then run through a series of graduated sieves (2.0, 1.0 and 0.5mm mesh), cleansed with water and then dried, sorted and stored in sterile jars (Ampella, 2009, Murie and Lavigne 1985, Harvey,1987, Torok,1994, Oxman, 1995, Phillips,2005). Each prey taxa has distinctive characteristics (e.g., fish otoliths, cephalopod parts, skate denticles, shell parts, crab carapace).

4.2.6. Methods of Analysis

In order to integrate benthic micro-faunal community assemblages and habitat characteristics with the abundance of shellfish and finfishes quantified by the fisheries independent surveys, non-parametric statistics were used. This enabled us to incorporate the different types of data and augment them with multiple additional factors using Primer7® (Clark and Gorley 2015). Factors were created by incorporating both the sediment characteristics and the categorical habitat classification from still grabs. Each station was treated as a sample and each physical characteristic (% Sand, % Silt, % Gravel, % Clay, % Loss On Ignition (LOI), Eelgrass, Cobble, Shell, and Algae) as a variable. Data were normalized by variables and a Principal Components Analysis (PCA) was performed to determine which variables contributed the most to describe the habitats present. A resemblance matrix was created using Euclidean distances before a cluster analysis and a non-metric multi-dimensional scaling (nMDS) plot was applied to create habitat groups. This created a factor for habitat (figure 4.3).

Factors describing the micro-faunal community assemblages across the stations were created by treating each station as a sample and the species abundance as variables. Date was standardized across samples and was transformed using square root. A PCA was performed to determine which variables contributed the most to the community assemblages. A resemblance matrix was created using Bray-Curtis Similarities and a cluster analysis and a nMDS plot was utilized to create micro-invertebrate community assemblage groups (figure 4.3).

Fish and shellfish communities are intrinsically linked to food availability and habitat type. The distribution of fish and shellfish communities found in the fisheries independent surveys were tested against the micro-faunal community assemblages and habitat categories. Trawls and dredges were assigned the micro-faunal community assemblage and habitat category which was spatially closest. In addition, distance of sample event (trawl or dredge) from the inlets was also tested. Each event represented a sample and species abundance was considered a variable. As trawling and dredging are specific to targeted habitats and/or species, each trawl sample was analyzed separately for invertebrates and finfishes (squid were included in the finfish as their presence in the system and capture-ability in the trawl more mimics other finfishes than shellfish).

Trawls and dredge data was transformed by square root and a PCA was performed to determine which species contributed the most to the community assemblage. A resemblance matrix was created using Bray-Curtis Similarities and an nMDS plot was utilized to identify similarities across stations. To test the factors of the microfaunal community assemblages, habitat and distance from inlet, Analysis of Similarities (ANOSIM) were performed to determine which factors contribute to the distribution of each fish or shellfish community.

Seal distribution and haul outs were identified from the aerial surveys and fisheries independent stations were characterized as either adjacent (within 200 m) or away (>200 m) from a seal haul out. Fish communities from trawl data were plotted out on an nMDS plot and proximity to seal haul outs classified each station. This allowed for visual interpretation of fish communities as it relates to seal haul outs.

4.3. Results

4.3.1. Benthic Sampling

Between June 24th and August 1st 2014, forty-eight stations within Pleasant Bay were sampled resulting in a total of 144 sieved and preserved biological samples (three replicates per station), 48 sediment samples, photographic and video data at each station (figure 4.2). PCA indicated that five habitat descriptors (LOI, Eelgrass, Algae, Sand and Shell) that contributed the most in describing each station (table 4.1). These factors were quantified from still imagery obtained from the video and sediment analysis identified stations into six habitat gradient types (figure 4.4, table 4.2) as determined by cluster analysis and plotted in a multi-dimensional scaling space (figure 4.3). The six clusters create descriptors (A-F) that are gradients of the habitat characterisics that were measured (i.e. Sand, Clay, Silt, Gravel, LOI, Eelgrass, Algae) (table 4.2). By creating habitat clusters (A-F), this allows for the use of habitat as a factor in which all variables are considered.

Micro-invertebrate community assemblages varied across and between stations. A total of 148 species were identified in Pleasant Bay. Cluster analysis, as represented on nMDS plot (figure 4.5), created seven micro-invertebrate communities groups across the stations (figure 4.6). PCA indicates species that are driving community differences are indicative of different habitat types (table 4.3), as these groups have various community make ups with different proportions of micro-invertebrates. These different groups were classified as a-f, as another factor for comparing the trends discovered in the fisheries independent sampling.

Table 4. 1. Principal components analysis of habitat characteristics with major drivers indicated in red.

Variable	PC1	PC2	PC3	PC4	PC5
Silt %	-0.488	-0.053	-0.022	-0.050	0.106
Clay %	-0.461	-0.067	-0.028	-0.055	0.163
LOI	-0.350	-0.037	0.271	0.022	0.356
Algae	-0.311	-0.398	-0.148	0.117	-0.566
Eelgrass	-0.062	0.305	-0.875	-0.246	0.145
Cobble	0.136	-0.344	0.147	-0.903	0.085
Gravel %	0.153	-0.614	-0.291	0.089	-0.190
Shell	0.221	-0.472	-0.163	0.309	0.665
Sand %	0.485	0.145	0.065	0.040	-0.093

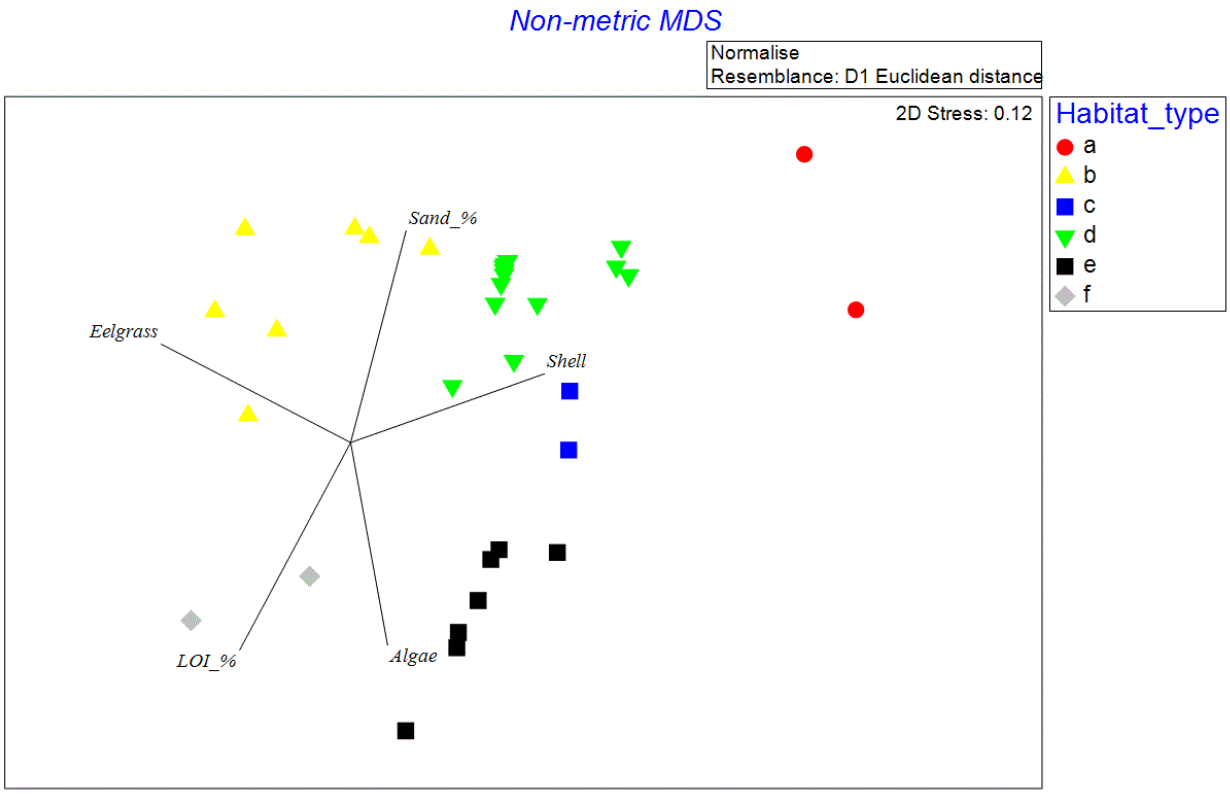

Figure 4.3. Non-Metric Multidimensional Scaling (nMDS) plot of habitat characteristics. Each symbol represents a station and colors indicate habitat type. Vectors indicate type of habitat represented.

Table 4. 2. Habitat types (A-F) as determined by Cluster analysis with composition of habitat descriptors (Gravel – Algae) that constitute each Habitat type as determined by non-parametric statistics.

	Habitat Types					
	a	**b**	**c**	**d**	**e**	**f**
Gravel	2.6 - 23.1 %	0 - 3.7%	10.3 - 21.3 %	0 - 7.8%	0 - 3.61 %	0 - 0.34 %
Sand	75.3 - 89.2 %	11.3 - 96.2%	77.4 - 87.6 %	25.4 - 100 %	4.1 - 96.4 %	5.2 - 96.9 %
Silt	1.4 - 4.7 %	1.3 - 65.8 %	1.1 - 1.8 %	0.0 - 64.4 %	0.0 - 83.1 %	2.45 - 84.2 %
Clay	0.14 - 0.73 %	0.2 - 21.6 %	0.2 - 0.3 %	0.0 - 10.2 %	0 - 23.32%	0.3 - 10.5 %
LOI	0.75 - 1.22 %	0.5 - 8.6 %	0.52 - 1.02 %	0.00 - 1.76%	0.0 - 17.77	15.2 - 22.4 %
Eelgrass	0%	25 - 100%	0%	0 - 25%	0%	0%
Cobble	0%	0%	0 - 25%	0 - 25%	0%	0%
Shell	50%	0 - 25%	0 - 25%	0 - 25%	0%	0%
Algae	0-25%	0-25%	25 - 75%	0 - 25%	0 - 75%	0 - 25%

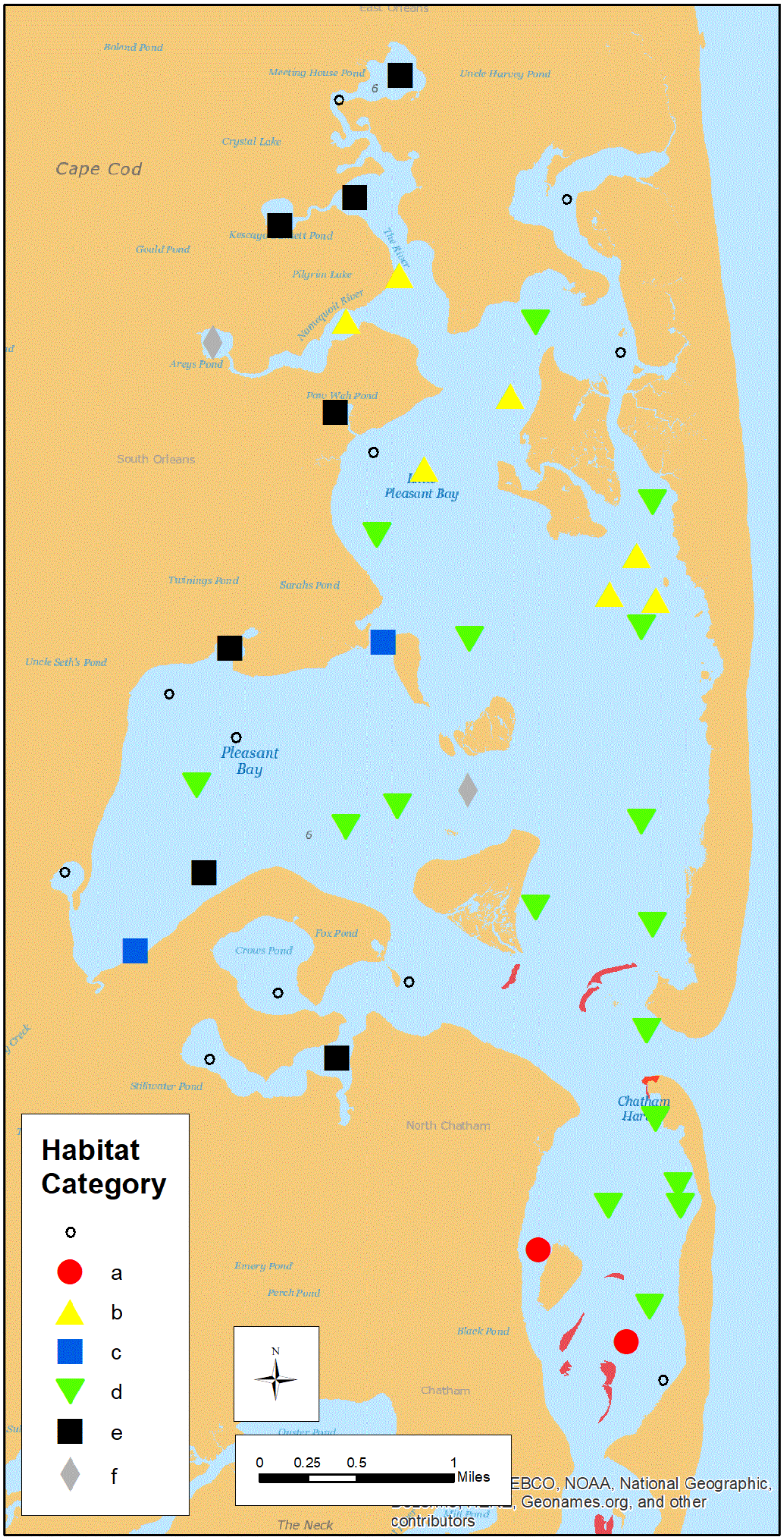

Figure 4.4. Habitats across stations as determined by cluster and principal components analysis.

Table 4. 3. Major species driving community assemblages according to cluster analysis and principal components analysis.

Species	Type	Habitat	Comment
Ampelisca spp	Amphipod	Algae/Eelgrass	Tube- building
Capitellidae	Polychaete	Soft sediment	Small surface-burrowing
Circeis spirillum	Polychaete	Eelgrass	Hard calcareous shell
Gemma gemma	Bivalve	Soft sediment	Mollusk bed
Streblospio bendicti	Polychaete	Soft sediment	Small tube building worm
Tellina agilis	Bivalve	Soft sediment	Mollusk bed
Cirratulidae	Polychaete	Coarse sediment	Deposit feeder
Nephtys spp	Polychaete	Well sorted medium and fine sand	burrowing

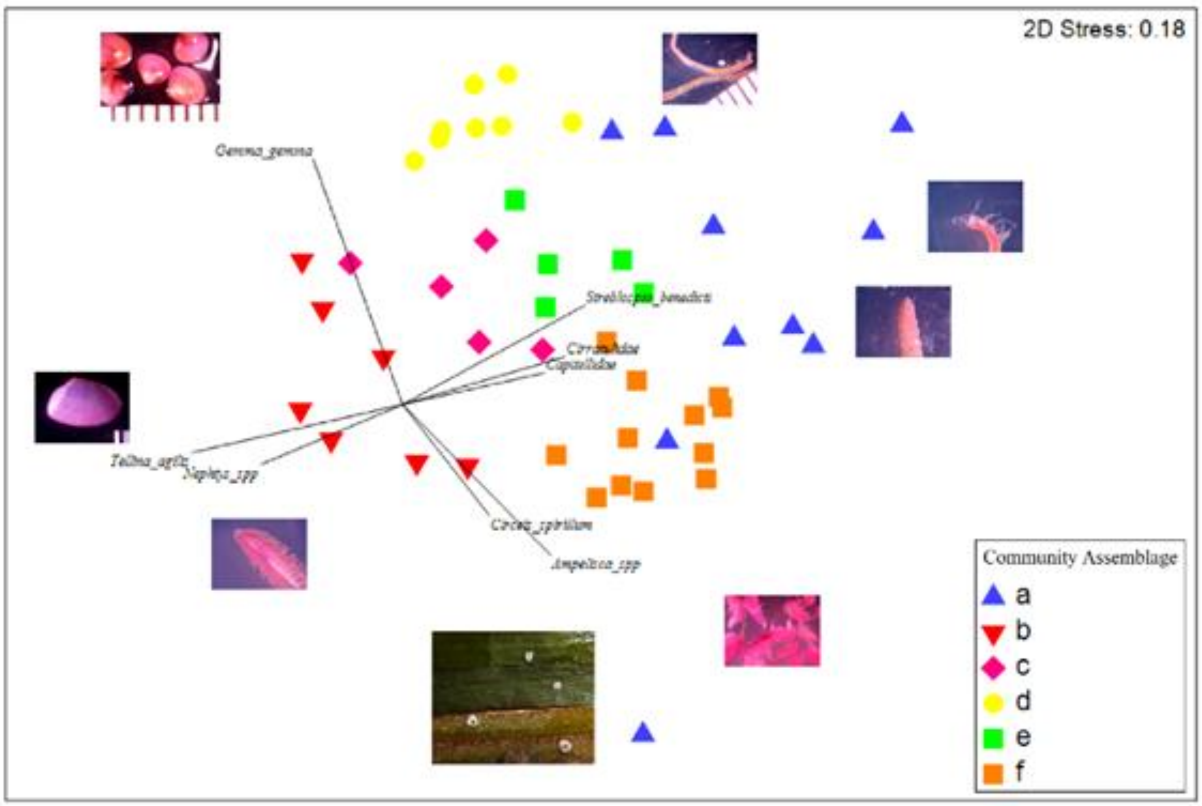

Figure 4.5. Non-Metric Multidimensional Scaling (nMDS) plot of microfaunal community assemblages. Each symbol represents a station and colors indicate a different community assemblage. Vectors indicates what species are driving the distribution.

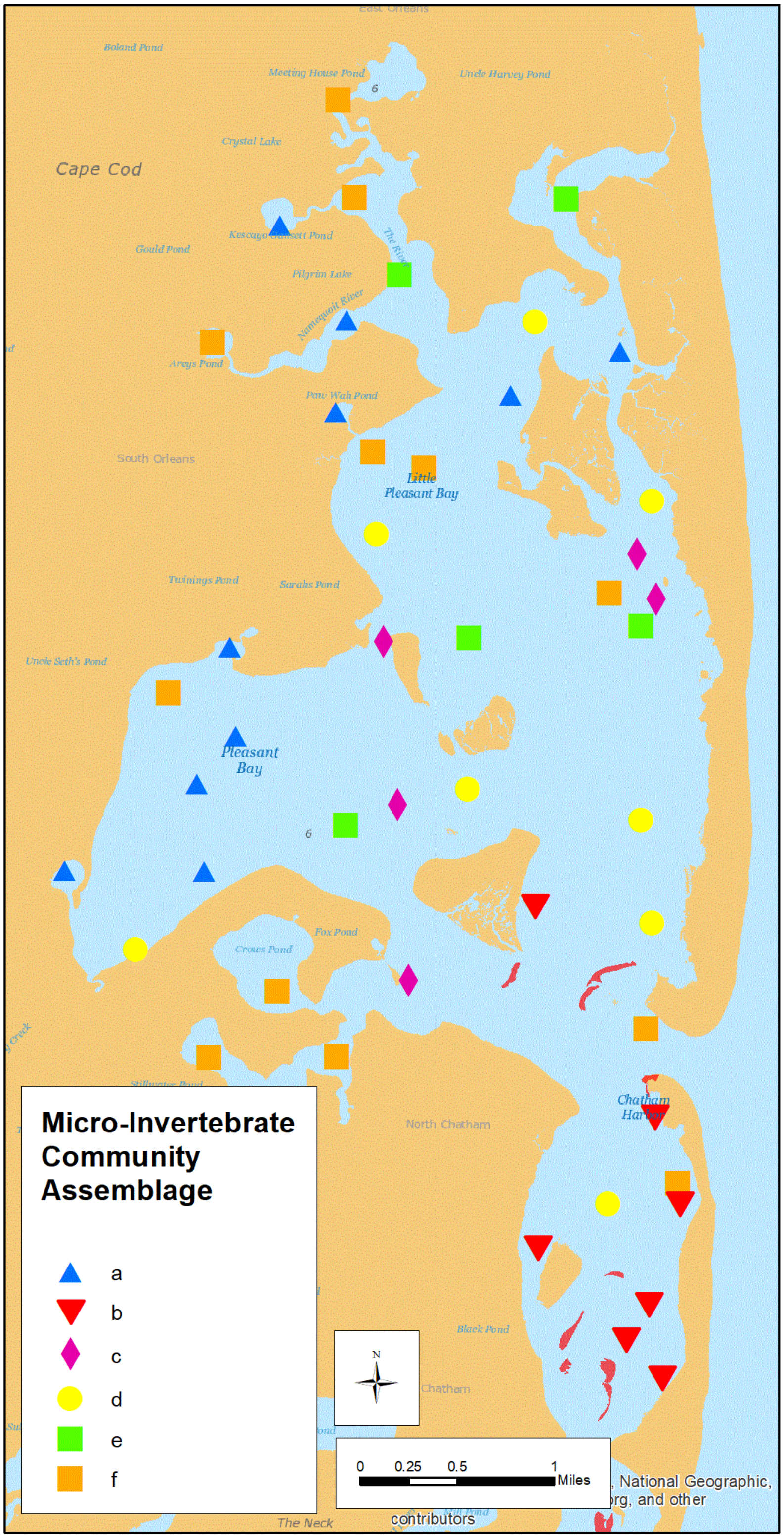

Figure 4.6. Micro-Invertebrate community assemblages across stations as determined by cluster and principal components analysis.

4.3.1. Fisheries Independent Sampling

Trawl and Dredge Sampling

The most commonly captured organisms captured by the trawl were the fishes fourspine stickleback (*Apeltes quadracus*: n = 2,132), young-of-the-year (YoY) Atlantic herring (*Clupea harengus*: n = 172), and Atlantic silverside (*Menidia menidia*: n = 111) and rock crabs (*Cancer irroratus*: n = 356). These five species accounted for 75% of the total catch. The high relative abundance of the fourspine stickleback was biased upwards by one tow with very high abundance (that was not repeated) in eelgrass habitat in September 2015, although it would still have been the dominant species with that sample removed from the data. The most commonly captured organisms captured by the dredge were rock crabs (*Cancer irroratu*s: n = 256), sea stars (*Asterias sp*.: n = 234), the oyster drill (*Urosalpix cinerea*: n = 64), and the fourspine stickleback *A. quadracus* (n = 181). These four species accounted for 66% of the total catch.

More dredge samples were collected than trawl samples due to the tendency of the trawl to become clogged with eelgrass or marine algae in some areas. Species diversity of trawl samples was greater than seine and dredge catches.

Fish communities were pooled by station and examined for temporal trends. As the habitat characteristics and benthic micro-invertebrate communities are a snapshot in time, this presents itself as an opportunity to test how useful these characteristics are when measured with a sampling design disregarding temporal aspects.

Shellfish communities captured in trawl sampling indicated seven species were drivers of community distributions across the stations (figure 4.7). These communities were tested against the same factors: habitat, micro invertebrate communities and distance from inlet, with an ASOSIM indicating that benthic micro-invertebrate communities are a significant factor (significance level of sample statistic: 0.1%) contributing 22.4% to the overall distribution.

Shellfish communities captured in the dredge sampling indicated five species that are descriptive the community assemblage across the stations (figure 4.7). An ANOSIM showed that benthic micro-invertebrate communities are a significant factor (significance level of sample statistic: 0.1%) contributing 37.2% to the overall distribution.

Fish communities from trawl sampling were driven by six species across stations (figure 4.8). These communities were tested against factors of habitat, micro-invertebrate communities and distance to inlet. ANOSIM indicated that both benthic micro-invertebrate communities and distance from inlet were significant factors (significance level of sample statistic: 0.1%) contributing 22.8% and 26.5% respectively to the overall distribution of fish. Finfish were not tested from the dredge sampling as fish sample size was low and dredge is inefficient at constantly capturing finfishes.

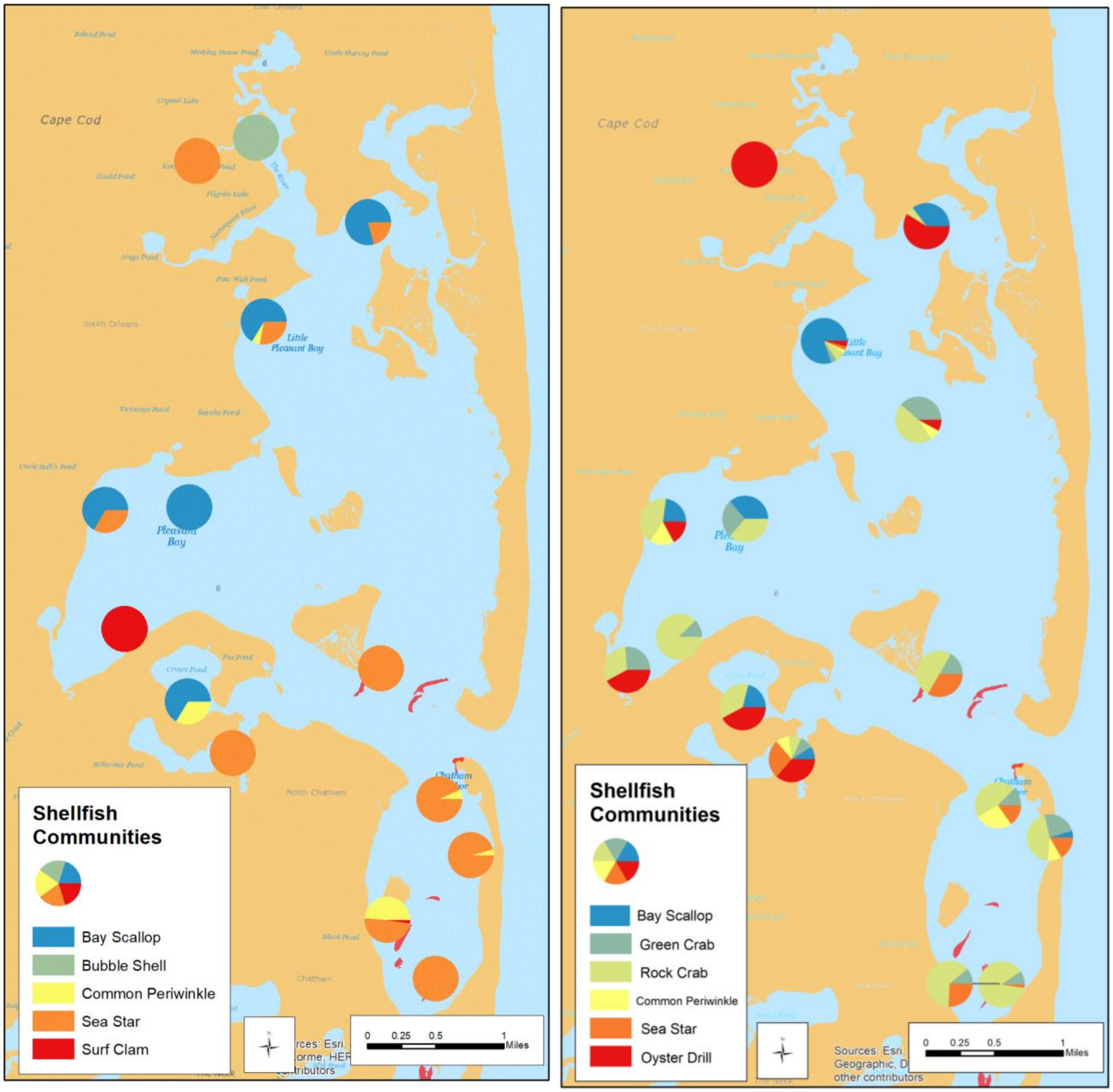

Figure 4.7. Shellfish communities across stations for Trawl (Left) and Dredge (right) with species that drive the community composition present.

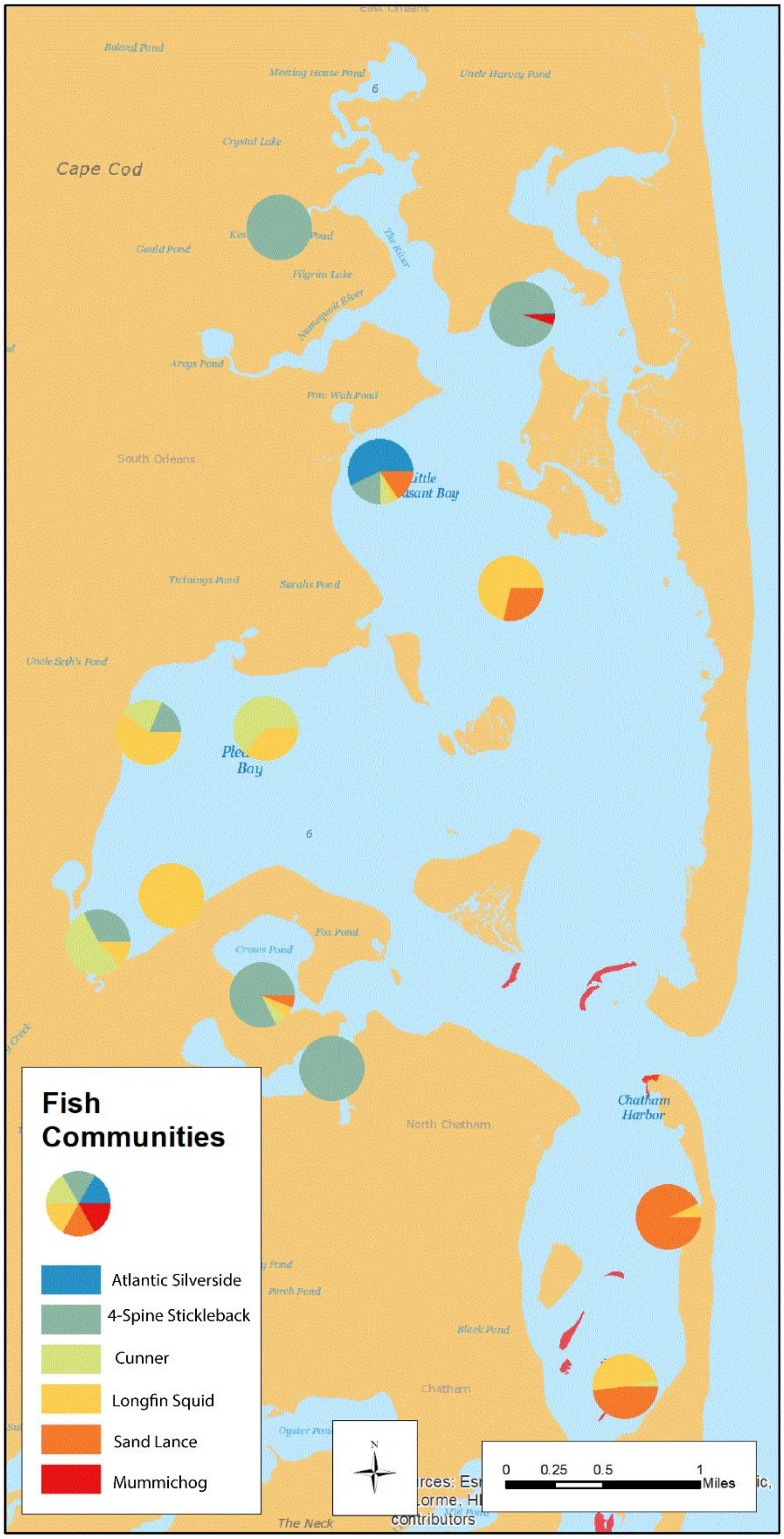

Figure 4.8. Finfish communities across stations for Trawl sampling with species that drive the community composition present

4.3.2. Seals

Finfish data from trawl sampling were pooled into two categories: adjacent and away from seal haul out locations and fish communities were plotted in non-dimensional space for visual interpretation (figure 4.9). Visual inspection indicates clear trends and trajectories of the species driving the trend identify sand lance and squid as community drivers of stations near seal haul outs. This corresponds to diet data collected in Pleasant Bay.

Hard parts analysis of harbor seals and gray seals found sand lance to be the most frequently occurring species. Longfin squid were observed to be the second most frequently occurring species in gray seals. Longfin squid and sand lance were the two species that best described the fish communities around the haul outs as indicated by the fisheries independent sampling (figure 4.8).

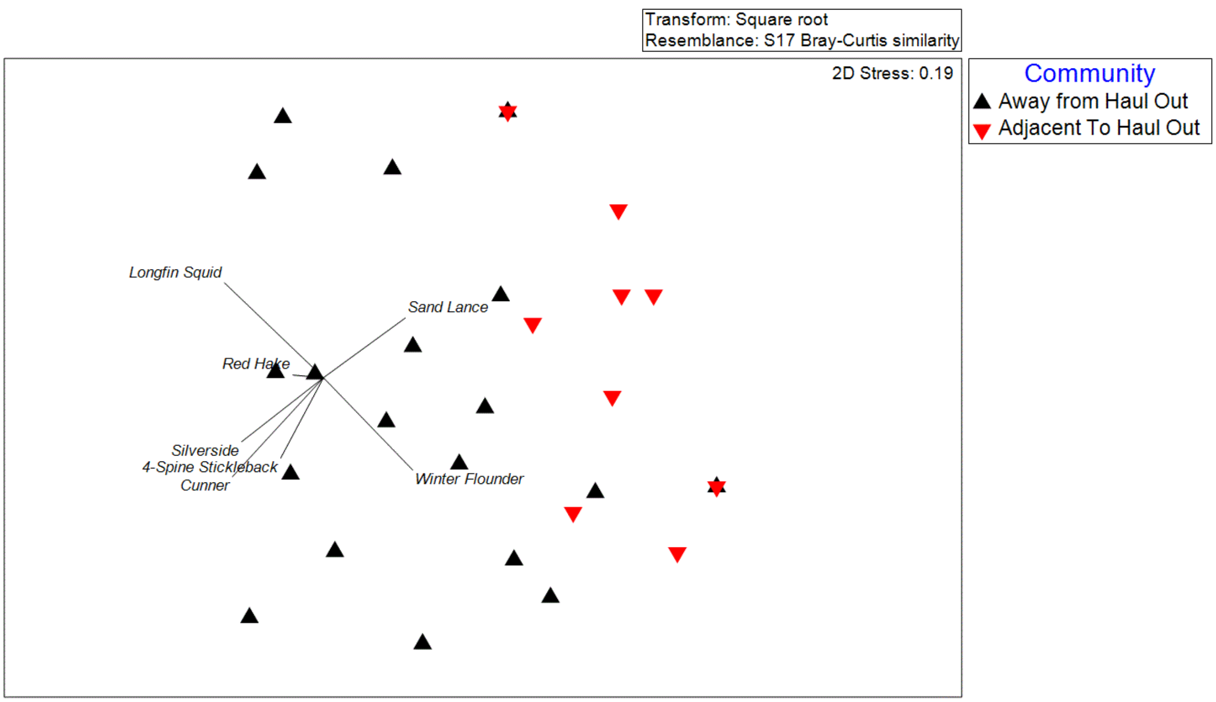

Figure 4.9. Non-Metric multidemensional scaling (nMDS) plot of fish communities from fisheries independent trawl sampling with adjacent to seal haul outs (Red) and away from seal haul outs (Black). Each symbol represents different fish communities. Vectors indicate species that are driving the distribution.

To identify differences in gray and harbor seal diet sand lance were removed, as almost every seal with identifiable parts had consumed sand lance. The seal diet (presence and absence) was plotted in non-dimensional space (figure 4.10). An ANOSIM was conducted to identify if the differences in observed diet were statistically significant. The ANOSIM determined that harbor seals and gray seals had different diets which were statistically significant, with a sample statistic (R) of 0.213 and a significance level of 0.1%.

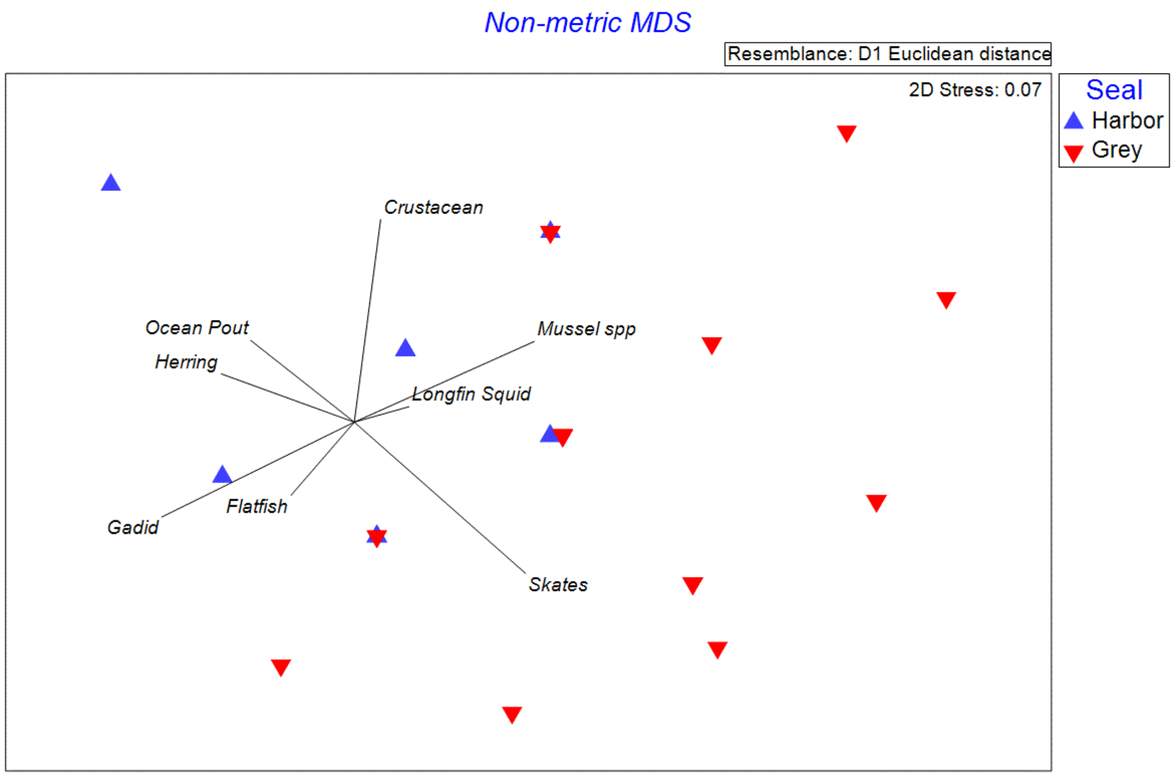

Figure 4.10. Non-Metric multidimensional scaling (nMDS) plot of diet from hard parts analysis of harbor seals (Blue) and gray seals (Red). Each symbol represents different species observed in the hard parts analysis. Vectors indicate what species are driving the distribution.

4.4. Discussion

Habitats are an ecological or environmental area inhabited by a particular species or group of species (ICES 2006). A habitat is made up of both physical (e.g. grain size, temperature, light, salinity) and biotic (e.g. food availability, presence of predators) factors and can be created by ecosystem engineers (eelgrass, oysters, tube forming worms) (ICES 2006). Delineation of habitats is difficult as they are often gradients of different descriptors and are dependent on mapping and sampling resolution (Legare and Mace 2017).

The approach described here integrates both categorical habitat information from video and quantitative information from sediment analysis, using non-parametric statistics. This method allowed for straightforward interpretation and grouping of habitat descriptors and created a more encompassing descriptor. For example, allowing the statistics to create habitat categories (a-f, figure 4.5) from categorical and sediment descriptors allowed us to describe the habitats that exist in Pleasant Bay (i.e. Sand/Eelgrass, Algae/LOI, Bare/Sand; table 4.2).

The abundances of fish and shellfish are both influenced by habitat and resource availability. Benthic micro-invertebrate communities are indicative of both the health of the system and the habitat present, and are a food resource utilized by fish and shellfish. The creation of station descriptors categorizing benthic micro-invertebrate assemblages as various groups (6 were used here) allows for additional analysis of the fisheries independent survey.

Fisheries surveys are limited by the habitat being sampled and the sampling method. Trawls, which occupy the water column and drag lightly along the bottom, are best at capturing large mobile invertebrates and fish. Dredge surveys on the other hand are best at capturing benthos associated invertebrates. Due to the difference in catchability between sampling methods, it was necessary to separate fish and shellfish in the dredge and trawl surveys. In doing so, shellfish distribution emerged to be significant when compared to both distance from inlet (describing a gradient of open ocean species to estuarine associated species) and across different benthic invertebrate community assemblages emerged. The micro-invertebrate assemblages were statistically significantly linked to the shellfish communities as the micro-invertebrate assemblages represent habitat types present and the food source available to the shellfish. Trawl surveys indicated a similar trend with fish communities distributed in a gradient from the inlet and across resources available as indicated by the micro-invertebrate assemblages.

The distribution and abundance of commercially, recreationally, and ecologically important species is known to be connected to the habitat and resources available. For example, horseshoe crabs are known to consume *Gemma gemma* clams and may move according to their abundance (Bottom 1984). The polychaete worms, *Capitella sp*., are known to be an indicator of disturbance as they are highly opportunistic (Blake et al. 2009). Stickleback, silversides and mummichogs are all known to find niche habitats within marsh systems, adapting to different food availabilities (Deegan and Garret 1997). These fish act as a forage base for larger predators such as winter flounder and squid which, along with sand lance, have been shown to be an important part of the diet of top predators of the Pleasant Bay system, such as the harbor and grey seal. The distributions and abundances of habitat, and invertebrates vary across the physical, seasonal, and water parameters further examination and connection to water monitoring programs is needed.

Further examination of fish communities indicates that the species driving fish community trends adjacent to seal haul-outs are the same species that are present in seal diet analysis. Hard parts analysis of seal scat indicated that both gray seals and harbor seals have a high frequency of occurrence of sand lance in their diet. Longfin squid was the second most occurring species in gray seal scat. These two species, sand lance and longfin squid, are the species that best describe the fish communities in trawl sampling adjacent to seal haul-outs. Other species present in seal diet included Gadid spp. and flounder. Trawl sampling adjacent to the seal haul-outs where scat samples were collected, captured species such as pollock, red hake, and winter flounder. The fisheries independent surveys and the seal diet surveys indicate that seals are potentially utilizing the resources in Pleasant Bay and the hard parts analysis indicates that they are consuming what is seasonally abundant within the system.

These surveys and analyses serve as a start and a baseline for future work. Future sampling and monitoring will further unlock links between seasons, habitats and abundances and allow linkages to be made to anthropogenic influences on Pleasant Bay. Utilizing multiple sets of data, as presented here, enables us to observe long-term changes. As the habitat changes, inlets shift, sea levels rise and temperatures warm, both the species composition of benthic communities and the distribution of habitats will change.

4.5. Recommendations

Using multiple data sets to identify trends in abundance of species across habitats is difficult. By employing new statistical and geographic techniques, this environmental assessment demonstrates that an interdisciplinary ecosystem analysis is possible and serves as a template for future work.

Future work on understanding the annual and inter-annual spatial-temporal movements of key species would be valuable. This would include increasing the effort of the fisheries independent survey, tagging and tracking of animals, and molecular techniques (e.g. genetics and stable isotope). Developing an understanding of movement of species within the bay and their resource needs will allow researchers and managers to understand local and regional connectivity of Pleasant Bay.

Further integration of these types of data with water quality monitoring and other monitoring programs will increase the statistical robustness of the described relationships. As the inlets and geology changes throughout the system and due to the location of Pleasant Bay at the intersection of Gulf of Maine and mid-Atlantic basin, it will be important to monitor this system in the context of geological and ecological change and sea-level rise. Monitoring and surveys using a trans-disciplinary approach, integrating abiotic and biotic factors, can make Pleasant Bay a model system for ecosystem management.

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