Rhode Island Oyster Restoration
Minimum Monitoring Metrics and Assessment Protocols

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Prepared for the Rhode Island Shellfish Technical Working Group

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1.0 Introduction

The eastern oyster, *Crassostrea virginica,* is an economically important bivalve mollusk and a key component of our coastal ecosystem. Oysters were once the primary fishery of Narragansett Bay during the early 1900's, with annual landings exceeding 14 million pounds (DeAlteris *et al.* 2000). Overharvest, habitat and water quality degradation coupled with the spread of disease has depleted our local and regional stocks to near extinction (Beck *et al.* 2011).

Federal, State and local non-profit organizations have long recognized both the ecological and socioeconomic importance the oyster represents to Rhode Island. Oyster restoration programs in RI date to the early 1900's (Rice *et al.* 2000), and have been making considerable progress and gaining popularity in the past decade. Oyster restoration has been implemented by federal, state, academic and non-governmental agencies, with cooperation and involvement from both the wild-harvest and aquaculture industries. Increasingly, the general public's interest in oyster restoration in the state has grown, and the recent trend suggests that oyster restoration in the state is likely to remain a high priority, involving most stakeholders mentioned above.

Despite the increase in shellfish restoration activities in Rhode Island, careful monitoring of the restored populations and associated habitat often takes a back seat to efforts of introducing shellfish into estuaries. Although the lack of stringent monitoring of restoration programs is not a local phenomenon (Luckenbach & Coen 2003; Bernhardt *et al.* 2005), monitoring is a fundamental part of the restoration process and therefore a recommended requirement for all Rhode Island oyster restoration projects. Without adequate monitoring, it is difficult to discern the individual project performance or ecological impact of a project (Brumbaugh *et al.* 2006), thus making it problematic to learn from mistakes or progress towards more efficient restoration methods. Increased systematic monitoring of shellfish restoration allows for informed project selection, adaptive management, and technique selection, ultimately improving restoration practices.

The Rhode Island Shellfish Technical Working Group (RISTWG) is a volunteer, advisory council to the Rhode Island Coastal Resource Management Council that was created to provide a framework for coordination and communication between the agencies and groups involved in various shellfish restoration activities. The RISTWG is represented by federal, state, NGO, wild-harvesters, and aquaculturists, acting as a centralized body commenting on Rhode Island shellfish restoration activities and collaboratively working together to further state-wide shellfish restoration planning, prioritization and goal setting. This guide does not attempt to describe habitat selection or provide information on restorable areas. Restoration practitioners are encouraged to seek advice from the RISTWG on these issues prior to embarking on an oyster restoration project. This document, developed by the RISTWG, is meant to serve as a guide to identify minimum monitoring metrics for *all* oyster restoration projects that

are implemented in state waters and suggest protocols for systematic, standardized monitoring for restoration practitioners in Rhode Island.

Oyster restoration in Rhode Island is aimed at building self-sustaining populations; therefore monitoring priorities should focus on documenting changes in the general population structure, e.g. survival, growth rates, recruitment and health status (disease prevalence). While more advanced monitoring and research can answer important questions regarding shellfish restoration, it is generally not practical or necessary to collect such data at all restorations sites. The metrics described in sections II and III of this guide are suggested as the minimum physical and biological monitoring requirements used on *every* project by which to gauge project-level performance. Some restoration projects will require more intense monitoring to address specific issues, ancillary goals and/or research questions. Examples of this level and type of monitoring, which in some instances may allow for increased evaluation of ecosystem services provided or advance the science of shellfish restoration, are provided section IV, but are not meant to be an exhaustive list. The minimum metrics described herein should not limit additional monitoring and research activity. The RISTWG recognizes that future state-wide shellfish restoration efforts and broader restoration activities will help guide and further refine state-wide restoration goals and priorities.

II. Pre-Restoration

2.0 Baseline Survey

Collecting baseline data before any restoration treatments are established is important in order to decipher changes in population structure and habitat during and after restoration (Brumbaugh *et al.* 2006). Baseline monitoring metrics will vary depending on the goals of the restoration project but at a minimum the following should be employed.

Oyster Density - Abundance: Monitoring baseline oyster density will provide a quantitative comparison of abundance before and after restoration. Oyster abundance will be standardized to number of individuals per m^2. A common method of collecting this information involves the use quadrat sampling (Luckenbach & Ross 2003; Hancock *et al.* 2005, 2006, 2007; Brumbaugh *et al.* 2006; DeAngelis *et al.* 2008, 2009) and is described in section 3.1.

Recruitment: Monitoring oyster recruitment to the site prior to restoration may provide insight to post-restoration recruitment success as well as a comparative index of recruitment before and after restoration. It may also provide valuable information regarding type and method of restoration to be performed. For example, lack of recruitment may warrant addition of broodstock to the site. On the contrary, if the system is not recruitment limited substrate enhancement may be a more appropriate tool for restoration. Monitoring recruitment can be accomplished by either the use of artificial spat collectors as described in section 3.3 or monitoring natural recruitment within the quadrats sampled for

oyster abundance (sections 3.1 & 3.2). While more time consuming, the use of artificial spat collectors may provide a more robust analysis of larval availability in the system of interest.

Disease: High prevalence of disease can affect the health of the population, decreasing reproductive effort and/or causing mortality, thus reducing success of restoration efforts. If oysters are present at the site of restoration it is prudent to obtain data on presence of disease and pathogen loads. If funding is limited and samples are not attainable it may be possible to contact local shellfish pathologists (see Appendix A) for historical information on disease prevalence at the location of interest.

III. Essential Monitoring

Essential monitoring is focused on documenting changes in the general population structure to gauge project-level performance. The metrics described in this section are suggested as the minimum physical and biological requirements to be collected on all projects.

3.0 Site Description

A comprehensive description of the site prior to restoration and at each monitoring event will provide a platform for comparative analysis of the physical and, if available, chemical characteristics between sites. It may also provide understanding of site specific performance of oyster populations. This information can be helpful to practitioners in deciding best habitats for future site selection. The following characteristics should be routinely monitored throughout the duration of the restoration project and during long term monitoring.

Physical and Chemical Characteristics of H_20: Physical and chemical conditions of a restoration site can be influencing factors on project success. Point data is typically not robust enough to analyze the effect on bivalves, and developing a large temporal data set of these characteristics can be cost and time prohibitive. Nonetheless, practitioners should attempt to retrieve as much of this data as possible, including: temperature, salinity, dissolved oxygen, pH and chlorophyll a. In recent years, temperature loggers have become increasing affordable, and are encouraged to be deployed at restoration sites. Practitioners are encouraged to communicate with environmental and community groups in an effort to utilize potential water quality and chemical data (e.g. Salt Ponds Coalition, Narragansett Bay Estuary Program, Watershed Watch, See Appendix B). All data should be converted to metric units.

Benthic Substrate & Depth: Site descriptions should also include information regarding the substrate the oyster bed was created on and dominant macro-algae cover. Benthic substrate types can be categorized based on grain size and/or composition (i.e. mud, sand/silt, sand/cobble, rocky). Percentages of substrate and macro-algae coverage should be recorded. This can be accomplished during quadrat sampling (section 3.1) by estimating percent composition of substrate type and macro algae within each quadrat sampled. To achieve consistency between practitioners, standard cover classes can be assigned

that provide a range of percent cover within each quadrat. Once the percent composition of substrate or algae is determined, as depicted in Appendix C, the midpoint value should be recorded (adopted from Carlisle *et al.* 2004). It is necessary to record the size of the quadrat which sample estimates are derived from.

If substrate enhancement was practiced prior to or during restoration the following parameters should be recorded: 1) date deployed and form of substrate used (i.e. surf clam shell, oyster shell, limestone marl, Reef ball TM, etc.), 2) clutching density (vol. material m^{-2}), 3) vertical relief of substrate and 4) total area (m^2) enhanced. Depth of the seeded site should also be included in the site description. This should describe whether the site is subtidal, intertidal or both, as well as water depth at mean low tide (m).

Seeding History: Seeding history is important to allow practitioners to survey mortality and growth of the cohort, particularly during the first year. Seeding history should include: date and number of animals seeded, average size of seed (mm) and planting density (# oysters m^{-2}). The origin of oysters should also be recorded as this can be useful information for tracking disease resistance and performance of a given genetic line.

Location & Footprint: Describing the general location of the restoration site is a straight forward exercise of providing the latitude and longitude of the center point and corner boundaries of the designed restoration area, as well as a common name of the body of water. If multiple oyster beds exist within the restoration site, each bed should be assigned a unique identifier and latitude and longitude recorded for each bed. All coordinates should be provided in decimal degree (DDD.DDDD) format (e.g. Bissel Cove, Bed 1: 41.54615 N, 71.42942 W).

Figure 1. Yellow dots depict discrete oyster beds; green polygon depicts large seeded area with even distribution.

It is important to accurately measure the area of the seeded footprint, as this is critical to estimating total population abundance within the restoration site. Sites should be laid out with pre-determined boundaries in a geometric shape, therefore site boundaries can be reestablished using a handheld Global Position System (GPS) and diving to determine limits of oysters seeded or substrate placed in previous years. Two general methods of seeding have been practiced in Rhode Island, resulting in varied spatial configurations of oyster beds. The first is exemplified through repeatedly seeding a large area (~1,000-3,000 m^2) with even distribution of oysters planted throughout the area. The second method is seeding a patch-work of discrete, small (~5-15 m^2) but highly dense oyster beds with negligible oyster presence between beds (Figure 1). In both cases oyster beds should be delineated, boundaries measured and corner boundaries marked and/or latitude longitude recorded. To accurately characterize oyster abundance

and density within discrete beds it is necessary to measure the footprint of each bed rather than the entire area of the site boundary they encompass.

Various methods have been used to define reef boundaries including: aerial imagery (Grizzle & Castagna 2000), side scan sonar (Luckenbach & Ross 2003), hydro-acoustic techniques (DeAlteris 1988), towed video (Grizzle *et al.* 2005), and walking the perimeter of intertidal reefs with surveying equipment (Coen & Walters 2007). Due to shallow depths and often low-lying reefs within restoration sites in Rhode Island, using hydro-acoustic, side scan sonar or video analysis to determine seeded areas may be challenging. Considering the current physical attributes of Rhode Island restoration sites, diving to determine the extent of oysters seeded and manually measuring bed perimeters is the most accurate, cost effective and transferable method of site delineation for practitioners. It is important to take note of oyster distributions and estimates of density during site delineation, as this will provide the basis for sampling design. Site boundaries and seeded area should be observed annually and re-established when necessary.

3.1 Oyster Density – Abundance

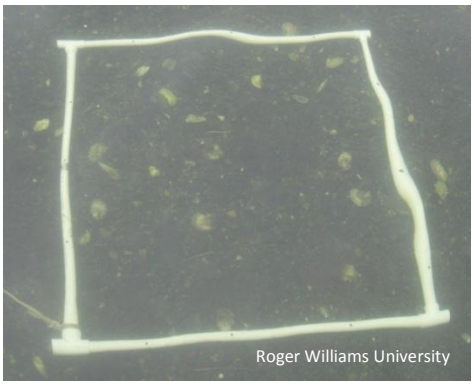

Figure 2. 1 m^2 quadrat deployed on a natural oyster bed.

Quadrat sampling is a common method used to provide quantitative estimates of abundance and survivorship of individual cohorts (Luckenbach & Ross 2003; Hancock *et al.* 2005, 2006, 2007; Brumbaugh *et al.* 2006; DeAngelis *et al.* 2008, 2009). Size of quadrats will vary depending on relative density of oysters observed during site delineation, whereas 1 m^2 quadrats are appropriate for sampling low density populations and 0.25 m^2 quadrats are adequate for sampling high density populations (Figure 2). Excavate all oysters from quadrats and enumerate live and recently dead (hinge still intact "boxes") oysters within each quadrat. Be careful to look for new recruits on oyster shells and other substrates within the quadrat (Figure 3). Keep a separate tally of new recruits, as this will enable recruitment densities to be calculated independently from oysters seeded. As an indicator of predation, record number of boxes with drill holes and boring sponge. After sampling, attempts should be made to replace all live and dead oysters back in the quadrat they were sampled from. Sampling design will vary depending on spatial distribution of oyster beds within a given site, as depicted in Figure 1, and can be broken down into a tiered system as follows.

Figure 3. Example of new recruitment or 'overset'.

Tier 1 - *Discrete oyster beds within a restoration footprint*: When a patchwork of discrete oyster beds exists with negligible oyster presence between beds, practitioners should treat each bed as an individual strata and sample beds independently from one another, thus reducing variance by minimizing zero quadrats. Quadrats should be distributed evenly in a haphazard, unbiased manner within each bed. Oyster abundance for each bed will be calculated from mean densities sampled, using bed area as a basis for extrapolation. Total oyster abundance in the given site will be the sum of oyster abundance in all individual beds.

Tier 2 - *Large area with even oyster distribution*: When large areas with relatively even oyster distributions exist, practitioners should treat the entire area as a single stratum, distributing quadrats evenly in a haphazard, unbiased manor. Total oyster abundance will be calculated within the bed from mean densities sampled, using total area of the sampled stratum as a basis for extrapolation.

Sampling methodologies can be adapted from Tier 1 to Tier 2, as sites change over time as a function of repetitive seeding, mortality, dispersal of oysters or other influencing factors. Sampling effort or number of replicate quadrats required will differ depending on variability of oyster distributions within given sites. Determining optimum sample size based on desired level of precision in the estimated mean can be derived from variance among samples. In essence, this requires two sampling events for each site; the first to determine variability among sample means, and second to sample for population characteristics of interest. If this is not feasible, it is suggested to obtain an adequate number of samples from each site or individual oyster bed, usually 2 - 10 % of the total seeded footprint. When sampling small oyster beds (~5-15 m^2), a minimum of three replicate quadrats should be used. Sites should be sampled annually, and timing of sampling should be held as consistent as possible from year to year.

3.2 Length Distribution

Length distributions provide the ability to delineate cohorts (i.e. multiple age classes), yearly growth rates, and the contribution of shell structure, a measure of shell accretion through growth and recruitment (Mann & Powell 2007). Oysters from quadrat samples can be used to determine length distributions. Before returning oysters back to the water measure the valve length (longest point from hinge to lip, Figure 4) of all live oysters and 30 boxes to the nearest millimeter using calipers. A sub-sample of 50 live oysters per quadrat is sufficient when densities are high. While sampling oysters, be careful to look for and measure new recruits within the quadrat, as length alone may not distinguish new recruits. Recruits can be distinguished from restoration oysters, as they may set over an existing oyster (over-set, Figure 3) or attach to a substrate not used in the remote setting process. To determine actual recruitment success and densities at the site, keep a separate tally of new recruits.

Figure 4. Measuring the valve length of an oyster from umbo to lip.

3.3 Recruitment

A key condition for measuring restoration performance is natural recruitment to the population. The objective of recruitment monitoring is to document the occurrence of spat settling on or near the restoration site, along with the magnitude and timing of the recruitment events (Hancock *et al.* 2006). Various materials and configurations have been used in Rhode Island for oyster spat collectors including: surf clam shell (*Spisula solidissima*), placed in 91cm x 91cm bottom trays (Hancock *et al.* 2006); five pieces of 10cm^2 x 1.3cm thick Hardibacker® hung in mid water column (Hancock *et al.* 2006, 2007); mesh bags containing *S. solidissima* shells hung in mid water column (DeAngelis *et al.* 2009; Leavitt, Pers. Comm.); and Chinese hats dipped in cement, hung in mid water column (Leavitt, Pers. Comm.; Doiron 2008). Documentation of oyster recruitment events have been limited in Rhode Island during the past decade, but should be monitored to track changes over time, particularly with the increase of broodstock from restoration activities.

Using mesh bags filled with surf clam shells is a cost-effective method of measuring relative settlement. Each collector is defined as a single bag of *S. solidissima* shells suspended in mid water column, as not to contact the benthic substrate at low tide (Figure 5). Each collector should contain approximately 10 single valves of *S. sollidissima*. Local hydrodynamics and wind patterns can dictate the dispersal and eventual locality of bivalve larvae at time of settlement, therefore, whenever possible locations of spat collectors should be based on this information. Thorough hydrodynamic models are not available for all bodies of water in Rhode Island, however much information can be gained from local tide books (e.g. Eldridge Tide and Pilot Book, White Instruments Inc.) coupled with knowledge of local bathymetry and seasonal wind patterns. To capture temporal variations in settlement, deployment and retrieval should take place on a rotating schedule every three weeks during the spawning season (June through September). If this is not feasible, spat collectors should be deployed and retrieved at the beginning and end of the spawning season, respectively.

Figure 5: Depiction of spat collector. Mesh bag filled with *S. solidissima* shell, suspended in mid water column.

The mean number of spat per collector can be converted to settlement indices (SI) to compare recruitment at each site. Settlement indices per collection for each site (SI_t) can be standardized with the following equation.

$$SI_t = \frac{\sum x/n}{w}$$

Where: x = number of spat per collector, n = number of collectors, and w = number of days deployed divided by 7 (modified from Southworth *et al.* 2010).

Annual settlement indices for each site (SI_a) can be standardized to:

$$SI_a = \sum SI_t / N$$

Where: N = number of collections per year (modified from Hancock *et al.* 2006). It is important to realize these indices can demonstrate relative magnitudes of abundance and distribution patterns but do not represent actual settlement rates on the bottom (Brumbaugh *et al.* 2006). A more appropriate measurement of recruitment rates on the bottom can be calculated from number of new recruits or overset per m^2, derived from density monitoring (section 3.1 & 3.2).

3.4 Disease

Diseases affecting oysters are a constant threat to the population. If funding is available, monitoring pathogen loads at restoration sites should be conducted before, during and after restoration to assess the impact of pathogens on the success of each site. A minimum of 30 individuals between 60-90 mm valve length should be sampled annually, in late summer to early fall, within the proximity of each restoration site. Samples should be accompanied with labels clearly describing the following: date collected, site owner, seed source, latitude-longitude (DDD.DDDD), temperature (˚C) and salinity (ppt). It is recommended all samples should be examined for prevalence and intensity of Dermo (*Perkinsus marinus)*, MSX (*Haplosporidium nelsoni*) and SSO (Seaside organism). Additional disease monitoring should be considered when high mortality not attributed to predation is experienced or when recently dead, gaping oysters are observed. The ability to attribute mortality to disease depends, in-part, on the level of confidence of ruling out other factors such as predation, thus, monitoring abundance of predators within restoration sites is highly recommended (see section 4.1). If sampling specifically for SSO, it is recommended to sample twice a year, once in the spring and once in the fall. All sampling should be coordinated with local shellfish pathologists (see Appendix A).

IV. Beneficial Monitoring

Collecting data beyond general population structure is often necessary to gauge project-level performance and outcomes. While it is not feasible to collect such data at all restoration sites, controlled experiments can provide valuable information to better understand factors affecting oyster restoration outcomes and ecosystem services of restoration (e.g. sources of stress and ecosystem functions/interactions). The following is not meant to be an exhaustive list or provide finite methodologies; instead, it briefly discusses some examples of beneficial monitoring that may be considered for implementation on restoration projects, depending on the projects objectives, geographic location and scope. Ultimately, it is the responsibility of the investigators to design sound

experiments to answer questions of interest. To be useful in gauging project performance, parameters should be measured prior to starting restoration and after a predetermined period of time.

4.0 Reproductive Effort

Monitoring oyster recruitment does not provide a direct indication of reproductive development in the animal, nor does it provide an indication of larvae in the water column which did not survive to settlement. The following methods can be used to collect discrete data on the reproductive effort of *C. virginica.*

Condition Index: Condition index is a measurement of soft tissue in an individual bivalve, normalized to a dimension of the oyster's shell (Mann *et al*. 1978). As the majority of variation in seasonal mass is associated with gonadal development, condition index can be used as an indirect indicator of gonadal maturation. Common methods used to assess condition indices involve the collection of bivalves over the course of the reproductive cycle with a schedule that captures seasonal changes. Samples collected from a given site every three weeks, May through September, will provide an adequate view of the reproductive cycle. A gravimetric condition index can be employed using the procedures and formula of Crosby and Gale (1990).

$$CI = [\text{dry soft tissue weight (g)} \times 1000] / [\text{total weight (g)} - \text{shell weight (g)}]$$

All weights should be estimated to the nearest 0.1 g. Tissue should be dried at 80˚C for 48 hours while the shell air dries for the same time period.

Gonadal Index: Collected over the spawning season (May through September), gonadal indices provide direct microscopic examination of the development of reproductive organs of the bivalve. Procedures can follow those discussed in Morraquin-Mora and Rice (2008), Howard and Smith (1983) and Eversole (1997). Oysters can be shucked, a cross section of gonadal tissue removed, placed in histology cassettes and fixed in formalin. In order for examination samples need to be stained, embedded in wax,

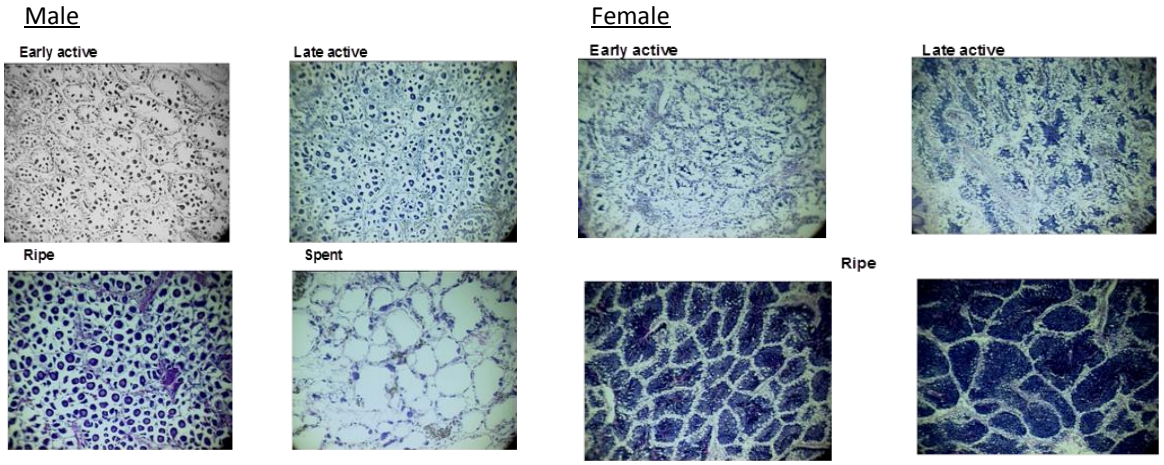

Figure 6: Gonadal stages of the northern quahog. Inactive=0, Early active=1, Late active=2, Ripe=3, Spent=1. Regression=0. From Eversol 1997 and Morraquin-Mora and Rice 2008.

microtomed and mounted on slides. This can be accomplished in house or sent to a histological laboratory. To facilitate analysis, gonadal tissue is assigned an index according to the stage of development, similar to that depicted in figure 6.

<u>*Larval Monitoring*</u>: Larval monitoring provides data on estimates of veliger stage oyster abundance present in the water column at or near restoration sites. Such data can help discern whether recruitment failure is due to lack of available larvae or a bottleneck between larval stage and settlement. Cost effective techniques include: filtration of known volumes of water through plankton nets in mid to lower water column and enumerating veliger larvae (Wood & Hargis 1971; Shanks & Brink 2005; DeAngelis *et al.* 2008). To provide an adequate window to capture larvae in the water column, sites should be sampled with even frequency (1-2 times per week) throughout the spawning season (May through September). Samples can be examined under a microscope and larvae enumerated through the use of a Sedgwick-rafter cell or Hemocytometer, where mean number of larvae per sample can be extrapolated to larvae m^{-3}. Deciphering species of bivalve larvae can be extremely challenging and limit effectiveness of larval monitoring. Species-specific morphometrics (length–width ratios) and birefringence patterns (Figure 7), derived through the use of refracted polarized light, can be used to help mitigate this problem (Mingione 2008; Mingione 2011). A guide to sampling techniques and identification of bivalve larvae is available from Aucoin *et al.* (2004).

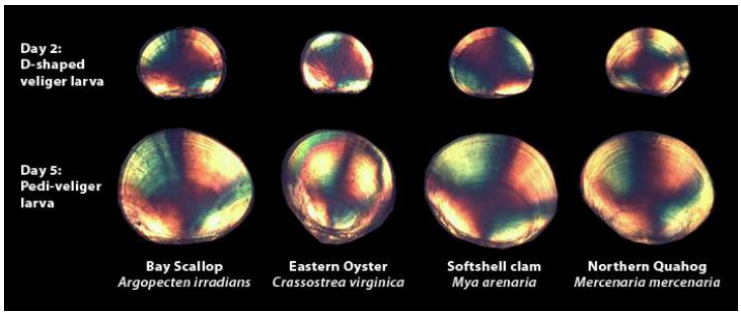

Figure 7: Differences in birefringence patterns of larval bivalve species (Mingione 2008).

A more robust but expensive method includes the use of Larval Identification and Hydrographic Data Telemetry (LIHDaT). The LIHDaT system uses computer software to interoperate species-specific birefringence patterns and can provide accurate estimates of bivalve larvae within water samples. (Tiwari & Gallager 2003; Mingione 2011). It is also possible to identify bivalve larvae through the use of electron microscopy (Lutz *et al.* 1982), DNA based methods (Bell & Grassle 1998; Hare *et al.* 2000; Larsen *et al.* 2005) and immunological methods (Garland 2000).

4.1 Sources of Stress - Mortality

Better understanding local sources of stress on oyster populations can lead to adaptive management and ultimately more successful projects. The following are sources of stress on oyster reefs that can be documented with relative ease. This is not a comprehensive list of stressors, rather a starting point for discussion.

<u>*Sedimentation*</u>: Heavy sedimentation has shown to increase mortality of oysters (Lenihan & Thayer 1999) and reduce settlement. Documenting sedimentation rates and how they relate to growth, recruitment and reproduction can impact decisions on future placement of restoration projects.

Documenting sedimentation rates is a common practice and can be accomplished with a suite of sediment traps as described in, McNinch (1997) and Blomqvist & Hakanson (1981).

Predation: Predation clearly impacts the success of oyster restoration. Documenting relative abundance of predators at restoration sites coupled with quantitative evidence of predation (cracked or damaged boxes) can shed light on the influence predators have on survival at given sites.

Quantifying relative abundance of mobile predators at restoration sites can be accomplished through various approaches. These methods include: dive transects (Luckenbach & Ross 2003), use of lift nets (Wenner *et al.* 1996; Tolley & Volety 2005), flume weirs (Knieb 1991; Wenner *et al*. 1996; Coen & Luckenbach 2000), throw traps (Glancy *et al.* 2003), and trays embedded in the reef and removed and sampled at pre-determined intervals (Wenner *et al.* 1996; Luckenbach & Ross 2003; Rodney & Paynter 2006). To quantify a relative index of dominant predators at each site, the use of 25 m transects is recommended. A minimum of 3 transects per site should be deployed in a simple random design prior to any other sampling activities. To avoid overlap of sampled area, each transect should be laid out in a north-south orientation. Number of predators including: blue crabs, green crabs, spider crabs, starfish, oyster drills and mud crabs should be enumerated 1 m from either side of the transect.

4.2 Ecological Benefits

A needed goal of restoration monitoring is establishing and quantifying ecological benefits associated with restored oyster populations. Healthy oyster reefs provide critical ecological functions by: providing structural habitat for marine organisms, improving water quality by filtering excess nitrogen from estuaries and protecting shorelines from erosion by stabilizing sediments (Brumbaugh *et al.* 2006). Coupled with ecological benefits, oyster reefs can provide a significant socioeconomic value to the region. They can increase fisheries and associated infrastructure both directly through harvest of oysters and indirectly through providing nursery grounds resulting in increased fish production (Peterson *et al.* 2003).

Documenting and quantifying the benefits associated with oyster restoration will help further our knowledge of the role oysters play as ecosystem engineers, as well as provide a tool to increase public awareness as to their local importance. Community involvement and awareness of restoration programs and associated benefits is critical in fostering environmental stewardship and long term support.

Associated Fauna: Comparative surveys documenting faunal assemblages of restored and non-restored oyster habitat can offer information on the habitat value oysters provide. A suite of methods exists for documenting both sessile and mobile fauna. Methods outlined in surveying mobile predator abundance (section 4.1) can also be used to document motile reef inhabitants. A common method of surveying sessile epifauna and infauna employs the use of trays embedded in the reef and removed and sampled at pre-determined intervals (Wenner *et al.* 1996; Luckenbach & Ross 2003; Rodney & Paynter 2006).

Effects on Water Quality: Brumbaugh *et al.* (2006), provides an overview of laboratory and field studies that have documented filter-feeding bivalves capacity to reduce particulates in overlying water (Verwey 1952; Haven & Morales-Alamo 1970; Asmus & Asmus 1991; Dame 1996). The ability of shellfish to reduce total suspended solids (TSS) and cholophyll *a* (Chl-a) have been documented by Haamer and Rodhe (2000), Cressman *et al.* (2003), and Nelson *et al.* (2004).

Measuring TSS in a water sample can be accomplished by filtering a known volume of water though a pre-weighed glass fiber filter, then weighing again after drying to remove all water. The gain in weight is representative of the dry weight of the particulates derived from the water filtered, typically expressed in mg/L.

$$TSS\ (mg/L) = (A - B)\left(\frac{1000}{C}\right)$$

Where: A = End weight of filter (g)
B = Initial weight of filter (g)
C = Volume of H_2O filtered (L)

Basic methodology of measuring Chl-a involves the filtration of a known volume of water through a glass fiber filter. The pigments are extracted with a solvent (acetone or alcohol) and measured spectophotometrically by determining the absorbance of the extract at various wavelengths. A variety of handheld units are also available to measure Chl-a in-situ.

V. Conclusion

Rhode Island has long recognized the ecological and socioeconomic role the oyster plays in the state. There is a growing interest, participation and investment in oyster restoration in Rhode Island, with direct involvement from many organizations and a multitude of private and public stakeholders. Various cooperative and collaborative efforts of coordinating oyster restoration throughout the state are being made. With the increased participation, it is important to define a standardized method of monitoring restoration outcomes. This will allow for cross site and cross program analyses, providing the opportunity to evaluate successes and failures, ecological impacts and ultimately improve the future of oyster restoration in Rhode Island.

VI. References

Asmus, R.M., Asmus, H. 1991. Mussel beds: Limiting or promoting phytoplankton? *J. Exp. Mar. Biol. Ecol.* 148: 215-232.

Aucoin, F., Doiron, S., Nadeau, M. 2004. Guide to sampling and identifying larvae of species of maricultural interest. Guide Publication G005. Government of New Brunswick and Quebec. ISBN: 1-55137-660-8. 73pp.

Beck, M.W., Brumbaugh, R.D., Airoldi L., Carranza, A., Coen, L., Crawford, C., Defeo, O., Edgar, G., Hancock, B., Kay, M., Lenihan, H., Luckenbach, M., Toropova, C., Zhang, G., Guo, X. 2011. Oyster reefs at Risk and Recommendations for Conservation, Restoration, and Management. *BioScience* 62(2): 107-116.

Bell, J.L., Grassle, J.P. 1998. A DNA probe for identification of the larvae of commercial surf clam (*Spisula solidissima*). *Molec. Mar. Bio. Biotechnol.* 2: 129-136.

Bernhardt, E.S., Palmer, M.A., Allan, J.D., Alexander, G., Barnas, K., Brooks, S., Carr, J., Clayton, S., Dahm, C., Follstad-Shah, J., Galat, D., Gloss, S., Goodwin, P., Hart, D., Hassett, B., Jenkinson, R., Katz, S., Kondolf, G.M., Lake, P.S., Lave, R., Meyer, J.L., O'Donnell, T.K., Pagano, L., Powell, B., and Sudduth, E. 2005. Synthesizing U.S. River Restoration Efforts. *Science* 308: 636-637.

Blomqvist, S., Hakanson, L. 1981. A review on sediment traps in aquatic environments. *Archiv fur Hydrobiologie* 19(1): 101-132

Brumbaugh, R. D., Beck, M.W., Coen, L., Craig, L., Hicks, P. 2006. A practitioners' guide to the design and monitoring of shellfish restoration projects: an ecosystem approach. The Nature Conservancy, Arlington, VA, 28pp.

Carlisle, B.K., Baker, J.D., Hicks, A.L., Smith, J.P., Wilbur, A.L. 2004. Cape Cod Salt Mash Assessment Project; Final Grant Report, Volume 1: Relationship of salt marsh indices of biotic integrity to surrounding land use, 1999 Boston MA. Massachusetts Office of Coastal Zone Management. 45pp.

Coen, L., Luckenbach, M. 2000. Developing Success criteria and goals for evaluating oyster reef restoration: ecological function or resource exploitation? *Ecological Engineering* 15: 323-343.

Coen, L., Walters, K. 2007. Oyster Restoration Workgroup: Reef Size. http://www.oyster-restoration.org/metrics/reefsize.php.

Cressman, K.A., Posey, M.H., Mallin, M.A., Leonard, L.A., Alphin, T.D. 2003. Effects of oyster reefs on water quality in a tidal creek estuary. *Journal of Shellfish Research.* 22(3): 73-762.

Crosby, M.P., Gale, L.D. (1990). A review and evaluation of bivalve condition index methodologies with a suggested standard method. *Journal of Shellfish Research.* 9(1): 233-237.

Dame, R.F. 1996. Ecology of Bivalves: An Ecosystem Approach. CRC Press, Boca Raton, FL. 254pp.

DeAlteris, J.T., Gibson, M., Skrobe, L.G. 2000. Narragansett Bay Summit 2000 White Paper: Fisheries of Rhode Island. Narragansett Bay Estuary Program, Providence, RI. 48pp.

DeAlteris, J.T. 1988. The application of hydroacoustics to the mapping of subtidal oyster reefs. *Journal of Shellfish Research* 7: 41-45.

DeAngelis, B., Griffin, M., Kocot, M., Turek, J., Lazar, N. 2009. *North Cape* shellfish restoration program, 2008 annual report. Rhode Island Department of Environmental Management, National Oceanic and Atmospheric Administration, and U.S. Fish and Wildlife Service. 54pp.

DeAngelis, B., Hancock, B., Kocot, M., Turek, J., Lazar, N. 2008. *North Cape* shellfish restoration program, 2007 annual report. Rhode Island Department of Environmental Management, National Oceanic and Atmospheric Administration, and U.S. Fish and Wildlife Service. 44pp.

Doiron, S. 2008. Reference Manual for Oyster Aquaculturists. Government of New Brunswick, Agriculture Aquaculture and Fisheries. ISBN 978-1-5471-094-2. 76pp.

Eversole, A.G. 1997. Gametogenesis of *Mercenaria mercenaria, M. campechiensis* and their hybrids. *The Nautilus*. 110(4): 107-110.

Garland, E.D. 2000. Temporal variability and vertical structure in larval abundance: the potential roles of biological and physical processes. Doctoral Dissertation. Massachusetts Institute of Technology/Woods Hole Oceanographic Institution, Woods Hole, MA. 213pp.

Glancy, T.P., Frazer, T. K., Cichra, C. E., Lindberg, W. J. 2003. Comparative patterns of occupancy by decapod crustaceans in seagrass, oyster, and marsh-edge habitats in a Northeast Gulf of Mexico estuary. *Estuaries* 26(5): 1291-1301.

Grizzle, R., Castagna, M. 2000. Natural intertidal oyster reefs in Florida: Can they teach us anything about constructed/restored reefs? *Journal of Shellfish Research* 19: 609.

Grizzle, R.E., Ward, L.G., Adams, J.R., Dijkstra, S.J., Smith, B. 2005. Mapping and Characterizing Oyster Reefs Using Acoustic Techniques, Underwater Videography, and Quadrat Counts. Proceedings Volume, Symposium on the Effects of Fishing Activities on Benthic Habitats: Linking Geology, Biology, Socioeconomics, and Management.

Haamer, J., Rodhe, J. 2000. Mussel *Mytilus edulis* (L.) filtering of the Baltic Sea outflow through the Öresund - An example of a natural large-scale ecosystem restoration. *Journal of Shellfish Research.* 19: 413-421.

Hancock, B., DeAngelis, B., Turek, J., Lazar, N. 2007. *North Cape* Shellfish Restoration Program 2006 annual report. Rhode Island Department of Environmental Management, National Oceanic and Atmospheric Administration, and U.S. Fish and Wildlife Service. 66pp.

Hancock, B., Costa, D., Ryan, K., Turek, J., Lazar, N. 2006. *North Cape* Shellfish Restoration Program 2005 annual report. Rhode Island Department of Environmental Management, National Oceanic and Atmospheric Administration, and U.S. Fish and Wildlife Service. 72pp.

Hancock, B., Holly, J., Turek, J., Lazar, N. 2005. *North Cape* Shellfish Restoration Program 2004 annual report. Rhode Island Department of Environmental Management, National Oceanic and Atmospheric Administration, and U.S. Fish and Wildlife Service. 43pp.

Hare, M.P., Palumbi, S.R., Butman, C.A. 2000. Single-step species identification of bivalve larvae using multiplex polymerase chain reaction. *Mar. Biol.* 137: 953-961.

Haven, D., Morales-Alamo, R. 1970. Filtration of particles from suspension by the American oyster, *Crassostrea virginica*. *Biol.Bull.* 139: 248-264.

Howard, D.W., Smith, C.S. 1983. Histological Techniques for Marine Bivalves Mollusks. NOAA Technical Memorandum NMFS-F/NEC-25. U.S. Department of Commerce. 97pp.

Kneib, R.T. 1991. Flume weir for quantitative collection of nekton from vegetated intertidal habitats. *Mar. Ecol. Prog. Ser.* 75: 29–38.

Larsen, J.B., Fischer, M.E., Ramussen, L.J., Hansen, B.W. 2005. Single-step nested multiplex PCR to differentiate between various bivalve larvae. *Mar. Biol.* 146: 1119-1129.

Leavitt, D. Associate Professor. Roger Williams University. Bristol, RI.

Lenihan, H.S., Thayer, G.W. 1999. Ecological effects of fishery disturbance to oyster reef habitat in eastern North America. *Journal of Shellfish Research* 18: 719.

Luckenbach, M.W., Coen, L.D. 2003. Oyster reef habitat restoration: A review of restoration approaches and an agenda for the future. *Journal of Shellfish Research* 22:341.

Luckenbach, M.W., Ross, P.G. 2003. An Experimental Evaluation of the Effects of Scale on Oyster Reef Restoration: Final Report Submitted VA Sea Grant Consortium. 120pp.

Lutz, R., Goodshell, J., Castagna, M., Chapman, S., Newell, C., Hidu, H., Mann, R., Jablonski, D., Kennedy, V., Siddall, S., Goldberg, R., Beattie, H., Galmagne, C., Chestnut, A., Partridge, A. 1982. Preliminary observations on the usefulness of hinge structures for identification on bivalve larvae. *Journal of Shellfish Research*. 2(1): 65-70.

Mann, R., Thorp, J.H., Gibbons, J.W. 1978. A comparison of morphometric, biochemical, and physiological indexes of condition in marine bivalve mollusks. In: Energy and environmental stress in aquatic systems. DOE symp. Ser. (no. 48). Publ. by: Technical Information Center, US Dep. Energy, Oak Ridge, TN (USA). 484-497.

Mann, R., Powell, P.N. 2007. Why oyster restoration goals in the Chesapeake Bay are not and probably cannot be achieved. *Journal of Shellfish Research. 26(4): 905-917.*

McNinch, J.E. 1997. Sedimentary processes and formation of a barrier island cape. Doctoral Dissertation. University of North Carolina at Chapel Hill, North Carolina, USA.

Mingione, C.T. 2008. Shellfish's mysterious pathways to adulthood. Oceanus. Dec. 2008.

Mingione, C.T. 2011. Species-Specific patterns in bivalve larval supply to a coastal embayment. Doctoral Dissertation. Massachusetts Institute of Technology/Woods Hole Oceanographic Institution, Woods Hole, MA. 242pp.

Morroquin-Mora, D.C., Rice, M.A. 2008. Gonadal cycle of northern quahogs, *Mercenaria mercenaria* (Linne, 1758), from fished and non-fished subpopulations in Narragansett Bay. *Journal of Shellfish Research.* 27(4):643-652.

Nelson, K.A., Leonard, L.A., Posey, M.H., Alphin, T.D., Mallin, M.A. 2004. Using transplanted oyster beds to improve water quality in small tidal creeks: A pilot study. *J. Exp. Mar. Biol. Ecol.* 298: 347-368.

Peterson, C.H., Grabowski, J., Powers, S.P. 2003. Estimated enhancement of fish production resulting from restoring oyster reef habitat: quantitative valuation. *Mar. Ecol. Prog. Ser*. 264: 249-264.

Rice, M.A., Valliere, A., Caporelli, A. 2000. A review of shellfish restoration and management projects in Rhode Island. *Journal of Shellfish Research* 19(1): 401-408.

Rodney, W. Paynter, K.T. 2006. Comparisons of macrofauna assemblages on restored and non-restored oyster reefs in mesohaline regions of Chesapeake Bay in Maryland. *J. Exp. Mar. Biol. Ecol.* 335: 39-51.

Shanks, A.L., Brink, L. 2005. Upwelling, downwelling, and cross-shelf transport of bivalve larvae: test of a hypothesis. *Mar Ecol. Prog. Ser.* 302: 1-12.

Southworth, M., Harding, J.M., Mann, R. 2010. The status of Virginia's public oyster resource 2009. Mulluscan Ecology Program, Virginia Institute of Marine Science, Gloucester Point, Virginia. 52 pp.

Tiwari, S., Gallager, S. 2003. Machine learning and multiscale methods in the identification of bivalve larvae. In: Proceedings fo the Ninth IEEE International Conference on Computer Vision. Vol. 2. IEEE Computer Society Press, Los Alamitos. 1-8.

Tolley, G.S., Volety, A.V. 2005. The role in oyster habitat use of oyster reefs by resident fishes and decapod crustaceans. *Journal Shellfish Research* 24(4): 1007-1012.

Wenner, E., Beatty, H.R., Coen, L.D. 1996. Method for quantitatively sampling nekton on intertidal oyster reefs. *Journal Shellfish Research* 115: 769–775.

Wood, L., Hargis, W.J. 1971. Transport of bivalve larvae in a tidal estuary. In: Fourth European Marine Biology Symposium. Publ. by: Syndica of Cambridge University Press, London. 29-44.

Verwey, J. 1952. On the ecology and distribution of cockle and mussel in the Dutch Wadden Sea, their role in sedimentation and the source of their food supply. *Arch. Neerl. Zool*. 10: 172-239.

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Appendix B: Contacts – Water Quality Monitoring Groups in Rhode Island

Narragansett Bay Estuary Program
URI Bay Campus
Narragansett, RI 02882
(401) 824-6233
www.nbep.org

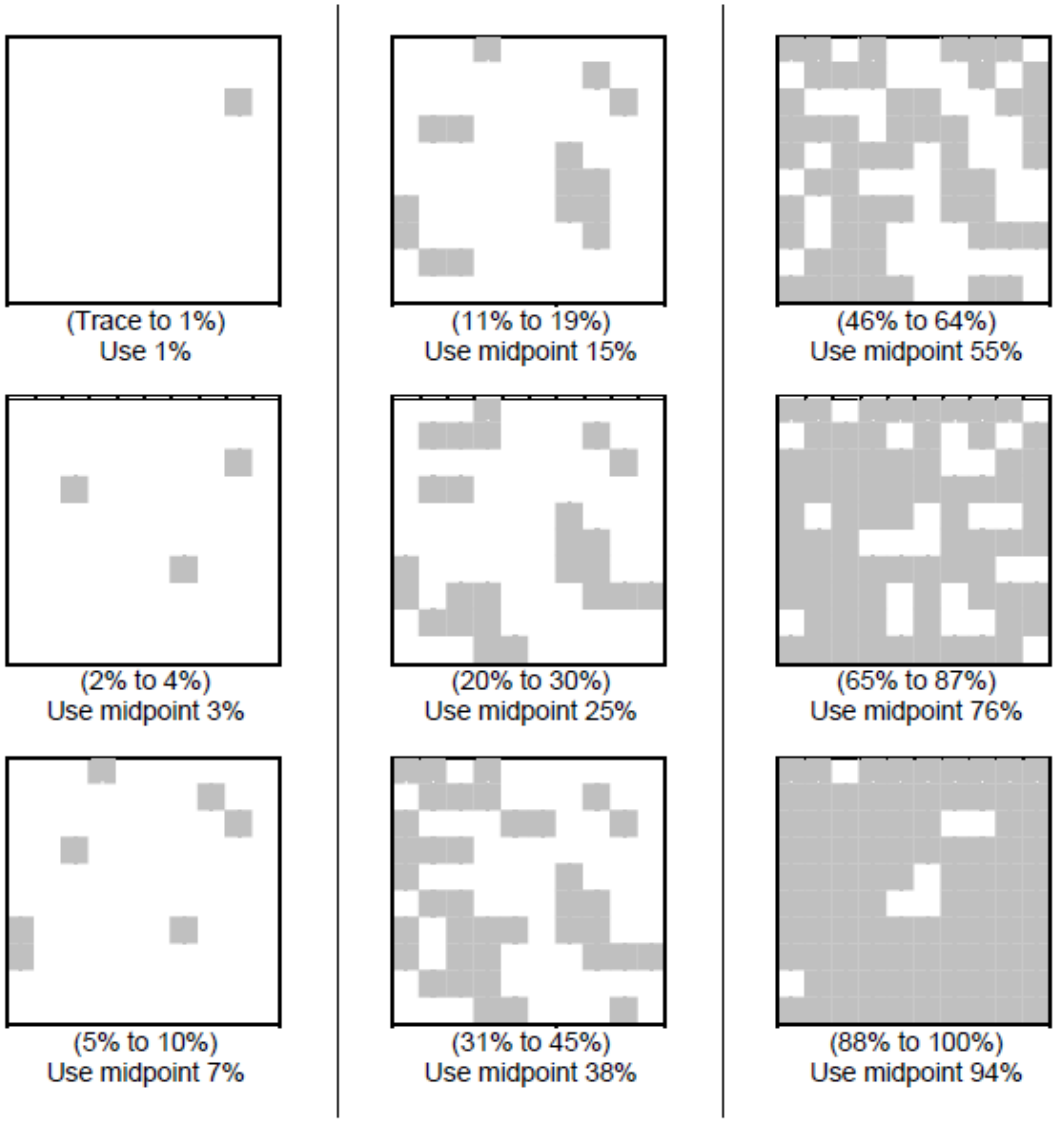
Narragansett Bay National Estuarine Research Reserve
Prudence Island, RI 02872
(401) 683-6780
www.nbnerr.org

Salt Ponds Coalition
Charlestown, RI 02813
(401) 322-3068
www.saltpondscoalition.org

Watershed Watch
URI Coastal Institute
Kingston, RI 02881
www.uri.edu/ce/wq/ww/

Appendix C: Standard Cover Classes

Depiction of nine standard cover classes to determine percent composition of benthic substrate or macro algae within each quadrat sampled. Determine appropriate percent cover based on the figures below and record the midpoint (Adopted from Carlise *et al*. 2004).



Carlisle *et al.* 2004

Monitoring Outline

Essential Monitoring: Recommended for all restoration sites

1. **Site Description**
    a. Physical – Chemical: If possible measure during each site visit.
        - Temperature (˚C)
        - Salinity (ppt)
        - Dissolved O_2 (mg/L)
    b. Depth: Intertidal or sub-tidal - Depth at mean low water (m).
    c. Benthic Substrate:
        - Percent coverage of substrate type (Mud, Sand/Silt, Sand/Cobble, Rocky)
        - Percent coverage of dominant macro-algae
    d. Substrate Enhancement:
        - Date of enhancement & material used
        - Density (vol. material m^{-2})
        - Vertical relief
        - Total area (m^2) enhanced
    e. Seeding History:
        - Date & number seeded
        - Average size of seed (mm)
        - Approximate seeding density (# oysters m^{-2})
        - Origin of seed (i.e. hatchery)
    f. Location:
        - Lat-Lon (DDD.DDDD) of center point of each seeded bed within restoration site
        - Unique identifier for each bed
        - E.g.: Bissel Cove, Bed 1 41.54615N, 71.42942W
    g. Footprint:
        - Dive to determine extent of oyster seed or clutched area of each bed
        - Record Lat-Lon and/or mark corners if appropriate
        - Measure dimensions of boundary (m)
        - Observe site boundaries annually, re-establish when necessary

2. **Quadrat sampling: Oyster Density & Length Distribution**
    a. Quadrat Size: 1 – 0.25 m^2 depending on oyster density
    b. Sampling Effort:
        - Optimize sample allocations based on variance of the sampled mean
        - Or: sample 2 – 10% of total seeded footprint - At least 3 replicate quadrats per oyster bed
    c. Sampling Design:
        - Distribute quadrats evenly in haphazard manor – within each oyster bed
        - Number of live and recently dead (hinge still intact) oysters in each quadrat
        - Number of recruits in each quadrat – separate tally
        - Number of oysters with drill holes or boring sponge
        - Valve length (mm) of 50 live and 30 dead oysters per quadrat
        - Return oysters to quadrat

3. **Recruitment**
    a. Artificial spat collectors:
        - Mesh bags filled with 10 individual surf calm valves - One bag per collector
        - Hung in mid water column
        - Spatially distributed throughout body of water
        - Deployment from June – October
        - Collect on 3 week rotating schedule, Or: end of season
        - Settlement indices per retrieval:

$$SIt = \frac{\sum x/_n}{w}$$

Where: x = number of spat
n = number of collectors
w = days deployed divided by 7

        - Annual settlement indices:

$$SIa = \sum SIt \; / \; N$$

Where: N = number of collections per year

    b. Recruitment to site:
        - Number of recruits per quadrat during density sampling – separate tally

4. **Disease Monitoring**
        - 30 oysters (60-90 mm valve length) sampled within each restoration site
        - Collect samples mid-August through September annually
        - Test for Dermo, MSX and SSO
        - Coordinate with local pathologists

Beneficial Monitoring

1. **Reproductive Effort**
    a. Condition Index: Provides index of changes in soft tissue mass
        - Samples collected every 3 weeks (May-September)
        CI = [dry soft tissue weight (g) X 1000] / [total weight (g) - shell weight (g)]
        (Crosby and Gale 1990)
    b. Gonadal Index: Direct microscopic measurement of gonadal tissue
        - Samples collected every 3 weeks (May – September)
        - Cross section of gonad removed, stained and mounted on slide
        - Gonadal tissue assigned an index depending on stage of development
        (Eversole 1997, Moraquin-Mora & Rice 2008)
    c. Larval Monitoring: Estimate density of veliger stage larvae in water column
        - Collect samples (1-2 X per week) over reproductive season (May-September)
        - Filtration of know volume of water and enumeration of larvae
        - Larval identification through morphometrics and birefringence patterns
        (Wood & Hargis 1971; Tiwari & Gallager 2003; Shanks & Brink 2005; DeAngelis *et al.* 2008)

2. Sources of Stress – Mortality

a. Sedimentation:
   - Sediment traps to measure sedimentation rates at restoration sites (McNinch 1997; Blomqvist & Hakanson 1981)

b. Predation: Relative abundance of predators at restoration sites
   - Three 25 m transects per site
     - Enumerate predators within 1 m of either side of transect
   - Lift nets (Tolley & Volety 2005; Wenner *et al.* 1996)
   - Flume wiers (Wenner *et al.* 1996; Coen & Luckenbach 2000; Knieb 1991)
   - Throw traps (Glancy *et al.* 2003)
   - Sampling trays (Wenner *et al.* 1996; Luckenbach & Ross 2003; Rodney and Paynter 2006)

3. Ecological Benefits

a. Associated Fauna: Compare biodiversity of restored and non-restored sites
   - Mobile fauna
     - Dive transect (Luckenbach & Ross 2003)
     - Lift nets (Tolley & Volety 2005; Wenner *et al.* 1996)
     - Flume wiers (Wenner *et al.* 1996; Coen & Luckenbach 2000; Knieb 1991)
     - Throw traps (Glancy *et al.* 2003)
   - Sessile epifauna
     - Sampling trays (Wenner *et al.* 1996; Luckenbach & Ross 2003; Rodney & Paynter 2006)

b. Effects on Water Quality:
   - Reduction of suspended particulates (Verwey 1952; Haven & Morales-Alamo 1971; Asmus & Asmus 1991)
   - Reduction in total suspended solids and Chlorophyll *a* (Dame 1996, Haamer & Rodhe 2000; Cressman *et al.* 2003; Nelson *et al.* 2004)