Massachusetts Estuaries Project

Benthic Monitoring Report:

Pleasant Bay 2021

Prepared for:
Massachusetts Department of Environmental Protection

MassDEP
Commonwealth of Massachusetts
Department of Environmental Protection

June 2021

BENTHIC MONITORING REPORT:
PLEASANT BAY 2021

for

MassDEP Massachusetts Estuaries Project

Benthic Monitoring

Submitted to

Massachusetts Department of Environmental Protection
Massachusetts Estuaries Project
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1 Introduction

The Massachusetts Estuaries Project (MEP) was established in 2001 to monitor and protect estuarine ecosystems in southeastern Massachusetts embayments. The technical reports produced from these embayment assessments documented embayment specific baseline water quality, habitat health, and identified the actions required to restore nutrient impaired waters for approximately 70 embayments. MEP provided technical guidance in support of policies on nitrogen loading to embayments, wastewater management decisions, and establishment of nitrogen Total Maximum Daily Loads (TMDLs) for over 30 estuaries. The four watershed communities of Pleasant Bay (Chatham, Harwich, Orleans, and Brewster) have begun the process of integrated water resources management planning or completed preparation of Comprehensive Water Resources Management Plans (CWRMPs) or Watershed Management Plans (WMPs). With implementation of the TMDLs and community measures, Massachusetts Department of Environmental Protection (MassDEP) identified a need to review and update the benthic monitoring procedures that were created in 2003 as part of the MEP Quality Assurance Project Plan (QAPP; Howes and Samimy 2003, Howes and Samimy 2005).

In 2017, MassDEP began the review process of the MEP Benthic Monitoring Program. Following a thorough review of MEP documents, relevant regional and federal benthic monitoring programs, and current scientific literature a tiered approach for previously assessed embayments and a baseline approach for unassessed embayments was recommended. New draft guidance documents for the collection of post-TMDL implementation and future baseline MEP benthic monitoring data were developed for the recommended approaches. The new guidance documents include a Marine Benthic Monitoring QAPP (Rutecki and Nestler 2019), Field Standard Operating Procedure (SOP; Sweeny and Rutecki 2019a), and Laboratory SOP (Sweeny and Rutecki 2019b) that describe the study objectives, field and laboratory techniques, data quality requirements and assessments, and data management for future MEP marine benthic monitoring. The goal of these documents is to develop guidelines and procedures that can be used by parties outside of MassDEP to collect benthic data that will be of sufficient quality to assess embayment conditions and be used in management decisions.

The MEP Marine Benthic Monitoring Pilot Field Study (Pilot Field Study) is being conducted to test the approaches and procedures described in the new draft guidance documents, and to obtain current benthic infaunal data for the embayments selected for the study. The development of a pilot field study is vital to verify the approaches and new documents will produce quality benthic data for MassDEP and coastal communities that 1) assess current embayment health, 2) are comparable between assessments and embayments, and 3) aid in future management decisions. MassDEP selected West Falmouth Harbor and the Pleasant Bay System, two previously assessed embayments with established TMDLs, and Wellfleet Harbor, a previously assessed largely unimpaired embayment, for the Pilot Field Study. West Falmouth Harbor was surveyed in 2019, the Pleasant Bay System in 2020, and Wellfleet Harbor will be surveyed in 2021. MassDEP selected Pleasant Bay for the Pilot Field Study because the embayment was previously assessed in the MEP in the fall of 2003 (Howes et al. 2006) and has established TMDLs.

The Pleasant Bay embayment system is the largest on Cape Cod, encompassing parts of four towns (Chatham, Harwich, Orleans, and Brewster; Figure 1). It is a valuable environmental ecosystem, designated by the state and recognized as an Area of Critical Environmental Concern (ACEC) in 1987. Although historically referred to as an estuary, Pleasant Bay is technically a lagoon (Hughes and Mittermayr 2018). By definition, an estuary is an embayment where salt water and freshwater inputs from rivers mix. However, there is little freshwater input to Pleasant Bay resulting in a lack of estuarine characteristics. The Pleasant Bay watershed is made of highly permeable substrates resulting in relatively low rainwater runoff. Most of the freshwater inflow is via groundwater discharge or groundwater fed surface water flow (e.g. streams to the head of Paw Wah Pond and Lonnies Pond).

Pleasant Bay is comprised largely of salt marsh and tidal flats and includes several large open water areas, small tributary sub-embayments, and the Bassing Harbor sub-system (Figures 2 – 4; Howes et al. 2006). Pleasant Bay is separated from the Atlantic Ocean by a barrier beach that is subject to frequent changes, largely due to coastal storms. Breaches through the barrier beach have occurred throughout the years creating islands and increased tidal flow to the estuary (Giese et al. 2009). The two most recent breaches formed the North (2007) and South (1987) Inlets which are both in the Chatham portion of the Pleasant Bay System (Giese et al. 2009). Changes in tidal flow to Pleasant Bay that result from changing geomorphology can affect the magnitude of nitrogen loading impacts on estuarine health in this embayment system. For example, if the inlet becomes restricted or migrates south causing a reduction in tidal flushing, nitrogen loading would increase (Howes et al. 2006).

Pleasant Bay supports a variety of recreational uses including boating, swimming, and shellfishing and is designated as an SA water under 314 CMR 4.00 Massachusetts Surface Water Quality Standards. SA water is designated for the following uses: marine fish, shellfish and wildlife habitat, shellfish harvesting for direct human consumption, recreation and all other legitimate uses including navigation. The Pleasant Bay System is currently designated as Outstanding Resource Waters (defined as habitat that should be protected) under the state surface water regulations of 1995 (Cape Cod Commission 2017). The Pleasant Bay System contains several benthic and shellfish habitat types including eelgrass (*Zostera marina*), softshell clam (*Mya arenaria*), quahog (*Mercenaria mercenaria*), bay scallop (*Argopecten irradians*), and razor clam (*Ensis directus*). The health of the harbor, like many embayments in the region, has declined in recent decades due to nutrient pollution entering the system from nearby development, as evidenced by increasing algal growth and loss of eelgrass beds (Howes et al. 2006, Costello and Kenworthy 2011).

A Pleasant Bay Resource Management Plan was adopted in 1998 by the Towns of Orleans, Chatham, and Harwich forming the Pleasant Bay Alliance. The Alliance and its member towns have implemented numerous measures over the years with the goal of improving the health of Pleasant Bay. The measures include the establishment of the Pleasant Bay Citizen Water Quality Monitoring Program, Guideline for Permitting Docks and Piers in Pleasant Bay, upgrading existing treatment facilities, sewer system construction in the Pleasant Bay watershed, the Muddy Creek Wetland Restoration, and the Pleasant Bay Composite Nitrogen Management Analysis (Cape Cod Commission 2017; Ridley and Associates Inc. 2018). Information on this work to restore the health of the Pleasant Bay System can be found at www.pleasantbay.org.

This report provides the results of the 2020 MEP Marine Benthic Monitoring Pilot Field Study conducted in Pleasant Bay. The report includes a comparison with the previous Pleasant Bay MEP assessment presented by Howes et al. (2006), the 2020 MEP West Falmouth Harbor Monitoring Pilot Field Study, and the Friends of Pleasant Bay Marine Ecosystem Assessment (Hughes and Mittermayr 2018).

Figure 1. The location of Pleasant Bay, Massachusetts.

2 Methods

The Pilot Field Study conducted in Pleasant Bay followed the Previously Assessed Embayment Tier 2 approach and corresponding field methodologies selected during the planning phase and outlined in the Embayment-Specific Study Plan for Pleasant Bay (Sweeny and Rutecki 2019c). The survey was comprised of four components: water quality measurement profiles, digital images, benthic infauna, and sediment conditions (grain size and total organic carbon [TOC]). Benthic sampling stations used in the current assessment were consistent with the benthic infaunal sampling stations used in the previous 2003 assessment. The latitude and longitude coordinates of the 2003 benthic infaunal sampling stations used by Howes et al. (2006) were redefined through georeferencing techniques in ArcGIS[1] (Table 1; Figures 2 - 4).

Detailed descriptions of the field and laboratory methods are contained in the draft MEP Benthic Monitoring QAPP (Rutecki and Nestler 2019), the draft MEP Marine Benthic Monitoring Field SOP (Sweeny and Rutecki 2019a), and the draft MEP Marine Benthic Monitoring Laboratory SOP (Sweeny and Rutecki 2019b). The recommendations from the 2020 MEP West Falmouth Harbor Benthic Monitoring Report (Sweeny and Rutecki 2020) were incorporated into the Pleasant Bay study. A brief overview of the methods, focused on information specific to this survey, is provided below in Section 2.1 to 2.3.

2.1 Field Methods

Sampling was conducted at the following 33 Pleasant Bay stations on September 17, 18, 23, 24, and 29, 2020:

- <u>17 September</u> - the three Round Cove (RCV1, RCV2, and RCV3) stations, Quanset Pond (QP3), Pleasant Bay (PB20) and Bassing Harbor (BH),
- <u>18 September</u> - the three Meetinghouse Pond stations (MP47, MP48 and MP49), the two locations in The River (TR50, TR52) and in Lonnies Pond (LP53),
- <u>23 September</u> - two stations each at The River and Pochet (TR26, TR45, P39, and P41, respectively) along with stations in Paw Wah Pond (PWP), Areys Pond (AP22/23) and Little Pleasant Bay (LP43),
- <u>24 September</u> - a total of seven stations in Pleasant Bay (PB6, PB32, and PB16), Crows Pond (CPE, CPW), Chatham Harbor (CH12) and Little Pleasant Bay (LBP), and
- <u>29 September</u> - Little Pleasant Bay (LP35, LP36, and LP37), Pleasant Bay (PB14), Muddy Creek (MCM), Frost Fish Creek (FFC) and Ryders Cove (RC).

All stations except ten were assessed as planned. Of these ten stations, four stations (RCV3, RCV1, PWP46, and AP22/23) were in mooring fields with high boats densities and needed to be slightly relocated to ensure vessel and crew safety. Three stations needed to be moved as the re-identified coordinates placed the stations on land (QP3) or on shoals that would be exposed at low tide (PB14, PB32). The station located at Pochet (P41) was moved since it was in the middle of a shellfish growing

[1] Latitude and longitude coordinates for the 2003 MEP benthic infaunal stations are unavailable. As a result station locations were re-identified by importing the image with MEP stations into ArcGIS. Control points for each station were selected from the image to the referenced map coordinate layer. The station locations were then digitized and the coordinates were exported.

area. The access review determined that the original stations for Frost Fish Creek and Muddy Creek were on the west side of Rt. 28 and not currently accessible by boat. New locations for these stations were situated as close as possible to the originals and approved by MassDEP. The two stations in Muddy Creek were combined into a single location and taken at the mouth of the creek (MCM; Figure 3). As a result eight stations were outside of the 30-m target radius (i.e. Stations QP3, PB14, AP22/23, PB32, P39, P41, FFC, and MCM). Water quality profiles, digital images, triplicate infaunal samples, and one sediment sample were collected at the 32 stations located in Pleasant Bay (Figures 2 - 4). Bottom sediment for benthic infauna and sediment condition were collected using a 0.04-m^2 Ted Young-modified Van Veen grab sampler. At LPB only underwater image and water quality sampling was conducted due to the presence of eelgrass (Figure 5). No grab samples were collected at this station in accordance with Section B2.2.3 of the draft MEP Marine Benthic Monitoring QAPP (Rutecki and Nestler 2019).

A Garmin GPSMAP 78 with WAAS (Wide Area Augmentation System; accuracy +/-2 meter [m]) on a field computer running Nobeltec VNS (Visual Navigation System) was used to acquire coordinates at the location of each sample. Comparisons among sampling coordinates and target station locations confirm that all sampling was conducted within 30 m of the target locations (except where noted above).

Water quality measurement profiles were taken using an YSI 6820 V2 multi-parameter water quality sonde with data recorder and temperature, dissolved oxygen (DO), pH, and salinity/conductivity probes. Measurements were collected following the depths and protocol specified in the draft Marine Benthic Monitoring QAPP and draft Field SOP (Rutecki and Nestler 2019, Sweeny and Rutecki 2019a).

Digital video images for each sampling location were recorded using a Sony HC3 HD camera in a Light-n-Motion waterproof housing attached to a stainless steel frame (15.5 inches by 15.5 inches) with scaling lights set 4 inches at 1 meter (m) apart. A GoPro Hero 3+ was also attached to the camera frame to provide digital still images and camera redundancy. Due to elevated turbidity at many of the stations, the camera used for the underwater images was positioned closer to the bottom than the 1 m outlined in the draft Marine Benthic Monitoring QAPP and draft Field SOP. The turbidity also prevented the use of additional lights on the camera frame as they produced backscatter and prevented a clear view of the bottom. Digital images were collected but only reviewed to provide documentation of eelgrass and a general visual description of the bottom at the sampling locations.

Figure 2. Map of benthic infaunal sampling locations in Pleasant Bay.

Figure 3. Map of benthic infaunal sampling locations in the Bassing Harbor sub-system. The red diamonds in Muddy Creek indicate the planned station locations that were not accessible. The blue diamond at the mouth of Muddy Creek indicates the location of the actual station sampled. The red diamond in Frost Fish Creek indicates the planned station location that was not accessible. The purple diamond in the lower Frost Fish Creek indicates the location of the actual station sampled.

Figure 4. Map of benthic infaunal sampling locations in the Round Cove sub-embayment.

Figure 5. Images of eelgrass observed at Station Little Pleasant Bay (LPB).

Table 1. Listing of Preliminary Field Data from Pleasant Bay 2020 Survey (PB-2020).

BASIN	STAT_ID	STAT_ARRIV_LOCAL	BEG_LATITUDE	BEG_LONGITUDE	NAV_QUAL	DEPTH_TO_BOTTOM	DEPTH_UNIT_CODE	COMMENTS
Meetinghouse Pond	MP47	9/18/2020 10:40	41.780317	-69.967017	+/- 2m	1.5	m	
	MP48	9/18/2020 11:29	41.781367	-69.965867	+/- 2m	3.3	m	
	MP49	9/18/2020 12:11	41.780150	-69.964467	+/- 2m	7.6	m	
The River	TR26	9/23/2020 14:24	41.762100	-69.969817	+/- 2m	0.9	m	
	TR45	9/23/2020 10:56	41.766167	-69.963500	+/- 2m	2.8	m	
	TR50	9/18/2020 13:05	41.779100	-69.971200	+/- 2m	2.6	m	
	TR52	9/18/2020 15:07	41.938150	-69.969500	+/- 2m	2.2	m	
Pochet	P39	9/23/2020 12:41	41.937917	-69.949100	+/- 2m	0.5	m	
	P41	9/23/2020 13:30	41.761517	-69.946500	+/- 2m	0.5	m	
Lonnies Pond	LP53	9/18/2020 14:24	41.769850	-69.976933	+/- 2m	4.3	m	
Areys Pond	AP22/23	9/23/2020 15:05	41.760317	-69.982533	+/- 2m	3.4	m	
Paw Wah	PWP46	9/23/2020 16:10	41.755583	-69.969733	+/- 2m	1.3	m	
Little Pleasant Bay	LPB	9/24/2020 11:58	41.746500	-69.965433	+/- 2m	1.6	m	eelgrass
	LPB37	9/29/2020 11:30	41.738717	-69.956283	+/- 2m	0.7	m	
	LPB35	9/29/2020 12:06	41.740817	-69.941350	+/- 2m	1	m	
	LPB36	9/29/2020 12:39	41.749217	-69.939233	+/- 2m	0.7	m	
	LPB43	9/23/2020 11:44	41.757083	-69.953033	+/- 2m	0.7	m	
Pleasant Bay	PB6	9/24/2020 12:28	41.726833	-69.982450	+/- 2m	4.5	m	
	PB14	9/29/2020 10:11	41.717217	-69.940450	+/- 2m	2.2	m	
	PB16	9/24/2020 15:20	41.718483	-69.949350	+/- 2m	2.1	m	
	PB20	9/17/2020 15:17	41.725600	-69.966217	+/- 2m	2.3	m	
	PB32	9/24/2020 16:00	41.726333	-69.940983	+/- 2m	1.6	m	
Quonset Pond	QP3	9/17/2020 14:27	41.737600	-69.980850	+/- 2m	3	m	
Round Cove	RCV2	9/17/2020 11:35	41.720450	-69.996917	+/- 2m	2.6	m	
	RCV3	9/17/2020 12:35	41.721133	-69.996500	+/- 2m	2.7	m	
	RCV1	9/17/2020 13:22	41.720117	-69.997900	+/- 2m	1.8	m	
Bassing Harbor	BH	9/17/2020 16:12	41.707183	-69.971317	+/- 2m	1.4	m	
	CPE	9/24/2020 13:40	41.713750	-69.975500	+/- 2m	4.2	m	
	CPW	9/24/2020 14:12	41.713950	-69.977333	+/- 2m	3.8	m	
	MCM	9/29/2020 13:31	41.713967	-69.994867	+/- 2m	1.2	m	
	FFC	9/29/2020 14:32	41.703067	-69.969050	+/- 2m	0.8	m	
	RC	9/29/2020 15:17	41.708483	-69.983100	+/- 2m	3	m	
Chatham Harbor	CH12	9/24/2020 10:30	41.700733	-69.935933	+/- 2m	1.4	m	

Beginning latitude and longitude coordinates are in decimal degrees.

2.2 Laboratory Methods

Laboratory methods were consistent with the draft MEP Benthic Monitoring QAPP (Rutecki and Nestler 2019) and the draft MEP Marine Benthic Monitoring Laboratory SOP (Sweeny and Rutecki 2019b) with one exception (see below *Gemma gemma* subsampling). Two infauna samples were randomly selected for processing, while the third was archived. A total of 64 benthic samples from Pleasant Bay were sorted. Organisms were sorted and identified to the lowest possible taxonomic level using a dissecting microscope. Each distinct taxon was saved separately in a labeled vial with reagent alcohol and archived in a reference collection as directed under Section B4.1 of the draft MEP Benthic Monitoring QAPP. Counts were standardized to densities per square meter (m^2) of bottom.

The following subsampling procedure was employed on samples with very high abundance of juvenile *G. gemma* (bivalve). During the first steps of processing, each sample was gently elutriated to separate the light from the heavy material and then passed through multiple sieves to separate the heavy material into homogeneous sizes. This process indicated that twenty Pleasant Bay samples had large quantities of sand captured on the 0.5 mm mesh screen which had large numbers of the very small bivalve, *G. gemma*, remaining. Due to the dominance of *G. gemma* in the remaining sand, subsampling was conducted to facilitate the counting effort and estimate the number of *G. gemma*.

For samples that contained very high abundance of *G. gemma*, following removal of the preservative, samples were gently washed and the light material was elutriated from the matrix and set aside for sorting. To separate the heavy material into homogeneous sizes to facilitate sorting, the remaining material was sieved through a stack of different sized sieves finishing with a 0.5mm sieve at the bottom of the stack. The sand retained on a 0.5 mm mesh was spread out evenly in a pan marked off with 36 squares. To sort 1/4 of the sand material, a random numbers table was used to select 9 squares of material out of the total of 36 squares (or ¼). Material from the 9 squares was removed from the pan and all *G. gemma*, as well as any incidental remaining molluscs were counted. Raw counts were multiplied by 4 to achieve the estimate for the entire sample and entered on the data sheet.

Grain size samples were analyzed following Section III of the draft MEP Marine Benthic Monitoring Laboratory SOP (Sweeny and Rutecki 2019b). Samples were held longer than the 28 days outlined in the draft QAPP and were frozen to extend the hold time. Grain-size distributions were not altered due to the change in preservation method and no other parameters were analyzed from these samples. One sediment sample (50 mL total volume) from each station, for a total of 32 samples were analyzed in the laboratory.

Grain size was classified following the Coastal and Marine Ecological Classification Standard (CMECS) mineral grain size descriptors adopted from Wentworth (1922; FGDC 2012) and reported as a percentage by weight in six categories as follows:

- Very coarse sand = sum of 2 mm and 1 mm sieve material
- coarse sand = 500 µ to < 1 mm
- medium sand = 250 µ to < 500 µ
- fine sand = 125 µ to < 250 µ
- very fine sand = 63 µ to < 125 µ
- silt = <63 µ

Marine and estuarine sediments generally consist of a mixture of grain sizes. For example, silty sand is defined as the combination of the three smallest sediment size classifications: fine sand, very fine sand, and silt.

Sediment samples for TOC followed the draft MEP Benthic Monitoring QAPP for preservation and hold times. Analytical methods for TOC followed the Lloyd Kahn Method (Kahn 1988).

2.3 Data Analysis

Benthic infauna data were analyzed for the following community parameters: abundance, Shannon-Wiener diversity index (H'), Pielou's evenness (J'), Margalef's diversity index (D_{mg}), Simpson, and Average Taxonomic Distinctiveness (ATD), using the PRIMER v5 (Plymouth Routines in Multivariate Ecological Research) software program (Warwick and Clarke 1991, Clarke and Gorley 2001). Shannon-Weiner (H') was calculated using log base e-transformed data.

Multivariate analyses were performed using PRIMER v5 software to examine spatial patterns in the overall similarity of benthic assemblages in Pleasant Bay (Clarke 1993, Clarke and Warwick 2001). These analyses included classification (cluster analysis) by hierarchical agglomerative clustering with group average linking and ordination by non-metric multidimensional scaling (MDS). Bray-Curtis similarity was used as the basis for both classification and ordination. Similarity measures compare counts within each taxon between all possible pairs of samples. Values range from 0, when two samples have no taxa in common, to 100 when two samples are identical in taxa and counts within taxa. MDS outputs a two-dimensional plot where spatial proximity illustrates relative similarity between samples and is interpreted by the closeness of the samples. Clarke (1993) suggested that a stress level less than 0.20 (shown in the upper right corner of the plot) indicates that a potentially useful two-dimensional representation has been achieved. The results are also presented with a hierarchical clustering tree diagram (a dendrogram), with the x-axis representing the full set of samples, and the y-axis defining a similarity level at which two samples or groups are considered to have fused (Clarke and Warwick 2001). For the purpose of reducing the influence of high-density outliers, densities were square-root transformed before calculating similarity. The square-root transformation decreases the influence of the most abundant species so that rare species factor in more heavily when calculating similarity.

US M-AMBI (multivariate AZTI Marine Biotic Index in United States coastal waters) was calculated following Pelletier et al. (2018) to determine Pleasant Bay sub-embayment and embayment soft bottom habitat health. Modifications to the existing M-AMBI taxonomic classification (Ecological Grouping [EG]) were made prior to using the program utilizing the taxonomic list and corresponding EGs established by Pelletier et al. (2018) to be specific for the northeast US region. Each taxon identified is classified as EG I, II, III, IV, or V, with I taxa being considered those found in healthy benthic habitats, and V taxa inhabiting low quality habitat. The available published EG taxonomic list is for European studies, and some classifications are not the same as those for other regions. The taxonomic EG list specific to the northeast US region was provide by M. Pelletier (personal communication 2019).

The data were prepared for US M-AMBI by first coding each station in Pleasant Bay as polyhaline (salinity range from 18 to <30 parts per thousand [ppt]) and then assigning each taxon with the Northeast United States EG codes (categories I-V). Some taxa in the Pleasant Bay samples were not included in the data set because no EG code was available for this region at this time (i.e. Oligochaetes and Nemertea), or the specimens were not able to be identified to a low enough taxonomic level (i.e.

Gastropoda and Bivalvia). The Biological Index (BI) was then calculated for each sample using the following formula:

BI = 0*%EG(I) + 1.5*%EG(II) + 3*%EG(III) + 4.5*%EG(IV) + 6*%EG(V)

Species richness (S) and Shannon-Weiner diversity index (H') were calculated for all species (including Oligochaetes, Nemertea, etc.) using PRIMER. These four parameters (salinity code, BI, S, and H') were then run through the R script for the Northeast United States provided by M. Pelletier (personal communication 2019). The output number corresponding to benthic health condition falls within the following categories: Bad (<0.20), Poor (0.20 to 0.39), Moderate (0.39 to 0.53), Good (0.53 to 0.77), and High (>0.77).

3 Results and Discussion

3.1 Water Quality

Water quality in Pleasant Bay was characterized in 2020 by measuring four parameters at each of the 33 sampling locations: water temperature, DO, pH, and salinity (Appendix A). As mentioned above, Pleasant Bay is designated as SA waters. The criteria for SA waters states DO shall not be less than 6.0 mg/L, temperature shall not exceed 29.4°C (85°F) nor a maximum daily mean of 80°F (26.7°0C), and pH shall be between 6.5 and 8.5 standard units and not more than 0.2 standard units outside of the natural background range (314 CMR 4.00). The majority of the water quality readings recorded during this survey met the SA water quality criteria. Four DO readings at two Meetinghouse Pond stations fell below 6.0 mg/L (one at Station 48 and three at Station 49 [Appendix A]). All DO readings during the survey were above 5.0 mg/L.

3.2 Sediment Composition

Sediment conditions in Pleasant Bay were characterized in 2020 by measuring two parameters at each sampling location where grab samples could be collected: (1) grain size and (2) total organic carbon (Table 2). In addition, the following field observations of the bottom conditions were recorded. Sediments in the Pleasant Bay Estuary ranged from sand at stations in Pleasant Bay (PB20, 14, 37), Little Pleasant Bay (LP43, 37, 36, 35), and Chatham Harbor (CH12) to soft black mud with a sulfur smell in Round Cove (RCV1, RCV2, and RCV3) and Meetinghouse Pond (MP47, 48, 49). Loose algae masses were observed in Meetinghouse Pond (MP47) and Lonnies Pond (LP53). Algae clumps were observed in Ryders Cove (RC). Sponge material was sporadically observed at Lonnies Pond (LP53) and Crows Pond (CPE and CPW). A horseshoe crab (*Limulus polyphemus*) was observed at two stations, TR45 and TR50. Eelgrass was observed at station LPB and along the channel edges while entering Crows Pond. Large patches of eelgrass were observed while navigating to Station 37 in Little Pleasant Bay (LP37) although no patches were observed at the station.

3.2.1 Grain Size Analysis

Surface sediments collected at 32 sampling locations in 2020 contained a range of sand and silt sediments summarized in Table 2 and Figure 6 below. Percentage of sediment types in the Pleasant Bay samples varied within and among basins; overall sediments were less silty in the stations in the Outer Bay, with the most direct access to tidal flushing (Little Pleasant Bay Stations 43, 36, 35, and 37; Pleasant Bay Stations 20, 16, and 14; Muddy Creek Mouth station; and Chatham Harbor Station 12; Table 2,

Figure 6). The exception was Station 50 in The River basin, which also had a relatively low percentage of silt, but was located in the Inner Bay to the north, with little access to tidal flushing. Percent silt ranged from 0 at PB16 to 84.9% at Meetinghouse Pond station MP49 (located in the Inner Bay). These results are consistent with surface sediments observed in Pleasant Bay by Hughes and Mittermayr (2018; Figure 7).

In general, higher percentages of organic matter deposition (e.g. silt) to the sediments result in a relatively lower benthic habitat quality (Howes et al. 2006). Silty sediments are generally inhabited by low-diversity, shallow-dwelling organisms compared to high-diversity deep-burrowing organisms found in more sandy sediments (Howes et al. 2006). However, sediments that are predominantly coarse sand due to high levels of tidal flushing or scouring, as observed at the Chatham Harbor Station 12 located near the breach, may also have relatively low diversity.

3.2.2 Total Organic Carbon

Organic matter in sediments can form water-soluble and water-insoluble complexes with metal ions and hydrous oxides, interact with clay minerals and bind particles together, adsorb and desorb both natural and man-made organic compounds, and absorb and release nutrients (Schumacher 2002). Therefore, total organic carbon (TOC) is an important parameter in characterizing the health status of a site because the level of TOC can markedly influence how chemicals will react in the sediment (Schumacher 2002). Three basic forms of carbon may be present in sediments: elemental carbon (from charcoal, soot, graphite, and coal), inorganic carbon (from geologic or soil parent material sources), and organic carbon (derived from the decomposition of plants and animals). In addition to the naturally occurring organic carbon sources, anthropogenic activities can also increase the total carbon content to sediment. For example, spills or releases of contaminants into the environment increase the total carbon content in the sediment. In general, though, the total carbon contribution from contaminants to the total organic carbon content in sediment is relatively small to negligible unless a fresh spill has occurred (Schumacher 2002).

TOC in the stations sampled in Pleasant Bay were variable ranging from <0.05% in the Pleasant Bay Basin (Station 16) and Chatham Harbor (Station 12) to 5.61% in Meetinghouse Pond (Station 47; Table 2; Figure 8). In general, TOC was highest in the inshore locations (e.g. Paw Wah Pond Station 46 [4.77%], Areys Pond Station AP 22/23 [4.31%] and Lonnie's Pond Station 53 [3.86%]) with relatively low levels of tidal flushing and decreased in the outer Pleasant Bay locations (e.g. Pleasant Bay [0.65%], Little Pleasant Bay [0.25%], Pochet [1.39%], and Chatham Harbor [<0.05%]). Higher TOC values were associated with a higher percent silt (Figure 8).

Table 2. Results for Pleasant Bay sediment grain size and TOC in 2020.

Sediment	MP47	MP48	MP49	TR50	TR52	TR45	TR26	LP53	AP22/23	PWP46	P39	P41	LPB43	LPB36	LPB35	LPB37
Very coarse sand	1.3	1.3	1.9	11.2	6.5	2.0	4.2	1.4	0.8	8.9	4.7	6.3	1.5	9.8	0.7	1.0
Coarse sand	5.3	3.9	2.2	28.1	0.7	7.6	8.7	3.6	12.8	9.6	7.3	29.2	34.5	61.0	28.8	60.9
Medium sand	12.6	8.5	5.4	36.3	7.2	9.3	17.1	8.2	11.8	30.7	26.9	37.9	51.8	24.5	60.7	31.6
Fine sand	21.3	5.8	1.8	16.0	9.1	1.8	7.9	9.6	8.9	12.3	22.3	3.8	5.7	1.8	6.4	3.4
Very fine sand	7.1	3.1	3.8	3.8	7.2	18.7	13.6	17.3	8.1	12.2	15.2	4.7	2.0	1.3	1.9	2.1
Silt	52.4	77.5	84.9	4.6	69.4	60.5	48.4	59.9	57.6	26.3	23.7	18.0	4.5	1.6	1.5	1.0
TOC	5.6	4.0	4.1	0.8	3.4	2.6	2.8	3.9	4.3	4.8	2.1	0.7	0.4	0.2	0.2	0.2
Sediment	QP3	PB6	PB20	PB32	PB16	PB14	RCV1	RCV2	RCV3	CPW	CPE	MCM	RC	BH	FFC	CH12
Very coarse sand	0.7	1.9	10.9	2.3	4.9	1.1	3.1	3.0	1.1	12.5	5.7	24.8	1.0	1.0	3.5	10.9
Coarse sand	1.8	0.7	31.7	1.8	78.1	20.1	5.9	2.3	1.0	16.8	4.1	50.3	13.2	0.9	3.0	60.6
Medium sand	8.4	1.7	42.1	5.4	16.3	62.6	7.8	7.3	5.6	10.6	6.1	23.4	38.7	3.1	35.6	26.9
Fine sand	10.3	2.9	11.3	13.1	0.6	8.5	6.5	5.8	8.4	22.4	7.4	1.2	13.0	3.4	6.0	1.4
Very fine sand	10.9	11.3	3.3	41.9	0.0	5.6	14.2	16.2	14.2	1.4	21.0	0.1	10.1	14.1	15.8	0.0
Silt	67.8	81.5	0.6	35.6	0.0	2.0	62.5	65.5	69.6	36.2	55.8	0.2	24.0	77.5	36.0	0.2
TOC	3.1	2.1	0.1	0.9	ND	0.2	2.9	3.2	3.3	1.3	2.0	0.1	3.8	1.8	4.1	ND

ND = Non-detectable level (<0.05%)

Basins are delineated by black outline.

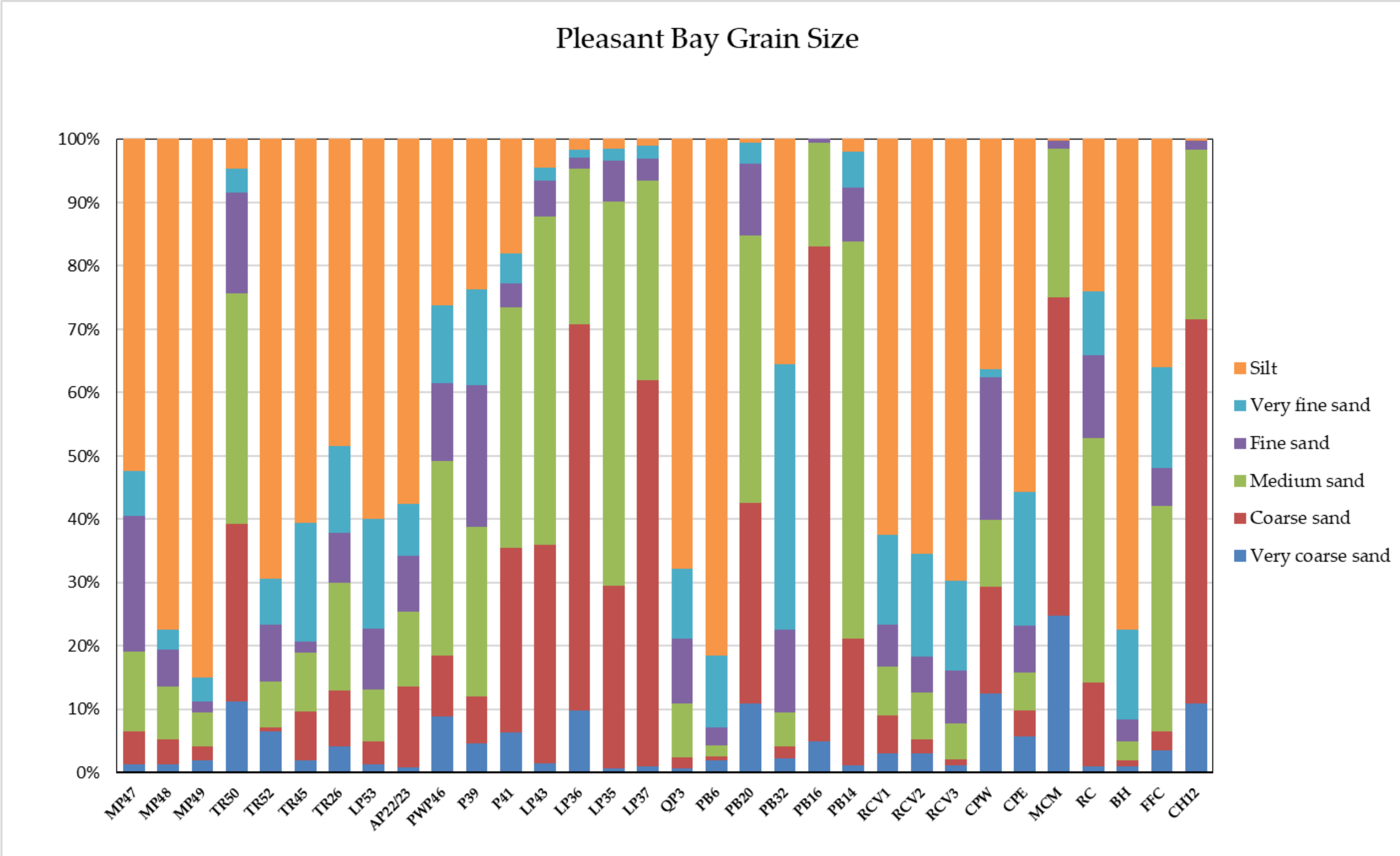


Figure 6. Pleasant Bay grain size analysis, September 2020.

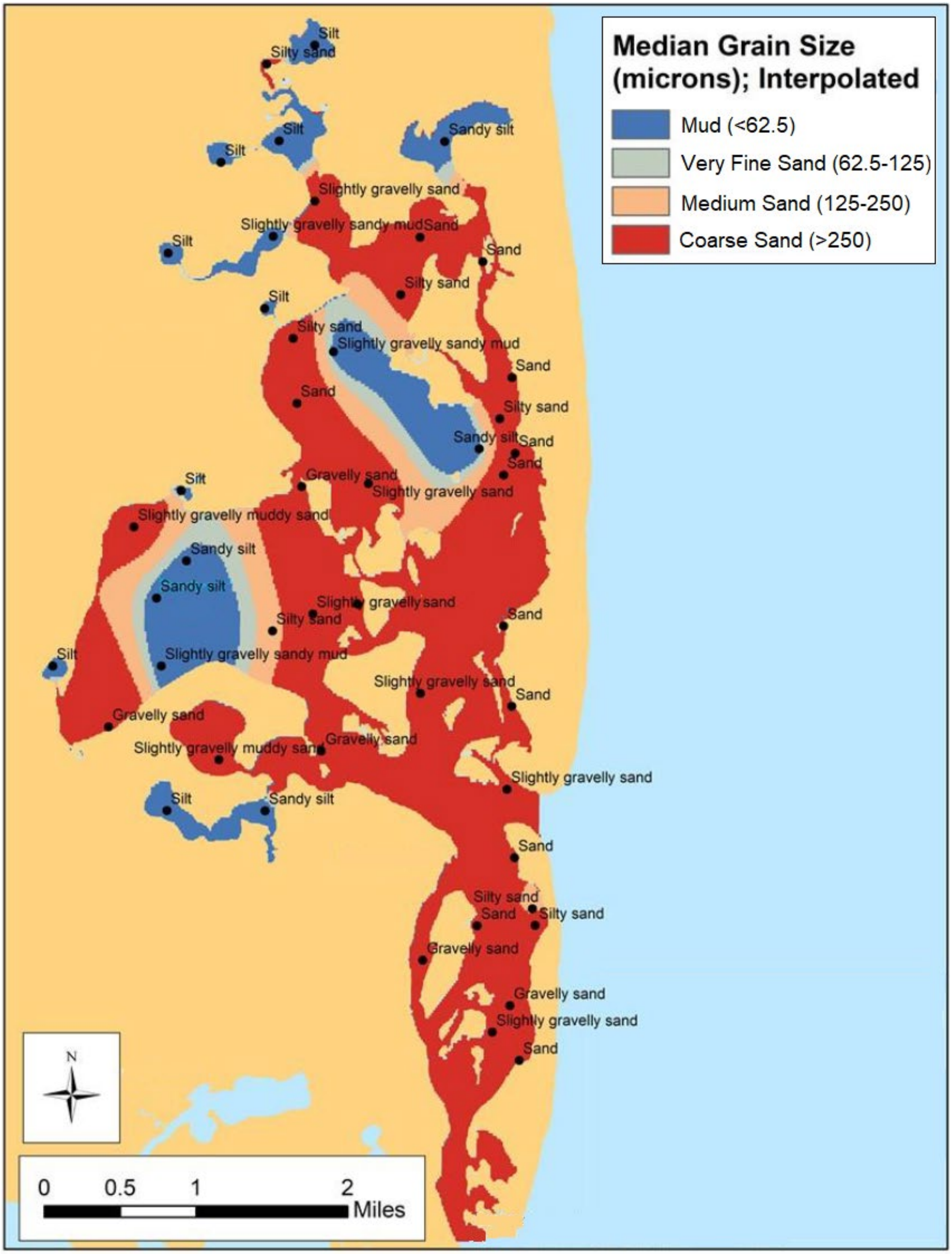


Figure 7. Median grain size in microns (interpolated) for Pleasant Bay (Source: Hughes and Mittermayr 2018, Figure 1.19).

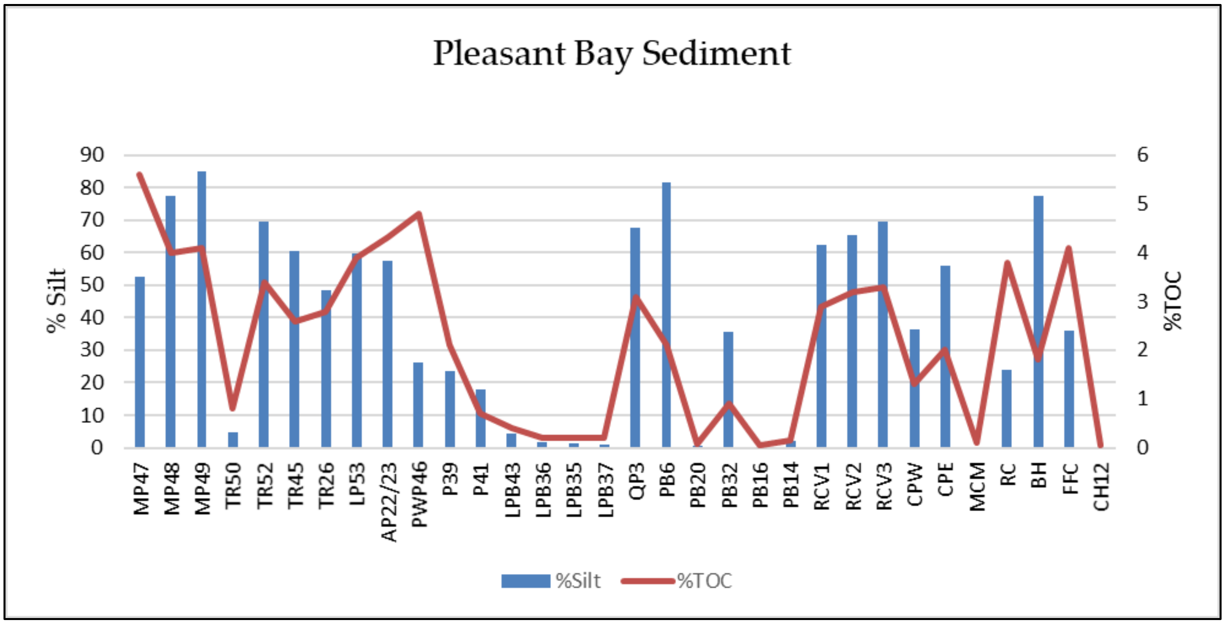


Figure 8. Pleasant Bay sediment, percent silt and TOC, 2020.

3.3 Underwater Digital Images

Digital photographs and video were taken at each station in Pleasant Bay. Still photographs for each sub-embayment can be found in Appendix B. Images of representative habitat types found within Pleasant Bay are provided below in Figure 9. Images of the eelgrass observed at Station Little Pleasant Bay (LPB) are shown above in Figure 5. Eelgrass was not observed at the other sampling locations in 2020. This is consistent with results from the MassDEP Eelgrass Mapping Project that indicate eelgrass has declined in some areas of Pleasant Bay from 2006 to 2019 (Costello and Kenworthy 2011, MassDEP 2021).

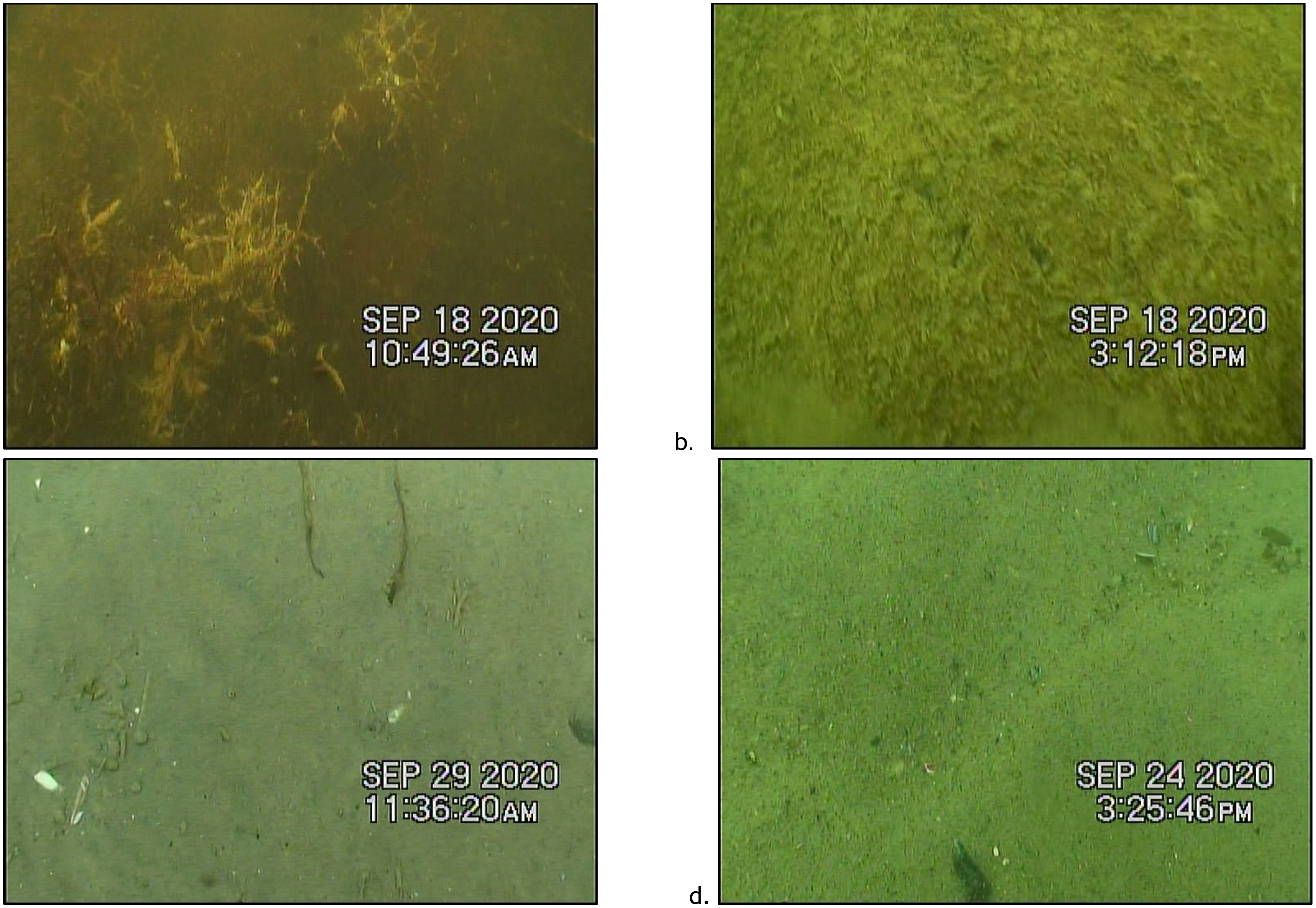


Figure 9. Images of Pleasant Bay bottom habitat: a) Station 47 (macroalgae), b) Station 52 (infaunal tubes), c) Station 37 (sandy sediment), d) Station 16 (coarse sand in a dynamic area), e) Station 14 (medium-coarse sand), f) Ryders Cove (silty-sand with macroalgae) and g) Bassing Harbor (silty sediment).

e.


f.


g.


Figure 9. (Continued)

3.4 Benthic Infauna Community

The 2020 Pleasant Bay benthic samples contained a total of 156 taxa, representing nine phyla (Table 3). The Pleasant Bay benthic communities were characterized based on the following macroinvertebrate metrics: number of species (S), abundance (N), species richness (Magalef, D_{mg}), diversity (Shannon-Weiner, [H’] and Simpson’s index [1-λ]), and evenness (Pielou, J’; Table 4). In addition, Average Taxonomic Distinctness (ATD), cluster and non-metric multidimensional scaling (MDS) analyses, and US M-AMBI are presented to assess spatial and temporal trends in community composition within and between sub-embayments, and eventually between estuaries. Due to the complexity and size of Pleasant Bay, the cluster and MDS analyses are presented first to provide groups based on similarity for which the remaining metrics could be discussed in the report. Since US M-AMBI incorporates several of the above metrics (i.e. species number, Shannon-Weiner diversity H’, salinity category, and BI score [see Methods section above]), US M-AMBI was used as an overall summary of the benthic habitat health status.

3.4.1 Cluster and non-metric multidimensional scaling (MDS)

Pleasant Bay is a relatively large complex embayment. As indicated above, benthic infaunal grabs were collected at 32 stations in 12 sub-embayments or basins within Pleasant Bay. Multivariate analyses were used to assess spatial patterns in the infaunal assemblages at the Pleasant Bay sampling stations. Replicates within each station exhibited high similarities in community structure and grouped together. The cluster analysis identified four main assemblages in the Pleasant Bay benthos (Figure 10). The patterns identified through cluster analysis were confirmed in the MDS ordination plot (Figure 11). Spatial patterns in the faunal assemblages of Pleasant Bay reflect a gradient from inner embayments and Bay areas to the main Bay and outer Bay. Group I (Meetinghouse Pond Stations 47 and 48) and Group II (Meetinghouse Pond Station 48, Areys Pond, Lonnie’s Pond, and Ryder’s Cove) assemblages are located in inner embayments and Bay areas. The Group III assemblage contains a mix of stations from areas in the inner Bay, main Bay, and outer Bay but generally represents the assemblages found in the Main Bay areas (e.g. Pleasant Bay, The River, Little Pleasant Bay, Bassing Harbor sub-system). Group III consists of two groups containing of several subgroups. Group IIIA was divided into three subgroups (IIIA1, IIIA2, and IIIA3), and Group IIIB was divided into two subgroups (IIIB1 and IIIB2). Additionally, Station 6 and Station 32 in Pleasant Bay were not grouped with other subgroups but were part of the larger Group IIIA, and Station 37 in Little Pleasant Bay was not grouped in a subgroup but was part of Group IIIB. Group IV consisted of Station 12 in Chatham Harbor and Station 16 in Pleasant Bay, two very dynamic areas in the outer Bay.

Percent silt for each station within the Bay was superimposed on the MDS plot (Figure 12). Groups I and II with stations in the inner reaches of the Bay (Meetinghouse Pond, Areys Pond, Lonnies Pond, and Ryders Cove) have high levels of silt. Group III with stations in the main Bay has a range of percent silt, with stations containing the Group IIIA assemblages having higher silt levels than stations with the Group IIIB assemblages. Group IV, the outer Bay stations 12 and 16, contain almost no silt. These two stations are near the breach in the south of Pleasant Bay and are exposed to high tidal flushing.

Table 3. Taxonomic list for Pleasant Bay benthos, 2020.

Taxonomic Group	Scientific Name	Taxonomic Group	Scientific Name
Polychaeta	Aglaophamus sp.	Polychaeta	Polycirrus sp.
	Alitta virens		Polydora cornuta
	Ampharete oculata		Polygordius jouinae
	Archiannelida		Prionospio cirrifera
	Aricidea (Acmira) catherinae		Prionospio heterobranchia
	Armandia sp.		Prionospio sp.
	Brania sp.		Prionospio steenstrupi
	Brania wellfleetensis		Protodrilidae
	Capitella capitata		Pygospio elegans
	Capitella jonesi		Sabaco elongatus
	Capitellidae		Salvatoria clavata
	Cirratulidae		Scolelepis squamata
	Clymenella torquata		Scoletoma tenuis
	Clymenella zonalis		Sphaerosyllis taylori
	Drilonereis longa		Spiophanes bombyx
	Eteone sp.		Streblospio benedicti
	Euclymene collaris		Streptosyllis verrilli
	Eumida sanguinea		Stygocapitella sp.
	Exogone dispar		Syllidae
	Fabricia stellaris		Syllides setosa
	Glycera americana		Syllides sp.
	Glycinde solitaria		Terebellidae
	Gyptis vittata		Tharyx acutus
	Harmothoe extenuata	Oligochaeta	
	Heteromastus filiformis	Hirudinea	
	Hypereteone heteropoda	Gammarid Amphipod	Acanthohaustorius millsi
	Hypereteone lactea		Ampelisca abdita
	Leitoscoloplos robustus		Ampelisca sp.
	Leitoscoloplos sp.		Ampelisca vadorum
	Lysidice ninetta		Ampelisca verrilli
	Maldane sarsi		Apocorophium acutum
	Maldanidae		Corophiidae
	Marenzelleria viridis		Corophium sp.
	Mediomastus ambiseta		Cymadusa compta
	Microphthalmus aberrans		Elasmopus levis
	Microphthalmus sczelkowii		Eobrolgus spinosus
	Neanthes arenaceodentata		Gammarus lawrencianus
	Nereididae		Gammarus mucronatus
	Nicolea venustula		Hyale plumulosa
	Notomastus latericeus		Idunella clymenellae
	Oxydromus obscurus		Lysianopsis alba
	Parahesione luteola		Melita nitida
	Paranaitis speciosa		Microdeutopus anomalus
	Paraonis fulgens		Microdeutopus gryllotalpa
	Parasabella microphthalma		Microprotopus raneyi
	Pectinaria gouldii		Monocorophium acherusicum
	Pholoe minuta		Phoxocephalus holbolli
	Phyllodoce mucosa		Rhepoxynius epistomus
	Pista sp.		Unciola irrorata
	Polycirrus eximius		Unciola serrata

Table 3. Continued.

Taxonomic Group	Scientific Name	Taxonomic Group	Scientific Name
Isopod	Asellota	Bivalvia	Astarte sp.
	Chiridotea arenicola		Bivalvia
	Chiridotea tuftsii		Chione sp.
	Edotia montosa		Gemma gemma
	Edotia sp.		Limecola balthica
	Erichsonella attenuata		Lyonsia arenosa
	Erichsonella filiformis		Lyonsia hyalina
	Erichsonella sp.		Macoploma tenta
	Exosphaeroma diminutum		Mactromeris polynyma
	Idotea balthica		Mercenaria mercenaria
	Idotea phosphorea		Mytilidae
	Idotea sp.		Nucula proxima
	Janira alta		Pitar morrhuanus
	Janira maculosa		Solemya sp.
	Janiridae		Solemya velum
Caprellid amphipod	Caprella sp.		Spisula solidissima
	Caprellidae		Tellina sp.
Cumacea	Leucon americanus		Yoldia limatula
	Oxyurostylis smithi		Yoldia sp.
Mysid shrimp	Mysis gaspensis	Ascidiacea	Ascidia sp.
	Heteromysis formosa		Ascidiacea
Decapod	Callinectes sp.	Anthozoa	Actiniaria
	Crangon septemspinosa		Ceriantharia
Tanaidacea	Hargeria rapax		Nematostella vectensis
Pycnogonida	Anoplodactylus petiolatus		Urticina felina
Horseshoe crab	Limulus polyphemus	Bryozoa	Amathia sp.
Gastropoda	Ameritella agilis		Botryllus schlosseri
	Crepidula fornicata	Nematoda	
	Ecrobia truncata	Turbellaria	
	Eupleura caudata	Nemertea	
	Haminella solitaria		
	Hydrobia sp.		
	Odostomia eburnea		
	Odostomia sp.		
	Tritia obsoleta		
	Turbonilla interrupta		
	Turbonillinae		

Table 4. Mean 2020 Pleasant Bay infaunal community parameters by station.

Area	Group	Sample	S	N	d	J'	H'(loge)	1-Lambda'
Inner Bay	Group I	MP49	9	138	1.63	0.96	2.10	0.87
		MP47	12	2875	1.38	0.57	1.42	0.61
	Group II	MP48	5	900	0.59	0.53	0.85	0.46
		AP22/23	9	850	1.19	0.62	1.36	0.62
		LP53	16	3688	1.83	0.41	1.15	0.44
		RC	16	3338	1.85	0.54	1.50	0.67
		TR52	26	14563	2.61	0.42	1.36	0.54
Main Bay	Group IIIA1	FFC	22	15238	2.18	0.56	1.73	0.75
		QP3	22	19600	2.13	0.45	1.40	0.64
		BH	40	17975	3.98	0.66	2.42	0.86
	Group IIIA2	PWP46	38	34200	3.54	0.58	2.10	0.80
		TR26	37	35663	3.44	0.44	1.61	0.67
		TR50	32	51513	2.86	0.57	1.97	0.80
		CPW	46	68775	4.04	0.56	2.13	0.80
		CPE	50	56450	4.48	0.49	1.93	0.73
		P39	39	47838	3.53	0.62	2.28	0.84
		TR45	36	133638	2.97	0.33	1.17	0.45
		P41	49	77725	4.26	0.38	1.47	0.51
	Group IIIA3	RCV1	20	58675	1.73	0.64	1.91	0.75
		RCV2	28	120588	2.31	0.51	1.69	0.74
		RCV3	24	37300	2.19	0.60	1.90	0.80
		PB6	31	34063	2.88	0.51	1.76	0.76
		PB32	29	137813	2.37	0.25	0.86	0.38
	Group IIIB1	PB20	25	5100	2.81	0.73	2.34	0.84
		PB14	25	7563	2.69	0.52	1.69	0.65
	Group IIIB2	MCM	31	24488	2.97	0.52	1.80	0.71
		LPB36	27	28913	2.53	0.36	1.19	0.47
		LPB35	24	40463	2.17	0.27	0.85	0.33
		LPB43	35	32250	3.28	0.43	1.53	0.55
		LPB37	38	147588	3.11	0.14	0.50	0.17
Outer Bay	Group IV	PB16	10	7925	1.00	0.54	1.23	0.60
		CH12	10	3200	1.12	0.54	1.24	0.57

S = number of taxa, N = number of individuals, d = Margalef's species richness, J' = Pielou's evenness, H' = Shannon-Weiner diversity index, and 1-λ = Simpson diversity.

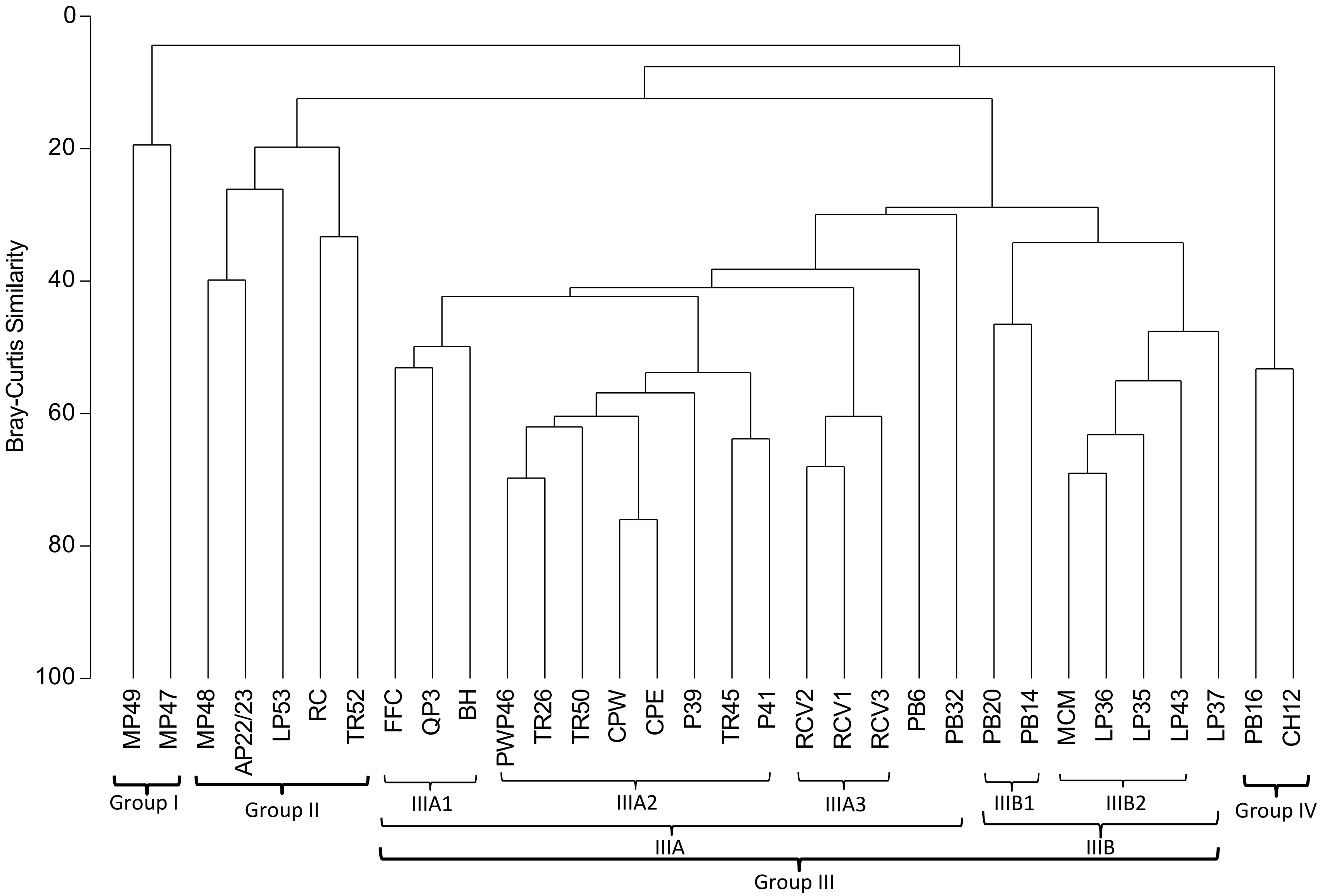


Figure 10. Cluster analysis results of the 2020 Pleasant Bay infaunal samples.

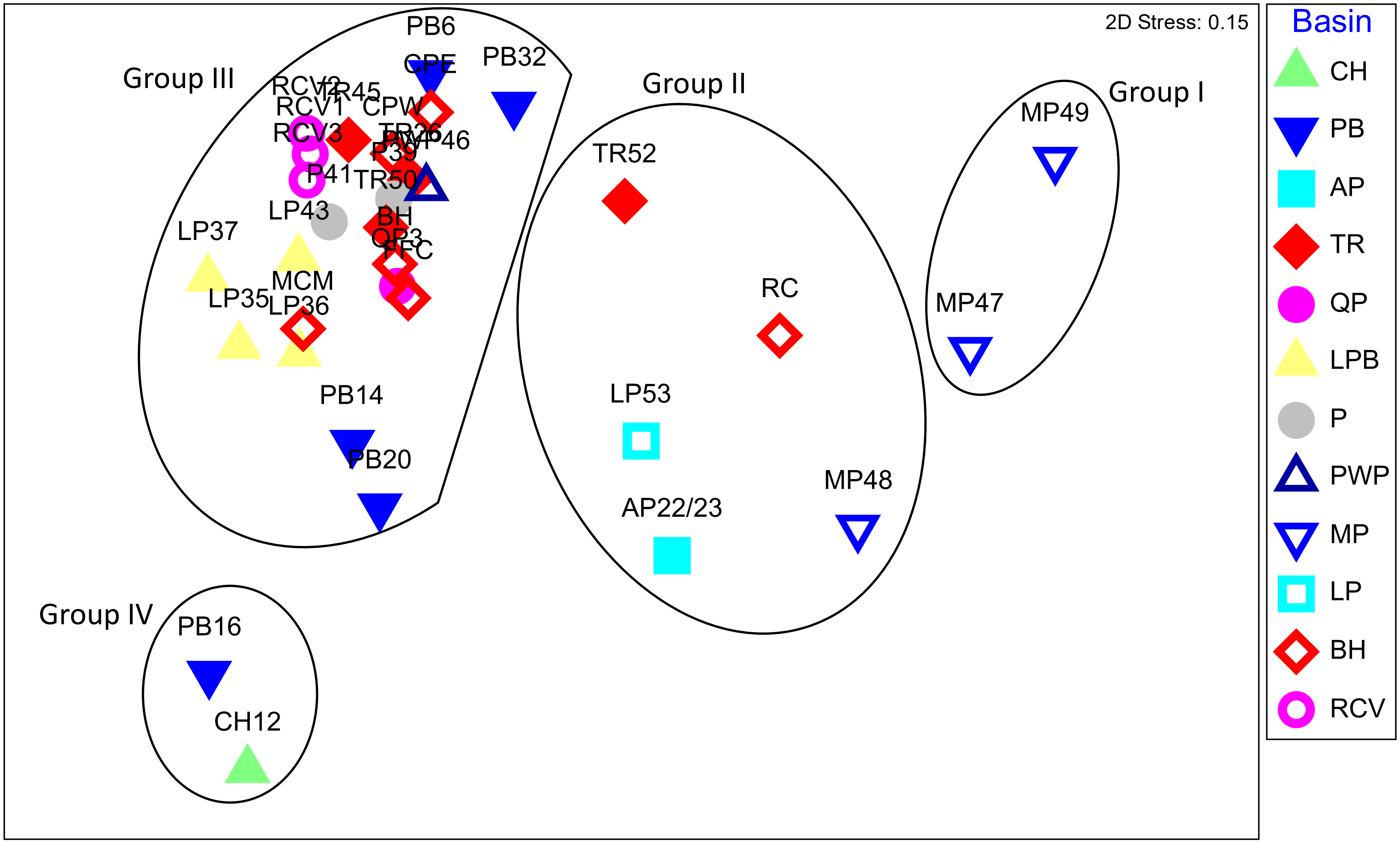


Figure 11. MDS ordination plot of Pleasant Bay 2020 infaunal benthic samples. Each point on the plot represents one of the 32 stations. The symbols represent the 12 basins within Pleasant Bay (Chatham Harbor, Pleasant Bay, Areys Pond, The River, Quonset Pond, Little Pleasant Bay, Pochet, Paw Wah Pond, Meetinghouse Pond, Lonnies Pond, Bassing Harbor, and Round Cove).

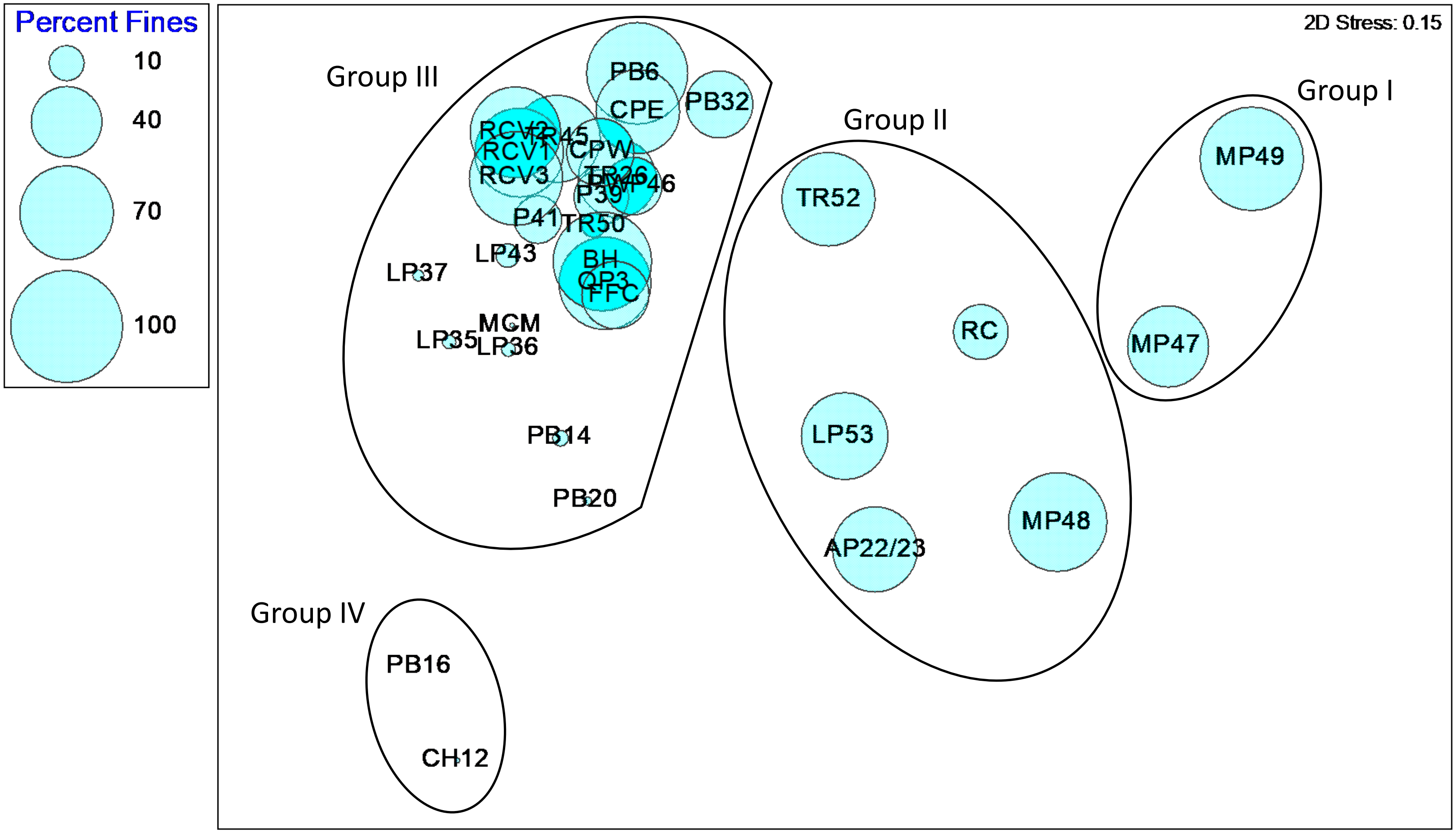


Figure 12. Percent fine sediments superimposed on the MDS ordination plot of the 2020 Pleasant Bay infauna samples. Each point on the plot represents one of the 32 stations; similarity of species composition is indicated by proximity of points on the plot. Faunal assemblages (Groups I-IV) identified by cluster analysis are circled on the plot. The ordination and cluster analysis are both based on Bray-Curtis Similarity.

3.4.2 Dominant taxonomic groups and species

A total of 101,868 individuals from 156 taxa were identified in the 2020 Pleasant Bay benthic samples (Table 3). These taxa represented nine phyla: Annelida (aquatic earth worms and bristle worms), Mollusca (bivalves and snails), Arthropoda (shrimp and crabs), Bryozoa (moss animals), Cordata (tunicates), Cnidaria (sea anemones), Nematoda (roundworms), Platyhelminthes (flat worms), and Nemertea (ribbon worms). In the 2020 Pleasant Bay samples, 73 taxa of polychaetes, 25 taxa of gammarid amphipods, and 18 species of bivalves were identified. Twenty-four taxa comprised 95% of all individuals. The most abundant taxa were *Gemma gemma* (41,745 individuals) and *Ampelisca* sp. (25,076 individuals).

Macroinvertebrates are valuable indicators of pollution due to their relatively sedentary life history and predictably responds to contaminants and eutrophication pollution (Scott 1990; Pelletier et al. 2010). Pelletier et al. (2010) identified benthic invertebrates that could be used as indicator species to detect the presence (pollution-tolerant species) or absence (pollution sensitive species) for various habitats including polyhaline mud and polyhaline sand that are present in Pleasant Bay. The pollution-sensitive and pollution-tolerant indicator species identified in the 2020 Pleasant Bay samples are presented in Table 5.

The following discussion on the dominant taxonomic groups and taxa will be based on the three main bays areas identified in the cluster and MDS analyses as follows:

Inner Bay – Group I (Stations MP49, MP47) and Group II (Stations MP48, AP 22/23, LP53, RC and TR52)

Main Bay – Group III (Stations FFC, QP3, BH, PWP46, TR26, TR45, TR50, CPW, CPE, P39, P41, RCV1, RCV2, RCV3, PB6, PB14, PB20, PB32, LPB35, LPB36, LPB37, LPB43, MCM)

Outer Bay – Group IV (Stations PB16 and CH12)

The top five taxonomic groups for each of the three Bay areas are presented in Table 6 and Figure 13. Polychaetes were among the top three numerical dominants in all three areas, and were the numerically dominant group in the Inner (56%) and Outer Bay (86%). Bivalves were the most numerically dominant in the Main Bay (43%; Table 6, Figure 13).

The top five dominant species differed between the Inner Bay, Main Bay, and Outer Bay areas (Table 7, Figure 14). The most abundant taxa in the Inner Bay were *Capitella capitata*, *Ampelisca* sp., *Erichsonella* sp., *Microdeutopus gryllotalpa*, and *Streblospio benedicti*. The polychaete *Capitella capitata* contributed 45% to the total abundance and comprised 81% of all polychaetes identified in the Inner Bay (Table 7). *C. capitata* is a common, pollution-tolerant indicator species for polyhaline mud habitats (Pelletier et al. 2010; Table 5). The gammarid amphipod genus *Ampelisca* sp. contributed 15% to the abundance of this area (Table 7). A majority (71%) of the *Ampelisca* were small juveniles that were only able to be identified to the genus level; the remaining 29% were identified to species, *A. vadorum. A. vadorum* builds tubes in medium to coarse sands in protected bays and estuaries. The species is common in eelgrass or stable sands in higher salinities (Bousfield 1973). *Erichsonella* sp., an isopod common in eelgrass beds of shallow bays and estuaries (Schultz 1969), accounted for 7% of the total abundance in this area. Although eelgrass was not observed at the stations in which *Erichsonella* sp. was identified, there was eelgrass observed at the nearby station LPB. *Microdeutopus gryllotalpa*, an invasive gammarid amphipod, accounted for 6% of the area's abundance. This amphipod was introduced to North America and is known to have an established population in Massachusetts waters. It builds loosely constructed tubes of mucus on vegetation and hard structure. *M. gryllotalpa* is often associated with man-made and eutrophic habitats in non-native ranges (Bousfield 1973, Wells et al. 2014). *S. benedicti* accounted for 5% of the area's abundance. S. benedicti occurs in mudflats and soft sediments of estuaries and coastal waters, and tolerates a broad range of temperatures and salinities. *S. benedicti* is tolerant to high organic content and pollution, flourishing in disturbed environments. It is considered an opportunistic pioneering species. The taxa recorded in the Inner Bay area did not include pollution-sensitive species that may occur in polyhaline mud or sand (Table 5) suggesting a relatively poor quality habitat.

The top five numerical dominant taxa in the Main Bay area were *Gemma gemma* (42%), *Ampelisca* sp. (including *A. vadorum*, *A. abdita*, and *A. verrilli*; 25%), *Monocorphium acherusicum* (4%), Oligochaeta (3%), and *S. benedicti* (3%). The bivalve *G. gemma* (Amethyst gem clam) made up the vast majority (99%) of all bivalves recorded in this area. Although not numerically dominant, four other pollution-sensitive indicator species, tannaid amphipod *Hargeria rapax*, bivalves *Astarte* sp. and *Nucula proxima*, and polychaete *Salvatoria* (previously *Brania*) *clavata* were recorded in the Main Bay (Table 7) indicative of relatively good quality habitat. There were also three pollution-tolerant species recorded, *C. capitata*, polychaete *Marenzelaria viridis*, both found in mud sediment, and polychaete *Mediomastus ambiseta*, found in sand habitat. The presence of both pollution-sensitive and pollution-tolerant species suggests that the Main Bay contains stations with varying degrees of benthic health.

The five most abundant taxa in the Outer Bay area were *Armandia* sp. (59%), *S. clavata* (24%), oligochaetes (8%), *Brania wellfleetensis* (4%), and Nemertea (3%; Table 7). The top three taxa contributed 87% to the total abundance. *S. clavata* is a pollution-sensitive indicator species in polyhaline mud habitats (Pelletier et al 2010; Table 5). Although not numerically dominant, the pollution-sensitive *G. gemma* was recorded in both stations in the Outer Bay area. The presence of pollution-sensitive species (and no pollution-tolerant species) would normally be indicative of a relatively healthy benthic habitat. While these stations had almost no silt and non-detectable TOC (Table 2) indicating very little pollution, the habitat at these stations is not considered good quality due to the high tidal exchange, constantly clearing the top sediment layers and organisms.

Table 5. Pollution-sensitive and pollution-tolerant indicator species recorded in Pleasant Bay benthos, 2020.

Taxon	Taxonomic Group	Pollution Sensitive/Tolerant	Habitat Type	Recorded in Pleasant Bay (Station or Basin*)	Recorded in Pleasant Bay (Station Grouping)
Hargeria rapax	Tanaid (shrimp-like crustacean)	Sensitive	Polyhaline mud	TR50	Main Bay
Gemma gemma	Bivalvia	Sensitive	Polyhaline mud	MP47, RC	Inner Bay
				PB, TR, QP, LPB, P, PWP, BH*	Main Bay
				CH12, PB16	Outer Bay
Astarte sp.	Bivalvia	Sensitive	Polyhaline mud	PB20, LPB35, CPW	Main Bay
Nucula proxima	Bivalvia	Sensitive	Polyhaline mud	PB6	Main Bay
Salvatoria clavata	Polychaete	Sensitive	Polyhaline mud	PB, LPB, P, PWP, TR, BH*	Main Bay
				CH12 and PB16	Outer Bay
Capitella capitata	Polychaete	Tolerant	Polyhaline mud	PB, TR, QP, LPB, P, PWP, BH*	Main Bay
				LP53, RC, AP22/23	Inner Bay
Marenzellaria viridis	Polychaete	Tolerant	Polyhaline mud	PB14, PB20, PB32, LPB43	Main Bay
Mediomastus ambiseta	Polychaete	Tolerant	Polyhaline sand	TR, QP, LPB, P, PWP, PB, BH*	Main Bay

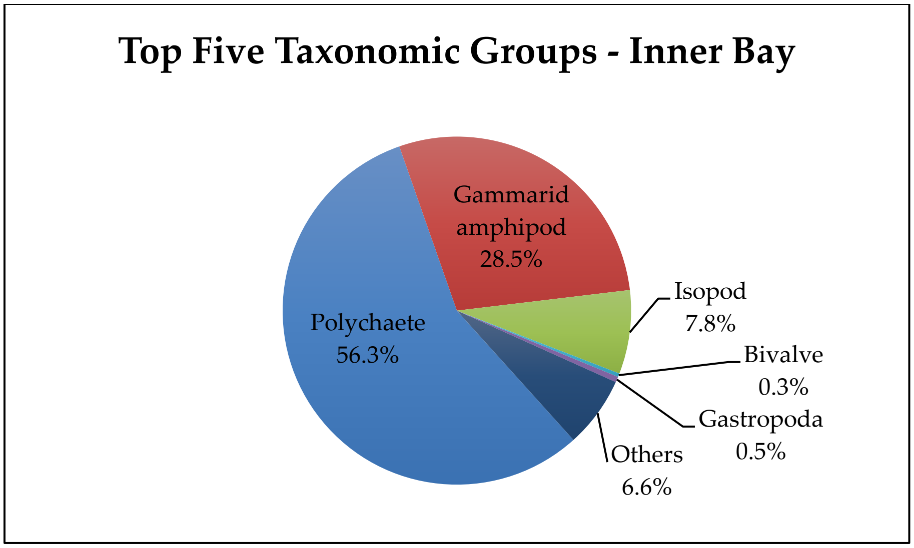
Pelletier et al. 2010. *Basins were presented to save space when the organism occurred in a majority of stations: PB = Pleasant Bay, TR = The River, QP = Quonset Pond, LPB = Little Pleasant Bay, P = Pochet, PWP = Paw Wah Pond, BH = Bassing Harbor.

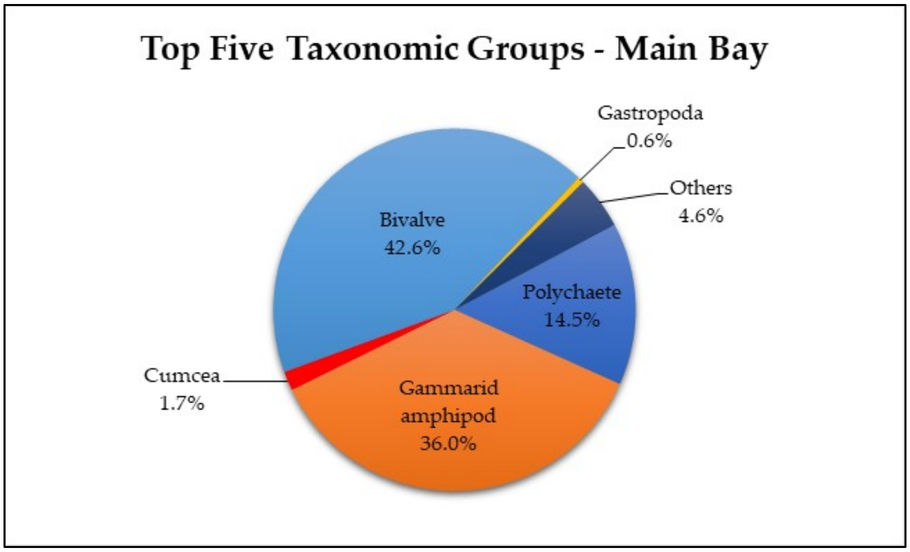
Table 6. Percent contribution of taxonomic groups in the Inner, Main, and Outer Bay areas in Pleasant Bay, 2020.

Taxonomic Group	Inner Bay	Main Bay	Outer Bay
Polychaete	56.3	15.0	86.0
Gammarid amphipod	28.5	36.0	
Isopod	7.8		1.3
Cumacea		1.7	
Bivalve	0.3	42.6	1.9
Gastropoda	0.5	0.6	
Nemertea			2.8
Others	6.6	4.6	7.9

Table 7. Percent contribution of the top five species in the Inner, Main, and Outer Bay areas in Pleasant Bay, 2020.

Taxonomic Group	Taxa	Inner Bay	Main Bay	Outer Bay
Polychaeta	*Capitella capitata*	45.4%		
	Streblospio benedicti	4.9%	2.7%	
	Armandia sp.			59.1%
	Salvatoria clavata			20.4%
	Brania wellfleetensis			4.3%
Amphipoda	*Ampelisca* sp.	15.4%	24.7%	
	Microdeutopus gryllotalpa	6.3%		
	Monocorophium acherusicum		3.7%	
Isopoda	*Erichsonella* sp.	6.6%		
Bivlavia	*Gemma gemma*		42.3%	
Oligochaeta	Oligochaeta		3.0%	7.8%
Nemertea	Nemertea			2.8%
Others		21.4%	23.5%	5.6%





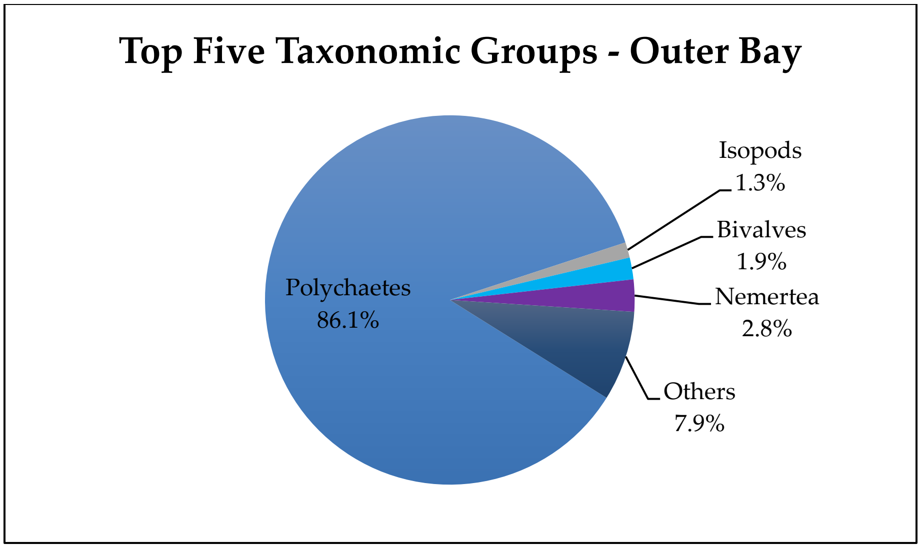


Figure 13. Percentage of benthic groups in Pleasant Bay: Inner Bay (top), Main Bay (middle) and Outer Bay (bottom), June 2020.

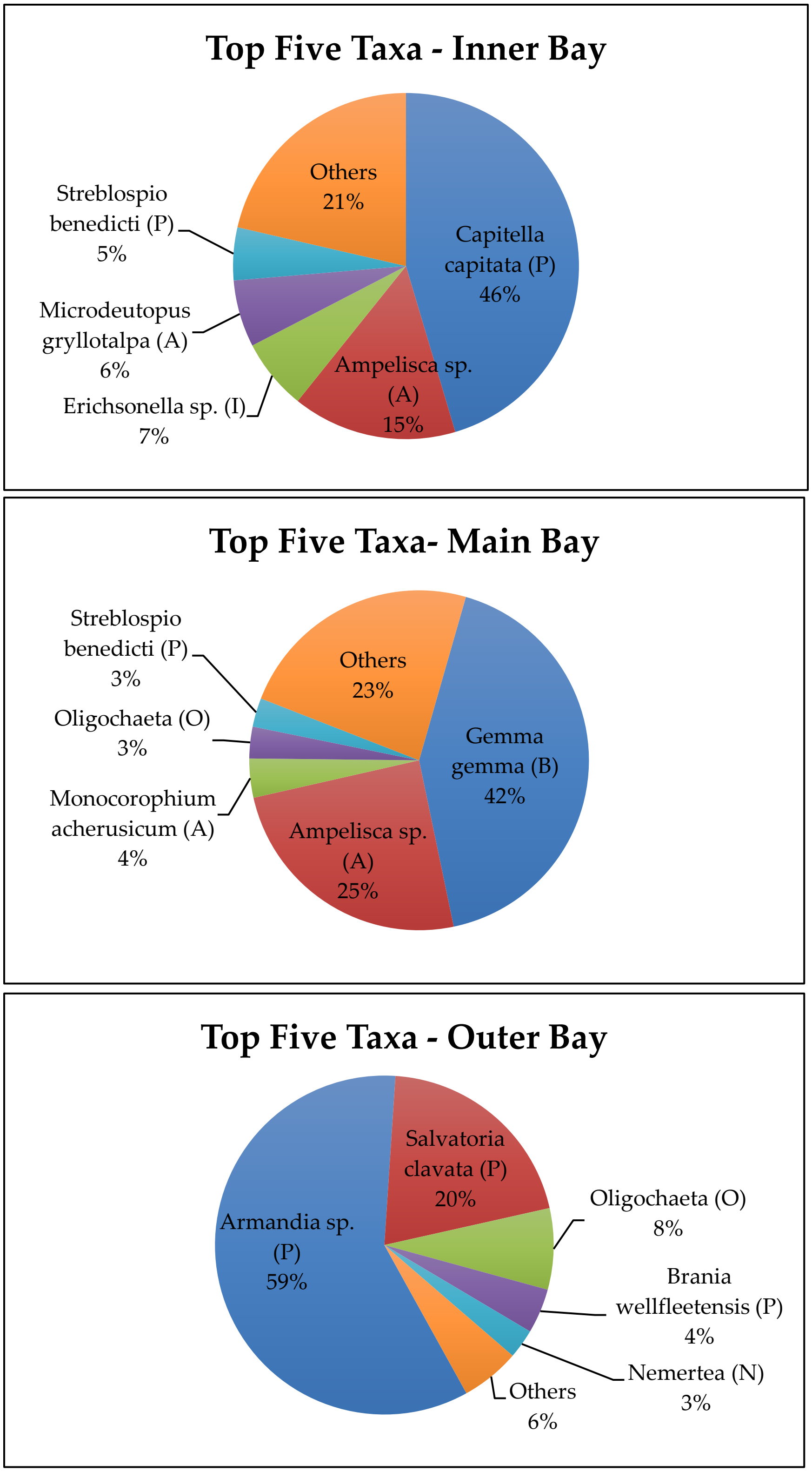


Figure 14. Top five taxa in Pleasant Bay - Inner Bay, Main Bay, and Outer Bay. P = Polychaete, A = Gammarid Amphipod, I = Isopod, O = Oligochaete, B = Bivalve, and N = Nemertea.

3.4.3 Diversity, richness, and evenness indices

In general, in terms of the number of species and abundance, the Main Bay appears to have higher quality benthic habitat compared to the Inner Bay and Outer Bay. The mean number of taxa was higher in the Main Bay (mean of 32 taxa, ranging 20 to 50 taxa per sample) compared to the Inner Bay (mean of 13 taxa, ranging from 5 to 26 taxa per sample) and the Outer Bay (mean of 10 taxa, with 10 taxa in each of the two samples; Table 4). The number of individuals was higher in the Main Bay (mean of 53,627 individuals, ranging from 5,100 to 147,588 individuals per sample) compared to the Inner Bay (mean of 3,765 individuals, ranging from 138 to 14,563 individuals per sample) and the Outer Bay (mean of 5,563 individuals, ranging from 3,200 to 7,925 individuals per sample; Table 4).

Overall, diversity, richness, and evenness indices indicated a relatively healthier habitat in the Main Bay, followed by the Inner Bay, and the lowest habitat conditions in the Outer Bay. The Shannon-Wiener diversity index is a function of the number of different taxa in a sample, the number of individuals per taxa, and the total number of individuals. H' increases with the number of species in the community and when a more even distribution of numbers among taxa is found. H' ranges from 0 when only one species is present to 5.0 when many taxa are found in equal numbers of individuals. Evenness is another expression of how individuals are distributed among different species or taxa. Pielou's evenness index (J') ranges from 0 to 1 and is essentially the reverse of dominance and therefore a sample with low evenness would be highly dominated by a small number of the taxa present. The average Shannon Wiener diversity index was higher in the Main Bay (1.66 with a range of 0.8 to 2.4) compared to in the Inner Bay (1.39, range of 0.8 to 2.1) and the Outer Bay (1.23). Similarly, Margalef's species richness (D_{mg}), and Simpson's diversity (1-λ) indices indicated that the Main Bay had higher habitat quality (e.g. higher richness and diversity, and lower evenness) compared to the Inner and Outer Bays (Table 4). Average Pielou's evenness (J') was lowest in the Main Bay (0.48) compared to the Inner Bay (0.58) and Outer Bay (0.54). This was due to the very high abundance of *G. gemma* in the Main Bay which resulted in relatively low evenness.

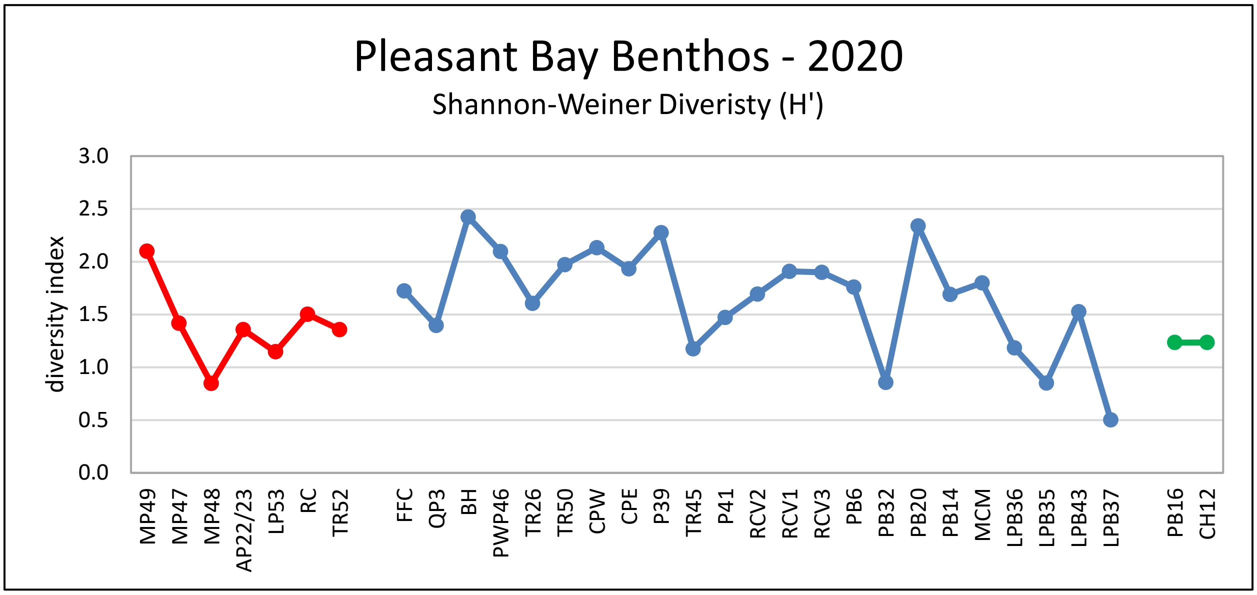


Figure 15. Shannon-Weiner diversity indices for Pleasant Bay Benthos, 2020: Inner Bay (red line), Main Bay (blue line), and Outer Bay (green line).

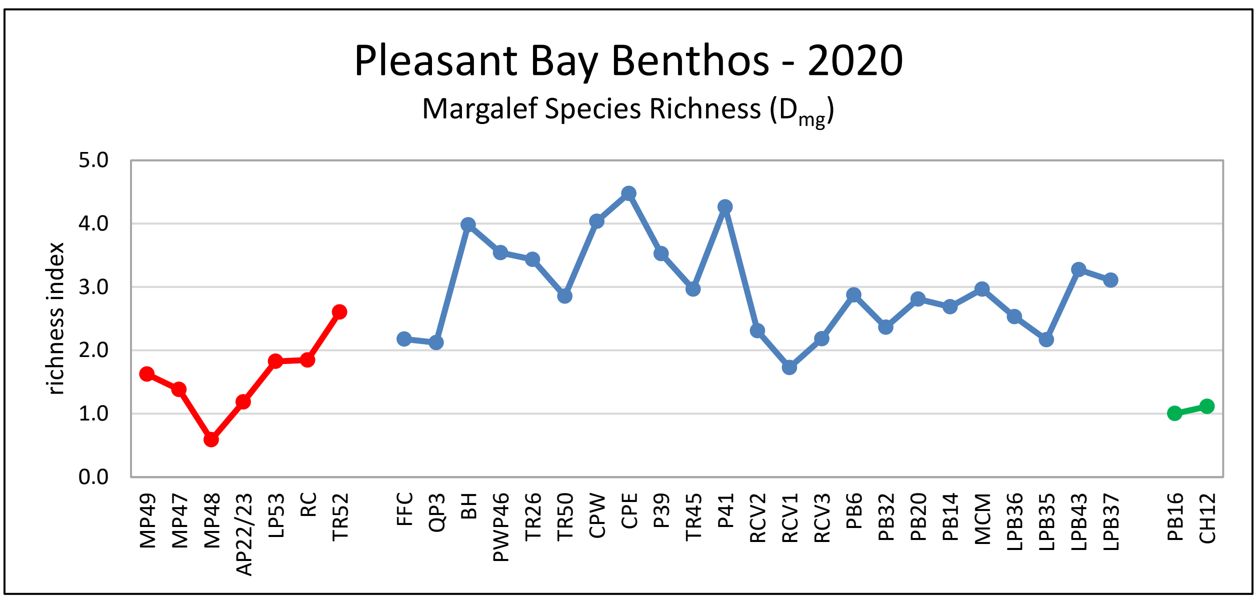


Figure 16. Margalef's species richness indices for Pleasant Bay Benthos, 2020: Inner Bay (red line), Main Bay (blue line), and Outer Bay (green line).

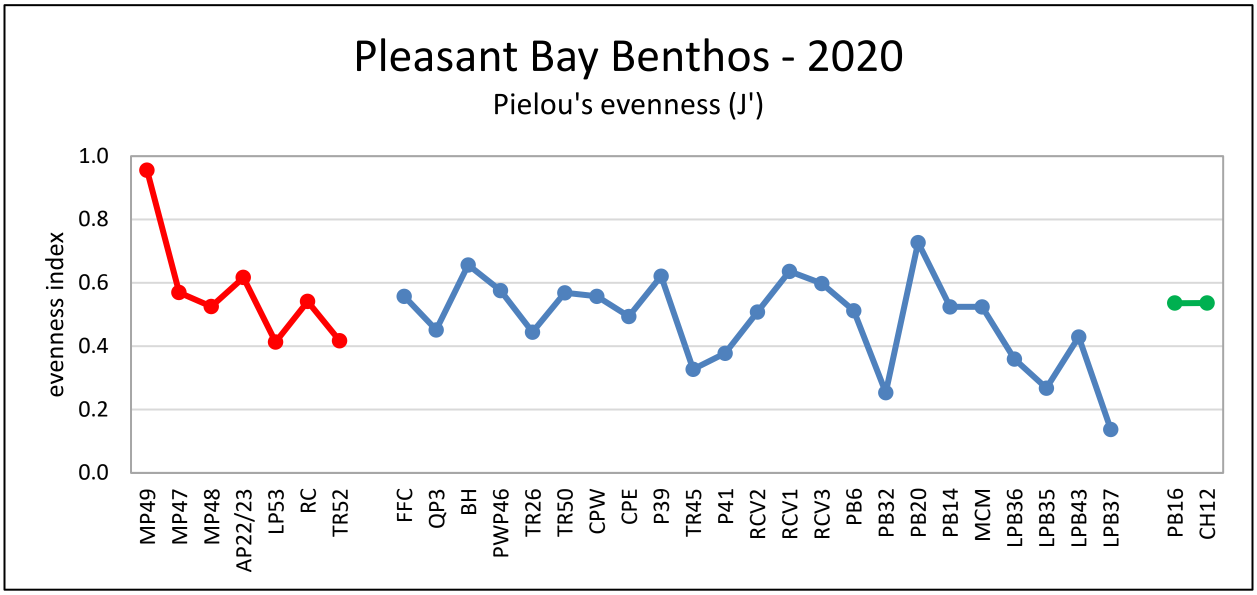


Figure 17. Pielou's evenness indices for Pleasant Bay Benthos, 2020: Inner Bay (red line), Main Bay (blue line), and Outer Bay (green line).

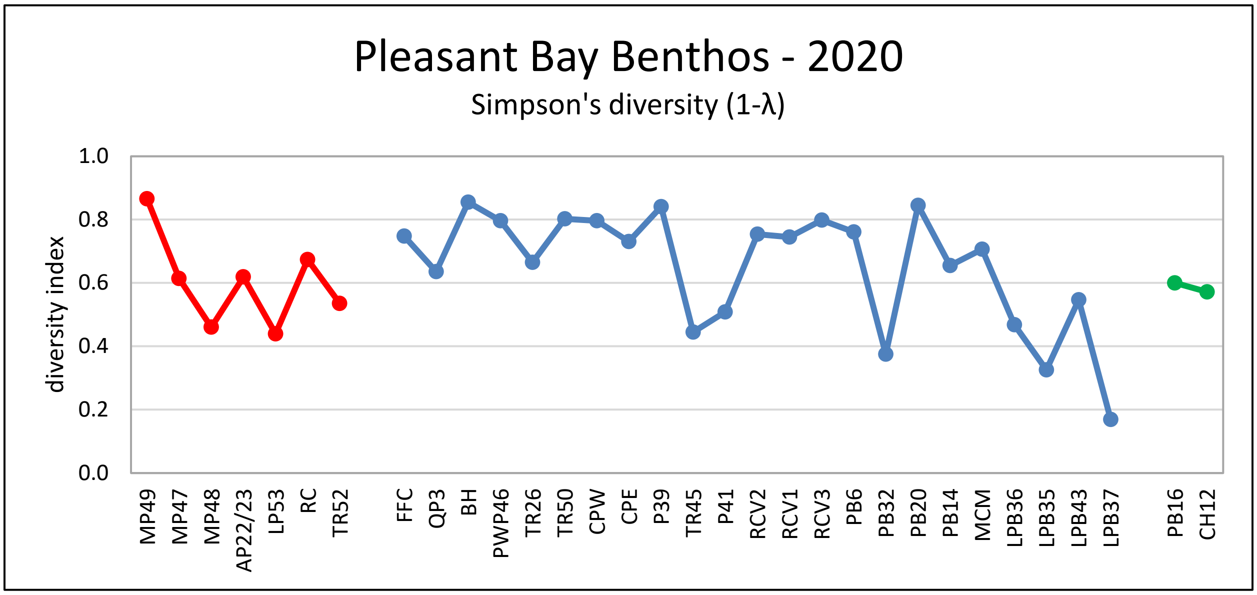


Figure 18. Simpson's diversity indices for Pleasant Bay Benthos, 2020: Inner Bay (red line), Main Bay (blue line), and Outer Bay (green line).

3.5 Average Taxonomic Distinctness (ATD)

Taxonomic distinctness is a biodiversity calculation used to indicate the relatedness of organisms based on Linnaean classification system. Average Taxonomic Distinctness (ATD) is a relatedness measure that can only be calculated from simple species lists (e.g. Phylum, Class, Order, Family, Genus, and Species) but also possesses a robustness to the varying number of species in the lists. More specifically, mean values are unchanged in different-sized sublists generated by random sampling from a larger list. This suggests that it is valid to compare Delta+ over historic time or biogeographic space scales, under conditions of variable sampling effort.

Average taxonomic distinctness (Delta+) for the Pleasant Bay benthos is represented in the funnel plot showing the 95% upper and lower limits of the expected range of diversity (Figure 19). Results indicate that while most samples are within the expected range, five stations (Bassing Harbor [BH], Little Pleasant Bay [LPB] 43 and 36, Muddy Creek [MCM], and Meetinghouse Pond [MP] 49) were below the expected range of biodiversity. Although four of these stations have relatively high number of taxa (BH = 40 taxa, LPB43 = 35 taxa, LPB36 = 27 taxa, and MCM = 31 taxa) there is a disproportionate level of representation from a few families (primarily Spiondae, and Syllidae). MP49 has a relatively small number of taxa (9) and two of these are from the same genus of isopod *Erichsonella*, which would account for this station's reduced taxonomic distinctness. These ATD results appear to be slightly inconsistent with the other community parameters examined (e.g. Shannon- Weiner diversity and Pielou's evenness [Table 4]) for the stations with the relatively high numbers of taxa, therefore a review of this and other taxonomic distinctness tests is recommended to ensure that the measure selected provides the most accurate and useful data.

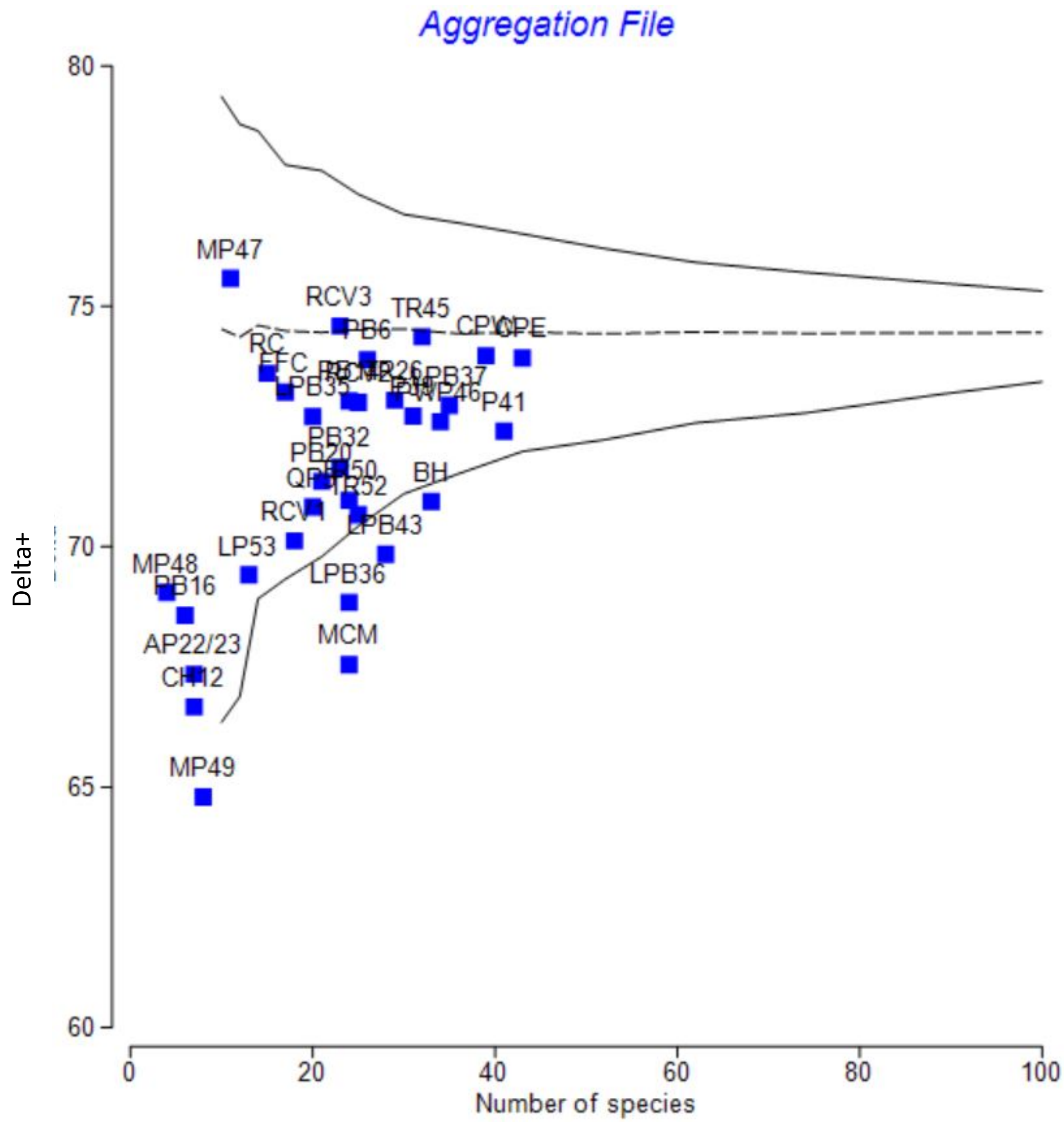


Figure 19. Pleasant Bay Average Taxonomic Distinctness (Delta+) for all stations.

3.6 US M-AMBI

US M-AMBI results for Pleasant Bay indicate that the health of the benthic community in Pleasant Bay ranges from Poor to High (Table 8, Figure 20). US M-AMBI scores in the Inner Bay ranged from Poor (42% of the stations) to Moderate (58% of the stations). The Poor stations were in Lonnies Pond (LP53), Meetinghouse Pond (MP48), Round Cove (RC), and Station 52 (TR52). The US M-AMBI scores are consistent with the sediment grain size and TOC results observed at these stations that indicated relatively poor benthic habitat. The sediment at LP53 and MP48 had relatively high percentages of silt (≥60%) and RC had a moderate percentage of silt (24%). These stations had moderate levels of TOC ranging from 3.8 to 4.0% (Table 2).

The Main Bay ranged from Poor (4% of the stations) to High (7% of the stations), with the majority of stations categorized as Good (85%, Table 8). The station categorized as Poor was Station 32 in the Pleasant Bay basin with sediments containing 36% silt and 0.9% TOC (Table 2). The three stations with High US M-AMBI scores were Bassing Harbor (BH), Crow's Pond West (CPW), and Station 41 in Pochet. Interestingly, the percent silt was high at the BH station (78%) and moderate at the CPW (36%) and Pochet (18%) stations. The TOC at all three stations was low ranging from 0.7% at Station 41 to 1.8% at Bassing Harbor (Table 2). A closer look at the Bassing Harbor benthic community indicated moderate to moderately high densities of several taxa: *A. vadorum*, *Ecrobia truncata* (gastropod), *G. gemma*, and the following polychaetes: *Leitoscoloplos robustus, Scoletoma tenuis, S. benedicti*, and *Tharyx acutus*. Most of these species are either deposit feeders or associated with silty fine sand sediments (Steimle 1982, Dauer et al. 1981, Rice et al. 1986, Pelletier et al. 2010). Thus, reaffirming that the percentage of silt alone is not necessarily a good indicator of benthic habitat health. The stations categorized as Good had a wide range of sediment grain size and TOC results. For example, Stations 35, 36, and 37 in Little Pleasant Bay were predominately medium-coarse sand (>85%) with low TOC (0.2%), while the three stations in Round Cove were predominately silt (>60%) with moderate TOC (approximately 3%, Table 2). The range of sediments in Pleasant Bay associated with Good benthic health condition indicates the importance of other factors, including water quality and flushing, in the health of these benthic communities.

The Outer Bay was similar in health status to the Inner Bay, ranging from Poor (25% of the stations) to Moderate (75% of stations). However unlike the Inner Bay where poor habitat appears to be associated with high silt, the relatively poor habitat at the two stations in the Outer Bay is produced by their position near the North Inlet which results in a very dynamic environment with unstable sediments. The dynamic sediment facilitates colonization of tolerant or opportunistic species generally associated with disturbed or early successional communities.

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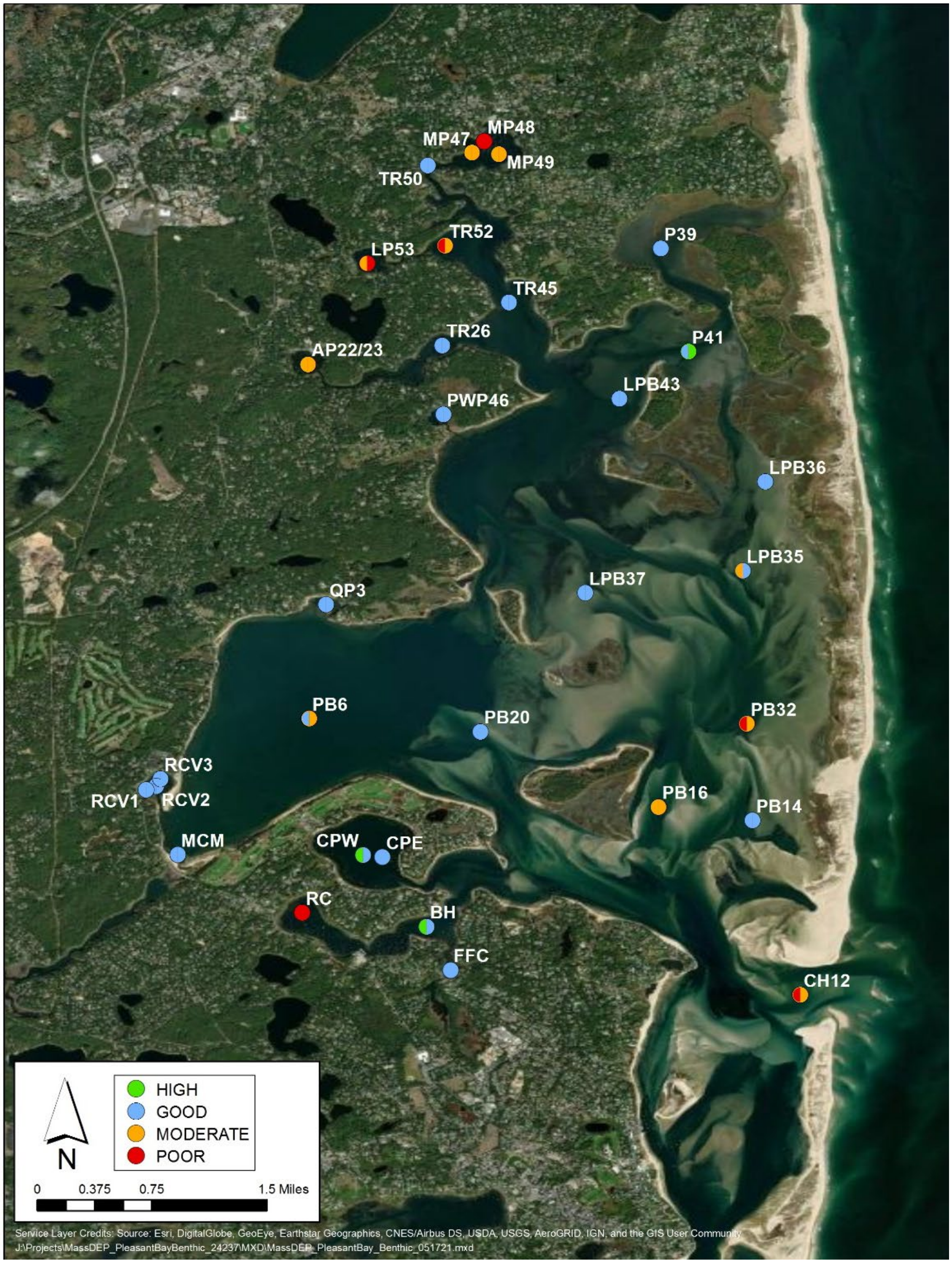

Figure 20. Summary of US-M AMBI results for Pleasant Bay benthos, 2020. The circles at each station location represent qualitative AMBI scores Poor, Moderate, Good, and High for each station. Each circle represents the 2 replicate samples at each station, Replicate 1 on the left and Replicate 2 on the right half of each circle.

Table 8. US M-AMBI score and category for Pleasant Bay benthic samples.

Basin	Station	BI	S	H'	M-AMBI	M-AMBI CATEGORY
Main Bay	FFC_1	0.93	17	1.691	0.603	GOOD
	FFC_3	0.9	16	1.718	0.601	GOOD
	QP3_1	0.98	17	1.315	0.544	GOOD
	QP3_2	1.14	13	1.552	0.544	GOOD
	BH_2	1.25	37	2.376	0.837	HIGH
	BH_3	2.08	32	2.408	0.77	GOOD
	PWP46_1	2.18	32	2.131	0.723	GOOD
	PWP46_3	2.8	25	1.806	0.596	GOOD
	TR26_1	2.2	27	1.612	0.607	GOOD
	TR26_3	2.04	26	1.532	0.595	GOOD
	TR50_2	1.76	29	1.939	0.691	GOOD
	TR50_3	1.15	23	1.787	0.651	GOOD
	CPW_1	1.99	42	2.082	0.796	HIGH
	CPW_2	2.96	30	1.966	0.649	GOOD
	CPE_1	2.76	38	1.949	0.713	GOOD
	CPE_3	2.92	36	1.849	0.677	GOOD
	P39_1	1.74	28	2.012	0.696	GOOD
	P39_3	1.78	31	2.201	0.744	GOOD
	TR45_1	0.81	34	1.209	0.658	GOOD
	TR45_2	0.53	27	1.077	0.6	GOOD
	P41_1	0.53	29	1.021	0.605	GOOD
	P41_3	0.87	44	1.803	0.818	HIGH
	RCV2_2	1.68	24	1.707	0.623	GOOD
	RCV2_3	2.08	20	1.721	0.579	GOOD
	RCV1_1	1.48	16	1.685	0.571	GOOD
	RCV1_2	1.44	16	1.884	0.603	GOOD
	RCV3_1	2.07	20	1.819	0.594	GOOD
	RCV3_3	2.05	22	1.961	0.631	GOOD
	PB6_2	2.72	22	1.7	0.562	GOOD
	PB6_3	2.8	19	1.64	0.527	MODERATE
	PB32_1	3.26	21	0.774	0.39	POOR
	PB32_2	2.68	24	0.8848	0.454	MODERATE
	PB20_1	1.31	18	2.074	0.652	GOOD
	PB20_3	1.61	19	2.215	0.667	GOOD
	PB14_1	1.18	12	1.56	0.536	GOOD
	PB14_2	0.9	24	1.71	0.658	GOOD
	MCM_2	0.29	21	1.76	0.671	GOOD
	MCM_3	0.37	26	1.777	0.706	GOOD
	LPB36_1	0.32	20	1.234	0.582	GOOD
	LPB36_3	0.24	22	1.073	0.576	GOOD
	LPB35_2	0.12	15	0.8297	0.493	MODERATE
	LPB35_3	0.17	23	0.8466	0.552	GOOD
	LPB43_1	0.67	28	1.594	0.679	GOOD
	LPB43_3	0.57	27	1.416	0.649	GOOD
	LPB37_1	0.13	30	0.3698	0.531	GOOD
	LPB37_2	0.22	25	0.7265	0.545	GOOD

Table 8. Continued.

Basin	Station	BI	S	H'	M-AMBI	M-AMBI CATEGORY
Inner Bay	MP49_1	1.71	5	1.475	0.449	MODERATE
	MP49_3	1.5	4	1.386	0.438	MODERATE
	MP47_2	1.32	9	1.269	0.464	MODERATE
	MP47_3	1.45	9	1.494	0.492	MODERATE
	MP48_1	2.37	4	0.9515	0.333	POOR
	MP48_2	2.49	3	0.7374	0.288	POOR
	AP22_23_2	2	6	1.586	0.46	MODERATE
	AP22_23_3	0.75	7	1.049	0.441	MODERATE
	LP53_1	2.85	14	1.047	0.399	MODERATE
	LP53_2	3.28	10	1.077	0.356	POOR
	RC_1	3.78	11	1.319	0.378	POOR
	RC_2	5.07	8	0.9359	0.241	POOR
	TR52_1	5.36	12	0.9448	0.258	POOR
	TR52_3	4.56	23	1.452	0.45	MODERATE
Outer Bay	CH12_1	1.28	5	0.6859	0.348	POOR
	CH12_2	1.14	8	1.243	0.461	MODERATE
	PB16_2	1.24	7	1.121	0.431	MODERATE
	PB16_3	1.25	8	1.27	0.46	MODERATE

BI = Calculated Biological Index (see methods section), S = number of individuals, H' = Shannon-Wiener diversity index

3.7 Comparison with Previous Assessments

Massachusetts Estuaries Program – Pleasant Bay, 2003

The MEP Pleasant Bay 2003 assessment (Howes et al. 2006) and the current 2020 assessment cannot be directly compared as the benthic infaunal community parameters (number of species, total number of individuals, Shannon-Weiner diversity index H', and evenness [E]) presented by Howes et al. (2006) represent a surface area of 0.0625 m^2. The current assessment results were standardized to number per 1 m^2. Sampling methods for the 2003 benthic assessment including the method used to obtain the 0.0625 m^2 sample area are not described in the report. Therefore, it is unknown if this area was a subsample of a 0.1 m^2 Van Veen grab or a different grab type (e.g. Eckmann grab). The surface area was only noted in the title of the results table VII-8 in Howes et al. (2006). The benthic habitat parameters presented by Howes et al. (2006) are summarized in Table 9.

In addition, the 2003 Infaunal Indicators (Healthy [H], Moderately Impaired [MI], Significantly Impaired [SI], and Severely Degraded [SD]) were based on different parameters (dissolved oxygen, chlorophyll a, macroalgae, eelgrass, and number of infaunal animals) than US M-AMBI, and thus cannot be directly compared. US M-AMBI is calculated using ecological groupings based on pollution tolerance and sensitivity of each taxa, salinity, and Shannon-Weiner diversity.

Overall, the 2003 survey indicated that the benthic habitats in the inner basins (Meetinghouse Pond, Lonnies Pond, Areys Pond, Round Cove, Quonset Pond, Paw Wah Pond, Upper Muddy Creek) were generally significantly to severely impaired. Paw Wah Pond was essentially devoid of benthic animals and Areys Pond, Quonset Pond, and Upper Muddy Creek had significantly depleted benthic populations. The other inner basins (Lonnies Pond and Meetinghouse Pond outlet) were able to support benthic infauna, but the community was dominated by opportunistic species (*Capitella* and *Streblospio*) indicative of very high organic matter loading or by intermediate stress indicator species (Gemma, Amphipods; Howes et al. 2006).

The benthic habitats in the larger main basin of Little Pleasant Bay indicated a moderate level of stress from organic matter loading and oxygen depletion, with a pattern of decreasing habitat quality moving from the shore to depths. The Chatham Harbor habitat had only moderate numbers of individuals and species, however this was caused by the dynamic nature of the bottom sediments, due to the high tidal velocities, rather than nutrient related impairment (Howe et al. 2006).

A separate analysis of the habitat quality within the Bassing Harbor sub-system (Ryder Cove, Bassing Harbor, Frost Fish Creek, Crows Pond, and Muddy Creek) was conducted in 2001. The majority of the habitat surveyed within the Bassing Harbor System appeared to have productive and diverse benthic animal communities. The lower regions (those nearest the inlet to Bassing Harbor) showed higher habitat quality, with intermediate to low stress, likely due to a decreased level of nitrogen inputs and tidal flushing from Pleasant Bay (Howes et al. 2006). The tidally restricted Frost Fish Creek had very poor habitat quality with a heavy nutrient and organic matter load. The larger basin within the Bassing Harbor System was generally characterized as intermediate habitat quality. The highest quality habitat areas found in the Bassing Harbor System were Crows Pond and Bassing Harbor (Howes et al. 2006). These infauna indicator analysis results were consistent with the levels of nitrogen and oxygen depletion within these systems in 2001. In addition, the sediment survey results largely supported the concept of high organic matter loading within the upper poor-quality regions of the Town of Chatham embayments (Howes et al. 2006).

Despite the differences in benthic parameters between the 2003 and 2020 surveys, a general comparison can be made. Over the past 17 years, there were two stations in which the benthic habitat has clearly improved. Paw Wah Pond (PWP46) was categorized as SD in 2003, with 2 species and 3 individuals and in 2020 it was characterized as Good habitat, with 38 species and 34,200 individuals per 1 m^2. Similarly, Quonset Pond (QP3) was ranked as SI in 2003 (2 species and 48 individuals) and as Good (22 species and 19,600 individuals per 1 m^2) in 2020. These comparisons can be made because the 2003 results indicated habitat that was so poor that it would have been characterized as Bad or Poor under US M-AMBI, and the fact that in 2020 it was characterized as Good under US M-AMBI indicated improvement. There were several other stations that likely showed improvement (e.g. RCV 1, RCV 2, RCV 3, CPW, and CPE), however, without a clear distinction between the middle categories of the two indicator systems (e.g. MI versus Good and/or Moderate, SI versus Moderate and/or Poor) an appropriate comparison could not be made.

Station LPB is classified as Healthy (by 2003 ranking or High) because of the presence of eelgrass in 2020. It is unlikely that eelgrass was present at the station location in 2003 because benthic samples were taken, and the Little Pleasant Bay sub-embayment was categorized as Moderately Impaired. If eelgrass was not present in 2003, this would indicate a clear habitat improvement. Eelgrass is considered a sentinel species for indicating nitrogen loading in coastal embayments. Changes in eelgrass distribution over time provide a strong basis for evaluating increases or decreases in water quality and nutrient enrichment (Dennison et al. 1993, Short et al. 1995, Howes et al. 2003). Eelgrass provides several ecological functions including shelter and food for a variety of commercial, recreational, and ecologically important organisms, and sediment stabilization (Laney 1997, Thayer et al. 1997).

The quality of benthic habitat clearly decreased from 2003 to 2020 at three stations. Meetinghouse Pond (MP48) was previously ranked as SI and in 2020 was ranked as Poor with 5 species and 900 individuals per 1 m^2. Ryders Cove (RC) was categorized as Moderately Impaired in 2003 and has declined to a Poor habitat with 16 species and 3,338 individuals per 1 m^2. Both of these stations are located in the inner basins of Pleasant Bay, making them more susceptible to eutrophication and low dissolved oxygen conditions. The biggest change in benthic habitat quality in the study occurred at Station 12 in Chatham Harbor. In 2003, prior to the breach in the barrier beach directly to the east (North Inlet), this station was categorized as Healthy, although supporting only a moderate number of individuals and species. After the 2007 breach, the large tidal volume has remodeled this location in Pleasant Bay leaving a benthic habitat of coarse sand, with very few organisms, categorizing it as Poor in the 2020 assessment.

Howes et al. (2006) designated 3 primary sentinel stations for Pleasant Bay located at the head of Little Pleasant Bay, in Ryder's Cove (Station RC), and Bassing Harbor (Station BH) for eelgrass and infauna due the size of the Pleasant Bay system. The sentinel stations were sampled separately from the benthic infauna stations and there was no benthic station at the sentinel station in Little Pleasant Bay. The closest benthic station in the 2020 survey is LPB43. Sentinel stations are defined as those stations that once improved, would indicate similar improvement among the other sub-embayments within a system. According to the nitrogen loading model the sentinel station is an indicator for the status of the whole basin. In other words if the benthic habitat at the sentinel station is recovered (i.e. with eelgrass beds re-established), then the rest of the basin would also be recovered. However, Howes et al. (2006) warned that due to the size of the Pleasant Bay System and the relatively isolated nature of some of the small sub-embayments, it may be possible that a specific sub-embayment may or may not achieve its secondary threshold, even though the eelgrass threshold is reached at the sentinel stations.

As mentioned above, the criteria used to determine the infaunal indicators are not comparable to the US M-AMBI scores in this report, however, as there was no eelgrass observed in any of the sentinel (or near-sentinel) stations, these stations would not be categorized as "recovered" by Howes et al. (2006) standards. Using the US M-AMBI's benthic community health criteria at the sentinel stations may provide a way to document changes in the benthic health condition at these locations in addition to the use of eelgrass. The habitat health at Station RC declined, and was ranked as Poor in 2020. The habitat at Station LPB43 was ranked as Good, and the habitat at Station BH was Good/High suggesting a clear improvement. These US M-AMBI classifications can be used as the baseline for comparisons to future surveys at the sentinel stations.

Table 9. Pleasant Bay benthic health indicators in 2003 and 2020.

		2003 Assessment (Howes et al. 2006)[1]					2020 Assessment				
Sub-Embayment	Station	Total Number of Species	Total Number of Individuals	Shannon-Weiner Diversity (H')	Evenness (E)	Infaunal Indicator[2]	Total Number of Species	Total Number of Individuals /m²	US M-AMBI Score	Shannon-Weiner Diversity (H')	Pielou's evenness (J')
Meetinghouse Pond	MP47	6	672	1.60	0.62	SI	12	2,875	Moderate	1.42	0.57
	MP48	6	752	1.90	0.73	SI	5	900	Poor	0.85	0.53
	MP49	7	800	1.65	0.59	SI	9	138	Moderate	2.10	0.96
Lonnies Pond	LP53	9	897	1.73	0.54	SI/MI	16	3,688	Poor/Mod.	1.15	0.41
Areys Pond	AP 22/23	4	93	1.58	0.79	SD/SI	9	850	Moderate	1.36	0.62
The River	TR26	9	1,561	1.53	0.49	MI	37	35,663	Good	1.61	0.44
	TR45	4	105	1.28	0.74	MI	36	133,638	Good	1.17	0.33
	TR50	9	834	2.27	0.74	MI	32	51,513	Good	1.97	0.57
	TR52	9	2,448	2.43	0.78	MI	26	14,563	Poor/Mod.	1.36	0.42
Paw Wah Pond	PWP46	2	3	0.41	0.41	SD	38	34,200	Good	2.10	0.58
Quonset Pond	QP3	2	48	0.50	0.5	SI	22	19,600	Good	1.40	0.45
Round Cove	RCV1	5	397	1.21	0.52	SI/MI	20	58,675	Good	1.91	0.64
	RCV2	8	227	1.52	0.51	SI/MI	28	120,588	Good	1.69	0.51
	RCV3	5	296	1.21	0.52	SI/MI	24	37,300	Good	1.90	0.60
Muddy Creek	MCM	7	139	1.69	0.60	SI/SD	31	24,488	Good	1.80	0.52
Bassing Harbor	BH	16	633	3.06	0.77	H/MI	40	17,975	Good/High	2.42	0.66
	RC	18	136	1.81	0.43	MI	16	3,338	Poor	1.50	0.54
	CPW	30	287	3.76	0.77	MI	46	68,775	Good/High	2.13	0.56
	CPE	29	374	3.63	0.74	MI	50	56,450	Good	1.93	0.49
	FFC	5	125	1.53	0.66	SI	22	15,238	Good	1.73	0.56
Pochet	P39	9	480	2.98	0.94	H	39	47,838	Good	2.28	0.62
	P41	12	600	2.78	0.72	H	49	77,725	Good/High	1.47	0.38

Table 9. Continued.

		2003 Assessment (Howes et al. 2006)[1]					2020 Assessment				
Sub-Embayment	Station	Total Number of Species	Total Number of Individuals	Shannon -Weiner Diversity (H')	Evenness (E)	Infaunal Indicator[2]	Total Number of Species	Total Number of Individuals /m²	US M-AMBI Score	Shannon -Weiner Diversity (H')	Pielou's evenness (J')
	LPB37	10	944	2.26	0.68	MI	38	147,588	Good	0.50	0.14
	LPB43	9	724	2.40	0.81	MI	35	32,250	Good	1.53	0.43
Pleasant Bay	PB6	8	2,640	1.91	0.63	MI	31	34,063	Mod./Good	1.76	0.51
	PB14	6	116	2.00	0.77	MI	25	7,563	Good	1.69	0.52
	PB16	5	72	2.21	0.97	MI	10	7,925	Moderate	1.23	0.54
	PB20	4	208	1.35	0.68	MI	25	5,100	Good	2.34	0.73
	PB32	4	196	1.30	0.81	MI	29	13,7813	Poor/Mod.	0.86	0.25
Chatham Harbor	CH12	7	224	2.1	0.75	H	10	3,200	Poor/Mod.	1.24	0.54

Estimates of the number of species adjusted to the number of individuals and diversity (H') and Evenness (E) of the community allow comparison between locations. Samples represent surface area of 0.0625 m² and values are averages of grab samples a-c).

Infaunal Indicators: H = Healthy habitat conditions; MI = Moderate Impairment; SI = Significant Impairment; SD = Severe Degradation

[1]2003 samples represent surface area of 0.0625 m² and thus cannot be directly compared to the 2020 samples, which are standardized to number per square meter.

Friends of Pleasant Bay Ecosystem Assessment – Pleasant Bay, 2014-2017

The Friends of Pleasant Bay (FOPB) funded the Center for Coastal Studies (CCS) to conduct an ecosystem assessment of Pleasant Bay between 2014 and 2017. The goal of this research was to develop a dataset and baseline assessment of the present status of the natural resources of Pleasant Bay that could be used to develop a long-term habitat monitoring program (Hughes and Mittermayr 2018). A portion of this study focused on determining whether sediment type could be correlated to various benthic assemblages. Between June 24 and August 1, 2014, 48 stations within Pleasant Bay were sampled resulting in a total of 144 benthic infaunal samples (three replicates per station, 1 mm mesh size) and 48 sediment samples, along with water column profiles and photographic and video data for each station. Of these 48 stations, 15 stations overlapped with the Howes et al. (2006) MEP survey, however, these stations were not included in the report's statistical analysis as the data were not available at the time of writing (Hughes and Mittermayr 2018).

Overall, the study found 148 macroinvertebrate taxa but only 32 taxa comprised the top 95% of all individuals in the benthic communities (Hughes and Mittermayr 2018). A total of 67,167 individuals were recorded. The most abundant taxa in Pleasant Bay were the Amethyst Gem Clam (*G. gemma,* 18,659 individuals), and the gammarid amphipod *Ampelisca* sp. (16,658 individuals; Hughes and Mittermayr 2018). Hughes and Mittermayr (2018) examined the relationship between bottom sediment types and biological communities. The analyses indicated that Pleasant Bay was a complex system with a diversity of habitat types and organisms associated with them, including mud, sandy sediments, and eelgrass beds. They found that eelgrass in Pleasant Bay was the most productive habitat, with more than two times the number of individuals and almost twice the number of species as sandy bottom habitats. In non-eelgrass habitats three significant indicator taxa were identified, *Ampelisca* sp., *Tellina agilis* and Capitellidae. These taxa were also the most dominant in several of the benthic community cluster analysis groups suggesting that they play an important role in the overall composition of Pleasant Bay benthic communities. Hughes and Mittermayr (2018) suggested that sediment type alone could not fully explain the variability in the benthic infaunal species and that other biological and physical factors may be structuring benthic communities in Pleasant Bay. Possible examples they included were biotic interactions such as competition and predation, dominant benthic vegetation type, and water quality. The authors noted that water quality has been shown to influence benthic habitat quality in Pleasant Bay and factors such as dissolved oxygen likely play an important role in the composition of system's benthic communities.

The results from the MEP Pleasant Bay 2020 survey are very similar to those found by Hughes and Mittermayr (2018). A total of 156 taxa were identified and 24 taxa comprised 95% of all individuals. A total of 101,868 individuals were recorded and the most abundant were G. gemma (41,745 individuals) and Ampelisca sp. (25,076 individuals). Sediment grain size recorded in 2020 was consistent with the median grain size reported by Hughes and Mittermayr (2018; Figure 7). The multivariate analyses for the 2020 study identified 4 main benthic assemblages with Group III (Main Bay area) containing several sub-groups. Similar to Hughes and Mittermayr (2018), sediment grain size explained some of the differences in the community assemblages observed, for example Group I (Inner Bay) and Group IV (Outer Bay). Dissolved oxygen (DO) also explained some of the variability as the lowest bottom DO levels were observed in Meetinghouse Pond (5.2 to 6.5, Appendix A) and Lonnies Pond (6.4) which contain the Group I and Group II assemblages, while some of the highest DO values (>9.0; Appendix A) were observed at Stations 12 and 16, the Group IV assemblage. However, sediment grain and DO could not explain all of the variability as illustrated by the largest group, Group III, that contains a range of grain size and DO values within the sub-groups. The 2020 Pleasant Bay benthic assessment supports Hughes and Mittermayr (2018) finding that other biological and physical factors such as biotic interactions may be structuring benthic communities in Pleasant Bay.

4 Recommendations

The MEP Marine Benthic Monitoring Pilot Field Study is being conducted to test the approaches and procedures described in the new draft guidance documents. Overall, the Pilot Field Study conducted in both West Falmouth Harbor (2019) and Pleasant Bay demonstrated that the new draft guidance documents will successfully provide guidance to parties outside of MassDEP and produce quality benthic data that 1) assess current embayment health, 2) are comparable between assessments, and 3) will aid in future management decisions. The Pleasant Bay survey also demonstrated that US M-AMBI can be successfully used in large embayments with multiple and complex sub-systems. The results from this index will need to be interpreted in conjunction with other data collected during an assessment to determine if areas characterized as Poor or Bad are the result of anthropogenic or natural factors. Recommendations for future sampling in Pleasant Bay are presented below, and minor recommended revisions to the draft guidance documents are presented in Appendix C.

Recommendations for future sampling in Pleasant Bay include:

1. Sampling locations for several stations (e.g. Frost Fish Creek or Muddy Creek) should be reviewed and relocated to coordinates sampled during the 2020 survey. The locations identified for the 2020 Pilot Field Study were based on the benthic infaunal sampling stations established by Howes et al. (2006). The sampling locations in Frost Fish Creek and Muddy Creek are not currently accessible by boat. These areas may be accessible with a canoe or onshore access points, however these alternative access methods will prevent the use of a 0.04-m^2 Ted Young-modified Van Veen grab. If benthic samples are desired from the original locations in these areas, an alternative benthic sampler should also be considered. The sampling location at Station 41 (Pochet) should also be reconsidered as this area now contains a shellfish growing area. Conducting benthic monitoring in an active shellfish growing area will not provide data that would accurately represent the species or benthic health condition in adjacent areas.
2. Several sampling locations were adjusted due to the presence of mooring fields with high boat densities and a concern for vessel and crew safety. It is recommend for future Pleasant Bay assessments that embayments with mooring fields be included in the access review and a protocol established for moving stations at these locations. The protocol could include pre-established alternative sampling locations in the sub-embayment or basin.
3. Lastly, program and assessment objectives change over time, the sampling locations in Pleasant Bay should be re-evaluated to ensure that they continue to meet program study design and objectives.

5 Summary

Pleasant Bay is a large, ecologically complex system with many biological and physical factors influencing the benthic community assemblages and health status. The benthic habitats ranged from Poor to High, with the majority of stations sampled categorized as Good (61%). Most of the stations categorized as Poor were in the Inner Bay, generally areas with less tidal flushing and silty sediments (The River [TR52], Lonnies Pond [LP53], Meetinghouse Pond [MP48], and Ryder's Cove [RC]). Two other stations categorized as Poor were Station 32 in Pleasant Bay (PB32) and Station 12 in Chatham Harbor (CH12) which were exposed to a high tidal velocity, causing a relatively low quality habitat. Two of the three stations categorized as High were in the Bassing Harbor sub-system (south of the Main Bay), Bassing Harbor (BH) and Crow's Pond West (CPW). The third station categorized as having a High benthic health status was located in Pochet in the east portion of the Main Bay, Station 41 (P41).

Direct comparison between the 2003 MEP benthic infauna assessment (Howes et al. 2006) and the current benthic assessment was not possible due to a difference in sampling methods. The 2003 benthic community results were presented as number per 0.0625 m^2 while the 2020 assessment densities were standardized to number per 1.0 m^2. It is not clear if the 2003 sample area was a subsample or a different grab type, and neither the data nor methods were available to examine. In addition, the 2003 Infaunal Indicators were based on different parameters than US M-AMBI and thus could not be directly compared. Despite the differences between the 2003 and 2020 surveys, a general comparison was made that indicated that three stations showed habitat improvement (Stations 3 and 46, and Little Pleasant Bay) and three stations or basins showed a decline in habitat health (Stations 12 and 48, and Ryders Cove). Using the US M-AMBI's benthic community health criteria at the sentinel stations may provide a way to document changes in the benthic health condition at these locations that can be used for comparisons to future surveys at the sentinel stations.

The comparison of the MEP Pleasant Bay 2020 survey to Hughes and Mittermayr (2018) indicated very similar results between the two studies. The number of taxa were similar and the two most abundant taxa in both studies were *G. gemma* and *Ampelisca* sp. The multivariate analyses conducted by both studies indicated that sediment grain could not explain all of the variability of the benthic infaunal assemblages and that other biological and physical factors including tidal flushing, water quality, and biotic interactions may be structuring the benthic communities in Pleasant Bay.

Overall, the Pilot Field Study conducted in Pleasant Bay demonstrated that the new draft MEP Marine Benthic Monitoring guidance documents will successfully provide guidance to parties outside of MassDEP and produce quality benthic data that 1) assess current embayment health, 2) are comparable between US M-AMBI assessments, and 3) will assist in future management decisions.

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Appendix A. Water Quality Measurements in Pleasant Bay, September 2020.

Sub-embayment	Station	Depth (m)	Temp (°C)	DO (mg/L)	pH	Salinity (ppt)
Little Pleasant Bay	LPB	0.12	15.40	8.71	8.12	30.59
		0.54	15.36	8.83	8.11	30.60
		1.07	15.32	8.90	8.11	30.68
		1.24	15.31	8.92	8.11	30.73
	35	0.01	15.93	8.46	8.13	29.62
		0.16	15.92	8.61	8.13	29.74
	36	0.03	19.33	7.73	8.10	30.99
		0.08	19.33	7.83	8.10	31.04
	37	0.03	15.38	8.62	8.12	29.39
		0.03	15.37	8.63	8.12	29.40
		0.07	15.37	8.68	8.12	29.46
	43	0.14	13.38	8.52	8.01	28.69
		0.44	13.43	8.63	8.00	28.47
Pleasant Bay	6	0.09	15.06	8.49	8.12	30.76
		0.53	14.88	8.53	8.11	30.76
		1.01	14.81	8.53	8.10	30.82
		2.00	14.70	8.53	8.10	30.80
		3.02	14.21	8.65	8.13	30.69
		4.01	14.14	8.70	8.12	30.67
	14	0.02	15.40	8.40	8.12	28.32
		0.06	15.50	8.55	8.11	28.54
	16	0.13	16.30	9.07	8.15	32.10
		0.52	16.30	9.15	8.14	32.16
		1.07	16.31	9.20	8.14	32.18
		1.94	16.29	9.22	8.14	32.20
	20	0.20	17.18	8.77	8.19	30.82
		0.55	17.15	8.83	8.18	30.85
		1.00	17.09	8.88	8.18	30.86
		1.83	16.69	8.91	8.17	30.84
	32	0.10	16.92	10.00	8.19	32.54
		0.53	16.92	10.05	8.19	32.56
		0.95	16.92	10.08	8.19	32.58
		1.17	16.92	10.09	8.19	32.60
Quonset Pond	3	0.20	18.77	9.30	8.25	30.66
		0.58	18.78	9.34	8.24	30.71
		1.04	18.77	9.40	8.24	30.77
		1.99	17.98	8.94	8.18	30.71
		2.52	17.73	8.54	8.15	30.69

Appendix A. (continued)

Sub-embayment	Station	Depth (m)	Temp (°C)	DO (mg/L)	pH	Salinity (ppt)
Meetinghouse Pond	47	0.16	19.37	6.55	7.79	29.33
		0.51	19.39	6.52	7.78	29.35
		1.03	19.39	6.22	7.76	29.41
	48	0.18	19.37	6.47	7.80	29.66
		0.60	19.37	6.44	7.79	29.68
		1.12	19.37	6.42	7.79	29.69
		2.02	19.38	6.28	7.77	29.74
		2.83	19.42	5.18	7.69	29.94
	49	0.27	19.32	6.90	7.82	29.71
		0.53	19.33	6.82	7.82	29.72
		1.14	19.33	6.81	7.82	29.73
		2.09	19.34	6.78	7.82	29.75
		3.07	19.34	6.75	7.81	29.76
		4.04	19.34	6.72	7.81	29.77
		5.03	19.36	6.34	7.78	29.88
		5.97	19.36	5.96	7.76	30.10
		6.98	19.35	5.78	7.75	30.14
		7.16	19.35	5.67	7.75	30.16
The River	26	0.12	15.53	10.57	8.19	29.58
		0.44	15.10	10.53	8.19	30.05
	45	0.23	13.81	8.46	8.00	27.55
		0.52	13.72	8.44	7.99	27.54
		1.05	13.68	8.44	7.99	27.57
		1.98	13.46	8.42	8.00	27.62
		2.37	13.43	8.38	8.00	27.64
	50	0.29	18.86	6.59	7.83	30.47
		0.56	18.85	6.52	7.83	30.50
		1.07	18.82	6.48	7.83	30.54
		1.96	18.80	6.48	7.84	30.59
		2.17	18.78	6.46	7.84	30.63
	52	0.19	18.58	6.79	7.88	30.81
		0.52	18.58	6.75	7.88	30.83
		1.02	18.56	6.71	7.88	30.88
		1.71	18.31	6.69	7.89	31.08
Pochet	39	0.03	14.41	9.07	7.87	29.34
	41	0.13	14.09	8.77	7.97	29.67
Lonnies Pond	53	0.15	18.77	7.23	7.88	29.82
		0.48	18.81	7.13	7.86	29.94
		1.05	18.98	6.77	7.83	30.23
		1.96	19.12	6.32	7.80	30.46
		2.98	19.05	6.26	7.81	30.48
		3.86	18.93	6.37	7.82	30.48
Areys Pond	22/23	0.10	16.08	9.72	8.04	27.89
		0.52	15.87	9.82	8.04	28.08
		1.05	14.79	9.97	8.06	28.36
		2.01	14.41	8.14	7.89	28.77
		2.89	14.37	7.48	7.84	28.98
Paw Wah	46	0.07	16.24	9.42	8.16	30.91
		0.54	16.17	9.44	8.14	30.90
		0.83	15.30	8.72	8.08	30.56

Appendix A. (continued)

Sub-embayment	Station	Depth (m)	Temp (°C)	DO (mg/L)	pH	Salinity (ppt)
Round Cove	RCV1	0.28	18.25	8.39	8.12	29.67
		0.54	18.05	8.43	8.13	29.82
		1.01	17.89	8.43	8.12	30.05
		1.33	17.69	8.27	8.10	30.22
	RCV2	0.18	18.47	8.49	8.10	28.51
		0.55	18.49	8.51	8.08	28.65
		1.11	17.63	8.45	8.10	29.45
		2.09	17.59	8.43	8.12	29.56
		2.24	17.57	8.39	8.11	29.58
	RCV3	0.33	18.65	8.56	8.09	28.45
		0.54	18.61	8.64	8.08	28.56
		1.01	18.19	8.44	8.10	29.15
		1.99	17.63	8.38	8.10	29.86
		2.27	17.60	8.27	8.10	29.91
Bassing Harbor	Bassing Harbor	0.18	18.53	8.74	8.19	31.23
		0.58	18.41	8.78	8.18	31.26
		1.00	18.37	8.82	8.18	31.29
	Crows Pond East	0.13	15.78	8.83	8.17	31.38
		0.52	15.39	8.91	8.17	31.40
		1.01	15.30	8.98	8.17	31.41
		1.98	14.75	9.06	8.16	31.24
		2.99	14.72	9.15	8.17	31.21
		3.74	14.55	9.13	8.15	31.23
	Crows Pond West	0.11	15.71	9.10	8.20	31.63
		0.53	15.49	9.16	8.18	31.59
		1.01	15.08	9.24	8.18	31.51
		2.00	14.68	9.26	8.17	31.44
		2.98	14.60	9.24	8.17	31.40
		3.34	14.49	9.20	8.16	31.36
	Frost Fish Creek	0.02	19.21	8.14	8.09	31.09
		0.10	19.20	8.25	8.09	31.13
	Muddy Creek Mouth	0.03	17.69	8.37	8.12	30.69
		0.17	17.59	8.46	8.12	30.75
		0.23	17.42	8.39	8.11	30.75
	Ryders Cove	0.03	20.26	8.14	8.08	31.21
		0.16	20.40	8.27	8.08	31.21
		0.32	20.30	8.27	8.07	31.26
		0.65	20.43	8.28	8.08	31.28
		0.77	20.13	8.32	8.08	31.15
Chatham Harbor	12	0.03	14.89	8.83	8.13	29.60
		0.53	14.88	8.98	8.13	29.69
		0.91	14.88	9.02	8.12	29.73

Appendix B. Images of Soft Benthic Habitat from Pleasant Bay 2020 Survey.

a.

b.

Figure B-1. Images of soft bottom habitat in Meetinghouse Pond taken on September 18, 2020 at stations: a) 47 and b) 48.

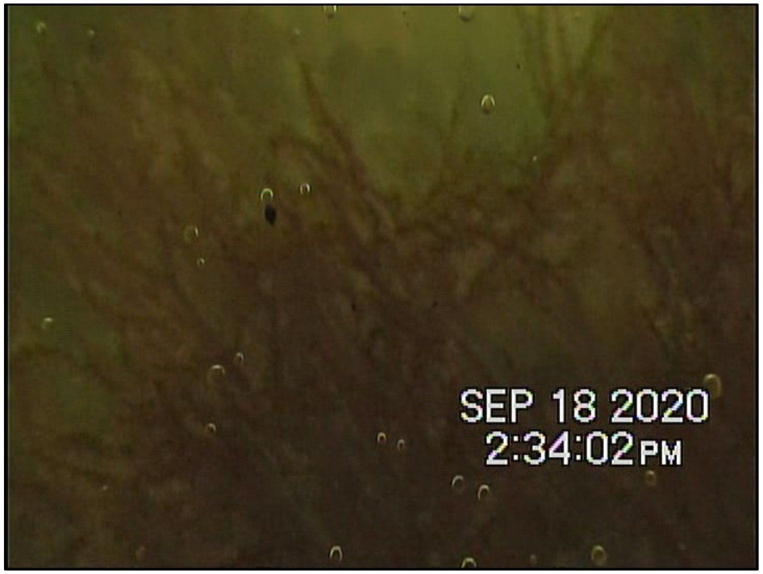
a.


b.


c.

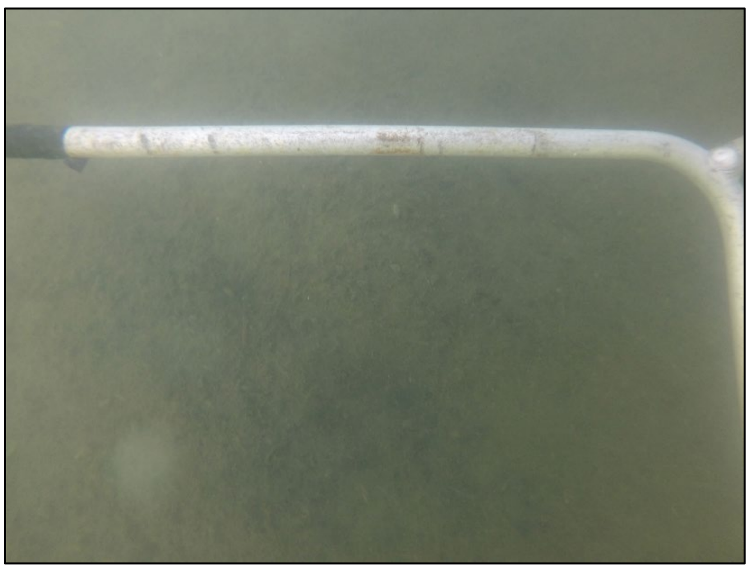

d.


Figure B-2. Images of soft bottom habitat in the Rivers taken on September 18 and 23, 2020 at stations: a) 50, b) 52, c) 45, and d) 26.



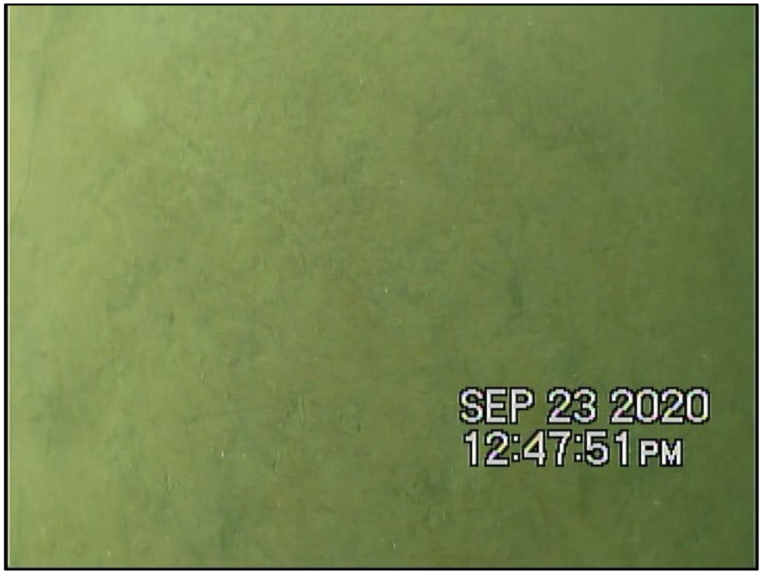
a.

b.

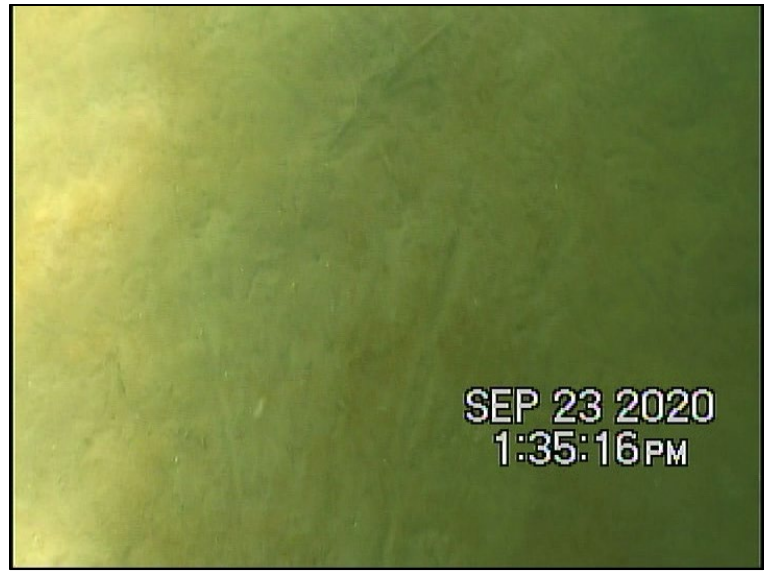


c.

Figure B-3. Images of soft bottom habitat in Lonnies Pond (a. Station 53), Areys Pond (b. Station 22/23), and Paw Wah Pond (c. Station 46, GoPro camera) taken on September 18 and 23, 2020.



a.



b.

c.

Figure B-4. Images of soft bottom habitat in Pochet taken on September 23, 2020 at stations a) 39 and b) 41, and Quanset Pond (c) taken on September 17, 2020.

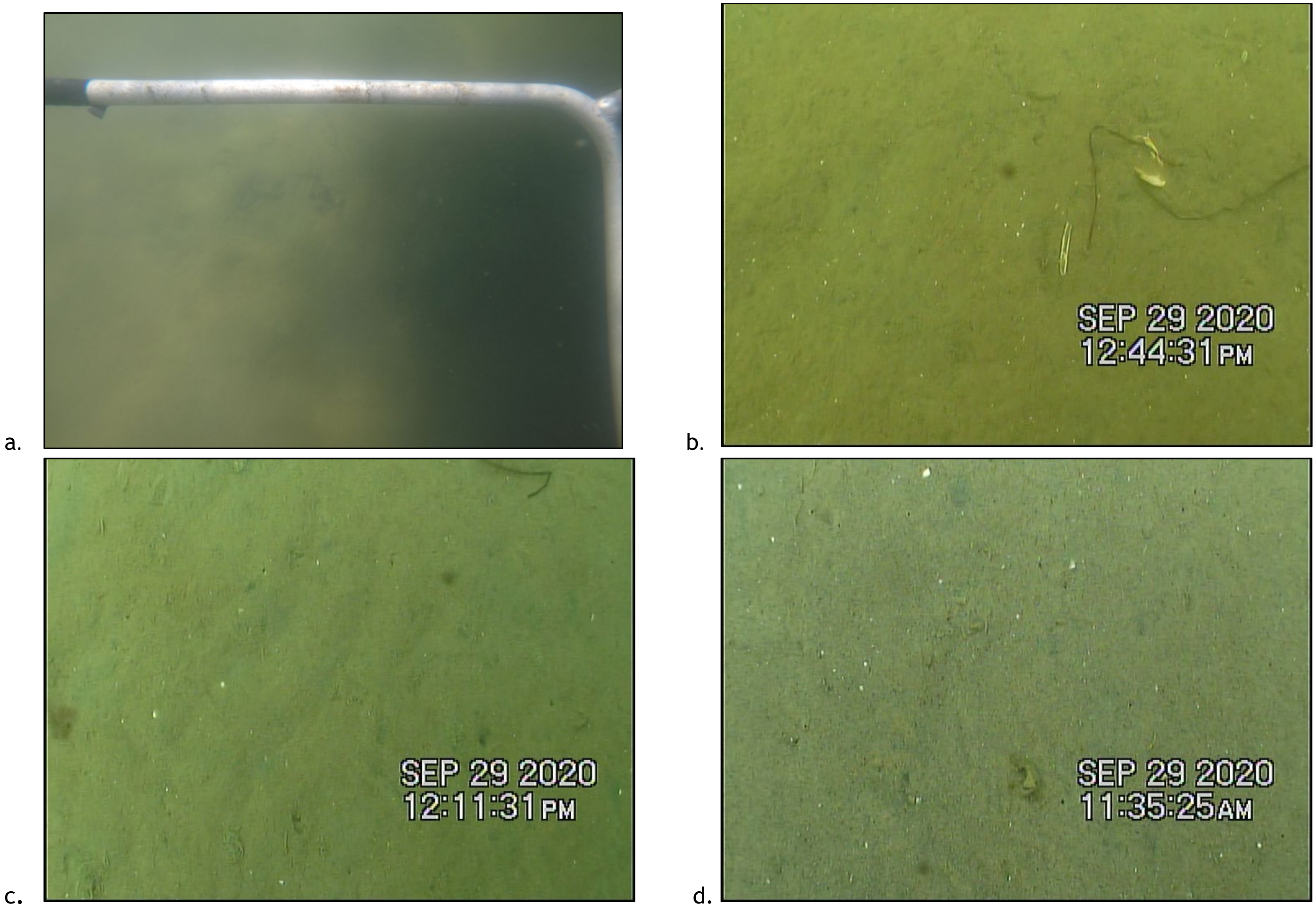
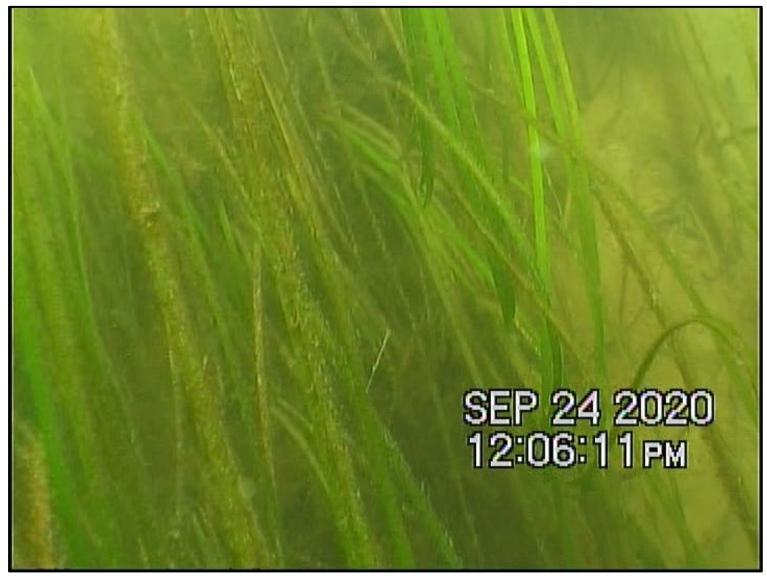


Figure B-5. Images of soft bottom habitat recorded on September 23 and 29, 2020 in Little Pleasant Bay at stations: a) 43 (GoPro camera), b) 36, c) 35, and d) 37. Eelgrass was observed at Station LPB (images e through h) on September 24, 2020.

e.

f.

g.



d.

Figure B-5. Continued.

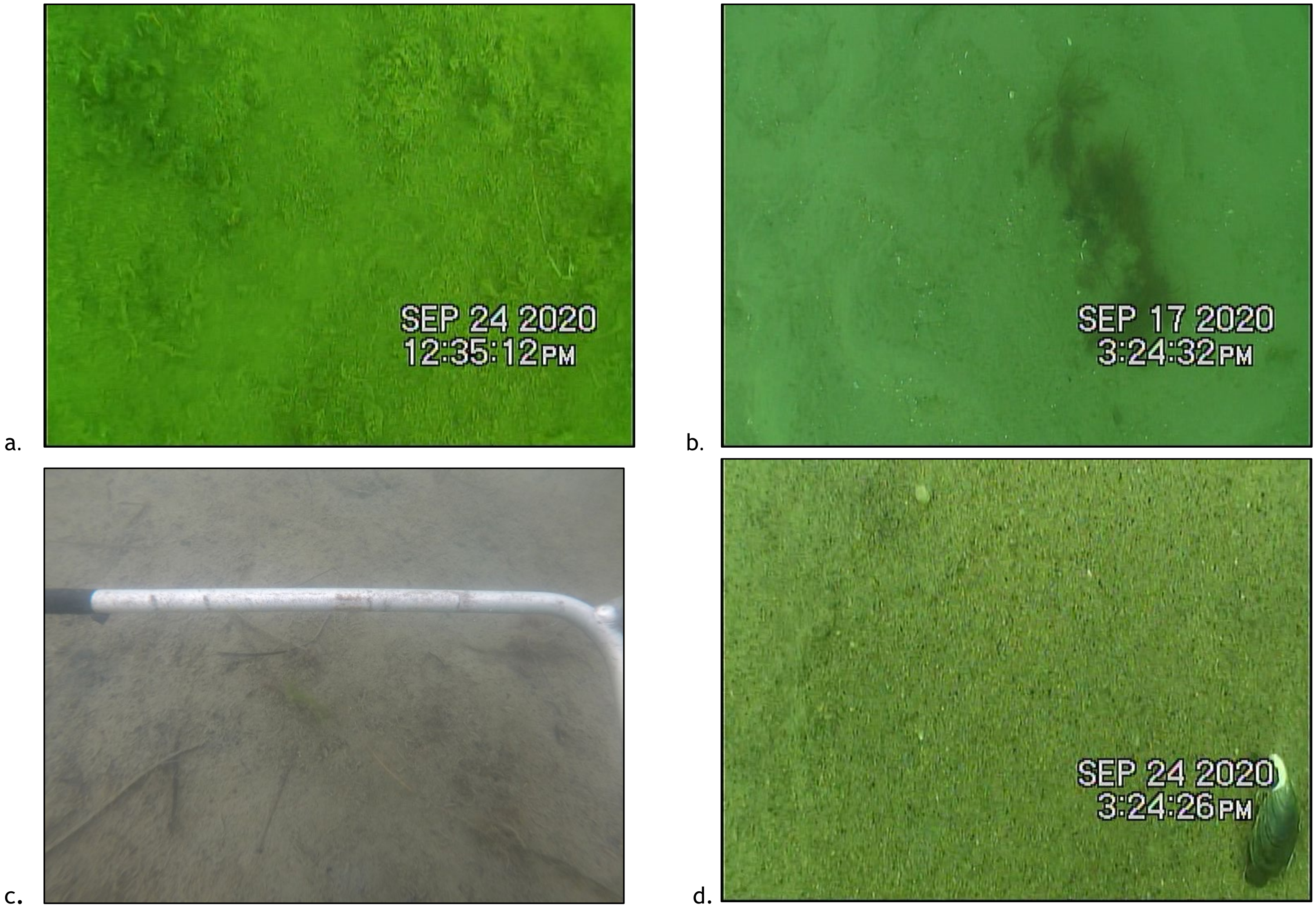


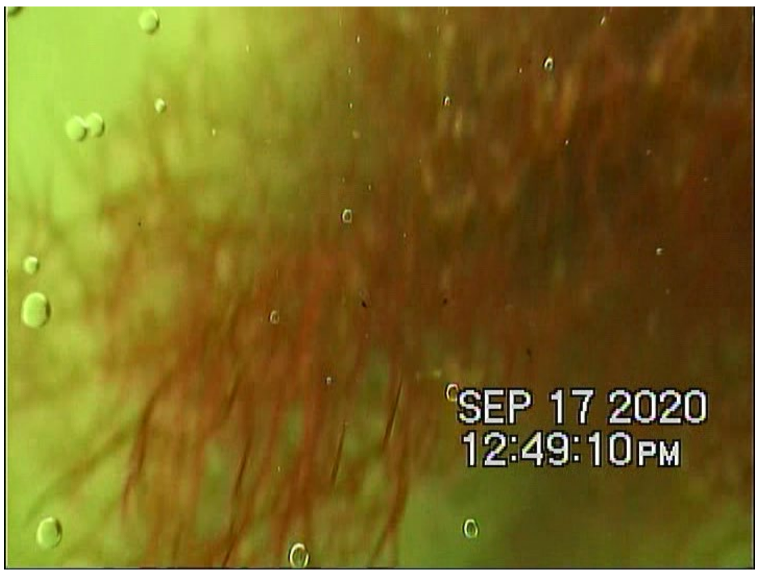
Figure B-6. Images of soft bottom habitat recorded on September 17, 24 and 29, 2020 in Pleasant Bay at stations: a) 6, b) 20, c) 32 (GoPro camera), d) 16, and e) 14.

e. 

Figure B-6. Continued.

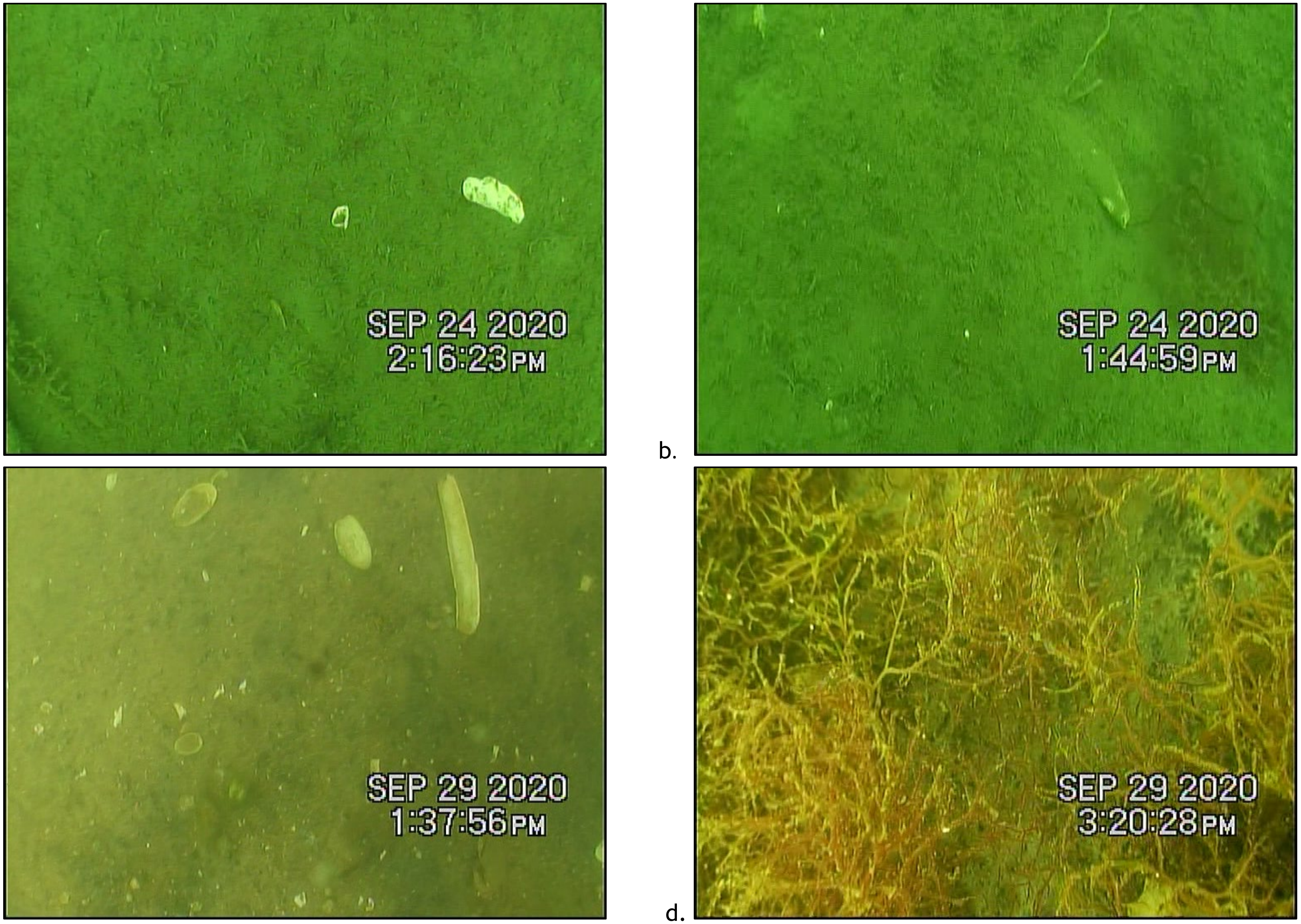
a.

b.



c.

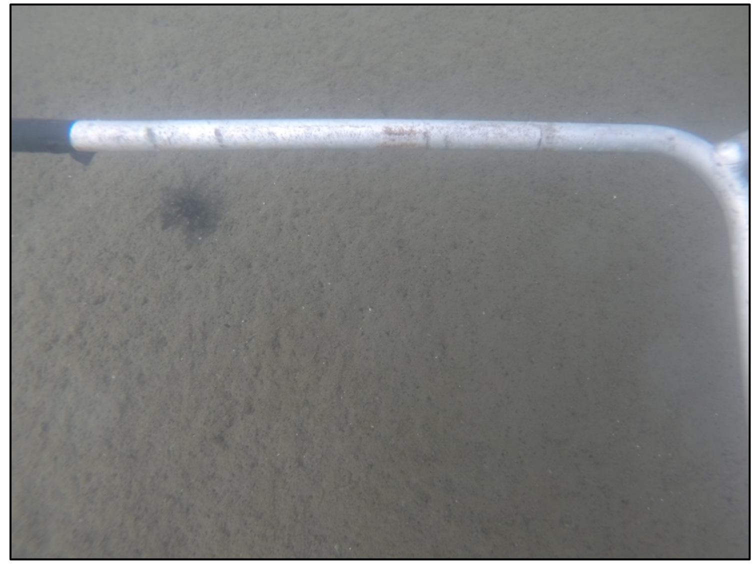
Figure B-7. Images of soft bottom habitat in Round Cove taken on September 17, 2020 at stations a) RCV1, b) RCV2, and c) RCV3.



a. b. c. d.

Figure B-8. Images of soft bottom habitat in Crows Pond (stations Crows Pond West [a] and East [b]), Muddy Creek (c), and Ryders Cove (d) taken on September 24 and 29, 2020.

a.


b.


c.


Figure B-9. Images of soft bottom habitat in Bassing Harbor (a), Frost Fish River Creek (b – GoPro camera), and Chatham Harbor (c – Station 12) taken on September 17, 24, and 29, 2020.

Appendix C. Recommended revisions to the draft guidance documents.

The MEP Marine Benthic Monitoring Pilot Field Study is being conducted to test the approaches and procedures described in the new draft guidance documents. Overall, the Pilot Field Study conducted in Pleasant Bay demonstrated that the new draft guidance documents will successfully provide guidance to parties outside of MassDEP and produce quality benthic data that 1) assess current embayment health, 2) are comparable between assessments, and 3) will assist in future management decisions. Below are several recommended revisions to the draft guidance documents to provide greater direction to the laboratory technicians, clarify procedures, and improve data quality.

The following revisions to laboratory analysis and data management procedures are recommended for the draft MEP Marine Benthic Monitoring QAPP and Laboratory SOP:

1. The draft QAPP and Laboratory SOP should be revised to include a protocol for subsampling benthic infaunal samples that contain an extremely high number of individuals that cannot be easy separated from the sediment matrix (e.g. juvenile bivalves approximately 0.5 mm in size). The protocol should contain a specific threshold value for when the procedure should be implemented, the percentage of the sample to processed in the subsample, guidance for random selection of subsample, and guidance on data handling and documentation.
2. The draft QAPP and Laboratory SOP should be revised to change the species identifier from Taxonomic Serial Number (TSN) to AlphiaID. This revision is recommended based on the latest National Coastal Condition Assessment (NCCA) taxonomic species list and the observation that currently the Integrated Taxonomic Information System (ITIS) is not being consistently updated for recent changes to the taxonomic nomenclature. AlphiaID is available from the World Register of Marine species (WoRMS) which is routinely maintained and continuously updated with the latest changes in the taxonomic nomenclature found in scientific literature.
3. The use of Average Taxonomic Distinctness (ATD, delta +), Total Taxonomic Distinctness (TTD), or another taxonomic distantness measure should be reviewed and the draft QAPP and Laboratory SOP documents updated to reflect the final selection. ATD is the average 'distance apart' of any two species or individuals chosen at random from the sample. TTD is the average taxonomic distance from species *i* to every other species, summed over all species in the sample. ATD was calculated for the 2019 West Falmouth Harbor study after communications with PRIMER-e which indicated that it was the preferred measure. The use of ATD was continued in the 2020 Pleasant Bay study. The two measures are similar but provide slightly different information and have different advantages and limitations. These differences should be re-evaluated along with the prevalence of measure use by other researchers. Additionally, advances in statistic software may provide other or new taxonomic distantness measures which should also be considered.
4. The data codes for STAT_ARRIV_LOCAL in Tables 22, 23, 30, and 32, and SAMPLE_DATE_TIME_LOCAL in Table 23 of the draft QAPP and the corresponding tables in the draft Laboratory SOP should be revised to change the format from MMDDYY:HH:MM to MM/DD/YY HH:MM.

5. The data codes for DATE_COLLECTED in Tables 25, 28, 29, and 31, and DATE_RECEIVED/DATE_COLLECTED/DATE_TAXON in Table 26, DATA_ANALYSIS in Table 23 of the draft QAPP and the corresponding tables in the draft Laboratory SOP should be revised to change the format from MMDDYY:HH:MM to MM/DD/YY HH:MM.
6. The following data code revision are recommended for the draft QAPP and the corresponding tables in the draft Laboratory SOP:
    a. For Table 34a gear code for the multiparameter sonde (MPS) should be added.
    b. For Table 25 TEMP variable description should be updated to say: Water temperature measured in °C.
    c. For Table 25 a DEPTH_UNIT variable should be added (Format: Character; Description: Unit of depth measurement for DEPTH).
    d. For Tables 25, 28, and 29 SITE_ID should be renamed to STAT_ID for consistency with other data deliverable formats.
    e. For Table 28 the following variables should be added: LAB_NAME (Format: Character; Description: Name of lab) and DATE_RECEIVED (Format: MM/DD/YY; Description: Date sample was received by lab).
    f. Header to Table 29 should be revised from "Sediment grain size and TOC analysis data codes." to "TOC and other parameters including contaminants with single result values." This change will better reflect the type of samples that could be included in a study using this format.
    g. Header to Table 26 should be revised from "Benthic macrofaunal and sediment sample data codes." to "Benthic macrofaunal and destructive sample data codes."
    h. In Table 26 'SEDIMENT' should be remove from list of SAMPLE_TYPE variable.
    i. Further revisions should be made to the Sediment Grain Size Result Data Code Table recommended in the West Falmouth Harbor Benthic Report to be consistent with Table 29. These revision including adding the following variables (variable Formats and Descriptions should follow Table 29): DATE_COLLECTED, ANALYSIS_TYPE, CONDITION_CODE, COND_COMMENTS, PARAMETER, METHOD, MDL, LRL, REASON, RESULT_QUAL, UNIT, QC_CODE, and COMMENT.