



# **Technical Report**

# FINAL

# Shellfish Aquaculture Demonstration Project Little Pond Monitoring 2013-2014 Oyster Deployment

To:

Jerry Potamis and Town of Falmouth Water Quality Management Committee Chair, Eric Turkington

From:

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### **Section I. Introduction**

Background: The Little Pond embayment system is located within the Town of Falmouth, on Cape Cod Massachusetts. The system has a southern shore surrounded by water from Vineyard Sound (Figure I-1). The watershed for this salt pond system is distributed fully within the Town of Falmouth. The present shape of the Little Pond embayment results from the drowning by rising sea level of valleys formed primarily via post-glacial erosion by groundwater fed rivers and streams. At present, Little Pond is a tidal embayment with a groundwater fed stream discharging to its headwaters. Almost all of the salt ponds on this stretch of the southern coast of Cape Cod mirror this configuration. As is typical with other Falmouth embayments (Great, Green, and Bournes Pond) Little Pond is separated from Vineyard Sound by a barrier beach, which was naturally breached and is now artificially maintained by jetties. The beach and the opening to the embayment are very dynamic geomorphic features due to the influence of littoral transport processes. Over the past century the Little Pond inlet has experienced varying degrees of occlusion thereby affecting tidal exchange and circulation within the salt pond. By example, Bournes Pond became very restricted and finally completely isolated from Vineyard Sound waters in the late 1970's/early 1980's and was re-opened with a fixed inlet in the mid 1980's. Currently, the inlet to Little Pond is periodically dredged to maintain the small tidal channel into the pond.

Similar to the Great, Green and Bournes Pond embayment systems, Little Pond is a mesotrophic (moderately nutrient impacted) to eutrophic (nutrient-rich) shallow coastal salt pond. The embayment is located within a glacial outwash plain, the Mashpee Pitted Plain, consisting of material deposited after the retreat of the Cape Cod Lobe of the Laurentide Ice sheet ~18,000 years ago. The outwash material is highly permeable and varies in composition from well sorted medium sands to course pebble sands and gravels extending down to about 17 m below mean sea level (Millham, 1993 and Millham and Howes, 1994). As such, direct rainwater run-off is typically rather low for these finger ponds and therefore, most freshwater inflow to these estuarine systems is via groundwater discharge or groundwater fed surface water flow (e.g. stream to the head of Little Pond, Coonamessett River to Great Pond, Backus River to Green Pond, etc.). Little Pond acts as a mixing zone for terrestrial freshwater inflow and saline tidal flow from Vineyard sound. However, the salinity characteristics of the salt pond varies with the volume of freshwater inflow as well as the effectiveness of tidal exchange with Vineyard Sound.

Along with the other salt pond embayments located on the south coast of Falmouth, Little Pond, constitutes an important component of the Town's natural and cultural resources. In addition, the large length to width ratio (8:1) greatly increases the potential for direct discharges from homes situated on the shore and decreases the travel time of groundwater from the watershed recharge areas to bay regions of discharge. The nature of enclosed embayments in populated regions brings two opposing elements to bare: as protected marine shoreline they are popular regions for boating, recreation, and land development; as enclosed bodies of water, they may not be readily flushed of the pollutants that they receive due to the proximity and density of development near and along their shores. In this context, Little Pond as well as other Falmouth embayments such as the Great, Green and Bournes Pond embayment systems along the Falmouth shoreline, are showing clear signs of eutrophication from high nitrogen loads in the groundwater and runoff from their watersheds.



Figure I-1. The Little Pond embayment system assessed by the Massachusetts Estuaries Project for system specific nitrogen threshold determination and to guide nutrient management in the associated watershed. Tidal waters enter the salt pond through one inlet to Vineyard Sound. Freshwaters enter from the watershed primarily through 1 surface water discharge (stream to the head of Little Pond) and direct groundwater discharge. *Nitrogen Loading:* Surface and groundwater flows are pathways for the transfer of landsourced nutrients to coastal waters. Fluxes of primary ecosystem structuring nutrients, nitrogen and phosphorus, differ significantly as a result of their hydrologic transport pathway (i.e. streams versus groundwater). In sandy glacial outwash aquifers, such as in the watershed to the Little Pond embayment system, phosphorus is highly retained during groundwater transport as a result of sorption to aquifer mineral (Weiskel and Howes 1992). Since even Cape Cod "rivers" are primarily groundwater fed, watersheds tend to release little phosphorus to coastal waters. In contrast, nitrogen, primarily as plant available nitrate, is readily transported through oxygenated groundwater systems on Cape Cod (DeSimone and Howes 1996, Weiskel and Howes 1992, Smith *et al.* 1991). The result is that terrestrial inputs to coastal waters tend to be higher in plant available nitrogen than phosphorus (relative to plant growth requirements). However, coastal estuaries tend to have algal growth limited by nitrogen availability, due to their flooding with low nitrogen coastal waters (Ryther and Dunstan 1971). Tidal reaches within Little Pond follow this general pattern, where the primary nutrient of eutrophication in these systems is nitrogen.

Nutrient related water quality decline represents one of the most serious threats to the ecological health of the nearshore coastal waters. Coastal embayments, because of their enclosed basins, shallow waters and large shoreline area, are generally the first indicators of nutrient pollution from terrestrial sources. By nature, these systems are highly productive environments, but nutrient over-enrichment of these systems worldwide is resulting in the loss of their biodiversity, aesthetic, economic and commercially valuable attributes.

Each embayment system maintains a capacity to assimilate watershed nitrogen inputs without degradation. However, as loading increases a point is reached at which the capacity (termed assimilative capacity) is exceeded and nutrient related water quality degradation occurs. As nearshore coastal salt ponds and embayments are the primary recipients of nutrients carried via surface and groundwater transport from terrestrial sources, it is clear that activities within the watershed, often miles from the water body itself, can have chronic and long lasting impacts on these fragile coastal environments.

Protection and restoration of coastal embayments from nitrogen overloading has resulted in the conduct of the Massachusetts Estuaries Project throughout southeastern Massachusetts and specifically in Falmouth, the focus of the project being on determining the nitrogen threshold (assimilative capacity) of coastal aquatic systems (e.g. Little Pond) for nitrogen. To establish the embayment specific threshold the MEP effort integrates site-specific data on nitrogen levels and the gradient in N concentration throughout the Little Pond system monitored by the Falmouth PondWatch Monitoring Program with site-specific habitat quality data (D.O., eelgrass, phytoplankton blooms, benthic animals).

As documented in the 2006 Massachusetts Estuaries Project (MEP) Nitrogen Threshold Analysis of Little Pond, the primary ecological threat to the natural resources of Little Pond is degradation resulting from nutrient enrichment. Loading of the critical nutrient, nitrogen, to the embayment waters has been greatly increased over the past few decades with further increases likely unless nitrogen management is implemented. The nitrogen loading to this and other Falmouth salt ponds, like almost all embayments in southeastern Massachusetts, results primarily from on-site disposal of wastewater. The Town of Falmouth has been among the fastest growing towns in the Commonwealth over the past two decades and does not have centralized wastewater treatment throughout the entire Town. At present Little Pond is beyond its ability to tolerate additional nitrogen inputs. It is presently showing habitat degradation consistent with nitrogen overloading. Although the Little Pond watershed is approaching buildout, nitrogen related degradation will likely increase slightly with further water quality degradation, unless nitrogen management is initiated and nitrogen loads. Fortunately, as Little Pond nitrogen loads are near their build-out rates, management options can be clearly defined based on the MEP nitrogen threshold established for Little Pond and options can be implemented with a high degree of certainty for restoration.

Project Need: As described above, estuarine water quality in towns throughout southern Massachusetts, inclusive of the Town of Falmouth, is impaired due to excessive nitrogen inputs from development within their watershed and needs to be improved in order to meet State Water Quality Standards. Towns are currently working on planning and implementing pilot studies to lower nitrogen levels within these impaired estuaries and are seeking lowest cost alternatives. The Massachusetts Estuaries Project (MEP) has explicitly and quantitatively documented nitrogen related habitat impairments to the main basins of Little Pond, Falmouth, MA, and developed a quantitative system specific modeling tool to assess the dynamics of the system as well as its response to nitrogen management alternatives. Results of the MEP analysis show that Little Pond is particularly sensitive to nitrogen inputs because of the comparatively small tidal range of Vineyard Sound and its restricted tidal inlet. Both tidal features result in a relatively low rate of tidal exchange (tidal flushing), which allows nitrogen and organic matter to accumulate in Pond waters, causing the eutrophication impairments noted above. As such, a broad range of management options should be considered external and internal to the function of the Little Pond system in order to ultimately meet the nitrogen threshold that would be supportive of healthy habitat. The Little Pond MEP report established a nitrogen threshold of 0.45 mg/L total nitrogen (TN) at a sentinel station (Pond Watchers station LP-2)<sup>1</sup>. The average (mean) TN concentration at LP-2 at the time of the MEP report (2006) was 0.898 mg/L. The threshold concentration was adapted by the Massachusetts Department of Environmental Protection into a total nitrogen load limit or Total Maximum Daily Load (TMDL) of 7 kg/day<sup>1</sup> and this TMDL was approved by USEPA.<sup>2</sup> MassDEP's TMDL for Little Pond does not stipulate how nitrogen levels are to be reduced, but reducing the watershed nitrogen sources (septic effluent, fertilizers, road runoff, etc) or increasing the rate of exchange with Vineyard Sound (enhanced flushing) are typically examined as alternatives. However, the Town of Falmouth has been investigating a series of new approaches for reducing nitrogen levels within its estuaries (innovated approaches to residential septic waste management, fertilizer bylaw, nitrogen intercepting PRBs).

Perhaps the most intriguing is the use of filter feeders as a form of in-estuary treatment (others are to lower watershed loads). The filter feeders generally considered are quahogs and oysters, as they grow well in the shallow warm estuaries of southeastern Massachusetts, and the mechanics of seeding and aquaculture are well established regionally. Oyster culture has been specifically deployed for lowering nitrogen levels in nearby Mashpee River, within the Popponesset Bay Estuary by both the Town of Mashpee and the Mashpee Wampanoag Tribe. SMAST staff has been supportive of these efforts from the beginning and have been working to quantify nitrogen removals for the past several years. Oyster culture within the Wellfleet Harbor Estuary has become well established, but only very recently is being looked at relative to nitrogen removal. The idea of using oysters for nitrogen reductions is not new, but probably the most well known analysis has resulted from the loss of oysters from the Chesapeake Bay. Nutrient and organic matter problems within Bay waters is now accepted to have resulted from

<sup>&</sup>lt;sup>1</sup> Massachusetts Department of Environmental Protection. February 7, 2008. FINAL Little Pond Embayment System Total Maximum Daily Loads For Total Nitrogen (Report # 96-TMDL-8 Control #246)

<sup>&</sup>lt;sup>2</sup> Approval of Little Pond System TMDL for Total Nitrogen (Report # MA96-TMDL-8, Control #246). March 3, 2008. Letter from Stephen S. Perkins, Director, Office of Ecosystem Protection, US Environmental Protection Agency, Region 1 to Laurie Burt, Commissioner, Massachusetts Department of Environmental Protection

increased inputs and the loss of this key filter feeder with the capability of filtering a high percentage of upper Bay water each day. Based upon studies of both natural systems and oyster aquaculture, there is no fundamental reason why oyster culture, if well established, could not have a significant impact on the nitrogen related water quality of Little Pond.

Less clear, however, is how the water quality benefits of oyster culture should be incorporated to TMDL compliance and development of Comprehensive Wastewater Management Plan (CWMP) development. The main policy/regulatory issue relative to these issues is to be able to quantify the nitrogen removal by an oyster deployment. It is easy to measure the nitrogen removed on harvest of the bivalves (meat and shells), but that is only a small portion of the N mass removed. On the order of 80%-90% of the particulate nitrogen removed from the water by an oyster, is deposited to the sediments as pseudofeces (not processed particulates). A conservative estimate would be that 50% of this is eventually denitrified (sent off as N<sub>2</sub> gas), effectively removing it from the estuary system. In an estuary with low tidal flushing like Little Pond, the nitrogen not denitrified may enter the ecosystem be taken up by phytoplankton (again), be filtered by other oysters with a portion being available for denitrification. In poorly flushed systems, this may occur several times with compounding nitrogen removals, that are many times the removal by ovster harvest alone. It is this combined effect that has been investigated and quantified for the Little Pond pilot project. Quantifying this number is important for planning purposes, but it is also fundamental to projecting removals to meet the TMDL. Simply put, the nitrogen removal per oyster deployment must be quantified in a manner that is scientifically defensible in order for MassDEP to match projected nitrogen removal by oysters to the quantitative requirements of the TMDL.

As agreed to by the Town of Falmouth and the SMAST-Coastal Systems Program (CSP) Science Team at the outset of the Shellfish Demonstration Project, the primary mission of the Town's Demonstration is to lay the foundation for establishing TMDL-nitrogen removal credit for oyster aquaculture. The Town selected the upper portion of Little Pond to examine its in-estuary nitrogen mitigation approach for improving water quality and habitat health. The upper estuary was selected for the following reasons:

- Nitrogen removal within the upper reaches of an estuary provides both local improvement, as well as lower nitrogen levels in down gradient basins. This distributes any beneficial effects over a larger area of estuarine habitat;
- Eelgrass restoration is primarily restricted to the lower main basin, placing culture gear in the lower basin would have to take into account shading of the bottom and other actions that would impede potential restoration of eelgrass habitat;
- Access through Narragansett Street is readily available.

It should be noted that the assessment of the effects of the moderate scale oyster deployment in Little Pond (2013) builds on foundational work undertaken by CSP-SMAST scientists over the past two decades, specifically for Little Pond:

 PondWatch – this program coordinated by CSP-SMAST staff since 1987 collects water quality data on Little Pond each summer in July and August on 4 dates. This data supported the basis for the Massachusetts Estuaries Project water quality modeling and nitrogen loading assessment. In addition, early PondWatch experiments on <u>oyster growth in Little Pond</u> were provided to the Town to assist its oyster pilot project planning. All data collected by CSP-SMAST for post-oyster deployment assessment will be directly comparable to the PondWatch results.

- Massachusetts Estuaries Project technical leadership by CSP-SMAST. The MEP conducted embayment specific habitat assessment, hydrodynamic and water quality modeling that serves as the underpinning of the Little Pond TMDL. The water quality and moored instrumentation approaches used by CSP-SMAST for the MEP analysis of Little Pond will be directly comparable to the CSP-SMAST post-oyster deployment assessment data.
- 3. Pre-oyster The CSP-SMAST scientific team on the present project also conducted detailed summer 2012 monitoring data collection from Little Pond at 9 sites (seven of which are in this RFP) and 12 dates (biweekly). This data forms the basis for the pre-oyster deployment baseline and also demonstrated the ability to collect samples within the necessary time-window to give a true-snapshot of the nitrogen related water quality gradient (TN, chlorophyll-a, particulate N, etc) along the long axis of Little Pond. This data was used by the Town for its permitting of the pilot project and the data collection in this proposal will be directly comparable to the CSP-SMAST pre-oyster deployment assessment data, both in sampling approach and laboratory assay.
- 4. Preparation for post-oyster assessment Prior to the finalization of water quality sampling plans, CSP-SMAST scientists collected 2012 water quality samples in May and June from Little Pond at each of the 9 stations and depths where sampling was conducted in the summer of 2013. These samples have been assayed, and yield directly comparable results to summer 2013 data. These samples were collected in concert with other programs conducted by SMAST throughout the upper Cape.

Specific characteristics of Little Pond, its function and its response to the initiation of oyster aquaculture in this system are further evaluated in the context of additional work undertaken by CSP scientists across the estuaries of southeastern Massachusetts, specifically:

- Scientific analysis of TN levels and eelgrass habitat quality, documenting TN reduction to restore habitat. A full assessment of the relationship of TN level to eelgrass habitat has been conducted by CSP-SMAST scientists to support management, where nitrogen reductions to reduce water column nitrogen levels are needed for restoration.
- Scientific analysis of TN levels and benthic animal habitat quality, documenting TN reduction for restoration. A full assessment of the relationship of TN level to infaunal habitat has been conducted by CSP-SMAST scientists to support management, where nitrogen reductions to reduce water column nitrogen levels are needed for restoration.
- 3. Scientific analysis of TN levels and accumulation of macroalgae, documenting at what levels and environments accumulation occurs. Much of this analysis was conducted in Falmouth estuaries and was conducted to support predictions of the impacts on macroalgal accumulations of lower nitrogen levels in estuarine waters.
- 4. Review of scientific analysis of oyster filtration and water clarity, on-going review of effects of nitrogen processing by bivalves on natural and artificial systems.

5. Assessment of over 60 estuaries under the MEP project including measurement and evaluation of benthic communities, sediment dynamics, surface water and stormwater inputs, water quality analysis, tidal dynamics, watershed delineation, watershed nitrogen loading, water quality modeling, and ecosystem assessments.

All of the information noted above directly relates to Little Pond nitrogen dynamics and the efficacy of the oyster monitoring project.

# Section II: Background Water Quality Monitoring in Little Pond

#### Post -MEP Data Synthesis of PondWatch Water Quality Data (2004-2011)

Previously under separate contract, the Coastal Systems Staff submitted the Post-MEP PondWatch water quality data (2005-2012) to the Town in electronic format with a Technical Memorandum<sup>3</sup>. The present Technical Report related to the Shellfish Demonstration Project provides the detailed 2013 water quality data, thus expanding the database.

The general findings of the PondWatch 2005-2012 monitoring indicated that although the Little Pond watershed is relatively small, it is densely developed with most commercial and residential property using on-site septic disposal of wastewater. The inlet is under-sized and periodically occluded by sand, restricting tidal flows. As a result TN levels tend to be high and the gradient in water quality within the pond is reduced, with the pond generally showing poor water quality throughout the tidal reach (LP-1, LP-2, LP-3) as depicted in Figure II-1. The result is that the estuarine basins of Little Pond are significantly impaired by nitrogen enrichment. The Sentinel Station (LP-2) has a nitrogen threshold of 0.45 mg TN/L, to generate TN levels of <0.42 in the lower basin (LP-3).

In contrast to other estuaries, Little Pond does appear to have rapid changes in TN as can be seen in the time-series TN record (Figure II-2). A rapid increase is particularly evident in the later years in the upper basin (LP-1), as well as other stations (Figure II-1). Overall the pattern is one of TN increasing throughout the estuary, but this cannot be evaluated on a year by year basis, as inter-annual variation is apparent. However, in general all basins showed increases in TN over the time-series consistent with changes in watershed nitrogen inputs, but also likely linked to changes in tidal flushing. Variations in tidal flushing are consistent with the observed inter-annual variation in salinity at each sampling station, which tended to be larger than observed in the other estuaries. Given the structure and location of the Little Pond inlet relative to local sand transport patterns under storm and non-storm periods it appears that occlusion of the inlet by sand deposition and periodic clearing during periods of higher tidal velocities are linked to the variations in TN levels. This is supported by the variation in salinity, where relatively large inter-annual variations were observed and in general lower salinities at a station were coupled to higher TN levels (Figure II-2). Keeping the existing inlet open and later widening it, should have a significant effect in lowering present TN levels and improving nitrogen related habitat quality within Little Pond.

Overall the temporal pattern of TN within Little Pond appears to follow the build-out projections of the MEP, with variation added due to variations in tidal flushing. Phytoplankton biomass/bloom activity did not show a clear pattern across the stations over 2005-2012. The restriction of tidal exchanges creates a situation somewhat like a pond, where water quality parameters show weaker gradients, i.e. tend to be more evenly distributed than in a well flushed estuary. The lack of gradient shows clearly in the chlorophyll a results. None-the-less, the level of chlorophyll a and TN are consistent with the level of habitat impairment noted by the MEP. There is no evidence of habitat improvement within this estuary and none is expected based on the water quality metrics.

<sup>&</sup>lt;sup>3</sup> B.L. Howes, S.J. Sampieri, D.G. Goehringer & R.I. Samimy. January 2014. PondWatch Nutrient Related Water Quality Little Pond, Oyster Pond and West Falmouth Harbor: SMAST POST-MEP Sampling. Coastal Systems Program Technical Memorandum to the Town of Falmouth Water Quality Management Committee.

Fortunately, the temporal pattern of change in key water quality metrics suggests a rapid response in water quality once nitrogen management alternatives (tidal flushing, nitrogen load reduction) are implemented.

# Pre-Oyster Pilot Project Nutrient Related Water Quality Monitoring Baseline (Summer 2012)

Coastal salt ponds and estuaries are among the most productive components of the coastal ocean. These circulation-restricted embayments support extensive and diverse plant and animal communities providing the foundation for many important commercial and recreational fisheries. The aesthetic value of these systems, as well as the freshwater ponds of a town, are important resources to both residents and the tourist industry alike. Maintaining high levels of water quality and ecological health in these aquatic systems (fresh and marine) is fundamental to the enjoyment and utilization of these valuable resources for all coastal communities.

As documented by the Massachusetts Estuaries Project analysis of the estuaries of the Town of Falmouth, many of the 14 estuaries within the Town are currently showing nutrient impaired water quality and resource loss. This is particularly true of Little Pond which has been documented to be nutrient enriched and eutrophic for almost 30 years. Based on the partnership between the Town of Falmouth and the Massachusetts Estuaries Project (MEP), through SMAST at UMass Dartmouth, a quantitative assessment of habitat quality in Little Pond was completed and an appropriate system specific nutrient threshold was determined for restoration of this estuary. The MEP found Little Pond to be currently supporting both impaired eelgrass habitat and benthic infaunal animal habitat. Restoration of habitat quality requires a lowering of nitrogen levels within Little Pond waters through reduction in inputs, increase in outputs (tidal flushing) or possibly alternatives to directly lower the nitrogen levels in pond waters (filter feeders such as oysters).

The Town of Falmouth has been moving forward on its Comprehensive Watershed Management Plan to restore the Town's estuarine resources for the citizens. As part of this effort the Town formed the Water Quality Management Committee to assist in the process. One innovative step by the Committee has been to investigate a variety of "non-traditional" alternatives to nitrogen management, including the use of oysters (filter feeders) as an inestuary approach to lowering nitrogen levels. Scientists at SMAST involved with the MEP have been working for almost a decade on this process and partnered with the Town of Falmouth on their Shellfish Demonstration Project. The Demonstration Project is being conducted in Little Pond.

In its prior work on non-traditional approaches to nitrogen management SMAST scientists had determined that a critical hurdle for acceptance of a new approach was to be able to quantify the mass of nitrogen removed per unit of alternative. In the case of oysters, this generally takes the form of mass removed per oyster or per million oysters, or per unit area of oyster aquaculture. This is needed to provide direct comparison to traditional approaches that have been quantified and are accepted, such as centralized wastewater treatment facilities. The Town of Falmouth agreed with the importance of quantifying the effectiveness of its Shellfish Demonstration Project on lowering nitrogen levels in Little Pond waters.

To this end, the Coastal Systems Program at SMAST undertook a pre-implementation baseline monitoring program during the summer of 2012, with the oysters slated for deployment in the summer 2013. What follows is a description of the sampling that was completed in Little Pond

in advance of the oyster aquaculture pilot project and to establish a solid pre-implementation water quality baseline. The emphasis of the summer 2012 monitoring was proof of concept on the monitoring approach and the establishment of the pre-oyster deployment baseline.

**Sampling Program:** The Coastal Systems Program has been responsible for the development and coordination of the majority of the estuarine and pond water quality monitoring programs across the southeastern Massachusetts region, inclusive of Cape Cod and the Islands as well as the analysis of the samples collected and synthesis of the resulting water quality data. As such, the CSP is able to leverage this comprehensive water quality database to further evaluate results obtained from the Little Pond post-oyster deployment monitoring results relative to potential long-term trends.

The 2012 Little Pond Pre-Oyster Water Quality Monitoring Program was established to:

- Develop the foundation (and context), a pre-oyster deployment baseline, for evaluating in-estuary nitrogen management by oysters and,
- Work out the details of obtaining an adequate snapshot of the nutrient gradient in Little Pond.

Sampling was conducted at nominal weekly intervals from July 8, 2012 to October 24, 2012. The sampling program was built around the existing PondWatch Water Quality Monitoring Program, which is under SMAST technical oversight. Additional sampling stations expanded on the historical 4 PondWatch Stations to add refined spatial coverage of the main nitrogen gradient within Little Pond (Figure 1). Stations were located by GPS and coordinates (LAT/LON) were obtained for each of the 9 stations sampled (Table 1). A total of 232 samples (plus additional field QA samples) were collected during the 14 sampling events (Table 2).

Sampling was conducted at 0.15 m depth and ~0.3 m above the bottom, at each of the 9 stations for nutrients and chlorophyll-a. Dissolved oxygen and Secchi depth was only measured at the 4 interspersed PondWatch stations. Sampling was conducted from a small boat during mid – ebb tide, consistent with standard PondWatch sampling and other water quality monitoring programs within the region (Mashpee, Barnstable, Yarmouth, etc), with the difference that sampling of all stations was completed within 1 hour to give a more accurate measure of the nutrient and chlorophyll gradient, as opposed to the traditional PondWatch sampling which has a slightly broader sampling window. The sampling was completed compatible with SMAST and MEP protocols.



Figure II-1. Sampling locations and proposed growing area in Little Pond as sampled during 2012 for development of the pre-oyster deployment base line. Total number of stations is 9 in order to quantify the "local" effect of the aquaculture pilot project. Four stations are traditional PondWatch stations shown as black dots (LP-Head, LP-1, LP-2, LP-3) and five stations are additional sampling sites shown as red triangles to quantify more localized effects of oyster aquaculture (LP-1.1, LP-1.2, LP-1.3, LP-2.1, LP-2.2). The proposed footprint of the oyster deployment is shown by the green polygon.

**Table II-1.** Coordinates of the pre-oyster deployment sampling in 2012. Station I.D.'s correspond to Figure II-1 and figures and data tables provided electronically. Note the tidal inlet is used as a reference.

STATION	COORDINATES*							
LP HEAD	041° 33.4576' N, 070° 35.4993' W							
LP1	041° 33.4078' N, 070° 35.4925' W							
LP1.1	041° 33.3404' N, 070° 35.4856' W							
LP1.2	041° 33.2865' N, 070° 35.4856' W							
LP1.3	041° 33.2315' N, 070° 35.4472' W							
L P 2	041° 33.1797' N, 070° 35.4101' W							
LP2.1	041° 33.1258' N, 070° 35.3758' W							
LP2.2	041° 33.0781' N, 070° 35.3565' W							
LP3	041° 32.9071' N, 070° 35.3579' W							
Inlet	041° 32.6540' N, 070° 35.3402' W							
* coordinates are in WGS 84								
"Inlet" does not have a	WQ station , it is for reference							

#### LITTLE POND PILOT STATION COORDINATES 2012

SMAST Technical Staff conducted the sampling with PondWatch support, both to assist the effort and as part of QA/QC procedures and to insure proper transport and delivery of samples to the Coastal Systems Analytical Facility<sup>4</sup>.

The physical parameters measured in the 2012 Little Pond samplings included: total depth, Secchi depth (light penetration), temperature, general weather, wind speed and direction, dissolved oxygen levels and observations of unusual events (fish kills, algal blooms, etc). Laboratory analyses included: salinity, nitrate + nitrite, ammonium, dissolved organic nitrogen, particulate organic carbon and nitrogen, chlorophyll-*a* and pheophytin-*a* and orthophosphate. Data were compiled and reviewed by the laboratory QA Officer and Project Manager for accuracy and evaluated to discern any possible artifacts caused by improper sampling technique. All data issues were resolved, unless noted in the database (note that none are noted in the 2012 data).

**Table II-2.** Sampling Schedule for Summer 2012 Little Pond Pre-Oyster Baseline. Station I.D.'s refer to those shown in Figure II-1. Data from these sampling events has been provided to the Town of Falmouth electronically (Excel format).

Little Pond Station Sampling 2012 for Pre-Oyster Water Quality Baseline										
Date	Head	LP-1	LP-1.1	LP-1.2	LP-1.3	LP-2	LP-2.1	LP-2.2	LP-3	
7/8/2012	2	2				2			2	
7/22/2012	2	2				2			2	
7/30/2012	2	2	2	2	2	2	2	2	2	
8/5/2012	2	2	2	2	2	2	2	2	2	
8/15/2012	2	2	2	2	2	2	2	2	2	
8/20/2012	2	2	2	2	2	2	2	2	2	
8/26/2012	2	2	2	2	2	2	2	2	2	
9/7/2012	2	2	2	2	2	2	2	2	2	
9/12/2012	2	2	2	2	2	2	2	2	2	
9/17/2012	2	2	2	2	2	2	2	2	2	
9/26/2012	2	2	2	2	2	2	2	2	2	
10/1/2012	2	2	2	2	2	2	2	2	2	
10/10/2012	2	2	2	2	2	2	2	2	2	
10/24/2012	2	2	2	2	2	2	2	2	2	
Total Samples =	28	28	24	24	24	28	24	24	28	
Total 2012 Summer Sam	ples =	232								
Standard PondWatch Sa	mples we	ere proces	ssed by SM	AST not	associated	d with the	Oyster Pi	lot Project		

<sup>&</sup>lt;sup>4</sup> The Coastal Systems Analytical Facility is sited within the School for Marine Science and Technology, UMASS-Dartmouth at 706 S. Rodney French Blvd, New Bedford, MA. 02744 (Sara Sampieri, 508-910-6325; ssampieri@umassd.edu). The laboratory supports a full range of environmental assays, with detection limits suited for natural waters. The laboratory data is accepted for both research and regulatory (USEPA, MassDEP, MCZM, NOAA) projects.

**Brief Description of Findings and Conclusions:** Although the Technical Memorandum previously submitted to the Town of Falmouth was meant only to present the baseline data, SMAST reviewed the data for suitability toward the intended purpose (oyster aquaculture pilot project) and to assess any issues that might need to be addressed or might inform the post-oyster deployment water quality monitoring effort.

*Dissolved Oxygen*. Dissolved oxygen (DO, Table II-3 and II-4) was measured as part of standard PondWatch sampling, using YSI meters and oxygen electrodes, calibrated before and after use. During the summer 2012 sampling events oxygen levels were consistently at or slightly below Commonwealth of Massachusetts water quality standards (>6 mg L<sup>-1</sup>). This was not, however, the case on the 7/22/12 sampling date where DO was observed to be well below the >6 mg/L threshold throughout the estuary. Additionally, it is important to note that on the 8/5/12 sampling date, DO concentrations in the bottom water at two sampling stations (LP-1 and LP-2) show near anoxic to hypoxic conditions. It is also important to mention that higher resolution time-series measurements have found periodic hypoxia in Little Pond bottom waters, emphasizing the need for time-series measurements. The grab sampling program does indicate that prolonged hypoxia may have been occurring in Little Pond on a limited basis during the 2012 field season.

Dissolved Oxygen in Little Pond Sampled by PondWatch Summer 2012										
	Head	ad LP-1 LP-2				LP-3				
	Surf	Surf	Btm	Surf	Btm	Surf	Btm			
7/8/2012	5.68	5.69	5.57	5.27	6.08	5.11	7.26			
7/22/2012	0.16	0.09	0.05	0.05	0.12	4.00	2.48			
7/30/2012										
8/5/2012	6.00	8.45	0.80	7.66	1.50	6.62	5.11			
8/15/2012										
8/20/2012										
8/26/2012	4.10	4.23	4.85	6.71	6.50	8.33	7.30			
9/7/2012										
9/7/2012										
9/12/2012										
9/17/2012										
9/26/2012										
10/1/2012										
10/10/2012										
10/24/2012										
Diss	olved Oxy	gen only C	collected o	n PondWa	tch Samplii	ng Dates				

#### Table II-3 - Summary of Summer 2012 Dissolved Oxygen (mg/L) Results for Little Pond Sampling

ID	Depth	Date	TIME	TEMP ©	SAL (ppt)	DO (mg/L)	% Sat
LP HEAD	S	7/8/2012	9:18	28.2	29.8	5.68	86%
LP1	S	7/8/2012	9:23	27.8	28.5	5.69	85%
LP1	В	7/8/2012	9:24	27.7	30.2	5.57	84%
LP2	S	7/8/2012	9:36	27.7	29.5	5.27	79%
LP2	В	7/8/2012	9:37	26.6	30.6	6.08	90%
LP3	S	7/8/2012	9:48	27.1	28.7	5.11	75%
LP3	В	7/8/2012	9:49	24.8	30.6	7.26	104%
		- ( (					
LP HEAD	S	7/22/2012	7:22	25.6	28.6	0.16	2%
LP1	S	7/22/2012	7:28	25.0	28.6	0.09	1%
LP1	В	7/22/2012	7:28	25.0	30.6	0.05	1%
LP2	S	7/22/2012	7:45	25.0	28.2	0.05	1%
LP2	В	7/22/2012	7:45	25.9	30.4	0.12	2%
LP3	S	7/22/2012	7:57	24.0	28.4	4.00	56%
LP3	В	7/22/2012	7:57	24.2	30.5	2.48	35%
	S	8/5/2012	6.23	23.3	23	6.00	71%
I P1	S	8/5/2012	6:26	23.3	2.5	8.45	109%
	B	8/5/2012	6:26	27.5	27.3	0.45	11%
1P2	S	8/5/2012	6:35	27.4	13.6	7.66	105%
1P2	B	8/5/2012	6:35	27.1	28.2	1.50	22%
1 P3	S	8/5/2012	6:44	27.1	25.2	6.62	96%
LP3	B	8/5/2012	6:44	27.4	30.1	5.11	76%
		-, -,					
LP HEAD	S	8/26/2012	9:50	24.6	23.0	4.10	56%
LP1	S	8/26/2012	9:55	24.2	29.0	4.23	60%
LP1	В	8/26/2012	9:56	27.4	30.0	4.85	72%
LP2	S	8/26/2012	10:04	26.1	27.5	6.71	97%
LP2	В	8/26/2012	10:05	27.3	30.3	6.50	97%
LP3	S	8/26/2012	10:16	24.3	26.8	8.33	116%
LP3	В	8/26/2012	10:17	26.9	30.7	7.30	109%

### Table II-4 - Results for Little Pond Sampling (summer 2012) Summary of Dissolved Oxygen (mg/L), Temperature, Salinity and % Saturation

<u>Nutrient Related Water Quality</u>. The main purposes of the 2012 sampling were to (1) develop a baseline for comparison to post-oyster deployment conditions (2) examine the pre-oyster water quality conditions and (3) define the water quality gradient within Little Pond.

Examination of the dissolved inorganic nitrogen (DIN) concentration data indicated that future evaluation of the nitrogen gradient needed to account for the salinity of pond waters (Figure II-2). During the 2012 samplings, it became clear that the pond periodically freshens due to storm events, possibly enhanced by tidal restriction. The cause would be high freshwater inflows with high DIN levels. The result is that there was significantly higher DIN in the upper and mid pond basins under fresher conditions (<25 ppt) and lower levels at higher salinities (>25 ppt) (Figure 2). Not surprising, the difference in DIN with salinity was most pronounced at the headwaters and diminished down-gradient to 0.07 mg/L by station LP-2.2. Accounting for salinity will allow for better assessment of oyster effects, versus independent external effects associated with precipitation. It should also be noted that while it is reasonable to see high DIN under low salinity conditions due to large inflow of freshwater in the upper reaches of the estuary, DIN can drop towards the inlet both because of the dilution that occurs in this area, as well as because of uptake by phytoplankton. Therefore, increases in particulate organic nitrogen (PON) concentration from the Head to LP-1 could be related to a decrease in DIN concentrations as DIN is taken up by phytoplankton. However, this inverse relationship can be obscured by other processes taking place in the estuary. For example, PON can settle out of the water column thereby decreasing PON while DIN decreases or PON can be released from sediments and show an increase even while DIN shows a decrease. The salinity effect was also found in the total nitrogen (TN) gradients (Figure II-3), but was much smaller, since DIN is only a relatively small fraction of the TN pool by station LP-1 and down-gradient stations (Figure II-4).

Careful review of phytoplankton biomass (chlorophyll-*a* + pheophytin-*a*) and total nitrogen (TN) data revealed a strong relationship. This confirms the assertion that lowering TN levels will reduce eutrophication in Little Pond (Figure II-5). Changes in relationships like this one and in the nutrient gradients or fractionation will be used as corroboration of findings from the post-oyster deployment sampling.



Figure II-2. Average dissolved inorganic nitrogen (DIN) gradients in Little Pond at high and low salinity. Stations refer to locations in Figure 1.



Figure II-3. Average total nitrogen (TN) gradients in Little Pond at high and low salinity. Stations refer to locations in Figure 1. TN threshold set by MassDEP is 0.45 mg/L to support eelgrass restoration. System remains above its TN threshold.



Figure II-4. Fractionation of the Total Nitrogen pool (top line is TN) in Little Pond over the 2012 field season. Headwaters are strongly influenced by watershed inputs which are high in DIN. In general, organic forms (DON and PON) dominate the TN pool throughout the pond. Oysters will have a strong immediate effect on TN and DIN levels.



Figure II-5. A strong relationship between phytoplankton biomass (chlorophyll-*a* + pheophytin-*a*) and total nitrogen (TN). This strongly supports the contention that lowering TN levels will reduce eutrophication in Little Pond.

#### Recommendations based on 2012 pre-oyster deployment sampling results:

- Deploying oysters in the upper water column around Station LP-2 appears to be optimal for assessing their effect on nitrogen levels in Little Pond.
- Nitrogen gradients (especially DIN) need to account for changes in pond salinity.
- Changes in biogeochemical relationships, not just TN concentration are important for determining oyster effects.
- A tide gauge should be placed within Little Pond main basin to allow assessment of periodic additional restrictions to tidal flows (SMAST can use its available gauges for this on request).

# Section III. Water Quality Assessment 2013 Post Oyster Deployment

In advance of the Town of Falmouth initiating the Shellfish Demonstration, a sampling plan was developed by the Coastal Systems Program Technical Team to quantify potential nitrogen removal by the proposed project in Little Pond. The sampling and chemical analyses followed the same procedures and methodologies as is employed for the PondWatch water quality monitoring, the MEP and most importantly, for the pre-oyster baseline analysis completed in the summer of 2012. The sampling design collected spatial data beyond historical PondWatch sampling (7 versus 4 stations, respectively), in order to better quantify the nutrient related water quality gradient and in particular, nitrogen species and total chlorophyll-a pigment.

The 2013 water quality sampling season was considered to be a transitional year, bridging the gap between "no" oyster effect and a "full" oyster effect anticipated in subsequent years beyond year 1 (2013) when the oysters were first introduced to the system. As the water quality monitoring program for the oyster project was initially envisioned, the anticipated methodology would be to overlay the 2013 spatial and temporal data set over the 2012 dataset and see agreement in the May/June timeframe with a growing divergence in summer and fall samplings. As it evolved, inter-annual variation and seasonal variation resulted in the main focus of the analysis being to quantify changes in concentrations of key constituents along the main axis of the pond (from above the oyster deployment area to below) and examine differences in the gradients of nutrient-related parameters between 2013 vs. 2012. These comparisons allowed evaluation of any effect of the oysters even though the absolute concentrations of some parameters varied from year to year and season to season. Also, the 2012 and 2013 data form the basis for comparison to subsequent years and guide the future deployments of oyster aquaculture rafts in out years.

Two critical objectives of the 2013 water quality monitoring effort were: 1) the quantification of the effects of oyster cultivation in Little Pond and 2) document water quality improvements resulting from such a nitrogen management approach. To achieve these objectives, the water quality data collected during the 2013 growing season was compared to the 2012 results, prior PondWatch data and the nitrogen thresholds established by the Massachusetts Estuaries Project. The comparison is geared specifically to assist with adaptive nitrogen management planning to meet the nitrogen threshold for the Little Pond system as established by the Massachusetts Estuaries Project.

Per the scope of work, samples were analyzed for the following parameters: temperature, total nitrogen (nitrate + nitrite, ammonia, dissolved organic nitrogen, particulate organic nitrogen, dissolved organic nitrogen), chlorophyll-a, pheophytin-a, orthophosphate, salinity, temperature, dissolved oxygen, transparency (secchi depth), alkalinity. As is done during all water quality sampling , weather and tide-status were documented to assist in the interpretation of results. Additionally, sampling was completed synoptically during a narrow (~2 hr.) window to minimize the effects of a changing tide (ebb tide sampling) as well as weather-related effects on samples (e.g. significant precipitation events). Salinity measurements were correlated to rainfall and other relevant parameters.

The proposed sampling locations established for the oyster monitoring project build upon the existing PondWatch monitoring stations for data collection ("LP" in Figure III-1) utilized by the Massachusetts Estuaries Project (MEP), but were to be a down-sampling from the predeployment baseline conducted in 2012. CSP staff decided that down-sampling would cause difficulties in interpretation, especially if some 2012 stations were not sampled in 2013 or their locations were moved. As a result CSP maintained the sampling program from 2012 in 2013 allowing the full suite of sampling locations for comparative analysis to be performed. Proposed locations for sampling included: LP-1.1, LP-1.2, LP-2.1 and were located specifically for the purposes of the oyster demonstration project (Table III-1, III-2). The 2 additional stations (LP-1.3 and LP-2.2) were sampled throughout the 2012 and 2013 monitoring program, to increase the accuracy of the results.

The pre-oyster deployment sampling demonstrated that a higher station density (9 stations) allowed detection of smaller differences than coarser scale sampling. In 2012 sampling, stations were situated to give 3 near-field stations both up-gradient and down-gradient of the ovster deployment area. This configuration yields a more robust assessment of changes in gradient due to passage of water through the oyster deployment area. Also, three stations is the minimum to detect trends, which is why 3 stations (up-gradient and down-gradient) were sampled in 2012 and 2013. Further, given the significance of the results of the trend analysis (see below), it likely that the MEP water quality model might usefully be brought to bear to help determine the TN mass reduction by the oysters (kg N/reach/unit time from flow in model). This would be initiated after the larger scale oyster deployment in 2014. Results of the numerical modeling is greatly improved by having high resolution spatial data. The ability to directly compare year to year, date to date and site to site greatly increases the probability of determining the "oyster effect" and evaluating nitrogen removal relative to regulatory requirements. The CSP science team strongly supports the continuation of high resolution spatial data collection in the near-field, and possibly removing a far-field station in the future (Table III-2).



Figure III-1. Sampling locations and growing area in Little Pond sampled during the 2012 base line and 2013 growing season. SMAST determined that 9 stations (rather than 7) would enable more robust quantification of the localized effect of the Shellfish Demonstration and would provide direct comparison to the pre-oyster deployment 2012 baseline which also sampled these 9 stations. Four stations are PondWatch stations (LP-Head, LP-1, LP-2, LP-3) and five stations are additional sampling sites to quantify more localized effects of oyster aquaculture (LP-1.1, LP-1.2, LP-1.3, LP-2.1, LP-2.2).

Table III-1. Proposed dates and stations from which water samples were to be collected for the 2013 oyster demonstration project.

													TOTAL
													(surface and
STATION	June 1-15	June 16-30	July 1-15	July 16-31	Aug 1-15	Aug 16-31	Sept 1-15	Sept 16-30	Oct 1-15	Oct 16-31	Dec 1-15	April 1, 2014	bottom
LP-Head	2	2	2	2	2	2	2	2	2	2	2	2	24
LP-1	2	2	2	2	2	2	2	2	2	2	2	2	24
LP-2	2	2	2	2	2	2	2	2	2	2	2	2	24
LP-3	2	2	2	2	2	2	2	2	2	2	2	2	24
LP-1.1	2	2	2	2	2	2	2	2	2	2	2	2	24
LP 1.2	2	2	2	2	2	2	2	2	2	2	2	2	24
LP-2.1	2	2	2	2	2	2	2	2	2	2	2	2	24
ALL													168

Table III-2 Summary of Year 1 Oyster Demonstration Project sampling (2013 growing season). Total number of stations is 9 in order to quantify the localized effect of the aquaculture experiment and to provide direct comparison to the pre-oyster deployment baseline which also sampled these 9 stations during the same May 1 – October 31 period. Four stations are PondWatch stations (LP-Head, LP-1, LP-2, LP-3) and five stations are additional sampling sites to quantify more localized effects of oyster aquaculture (LP-1.1, LP-1.2, LP-1.3, LP-2.1, LP-2.2). Two additional sampling events (December and April) are factored into the year 1 sampling program per the Town of Falmouth Conservation Commission Order of Conditions. The number two (2) indicates that surface and bottom water samples were collected. FD = Field Duplicate Sample.

	Little Pond Station Sampling 2013								
Date	Head	LP-1	LP-1.1	LP-1.2	LP-1.3	LP-2	LP-2.1	LP-2.2	LP-3
5/3/2013	1 + FD	2	2	2	2	2	2	2	2
5/17/2013	1	2	2	2	2	2	2	2	2 + FD
6/4/2013	1	2 + FD	2	2	2	2	2	2	2
6/18/2013	1	2	2	2	2	2	2	2	2 + FD
7/1/2013	1	2	2	2	2 + FD	2	2	2	2
7/18/2013	1	2	2	2	2	2	2 + FD	2	2
7/31/2013	1 + FD	2	2	2	2	2	2	2	2
8/15/2013	1 + FD	2	2	2	2	2	2	2	2
8/29/2013	1	2	2	2	2	2 + FD	2	2	2
9/16/2013	1 + FD	2	2	2	2	2	2	2	2
10/1/2013	1	2	2	2	2	2 + FD	2	2	2
10/16/2013	1	2	2	2	2	2	2	2	2 + FD
12/8/2013				2	2	2	2 + FD	2	2
Total Samples per Station=	16	25	24	26	27	28	28	26	29
Total 2013 Summer Samples=	229								

#### Results:

The major question to be addressed relates to whether the oysters make a discernable change in the water column nitrogen related constituents in Little Pond. Since oysters were introduced to the surface waters of the pond on July 10, 2013 and removed in late October, this was the primary period for evaluation. The analytical approach involved comparing the gradients in water quality parameters both during the oyster deployment (2013) and during the same period in 2012 when no oysters were deployed. Since the oysters were deployed in floating bags, initially as small spat, their effect over the first month in the pond is expected to be negligible. Therefore, the period for gauging the effect of the ~1 million oysters deployed was July 31, 2013 through the end of October, while the oysters were larger and actively feeding. Analysis using the entire dataset (May-December) was also performed, but the results were confounded by high rainfall in May and June and very cold temperatures (low activity) in December, coupled with the absence of the oysters in May, June and December. Therefore, future monitoring does not need to include these sampling dates in order to effectively determine any impacts on water quality caused by the Shellfish Demonstration Project.

#### **Stratification of Little Pond**

In both 2013 and 2012, the Little Pond water column showed moderate stratification<sup>5</sup> (Figure III-2). Stratification of an estuarine water column is most commonly the result of salinity differences from the surface to the bottom waters. This salinity difference results from fresh waters entering from the watershed and differentially freshening the surface water, and marine waters entering from the tidal inlet differentially increasing the salinity of bottom waters. A secondary factor in the density difference is the occurrence of colder bottom waters overlain by warmer surface waters. The salinity difference is clear in both 2012 and 2013 results (Figure III-2). The salinity differences exist at all stations, but diminish moving away from the major freshwater inflows in the pond's upper reaches. While the pond was periodically hypoxic, periodic mixing did occur which aerated the bottom waters. Although it is difficult to quantify, it appears that stratification focused the effects of the oysters on the surface layer, since the oysters were deployed in floating bags at the pond surface. From the water quality sampling it is clear that the surface and bottom layers of the pond water column are not tightly coupled through the warmest months of the study when the oysters were having their greatest effect (see below).

The stratification of surface and bottom waters can also be clearly seen in the key oyster food indicators of total chlorophyll *a* pigment and particulate organic carbon (Figure III-3). In both cases the higher phytoplankton production in the surface layer, due to light availability, can be seen in the significantly higher concentrations in surface versus bottom waters at almost all sampling locations in 2013. The same effect was seen in 2012 over the same time period, but with smaller differences.

<sup>&</sup>lt;sup>5</sup> Well mixed conditions show similar salinity and temperature at surface and bottom, while moderate stratification shows higher salinity (1-2 PSU) at bottom than surface.

#### Chlorophyll, Particulate Organic Carbon (POC) and Fluorescence Data Analysis

#### Particulate Organic Carbon and Total Pigment Data

Data for both total chlorophyll *a* and POC<sup>6</sup> shows higher phytoplankton concentrations in the stations upgradient (LP-1.2) of the deployment area (LP-1.3). Concentrations on the ebbing tide decrease measurably at LP-1.3, with a continued decline at LP-2, downgradient of the Demonstration Site. This was clearly seen in the surface waters where the decline from upper to lower station was 0.64 mg C/L for POC and 6.7 ug/L for Total Chlorophyll *a*. This decline was not seen across the bottom waters across these sites, further supporting the conclusion that the decline is due to the removal of particles by the oysters in the surface layer, with deposition likely downgradient as the particles settle. The decline occurred over the three "near-field" stations as would be expected as water flows downgradient through the oyster area on the ebbing tide.

Analysis of the 2012 water quality results from these same 3 sites, when there were no oysters deployed, shows no significant decline in POC or Total Chlorophyll *a*. This is consistent with the conclusion that the pattern in 2013 resulted from the addition of oysters and not from a natural gradient within the estuary (Figures III-3 and III-4).

#### Chlorophyll a and Pheophytin a Data

When oysters filter phytoplankton and process the particles, the chlorophyll *a* in the phytoplankton chloroplasts is typically degraded to pheophytin *a*. This can also occur when zooplankton graze on phytoplankton cells and is a well-documented effect of grazing.

Examining the chlorophyll *a* and pheophytin *a* gradients in the 2013 and 2012 monitoring indicates that there is intense "grazing" of phytoplankton in the surface layer in 2013, but not in 2012, and not in the bottom waters (Figures III-5 and III-6). In 2013 there is a decline in chlorophyll *a* concentration through the near-field, with concentrations upgradient > oyster site > downgradient station on the outgoing tide. The bottom layer did not show this pattern. The 2012 data showed the opposite, with chlorophyll *a* increasing to a peak at the site of the next year's oyster deployment and the pheophytin *a* showing a continuing decline through the area. Of particular note in 2013, the pheophytin *a* "spiked" within the oyster deployment area, compared to upgradient and downgradient sites. These measurements are consistent with the oysters actively feeding (deployment area) where phytoplankton are observed to have been consumed (chlorophyll *a* declines) and "digested" (spike in pheophytin *a*) and that this activity is mainly seen in the surface layer due to water column stratification at the time of the measurements. That these patterns were not observed in 2012 when the oysters were not deployed is also consistent with the effect resulting from oysters rather than a natural gradient within the pond.

<sup>&</sup>lt;sup>6</sup> There is a linear relationship between total chlorophyll a and POC in Little Pond water as in many other estuaries on Cape Cod. This was presented in the 2012 Pre-Oyster Deployment Monitoring Report [B.L. Howes, S.J. Sampieri and R.I. Samimy. 2013. Nutrient Related Water Quality Monitoring Baseline for Gauging Nitrogen Removal by Town of Falmouth Oyster Pilot Project in Little Pond Summer 2012, Technical Memorandum from the Coastal Systems Program-SMAST to the Town of Falmouth.].

These results related to oyster feeding through filtration of water column particles (phytoplankton) is supported by the time-series measurements of fluorescence and light attenuation.

#### Fluorescence Data

Although the mooring data varies considerably throughout the three month deployment, there were generally trends seen from the head of the pond down to the mouth. Highest chlorophyll was generally upgradient of the oyster beds. Chlorophyll levels declined through the Demonstration Site itself (LP-1.3), with the lowest levels at LP-2, immediately downgradient of the Demonstration Site. The extent of this depletion is not quantifiable from the time-series measurements, as the observed increase in pheophytin *a* in this region partially obscures the loss of chlorophyll *a* (when measured by in situ fluorescence). While the general trend is decreasing concentrations from the head to the mouth of the pond, during bloom conditions chlorophyll concentrations were higher directly upstream of LP-1.3. Spatial trends in chlorophyll concentrations were lower chlorophyll a levels within the Demonstration Site. Moreover, continuous fluorescence sensors for chlorophyll do not differentiate between chlorophyll *a* and pheophytin *a*, thus the magnitude of the changes may be reduced.

In addition, the removal of particles is expected to result in a change in water clarity. Timeseries measures of water clarity were consistent with the time-series fluorescence measures and the water quality grab sampling results, in that the water showed increased clarity within the Demonstration Site (LP-1.3) relative to upgradient and downgradient sites. Independent measurements demonstrate a phenomenon occurring within the oyster deployment area, consistent with oysters removing significant amounts of phytoplankton and particulates from the water column. This is expected as it is a direct result of feeding, which is further evidenced by the localized spike in pheophytin *a*.

Additional details on fluorescence provided in see Section IV.

#### **Nutrient Data**

The removal of phytoplankton/particles by the oysters could also be seen in the total nitrogen (TN) results from the 2013 post-oyster deployment versus 2012 pre-oyster deployment. In 2013 TN showed a decline on the ebbing tide through the deployment area. A peak in chlorophyll a is seen at LP-1.2, and rapidly declines at LP-1.3 with a minimum concentration seen at LP-2. In 2012, the concentration of chlorophyll *a* showed relatively small change from station to station (i.e. flat gradient; Figures III-7 and III-8). The "oyster effect" was not as large in the TN data as in the other parameters noted above (Figures III-2 thru III-6), but is still clearly present and shows a similar pattern as chlorophyll *a*, peaking at LP-1.2, upgradient of the Demonstration Site and declining at LP-1.3, within the oyster deployment area. TN is comprised of a large dissolved organic nitrogen fraction which is relatively constant throughout the pond waters, presenting a large relatively inactive background nitrogen pool. This background obscures the effects of the oysters because their impact is primarily on particulate and inorganic nitrogen. In order to discern the oysters' potential impact on TN constituents, the TN pool was fractionated into particulate nitrogen and inorganic nitrogen.

Inorganic nitrogen can be generated in several ways. It can be directly excreted by the oysters from the digestion of particulate organic nitrogen (phytoplankton) to the surrounding water, or it

can be formed from the decomposition of particulate matter some of which is released and some of which is oxidized to nitrate prior to entering the water column. Examining the data for ammonium and nitrate+nitrite (NOx), there is no clear increase in either nitrogen species within the deployment area. The only evidence of a potential effect of the oysters stems from comparing the 2013 and 2012 (pre-deployment) data (Figures III-9 and III-10). In 2012 the NOx and ammonium showed smooth gradients, with levels declining from the headwaters to the tidal inlet. In contrast, in 2013 the gradients ammonium tended to show a break from above to below the oyster deployment area. It is possible that the relatively low levels of inorganic nitrogen and the rapid uptake by phytoplankton (once released to the water column) obscure any clear pattern associated with the oysters. At this point these data are supportive but not compelling. In contrast, the PON results showed a clear pattern of removal by the oysters, consistent with the observations of chlorophyll a and particulate carbon discussed above. It appears that removal of particulate nitrogen by oysters is the predominant cause of the reduction in TN. However, at this time the fate of this particulate nitrogen is not yet quantified.

Particulate nitrogen once removed by oysters can be consumed and digested with incorporation into tissue or excreted as waste, this represents a small fraction of what the oysters collect with the result that a substantial amount of pseudo-feces are generated. These pseudo-feces once released can be transported in tidal waters as additional particulates (TSS) or reach the pond sediments where the nitrogen is remineralized and either released to the water column as inorganic nitrogen and consumed in part or fully by phytoplankton and bacteria or be denitrified (coupled nitrification-denitrification). This latter process is routinely measured at SMAST and our quantification of this removal resulted in MassDEP granting in the Town's wastewater discharge permit a 40% removal of nitrogen entering Mashapaquit Creek in West Falmouth.

One concern related to potential negative effects of the oysters on the pond system stems from the above mentioned potential increase in deposition of particles to the sediments. This would result from a strong localized change in organic matter deposition and one result might be low oxygen. While in 2013 Little Pond experienced hypoxia, this phenomenon occurred both prior to, and after the deployment of the oysters. Further, hypoxia was not focused within the deployment area. It is possible that oxygen conditions improved in both surface and bottom waters on the ebbing tide as the water passed through the oyster deployment site. Improved oxygen condition in the Demonstration Site paralleled the decline in particulate organic matter. While there are a myriad of potential causes for this pattern in oxygen level, it is clear that the oysters were not resulting in a depletion of oxygen in the surface or bottom waters. This is further supported by the historical oxygen measurements by the MEP and PondWatch which indicate the existence of periodic summertime hypoxia in Little Pond for over two decades.

#### **Conclusions:**

The water quality monitoring results clearly document that the deployment of oysters in Little Pond did produce modest water quality improvements near-field to the deployment area. The primary mechanism of this water quality improvement appears to be the uptake of phytoplankton by the oysters.

Lines of evidence from nutrient data (TN, PON, POC), chlorophyll *a*, fluorescence and turbidity analyses (detailed below in Section IV below) indicate that the oysters are removing particles from the surface layer of Little Pond, with the effect clearly discernible in the near-field region (LP-1.2, 1.3, 2). It is likely that tidal action, dilution and other biologic processes tend to predominate in other areas, muting the oyster effects at these monitoring stations.

Several independent measurements support this conclusion:

- Nutrient data show a decrease in water column total nitrogen (TN) at LP-1.3, within the oyster deployment area compared to upgradient sites on the ebbing tide.
- Nutrient data shows lower POC and PON at LP-1.3 than in stations above the Demonstration Site.
- Lab analysis of water samples for chlorophyll *a* shows reduced phytoplankton at LP-1.3 on the ebbing tide.
- Lab analysis of water samples for pheophytin *a* show an increase within the deployment area, which is the result of the degradation of phytoplankton chlorophyll *a* to pheophytin as oysters feed.
- Time-series measurements from moored instruments at 4 sites show that chlorophyll *a* increased above, and declined across the Demonstration Site.
- Time-series measurements from moored instruments at 4 sites (Section IV) show an improvement in water clarity (i.e. decreased turbidity).
- The moderate stratification of the water column focused the "oyster effect" in the surface layer where they were deployed. Comparisons between 2013 (oyster deployment) and 2012 (no oyster deployment) demonstrate that it was the oysters, and not natural gradients that were producing the observed effects in the region nearfield to the deployment area.

While there were initial theoretical questions regarding the potential for oyster aquaculture to negatively impact oxygen dynamics, data from 2012, as well as the 2006 Little Pond MEP Report and earlier data documents that hypoxia is a condition in Little Pond preceding the oyster deployment by decades. Moreover, the oysters were effectively separated from the bottom water by the moderate stratification. The pattern of bottom water oxygen suggested that the oysters may have had a positive effect (if any) on oxygen conditions. Based on the water quality monitoring, the oyster deployment was not found to have any negative ecological impacts on the estuarine habitats within the Little Pond System.

The 2013 oyster deployment consisted of 1 million oysters. In 2014, the deployment of an additional 1 million oysters, for a total of 2 million oysters, should lead to stronger trends being observed in the associated monitoring data. With this additional oyster activity, some quantification of the nitrogen removal should be possible as flow measurements are proposed.

#### **Recommendations:**

(1) Measure the stimulation of denitrification in sediments resulting from the delivery of particles to the sediments from oyster feeding. SMAST has already performed these types of quantitative measures to support Town permits in other estuaries.

(2) Quantify the amount of particulate capture and deposition from oysters in the oyster deployment area in 2014. This would be accomplished by a trapping system deployed periodically with the oyster bags/cages.

(3) Conduct a passive inert tracer study to quantify the amount of phytoplankton/PON removed by the oysters separate from dilution as water parcels move downgradient through the deployment area.
(4) Do not change the location of stations in the upper pond, but realign far-field stations or create new stations to increase the data collection density in the near-field. Ideally, a station would be added between the existing PondWatch Station (LP-2) and the recently created station LP-2.1. New station could be labeled (LP-2.0)

(5) Early season (May) and late season (post-oyster removal) sampling did assist in the analysis of whether there is any "oyster effect". Sampling should concentrate on the several months where the oysters are most active. Rainfall and storm events confounded early season results and December data collected when the biological systems within the pond are relatively inactive, limited the utility of these data. Therefore, future monitoring does not need to include these sampling dates in order to effectively determine any impacts on water quality caused by the Shellfish Demonstration Project.

(6) The effect of the oysters was clear, but limited in extent. Doubling the number of oysters, deploying them earlier and having larger oysters from 2013 redeployed should greatly enhance quantification of the effects and also provide support for scaling the results for other deployments.

(7) Following the larger scale oyster deployment in 2014 it may be that the MEP water quality model might usefully be brought to bear to help determine the TN mass reduction by the oysters (kg N/reach/unit time from flow in model). This should only be considered if the 2014 data show significant reductions and will support a numerical modeling scenario.

(8) Flow and TN and PON concentration measurements should be collected above and below and within the oyster deployment area over 3 tidal cycles to show removal on both the ebbing and flooding tide.

(9) Add CO<sub>2</sub> concentration via alkalinity analysis to battery of water quality parameters being measured.



Figure III-2. Water column salinity averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA. Water was collected near surface (0.15 m depth) and near bottom (0.3 m above) in, Top 2013 (oysters) and Btm, 2012 (no-oysters), The water column was moderately stratified in both years..



Figure III-3. Water column phytoplankton biomass metric averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013. (Top) Total Chlorophyll a and (Btm) Particulate Organic Carbon. Both parameters relate to both filtration and fecal production by oysters. Bar shows most likely area of "oyster effect".





Figure III-4. Water column phytoplankton biomass metric averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2012. (Top) Total Chlorophyll a and (Btm) Particulate Organic Carbon. Both parameters relate to both filtration and fecal production by oysters in 2013, but 2012 served as a no-oyster control summer for comparison to 2013 (Figure 3). Horizontal bar shows



Figure III-5. Water column phytoplankton pigment averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013. (Top) active Chlorophyll a and (Btm) initial degradation product of Chlorophyll a, generally associated with phytoplankton senescence or invertebrate grazing/feeding. Shifts from Chlorophyll a to Pheophytin a can be used as an indicator of oyster feeding.



Figure III-6. Water column phytoplankton pigment averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2012. (Top) active Chlorophyll a and (Btm) initial degradation product of Chlorophyll a, generally associated with phytoplankton senescence or invertebrate grazing/feeding. As there were no oysters deployed, shifts from Chlorophyll a to Pheophytin a were judged to be associated primarily with phytoplankton senescence.



Figure III-7. (Top Panel) water column total nitrogen and (Bottom Panel) phytoplankton biomass (total chlorophyll a pigment) averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013.



Figure III-8. (Top Panel) water column total nitrogen and (Bottom Panel) phytoplankton biomass (total chlorophyll a pigment) averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2012.





Figure III-9. Water column inorganic nitrogen averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013. (Top Panel) water column nitrate+nitrite and (Bottom Panel) ammonium. Bar represents region of maximum change.





Figure III-10. Water column inorganic nitrogen averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2012. (Top Panel) water column nitrate+nitrite and (Bottom Panel) ammonium. Bar represents region of projected maximum change when oysters are deployed..



Figure III-11. Water column bioactive nitrogen averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013. (Top Panel) water column dissolved inorganic nitrogen (ammonium+nitrate +nitrite) and (Bottom Panel) particulate organic nitrogen primarily associated with phytoplankton cells living and dead. Bar represents region of maximum change.



Figure III-12. Water column bioactive nitrogen averages from the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2012. (Top Panel) water column dissolved inorganic nitrogen (ammonium+nitrate +nitrite) and (Bottom Panel) particulate organic nitrogen primarily associated with phytoplankton cells living and dead. Bar represents region of maximum change.



Figure III-13. (Top Panel) Bottom water oxygen minimum measured during the high frequency grab sampling surveys conducted in Little Pond, Falmouth MA from July 31 (nominally August) through late October, 2013. (Bottom Panel) particulate organic carbon concentration presented as an indicator of potential oxygen uptake (as substrate or as phytoplankton. Bar indicates region of maximum change.

## Section IV. Time-Series DO/CHLA Moorings (Intensive Sampling for Little Pond Oyster Demonstration Project)

*Time-series Mooring Deployment and Sampling:* Bottom moored sondes capable of high frequency measurements of both dissolved oxygen and chlorophyll-*a* were deployed July 1, 2013 at locations noted on the accompanying map (Figure IV-1) 30 cm from the sediment surface. On July 9 and 10 physical samples were collected at each site and the DO Upper B mooring was moved to the location labeled DO Upper A BOTTOM to reflect amendments made in the original scope of work.

Figure IV-1 shows the deployment locations and nomenclature used throughout the study as follows:

- LP Upper B **BOTTOM**
- LP Upper A **BOTTOM**
- LP Oyster Raft SURFACE
- LP Lower A BOTTOM
- LP Lower B **BOTTOM**

At one week intervals calibration samples were collected at the specific depth and location of each sonde. Sondes were then inspected, cleaned and the data was downloaded. Sondes were returned to the moorings and secured. Calibration sampling included triplicate Winkler samples for dissolved oxygen determination as well as whole water for chlorophyll extraction. Calibration samples were used to correct time series data for slight calibration drift during the deployment.

*Time-series Sensor and Sonde Data Results and Discussion:* Dissolved oxygen levels were generally low after the first few days of deployment and remained anoxic or hypoxic for a majority of the time, with the exception of the LP Oyster Raft SURFACE sonde which experienced little influence from sediment oxygen demand. Low bottom water dissolved oxygen appears to be the result of water column salinity and temperature stratification (Figure IV-2 and IV-3). The Oyster Raft sonde, located at the surface, had consistently lower salinity and higher temperatures than all other sondes which were moored near the sediment surface, thus confirming a potential barrier to oxygen exchange with the bottom water. The remaining four sondes all had similar temperatures and salinities. Individual plots of temperature, salinity and depth are located in Appendix 1.

The physical parameters of salinity, temperature and depth create an environment that is sensitive to changes in primary production and respiration. Fresh water from head waters and surface runoff both enhances stratification and stimulates primary production through nutrient inputs. Thus, large salinity drops at the surface moored Oyster Raft sonde are followed by increases in phytoplankton chlorophyll (Figure IV-4). Since this sonde was located at the surface, above the stratified layer, the oxygen produced by the additional phytoplankton during the day result in late afternoon spikes in oxygen concentration well above air saturation. During the dark hours, when that same additional phytoplankton respires, water column oxygen drops below saturation. The result was large diurnal swings in dissolved oxygen. Unlike the bottom moorings (Figures IV-5 through IV-8) however, the dissolved oxygen concentrations rarely reach anoxia because exchange of oxygen with the atmosphere was not impeded by stratification.

Bottom moorings (Figures IV-5 through IV-8) received only a fraction of the light received at the surface thereby decreasing primary production by phytoplankton and concomitantly decreasing oxygen production during daylight hours. In addition, these instruments were close to the sediment surface which continuously respired water column oxygen. Thus, compared to the surface, bottom water oxygen concentrations were depressed, often to the point of anoxia, while diurnal excursions in oxygen concentration were of a similar magnitude throughout much of the deployment. Low levels of sulfide were detected in the bottom water during the July 9 sampling and were episodic throughout most of the deployment period. Particulate sulfur was observed in the bottom water on July 10 and July 19 indicating persistent anoxic conditions.

Large blooms of phytoplankton were associated with rain events bring nutrients into the pond, most notably at the beginning of the mooring record, which coincided with a storm which dropped several inches of rain in a single day. Salinity gradients produced by the fresh water stream at the head of Little Pond suggest that much of the nutrient inputs come from this source. The decreasing concentration of chlorophyll during bloom conditions observed with increasing distance from the head waters was consistent with this process. (See Figures IV-5 through IV-7, ~7/29/13). It should be noted that the large diurnal oscillations in chlorophyll concentrations are mainly an artifact of using the proxy measurement of fluorescence (standard method) to measure chlorophyll concentrations. The strength of the fluorescent response to sunlight (and the pulse of light produced by the sonde prior to measurement) by individual phytoplankton tends to be lower during daylight hours when the chlorophyll is more likely to be saturated with light than at night when ambient light is absent. Thus, the large concentration changes over a day/night cycle. While this is generally not accounted for by many users of YSI moorings, it is a phenomenon noted by many researchers.

Chlorophyll concentrations decreased somewhat during the latter part of the deployment. Cooler water, which can hold more oxygen, had the effect of generally increasing dissolved oxygen concentrations in the bottom water as well. Most interesting, with respect to the oyster transplants, was that once all the oysters were placed in the pond there appeared to be a decrease in the amplitude of phytoplankton blooms downstream of the rafts. Although we cannot ascribe causation at this time the observation is consistent with the hypothesis that oyster culture would tend to remove phytoplankton from the water. There was, however, no clear relationship between decreased phytoplankton and elevated bottom water dissolved oxygen. Little Pond represents a shallow, eutrophic estuary in which sediment respiration makes a large contribution to the overall water column oxygen budget, so if the observed decrease in phytoplankton density downstream from the oyster rafts turns out to be causally related it may take several years before the decreased organic load to the sediment is reflected in the water column oxygen concentrations.

It is clear that the oxygen conditions were showing depletion at both the surface and bottom, but the observed depletions did not appear to be related to the oysters that have been placed in the pond. Conditions were poor before oyster introduction and either remained poor or improved throughout the summer. Oxygen conditions were no worse within the oyster culture area (Upper A (Bottom)) than anywhere else, in fact better than some suggesting that accelerated deposition of fecal pellets below the rafts was either not an important factor or that the observed poor water quality left little margin for detecting deleterious effects.



Figure IV-1. Final mooring locations reflecting amended scope of work.

**Mooring Temperatures** 



Figure IV-2. Time series record of temperature for all moorings.



**Mooring Salinities** 

Figure IV-3. Time series record of salinity for all moorings.





Figure IV-4. Dissolved oxygen and chlorophyll time series data plots for surface deployed Oyster Raft sonde. Red dots denote calibration samples.



Figure IV-5. Dissolved oxygen and chlorophyll time series data plots for surface deployed Upper B (Bottom) sonde. Red dots denote calibration samples.



Figure IV-6. Dissolved oxygen and chlorophyll time series data plots for surface deployed Upper A (Bottom) sonde. Red dots denote calibration samples.





Figure IV-7. Dissolved oxygen and chlorophyll time series data plots for surface deployed Lower A (Bottom) sonde. Red dots denote calibration samples.





Figure IV-8. Dissolved oxygen and chlorophyll time series data plots for surface deployed Lower B (Bottom) sonde. Red dots denote calibration samples.

Comparing time series data collected over the summer of 2013 with data collected with similar protocols in 2002, it is obvious that water quality conditions have degraded significantly over the last decade (Figures IV-9 and IV-10). Tidal phases were similar and water temperatures were similar. Salinity, however, was much lower in 2013, on average about 2 ppt lower than observed in 2002.

Oxygen concentrations in 2002 reached hypoxia, but never declined to zero, whereas in 2013 over the same time span oxygen concentrations regularly reached sub-milligram per liter levels. Chlorophyll concentrations were also significantly higher in 2013 than in 2002, occasionally as much as six fold higher. A statistical summation is provided in Table 1, similar to that presented in the MEP report on Little Pond. The differences are dramatic. For example, in 2002 conditions were hypoxic (<3mg/L) or worse 2% of the time, while in 2013 that number increased to nearly 70%. Similar results can be seen with the chlorophyll concentrations (Table 2) which exceeded 15ug/L 12% and 26% of the time for 2002 and 2013, respectively.



Figure IV-9. Time series temperature (upper panel) and salinity (lower panel) measurements performed in 2002 (blue/upper trace) and 2013 (red/lower trace) at Upper B (Bottom).



Figure IV-10. Time series dissolved oxygen and chlorophyll measurements performed in 2002 and 2013 at Upper B (Bottom).

Table IV-1. Dissolved oxygen summary for sonde deployments at Lower A (Bottom) in Little Pond for 2002 and 2013.

Dissolved Oxygen Statistics for 2002 and 2013 Mooring Deployments							
			Total	<6 mg/L	<5 mg/L	<4 mg/L	<3 mg/L
			Deployment	Duration	Duration	Duration	Duration
	Start Date	End Date	(Days)	(Days)	(Days)	(Days)	(Days)
Little Pond Upper	7/14/2002	8/11/2002	28.0	5.85	3.77	1.56	0.53
			Mean	0.19	0.15	0.09	0.05
			Min	0.01	0.02	0.02	0.02
			Max	0.90	0.50	0.25	0.10
			S.D.	0.22	0.12	0.06	0.03
Little Pond Lower A	7/1/2013	9/22/2013	83.0	75.75	71.46	64.54	55.88
			Mean	6.89	3.76	2.08	1.43
			Min	0.08	0.04	0.04	0.04
			Max	27.75	26.83	22.46	22.29
			S.D.	9.66	7.00	4.82	3.88

Table IV-2. Chlorophyll summary for sonde deployments at Lower A (Bottom) in Little Pond for 2002 and 2013.

Chlorophyll Statistics for 2002 and 2013 Mooring Deployments								
			Total	>5 ug/L	>10 ug/L	>15 ug/L	>20 ug/L	>25 ug/L
			Deployment	Duration	Duration	Duration	Duration	Duration
	Start Date	End Date	(Days)	(Days)	(Days)	(Days)	(Days)	(Days)
Little Pond Upper	7/14/2002	8/11/2002	28.0	25.75	13.25	3.46	0.46	0.00
			Mean	1.61	0.29	0.19	0.11	N/A
			Min	0.04	0.04	0.04	0.08	0.00
			Max	11.75	1.83	0.63	0.21	0.00
			S.D.	2.82	0.35	0.17	0.06	N/A
Little Pond Lower A	7/1/2013	9/22/2013	83.0	76.42	44.46	21.58	9.04	3.42
			Mean	2.47	0.46	0.24	0.18	0.16
			Min	0.08	0.04	0.04	0.04	0.04
			Max	18.04	4.63	1.54	1.00	0.67
			S.D.	3.94	0.71	0.28	0.19	0.14

The differences in observed salinity from 2002 to 2013 (Figure IV-9) suggest that there may be occlusion of the tidal inlet leading to decreased flushing and slightly fresher water. However, statistical analysis of the depth records from the two deployments showed no difference in the range of the tidal excursions (Table 3, see highlighted regions). Closer examination of the tidal records with reference to the offshore tidal records provides evidence that the inlet was similarly occluded during both periods (Figure IV-11), as the tidal histograms show similar tide ranges in 2002 (left) and 2013 (right), as did the gauges deployed within Little Pond (Table IV-3). It is therefore more likely that the differences in salinity reflect differences in freshwater input in July and early August in the 2 years.

Table IV-3. Tidal ranges for moorings deployed in Little Pond for 2002 and 2013.

	Station		Mean	Std Dev	Range	Max	Min
Year	ID	# Obs.	(m)	(m)	(m)	(m)	(m)
2002	Lower A	2258	1.336	0.0649	0.41	1.559	1.149
2013	Upper B	2043	1.233	0.0732	0.423	1.482	1.059
2013	Upper A	2041	1.308	0.0653	0.421	1.56	1.139
2013	Lower A	2259	1.336	0.0649	0.41	1.559	1.149
2013	Lower B	2042	1.39	0.0651	0.717	1.64	0.923

**Tidal Statistics from Moorings** 



Figure IV-11: Histograms comparing NOAA tide predictions for Falmouth Heights with mooring deployed in 2002 and 2013.

**Conclusions of DO / CHLA Mooring Deployment:** The mooring study indicates that surface oxygen conditions were conducive to oyster survival and chlorophyll levels were generally high >10-15 ug/L providing ample food supply. Temperature and salinity are important factors to consider, particularly during transplant operations. Temperatures as high as 34°C and surface salinities as low as 14 ppt could severely stress immature oysters that are acclimated to conditions similar to offshore waters.

The data showed generally poor oxygen conditions and moderate to high chlorophyll levels throughout the transplant area. These conditions were present before the introduction of oysters to the pond and did not appear to change as a result of the oyster addition. In fact, during the last half of the deployment there appeared to be a decrease in phytoplankton abundance south of the oyster rafts suggesting that as the oysters grew larger and filtered more water that they may noticeablely reduce the water column burden of phytoplankton. One specific concern was that accelerated deposition of organic matter under the oyster rafts might adversely affect the benthos. No indication of this phenomenon was apparent in the sonde

records, though the poor water quality observed leaves little margin for detecting deleterious effects. Based upon the data collected the instrument placement was appropriate to detect any possible influence on water quality resulting from the oyster culture. It is likely that the benefits of oyster culture on water column clearance of phytoplankton suggested by the data would be enhanced by the introduction of more oysters or oysters of a larger size class. Any deleterious effect this may have on the benthic environment is still uncertain, so continued monitoring would be recommended. Intensive monitoring would also serve to validate the beneficial effects suggested by this pilot project.

Analysis of Turbidity in Little Pond: Turbidity is a measure of the water clarity and was determined at each of the 4 final mooring locations (Figure IV-12) using HOBO® Temperature/Light Pendant Data Loggers (UA-002-64). These light pendants were permanently positioned just below the waters surface (0.2m) and close to the bottom at a depth of 1m at each station. Data was compiled from July 2-October 2, 2013 for all sites except LP OYS2, which was added on July 9. There was a gap in data collection from August 8-15 due to technical difficulties and the compiled data reflects these time frames.



Figure IV-12 Site locations of the water quality monitoring (in red) and dissolved oxygen/chlorophyll moorings (in blue) for Little Pond.

The light pendants measured light intensity in units of  $\Box$  E/m2/s every 5 minutes and profiles were used to calculate the percent surface irradiance and the light extinction coefficient (k) within the water column. The light extinction coefficient, k was calculated using the Beer-Lambert Law which describes the logarithmic decay of light through a medium; the larger the light extinction coefficient, the more rapid the loss of light through the water column. In contrast, small light extinction coefficients reflect greater light transmission through the water column. The mean daily light intensity was also calculated using only the active photoperiod. In addition to the discrete light profile measurements, a water sample was collected 30cm from the bottom and filtered for total suspended solids, TSS. The site located within the oyster rack only included a YSI DO/chla sonde, at the surface. The corresponding water sample was also collected at the surface for chlorophyll and TSS.

The light characteristics using the light profiles and water quality samples taken at the moorings pertaining to Little Pond during the oyster pilot project are shown below (Table IV-4). The highest light penetration was located at the site closest to the oyster rack, LP OYS2 (Figure IV-12) and the light extinction coefficient was markedly lower at the same site closest to the oysters, indicating the oysters influenced the turbidity of the water (Figure IV-13). There was a distinctive linear trend of decreasing light extinction coefficients from the head to the mouth of Little Pond when excluding the site closest to the oyster rack (r2=0.99) giving a positive indication for the oysters improving the water clarity. The total suspended solids and total chlorophyll pigments do not however show a relationship with the light extinction coefficients or light penetration.

Table IV-4: Results from the continuous light profiles measurements from Little Pond showing the mean light intensity, penetration, and the light extinction coefficients along with the total suspended solids and total chlorophyll pigments at each station.

	Approxiamate				Beer-Lambert		
	distance		Light	Mean	Light extiction coefficient	TSS	Total Chlorophyll-a
Station	from head (ft)	Depth	Intensity (µE/m²/s)	Light Penetration	k	(mg/L)	Pigments (µg/L)
LP Upper A	1500	Surface	231.66				
		Bottom	63.69	27.69%	1.92	26.79	11.93
LP OYS2	1700	Surface	174.59				
		Bottom	55.22	32.83%	1.74	32.00	11.68
LP Oyster rack	1850	Surface	ND	ND	ND	20.68	20.78
LP Lower A	2050	Surface	217.73				
		Bottom	60.11	27.49%	1.82	14.17	14.08
LP Lower B	2750	Surface	208.33				
		Bottom	60.34	30.09%	1.64	31.24	9.68



Figure IV-13: Results showing mean light extinction coefficients calculated using the light profiles at each site in Little Pond.

Turbidity can be related to the tides, with the highest tides producing the largest water volume and thus more turbid waters, and the opposite concept being true for the lowest tides. The spring tides produce the strongest high and lowest low tides and occur during the full and new moons, with the extreme tides occurring on the new moon. Neap tides have the smallest amplitude high and low tides and occur during the first and third quarter moons. The tidal amplitude in Vineyard Sound in the vicinity of Little Pond is relatively small though, around 1.5 feet. By looking at the light intensity of the stations profiles, you can see the surface and bottom are correlated, but a relationship between turbidity and tides over the duration of the monitoring effort is unclear (Figure IV-14).



Figure IV-14: Light intensity time series for the site closest to the mouth of Little Pond, LP Lower B, in conjunction to the moon phase which reflects tidal amplitude.

Displaying a direct correlation of turbidity and tides in the entire data set is not concise enough to show a direct connection, but by using the extreme high tide and low tides during a new moon and a specific light profile can show the influence of tides on turbidity more closely. The new moon tides during this project occurred on July 8, August 6, and September 5, which coincided with extreme low tides between 5-6:00 a.m./p.m. and high tides between 11:30-noon. When plotting each station's light intensity and light penetration for these days it appears the photoperiod plays an over-arching role in light (Figure IV-15). There is about 25% light penetration to the bottom during the extreme low tides, but the most penetration occurs at 12:25 p.m. It is interesting to note though that there is a dramatic drop in light penetration from 11:50-12:20 when the sun is at its zenith, coinciding with the extreme high tide predicted for 12:05 a.m. for July 8. This illustrates that the extreme high water did reduce the light intensity and penetration, thus increasing the turbidity for a window of time. It is more difficult to pinpoint a decrease in turbidity with the extreme low tides occurring at 6:44 a.m. and 6:57 p.m. since the sunrise and set were so close to these times (5:43 a.m. & 7:52 p.m. respectively).



Figure IV-15: Light intensity and penetration during the extreme high/low spring tide of the new moon on July 8, 2013.

Another potential contributor to turbidity could be precipitation and wind strength and direction. A due south wind would drive boundary water from Vinevard Sound into Little Pond. During the summer, there is predominantly a southwest wind. During the duration of the Little Pond Oyster project there were several rain events (Table IV-5), but most occurring in the twilight or overnight hours when light measurements are low or zero. A substantial rain event occurred on July 26 where the effects of rain on turbidity could be explored, yet there was no direct and immediate relationship that shows an increase in turbidity (higher light extinction coefficient or decreased light penetration) with precipitation (Figure IV-16). The same trends were seen in all four stations and this could be due to lower photosynthetically active radiation (PAR) emitting from the sun that is typical on cloudy, rain events. By looking and the days preceding and following the storm event and the corresponding average PAR measurements, it can be seen that the average PAR makes a significant difference in the amount of light penetration possible (Figure IV-16). There was no rain recorded on July 25, but the average PAR was the similar on July 26 (274 & 273 umol/m2/sec respectively) when there was a rain event. In addition, both days had very similar light penetration (Figure IV-16), suggesting changes in turbidity here are more dependent on incoming light than rain and storm run-off. The average PAR measurements were measured at the NERRS meteorological station in Waquoit Bay.

Table: IV-5: Date and time of rain events and corresponding precipitation and wind characteristics from a meteorological station in Waquoit Bay supplied by the NOAA NERR Centralized Data Management Office http://cdmo.baruch.sc.edu/get/export.cfm from July-October 2013.

	Data	Timo	Duration	Cumulative Procingtation (mm)	Wind Speed (m/s)	Max Wind (m/s)	Average Wind	Avera	ge Wind
	Dale	Time	Duration	Frecipatation (IIIII)	Speed (III/S)	winu (iii/s)	Direction (degrees)	(caruma	
	7/1/2013	14:45	0:15	13.7	1.6	4.7	222	SW	<b>×</b>
7	7/10-7/11/13	23:45-0:15	1:30	2.5	1.3	4.7	249	WSW	<b>X</b>
	7/21/2013	4:45-6:00	1:15	14.2	1.1	2.6	230	SW	1
7	7/25-7/26/13	22:30-17:45	19:15	15.7	2.4	5.9	187	S	
	7/28/2013	2:30-4:30	2:00	5.3	0.8	2.1	112	ESE	•
	8/13/2013	11:30-14:30	9:15	2.8	0.7	2.1	166	SSE	•
	8/18/2013	17:45-20:45	3:00	1.1	0.3	0.9	118	ESE	
	8/27/2013	5:00-10:00	5:00	16.5	0.5	1.5	185	S	<b>†</b>
	9/1/2013	1:15-2:15	2:00	14.7	1.9	6.3	238	SW	ѫ
1	9/3/2013	15:45-19:30	3:45	29.5	0.8	2.4	177	S	<b>↑</b>
	9/13/2013	4:30-9:30	5:00	8.9	0.9	2.7	251	WSW	×



Figure IV-16: Light extinction coefficient relative to the cumulative precipitation during the July 26 and 28 rain events at the Little Pond station LP Lower B (umol/m2/sec).

*Water Temperature:* Temperature was measured every 5 minutes using the HOBO temperature and light pendants and averaged from July 2-October 2, 2013 for all sites except for LP OYS2, which was added on July 9, 2013. There was a gap in the data collection from August 8-15, 2013. The temperature was averaged for each station and depth reflecting the above time frames (Table 4.6 below). Light sensors were not deployed at the surface Oyster Rack station.

Station	Depth	Temperature °C
LP Upper A	Surface	25.01
LP Upper A	Bottom	24.57
LP OYS2	Surface	24.83
LP OYS2	Bottom	24.46
LP Lower A	Surface	25.02
LP Lower A	Bottom	25.54
LP Lower B	Surface	24.69
LP Lower B	Bottom	24.42

## Section V: Stormwater Sampling

Stormwater is an intermittent contributor of nitrogen to estuaries depending on the watersheds to the stormwater discharge points, the intensity of storms, the type of land use, and the treatment design in the stormwater structures. Depending on these factors, stormwater can be an important component of the overall nitrogen budget of a pond. In order to address this potential nutrient input, CSP-SMAST staff undertook a preliminary survey of the Naragansett Street stormwater pipe to ascertain if the discharge is a significant local point source.

CSP scientists determined and measured first-flush stormwater runoff flow at the identified discharge point during a minimum of three (3) storm events. Per the scope of work, measurement of storm events were to be focused on the summer months (as possible) to best gauge nitrogen impacts during the most active ecosystem period. Ultimately, based on rainfall conditions during the summer 2013, two events were sampled during the summer (July and August) and one storm event was sampled in October.

First-flush runoff, typically, contains the majority of the contaminant load associated with stormwater runoff. First-flush determination included determining pre- and post-storm flows. Measured flows were compared to storm precipitation volumes determined from nearby local sources. It is important to note that the stormwater pipe was examined during the proposal development process and found to not be flowing, indicating that there is not likely to be any base flow discharge as would be the case with inflow/infiltration to the pipe. As such, the storm drain pipe outlet (Figure V-1) only flows during storm events, and this was confirmed by the project team. Additionally, the discharge end of the pipe was confirmed to be fully exposed during at least 2/3 of any given tidal cycle thus allowing storm samples to be taken without the influence of tidal waters mixing with stormwater in advance of collection. Collected stormwater samples were analyzed for standard MEP nitrogen constituents, including total nitrogen, NH<sub>4</sub>, NO<sub>3</sub>+NO<sub>2</sub>, DON and PON, as well as ortho-phosphorus and total phosphorus. Data was retrieved by CSP/SMAST staff in accordance with accepted quality control and quality assurance procedures utilized by the MEP.



Figure V-1 Narragansett Street Stormwater Drain discharge into Little Pond fully exposed 2/3 of the tidal cycle.
**Stormwater Sampling Results (2013):** Storm water can adversely affect water quality. Storm drains collect water from larger surface areas and channel those waters into point source discharges in receiving waters. One storm drain discharges into Little Pond. The outlet to Little Pond, located near the high tide line at the end of Narragansett Road, is the end of a series of interconnected storm drains and dry wells (Figure V-2). Storm water flows into each of the drains, but also flows between them with delays at each drain as the dry well sumps fill before overflowing into the connecting pipes.

To assess the impact of the storm drain on nutrient related health of Little Pond and the viability of oyster propagation in the pond a HOBO accelerometer (to determine when the drain was flowing) was mounted on a PVC pipe in such a way that it could rotate freely along one axis when deflected by flowing water. Rainfall measurements by the Waquoit bay Estuarine Reserve (WBNERR), located 6.2 km away, were used to estimate rainfall amounts at Little Pond (Figure V-3). Unlike the WBNERR data rainfall was not summed for calendar days, but rather for discreet events. Measurable rainfall separated by more than 5 hours was considered an unique event.



Figure V-2. Schematic of storm drain configuration and storm water flow direction. Storm drains 1-4 have large dry wells which delay water flow to pond. HOBO accelerometer hung free to move with storm water approximately 5 mm from the drain pipe exiting storm drain 4 towards the Little Pond outlet.



Figure V-3. Cumulative rainfall record (15 minute intervals) from Waquoit Bay Estuarine Reserve Weather Station located 6.2 km NNE of Little Pond outlet shown in red for the entire deployment record. In black is HOBO accelerometer record (5 minute interval). 1G corresponds to the accelerometer hanging vertically. Deviations below 1 G indicate deflection from vertical in response to water flow and/or debris.

The HOBO accelerometer (HOBO Pendant G) logged data every 5 minutes and is plotted with rainfall data in Figure 2. By convention 1 G corresponds to the logger being vertical. Lower G measurements reflect deflection of the logger from vertical. Water striking the side of the logger during periods of flow deflected the logger from vertical and produced measures less than unity. However, we found that debris in the storm drain sometimes did not allow the logger to return to a vertical position and the apparatus was frequently visited by an opossum that lived in the storm drain when it was not flowing.

The interconnected storm drains and dry wells, and their variable infiltration rates, made direct correlation of rainfall and storm drain flow impossible. However, analysis of discreet events did allow us to place bounds on conditions necessary for flow to the pond. Details of 4 rain events spanning the range of precipitation amounts are shown in Figure V-4. Typically at least 3 mm of rain had to fall before storm flow was observed. In addition, the rate of rainfall had to meet or exceed 2 mm/hour. Onset of flow was rapid once these conditions were met and flow terminated within 30 minutes of when rainfall stopped.



Figure V-4. Plots of cumulative rainfall (red) and HOBO accelerometer response (black) for 4 discreet rain events throughout for the range of total rainfall time courses observed.

Flow measurements were attempted on each of the three dates that samples were collected (Table V-1). However high tides and shallow laminar flow required many temporary modifications of the drain outlet before flow was finally measured on October 8. Cinder blocks and expanding insulation was used to create a 7.5 cm wide flume so that adequate depth of

flowing water was available for velocity measurements. Flow on October 8 was estimated at 660L/h and lasted just under 1 hour. This flow rate also appears to represent the detection limit of the HOBO logger as no deflection was observed.

Compared to estimates presented in the Little Pond MEP report, the TN concentration associated with this event was very close at 1.8 mg/L TN versus the estimated 1.5 mg/L for all impervious surfaces. The highest recorded concentration for the storm drain was 4.3 mg/L on 8/13. Both of these numbers, however, represent the highest concentrations possible for initial flow from the storm drain.

Storm water concentrations of nutrients reflect the relatively stable concentrations found in rain water as well as the accumulation of organic matter in the drain system (and its decay) between storms. The relationship between nutrient concentrations and the length of time since last rainfall did show a positive relationship (longer interval, higher concentration), but the small number of samples precluded statistical analysis. The number of days since the last rainfall, 0.9, 3.8, 4.3 days were associated with events on 10/8, 8/13 and 7/26, respectively.

Nitrogen loading to Little Pond from impervious surfaces was estimated at 510kg/y total, with half (249 kg/yr) within 10 yr travel time to the estuary (from MEP Report). This latter number is most comparable to the storm drain study results discussed here. The storm drain discharge load from the single event on 10/8 was estimated at about 1 gram (0.997gr TN; 8mm rainfall, volume=660L). The MEP uses different source strengths for roadways and driveways, yielding TN loads, based on the 660L measured flow, of 0.99 and 0.50 grams, respectively. It is not possible to determine the amount of flow from driveways versus roadway in this study as most water did not exit through the pipe (see below). In addition, the storm drain accumulates detritus and animal waste (animals were observed living in the Narragansett Street piping), which add to the TN load. None-the-less, the measured load (0.997 gram) fits quite well with the MEP estimates (0.99 – 0.50 gram).

More important, the storm drain system is infiltrating most of the water which enters it. We estimated the impervious area feeding water to the Narragansett Street storm drain system at 4,734 m<sup>2</sup> or 1.17acres. This area includes roadways, driveways, and parking areas on Narragansett as well as road ways extending 50 meters from the intersection of Narragansett Rd. and Maravista Rd. During an 8 mm rain event, the outflow from the drain should be ~38,000 L or more than 50 times the measured flow. This results from infiltration basins within the storm drain system designed to infiltrate rainwater rather than just divert it from the roadway into the pond. So while the load per m2 is consistent with the MEP results, the load to the pond from the entire drain system could not be measured for comparison to the MEP results. While the storm drain does prevent flooding of the roadway and resident's yards during rain events, the storm drain does not appear to represent a major source of nitrogen to Little Pond.

		PO4	TP	NH4	NOx	DIN	DON	TN	POC	PON	TON
Date	TIME	(uM)	(uM)	(uM)	(uM)	(uM)	(uM)	(uM)	(uM)	(uM)	(uM)
7/26/2013	0:45	6.58	16.23	26.28	42.95	69.23	68.23	204.7	1043.3	67.13	135.35
8/13/2013	15:40	11.63	15.97	74.94	81.78	156.72	122.12	309.0	430.3	30.12	152.23
8/13/2013	15:50	10.26	12.45	66.37	77.29	143.66	115.49	293.3	468.6	34.12	149.61
10/8/2013	0:35	5.54	9.53	12.78	43.51	56.29	57.96	148.8	491.1	34.56	92.52
10/8/2013	0:50	5.96	9.19	12.15	46.22	58.37	49.42	129.6	292.0	21.85	71.27

Table V-1. Constituents and concentrations of first flow from storm drain into Little Pond.

## Section VI: Benthic Animal Community Analysis

**Overview of Biological Health Indicators:** There are a variety of indicators that are used in concert with water quality monitoring data for evaluating the ecological health of embayment systems. The best biological indicators are those species which are non-mobile and which persist over relatively long periods, if environmental conditions remain constant. The concept is to use species which integrate environmental conditions over seasonal to annual intervals. This approach is particularly useful in environments where high-frequency variations in structuring parameters (e.g. light, nutrients, dissolved oxygen, etc.) are common, making adequate field sampling difficult.

As a basis for a nitrogen threshold determination (assimilative capacity of the estuary) such as that developed by the Massachusetts Estuaries Project, ecological assessment focuses on major habitat quality indicators: (1) bottom water dissolved oxygen and chlorophyll-*a*, (2) eelgrass distribution over time, and (3) benthic animal communities. Dissolved oxygen depletion is frequently the proximate cause of habitat quality decline in coastal embayments (the ultimate cause being nitrogen loading). However, oxygen conditions can change rapidly and frequently show strong tidal and diurnal patterns. Even severe levels of oxygen depletion may occur only infrequently, yet have important effects on system health. To capture this variation, dissolved oxygen sensors are deployed at strategic locations throughout the system under investigation. During the MEP analysis of Little Pond, DO moorings were deployed within the upper portion of the Little Pond system, as well as closer to the inlet to Little Pond, to record the frequency and duration of low oxygen conditions during the critical summer period. These stations were located above and below the sentinel station established in Little Pond for setting the threshold nitrogen concentration. These historic MEP mooring deployments were mirrored and positioned in a similar manner relative to the oyster racks deployed in Little Pond.

In areas that did not support eelgrass beds, benthic animal indicators are typically used to assess the level of habitat health from "healthy" (low organic matter loading, high D.O.) to "highly stressed" (high organic matter loading-low D.O.). The basic concept being that certain species or species assemblages reflect the quality of their habitat. Benthic animal species from sediment samples are identified and the environments ranked based upon the fraction of healthy, transitional, and stressed indicator species. The analysis completed in Little Pond and other estuaries across southeastern Massachusetts is based upon life-history information on the species and a wide variety of field studies within southeastern Massachusetts waters, including the Wild Harbor oil spill (Hampson, 1978), benthic population studies in Buzzards Bay (e.g. Hampson, 1989) and New Bedford (SMAST, unpublished data), and more recently the Woods Hole Oceanographic Institution Nantucket Harbor Study (Howes *et al.* 1997). These data are coupled with the level of diversity (H') and evenness (E) of the benthic community in a given system and the total number of individuals to determine the infaunal habitat quality for that system (e.g. Little Pond).

**Ecological Health of the Little Pond System:** As mentioned in more detail above, the nutrient related ecological health of an estuary can be gauged by the nutrient, chlorophyll, and oxygen levels of its waters and the plant (eelgrass, macroalgae) and animal communities (fish, shellfish, infauna) which it supports. For the Little Pond embayment system, the overarching MEP assessment that yielded the system specific nitrogen threshold was based upon data from the water quality monitoring database, surveys of eelgrass distribution, benthic animal communities, sediment characteristics, and dissolved oxygen records conducted during the summer and fall

of 2003. These data form the basis of an assessment of the system's health which serves as a baseline point of comparison to more recent (2013) benthic surveying conducted as part of the monitoring for the oyster aquaculture pilot project.

Under the Massachusetts Estuaries Project (Little Pond data collection 2003, MEP analysis completed in 2006), quantitative sediment sampling was conducted at 8 locations throughout the Little Pond System (Figure VI-1). In some cases multiple assays were conducted. In all areas and particularly those that do not support eelgrass beds, benthic animal indicators were used to assess the level of habitat health from healthy (low organic matter loading, high D.O.) to highly stressed (high organic matter loading-low D.O.). The basic concept is that certain species or species assemblages reflect the quality of the habitat in which they live. Benthic animal species from sediment samples are identified and ranked as to their association with nutrient related stresses, such as organic matter loading, anoxia, and dissolved sulfide. The analysis is based upon life-history information and animal-sediment relationships (Rhoads and Germano 1986). Assemblages are classified as representative of healthy conditions, transitional, or stressed conditions. Both the distribution of species and the overall population density are taken into account, as well as the general diversity and evenness of the community.

It should be noted that, given the loss of eelgrass beds, the Little Pond System (at the time of the MEP data collection, 2003) was showing clear signs of nutrient related impairment from over enrichment by nitrogen. However, to the extent that the Little Pond system can still support healthy infaunal communities, the benthic infauna analysis is important for determining the level of impairment (moderately impaired → significantly impaired → severely degraded) as well as serve as a bench mark for long term improvement of habitat quality from the implementation of nitrogen management alternatives such as shellfish aquaculture. The 2003 benthic infaunal survey completed by the MEP was critical to the establishment of site-specific nitrogen thresholds. A summary of the MEP benthic infaunal analysis is provided below.

**Summary of MEP Benthic Infaunal Analysis (2003):** In the MEP assessment, analysis of the evenness and diversity of the benthic animal communities was used to support infaunal density data and the natural history information. The evenness statistic can range from 0-1 (one being most even), while the diversity index does not have a theoretical upper limit. The highest quality habitat areas, as shown by the concomitant oxygen and chlorophyll records and eelgrass coverage (MEP 2003), have the highest diversity (generally >3) and evenness (~0.7). The converse is also true, with poorest habitat quality found where diversity is <1 and evenness is <0.5.

The MEP 2003 infauna study was fully consistent with the nitrogen data and oxygen and chlorophyll-a records collected in the summer of 2003 and the loss of eelgrass from this system, showing nitrogen related habitat impairment and only a relatively modest gradient between the upper and lower reaches of the overall system. The infaunal community data indicated that based on the 2003 data, Little Pond was moderately to severely degraded throughout its estuarine reach (Table VI-1). The Lower basin stations (LP-6, 7, 8) nearest the tidal inlet showed generally higher numbers of individuals, higher diversity and evenness than the upper estuarine reach stations (LP-2,3,4). On average, the lower reach had more than 2 times the individuals (615 vs. 257) distributed among 1.7 times the species resulting in a more productive and diverse community. The evenness scores appear similar, however, it relates more to the small number of species rather than to the habitat quality in the comparison. While the trend is clear, it is important to note that these indicators from the lower basin were much lower than indicative of higher quality systems where diversities generally range from 2.5 to 3 and above and evenness is generally >0.7. Equally important, the dominant species in both the upper and lower reaches were indicative of poor quality habitat. The infaunal populations throughout the

Pond in 2003 were dominated by small polychaete worms, specifically Capitella and Streblospio. These species are typical of highly organically loaded sediments. Their life histories allow them to persist in a stressful environment. The lack of larger longer-lived species was equally supportive of the conclusion that Little Pond was a significantly impaired system based on the 2003 infaunal survey.

Table VI-1. Benthic infaunal community data (MEP data from 2003) for the Little Pond embayment system. 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 0.018 m2).

Sub-		Total Actual	Total Actual	Species Calculated	Weiner Diversity	Evenness			
Embayment	Location	Species	Individuals	@75 Indiv.	(H')	(E)			
Little Pond System									
	Sta. 1	8	697	5.13	1.21	0.40			
	Sta. 2	6	227	3.99	1.08	0.42			
	Sta. 3	4	216	3.82	1.59	0.79			
	Sta. 4	3	328	2.99	0.81	0.51			
	Sta. 5	3	696	2.80	0.37	0.24			
	Sta. 6	7	668	5.14	1.30	0.46			
	Sta. 7	8	560	5.20	1.30	0.43			
	Sta. 8	7	616	6.32	1.57	0.56			



Figure VI-1. Aerial photograph of the Little Pond system showing location of MEP (2003) benthic infaunal sampling stations (red symbol).

**Benthic Infaunal Analysis (2013 Oyster Aquaculture Pilot):** Changes in the Little Pond ecosystem due to the oyster culture may impact the benthic community, which was a key measure of ecosystem status during the MEP assessment. In order to more holistically measure the impact of the oyster culture demonstration, CSP-SMAST assessed and quantified the impacts of the oyster culture on the benthic community in the oyster aquaculture area.

The benthic animal community throughout Little Pond (including where the oysters were deployed) were characterized and quantified per the scope of work for the oyster monitoring project. The locations were selected during an initial field reconnaissance, and the following spatial pattern was chosen:

- 2 sites above the oyster deployment site,
- 2 sites within the oyster deployment area,
- 2 sites below the oyster deployment area.

This plan placed 2 up-gradient and 2 down-gradient sampling sites near the benthic infaunal sampling sites previously established by the MEP in 2003 (Figure VI-2). This placement was important since both the up-gradient and down gradient sites showed relatively poor animal habitat in 2003 and any potential assignment of impacts due to the oyster deployment must be made in that light.

Consistent with sampling undertaken by the MEP (for cross comparison purposes) samples collected in 2013 were obtained using a Young modified Van Veen grab at each site. Analysis included both the species and numbers of individuals per species within each sample. Key metrics such as Weiner Diversity and Evenness were also calculated and directly compared to the MEP 2003 benthic infauna analysis completed in Little Pond prior to this oyster cultivation project. Benthic communities were evaluated relative to the total nitrogen – species numbers relationship developed for Cape Cod estuaries by CSP-SMAST, as well as traditional established indicators. All procedures followed the MEP analytical approach and the USEPA/MassDEP approved MEP QAPP developed by CSP-SMAST.



Figure VI-2. Benthic infauna station maps for pre (MEP 2003) and post (2013) oyster project in Little Pond, Falmouth, MA.

### Benthic Animal Community Results (2013):

The 2013 infauna results (Tables VI-2 and VI-3) document a nutrient impaired habitat for benthic infauna throughout Little Pond. This is consistent with the 2013 measured hypoxia throughout all pond basins and the high total nitrogen, phytoplankton biomass (chlorophyll a) and particulate organic matter in pond waters. The observed infauna communities are also consistent with the gradient of declining water quality from the tidal inlet to the upper tidal reach of Little Pond. Although the 2013 results show a potential improvement in habitat quality from 2003 to 2013, based on numbers of species and individuals and diversity, conditions remain poor. The increase in species was due to the presence of only a few rare species and the increase in numbers was due the dominance of opportunistic species dominate the community frequently, accounting for more than 80% of the population. These species are generally small polychaete worms, specifically Capitella and Streblospio (as in 2003) and are typical of highly organically loaded sediments. Their life histories allow them to persist in a stressful environment. The obverse was also found, i.e. longer-lived larger species were absent. Based on both findings it is clear that Little Pond remains a significantly impaired system.

Nonetheless, the 2013 data were examined for any potential response to the oyster deployment and none could be documented. Upon initial examination, there is a depression in habitat quality in the region associated with the deployment, however, this depression in species numbers and number of individuals was also seen in the 2003 survey conducted by the MEP (see above). As such, a negative oyster effect cannot be concluded based on infauna habitat analysis. The difficulty in determining a negative or positive effect of oysters on benthic animal habitat in Little Pond appears to result from the very poor quality of the habitat at present. As the pond improves or perhaps, in a higher quality environment, it is likely that effects would be more evident.

Station			Species	Weiner	Evenness					
Location Species		Individuals	@75 Indiv.	(H')	(E)					
Little Pond Embayment System										
LP1.1	11	975	5	1.380	0.399					
LP1.2	7	573	4	1.009	0.359					
LP Oys Rack	7	333	5	1.482	0.528					
LP2	9	203	8	2.116	0.668					
LP2.2	9	1176	5	0.982	0.310					
LP3	14	913	9	2.471	0.649					

Table VI-2. Benthic infaunal data collected for the 2013 oyster demonstration project.

Table VI-3. Comparison of benthic infaunal data collected in 2003 for the MEP and 2013 data collected for the oyster demonstration project.

				_	_	Weiner	Weiner	_	_
		Total Actual	Total Actual	Total Actual	Total Actual	Diversity	Diversity	Evenness	Evenness
Location ID	Location ID	Species	Species	Individuals	Individuals	(H')	(H')	(E)	(E)
2003	2013	2003	2013	2003	2013	2003	2013	2003	2013
Little Pond Estuary									
LP3	LP1.1	4	11	216	975	1.59	1.38	0.79	0.399
LP4	LP2	3	9	328	203	0.81	2.12	0.51	0.668
LP5	LP2.2	3	9	696	1176	0.37	0.98	0.24	0.310
LP6	LP3	7	14	668	913	1.30	2.47	0.46	0.649

## **Section VII. Conclusions**

#### **Conclusions:**

Overall, the water quality results clearly document that the deployment of oysters in Little Pond did produce modest improvements in the nearfield to the deployment area. The oysters appear to have removed phytoplankton (lower chlorophyll a, POC and PON) resulting in greater water clarity. That this was from feeding activities was seen in the increase in pheophytin within the deployment area, which is the result of the degradation of phytoplankton chlorophyll a to pheophytin a. The decrease in chlorophyll a and increase in water clarity were also documented by time-series measurements from moored instruments at 4 sites (Section IV). The removal of particulates resulted in a decrease in water column TN, as well. The moderate stratification of the water column focused the "oyster effect" in the surface layer where they were deployed. Comparisons between 2013 (ovster deployment) and 2012 (no ovster deployment) indicated that it was the oysters not natural gradients that were producing the observed effects in the region nearfield to the deployment area. While there were initial theoretical concerns over the oysters negatively impacting oxygen dynamics, hypoxia preceded the oyster deployment, the oysters were effectively separated from the bottom water by the moderate stratification and the pattern of bottom water oxygen suggested that the oysters may have had a positive effect (if any) on oxygen conditions. Based on the water quality monitoring, the oyster deployment was not found to have any negative ecological impacts on Little Pond, but was found to improve water quality conditions and therefore potentially associated habitat quality. However, the effects were localized and placed upon the background conditions of a eutrophic shallow estuary.

Chlorophyll concentrations decreased somewhat during the latter part of the mooring deployment period in Little Pond. Most interesting, with respect to the oyster transplants, was that once all the oysters were placed in the pond there appeared to be a decrease in the amplitude of phytoplankton blooms downstream of the rafts. Although we cannot ascribe causation at this time the observation is consistent with the hypothesis that oyster culture would tend to remove phytoplankton from the water. There was, however, no clear relationship between decreased phytoplankton and elevated bottom water dissolved oxygen. Little Pond represents a shallow, eutrophic estuary in which sediment respiration makes a large contribution to the overall water column oxygen budget, so if the observed decrease in phytoplankton density downstream from the oyster rafts turns out to be causally related it may take several years before the decreased organic load to the sediment is reflected in the water column oxygen concentrations.

It is clear that the oxygen conditions were showing depletion at both the surface and bottom, but the observed depletions did not appear to be related to the oysters that have been placed in the pond. Conditions were poor before oyster introduction and either remained poor or improved throughout the summer. Oxygen conditions were no worse within the oyster culture area (Upper A (Bottom)) than anywhere else, in fact better than in some areas, suggesting that accelerated deposition of fecal pellets below the rafts was either not an important factor or that the observed poor water quality left little margin for detecting deleterious effects.

Through the deployment of near continuous recording of light penetrating the water column it was possible to determine the light characteristics using the light profiles and water quality samples taken at the moorings in relation to the oyster pilot project. The highest light penetration was located at the site closest to the oyster rack, LP OYS2 and the light extinction coefficient was markedly lower at the same site closest to the oysters, indicating the oysters

influenced the turbidity of the water. There was a distinctive linear trend of decreasing light extinction coefficients from the head to the mouth of Little Pond when excluding the site closest to the oyster rack ( $r^2$ =0.99) giving a positive indication for the oysters improving the water clarity. The total suspended solids and total chlorophyll pigments did not however show a relationship with the light extinction coefficients or light penetration. It is important to note that additional measurements should be done in this regard and relative to turbidity and the racks as decreases in turbidity can be associated with meteorological events (strong wind) as well as strong tides (spring) which can mask the positive effects the oysters maybe having on water clarity. Initial measurements indicate that rain did not have a measureable effect on turbidity.

In regard to the flow and TN-load measurements completed for the stormwater discharge to Little Pond, nitrogen loading to Little Pond from impervious surfaces was estimated at 510kg/y, whereas the load from the single event on 10/8 was estimated at just under 1 gram (0.997 mg TN). From this data it seems likely that the loading to the pond from the storm drain is both within the average estimates for the watershed and insignificant relative to the attenuated total load to the pond of 7569 kg/y. No further investigation is being recommended relative to this specific stormwater outfall.

In 2013, based on infaunal sampling undertaken in conjunction with the deployment of the Oyster aquaculture rafts, opportunistic species dominate the community frequently with accounting for more than 80% of the population. Consistent with results from the 2003 infaunal survey undertaken in Little Pond by the MEP, these species (2013) are generally small polychaete worms, specifically Capitella and Streblospio and are typical of highly organically loaded sediments.

None-the-less, the 2013 infaunal data were examined for any potential response to the oyster deployment and none could be documented. At initial examination, there is a depression in habitat quality in the region associated with the deployment, however, this depression in species numbers and number of individuals was also seen in the 2003 survey conducted by the MEP. As a result a negative oyster effect cannot be concluded based on infauna habitat analysis.

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# Appendix 1.

Individual Plots of Temperature, Salinity and Depth











Date





















Date