Oyster Habitat Restoration

Monitoring and Assessment Handbook

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Executive Summary

Oyster reefs or beds are a globally imperiled marine habitat, with degradation primarily driven by anthropogenic factors such as overharvest, changes to hydrology and salinity regimens, pollution and introduced disease. While oyster restoration efforts have historically focused on improving harvests, in recent decades there has been an increasing recognition and better quantitative description of a broader array of ecological services provided by oysters. This has prompted many agencies and conservation organizations to re-focus their attention on restoring oyster habitat for these broader ecological functions and societal benefits. Benefits include production of fish and invertebrates of commercial, recreational and ecological significance, water quality improvement, removal of excess nutrients from coastal ecosystems, and stabilization and/or creation of adjacent habitats such as seagrass beds and salt marshes. Increasingly, these ecosystem services are cited as the principal or exclusive goal(s) of oyster restoration projects.

Despite increased restoration the restored reefs have often not been monitored to an extent that allows for comparison. A recent meta-analysis of oyster restoration projects in the Chesapeake Bay examined the available datasets from 1990 to 2007, analyzing over 78,0000 records from 1035 sites (Kramer and Sellner 2009, Kennedy et al. 2011). The analysis found that relatively few of the restoration activities were monitored, and that the restoration goals of many of the projects were not well-defined, with only 43% of the datasets including both a restoration and monitoring component. The authors concluded that the monitoring of this large body of work was inadequate, and they were unable to assess changes in oyster populations on the constructed reefs. Their recommendations were to implement all oyster restoration projects using experimental designs with robust sample size replication and quantitative pre- and post-restoration monitoring. Sufficient monitoring would allow for adaptive management during the post-construction phase, for assessing whether the project met its goals and related performance criteria or to determine whether restored reefs are achieving the stated ecosystem-based restoration goals.

To address this critical gap, a working group was formed that consisted of restoration scientists and practitioners from the Atlantic, Pacific, and Gulf coasts of the US. The aim of the group was to recommend monitoring techniques and performance criteria for both the eastern oyster (*Crassostrea virginica*) and the Olympia oyster (*Ostrea lurida*) that would allow for more extensive and consistent post-restoration assessment between projects on varying geographic scales. **With additional expert input, the working group developed recommendations for a set of Universal Metrics that should be monitored for all oyster restoration projects. The working group also developed guidelines for assessing optional Restoration Goal-based Metrics.** The Goal-based Metrics were not meant to be monitored for all projects, but could be monitored as needed to measure project performance and to advance the science of oyster habitat restoration depending on the availability of the necessary funding, capacity, and expertise. The Universal Metrics allow for the systematic assessment of the basic performance of restoration projects, whereas the Restoration Goal-based Metrics would allow practitioners to assess the performance of the restored reefs in meeting the ecosystem service-based restoration goal(s) associated with a project. Together, these metrics allow for the comparison of projects across a variety of scales and restoration approaches. Monitoring of the Universal Environmental Variables will also aid in the interpretation of Universal and Restoration Goal-based Metrics data collected through both pre- and post-restoration monitoring.

The Universal Metrics that should be monitored for every oyster restoration project include: (1) reef areal dimension; (2) reef height; (3) oyster density; and, (4) oyster size-frequency distributions. Performance criteria for the Universal Metrics are based on emergent structure (assessed as reef height), successful recruitment, and oyster density present at both short- and mid-term post-construction time frames. The following Universal Environmental Variables should also be monitored for every oyster restoration project to aid with interpretation of Universal Metrics data: (1) water temperature; (2) salinity; and, (3) dissolved oxygen (for subtidal reefs). Restoration practitioners that lack the equipment or capacity to conduct the minimum required monitoring should collaborate with others able to provide this capability such as local researchers from academic institutions and local, state or federal agencies.

Along with the Universal Metrics, the Restoration Goal-based Metrics are an optional set of monitoring guidelines provided to enable project managers to assess the following ecosystem service-based restoration goals: (1) brood stock and oyster population enhancement; (2) habitat enhancement for resident and transient species; (3) enhancement of adjacent habitats; and (4) water clarity improvement. This handbook is meant to be a living document that allows for future updates as monitoring methodologies and the state of the science evolve, and as meta-analyses of comparable data are undertaken. Restoration practitioners and other interested parties may at any time submit their comments and any suggestions for improvement to the handbook to ormetrics@gmail.com for consideration in future editions.

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Acknowledgements

The establishment of standardized restoration metrics has been a priority within the oyster restoration community for many years, and this handbook is built upon previous work (e.g., Thayer et al. 2003, 2005; Coen et al. 2004; Brumbaugh et al. 2006). We would like to express our appreciation to the leads of a 2004 workshop (Drs. Loren Coen, Keith Walters, Dara Wilber, Ms. Nancy Hadley and the participants) for providing a valuable starting point. To further develop this handbook, members of a steering committee from the University of South Alabama, The Nature Conservancy, Florida Atlantic University, and the NOAA Restoration Center convened a workshop in 2011 in Silver Spring, MD. The authors would like to thank the participants for their contributions through the 2011 workshop as well as for their valuable comments and expert input on various drafts of this document: Brian Allen (Puget Sound Restoration Fund), Dr. Denise Breitburg (Smithsonian Environmental Research Center), Dr. Dave Bushek (Rutgers University), Dr. Jonathan Grabowski (Northeastern University), Dr. Ray Grizzle (University of New Hampshire), Dr. Ted Grosholz (University of California-Davis), Dr. Megan La Peyre (U.S. Geological Survey and Louisiana State University), Dr. Mark Luckenbach (Virginia Institute of Marine Science), Dr. Kay McGraw (NOAA), Dr. Mike Piehler (University of North Carolina-Chapel Hill), Stephanie Westby (NOAA), Dr. Steve Geiger (Florida Fish and Wildlife Conservation Commission, Florida Fish and Wildlife Research Institute) and Dr. Philine zu Ermgassen (University of Cambridge). The authors would also like to thank Dr. Brad Andres (U.S. Fish and Wildlife Service), Felicia Sanders (South Carolina Department of Natural Resources), Barry Truitt (The Nature Conservancy), and Alex Wilke (The Nature Conservancy) for contributing sections pertaining to the monitoring of waterbird usage of oyster habitat, and Jim Lyons (U.S. Fish and Wildlife Service) and Matt Reiter (PRBO Conservation Services) for their helpful comments on that section. Drs. Mark Spalding and Philine zu Ermgassen contributed the section on defining natural oyster habitats, for which we are also grateful. Additionally, the authors would like to extend our gratitude to Stan Bosarge and Mike Dardeau, both of the Dauphin Island Sea Lab, for providing information concerning the use of various types of equipment discussed in this handbook. Valuable comment was provided on the merit of various sections to the restoration of Olympia oysters by the San Francisco Bay Native Oyster Working Group (SFBNOW). The handbook has also benefitted from thoughtful comments and expert input provided by numerous scientists, resource managers and restoration practitioners who reviewed previous drafts. The authors also appreciate editorial comments provided by Frances Pflieger of NOAA. Ultimately, the information and recommendations contained within this document are only possible because of the previous research, monitoring and careful documentation of outcomes from shellfish restoration projects undertaken by scores of scientists, resource managers and restoration practitioners, some of whose work is referenced herein.

Oyster Reef Restoration Monitoring and Assessment Handbook Steering Committee

Robert Brumbaugh (co-Chair) –
The Nature Conservancy

Sean Powers (co-Chair) - University of South Alabama, Dauphin Island Sea Lab

Lesley Baggett (lead author)- University of South Alabama, Dauphin Island Sea Lab

Loren Coen – Florida Atlantic University and College of Charleston

Bryan DeAngelis – The Nature Conservancy (formerly NOAA Restoration Center)

Jennifer Greene – The Nature Conservancy

Boze Hancock – The Nature Conservancy

Summer Morlock – NOAA Restoration Center

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Paul Gormont - Apertures, Inc.
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Chapter 1: Introduction

1.1 Brief Synopsis of oyster Declines Worldwide

In their assessment of the status of oyster reefs worldwide, Beck et al. (2011) estimated that 85% of oyster habitat has been lost globally and that the majority of remaining natural oyster populations are in poor condition (Figure 1. Map showing the global condition of oyster populations, with condition ratings based on the percent of current to historical abundance of oyster reefs remaining: < 50% lost (good); 50-89 % lost (fair); 90-99% lost (poor); > 99% lost (functionally extinct) (from Beck et al. 2009).Figure 1). In the United States, there has been an estimated 88% decline in oyster biomass and an estimated 63% decline in the spatial extent of oyster habitat over the past 100 years, with oyster population declines being greatest in estuaries along the Atlantic coast (zu Ermgassen et al. 2012a). For example, Wilberg et al. (2011) estimated that oyster abundance in Chesapeake Bay has declined 99.7% since the early 1800s. Although commercial landings of oysters in the Gulf of Mexico are the highest in the world (Beck et al. 2011), the region has suffered serious declines in overall oyster biomass (zu Ermgassen et al. 2012a) and abundance (Beck et al. 2011). In the United States, overharvesting is generally accepted as the primary factor in the decline of populations of both the eastern oyster, *Crassostrea virginica*, and the Olympia oyster, *Ostrea lurida*; however, other factors such as habitat loss or degradation from coastal development and dredging, non-native introductions including introduced diseases, pollution and sedimentation have also contributed in varying degrees to estuarine and regional-scale declines in oyster populations (e.g., Ewart and Ford 1993; Baker 1995; Coen and Luckenbach 2000; Beck et al 2011; Wilberg et al. 2011).

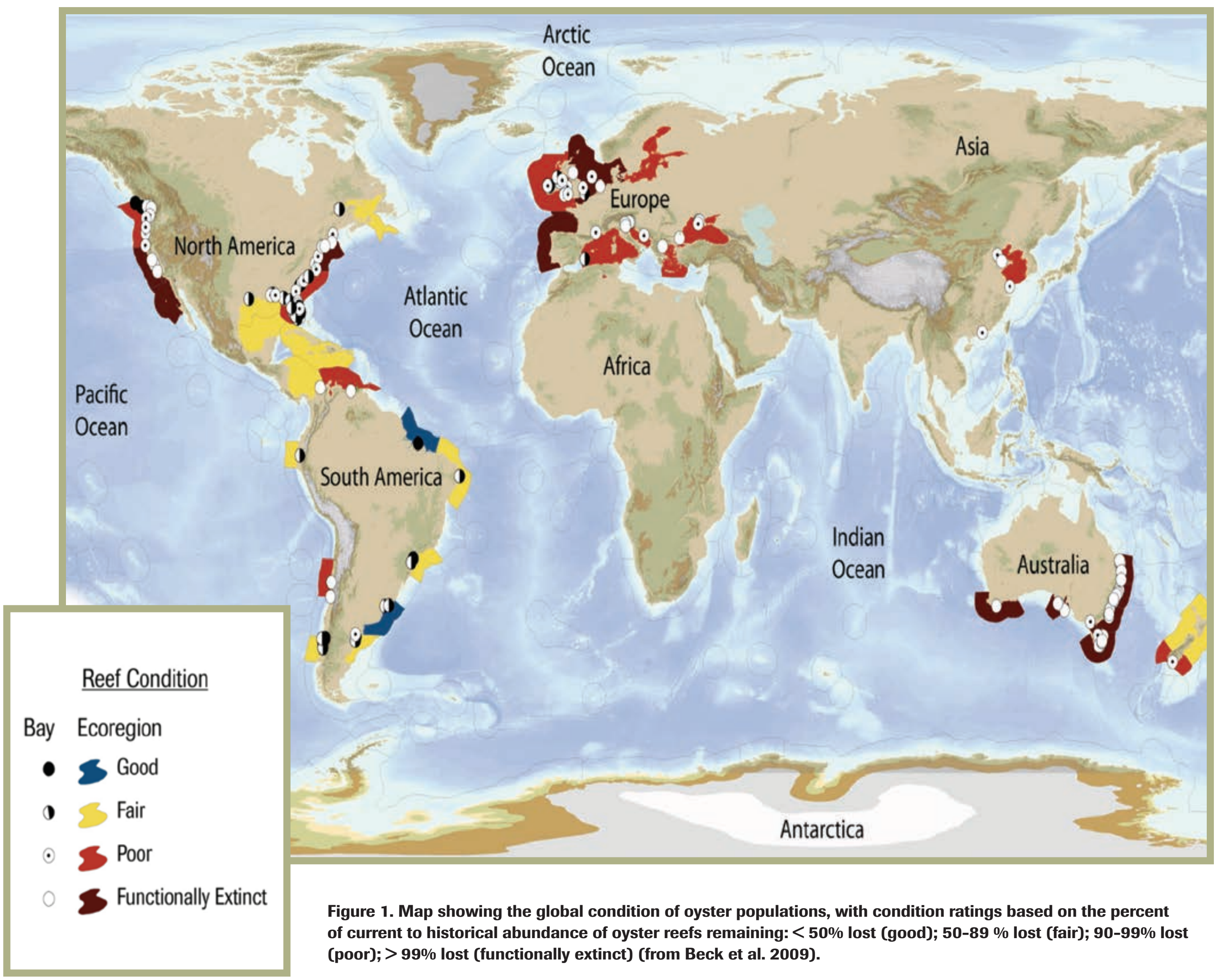

Figure 1. Map showing the global condition of oyster populations, with condition ratings based on the percent of current to historical abundance of oyster reefs remaining: < 50% lost (good); 50-89 % lost (fair); 90-99% lost (poor); > 99% lost (functionally extinct) (from Beck et al. 2009).

1.2 Oyster Restoration and Monitoring Efforts

In the past, the majority of oyster restoration efforts focused on recovering oyster fisheries and mitigating losses from natural and man-made disasters (e.g., Luckenbach et al. 2005, Coen et al. 2006; Beck et al. 2011); however, the increasing recognition of other valuable services provided by oysters has focused attention on restoring the ecological functions of oyster habitats (e.g., Peterson et al. 2003b, Luckenbach et al. 2005; Coen et al. 2007, Grabowski et al. 2012). In a review Grabowski and Peterson (2007) recognized seven ecosystem services provided by *C. virginica* habitats: "(1) production of oysters; (2) water filtration and concentration of pseudofeces; (3) provision of habitat for epibenthic invertebrates; (4) nutrient sequestration; (5) augmented fish production; (6) stabilization of adjacent habitats and shoreline; and (7) diversification of the landscape and ecosystem." The ecosystem services historically provided by *O. lurida* were likely similar to those provided by *C. virginica* and include: "(1) maintenance of a hardened substratum that served as benthic habitat for many species; (2) biofiltration of phytoplankton and sediment particles from the water column; (3) pelagic-benthic coupling resulting in enhanced secondary production of bivalve tissue and other associated organisms: and, (4) increased biotic diversity and foraging areas for invertebrates, fish, and shorebirds" (Groth and Rumrill 2009). Like other habitat restoration efforts, many oyster restoration projects now include enhanced ecosystem services provided by oyster habitat as their primary or even exclusive project goals, particularly those relating to fish habitat enhancement, enhancement of adjacent habitats, shoreline stabilization, and water quality improvements (e.g., Luckenbach et al. 2005; NRC 2007 ASMFC 2007; Coen et al. 2007; Trimble et al. 2009).

Unfortunately, many habitat restoration projects are often not monitored at all pre-construction with little post-construction monitoring to allow for: (1) comparison among restoration projects; (2) adaptive management; and (3) determination of whether their stated restoration goals were successfully achieved (Hassett et al. 2005, Clewell and Aronson, 2013). A recent analysis of the available data from oyster restoration in the Chesapeake Bay from 1990 to 2007 found that relatively few of the restoration activities were monitored, and that the restoration goals of the projects were often not well-defined (Kramer and Sellner 2009, Kennedy et al. 2011). Over 78,0000 records from 1035 sites were examined and only 43% included both restoration and monitoring. Among other things, Kennedy et al. (2011) were unable to answer basic questions from the available data concerning the success of restoration projects, the influence of scale on success, long-term trends in restoration success, and oyster disease resistance (in natural stock as well as selectively bred strains). This publication stressed the need for restoration projects to include clearly stated goals as well as quantitative sampling with adequate replication and sample sizes, and emphasized the importance of pre- and post-restoration monitoring. Unfortunately, the lack of adequate monitoring of oyster restoration activities is not limited to one region alone. However, efforts to develop local and regional oyster habitat restoration plans and protocols and criteria by which to judge the performance of oyster restoration projects on the Atlantic, Gulf, and Pacific coasts have been increasing (e.g. CSCC 2010, OMW 2011, Blake and Bradbury 2012, Boswell et al. 2012).

1.3 Development of this Handbook

To address the current problems with consistency and extent of monitoring of oyster restoration projects, a working group was formed to recommend monitoring techniques that would allow for systematic comparison among restored sites and that could be used to develop performance criteria. The working group consisted of restoration scientists and practitioners from the Atlantic, Pacific, and Gulf coasts of the US. The working group, with additional expert input, developed a set of Universal Metrics and Universal Environmental Variables[1] to be monitored for all oyster restoration projects. The working group also developed guidelines for additional Restoration Goal-Based Metrics, which are not meant to be monitored for all projects, but could be monitored depending on factors such as the current state of the science and information gaps, availability of additional funding, capacity, and expertise (Appendix 1). The Universal Metrics allow for the assessment of the basic project performance of restoration projects (e.g., reef area, height and persistence, and abundance, recruitment and size frequency of oysters), whereas the Restoration Goal-based Metrics allow practitioners to assess the performance of the restored reefs in meeting the ecosystem service-based restoration goal(s) associated with their project. Together, these metrics will allow for the comparison of restoration projects within and across regions, tidal elevations, and construction types, and will provide practitioners with valuable information towards adaptive management of restored reefs. Monitoring of an additional set of Universal Environmental Variables will aid in the interpretation of data collected through pre- and post-restoration project monitoring. Additional guidance is provided for a set of Ancillary Monitoring Approaches that can help identify problems with a site or a restoration effort and provide valuable information that can aid in the interpretation of data or help to improve subsequent efforts.

1 For the purposes of this handbook, a metric is defined as a measurement used to quantify a characteristic of a habitat, whereas a variable is a physical or environmental factor that is subject to change and may impact the habitat of study.

Universal Metrics and Universal Environmental Variables should be sampled for *every* oyster restoration project.

Restoration Goal-based Metrics are specific to ecosystem service-based restoration goals and are not sampled for every project. They may be considered for projects citing a particular restoration goal.

Ancillary Monitoring Considerations describe optional monitoring that practitioners may consider to obtain additional beneficial information associated with restoration performance.

The development of this handbook built upon previous documents that provide more general monitoring recommendations or site selection guidance (e.g., Thayer et al. 2003, 2005; Coen et al. 2004, 2006; Brumbaugh et al. 2006) by providing standardized metrics to be monitored for all oyster restoration projects, as well as suggested methodologies and guidance on the development of performance criteria. Practitioners should consult these, and other, previous publications for guidance concerning the design of oyster restoration projects and the selection of appropriate sites for restoration projects. Information regarding physical and biological parameters that should be considered in site-selection can be found in Coen et al. (2004). Brumbaugh et al. (2006) also provides information regarding site-selection, as well as design strategies for addressing certain stressors to oyster populations. Additionally, information specific to the restoration of oysters on the West coast of the United States, including site selection and project design, can be found in CSCC (2010) (available at http://www.sfbaysubtidal.org) and Peter-Contesse and Peabody (2005).

The development of this handbook also benefitted greatly from previous workshops on the topic held around the US (Atlantic, Pacific and Gulf) [see Coen et al. (2004) and NOAA Restoration Center (2007) for proceedings from two such workshops; also see http://www.oyster-restoration.org/workshops-meetings-related-to-oyster-restoration/ for a more complete listing of workshops and resulting information]. At a workshop convened by the steering committee responsible for this handbook in 2011, oyster reef restoration scientists and practitioners from around the U.S. met to discuss restoration goals, metrics and associated methodology to assess restoration performance criteria. Workshop participants also provided input on later drafts of this handbook.

Additionally, a draft of this handbook was made publically available for review over a period of several months at http://www.oyster-restoration.org/. The handbook benefited from valuable input received from a diverse group of restoration scientists and practitioners, fisheries managers, and local, state and federal entities. Drafts of the handbook were presented and discussed at five scientific conferences during 2012 (the Benthic Ecology Meeting, the Restore America's Estuaries National Conference, the Gulf Estuarine Research Society Meeting, the Bays and Bayous Symposium, and the International Conference on Shellfish Restoration). Together, these efforts resulted in a scientifically and publicly vetted handbook which provides specific and preferred guidance for measuring metrics in each category of ecosystem services, as well as universal metrics recommended for monitoring at all oyster restoration projects.

1.4 Target Audience

This handbook is intended for use by those who are relatively new to oyster restoration, experienced oyster restoration practitioners and scientists, and by restoration funding entities. Information contained within Chapter 2 will help guide newer practitioners in their efforts to monitor their restoration projects with appropriate scientific rigor; however, it is recommended that these practitioners collaborate with more experienced restoration practitioners and scientists in order to carry out scientifically sound assessments. For additional detail citations have been included as an introduction to the literature. Guidance provided by this handbook will benefit experienced restoration practitioners in that standardized monitoring will provide the data necessary for analyzing restoration trends and performance of a particular restoration project, as well as comparison across projects. Restoration funding entities should consider the metrics, both the Universal and Restoration Goal-Based Metrics, listed in this handbook when determining the amount and duration of funding provided for restoration projects.

Many oyster restoration projects incorporate outreach efforts to build federal, state, and local support for their work. These outreach efforts are vital to educating the public, resource managers, and funding entities about the many benefits that healthy oyster populations provide. Oyster restoration projects often incorporate hands-on volunteer opportunities. Volunteers can typically participate in a variety of activities in support of all types of restoration projects, ranging from bagging loose oyster shell and the placement of the bagged shell during reef construction, to both pre- and post-restoration monitoring activities (e.g., Brumbaugh et al. 2000a,b, Leslie et al. 2004; Hadley et al. 2010) (Figure 2). Although the emphasis of this handbook is on monitoring the biological and ecological aspects of oyster restoration, and not on public outreach, implementing public outreach activities should be considered an integral component of restoration projects. Evaluating public involvement and gauging the effectiveness of community participation and education efforts should be conducted to document these contributions and improve our knowledge of outreach effectiveness. Multiple restoration programs exist across the country that incorporate successful public outreach programs (see http://www.oyster-restoration.org/related-links/ for a list of some restoration programs that incorporate public outreach).

Figure 2A and B. (A) Volunteers aiding in the placement of bagged shell at Helen Wood Park in Mobile, AL. (B) A drop-off point for recycling oyster shell at the RI Dept. of Environmental Management property in Jerusalem, RI.

Chapter 2: The Oyster Habitat Restoration Monitoring and Assessment Handbook

2.1 How to use this Handbook

The intent of this handbook is to put forward a set of working guidelines that are broad enough to be applicable to natural and constructed oyster habitats around the US, but specific enough to inform the consistent monitoring of oyster restoration projects regardless of species or location. There is also substantial variation in most aspects of oyster restoration around the country. There are inherent differences between oyster species; numerous construction methods and materials employed for oyster reef restoration; differences in local reef architecture and habitat such as subtidal or intertidal reefs, fringing or patch reefs, harvesting history and age; or seasonal sea ice or freshwater flooding. As a result, some generalizations and simplifications in the definitions of restoration, oyster habitat and reef area, as well as in the suggested methodologies, have been used for this handbook. The generalized methodologies suggested in this handbook should be adapted to suit the habitat characteristics of each restoration project. The metrics and methodologies also range from fairly simple and inexpensive ones used to gather basic information to those that are relatively complicated. Despite any local variations that might be employed it is critical that there is as much consistency as possible in defining and monitoring these habitats, so that oyster restoration projects may be compared among and within regions, tidal elevations, construction types, etc.

In this chapter sections address the following: defining restoration (Section 2.2), defining natural and restored oyster habitats (Section 2.3), guidance for determining which of the various non-universal metrics should be sampled under various scenarios (Section 2.4), information on how to collect baseline data and on sampling control and reference sites (Section 2.5), basic information on how to sample Universal Metrics (Section 2.6), and an introduction to Basic and Restoration Goal-Based Performance Criteria (Section 2.7). Subsequent chapters present the metrics and suggested methodologies for the Universal Metrics (Chapter 3), Universal Environmental Variables (Chapter 4), Ancillary Monitoring Considerations (Chapter 5), and Restoration Goal-Based Metrics (Chapters 6-9).

2.2 Defining Restoration

For the purposes of this handbook, restoration is defined as:

"The process of establishing or reestablishing a habitat that in time can come to closely resemble a natural condition in terms of structure and function." (modified from Turner and Streever 2002)

This definition includes activities aimed at returning degraded oyster habitat to its prior condition, and the construction of new oyster habitats of various forms and construction materials, either natural or man-made (Haven et al. 1987; Dugas and Berrigan 1991; Luckenbach et al. 1999; Coen and Luckenbach 2000; Soniat and Burton 2005; Street el al, 2005; Brumbaugh et al. 2006). In the interests of consistency and brevity, all oyster habitat restoration or construction projects that fall within this definition, including those involving species that form beds rather than reefs (such as the Olympia oyster, *Ostrea lurida*), will be referred to as "restored reefs" in this handbook. This definition, however, does not include restoration in a fisheries management context.

2.3 Defining Oyster Habitat

2.3.1 Natural Habitats

Oyster reefs and beds may be intertidal or subtidal biogenic structures formed by oysters living at high densities and building a habitat with significant surface complexity (Galtstoff 1964; Chestnut 1974; Bahr and Lanier 1981; Holt et al. 1998; ASMFC 2007). Reefs are defined here as having significant vertical relief, >0.2 m above the surrounding substrate, while beds have lower relief, <0.2 m (Beck et al. 2009). There appears to be some species specificity to habitat formation, with certain species, such as the eastern oyster (*Crassostrea virginica*) tending towards building reefs while European and Olympia oysters (*Ostrea* spp.) are more commonly described as building beds (Peter-Contesse and Peabody 2005; ASMFC 2007; Jacobsen 2009; Meyer et al. 2010). Overall, such structures are accreting through the continuing deposition of shell material which is in turn degraded at varying rates (Powell et al. 2006; Mann and Powell 2007; Powell and Klinck 2007;

Figure 3A and B. (A) Fringing reef on Ossabaw Island, GA. (B) Patch reef in Chesapeake Bay.

Figure 4. Intertidal patch reef with oyster clusters on a muddy substrate, NC.

Green et al. 2009). These "shell budgets" are critical to developing carbonate dominated habitats (see Powell and Klinck 2007, Powell et al. 2006, 2012; Waldbusser et al. 2013). In some places it is likely that vertical accretion may be restricted by tidal exposure, leaving a non-accreting reef.

An oyster reef system is an area of ecologically connected reefs or beds and oyster shell dominated bottom, and may include small areas of bare mud, sand or shelly substrates or seagrass where these are considered integral to the overall ecology (discussed in ASMFC 2007; Coen et al. 2011a) (Figures 3 and 4). While reefs are normally an integral part of such diverse landscapes (Eggleston 1999; Eggleston et al. 1999; Micheli and Peterson 1999; Harwell et al. 2011; Puckett and Eggleston 2012), areas of oyster shell bottom with low densities of live oysters (1-10 m^{-2}) can also be classified as reef systems, noting that it may not be possible in many areas to differentiate natural from anthropogenic impacts or recovering versus degrading systems.

2.3.2 Restored Habitats

Oyster restoration projects can take many forms and use a variety of construction materials (Brumbaugh et al 2006). While the aim of some projects is to restore a natural but degraded reef to some former condition, other restoration projects involve constructing an entirely new reef structure, though often at historic reef sites[2]. Construction materials may include unconsolidated clean[3] shell (cultch), bagged clean shell, limestone or fossil shell, engineered concrete domes, metal shell containment structures filled with loose or bagged oyster shell, or other similar materials. Construction processes range from the direct placement of cultch or other construction materials to form a reef of a specific design and size, to spraying unconsolidated shell off barges across a project area, resulting in a reef with large spatial extent, but varying vertical relief and cultch density (Figure 5). When designing an oyster restoration project, practitioners should

2 Note that often so many changes have occurred since reefs were present that selecting a site should not hinge on past presence alone (Coen and Luckenbach 2000, Coen et al. 2004).

3 Shell should not contain any soft tissue to protect against disease transfer (see Bushek et al. 2004).

Figure 5A, B and C. (A) Limestone marl being placed as reef substrate in Matagorda Bay, Texas. (B) NC DMF vessel delivering marl to restoration sites. (C) Using a water cannon to spread oyster cultch over a restoration site in Dogfish Bay, WA.

consider local conditions in selecting the construction material and reef design, and should choose a site that maximizes the chance of successful restoration (for site selection considerations see Coen et al. 2004, Brumbaugh et al. 2006, Brumbaugh and Coen 2009, CSCC 2010, and http://www.oyster-restoration.org for additional information). Additionally, restoration projects should be designed so that the appropriate metrics can be assessed and statistical analyses can be performed on the resultant data.

2.4 Deciding What to Sample

The Universal Metrics (Chapter 3; Appendix I) and Universal Environmental Variables (Chapter 4, Appendix I) should be sampled for *every* oyster restoration project, regardless of the restoration goal(s) of that project. Sampling of the Universal Metrics allows for the basic performance of each reef to be assessed through time, while also allowing for comparisons of Universal Metrics with other projects across the US. Sampling of the Universal Environmental Variables also provides important information that can aid in the interpretation of data collected during reef monitoring activities. The timing of measuring some Universal Environmental Variables, particularly dissolved oxygen concentration, should be considered as they can vary over short time periods.

In addition to the Universal Metrics, this document contains guidance on some Restoration Goal-Based Metrics that can be used to monitor Ecosystem Service-Based Project Goals (see Chapter 6: Brood Stock and Oyster Population Enhancement; Chapter 7: Habitat Enhancement for Resident and Transient Species; Chapter 8: Enhancement of Adjacent Habitats; and, Chapter 9: Water Clarity Improvement) (Appendix I). If individual project goals are similar to the ecosystem service-based goals described within this handbook, then those metrics should be considered for monitoring, depending on factors such as: the availability of additional funding, the state of the science, capacity, and expertise. Additionally, guidance is provided for a set of Ancillary Monitoring Considerations (Chapter 5), which could provide supplemental and/or more detailed information concerning the performance of a given project.

Note that the monitoring protocols included in this handbook represent the **minimum recommended** level of sampling. Practitioners are encouraged to sample additional metrics and/or at increased frequencies and duration, if considered appropriate. Practitioners should fully consider the level of effort associated with any of the goal-based metrics when planning projects and applying for funding. While numerous benefits may result from additional sampling, restoration goals need to be based on realistic budgetary and capacity considerations. This handbook is not meant to discourage additional goal-based monitoring, but to make all parties cognizant of the level of effort and rigor necessary to properly assess the performance of the project in meeting any stated goals.

2.5 Baseline Data and Control and Reference Sites

To determine the ecological impact and to assess the performance of a project in meeting its restoration goals, it is necessary to perform both pre- and post-construction monitoring, with the inclusion of monitoring at a control site and a reference site if available (Aronson et al. 1993a,b; SER 2004). Pre-construction monitoring at a proposed restoration site is recommended in the year prior to construction to aid in site-selection (e.g., Coen et al. 2004; Burrows et al. 2005) and to document conditions (hydrological, ecological, etc.) before reef construction. Practitioners should also perform sampling at a control or natural reference site concurrently with sampling at the restored reef. Control sites are unaltered areas that mimic the pre-restoration conditions (e.g., sand or mud substrate) whereas natural reference sites, if available, would optimally be healthy, natural reefs characteristic of the restoration goal (Kennedy and Sanford 1999; Beck et al. 2011; zu Ermgassen et al. 2012a). In this context, control areas would allow for determination of the degree of local enhancement resulting from the project and reference areas could be used to determine if the restored reef is performing to the level of a healthy natural reef. Control and natural reference sites should have physical characteristics (e.g., flow, wave action, tidal range and exposure, salinity, proximity to open water, water temperature, freshwater influence, substrate type, water depth, etc.) similar to the restored site. In many areas a natural reference site will not be available.

If pre-restoration monitoring is not an option, the comparison between the restored and control sites is essential. This comparison should be augmented with comparison to a natural reef if available (Coen and Luckenbach 2000; O'Beirn et al. 2000).

For all metrics, sampling should be performed at the restoration site and a control and/or natural reference site in the year prior to construction, and during post-construction monitoring

2.5.1 Comparison with Control (unrestored area) Sites

Comparisons with control (unrestored) areas are best performed using a BACI (Before-After-Control-Impact) experimental design (Figure 6. Stewart-Oaten et al. 1986, Underwood 1994, Smith 2002). The BACI design entails monitoring at a control site (unrestored) and impact site (location of oyster restoration) both before and after the construction of the oyster reef. Pre-construction monitoring should be conducted at both sites prior to construction of the reef and post-construction monitoring be conducted long enough to encompass both the short (e.g., 1-2 years) and mid-term (e.g., 4 to 6 years) post-construction time frames (see Section 2.7 for further discussion).

2.5.2 Comparison with Natural or Reference Reefs

In areas where relatively healthy natural reefs are still present, it may be possible to compare the restored oyster reef(s) to a nearby existing natural reef(s) that is not subject to harvest (Coen et al. 1999a; Kennedy and Sanford 1999). As with the control site examples, a BACI design should be used, which requires that pre-construction monitoring be conducted at both reference and restoration sites prior to construction of the reef, and that post-construction monitoring be conducted long enough to encompass both the short (e.g., 1-2 years) and mid-term (e.g., 4 to 6 years) post-construction timeframes.

2.6 Sampling Techniques

For each of the metrics listed in this handbook, information is provided regarding; (1) required units for data collected, (2) suggested methodologies, and (3) frequencies of sampling. *It is imperative that data are recorded using the required units and with a mean and variance so that data may be compared among projects*. The methodologies listed for each metric

BACI Experimental Design

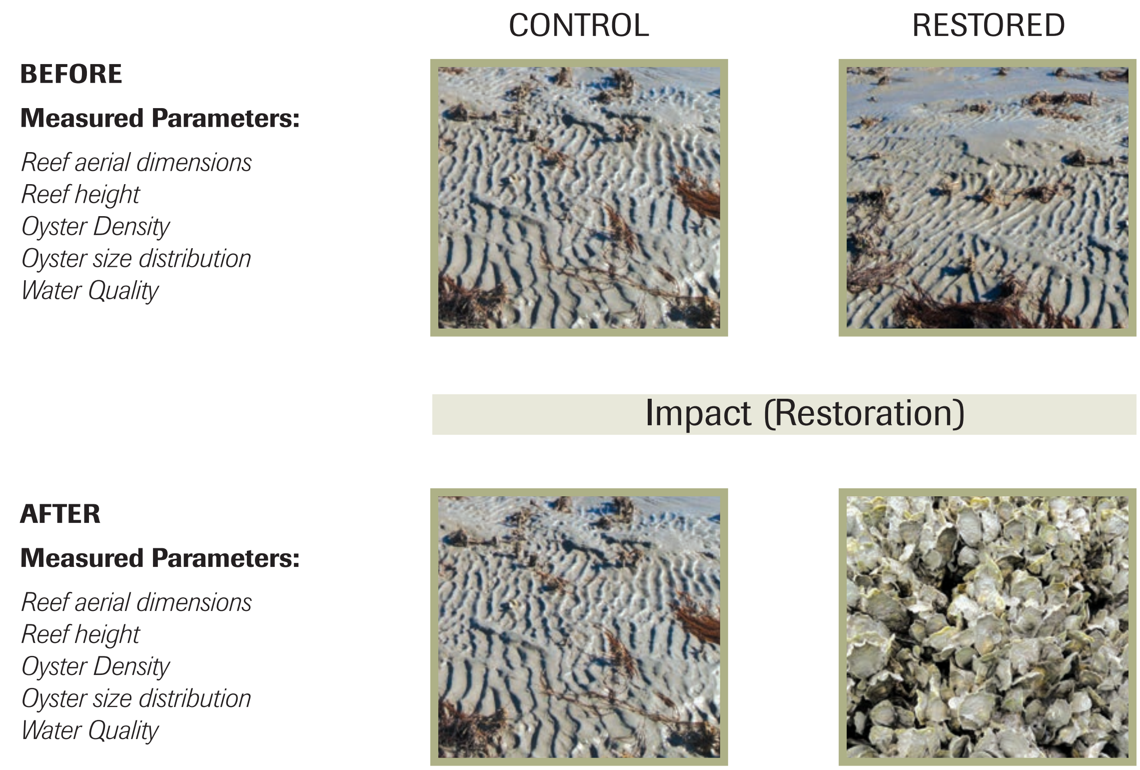

Figure 6. A visual representation of an example of a BACI (Before-After-Control-Impact) design experiment (see references above for more details).

are noted as 'suggested' because the high degree of variation among individual restoration projects makes it difficult to recommend a "one-size-fits-all" methodology for each or all of the metrics suggested here. Suggested methodologies in this handbook often include low (e.g., Grizzle 1990) and high-tech alternatives (e.g., Grizzle et al. 2005, 2008), as well as alternatives for various reef construction approaches and tidal elevations. If unable to use the suggested methodologies, alternative methodologies that have equal rigor and that can provide data in the specified units for that metric should be used. Projects should establish a statistical design for data analysis prior to construction.

For universal metrics data must be recorded using the required units. Where a mean is calculated a variance also needs to be provided, commonly provided as a standard error (Mean $\pm$ 1 SE).

The sampling frequency stated for a given metric is the minimum amount of monitoring necessary for comparability and statistical rigor, and it is encouraged that practitioners monitor more frequently and longer if that improves the resolution of their results.

The stated sampling frequency is the minimum amount of monitoring suggested, and practitioners are encouraged to monitor more frequently and over a longer timeframe where possible.

2.6.1 Random Sampling

To eliminate bias when collecting samples, it is best to use a true random sampling method in which each potential sampling point or location has an equal probability of being selected and that samples taken from one location do not influence samples in another location. Samples should also be 'independent' so they cannot be considered replicates or pseudoreplicates. A common way to perform random sampling is to determine the area to be sampled, then superimpose a numbered grid on an aerial photo or a diagram of the area to be sampled (e.g., reef area), then randomly select the locations to sample using a random number generator (Figure 7). Random number tables can be found online (e.g. http ://www.random.org/; http://www.randomnumbergenerator.com/; http://www.randomnumbers.info/) and in most statistics textbooks (e.g., Hurlbert 1984, Stewart-Oaten 1986; Coyer et al. 2011).

If true random sampling is not possible, then sampling should be conducted in a manner that does not bias the results (e.g., a bias would result from placing quadrats on only the 'best' parts of the reef). Common methods include 'haphazard sampling' [e.g., facing away from the reef and tossing a quadrat over the head and sampling wherever the quadrat lands, or sampling on a more systematic basis with samples being collected at pre-determined spacing intervals (e.g., every 2 m)] based on area to be sampled. In some instances it may be necessary to perform fixed sampling, in which the same marked locations are sampled at every monitoring event. If this is the case the initial sample locations that are revisited at subsequent monitoring events must be selected with the above considerations in mind. While the non-random methods are not as robust as true random sampling, they can be acceptable in situations where true random sampling is not possible.

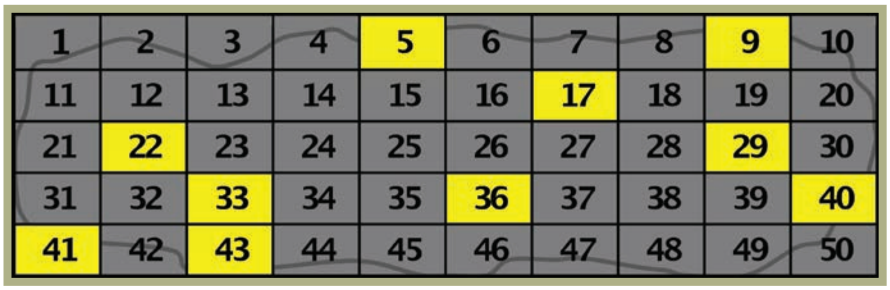

Figure 7. An example of a numbered grid superimposed on a diagram of a reef (in grey), with randomly selected sample locations noted in yellow.

2.6.2 Stratified Random Sampling

In some situations sample locations should be divided into strata. In this method, strata are delineated based on what the practitioner perceives as the source of variation in the target metric such as oyster density (e.g., oyster density might vary with reef height, distance from shore, orientation to mainland, percent coverage of cultch, etc.). Stratification recognizes that oyster reefs are typically not monolithic structures with oysters distributed uniformly throughout the project footprint. *A detailed explanation of how to assess oyster populations, size, recruitment abundance, etc., and the considerations for accurate estimates are provided in Chapter 3.*

The area of each stratum is defined and using the same grid method described above a set number of random sample locations are chosen within each strata. When performing stratified random sampling, the stratum in which each sampling point is located should be noted for analysis purposes.

For additional guidance we recommend that the restoration practitioner consult with a statistician or experienced ecologist, perhaps at a nearby academic institution, to assist with some of the more complex designs and analysis (e.g., Walters and Coen 2006).

2.6.3 Quadrat Sampling

Throughout the handbook, several methodologies are suggested that require the use of quadrats to obtain samples, with one quadrat equaling one sample or replicate. Unless otherwise noted, practitioners may select a quadrat size that is best suited for their particular project's characteristics. In general, it is suggested that a 0.25 m^2 quadrat (e.g., 0.5 m x 0.5 m) be used to collect samples; however, a larger or smaller size quadrat may be used depending on the density of oysters. For example, a larger 1 m^2 sized quadrat may be used in areas of low oyster density, below about 100 oysters m^{-2}, a 0.5m^2 quadrat could be used for moderate densities of 100 to 500 oyster m^{-2} and a smaller sized quadrat (e.g., 0.0625 m^2) in areas with high oyster densities above about 500 m^{-2}. Quadrat size should ultimately be a function of optimal statistical rigor (i.e., lowest standard error or narrowest confidence interval of the mean) and logistical considerations (e.g., time to process samples). Throughout the handbook quadrats will simply be referred to as "the quadrat," rather than by a specific size. For more information on methods to allow for more rapid work-up of samples see http://www.oyster-restoration.org/ .

2.6.4 Determining Sample Size

Oyster restoration projects vary in extent, construction material, morphology, form, and tidal depth. As such, the number of samples (e.g., quadrat samples, core samples, lift net samples, etc.) to be taken per reef for each metric will be different for each restoration project. Monitoring costs, particularly by advanced methods, can be high, especially when large areas need to be assessed. Attempts are therefore often made to keep sample replicates to a minimum. In some instances, however, accurate estimates can require large samples sizes (e.g. estimates of mean abundances in highly patchy populations; see Chapter 3 for further explanation). Sample size is ultimately a function of the logistical and financial feasibility of a given project. Proper replication is essential when sampling both the Universal and Environmental variables and Restoration Goal-based Metrics. Determining the relationship between sample number and sample variance will aid in determining the optimal number of samples that should be collected.

One sample is equal to one quadrat, one substrate basket or tray, one setting of a gillnet, etc. Sample size (n; the number of samples to be taken per reef) can be determined using the following equation (e.g., Quinn and Keough 2002):

$$n = z_\alpha^2 \sigma^2 / d^2$$

Where n is sample size, α is the significance interval (most often 0.05), z_α is the z-value from a standard normal distribution for the chosen α (1.96 for α=0.05), σ^2 is the variance of the population, and d is the maximum allowable absolute difference between the true population mean and the estimated population mean, often 30% of the sample mean (Quinn and Keough 2002). Sigma (σ) is usually unknown but may be estimated from the standard deviation (SD) of pilot samples (assuming that you are covering the range of densites or other measure with this sampling). To estimate $\boldsymbol{\sigma}$, take a minimum of five (replicate) pilot samples (see suggested methodologies for each metric) and determine the mean and standard deviation of the data obtained from these pilot samples. Obtain the variance (σ^2) by squaring the standard deviation, and then use this calculated variance in the above equation. Please note that sample size will be different for each of the metrics, so calculate sample sizes separately for each metric based on the pilot data obtained for that particular metric at each site. Based on standards that are commonly accepted in fisheries literature [confidence interval (CI) of 95%, with a maximum allowable distance (d) of 30% of the mean and α of 0.05] enough samples should be collected to ensure that the coefficient of variation (CV), which is the ratio of the standard deviation to the mean, is approximately 0.5.

Example sample size calculation:

Oyster densities (oysters/m^2) from five pilot samples: 16, 26, 35, 47, 64

Mean = 37.60; σ = 18.66, CV = 0.50

z_α for α of 0.05 and a 95% confidence interval = 1.96

d = 0.30 x 37.60 = 11.27

n = $(1.96^2 \times 18.66^2)/ 11.27^2$

n = 10.53 (sample size would be 11)

Sample sizes may also be calculated using various statistical programs and online sample size calculators can be found on various websites. The sample size will need to be re-calculated if there is any indication that the density (or other variable) at a site differs from the mean obtained from the pilot samples.

2.7 Assessing the Performance of an Oyster Restoration Project

Adopting a truly 'universal' set of performance criteria[4] for oyster restoration projects is difficult due to the variation in restoration projects, as well as differences in reef characteristics, depth, region, and species differences in life histories. As such, performance criteria will need to be defined on a per-project basis using the guidelines described in this section.

4 Performance criteria are tangible, measurable objectives to be accomplished within a proposed timeframe that indicate progress toward meeting the project goals. The criteria should include a **metric**, **target value**, and **timeframe**. Performance criteria may represent conditions at a reference site, and/or they may represent target conditions considering the surrounding land use or other local conditions. Performance criteria may also be known by other names (e.g., success criteria, performance standards, etc.).

2.7.1 Timeframe of Post-implementation Monitoring

During the project development period, practitioners should create a timeline for sampling and project performance that describes how they anticipate the project should progress over both the short- and mid-term timeframes. The short-term timeframe refers to the **minimum** monitoring period of one to two years post-construction and should include at least two recruitment phases (see Chapter 3.3 for information describing recruitment versus settlement). This timeframe is often dictated by the funding period of the project, and may be the only post construction monitoring feasible depending on funding constraints. The commencement of funding does not always coincide with the timing of larval settlement, so some knowledge of local recruitment periods (e.g., through pre-restoration pilot data) is important when determining the timing of project construction and subsequent monitoring. In many locations May to October is the period that covers larval settlement, though this period may be shorter at higher latitudes.

Unfortunately, the short-term timeframe, often dictated by funding, may not adequately capture the dynamic changes a restoration project may undergo beyond the funded years. The mid-term time frame is the period approximately four to six years post-construction, and is the preferred minimum timeframe for post-implementation monitoring. The mid-term timeframe is more of an ecologically meaningful period in which to assess performance. When preparing timelines, practitioners should address how the project will perform with regard to basic criteria (i.e. reef persistence, presence of live oysters, successful recruitment with multiple year classes) as well as criteria related to the ecosystem service-based restoration goal(s) of the project.

The **short-term timeframe** is the **minimum** post-implementation monitoring timeframe, and refers to the period one to two years post-construction which should include at least two recruitment phases.

The **mid-term timeframe** is the **preferred** post-implementation monitoring timeframe, and refers to the period approximately four to six years post-construction. The mid-term timeframe is more of an ecological timeframe by which to assess performance.

2.7.2 Basic Performance Criteria

Performance criteria should be established at the beginning of a given project for the purposes of assessing progress toward meeting the stated project goals, and allowing for adaptive management of the project or integration of lessons learned in the future (Brumbaugh and Coen 2009). Performance criteria are not meant to be a “grade” by which a project passes or fails, and should not be viewed as such. When developing basic performance criteria, practitioners should frame their goals as testable hypotheses with regard to data gained from either pre-construction baseline sampling or data obtained from sampling at control and/or natural reference sites. Basic project performance should be determined using appropriate statistical analyses, and as such, goals should be framed in a way that allows for these analyses.

The basic performance of a given restoration project should be determined by monitoring the Universal Metrics (Chapter 3) and is based on (1) the persistence of emergent structure (as assessed by reef height), (2) the density of oysters on the reef, and (3) evidence of successful recruitment (Coen et al. 2004; Burrows et al. 2005; Luckenbach et al. 2005; Powers et al. 2009) (more detailed information concerning basic performance criteria is included for each of the Universal Metrics in Chapter 3).

2.7.3 Restoration Goal-Based Performance Criteria

Performance criteria guidelines for Restoration Goal-based Metrics are listed with each individual metric. As with the basic performance criteria, practitioners should frame their goals as statistically testable hypotheses with regard to data gained from either pre-construction baseline data or data obtained from sampling at control and/or natural reference sites, and project performance should be determined using appropriate statistical analyses. For example, a restoration project whose goal was to improve water clarity could state their performance criteria as follows: “*We predict that chlorophyll a levels will be significantly lower at the restoration site than at the control site at both the short-term and mid-term monitoring milestones under comparable conditions*.”

Chapter 3: Universal Metrics

Monitoring of the Universal Metrics allows for the assessment of the basic performance of restoration projects (i.e., reef persistence, density and abundance of live oysters, successful multi-year recruitment) and the comparison of restoration projects within and across regions. The Universal Metrics should be sampled for **every** oyster restoration project, regardless of the restoration goal(s) for the project. There are four Universal Metrics: (1) reef areal dimensions; (2) reef height; (3) oyster density, and (4) oyster size-frequency distribution (Appendix I).

Universal Metrics

1. Reef areal dimensions
2. Reef height
3. Oyster density
4. Oyster size-frequency distribution

3.1 Universal Metric #1: Reef Areal Dimensions

The accurate determination of the reef areal dimensions is critical to estimating the amount of restored area, reef persistence through time, oyster population abundance and ultimately the quantity of the ecosystem services provided by the restored oyster reef (Coen and Luckenbach 2000; Grabowski and Peterson 2007; Grabowski et al. 2012; OMW 2011). Complications with comparing reef aerial dimensions, and thus the related oyster population estimates, are likely to arise across projects. Uncertainties typically arise because oyster reefs, both natural and constructed, are generally not monolithic structures with oysters distributed uniformly. *Also, practitioners need to recognize that because of the way that cultch or seed is placed, or the natural movement of reef construction material, the area initially designated and/or targeted for the restoration project may not precisely match the area(s) with restored oysters.* The reef areal dimension metric consists of two separate measurements, **project footprint** and **reef area** (Figure 8):

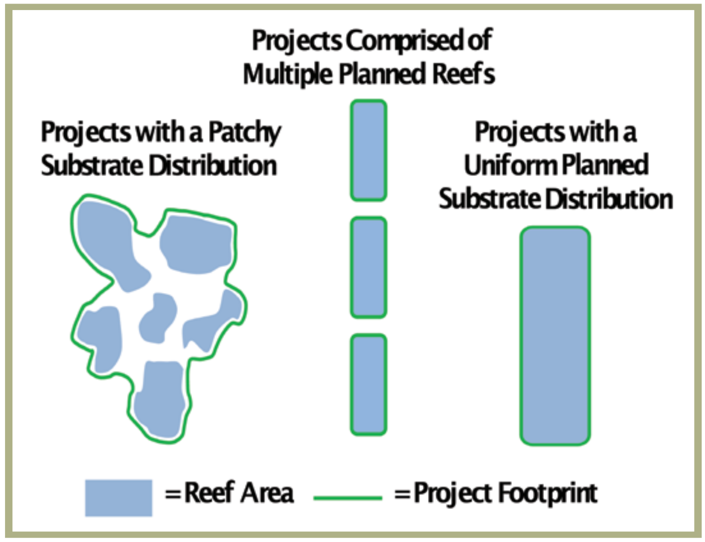

Figure 8. Examples of project footprints and reef areas in projects with a patchy substrate distribution (left), projects comprised of multiple, distinct planned reefs (middle), and projects with a single uniform substrate distribution (right). The green line denotes the outline of the project footprint while the blue shaded polygons denote the reef area. Note: the area originally targeted for the oyster restoration project (not shown here) may not precisely match the actual area with oysters.

The **project footprint** is *the maximum areal extent of the footprint of the reef.* This measurement ignores the possible initial patchiness of multiple smaller reefs that may be located within the 'reef complex' (Figure 8). Practitioners should note the project footprint may not precisely match the area originally designated and/or targeted for restoration, or the contractors assessment if deploying shell below the water line (e.g., spraying shell off a barge).

Reef area is *the actual area (summed) of patches of living and non-living oyster shell (or other construction material with and without live oysters) within the project footprint* (Figure 8). In some cases, the project footprint and the reef area may be the same. However, when patches occur either by project design or form inadvertently as unconsolidated shell is deployed, the project footprint area and actual reef area may be quite different.

Project footprint is the maximum areal extent of the reef.

Reef area is the actual area (summed) of *patches* of living and non-living oyster shell (or reef substrate with and without live oysters) within the project footprint.

Reef edge is defined as a continuous line where the percent coverage of substrate, either living or non-living material remaining above the sediment, is equal to or greater than 25%.

For determination of reef area, the edge of the reef is defined as a continuous line where the percent coverage of surficial living or non-living shell substrate (or alternate material) is equal to or greater than 25%. The original design of a restoration project should be considered when determining the areal dimensions of the restoration project (Figure 8). For example, if the original project design included three distinct reefs located several hundred meters apart, then it may be appropriate to consider them as three separate project footprints. As another example, if the original project design involved unconsolidated cultch deployed over a large area resulting in a patchy cultch distribution, then the project footprint may be the entire area over which the cultch was deployed and the reef area will be the summed areas of the individual patch reefs within that project footprint.

Required Units = m^2, Note accuracy of the measuring device (± 0.5m).

3.1.1 Suggested Methodologies for assessing reef dimensions

For Intertidal Reefs:

Project footprint: At a negative low tide, make continuous measurements while walking along the perimeter of the project footprint using at least a standard handheld GPS [differential GPS (dGPS) or a mapping/survey grade GPS with post-processing will increase accuracy] (Figure 9, Figure 10). Collect as many GPS locations as possible as the perimeter is walked; large numbers of data points may be required to accurately define the reef perimeter. Coordinates should be entered into mapping software (e.g., ArcGIS, ESRI, Redlands, CA), and the area of the plotted project footprint determined and reported in m^2. It may also be helpful to place poles or markers for reference when re-assessing the footprint (Twichell et al. 2007; SCDNR 2008; Seavey et al. 2012).

Figure 9. Determining project footprint and reef area using a backpack dGPS unit.

Reef Area: At a negative low tide, make continuous measurements using a GPS while walking the perimeter of each distinct patch reef, or reef substrate with and without live oysters (as above). Coordinates for each should be entered into mapping software (e.g., ArcGIS, ESRI, Redlands, CA), and the area of each patch determined and reported in m^2. For total reef area, sum the areas of the individual patches within the project footprint.

Alternative Methodology 1: The area of the overall project footprint and the area of the individual patches can also be determined using a surveyor's measuring wheel, laser rangefinder or transect tape (measure the perimeter, and maximum length and width to the nearest 0.5 m). Area measurements gained with this method may not be as accurate since patches are typically not uniform or easily delineated. Practitioners should still use a GPS to determine the latitude and longitude of the central locations of each patch reef and the total project footprint.

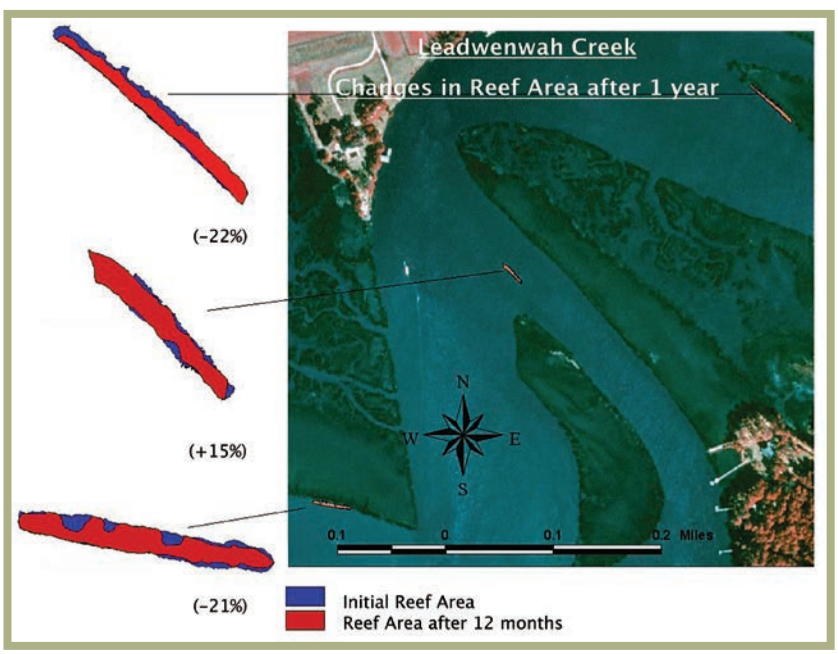

Figure 10. Changes in reef footprint through time on three restored intertidal reefs in SC documented using Trimble dGPS and GIS software (modified from Coen et al. 2011b).

Alternative Methodology 2: Estimates of the size of the project footprint and reef area may be obtained from high resolution aerial images of natural or constructed reefs. If using aerial imagery, use appropriate geo-referencing tools (e.g., permanent base stakes with markers that are visible from the air, or the presence of a permanent feature such as a dock) for scaling purposes. If possible, ortho-rectified aerial photographs (geometrically corrected to have a uniform scale) should be used. Practitioners should still determine the latitude and longitude of the central locations of each patch and the total project footprint.

For Subtidal Reefs:

Project footprint: The perimeter of the project footprint can be determined using either a side-scan or multi-beam sonar (Figure 11). Most side-scan and multi-beam sonar instruments will acquire GPS coordinates with the sonar data and output these data into one file which can then be exported into a mapping software package (e.g., ArcGIS or ERDAS Imagine); however, if the instrument does not have this capability, be sure to make continuous location readings using a GPS (see methodology above) while sonar readings are being taken. It is important to note that side-scan sonar does have some limitations, particularly in areas with high reflectivity. The lower frequencies of most side-scan sonar instruments can penetrate sediment layers and may indicate an exposed shell bottom where it is not actually present. For this reason, additional ground-truthing (taking bottom samples at regular intervals or diver investigations to determine presence/absence of reef substrate) is needed when using side-scan sonar to check the accuracy of the sonar readings.

If the water is too shallow for the use of a side-scan sonar, but still shallow enough to see the shelly bottom, mount a GPS in a fixed position in a kayak, canoe or other small craft and walk, paddle or pole the craft along the perimeter of the project footprint while taking continuous measurements with the GPS.

Reef area: Once the project footprint has been determined, run transects in a grid pattern (see below for determination of proper lane width). Use side-scan or multi-beam sonar while making continuous location readings using a GPS (if the sonar instrument does not provide continuous GPS readings) to determine location and area of individual patches within the project footprint.

For side-scan sonar, lane width is determined using the following formula:

$$\text{Lane width} = \text{Range} - (\text{Altitude} + \text{Overlap})$$

Where range is the scanning range of the instrument per channel (check manufacturer's specifications), altitude is the altitude (height) of the towfish above the seafloor (ideally this would be a value equal to 10% of the range), and overlap is the desired overlap between lanes (a suggested overlap is 10%).

For multi-beam sonar, lane width is determined using the following formula:

$$\text{Lane width} = \text{Total range} - \text{Overlap}$$

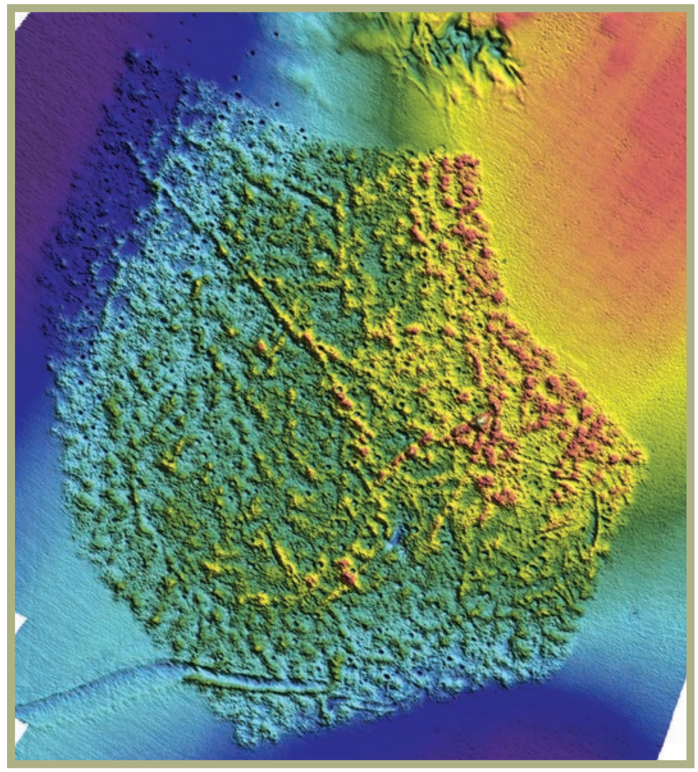

Figure 11. A mosaic of images obtained using multi-beam sonar showing the 'as-built' placement of cultch for reef restoration in Chesapeake Bay, VA.

Where total range is determined by the frequency used (generally 3 times the water depth, but check manufacturer's specifications) and overlap is the desired overlap between lanes (a suggested overlap is 10%).

If side-scan or multi-beam sonar are not available, and water depth is too great or visibility is too poor to see the bottom from the surface, determine project footprint and reef area using a quality echo sounder (a side-imaging depth finder that utilizes side-scan sonar technology) or a depth finder to detect changes in bottom relief in conjunction with a pole, tongs or other methods to verify the presence of shell (or other reef construction material). Some of these acoustic instruments will take continuous GPS measurements in conjunction with their readings; however, if the instrument does not have this capability, make continuous measurements of the perimeter of the reef complex using a GPS. Once the project footprint is determined, run transects in a grid pattern (see previous discussion of determination of proper lane width) taking bottom samples at regular intervals (2 m or greater, depending on depth and project footprint) using a pole or tongs to determine presence/absence of shell or other material. Locations of samples should be marked using a GPS.

Alternative Methodology: If the above technologies are not available, and the water depth is too great or water visibility is too poor to see the bottom from the surface, the area of the overall project footprint and the individual patches can also be determined by SCUBA using a transect tape (take measurements of length, width, and perimeter to the nearest 0.5 m). Area measurements may not be as accurate since patches are typically not uniform or easily delineated. Practitioners should still, however, use a GPS to determine the latitude and longitude of the central locations of each reef area and the total project footprint.

In all methodologies, to aid in the proper determination of project footprint (as well as reef area), SCUBA divers may swim the perimeter of the project footprint (or reef areas) and mark the boundaries at regular intervals with survey stakes that extend above the water surface or with floats. The locations of these markers should then be marked using a GPS.

3.1.2 Sampling Frequency

Measurements should be taken once prior to construction at both reference and construction sites, within three months post-construction (to document the as-built project footprint and reef areas), and one to two years post-construction. Ideally, measurements should also be taken four to six years post construction, with additional measurements taken after events that could alter reef area or oyster survival (e.g., hurricanes).

3.1.3 Performance Criteria

Performance Criteria = None

There are no performance criteria for this metric since gains in the project footprint and reef area may be due to the spreading of the original construction material (e.g., cultch) and not the accretion of the reef. However, documenting change in areas over time is important to assessing reef performance.

3.2 Universal Metric #2: Reef Height

Reef height is a measure of the mean height of the reef above the surrounding substrate (in relation to the substrate immediately adjacent to the reef, not the shoreline). Along with project footprint and reef area, measurement of reef height provides valuable information regarding changes in the reef over time, such as the persistence of a reef after construction (after possible subsidence and/or spreading) as well as the habitat provided for resident and transient finfish and invertebrate species. *In addition to reporting the mean reef height, also report the minimum and maximum reef heights*. If documenting more detailed information concerning the surface topography or rugosity of the reef is important these more detailed measurements can be taken; however, they are not Universal Metrics (Risk 1972; McCormick 1994; Commito and Rusignuolo 2000; Proffitt et al. 2013).

Required Units = m (cm for low relief reefs), Note the accuracy of the measuring device (in cm). For mid- and high relief reefs, devices that measure to a finer scale can be used however, report the data in m.

3.2.1 Suggested Methodologies

For Intertidal Reefs:

If available, equipment such as a total GPS station or RTK GPS (instruments used to find horizontal and vertical angles and distances) may be used to create a three dimensional graphic of the reef area and height. If this equipment is not available, other traditional survey equipment (e.g., level and rod, or transit pole and self-leveling laser) can be used to measure reef height[5] (Figure 12) . Take measurements every 1 m along the crest of the reef (every 5 m if reef length is >200 m). If the reef does not have a discernible crest, then take measurements along an approximate center line of the long axis of the reef. For ease of sampling, all measurements should be taken during low tide.

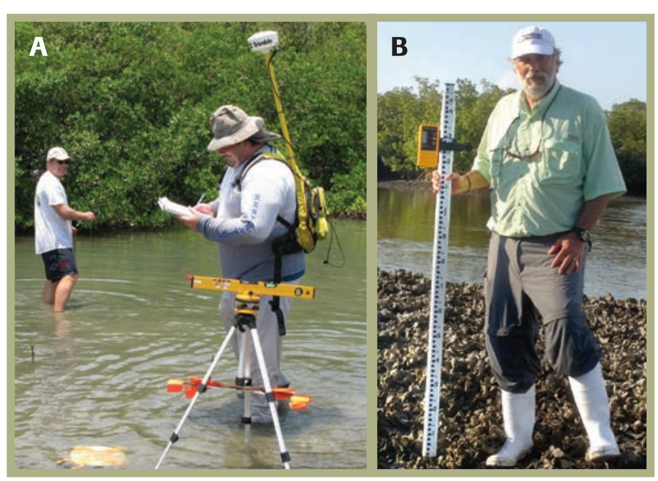

Figure 12. (A & B) Using a tripod-mounted laser level to measure reef height.

5 Performance criteria are tangible, measurable objectives to be accomplished within a proposed timeframe that indicate progress toward meeting the project goals. The criteria should include a **metric**, **target value**, and **timeframe**. Performance criteria may represent conditions at a reference site, and/or they may represent target conditions considering the surrounding land use or other local conditions. Performance criteria may also be known by other names (e.g., success criteria, performance standards, etc.).

For Subtidal Reefs:

Using a multi-beam sonar, interferometric side-scan sonar (or similar type of bathymetric side-scan sonar), a scientific echo sounder, or even a depth-finder, run a transect along the reef crest, taking height readings along the crest of the reef (Figure 13). If the reef does not have a discernible crest, then take readings along an approximate center line of the long axis of the reef. Use these measures to calculate mean reef height (± S.E.). If the restoration is made up of several smaller patch reefs, run transects in a grid pattern (see previous discussion of calculating proper lane width in Section 3.1.1) within the project footprint using one of the listed instruments to determine the heights along the patch reefs. Take the mean of the patch reef heights to determine overall mean reef height. Many of these acoustic instruments will take continuous GPS measurements in conjunction with depth readings. If the instrument does not have this capability, make continuous location readings using a GPS to 'mark' the locations of the reef height measurements. When sampling subtidal oyster reefs with these methods it is imperative that the tide height and the depth of water surrounding the reef are recorded at the time of sampling.

Alternative Methodology: If sonar, echo-sounders, or depth finders are unavailable, reef height may be determined by taking depth measurements using a sounding pole along the crest of the reef at regular intervals (2 m or greater, depending on project footprint). If the reef consists of smaller patch reefs, run transects in a grid pattern within the project footprint, taking depth measurements at regular intervals (2 m or greater, depending on project footprint) on previously identified patch reefs. Locations of all depth measurements should be marked using a GPS.

For Low Relief Reefs, Intertidal or Subtidal

In many natural or constructed oyster habitats, particularly on the west coast of the U.S., the relief of oyster reefs/beds is too low to be measured accurately using the methodologies listed above. If reef relief is low, reef height may be measured using a ruler, meter stick, or graduated rod that can measure on a cm scale. Record the height of the reef in relation to the surrounding substrate every 1 m along the crest of the reef (or every 5 m if reef length is >200 m). If the reef does not have a discernible crest, then take measurements along an approximate center line of the long axis of

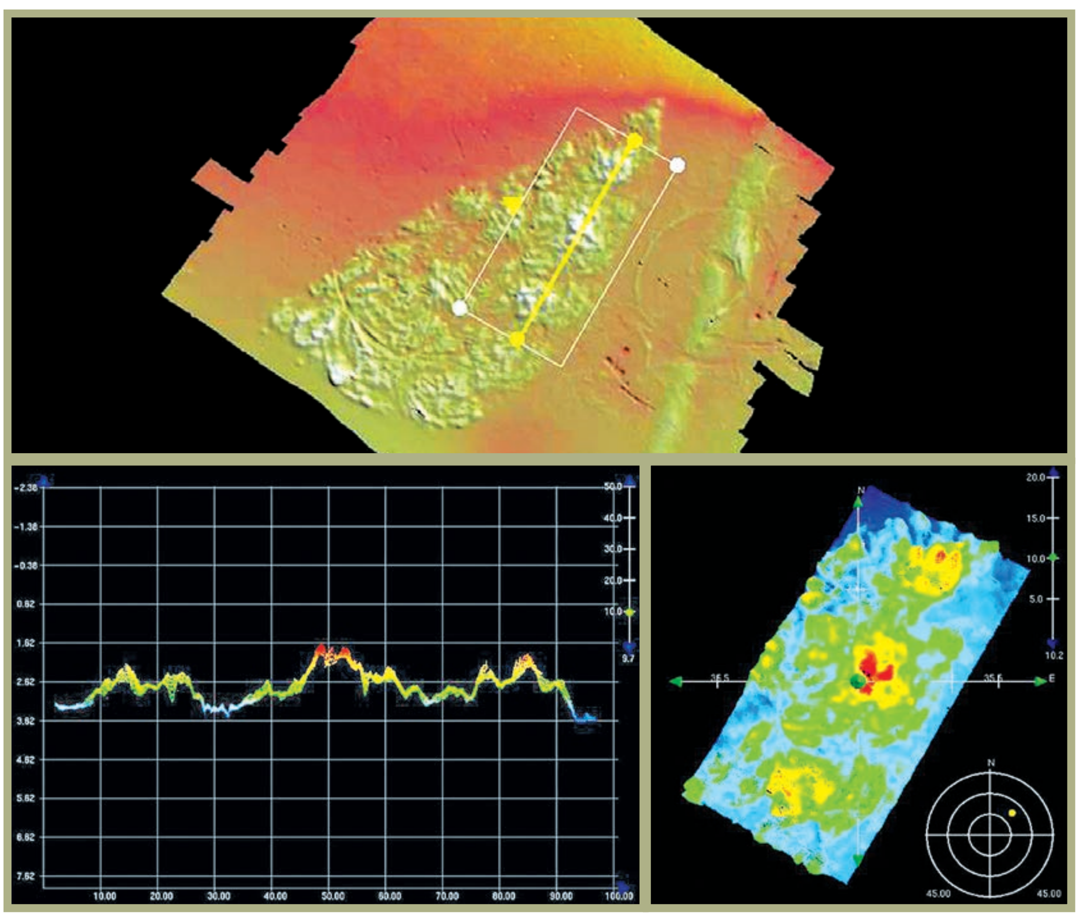

Figure 13. A multi-beam sonar mosaic (Top) with a 2D representation of cultch depth along a center transect (lower left) and a color contour plot of the site (lower right).

the reef. One method of obtaining measurements is by placing the ruler (or meter stick or graduated rod) vertically with careful contact at the sediment surface, and then using another ruler (or other thin, flat, rigid object) to lie horizontally on top of the shell and against the stick to make the observation. Take the average of these measurements to get mean reef height. If the reef is made up of several smaller patch reefs, take reef height measurements along each of the patch reefs, then calculate a mean overall reef height. For low relief reefs, height measurements should be reported in cm. (See http://www.directlinesoftware.com/survey.htm for descriptions of measures, surveying terms, abbreviations, water descriptions, etc.).

3.2.2 Sampling Frequency

Measurements should be taken once prior to construction, within three months post-construction (to document the as-built reef height), and one to two years post-construction. Ideally, measurements should also be taken four to six years post-construction, with additional measurements taken after events that could alter reef height or other metrics such as oyster survival (e.g., storms and hurricanes).

3.2.3 Performance Criteria

Performance Criteria = Positive or neutral change in reef height from original structure

Practitioners should set both a short-term and a mid-term goal of neutral or positive change from the original reef height (taken within three months post-construction). Reef construction material, form, and tidal location as well as oyster species characteristics should be considered when setting these goals. Practitioners should also check reef height data on natural and restored reefs in similar settings to aid in the determination of feasible goals. Restored reefs located in areas with historically low oyster densities, or restoration projects that utilize single oysters (rather than spat-on-shell) as seed oysters may not be able to attain a neutral or positive change in reef height; however, if a restoration project fails to meet the performance criteria, practitioners should not consider the project to be performing poorly if it is meeting the performance criteria of the oyster density metric. The reef height metric provides valuable information concerning the mid-term trajectory of the reef that can be used for adaptive management purposes.

3.3 Universal Metric #3: Oyster Density

Live oyster density is the number of live oysters, including recruits, per m^2. The mean density of live oysters provides information concerning oyster population size, survivorship, and recruitment of oysters on constructed or reference reefs.

Definition of settlement vs. recruitment. In this handbook the terms settlement and recruitment are clearly distinguished for the purposes of systematic oyster density measurements across projects. Settlement is simpler to define (although harder to measure perhaps) because it is a discrete event. Settlement occurs once the larva has become permanently attached to the substrate or has metamorphosed into its final benthic form (Wildish and Kristmanson 1997). Recruitment is more difficult to define because it includes settlement and some period of post-settlement survival, whose duration varies depending on the researcher's objective (Roegner 1991). Understanding the difference between settlement and recruitment is important because confusing the two could lead to very different measures being recorded.

Variation in settlement and recruitment can be attributed to influences in two broad categories: pre- and post-settlement factors (e.g., Olafsson et al. 1994; Wildish and Kristmanson 1997; Bartol and Mann 1997, 1999; Bartol et al. 1999; Kingsley-Smith and Luckenbach 2008). Pre-settlement processes, such as larval supply, larval competence and larval mortality directly influence settlement success. As has been assessed numerous times, settlement influences the horizontal distribution patterns of oysters (e.g., Bushek 1988). Post-settlement factors include predation, competition and physical stress, and influence recruitment success through post-settlement mortality.

It is strongly suggested that restoration projects monitor variation in settlement within a season, across more than one season, and at multiple locations, to account for temporal and spatial variability. Understanding the pattern and annual variability in settlement provides information on the availability of competent larvae but these measures are not considered a universal metric. For the purposes of this handbook, measurements of live oyster density should include recruits surviving to annual census (typically defined in this handbook as the end of the growing season) and typically at least 10 mm shell height for *C. virginica* and at least 3 mm for *O. lurida*.

Live oyster density is the number of live oysters, including recruits, per m^2

Measurements of live oyster density should include recruits surviving to annual census (typically the end of the growing season) and typically at least 10 mm for *C. virginica* and at least 3 mm for *O. lurida*.

Required Units = Mean density of live oysters, including recruits (individuals/m^2 $\pm$ S.E.)

3.3.1 Suggested Methodology

To estimate density, an adequate number of quadrat samples are taken from the reef area (or randomly selected patches if there are multiple patches of reef), and mean density is calculated. See section 2.6.4 for determination of an adequate sample size. Areal dimension is a Universal Metric, thus, reef area should also be delineated and area quantified for both intertidal and subtidal reefs. Oyster population size is estimated as the product of the mean density and the reef area. When underlying habitat strata explain a portion of the overall variance, stratified random sampling provides a more precise estimate of the total oyster abundance than simple random sampling for a given number of samples (see section 2.6.2, or OMW 2011).

Guidelines are provided in Section 2.6 to determine the desired sample size for the reef or number of patch reefs to be sampled. One quadrat excavated to a depth necessary to obtain all live oysters equals one sample. If using methodology other than quadrats, practitioners should report the efficiency of their sampling methods (the use of quadrat samples is assumed to be 100% efficient). There are several studies that address the accuracy of densities calculated from diver collected quadrat samples compared to those collected by tongs and dredges (e.g., Lenihan and Micheli 2000; Mann et al. 2004; Powell et al. 2007). We suggest the use of quadrats whenever possible or the use of grab samplers or tongs to sample a known area.

If possible, oysters collected from the reef for sampling purposes should be returned to the reef when measurements are completed in order to minimize impacts to the reef and to future sampling efforts (Figure 14). Conversely, practitioners may perform density counts without removing the oysters from the reef if the ability for accurate counts exists and the sample does not need to be used for size frequency measures.

Utilize the reef areal dimensions metric (Section 3.1) to determine the location and extent of the project footprint and reef area. Stratify samples if appropriate (e.g., reef height, etc. See section 2.6.2). Determine the adequate number of quadrat samples to be taken from the reef (or randomly selected patches if multiple patches exist) (see section 2.6.4). Collect quadrat samples by excavating to the depth necessary to obtain all live oysters within the quadrat. Utilize divers if necessary for subtidal reefs. Count all live oysters present within each sample, and record for calculation of mean density. Material from these quadrat samples is frequently used to obtain additional metrics such as size frequency (see section 3.4.1).

Figure 14. Restored reef at the Virginia Coast Reserve with a Quadrat (0.25m²) on an intertidal reef ready for removal of material to sample for resident organisms and oysters. Cultch and live oysters should be returned after measuring.

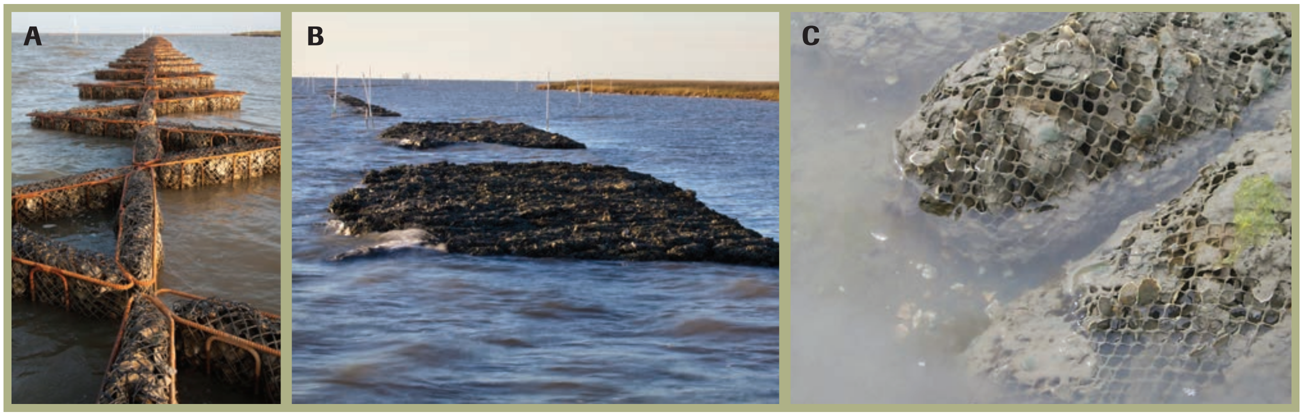

Figure 15. (a) Oyster reefs constructed of metal shell containment units in Mobile Bay, AL. (b) Bagged oyster shell near Dauphin Island, AL. (c) A close up of reef constructed of bagged oyster shell.

For Reefs Constructed of Rigid Structures or Bagged Shell:

In some instances, such as reefs constructed of metal structures or bagged shell (Figure 15), a standardized unit of sampling may be gained from removing a fixed unit of material (e.g., a bag of shell) from the reef for sampling. For reefs constructed of bagged shell or metal shell containment structures, stratify the reef area if appropriate (see section 2.6.2), and determine the adequate number of samples to be taken. Take random samples by removing a bag of shell; the area sampled per bag is determined by measuring the areal coverage of the measured bag, and should be converted to m^2. Count all live oysters present within each sample, and calculate mean density per unit area.

As oysters recruit to the substrate and grow it may not be possible to remove a bag of shell as in the early reef development stage, or when sampling from rigid structures (Figure 16). If this is the case, random samples can be taken using a quadrat rather than extracting a bag. For each quadrat sample, the quadrat should be placed on the surface of the reef structure and excavated to the depth necessary to collect all live oysters within the quadrat (for subtidal reefs use divers if necessary). Count all live oysters within each sample, and calculate mean density. Dense clusters of oysters will need to be separated to obtain accurate counts and measures. It may not be necessary to remove all oysters from within the quadrat if it is possible to count and measure all live oysters without removing them from the reef.

Figure 16. (a) Oyster growth on a reef constructed of metal shell containment units, TX. (b) An example of heavy oyster growth on a bag of shell cultch from a restored reef in FL after 12 months.

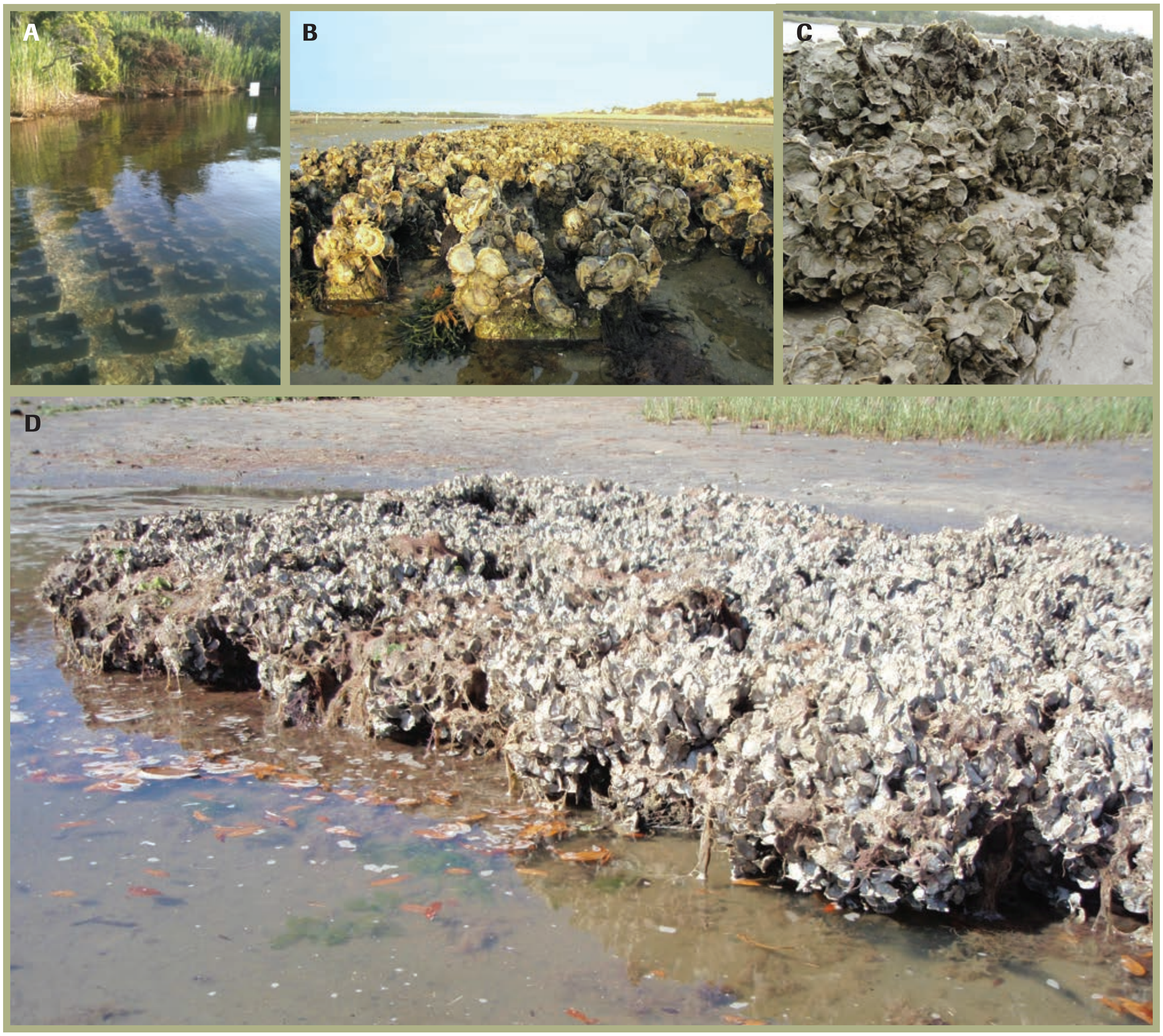

Figure 17. (a) Recently placed formed concrete blocks or 'oyster castles' in RI. (b) Oyster castles with oysters in Wellfleet Bay, MA. (c) Reef formed of oyster castles with growth in SC. (d) Reef formed of cement 'reef balls' with oyster growth in Tampa Bay, FL.

For Reefs Constructed of Cement Structures:

Often restored reefs are built by using adjacent or interlocking cement structures as substrate for reef development. To sample these structures for density, determine the adequate number of samples. Stratification of samples may include sampling quadrats near the base, at mid-height and near the crest of each structure (for subtidal reefs, use divers if necessary). Count all live oysters within each sample, and calculate mean density. It is not necessary to remove all oysters from within the quadrat if it is possible to count and measure all live oysters without removing them from the reef. Be sure to note the location (base, mid-height, or crest) at which each quadrat was sampled (Figure 17).

3.3.2 Sampling Frequency

At a minimum, sampling should be performed annually at the end of the oyster growing season (will vary by region, but is generally in the late summer or early fall). If possible, sampling should occur after newly settled oyster have grown to a size greater than (>) 10 mm for *C. virginica* and greater than (>) 3 mm for *O. lurida*. Efforts should be made to hold sampling dates as constant as possible across years and sites.

Figure 18. (a) Spat settled on an oyster shell. (b) Bagged shell for collecting natural spat settlement in lower Delaware Bay. (c) Spat set on bagged cultch.

3.3.3 Additional Methodology if Project Includes Seed Oysters

Live oysters are often added to restored reefs during construction to "seed" the reef with live individuals. In areas that are recruitment-limited these seed oysters are intended as brood stock to increase larvae availability and recruitment, and hasten reef development. Additionally, efforts have been made to seed reefs with specially bred disease-resistant oysters in the hope that the disease-resistant strains will enhance oyster populations through reduced mortality and by contributing disease-resistant progeny (Hare et al. 2006, Carlsson et al. 2008). Seed oysters may originate from hatcheries or from the wild, and may be in the form of spat-on-shell (Figure 18) or individual seed oysters. If hatchery produced seed oysters are used in a restoration project practitioners should take the source of the seed oysters into consideration in an effort to avoid genetic bottlenecking, and record the genetic/hatchery source as well as the nursery location. See Gaffney (2006) for a more thorough discussion of how genetic testing can be used in oyster restoration, as well as genetic issues to be considered in oyster restoration, and Brumbaugh et al. (2006) and Hedgecock (2011) for strategies to reduce potential genetic risks associated with stock enhancement. Regardless of the motives for using seed oysters, density of seed oysters should be monitored to the greatest extent possible.

Movement of oysters or cultch between water bodies poses the risk of introducing diseases or other unwanted organisms. Movement of restoration material is generally regulated by the state natural resource management agency, which should be consulted prior to moving any oysters or restoration material between water bodies (Mortensen et al. 2007, Hégaret, et al. 2008).

Required Units = Mean density of seed oysters (seed oysters/$m^2 \pm$ S.E.)

Suggested Methodology

Mean Density of Seed Oysters

Initial (post seeding) density of seed oysters can be determined one of two ways. Quadrat sampling can be performed (using the methodology described in section 3.3.1) within one week after deployment. Distinguish and count all seed oysters present in the density quadrats and calculate the mean number of seed oysters on a per m^2 basis. Alternatively, initial post seeding density can be estimated if the number of seed oysters deployed and the area over which they were deployed is known [i.e., mean seed oyster density = the number of seed oysters deployed ÷ the area (in m^2) over which they were deployed]. During later post-restoration monitoring, the only way to determine the number and density of surviving seed oysters with surety is through the use of genetic markers, a method that requires specialist knowledge and laboratory equipment [for more information on performing genetic analyses please refer to Milbury et al. (2004), Hare et al. (2006), or Carlsson et al. (2008)] or by marking seed oysters with an identifying paint. Because this is often not possible, the number and density of surviving seed oysters should be estimated from density sampling with robust length-frequency estimates (see Section 3.4), and annual cohorts distinguished by size class (Figure 19). Merging of size classes may make monitoring of an individual cohort impossible after one or two years, however, determining first and/or second year survival is very important in assessing performance of broodstock enhancement using seed oysters (Figure 19).

Natural settlement that occurs on the spat-on-shell after it has been deployed can artificially inflate survival estimates of seed oysters. This 'over-set' may be avoided by careful timing of seeding, however, estimating surviving seed oysters based on size-class will likely only provide information for one or two seasons. The mean number of surviving oysters should be reported on a per m^2 basis (±SE).

Sampling Frequency

The initial (post seeding) density of seed oysters should be determined immediately after deployment (i.e., within one week post-deployment). The density of surviving seed oysters can also be determined from the oyster density metric (this section) that is obtained annually at the end of the oyster growing season.

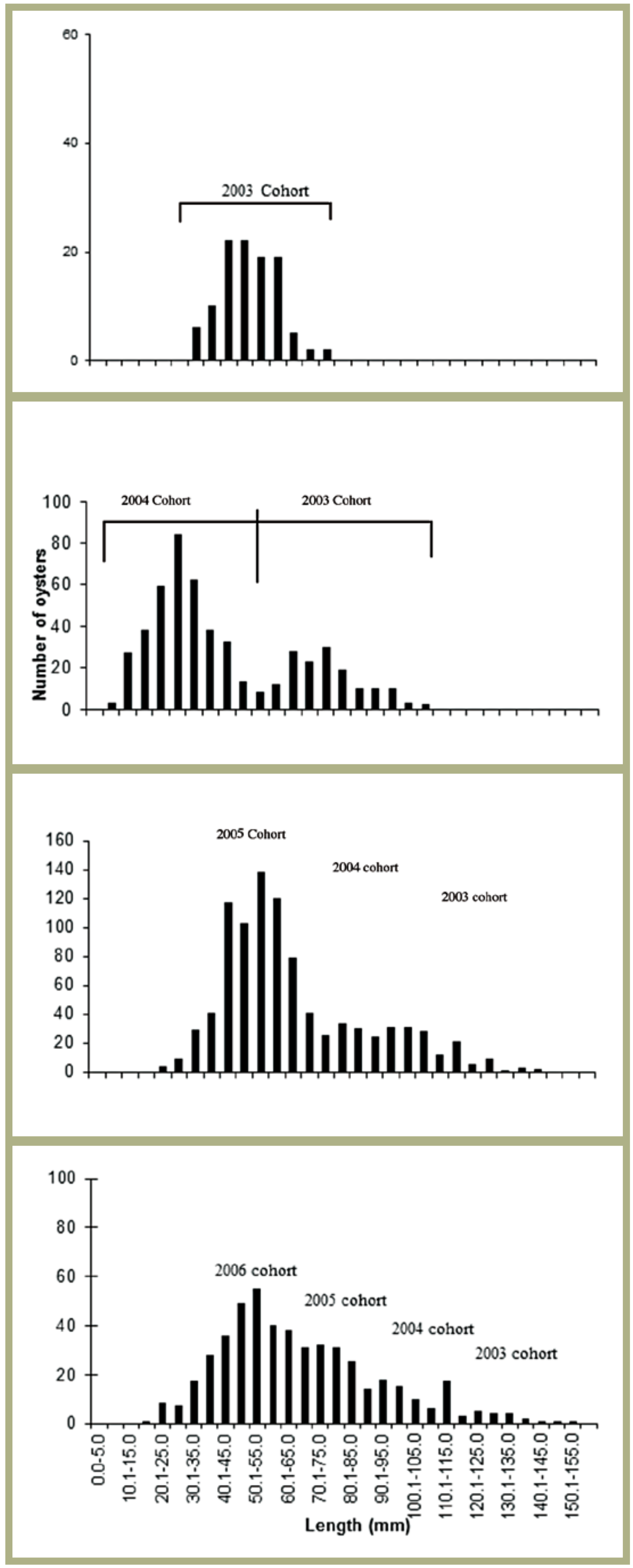

Figure 19. An example of size distributions of oysters sampled at a restoration site from 2004 to 2007 (sequential from top to bottom). Individual components of the 2003 cohort could easily be discriminated in 2004 and 2005, but not in subsequent samples. Mean size and survivorship of 1yr old animals can be determined in all samples except 2007.

3.3.4 Performance Criteria

Density Performance Criteria = Number of oysters per m²

When setting short- and mid-term oyster density goals, practitioners should refer to available density data for natural and/or restored reefs in similar settings as well as historical data. *Historical densities may be different than those we could expect to achieve today and target densities will vary by species, project type, and location*. It is therefore necessary to consider the full range of data available. There are numerous data sources available regionally through state fisheries management agencies, and nationally from zu Ermgassen at el. (2012a) (see Appendix II), which provides a compilation of recent and historical oyster densities. Practitioners should also consult with local and/or regional scientists and restoration practitioners to determine a target oyster density that is best suited for the restoration project and location. Practitioners need to consider the degree of oyster seeding (if any) performed during reef construction, and predicted mortality on those seed oysters when setting performance criteria.

Recruitment Performance Criteria = Number of recruits per m²

When setting short and mid-term recruitment goals, practitioners should refer to data for natural and restored reefs in similar settings, as well as any settlement and recruitment monitoring performed previously as part of the project. Recruitment is a vital component of a sustainable oyster reef. For an oyster reef to persist, the rate of shell accretion must exceed the rate of shell loss, also referred to as the shell budget (Powell et al. 2006, Mann and Powell 2007, Waldbusser 2013), and those balances should be considered when developing performance criteria.

Due to considerable local annual variability in recruitment rates, monitoring recruitment beyond the short-term time frame (1 – 2 years) is highly encouraged, if possible. To account for natural annual fluctuations in recruitment, monitoring extended to mid-term time frames (4 – 6 years) at minimum, is preferred.

3.4 Universal Metric #4: Oyster Size-Frequency Distribution

Oyster size-frequency distribution is a measure of how the oyster population is distributed across various size classes and provides information about oyster growth and the survivorship and mortality of cohorts.

Required Units = Mean shell height (SH) of live oysters (in mm); Mean percentage of measured oysters per size class (%) and/or number of oysters per size class.

3.4.1 Suggested Methodology

Oyster size-frequency distributions can be obtained from samples collected for the oyster density metric (Section 3.3) and a subset can be used for other measures such as Condition Index (Section 5.3.1). Using digital (or dial) calipers or a ruler, measure the shell height (the distance from the umbo to the distal margin of the shell) (Figure 20) to the nearest mm of at least 50 oysters from each sample (or enough oysters to equal a total of 250 oysters measured per reef).

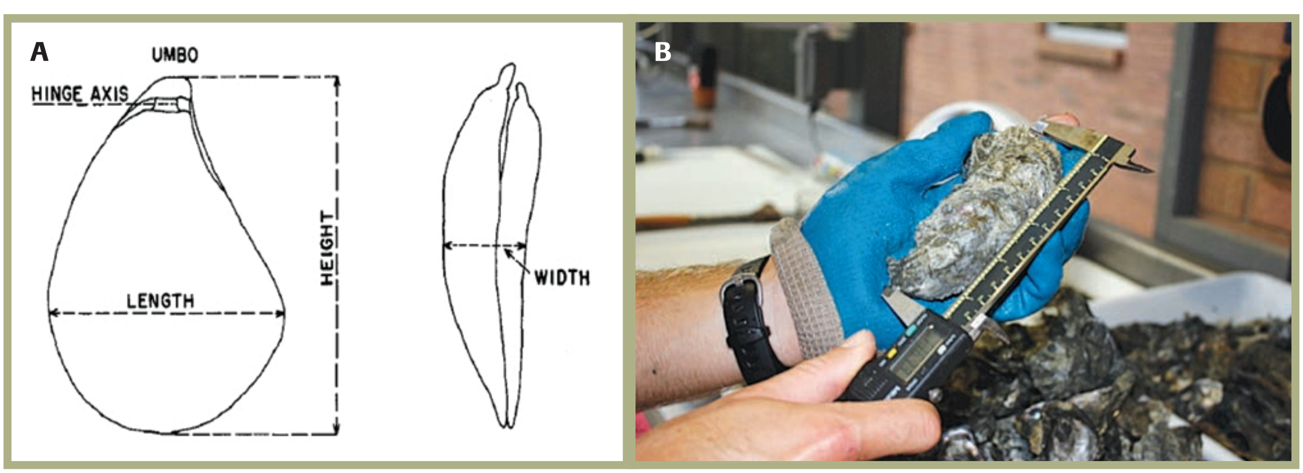

Figure 20. (a) Diagram of the height, length and width measurements of an oyster shell, from Galtstoff 1964, Chapter 2. (b) An example of shell height measurement in a lab.

Oysters to be measured per quadrat should be selected in an unbiased manner that correctly characterizes the size distribution of the population from the samples. One method to sub sample oysters for measurement is to break any clumps of oysters apart and spread out all oysters collected in the sample, dividing them in half, and then dividing the selected half again if necessary (Figure 21).

For *C. virginica*, measured oysters should be assigned to 5 mm size bins (e.g. ≤5mm, 6 to 10mm, 11 to 15mm, etc.). For *O. lurida*, measured oysters should be assigned to 3 mm size bins, (e.g. ≤3 mm, 4-6 mm, 7-9 mm etc.). For each reef, either calculate the percentage of measured individuals per size class (e.g., 50 individuals in a Size Class ÷ 250 measured individuals x 100 = 20%), or report the absolute number of individuals present in each size class (Figure 22).

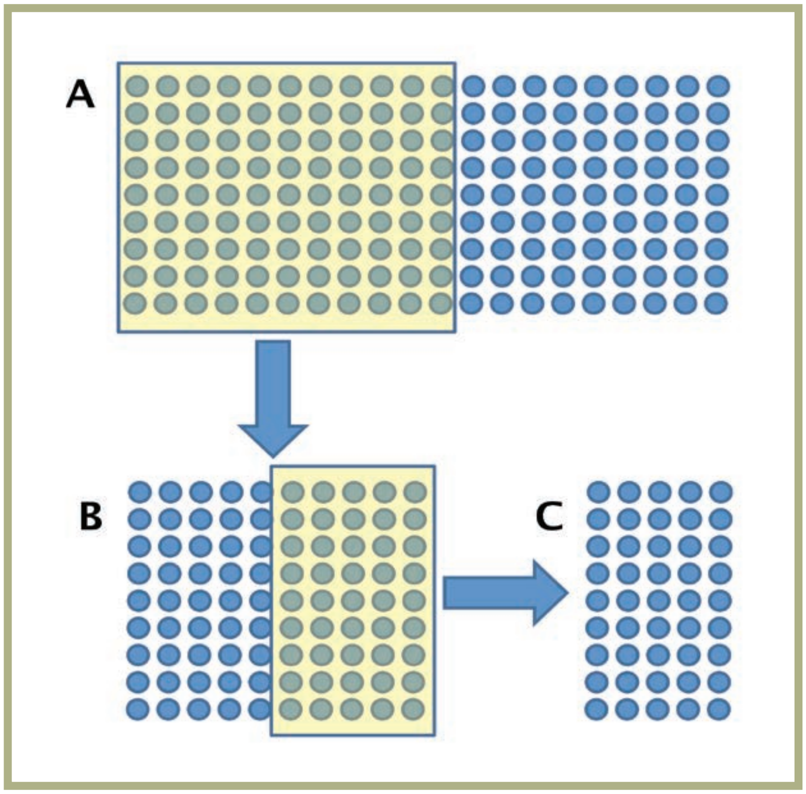

Figure 21. Dividing a quadrat sample for unbiased selection of representative oysters where individuals can be separated. All oysters in the quadrat sample should be spread out and divided in half (A). The selected half (yellow) should then be divided in half again (B), resulting in a reduced but unbiased sample of oysters to be measured.

3.4.2 Sampling Frequency

At a minimum, sampling should be performed annually at the end of the oyster growing season (will vary by latitude and region, but is generally in the late summer to early fall) in conjunction with sampling for oyster density (Section 3.3). If possible, sampling should occur after newly settled oysters have grown to a size greater than 10 mm for *C. virginica* and greater than 3 mm for *O. lurida*, and can be confidently classified as recruits (see section 3.3).

3.4.3 Performance Criteria

Performance Criteria = none

There are no performance criteria associated with the oyster size-frequency distribution metric. This metric can provide valuable information about the size (age) structure of the oyster population on the reef, growth (see Kraeuter et al. 2007), and can be used to evaluate the successful recruitment performance goal (Section 3.3.4).

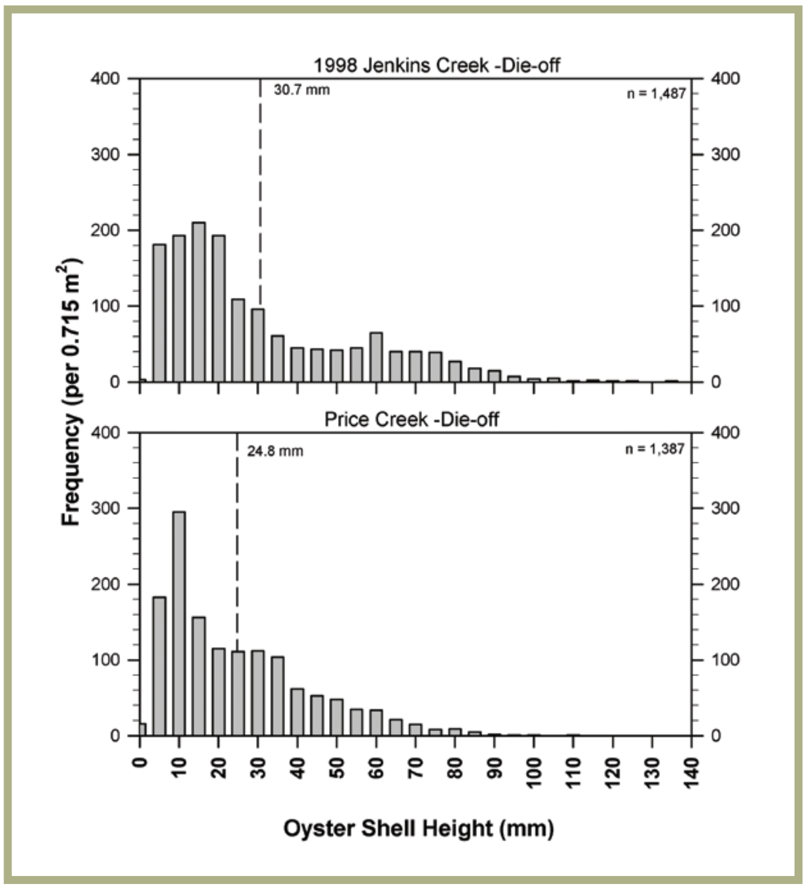

Figure 22. Example of plotted size-frequency data for oysters recruited into shell trays.

Chapter 4: Universal Environmental Variables

Universal Environmental Variables are parameters which should be monitored for *every* oyster restoration project, regardless of the restoration goal of that project (Appendix I). In that manner, they are similar to the Universal Metrics. There are no performance criteria for these Environmental Variables as they are meant to provide data to help explain potential impacts to the performance of restored reefs.

Monitoring stations (NOAA, USGS, EPA, or other government or academic group recording similar data) may be located near the restoration site and serve to provide the necessary data. Water temperature, salinity, and dissolved oxygen readings from those stations can be used. It is up to the practitioner to determine whether a monitoring station is a suitable surrogate for direct measurements of the Environmental Variables at the restoration site. If using data from a monitoring station, please note the distance and any possible influences on water quality or geomorphologic characteristics that may be present between the restoration site and the monitoring station. If a monitoring station is not available and the Environmental Variables must be sampled by restoration practitioners, it is important that all instrumentation used is properly calibrated. It is preferred that data measurements are continuous (either from a monitoring station or an *in situ* continuous monitor deployed by a practitioner). If continuous measurements not possible, measurements may be made less frequently, with the understanding that as the frequency of measurements decreases, the usefulness of the data also decreases. To assess the limitations of the data, practitioners should report the frequency and timing (i.e., season, tidal stage, time of day) of sampling.

4.1 Water Temperature

While the temperature tolerances of *C. virginica* and *O. lurida* may vary somewhat along their distribution ranges, temperature profoundly influences oyster reproduction, development and growth, and survival (Shumway 1996; Burrell 1986; Baker 1995; COSEWIC 2011). Additionally, higher water temperatures tend to increase the exposure of oysters, particularly *C. virginica,* to disease and predation (Shumway 1996).

Required Units = °C   Accuracy ± 1°C

4.1.1 Suggested Methodology

If no suitable monitoring station is nearby, water temperature measurements should be taken near the substrate as close to the reef as possible. Measurements should be reported in degrees Celsius and may be taken using a permanently deployed *in situ* instrument with a datalogger, a thermometer, or with other instrumentation (Figure 23).

Figure 23. Using handheld instrumentation to measure water temperature and salinity.

4.1.2 Sampling Frequency

Continuous water temperature measurements taken throughout the year at intervals of 15 to 60 minutes are preferred. If continuous data loggers are not available, water temperature measurements should be made every time other sampling is performed at the reef and in between sampling events as often as possible, and special efforts should be made to take measurements after storm events. It should be noted that continuous data loggers that record temperature and salinity at regular intervals are available at low cost.

4.2 Salinity

Although *C. virginica* occurs in a range of salinity from 5 to 40 practical salinity units (psu), their optimum salinity range is approximately 14 to 34 psu in habitats that typically include some fluctuation in salinity. *O. lurida* is not as tolerant of low salinities; for example, populations in Puget Sound WA have a lower salinity limit of 23-24 psu (Baker 1995). Like temperature, salinity influences *C. virginica's* growth and mortality, and, to a lesser degree, reproduction (Shumway 1996). Also, more so than temperature, higher salinities can be associated with greater instances of disease and predation in *C. virginica* (Ewart and Ford 1993; Shumway 1996).

Required Units = ppt (parts per thousand) or psu   Accuracy ± 1 ppt or 1 psu

(Note: Salinity measurements from an instrument, such as a CTD, that utilizes a conductivity ratio are unitless)

4.2.1 Suggested Methodology

If no suitable monitoring station is nearby, salinity measurements should be taken near the substrate as close to the reef as possible and should be reported in ppt or psu, with a 1 ppt or 1 psu resolution. Measurements may be taken using a permanently deployed *in situ* instrument with a datalogger, a refractometer, or with other instrumentation. Frequent calibration of instruments is important.

4.2.2 Sampling Frequency

Continuous salinity measurements at intervals of 15 to 60 minutes throughout the growing season are preferred. If continuous data loggers are not available, salinity measurements should be made every time other sampling is performed at the reef and in between sampling events as often as possible, and special efforts should be made to take measurements after storm events in order to document the extent of salinity fluctuations and the duration of lowered salinity. It should be noted that continuous data loggers that record temperature and salinity at regular intervals are available at low cost.

4.3 Dissolved Oxygen (subtidal reefs only)

Hypoxia ($O_2 < 2$ mg L^{-1}) and anoxia ($O_2 < 0.5$ mg L^{-1}) have been shown to have detrimental effects on the settlement, growth, and survival of oysters (e.g., Baker and Mann 1992; Johnson et al. 2009). Data on dissolved oxygen (DO) levels near oyster reefs could help explain low settlement or high mortality events. It is assumed that low DO is less likely to be a problem for intertidal oyster reefs.

Required Units = MG L^{-1}Accuracy ± 0.1 mg L^{-1}

4.3.1 Suggested Methodology

If no suitable monitoring station is nearby, dissolved oxygen measurements should be taken near the substrate as close to the reef as possible and should be reported in mg/L. Time of day and tidal stage during which the measurements were taken should be noted. Measurements may be taken using a permanently deployed *in situ* instrument with a datalogger, or with instrumentation such as a DO meter.

4.3.2 Sampling Frequency

Continuous dissolved oxygen measurements at intervals of 15 to 60 minutes are preferred, particularly during the period of peak water temperatures. Since DO levels often vary greatly between day and night (with levels being lower at night), practitioners should take both daytime and nighttime DO measurements if continuous measurements are not possible.

Chapter 5: Ancillary Monitoring Considerations

Several pieces of ancillary data can help identify problems with a site or a restoration effort. They are not essential to evaluating basic project performance, but often provide valuable information that can aid in the interpretation of data or help to improve subsequent efforts. The following section provides basic information on common ancillary monitoring that a practitioner may want to consider implementing as part of their monitoring plan. This section is not meant to be an exhaustive how-to manual in performing these monitoring techniques; but rather an introduction to the monitoring considerations and the literature that a practitioner can use to learn more about the specific monitoring technique.

5.1 Presence of Predatory, Pest, and/or Competitive Species

Species of vertebrates and invertebrates that prey upon, or compete for space with, oysters can negatively impact populations of *C. virginica* (see White and Wilson 1996 and references therein) and *O. lurida* (e.g., Baker 1995, COSEWIC 2011). Predators often include predatory gastropods, crabs, flatworms, sea stars, and fish, whereas sessile species such as bryozoans, barnacles, and mussels often compete with oysters for space (Baker 1995; White and Wilson 1996). Oyster pests include sponges, mud worms, pea crabs (White and Wilson 1996), and anomuran shrimps (Baker 1995). Sampling to quantify these organisms, using the appropriate methodology described in Chapter 7: Habitat Enhancement for Resident and Transient Species, can be performed to help understand the potential impacts from these species.

5.2 Disease Prevalence and Intensity

Oyster disease is cited as one of the major causes of oyster population decline, particularly along the Atlantic and Gulf coasts of the United States. Two diseases, Dermo and MSX, caused by the protozoans *Perkinsus marinus* and *Haplosporidium nelsoni,* respectively, can cause high levels of mortality among infected oyster populations. *Perkinsus marinus* is prevalent throughout the Gulf of Mexico and along the Atlantic coast up to Massachusetts. *Haplosporidium nelsoni* occurs primarily along the mid-Atlantic, but is also present from Florida to Maine (Ford and Tripp 1996).

Mortality due to oyster disease has also been documented on the West coast of the United States and Canada, and a variety of disease agents have been detected and/or associated with mortality events including *Hexamita nelsoni*, *Mikrocytos mackini*, *Haplosporidium* spp., and *Bonamia* spp., as well as a *Mikrocytos*-like protist and a hemic neoplasm (see Friedman et al. 2005 and COSEWIC 2011 and references therein).

Monitoring for the presence of oyster disease may not be necessary if disease prevalence and/or intensity are not thought to be high in or near the restoration area. If the restoration site is in a state that has a disease monitoring program and has monitoring sites near the restoration site, the restoration practitioner should consult with the staff of their state's disease monitoring program as to whether or not disease monitoring should be conducted. If the disease is suspected or known to be present at or near the restoration site(s), and state disease monitoring data are not available, then monitoring the presence and intensity of disease should be considered using the following methodology.

5.2.1 Suggested Methodologies

For Perkinsus marinus (Dermo)

Units = Disease Prevalence (%), Weighted prevalence (unitless)

Methodology

Randomly collect a minimum of 25 adult oysters from across the reef for analysis of Dermo prevalence and intensity (see Marques and Cabral (2007) for information regarding sample size determination for disease sampling). Oysters should be transported to a local testing lab (check with local universities or extension offices) as per the lab's instructions. Alternatively, if practitioners have the ability, they may determine disease prevalence using Ray's fluid thioglycolate method (Ray 1952; Bushek et al. 1994; Bobo et al. 1997).

Dermo infection intensity should be ranked according to Mackin's scale (Ray et al. 1953):

5 = Heavy infection

4 = Moderate to Heavy Infection

3 = Moderate Infection

2 = Light to Moderate Infection

1 = Light Infection

0.5 = Very Light Infection

Calculate disease prevalence as the number of diseased oysters per sample divided by the total number of oysters in the sample. The weighted prevalence is the mean infection intensity of the oysters in the sample.

For Haplosporidium nelsoni (MSX):

Units = Disease Prevalence (%), Weighted Prevalence (unitless)

Methodology

Randomly collect a minimum of 25 adult oysters from across the reef for analysis of MSX prevalence (see Marques and Cabral (2007) for information regarding sample size determination for disease sampling). If also testing for the presence of Dermo, use the same oysters collected for Dermo analysis. MSX disease prevalence may be determined either by the practitioner or by a research lab following the paraffin histology method described in Burreson et al. (1988) or by using PCR amplification (see Stokes et al. 1995) or other comparable techniques. Infection intensity should be assigned based on the number of pathogen cells present using the following scale:

Rare = less than 10 MSX plasmodia

Very light = 10-100 plasmodia

Moderate = 1-5 plasmodia per field of 100X view

Heavy = more than 5 plasmodia per field of 100X view

Calculate disease prevalence as the number of diseased oysters per sample divided by the total number of oysters in the sample.

An infection rating describing the disease progression should also be assigned using the following scale (Kim et al. 2006):

0 = No parasites present in tissues

1 = Parasites present only in digestive tract or gill epithelial tissue (10 or less plasmodia per 100X field of view)

2 = Parasites present only in digestive tract or gill epithelial tissue (11 to 99 plasmodia per 100X field of view)

3 = Parasites present in epithelium and subepithelium tissue (over 100 plasmodia per 100X field of view in gill or body tissue but less than 1 plasmodia per oil immersion 1000X field of view)

4 = Parasites evenly distributed in digestive tract or gill subepithelium and scattered throughout systemic tissue (over 100 plasmodia per 100X field of view in either body or gill tissue but 1 to 10 plasmodia per oil immersion 1000X field of view)

5 = Moderate systemic infection of parasites (average of 11 to 20 plasmodia per oil immersion 1000X field of view)

6 = Heavy systematic infection of parasites (average of over 20 plasmodia per oil immersion 1000X field of view)

The weighted prevalence should be calculated by taking a mean infection rating of the entire sample, including those with no detectable parasites or pathogens.

For West Coast Pathogens:

Units = Pathogen Prevalence (%)

Methodology

Randomly collect a minimum of 25 adult oysters from across the reef for analysis of pathogen prevalence (see Marques and Cabral (2007) for information regarding sample size determination for disease sampling). Oysters should be transported to a local testing lab (check with local universities or extension offices) as per the lab's instructions. Alternatively, if restoration practitioners have the ability, they may determine pathogen prevalence using the paraffin histology method described in Burreson et al. (1988) (also see Friedman et al. 2005 and Meyer et al. 2010) and either polymerase chain reaction (PCR) assays (see Meyer et al. 2010) or fluorescent *in situ* hybridization (see Friedman et al. 2005).

Calculate pathogen prevalence as the number of oysters in which pathogens are present per sample divided by the total number of oysters in the sample.

5.2.2 Sampling Frequency

At a minimum, sampling should be conducted annually during peak disease/pathogen occurrence (often during the fall) or monthly if the peak time of occurrence is unknown.

5.3 Oyster Condition Index

A common way to assess the effects of environmental conditions, such as salinity, temperature, tidal elevation, food quality, pollution, and presence of parasites, on oysters is by comparing condition index (CI) values among oysters exposed to differing conditions or located in various locations (e.g. Lawrence and Scott 1982, Rainer and Mann 1992, Rheault and Rice 1996).

Units = Condition Index (unitless)

5.3.1 Suggested Methodology

Oyster CI values can be determined using the same oysters measured for the oyster size-frequency distribution metric (see Section 3.4). A minimum of 25 adult oysters per sample should be assessed for condition index. For each oyster, blot shell as dry as possible and obtain a whole wet weight of the oyster (WWW) (g). After weighing, remove the tissue from the shell, blot the internal and external surfaces of the shell as dry as possible and weigh to obtain shell wet weight (SWW) (g). The SWW subtracted from the WWW yields the internal shell cavity capacity (g), which is considered equal to cavity volume (cm^3) when the density of the cavity contents (i.e., oyster tissue) is assumed to be 1g cm^{-3} (see Abbe and Albright 2003 for discussion). Dry the tissue of the oyster to a constant weight at 60°C (a minimum of 48 hours) and reweigh to obtain the tissue dry weight (TDW) (g). Calculate the CI using the following equation (Lucas and Beninger 1985, Rheault and Rice 1996):

$$CI = (TDW \times 100)/(WWW - SWW)$$

5.3.2 Sampling Frequency

Sampling frequency will depend on the performance or research question being addressed. At a minimum, sampling should be performed annually at the end of the oyster growing season (will vary by region); however, more frequent sampling (e.g., quarterly) may be preferable as it would be useful in discerning differences in condition index due to seasonality or other environmental factors (e.g., salinity, temperature, food availability).

5.4 Gonad Development Status

Gonad development status is a determination of the reproductive status of oysters present on a reef and, along with oyster density, size-frequency distribution, and sex ratio is a good indicator of a reef's potential for egg production.

Units = Percentage of reproductive adults (%)

5.4.1 Suggested Methodology

Randomly collect a minimum of 25 adult oysters (≥ 25 mm for *C. virginica* and ≥ 12 mm for *O. lurida*) with the size of the oysters collected being equally distributed across the available size range of oysters. Following the methodology of Southworth and Mann (1998), remove sections of the gonad and visceral mass from each oyster, fix in Bouin's solution, then dehydrate in alcohol. After dehydration, clear the alcohol from the tissues using xylene and then embed in paraffin wax. Slice the tissues into histological sections that are 7 to 10 μm thick and then stain in Delafield's solution, followed by counterstaining in eosin Y (see Humason 1962 for detailed staining and counterstaining methods) (Southworth and Mann 1998). After the staining process is complete, the gonad developmental stage for each oyster should be identified as either inactive or reproductive (active). See below for a description of gonadal development stages from Southworth and Mann (1998) (or also see Kennedy and Battle 1964):

Inactive

At this stage there are no follicles present peripheral to digestive gland and the sex of organism is undeterminable.

Active

Early Stages: In females, the eggs are not well developed. Oocytes are attached to the follicle wall and may contain a few nuclei, but no nucleoli. In males, there may be many follicles containing spermatogonia and spermatocytes, but no spermatozoa are present.

Late Stages: In females there are some free oocytes present, with the majority having distinct nuclei, but fewer than 50% having distinct nucleoli. In males, spermatids are predominately found in the follicles, but the follicles are not completely full. Within follicles, the characteristic swirling pattern of spermatozoa (tails oriented towards the center of the follicle) can begin to be seen.

Ripe: In females, free oocytes of similar sizes are present in the follicles, over 50% of which have a distinct nuclei and nucleoli. In males, characteristic swirling pattern of spermatozoa with their tails towards the center of the follicles is very evident.

Spent: In females, the eggs are of varying sizes and appear granular or as though they are breaking down and follicles are either partially or completely empty. In males, some phagocytes may be present and follicles are either partially or completely empty.

Calculate the percentage of reproductive adults by dividing the number of reproductive adults by the total number of oysters in the sample and then multiplying by 100.

5.4.2 Sampling Frequency

At a minimum, sampling should be conducted annually just prior to the peak reproductive season (will vary by region and species), and may be repeated again later in the reproductive season if needed. If discerning differences in gonad development due to seasonality or other environmental factors (e.g. salinity, temperature) is an important component of the project, more frequent sampling (e.g., monthly or quarterly) may be required.

5.5 Sex Ratio

The sex ratio is the ratio of male to female oysters present on the reef and, along with oyster density, size-frequency distribution, and gonad development status is a good indicator of a reef's potential for egg production. Also, since oysters are protandrous hermaphrodites, sex ratios can provide valuable information concerning generation times and the susceptibility of the population to collapse (Mann and Powell 2007).

Units = Unitless (ratio of males to females)

Figure 24. Collecting a gonad sample using a fine capillary tube.

5.5.1 Suggested Methodology

The same oysters collected for gonadal sampling (Section 5.4) may also be used for sex ratio sampling. If gonadal sampling is performed, oyster sex may be determined from those prepared gonad samples. If gonadal sampling and disease/pathogen testing are not performed, randomly collect at least 25 adult oysters (≥ 25 mm for *C. virginica* and ≥ 12 mm for *O. lurida*) with the size of the oysters collected being equally distributed across the available size range of oysters. Using a scalpel, remove a small section of gonad from each oyster and smear across a glass microscope slide, being sure to clean scalpel after each oyster. Alternatively, a Pasteur pipette or a fine capillary tube (Figure 24) can be gently inserted into the gonad to collect a sample that can be expelled onto a glass microscope slide (use a clean tube or pipette for each sample). Examine each slide under a microscope and determine sex of the oyster via presence of oogonia/ova or spermatocytes/spermatozoa. Calculate the sex ratio by dividing the number of males by the number of females.

5.5.2 Sampling Frequency

Sampling should be conducted just prior to the peak reproductive season (will vary by location and oyster species), and may be repeated again later in the reproductive season if needed.

5.6 Shell Volume for Determination of Shell Budget

For an oyster reef to persist, the rate of shell accretion must exceed the rate of shell loss (e.g., Mann and Powell (2007)). Shell accretion occurs through recruitment, growth and natural mortality (e.g., Mann and Powell 2007) whereas shell loss can be caused by taphonomic sources such as bioerosion, dissolution, and disarticulation (e.g., Powell et al. 2006) as well as habitat destruction and burial of shell (e.g., Mann and Powell 2007). Other factors such as disease and harvest also affect rates of shell accretion and loss (Powell and Klinck 2007, Powell et al. 2012). As restoration science progresses, more emphasis is being placed on understanding the processes involved in shell loss and accretion and their effects on restored or harvested oyster populations. The information obtained through the measurements of shell volume (see methodology below), combined with information on recruitment, natural mortality, and shell loss (due to harvest and/or taphonomic sources) can be used to calculate a shell budget (the balance between shell loss and accretion) (e.g., Powell et al. 2006) for a restored reef or other reef of interest. Further information on the intricacies of calculating shell budgets and their components can be found in Ford et al. (2006), Powell et al. (2006), Mann and Powell (2007), Powell and Klinck (2007), and Powell et al. (2012).

Units: L/m^2

5.6.1 Suggested Methodology

Using the same quadrats used for measures of oyster density and size frequency distribution (Sections 3.3 and 3.4), collect all shell present above the substrate (i.e., not buried beneath the sediment) within the quadrat. Separate the sample into the following components: live oysters, boxes [dead oysters with articulated valves (valves that are hinged together)], and cultch (e.g., oyster shell fragments, disarticulated valves, or clumps of shell that are not attached to live oysters) (Powell et al. 2006). Determine the shell volume of each of these components (be sure to remove the meat from the live oysters before determining their shell volume).

While there are several methods of determining the volume of shell, perhaps the easiest is the water displacement method. This method involves the use of either: (1) a bucket filled to brim with water and placed within an empty container (to catch overflow) or, (2) a bucket with a hole drilled ¾ of the way to the top and fitted with a tube to direct overflow from the hole to a container. In the tube-fitted bucket, the water level should be just at the bottom of the hole. For each quadrat sample, measure each component (live oysters, boxes, cultch) separately by placing the component in a mesh bag and slowly lowering the mesh bag into the bucket, being careful not to insert hands or any other object into the bucket. Collect the displaced water and pour into a graduated cylinder to determine the volume of water displaced. The volume of water displaced is equal to the shell volume and should be reported on a L/m^2 basis. To calculate a carbonate budget for a reef, samples of the subsurface shell component of the reef must also be collected (see Powell et al. 2012 for more details), and the subsurface shell volume determined.

5.6.2 Sampling Frequency

Measurements should be taken one to two years post-construction, at minimum. Preferably, measurements should also be taken four to six years post-construction, with additional measurements being taken after events that could alter reef height or oyster survival (e.g. hurricanes).

5.7 Percent cover of reef substrate

Measurement of the percent cover of reef substrate (both living and non-living) provides a quick visual estimate of the habitat available for oyster settlement. This measurement also provides information concerning smaller-scale patchiness of reef substrate within the larger project footprint/reef area.

Units = % (percent cover)

5.7.1 Suggested Methodology

Record a visual estimation of the percentage coverage of reef substrate (including living oysters and non-living hard substrate) within the same quadrats used for measures of oyster density and size frequency distribution (Sections 3.3 and 3.4). Percent coverage estimate must be made before oysters are excavated for the oyster density and size-frequency distribution samples. To aid in determination of percent coverage, a quadrat with a delineated (usually with string) grid pattern can be used. Count the number of squares in the grid in which shell is present, and from that determine the percentage of the substrate within the grid covered by shell (e.g. if the grid contains 50 squares of which 32 have shell present, then the percent coverage is 32/50= 64%; if the grid contains 100 squares of which 59 have shell present, then the percent coverage is 59%).

5.7.2 Sampling Frequency

Measurements should be taken once prior to construction, within three months post-construction (to document the as-built percent cover of reef substrate), and one to two years post-construction. Preferably, measurements should also be taken four to six years post-construction, with additional measurements being taken after events that could alter the percent coverage of reef substrate or oyster survival (e.g. hurricanes).

Chapter 6: Restoration Goal-Based Metrics: Brood Stock and Oyster Population Enhancement

Oyster restoration projects often involve the deployment of oyster stock for either brood stock or population enhancement. In projects with a goal of brood stock enhancement, oyster stock is deployed in areas where natural recruitment is low. The intended outcome being that the additional brood stock will generate increased recruitment that will enable the reef to perpetuate or will benefit nearby natural oyster habitat with increased recruitment. Projects may also have a goal of population enhancement for the benefit of increased harvest of oysters on nearby reefs. Similar to the goals of brood stock enhancement, fisheries enhancement can occur through indirect beneficial effects that restored reefs may have on nearby harvested reefs.

Additionally, efforts have been made to deploy specially bred disease-resistant stock in the hope that the disease-resistant strains will enhance oyster populations by contributing disease-resistant progeny. Oysters used in stock enhancement may originate from hatcheries or have been harvested from the wild, and may be in the form of spat-on-shell [multiple juvenile oysters (spat) attached to a shell substrate] or individual seed oysters. As mentioned in Section 3.3.3, there are several genetic issues (e.g. genetic bottleneck) to be considered with stock enhancement, and practitioners should be careful to select strains of stock that will maintain or enhance genetic diversity on the reef and at nearby areas of interest. See Gaffney (2006) for a more thorough discussion of genetic issues to be considered in oyster restoration, and Brumbaugh et al. (2006) for a summary of strategies to reduce potential genetic risks associated with stock enhancement.

While connectivity between the restored and nearby reefs is difficult to demonstrate (Kim et al. 2013), there are a few metrics that can be used to explore possible connectivity. If the project's brood stock and population enhancement restoration goals are focused only on the restored reef site and not also with nearby-reef connectivity, sampling only the Universal Metrics will be sufficient (See Sections 3.3 and 3.4, as well as methodology for projects utilizing seed oysters in Section 3.3.3). If the goal is brood stock and population enhancement for the purpose of improving nearby unrestored locations, then the metrics listed below should be sampled in addition to the sampling of Universal Metrics conducted on the restored reef. Therefore, the metrics below address only nearby-reef sampling, which should occur at areas of potential impact.

6.1 Metric #1: Nearby-Reef Oyster Density and Associated Size-Frequency Distributions

Restored oyster reefs with increased brood stock and increased populations may have beneficial effects on nearby reefs through the provision of larvae, with increased recruitment and increased oyster densities as a result. While this sort of connectivity is harder to document, post-restoration measurements of nearby-reef oyster density and size distribution, when compared to nearby-reef pre-construction data, can provide some insight to the impacts a restored reef may be having on nearby natural reefs. While this information can provide useful inferences on the contribution to recruitment the restored reef is providing to nearby habitat, practitioners should use caution in drawing direct connections between these types of source-sink relationships without further, more intensive analysis (e.g., genetic relationships, larval distribution models, etc.).

Required Units = Mean total density (individuals/m^2) (live oyster density including recruits, as described in section 3.3), Mean shell height of oysters (mm), Percentage (%) and/or number of measured oysters per size class

6.1.1 Suggested Methodology

Perform measurements of oyster density and size distribution at the area(s) of interest using the methodologies outlined in Sections 3.3 and 3.4. Recruitment is quantified using the oyster density and size distribution samples (see Section 3.3 for discussion of distinguishing settlement from recruitment). If quantifying settlement (e.g., temporal and spatial distribution), settlement tiles or shell trays of known area and depth, placed at areas of interest (e.g., at nearby natural reefs or at specific distances from the restored reef) can be utilized. Quantification of temporal and spatial spat settlement is not considered part of this metric, but may be valuable in understanding settlement dynamics of areas of interest, and thus is encouraged.

6.1.2 Sampling Frequency

At a minimum, sampling should be performed annually at the end of the oyster growing season (will vary by region, but is generally in the late summer or early fall). Sampling should occur after newly settled oyster spat have grown to a size greater than (>) 10 mm for *C. virginica* and greater than (>) 3 mm for *O. lurida*, and can be confidently classified as recruits (see section 3.3).

6.1.3 Performance Criteria

As monitoring progresses, there should be a trend of increasing oyster density on the nearby-reef site of interest, with an ultimate goal of having statistically greater oyster densities than those present pre-construction, and a density that is roughly equal to or exceeds that of a natural reference site.

6.2 Metric #2: Nearby-Reef Large Oyster Abundance

The number of resultant large (e.g., market-sized for *C. virginica*) oysters is of interest in many restoration projects designed to enhance oyster populations. Oyster size varies by species, but for the purposes of this handbook a large oyster is one that is equal to or greater than 3" (76 mm) in shell height for *C. virginica* (based on a common designation of market-size) and equal to or greater than 35 mm for *O. lurida*. It should be noted that if an area of potential impact is a harvested reef, then the abundance of large oysters may be low due to harvesting. Practitioners need to consider the impact of harvesting on the reliability of their data when designing their monitoring plan and areas of interest, and should consider performing pre- and post-harvest season sampling to assess the impact of harvesting.

Units = Mean Density (individuals/m^2)

6.2.1 Suggested Methodology

Obtain samples following the methodologies provided for oyster density (Section 3.3) and size-frequency distribution (Section 3.4), and count the number of large oysters present in the samples and report on a per m^2 basis.

6.2.2 Sampling Frequency

At a minimum, sampling should be performed annually at the end of the oyster growing season (will vary by region, but is generally in the late summer or early fall). Note that it may take several years for oysters to grow to large sizes.

6.2.3 Performance Criteria

As monitoring progresses, there should be a trend of increasing densities of large oysters at the nearby-reef site of interest, with an ultimate goal of having statistically more large oysters than at the control site or pre-construction conditions, or a density that is roughly equal to or exceeds that of a natural reference site.

Chapter 7: Restoration Goal-Based Metrics: Habitat Enhancement for Resident and Transient Species

Numerous coastal species, many of which are commercially or recreationally important, such as blue crab (*Callinectes sapidus*), red drum (*Sciaenops ocellatus*), gag grouper (*Mycteroperca microlepus*), black sea bass (*Centropristis striata*), and striped bass (*Morone saxatilis*) among others, utilize intertidal and subtidal oyster habitats for shelter and feeding or reproduction grounds (Coen et al. 1999b; Breitburg 1999; Breitburg et al. 2000; Peterson et al. 2003a and 2003b; ASMFC 2007). Species that are not commercially or recreationally important are still ecologically important in that they may feed on zooplankton or serve as prey for larger fish (Breitburg 1999; Coen and Luckenbach 2000; Harding and Mann 2000; Harding 2001), thus functioning as important links in the food chain. Some associated species may also contribute to water quality improvement through their feeding activities (Breitburg 1999; Coen and Luckenbach 2000). Oyster reefs also directly and indirectly provide food resources for numerous waterbirds (e.g., herons, oystercatchers, plovers, sandpipers, gulls, and terns) and aggregations of dead oysters can provide nesting and roosting sites.

The research questions associated with how natural and restored oyster reef habitat influences resident and transient species will vary greatly between projects. The metrics described in this chapter are focused on measuring the density of sessile and mobile invertebrates and vertebrates, including waterfowl. Traditional methods for recording species, abundance, density, and diversity of invertebrates and vertebrates on natural and restored oyster reefs include quadrats, substrate baskets and transects for invertebrates and resident finfish. Various types of nets have been used to sample fish and other nekton in oyster communities, including seines, lift-nets, breeder traps, and gill nets. Published information on growth rates of each species and empirical data on age-specific survivorship can also be analyzed for change in species and abundance over time. The per-unit-area enhancement of fish production and large mobile crustaceans expected from the addition of oyster reef habitat can then be calculated (see Peterson et al. 2003b; Grabowski and Peterson 2007).

7.1 Metric #1: Density of Selected Species and/or Faunal Groups

Selection of target species should be based on the individual restoration goal. For example, if the goal of the project is to increase biodiversity, then all species present should be quantified; but if the project goal is only to provide habitat for red drum, then only red drum need quantification. If the project goal is to increase the food supply for transients, then the relevant resident species should be quantified. Additionally, practitioners may also be interested in less desirable species such as predators, competitors, or invasive species and can quantify these species utilizing the applicable method(s) outlined below. See studies by Coen et al. (1999a, b), Berquist et al. (2006), Walters and Coen (2006) (quadrats and/or core samples), Grabowski et al. (2005) (core samples, trap samples, and gill nets), Luckenbach et al. (2005) (substrate baskets and gill nets), Rodney and Paynter (2006) (substrate baskets), Erbland and Ozbay (2008) (substrate baskets and lift nets), Wenner et al. (1996) and Coen et al. (1999a, b) (lift nets), Glancy et al. (2003) (throw-traps), Geraldi et al. (2009), Scyphers et al. (2011) (gillnets and seines) or similar studies for further information on sampling devices, as well as reviews on gear selection by Rozas and Minello (1997) and Coen et al. (1999b). It may be necessary to use multiple sampling methods and gear to properly assess the species and or/ faunal groups of interest. It is also important to note that habitat utilization by some species of resident and transient crustaceans, juvenile fish, and adult fish varies based on time of day (i.e., day vs. night) and, as such, practitioners should consider these potential differences when designing their sampling regime.

7.1.1a Suggested Methodologies for Invertebrates and Finfish

For Epifaunal Sessile Invertebrates:

Units = Density of each species (or lowest taxonomic grouping possible) (individuals/m^2), Wet weight by species (g/m^2), Shell height for non-oyster bivalves (mm), Percent cover (percent cover/m^2) for some organisms where density is not possible.

Figure 25. Mussels can often be as abundant as oysters in intertidal habitats. These mussels were collected in one 0.14 m² quadrat collected in South Carolina

Epifaunal sessile invertebrate samples can be obtained from oyster density and size distribution quadrat samples (Section 3.4). Remove all sessile invertebrates from the live and dead oyster shells collected in each quadrat sample (Figure 25). Organisms that cannot be removed without being destroyed (e.g., barnacles) should just be enumerated. Identify each sessile invertebrate to the lowest taxonomic level possible and enumerate. Blot all organisms as dry as possible and record a total wet weight (g) for each species.

Data should be reported on an individuals/m² and a wet weight g/m² basis. For non-oyster bivalve filter feeders such as mussels, the shell height of a subset of at least twenty individuals per species should be recorded. For some organisms, such as sponges, bryozoans and other colonial or encrusting organisms, density measurements will not be possible. For those organisms, percent cover should be recorded.

For Infaunal Invertebrates:

Units = Density of each species (or lowest taxonomic grouping possible) (individuals/m²), Wet weight by species (g/m²), Shell height for bivalves (mm)

At random points along the reef, take sediment core samples to a depth of 15 cm below the reef base using a 15 cm diameter core sampler. If reef is made up of several patch reefs, samples may be taken at random points across the project footprint. Additional core samples should be taken at random locations on a nearby mud flat or other nearby non-structured habitat. Samples should be washed through a 2mm mesh sieve (Figure 26). A smaller mesh size may be utilized (e.g., 500 μm) if desired, however, the portion retained on the 2 mm mesh sieve should be processed (identified and enumerated) first. All organisms retained on the sieve should be identified to the lowest possible taxonomic level and enumerated. Blot all organisms as dry as possible and record a total wet weight (g) for each species. Data should be reported on an individuals/m² and a wet weight g/m² basis. For bivalves, the shell height of a subset of at least twenty individuals per species should be recorded.

Figure 26. Rinsing a sample through a mesh sieve.

Figure 27. (a) Substrate baskets containing oyster shell prior to deployment. (b) Shell trays deployed on a fringing reef.

For Small Resident Mobile Fish and Invertebrates:

Units = Density of each species (or lowest taxonomic grouping possible) (individuals/m^2), Wet weight by species (g/m^2), Length (mm)

Substrate baskets (Figure 27) or trays (hereafter referred to as units) should be deployed on the reef and at the unrestored control site at random locations along the reef (or throughout the project footprint if the reef is constructed of several patch reefs). If the reef is a high relief reef, then units should be deployed near the reef crest, near the reef base, and in the sediment immediately adjacent to the reef. Additional units should be deployed at random locations on an adjacent mud flat or other nearby non-structured habitat. Units may be constructed of a material of the practitioner's choice (PVC, etc.) but must sample to a depth of 10 cm, have circulation/drainage holes located along the sides and bottom, and be lined with 1 mm mesh. Unit size should be scaled appropriately to the reef size and may range from 0.5 m x 0.5 m to 1 m^2 in size.

For each unit, a hole should be excavated in the reef/substrate and the unit should be placed in the hole so that it is flush with the reef/substrate surface. Excavated material should then be placed back into the unit. Units should remain in place for at least one month before collection. Before units are removed from the reef and sediments, their tops should be sealed with a small-mesh net or a cover to prevent organisms from escaping. Upon collection, contents of the units should be sieved through a 2 mm mesh sieve. A smaller mesh size may be utilized (e.g., 500 µm) if desired, however, the portion retained on the 2 mm mesh sieve should be processed (identified and enumerated) first. All organisms remaining on the mesh should be identified to the lowest possible taxonomic level, enumerated and measured. Record a total wet weight (g) for each species. Data should be presented on an individuals/m^2, wet weight g/m^2, and, if practical, length-frequency distribution per species.

Figure 28. Lift nets at (a) low tide before and (b) after being raised at high tide.

For Transient Crustaceans and Juvenile Fish:

Units = Density of each species (or lowest taxonomic grouping possible) (individuals/m^2), Wet weight by species (g/m^2), Length (mm)

Transient crustaceans and juvenile fish may be sampled in intertidal and shallow subtidal waters (<1.5 m) using either seines, lift nets, drop samplers, throw traps, or other similar gear (Figure 28). Sampling devices should sample an area of at least 1 m^2. If seine sampling, be sure to note the length of the seine and the distance traveled in order to determine the area sampled. Deeper subtidal waters may be sampled with visual surveys using video or divers. On-reef sampling is preferred, but if this is not possible, sampling should be performed immediately adjacent to the reef. If sampling adjacent to the reef, sampling should be performed at both the inshore and offshore sides of the reef. Samples should be taken at random locations along the reef or project footprint (if the reef is made up of several patch reefs). Additional samples should be taken at random locations on an adjacent mud flat or other non-structured habitat. Organisms collected should be identified to the lowest possible taxonomic level, enumerated and measured. Record a total wet weight (g) for each species. Data should be presented on an individuals/m^2, wet weight g/m^2 basis, and length-frequency distributions per species.

For Transient (typically larger) Fish:

Units = Catch per unit effort (CPUE) (individuals/hour), with wet weight (g/m^2) and Length (mm) by species.

Deploy gillnets (Figure 29) perpendicular to shore on both the nearshore and offshore side of reef (see Geraldi et al. 2009 and Scyphers et al. 2011 for examples of gillnet orientation). While 5 cm and 10 cm mesh sizes are commonly used, you may need to use a different mesh size for your target species. Mesh size may be determined using the following equation (Hubert 1996):

Mesh circumference* = optimum girth of target species/1.25

*Note that mesh circumference equals 2 x mesh stretch length

Nets should be set 1 hour before sunrise and sunset and have a 2 hour soak time. Upon retrieval, organisms retained in nets should be identified to the lowest possible taxonomic level, enumerated, measured, and weighed (wet weight). Data should be reported on a CPUE basis (individuals/hour) and by wet weight per species (g), and length frequency distribution per species.

Gillnets are not allowed in all states; check the regulations for the state in which your oyster restoration project is located and obtain any necessary permits to conduct the gillnet sampling.

Figure 29. Collecting finfish using a gillnet.

7.1.2a Sampling Frequency

At a minimum sampling should target the season that coincides with the maximum abundances of the target species, with repeated sampling performed during the expected time period. The abundance of transient species tends to be highly variable. In this case the sampling effort should be proportionately high in order to detect changes in relative abundance through time.

7.1.3a Performance Criteria

As monitoring progresses, there should be a trend of increasing relative abundance (CPUE) of the target species, with an ultimate goal of having statistically greater CPUE of target species than those present pre-construction or at the control site, or a relative abundance that is roughly equal to or greater than that of the natural reference site.

7.1.1b Suggested Methodologies for Waterbirds

Units: bird/ha (total and for each species or species group)

The number of observation points needed to evaluate waterbird use (Figure 30) will be determined by size of the restoration project footprint. For large projects, observation points should be spaced greater than or equal to (≥) 200 m apart within the bounds of the restored area and the control or natural reference area, which should also include all areas that could be affected by the reef restoration (i.e., landward mudflat development). If the project footprint or potential area of effect is small, an observation point should be established in the center of the area. Observation points should be accessible either by foot or boat and provide maximum visibility of the restored areas. If the project footprint or potential area of effect is small, or if more than one observation point can be accessed during low tide conditions, all observation points should be surveyed on each visit. If the restoration area is large or movement is restricted during low tide, survey one randomly selected observation point on each visit. If sampling occurs during a period when birds are primarily foraging and not roosting or nesting, observations should be made from the observation point; however, if sampling occurs during a period of bird roosting or nesting, then surveys of the observation points should be made remotely.

Figure 30. (a) Waterbirds utilizing intertidal oyster habitat and (b) American Oyster Catchers.

Depending on the number of observation points surveyed per visit, conduct as many scan samples as possible, at a maximum of one per hour, starting from one hour after high tide to one hour before low tide or vice versa (one hour after low tide to one hour before high tide). These repeated scans are necessary because waterbird use changes in response to changing tide level. During each scan, count the number of large waterbirds within a 100 m radius (r) of the observation point. This plot size is ideal because, at this distance, few birds will be obstructed from view by exposed oyster beds, which at low tide can hinder viewing from a greater distance. If the observer is unable to accurately estimate distance, markers can be placed at the edge of the radius by using a range finder. Identify all waterbirds to the species level; however, if shorebirds flush or are unidentifiable, record them to the nearest species group (e.g., yellowlegs, dowitchers, small calidrine sandpipers). In addition, categorize the behavior of every observed waterbird as feeding, roosting, or nesting, as well as the substrate on which the waterbird is standing (e.g., oyster bed, mud flat, sand bar, etc.). For ease, sampling events should utilize two observers, with one observer counting and observing behavior and a second observer serving as a scribe. Alternatively, a single observer can record data on a voice recorder.

Restored and control or natural reference sites should be surveyed within a few days of each other to reduce seasonal effects. Surveys should only be conducted under environmental conditions that permit identification of waterbirds with a good binocular or spotting scope (generally only up to light rain and moderate winds). Although it is important to survey roosting and nesting areas, observers should always use caution near these areas and minimize activities that alter birds' behaviors.

This procedure assumes that all species are detectable (visible on the mudflat or oyster reef) and that all of the restored and control or natural reference area is visible to the observer. Data should be reported on a birds/ha basis for total birds and by each species or species group. (Note: the formula for area is ϖr^2 and 1 hectare (ha) is equal to 10,000 m^2). Because multiple counts are performed during sampling events, data for the counts should be averaged per sampling event. Behavior data should be summarized as the proportions of observations in each of the three behavior classes (feeding, roosting, or nesting).

7.1.2b. Sampling Frequency

Because the abundance of migrant waterbirds changes through the spring and fall migration periods, counts should be repeated several times throughout the year. Following the recommendations of the International Shorebird Survey (ISS), the site should be surveyed three times during each of the three birding seasons. During the spring migration season (spring/early breeding period), the first count should be conducted between March 15 and April 6, the second between April 7 and April 29, and the third between April 30 and May 22. The last period should cover use of the site by breeding birds. For the fall season (late breeding/fall period), the first count should be conducted between July 15 and August 15, the second between August 16 and September 15, and the third between September 16 and October 15. To assess winter waterbird use of the site, surveys should be conducted once a month from December 1 to February 28. Counts should be made at least 14 days apart.

7.1.2b. Performance Criteria

As monitoring progresses, there should be a trend of increasing density of waterbirds, with an ultimate goal of having statistically greater densities of waterbirds than those present pre-construction or at the control site, or a density that is roughly equal to or greater than that of the natural reference site.

Chapter 8: Restoration Goal-Based Metrics: Enhancement of Adjacent Habitats

Habitat loss and disturbance are ranked worldwide as the top threats to biodiversity (Roberts et al. 2002; Thrush and Dayton 2002; Hoekstra et al. 2005; Airoldi and Beck 2007; Airoldi et al. 2008). Natural and anthropogenic hydrodynamic forces erode or disturb critical marsh-edge habitats (e.g., *Spartina spp.*, *Juncus spp.*), affect coastal sediment transport processes, and impact estuarine species (Grizzle et al. 2002; Wall et al. 2005). The loss and/or disturbance of marsh-edge habitat may reduce estuarine productivity and negatively impact numerous commercial and recreational fisheries that depend upon this unique habitat (Meyer et al. 1997). Additionally, hydrodynamic effects on these important fringing habitats may diminish their associated ecosystem services (Coen et al. 2007, 2011a; Cicchetti and Diaz 2000; Weinstein and Kreeger 2000; Grabowski and Peterson 2007; NRC 2007), such as their role as important buffers affecting water quality (Holland et al. 2004; Mallin and Lewitus 2004; Mallin et al. 2004; Long et al. 2006).

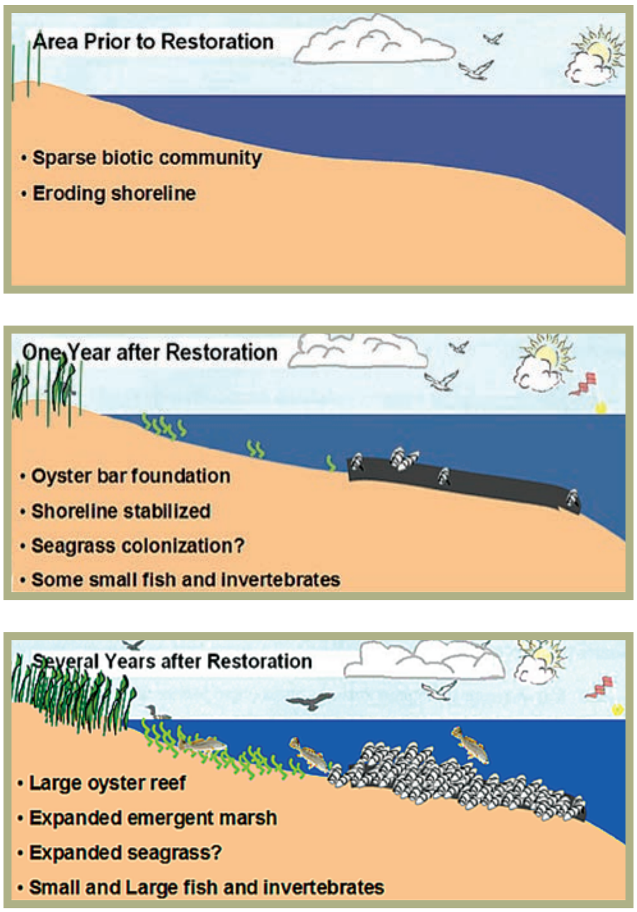

Figure 31. Possible effects of oyster restoration projects on adjacent habitats.

Intertidal and subtidal oyster reefs can help protect adjacent vegetated habitats from natural and anthropogenic-derived waves, currents and tides (e.g., Piazza et al. 2005; Scyphers et al. 2011). This lessening of wave action may also allow sediments to accumulate inshore (landward) of the reef, stabilizing the shoreline. This shoreline stabilization and sediment accumulation can benefit nearby marsh habitat by both protecting the marsh from erosion and even possibly allowing the marsh to expand due to the accretion of sediments. Oyster habitats may also aid in the creation or protection of submerged aquatic vegetation (SAV) habitat through sediment stabilization and improvements in water quality that often occur as a result of water filtration by the oysters (reviewed in Geraldi et al. 2009 and Coen et al. 2011a) (Figure 31).

Increasingly, oyster reefs are being incorporated into shoreline protection and restoration projects that are of a more natural design and utilize natural materials, such as rocks and logs, and vegetation, rather than bulkheads. Shoreline protection projects of this type are referred to as “living shorelines,” and in these projects constructed oyster reefs often serve as breakwaters designed to mitigate wave action and promote the inshore accumulation of sediments (Figure 31). In addition to shoreline protection, the oyster reef breakwaters also provide associated ecosystem services (e.g., habitat provision for fish and invertebrates, improved water clarity, etc.). Living shoreline-type projects that include oyster reefs as part of their design may also be considered to be oyster restoration projects and, as such, should be monitored accordingly. As with any shoreline protection or restoration project, knowledge of local tidal, flow, and sediment transport characteristics is key during the site-selection and design processes, and will aid in post-construction data interpretation.

Measurements of both shoreline loss/gain (the change in shoreline position) and shoreline profile/elevation change will allow practitioners to document the degree to which the oyster reef is abating erosion on the adjacent shoreline, and/or enhancing accretion of the shoreline. Measurements of the density of marsh plants (or mangroves or other shoreline plant habitat) can determine the effects the oyster reef may be having on nearby plant communities. The following is a general methodology for deploying the permanent transects required for measuring these metrics. More detailed

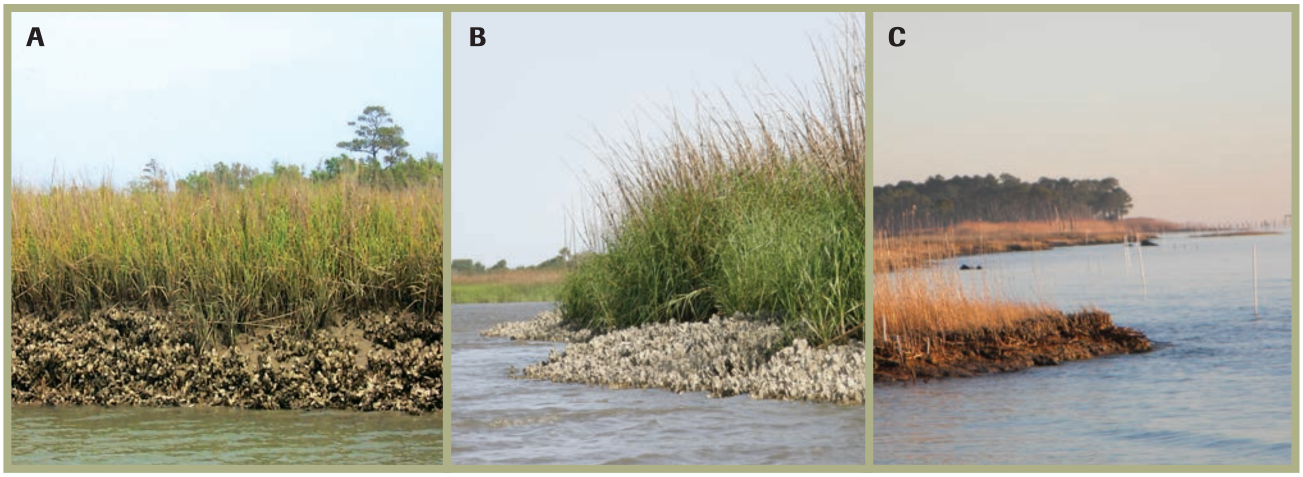

Figure 32. Images of various shoreline edges in saltmarshes. (A) South Carolina, (B) Georgia, (C) Alabama.

metric-specific methods are presented in the sub-section describing each metric. For the purpose of this handbook, the landward shoreline edge (hereafter referred to as the shoreline edge) is defined as the lower/seaward extent of the emergent macrophyte vegetation (Figure 32). Measurements should be performed on the shoreline adjacent to the constructed reef and at a control site with similar current and wave conditions. Practitioners should perform pre- and post-construction monitoring at both the construction site and the control site.

Establishing Permanent transects

Metrics contained within this chapter require the establishment of permanent transects. Permanent transects should be established along the length of the restored reef, with the 'base stake' starting at least 10 m inland of the shoreline edge (farther inland if project site is in an area subject to high erosion rates), and continuing to the constructed reef. Base stake locations should be marked with a dGPS (or a mapping/survey grade GPS with post-processing for sufficient accuracy). Transects should extend from the permanent base stakes along an established compass heading and extend to the restored reef (Figure 33). Permanent transects should also be established along an equal linear distance of shoreline edge at the control or natural reference site.

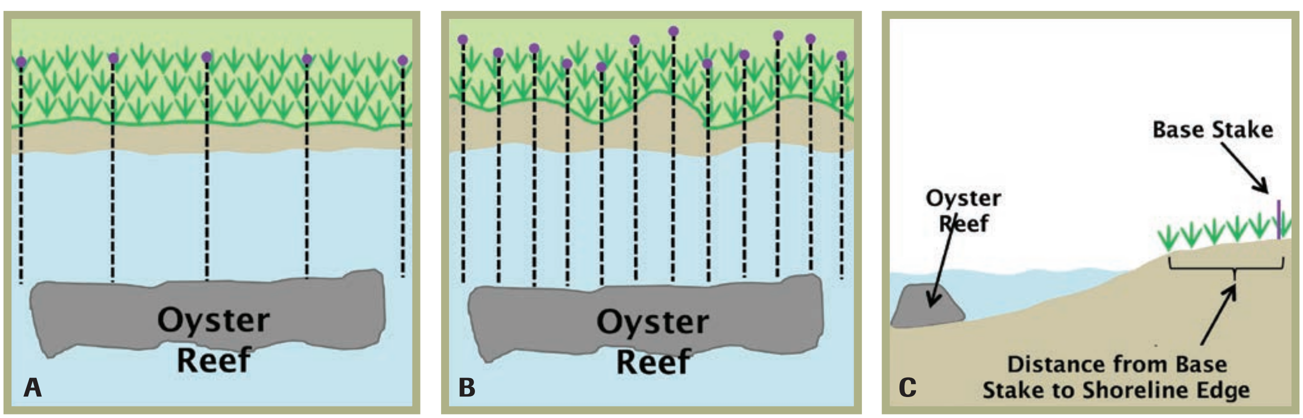

Figure 33. Overhead and cross-sectional views of example layouts of permanent transects (dashed black lines) and base stakes (purple dots). Shoreline edges (green lines) with a low degree of sinuosity or irregularity (A) will require fewer permanent transects, whereas shoreline edges with a high degree of sinuosity and irregularity (B) will require more permanent transects. When measuring shoreline loss/gain (C), practitioners should measure the distance from the base stakes (purple line) to the shoreline edge.

Due to the high degree of variability of shoreline characteristics both within and between restoration sites, the number of permanent transects that should be established will depend on the characteristics of a particular restoration site. For example, if the shoreline adjacent to the restoration site is generally linear, with a low degree of sinuosity or irregularity, then fewer permanent transects need to be established. If the shoreline adjacent to the restoration site has a greater degree of sinuosity and irregularity, then more permanent transects are needed to adequately document changes in shoreline.

With any permanent base stakes deployed in a coastal environment, there is a possibility of one or more stakes becoming dislodged and, as such, practitioners should reinforce the permanent base stakes appropriately. Permanent base stakes should be constructed of materials that will withstand harsh environmental conditions and should be driven as far into the substrate as possible (in some environments this may be several meters), leaving 1 m of the stake exposed. Practitioners should periodically check the permanent base stakes and reset or replace them as necessary, being sure to record the locations of any new base stakes with a dGPS.

8.1 Metric #1: Shoreline Loss/Gain (Change in Shoreline Position)

Units = Shoreline loss/gain (m/year)

8.1.1 Suggested Methodologies

The following methodologies represent low- and high-tech options for measuring shoreline loss or gain.

Using a dGPS:

Walk the shoreline edge adjacent to the constructed reef, taking continuous readings with a dGPS. It is preferred that the entire shoreline edge adjacent to the restored reef be measured, but if the project is very large or this is otherwise not possible, a minimum of 100 m of shoreline edge at each end and in the middle of the project site should be measured. Data should be entered into mapping software, such as ArcGIS, and mapped over a geo-referenced basemap. Repeat this process for an equal lateral distance of shoreline edge at the control or reference site.

Using a tape measure:

Using a tape measure, measure the linear distance from the permanent base stakes to the shoreline edge along the established transects. When taking measurements, practitioners should keep the length of measuring tape as taut as possible. Data should be entered into mapping software, such as ArcGIS, and mapped over a geo-referenced basemap.

Using Surveying Instrumentation:

If a practitioner is familiar with basic surveying techniques and is proficient with advanced surveying instrumentation (e.g., a Total Station or other instruments used to find horizontal and vertical angles and distances), then the practitioner may perform a topographic survey along each of the transects using these instruments. If using surveying instrumentation, data for the shoreline loss/gain metric and the shoreline profile/elevation change metric (Section 8.2) can be obtained simultaneously by performing a topographic survey. Take elevation/location measurements at regular intervals from the permanent base stakes to the shoreline edge (or continuing all the way to the constructed reef if also collecting information for the profile/elevation change metric) along each transect. Data should be entered into mapping software, such as ArcGIS, and mapped over a geo-referenced basemap.

Using Aerial Photography:

When using aerial imagery, be sure that you use appropriate geo-referencing tools (permanent base stakes with markers that are visible from the air, and the presence of a permanent feature) for measuring shoreline position and for scaling purposes. If possible, ortho-rectified aerial photographs (geometrically corrected to have a uniform scale) should be used. Using the aerial images, measure the linear distance from the permanent base stakes to the shoreline edge along the transects. Data should be entered into mapping software, such as ArcGIS, and mapped over a geo-referenced basemap.

8.1.2 Calculations

To determine shoreline loss/gain along each transect, subtract the current year's shoreline linear distance from the permanent base stake from the previous measurement's linear distance. Positive values denote land gain while negative values denote land loss.

8.1.3 Sampling Frequency

Measurements should be taken once prior to construction, within three months post-construction (to document the as-built project footprint and reef area), and annually thereafter. Additional measurements after events that could alter shoreline position (e.g., hurricanes) are also recommended.

8.1.4 Performance Criteria

As monitoring progresses, there should be a trend of decreasing shoreline loss, or shoreline gain, with an ultimate goal of having statistically less shoreline loss or greater shoreline gain than pre-construction conditions and at the control or reference site.

8.2 Metric #2: Shoreline Profile/Elevation Change

Units = Shoreline profile/elevation change (meters/year), Shoreline slope (i.e., rise/run) (unitless)

For the methodologies listed below, it is suggested that a dGPS be used to mark the locations of the measurements taken. For all methodologies, measurements should be taken at the project site and at a control site.

8.2.1 Suggested Methodologies

Using Surveying Instrumentation

If a practitioner is familiar with basic surveying techniques, then traditional surveying equipment such as a surveyor's level or laser level and graduated rod, can be used to create profiles and measure elevation change for each transect. Take elevation measurements at 1 m intervals, moving seaward from the permanent base stake to the reef, being sure to note location and elevation of the shoreline edge. Repeat this process for an equal linear distance of shoreline edge at the control site.

Similarly, if a practitioner is proficient with advanced surveying instrumentation (e.g., a Total Station or other instruments used to find horizontal and vertical angles and distances), then the practitioner may perform a topographic survey along each of the transects using these advanced surveying instruments. If using advanced surveying instrumentation, data for the shoreline profile/elevation metric and the shoreline loss/gain metric (Section 8.1) can be obtained simultaneously by performing a topographic survey. Using a Total Station or similar advanced surveying instrumentation, perform a topographic survey by taking elevation measurements at regular intervals (determined by the total transect length) along each transect from the permanent base stake to the reef, being sure to note location and elevation of the shoreline edge. Repeat this process for an equal linear distance at the control site.

8.2.2 Calculations

The mean elevation change can be calculated by taking the mean of the shoreline slopes of all transects. The shoreline profile (elevation change data per measured point) for each transect should be graphed as a line graph, and an overall shoreline profile for the site can be obtained by calculating the mean elevation change of all transects for each measured point.

8.2.3 Sampling Frequency

Measurements should be taken once prior to construction, within three months post-construction (to document the as-built project footprint and reef area), and annually thereafter. Additional measurements after events that could alter shoreline profile (e.g., hurricanes) are also recommended.

8.2.4 Performance Criteria

As monitoring progresses, there should be a trend of decreasing slope and increasing mean elevation at the shoreline, with an ultimate goal of having statistically lower shoreline slope or increased mean shoreline elevation than pre-construction conditions and at the control, or a decreased 'step' at the waters edge (e.g. Figure 34).

Figure 34: An oyster reef dampens wave action at Coffee Island, Alabama.

8.3 Metric #3: Density and Percent Cover of Marsh/Mangrove Plants

If the oyster restoration project is located seaward of a shoreline on which marsh or mangrove plants are located, and if your project is specifically documenting the effects of a restored reef on shoreline plant habitats, then measurements of the density and percent cover of marsh/mangrove forest plants should be performed.

Units = Density (live shoots/m^2), Percent coverage (%) of each species present

8.3.1 Suggested Methodology

Measurements of plant density should be taken along the transects every 2 m starting at the shoreline edge and extending to at least 4m inland of the marsh/mangrove forest edge. Measurements should be taken by placing a 1m x 1m quadrat on the substrate and counting the number of live stems of different species of marsh vegetation present within the quadrat (a 0.5 m x 0.5 m quadrat may be used if vegetation is particularly dense). In areas where mangroves are present, the use of a quadrat with four solid sides (e.g., a square constructed of PVC) is not possible. Instead, practitioners should mark out the corners of a 1 m^2 plot on the ground using surveyor's flags or PVC pipe segments to serve as a quadrat. If vines or other spreading species are present, a visual estimate of the percent of substrate covered by each of these species should be taken. Density measurements should be reported on a shoots/m^2 basis for each species, and percent coverage should be reported as a percentage for each species. Repeat this process for an equal linear distance of shoreline edge at the control or reference site.

8.3.2 Sampling Frequency

Sampling of marsh/mangrove vegetation should be performed annually at the end of the peak growing season.

8.3.3 Performance Criteria

As monitoring progresses, there should be a trend of increasing mean plant density and mean percent coverage, with an ultimate goal of having statistically greater mean plant density and mean percent coverage than pre-construction conditions and at the control site, or a mean density and mean present coverage that is roughly equal to that of the natural reference site.

8.4 Ancillary Monitoring Considerations for Enhancement of Adjacent Habitat

The ancillary monitoring considerations listed below may aid in understanding data gained from measurements of shoreline position and profile, or may provide information on secondary effects of oyster reefs. Not all areas are suitable for submerged aquatic vegetation (SAV) growth and, as such, the performance of a restored reef should not be judged on the presence/absence or coverage of SAV in areas near the reef. If a restored reef is located in an area with current or historic SAV coverage, or is located in an area where SAV restoration is occurring, measurements of SAV percent coverage and density can provide information as to how reef-induced improvements in water clarity and/or sediment stabilization may be affecting nearby habitats.

Measurements of wave energy and tidal water flows may provide information about why any changes in shoreline position and profile may be occurring, but reef-related alterations of wave energy and tidal flows do not, in themselves, guarantee a neutral or positive change in shoreline position and profile. In the future, wave energy and tidal water flow information might provide insight as to how much these forces need to be reduced in order to see reduced erosional effects on the shoreline, but currently their values do not directly translate into shoreline protection values.

8.4.1 Submerged Aquatic Vegetation

The presence of oyster habitat may increase SAV coverage through water clarity improvements and/or sediment stabilization. Sampling of SAV should be performed shoreward of restored reef and at a non-restored control site with similar current and wave conditions. Pre-construction data is recommended as well, if possible.

Units = Density (shoots/m^2), Percent coverage (Braun-Blanquet scale)

8.4.1.1 Suggested Methodology

Measurements should be taken at three locations (1 m shoreward of reef, midpoint between reef and shoreline, and 1 m seaward of shoreline) along transects that were previously established for the shoreline loss/gain (Section 8.1), shoreline profile/elevation change (Section 8.2), and marsh/mangrove plant density (Section 8.3) metrics. At each sampling location, place a quadrat on the substrate and count the number of SAV shoots present within each quadrat (SAV shoot density). Also, record a visual estimation of the percentage of substrate covered by the SAV within the quadrat (SAV percent coverage) using the modified Braun-Blanquet scale outlined in Fourqurean et al. (2001):

0 = no seagrass present in quadrat
0.1 = a solitary shoot, <5% cover
0.5 = less than 5 shoots, <5% cover
1 = greater than 5 shoots, <5% cover
2 = greater than 5 shoots, 5 – 25% coverage
3 = greater than 5 shoots, 25 – 50% coverage
4 = greater than 5 shoots, 50 – 75% coverage
5 = greater than 5 shoots, 75 – 100% coverage

Example photographs of various seagrass percent coverages may be found in Short et al. (2006). A SCUBA diver may be utilized for density and percent cover measurements if necessary. Density measurements should be reported on a SAV shoots/m^2 basis, and SAV percent coverage should be reported as a Braun-Blanquet score.

8.4.1.2 Sampling Frequency

Measurements of SAV should be performed annually near the end of the SAV growing season (generally in the late summer/early fall).

8.4.2 Wave Energy and Tidal Water Flows

Measurements of wave energy and tidal water flow should be performed seaward and landward of the restored reef to measure the decrease in wave energy and wave flow due to the presence of the reef, and also at a non-restored control site with similar current and wave conditions. Pre-construction data is recommended as well if possible.

8.4.2.1 Suggested Methodologies

Units = Tidal water flow (m/sec); Wave height (m)

Tidal Water Flow (i.e., tidal velocity)

Measurements of tidal water flows should be taken both seaward and landward of the reef at regular intervals along the length of the reef, and along an equal lateral distance and approximately equal depth at the natural reference or control site. Measurements should be made at mid-depth in the water column during flood and ebb tide using instrumentation such as an electromagnetic flow meter or an acoustic Doppler current velocimeter and should be reported in m/sec.

If instrumentation is not available, current velocity may be estimated by measuring the time it takes for an orange, a drifter or drogue to travel a known distance (report in m/sec). Report the mean tidal flow landward of the reef and the mean tidal water flow seaward of the reef in m/sec To determine the degree to which tidal water flow was affected by the presence of the reef, subtract the mean tidal water flow landward of the reef from the mean tidal water flow seaward of the reef.

Wave Energy (i.e., wave height)

Measurements of wave height should be taken both seaward and landward of the reef at regular intervals along the length of the reef, and along an equal lateral distance and approximately equal depth at the natural reference or control site. To measure wave height, stand adjacent to the reef and using a graduated surveyor's rod, measure the height of the wave crest and the height of the trough. To calculate wave height, subtract the trough height from the breaker height. Repeat this for 10 successive waves and report the mean in m. If possible, perform wave height measurements landward and shoreward of the reef concurrently. To determine the degree to which wave height was affected by the presence of the reef, subtract the mean wave height landward of the reef from the mean wave height seaward of the reef. Data loggers are also available that will record water depth on a short time interval. These records can be used to generate mean wave amplitude or a mean of the maximum minus minimum water depth calculated over multiple periods of minutes.

8.4.2.2 Sampling Frequency

Measurements of tidal water flow and wave energy should be made every time other sampling is performed at the reef, or at a minimum, quarterly.

Chapter 9: Restoration Goal-Based Metrics: Water Clarity Improvement

Bivalves such as oysters can play an important role in regulating local water clarity through their filtration activities. They can decrease turbidity, and thus improve water clarity, by removing seston, minute living (e.g., plankton) and non-living (e.g., sediment) particles, from the water column (see discussion in Grabowski and Peterson 2007, Kellogg et al. 2013, zu Ermgassen et al. 2012b, 2013). The decreased turbidity, along with the transfer of particulate material, including nutrients, from the water column to the sediment (benthic-pelagic coupling) provided by bivalve filtration, can have beneficial effects on nearby benthic habitats such as seagrass beds (Peterson and Heck 2001; Newell and Koch 2004; Wall et al. 2008; Booth and Heck 2009). Bivalves also aid in removing heavy metals, toxins, and fecal coliform from the water column through their filtration activities, and as such, have been utilized in the bioremediation of effects of industrial or other anthropogenic pollution (e.g., Gifford et al. 2005). To assess the performance of a restored oyster reef in improving water clarity, practitioners may choose to measure one or more of the following aspects of water clarity: (1) seston concentration; (2) chlorophyll *a* concentration; or (3) light penetration. Methodologies for these metrics are provided below. If oysters are used in bioremediation efforts, then practitioners may choose to sample additional parameters (e.g., toxins, heavy metals, bacteria) of interest.

While the effects of oyster habitats on water clarity may be measured directly through measurements of seston concentration, chlorophyll *a* concentration and light penetration, oyster filtration rates may be estimated using more indirect methods. For example, several studies have demonstrated a positive correlation between oyster biomass and biodeposition and/or filtration (Newell and Koch 2004; Sisson et al. 2011), and as such, oyster filtration rates can be estimated using oyster biomass. Oyster filtration rates can be affected by oyster size, as well as several environmental factors such as seston concentration and particle size, salinity, temperature, and water flow rates (e.g. Cerco and Noel 2005, 2007). There are several models used for estimating oyster filtration rates that take into account possible effects of various environmental factors (e.g. Cerco and Noel 2005, 2007). It is suggested that interested practitioners derive their own model of oyster filtration rates based on their available environmental data and any species-specific considerations. See Cerco and Noel (2005, 2007), Fulford et al. (2007), or zu Ermgassen et al. (2012b) for *C. virginica* filtration models that can be modified to suit your available data. For *O. lurida*, see zu Ermgassen et al. (2013) for a filtration model that is applicable to West Coast oyster habitats.

In addition to their positive effects on water clarity, oysters play an important role in coastal biogeochemical cycles by regulating carbon, nitrogen, and phosphorous fluxes through the sequestration of C, N, and P in their shells and tissues and by contributing to denitrification processes. While some of the nitrogen that oysters filter from organic matter in the water column is retained in their tissues, other nitrogen is delivered to the sediments in the form of biodeposits (feces and pseudofeces). The nitrogen present in these biodeposits may then be converted into nitrogen gas through nitrification and denitrification. This nitrogen gas diffuses from the sediment into the water column, and then into the atmosphere (sees Sisson et al. 2011 and references therein for more detailed information).

The methodologies for measuring the denitrification and nutrient fluxes associated with oyster reefs are developing, with likely advances in the near future. As a result, no standard technique for the measurement of denitrification is provided. This does not detract from the importance of denitrification by oyster habitats and the utility of measuring this ecosystem service. Sampling methodologies and analytical techniques for measuring nitrogen retention and removal in benthic habitats can be found at the following references: Byers et al. (1978), Seitzinger (1987), Kana et al. (1994), Miller-Way and Twilley (1996), Kana et al. (1998), Lavrentyev et al. (2000), An et al. (2001), Eyre and Ferguson (2002), Eyre et al. (2002), Gardner et al. (2006), Smith et al. (2006), McCarthy et al.(2007), Tobias et al. (2007), Hochard et al. (2010), Piehler and Smyth (2011) and Kellogg et al. (2013).

9.1 Metric #1: Seston and/or Chlorophyll a Concentrations

Seston concentration (includes total particulates and organic content) and chlorophyll *a* are commonly measured metrics in water quality studies. Sampling for seston and/or chlorophyll *a* concentrations usually involves the collection of water samples at various places on the reef, as well as immediately up-current and down-current of the reef; however, newer methodologies that involve the use of *in situ* fluorometry have been successfully performed in the field (Grizzle et al. 2006, 2008). Further information on sampling methodologies and analytical techniques used for water quality studies can be found at Judge et al. (1993), Welschmeyer (1994), Cressman et al. (2003), Nelson et al. (2004), Grizzle et al. (2006), Grizzle et al. (2008), Booth and Heck (2009), and Plutchak et al. (2010) to name a few. Practitioners can contact any water quality lab (often affiliated with a state agency, an academic institution, or a local cooperative extension) to process water samples.

Sampling of seston and/or chlorophyll *a* concentrations should be performed pre- and post-construction at the restored reef as well as at a control site or natural reference reef. Each sample set should be accompanied by a measurement of the water depth at the mid-point of the reef and by flow measurements. Flow rates may be determined by measuring the amount of time it takes for an orange, a drifter or drogue to cover a known distance and should be reported on a cm sec^{-1} basis (see Grizzle et al. 2006, 2008 for further information on measuring water flow in intertidal and shallow subtidal habitats). Flow rates may also be determined using an instrument such as an electromagnetic flow meter or an acoustic Doppler current velocimeter (see also section 8.4.2.1)..

Units = Total Particulates (mg/l); Organic Content (%); Chlorophyll *a* (µg/l)

9.1.1 Suggested Methodology

For Seston Concentration (Total Particulates and Organic Content)

For particulates, collect triplicate (n = 3) 1 liter water samples at three locations (midpoint of reef, 0.5 m up-current of reef, and 0.5 m down-current of reef) at 5-10 minute intervals following both slack ebb tide and slack flood tide. Ten sets of triplicate samples should be taken overall. Note the distances between the sample locations in m, as well as the water depth at the midpoint of the reef. When sampling, water samples should be taken mid-depth between the water surface and the reef surface and sampling bottles should be opened and closed under the water at the desired depth to avoid surface contamination. Each water sample should be filtered through a pre-weighed 1 µm GF/F or GF/C glass fiber filter, dried to a constant weight at 40°C, and weighed to determine total particulates (subtract filter weight from this value to determine total particulate weight). Total particulates should be reported in mg/l.

To determine organic content, combust filters at 450°C for four hours, then cool and weigh (Nelson et al. 2004; Grizzle et al. 2006; and also Judge et al. 1993). Organic content is calculated by subtracting the post-combustion weight from the pre-combustion weight. Values should be reported as percent organic content (calculated by dividing the total particulate value by the organic content value).

For Chlorophyll a

For chlorophyll *a*, collect triplicate 50 ml water samples at three locations (midpoint of reef, 0.5m up-current of reef, and 0.5m down-current of reef) at 5-10 minute intervals as early in both ebb tide and flood tide as possible. When sampling, water samples should be taken mid-depth between the water surface and the reef surface and sampling bottles should be opened and closed under the water at the desired depth to avoid surface contamination. Six sets of samples should be taken overall. Determine chlorophyll a content using the fluormetric technique of Welschmeyer (1994) (Nelson et al. 2004), and report values in µg/l.

9.1.2 Sampling Frequency

To obtain appropriate baseline data, sampling of seston and/or chlorophyll *a* concentrations should be conducted quarterly to discern seasonal differences in the year prior to reef construction. Post-construction sampling of seston and/or chlorophyll *a* should also be conducted quarterly to discern seasonal differences.

9.1.3 Performance Criteria

As monitoring progresses, there should be a trend of decreasing total particulates, organic content, and/or chlorophyll *a* values, with an ultimate goal of having statistically lower values than pre-construction conditions and at the control site, or roughly equal to that of the natural reference site. In addition, throughout the monitoring period, seston and/or cholorphyll a concentrations should be statistically lower on-reef and immediately down-current of the reef than concentrations found immediately up-current of the reef and at off-reef or control locations.

9.2 Metric #2: Light Penetration Measurements

By removing seston from the water column through their filtration activities, oysters increase water clarity, which in turn allows ambient light to penetrate more deeply into the water column. This increased light penetration can, in turn, have beneficial effects on SAV populations in areas where light had previously been limiting. While light penetration can be measured using low tech methods such as secchi discs and transparency tubes, handheld and *in situ* instrumentation that measures light intensity may also be used. It is important to note that each of these instruments measures a different aspect of light penetration (i.e., a secchi disc measures the depth to which light penetrates, whereas light sensors measure the light penetration at depth), and as such, practitioners should be consistent with the instrumentation used to measure light penetration and should report all data in the appropriate units.

Units = Vary depending on instrumentation used. See methodologies for units.

9.2.1 Suggested Methodology

Using in situ Light Sensors

Units = lux

Deploy an array of 5 light intensity sensors at roughly the center of the reef (or center of the project footprint if the reef consists of patch reefs), 50 m up-current of the reef, and 50 m down-current of the reef (Figure 35). In each array, the height of the sensors should be as follows:

1 sensor (the ambient light sensor) should be located so that it will not be submerged during high tide and data gained from it should be used to normalize the light field relative to ambient conditions.

2 sensors ("k" sensors) should be deployed 30.5 cm (12") above the reef condition sensors and data gained from these sensors should be used to calculate the k-coefficients across the reef.

2 sensors (reef condition sensors) should be deployed 10 cm above the reef surface.

All light sensors should be mounted so that the sensor is facing up (unless the instructions for that particular sensor state differently). Sensors should take continuous measurements every five seconds for one hour just after slack high tide (measurements should commence at the beginning of ebb tide). Measurements should be reported in lux and a grand mean should be calculated for each location. Please note that this methodology is meant to provide information on the relative differences in light penetration (in lux) between the restored site and pre-construction conditions or conditions at control or reference sites, and not the amount of photosynthetically active radiation (PAR) present.

Figure 35. A reef with light sensors deployed. Note: The sensors depicted have been deployed in a grid pattern, not as described in the text.

Using Handheld Instrumentation

Units = will vary by instrument, but may be lux or $\mu Es^{-1}m^{-2}$

The methods for measuring light penetration using handheld instrumentation will vary by the instrument used. Reference readings of ambient air irradiance should be taken in conjunction with irradiance readings taken at depth. Practitioners may either measure irradiance at regular depth intervals until the reef surface is reached in order to construct a light extinction curve, or only take irradiance readings just above the reef surface. Triplicate readings should be taken immediately up-current and down-current of the reef, as well as at three points on the reef (the mid-point and midway between the mid-point and both ends of the reef). Time of day and tidal stage during which measurements were taken should be noted.

Using a Secchi Disk

Units = Depth of disappearance (cm)

Secchi discs should be 20 to 30 cm in diameter and have alternating black and white quadrants (Figure 36), and be attached to a weighted line that is marked in 1 cm increments. To reduce water surface glare, readings should be taken from the shady side of the boat, or if in shallow waters, with the practitioner's back toward the sun. To take the readings, slowly lower the secchi disc into the water until it disappears. Then slowly raise and lower the disc slightly to determine the exact depth of disappearance, and record this depth. Triplicate readings should be taken immediately up-current and down-current of the reef, as well as at three points on the reef (the mid-point and midway between the mid-point and both ends of the reef). Time of day and tidal stage during which measurements were taken should be noted. Further information on measuring turbidity using a secchi disk can be found in the EPA Voluntary Estuary Monitoring Manual (Ohrel and Register 2006, http://water.epa.gov/type/oceb/nep/monitor_index.cfm).

Figure 36. Using a secchi disk to measure water clarity.

Using a Transparency Tube

A transparency tube is a clear plastic tube with marked units (preferably millimeters or centimeters), similar to a graduated cylinder, and with a black and white pattern on the bottom similar to that found on a secchi disk. To measure water clarity, collect a water sample using a bucket from mid-depth between the water surface and the top of the reef. Gently swirl the water in the bucket to homogenize the sample and while looking down into the tube carefully pour the water into the transparency tube, avoiding bubbles, until the black and white pattern at the bottom of the tube is no longer visible. Record the depth at which the pattern disappears. Triplicate readings should be taken immediately up-current and down-current of the reef, as well as at three points on the reef (the mid-point and midway between the mid-point and both ends of the reef). Time of day and tidal stage during which measurements were taken should be noted.

Further information on measuring turbidity using a transparency tube can be found in the EPA Voluntary Estuary Monitoring Manual (Ohrel and Register 2006, http://water.epa.gov/type/oceb/nep/monitor_index.cfm).

9.2.2 Sampling Frequency

To obtain appropriate baseline data, sampling for light penetration should be conducted quarterly in the year prior to reef construction. Post-construction sampling of light penetration should, at a minimum, be conducted quarterly to discern seasonal differences.

9.2.3 Performance Criteria

As monitoring progresses, there should be a trend of increasing light penetration, with an ultimate goal of having statistically greater values than pre-construction conditions and at the control site, or roughly equal to that of the natural reference site. Throughout the monitoring period, light penetration should be greater on-reef and immediately down-current of the reef than found immediately up-current of the reef and at off-reef or control locations.

Chapter 10: Closing Remarks

The field of restoration ecology is relatively new and developing rapidly. Increasingly, habitat restoration is recognized as an important, if not central, part of landscape-scale conservation efforts and ecosystem-based management (Schmitz 2012). Development of effective restoration methods that can be applied at a meaningful scale depends on the ability to set and measure progress toward clear, ecologically and socially meaningful goals. Measuring progress toward a common set of goals using widely adopted performance metrics not only allows project managers to infer progress toward goals at the project scale, but also helps with comparison across projects (Moreno-Mateos et al. 2012; Powell et al. 2008). A comparison of projects at a regional or national scale will help highlight successful techniques and advance the field of oyster restoration more rapidly.

The imperative for well-considered monitoring of oyster restoration projects progresses along a continuous scale. On the simplest level there is the necessity to understand whether a project has contributed towards restoration goals. For example, are there more oysters as a result of the work, or is there stable substrate persisting through time. Projects that add substrate or live oysters without monitoring even the short term performance of the work can no longer be justified (Brumbaugh et al 2006). At the intermediate level there is an obligation on the restoration community to learn as efficiently as possible from the expanding body of restoration being conducted nationally, and to an increasing extent internationally. A basic pre-requisite for being able to learn from the collective body of oyster restoration is the ability to compare between projects (Kennedy et al. 2011).

At the overview level it is becoming increasingly relevant to set goals for oyster restoration based not only on a concept of returning the environment to some historic level of oyster habitat (Beck et al. 2011, zu Ermgassen et al. 2012a), but also on the amount of the ecosystem services provided by oyster habitat that is desired for a particular estuarine system (Brumbaugh et al. 2010). For example a restoration goal could be restoring sufficient oyster reef to make oyster filtration the dominant impact on water clarity through filtration rather than purely through tidal exchange (zu Ermgassen et al. 2012b and 2013). Similarly oyster reefs are particularly efficient at removing the organically available nitrogen that causes the eutrophication and low oxygen conditions affecting many of our coastal bays and estuaries (Piehler and Smyth 2011), so restoration targets could be set to achieve a desired level of nitrogen removal. The fish production potential of oyster reefs could be used to set oyster habitat restoration targets for the production of recreationally and commercially important species of fish and crabs species (Grabowski et al. 2012).

While there is still much to be learned in the field of oyster habitat restoration, the metrics and methodologies outlined in this handbook are intended to bring further clarity to measuring performance. The handbook emphasizes the need for monitoring the basic Universal Metrics for every project and sets the stage for implementing higher level goal-based metrics on selected projects. Widespread adoption of and reporting on data based on these performance metrics will enable much needed additional analysis and comparison of the economic and ecological return on restoration investments, and will continue to increase the efficacy of restoration in the future.

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Appendix I: Tables of Universal Metrics, Universal Environmental Variables, and Restoration Goal-based Metrics

Universal Metrics

Metric	Methods	Units	Frequency	Performance Criteria
Reef Areal Dimension: Project Footprint	Measure maximal aerial extent of reef using GPS, surveyor's measuring wheel or transect tape, or aerial imagery; subtidal use sonar, or SCUBA.	m^2	Pre-construction, within 3 months post-construction, and minimum 1-2 years post-construction; preferably 4-6 years. After events that could alter reef area.	None
Reef Areal Dimension: Reef Area	Measure area of each patch reef using GPS, surveyor's measuring wheel or transect tape, or aerial imagery; subtidal use sonar or depth finder with ground trothing, or SCUBA. Sum all patches to get total reef area.	m^2	Pre-construction, within 3 months post-construction, and minimum 1-2 years post-construction; preferably 4-6 years. After events that could alter reef area.	None
Reef Height	Measure using ruler, graduated rod and transit, or survey equipment; subtidal use sonar or depth finder.	cm	Pre-construction, within 3 months post-construction, and minimum 1-2 years post-construction; preferably 4-6 years. After events that could alter reef area.	Positive or neutral change
Live Oyster Density/ Recruitment Density	Utilize quadrats. Collect substrate to depth necessary to obtain all live oysters within quadrat, and enumerate number of live oysters, including recruits. If project involved the use of seed oysters, enumerate all seed oysters present in quadrat.	ind/m2	Immediately after deployment if using seed oysters. Otherwise, annually at the end of oyster growing season (will vary by region), 1-2 years at minimum; preferably 4-6 years.	Based on short- and long-term goals developed using available regional and project-type data, as well as current and/ or historical local/regional densities.
Size-Frequency Distribution	Measure shell height of at least 50 live oysters per oyster density sample.	mm (size), number or % per bin (size dist.)	Annually at the end of oyster growing season (will vary by region) in conjunction with oyster density sampling, at a minimum.	None

Universal Environmental Variables

Metric	Methods	Units	Frequency
Water temperature	Measure above substrate close to reef using *in situ* instrumentation, a thermometer, or other handheld instrumentation.	°C	Continuous (preferred), otherwise as often as possible
Salinity	Measure above substrate close to reef using *in situ* instrumentation, a refractometer, or other handheld instrumentation.	ppt or psu	Continuous (preferred), otherwise as often as possible
Dissolved oxygen (subtidal only)	Measure above substrate close to reef using *in situ* instrumentation or handheld instrumentation.	mg/L	Continuous (preferred), otherwise as often as possible

Restoration Goal-based Metrics

Metric	Methods	Units	Frequency	Performance Criteria
Goal: Brood Stock and Oyster Population Enhancement				
Nearby-Reef Density and Associated Size-Frequency Distribution	Follow methodologies outlined in Chapter 3 for determining reef aerial dimensions, density, and oyster size frequency.	ind/m2 (density), mm (size)	Annually at the end of oyster growing season (will vary by region), at a minimum. Sampling should occur after newly settled oyster spat can be confidently classified as recruits.	Trend of increasing oyster density on the nearby-reef site of interest, with an ultimate goal of having statistically greater oyster densities than those present pre-construction, and a density that is roughly equal to or exceeds that of a natural reference site.
Nearby-Reef Large Oyster Abundance	Follow methodologies outlined in Chapter 3 for determining reef aerial dimensions, density, and oyster size frequency. Determine the number of large (≥76 mm for *C. virginica;* ≥35 mm for *O. lurida*) oysters.	ind/m2	Annually at the end of oyster growing season (will vary by region), at a minimum. Note that it may take several years for oysters to grow to large sizes.	Trend of increasing densities of large oysters at the nearby-reef site of interest, with an ultimate goal of having statistically more large oysters than at the control site or pre-construction conditions, or a density that is roughly equal to or exceeds that of a natural reference site.

Restoration Goal-based Metrics (cont.)

Metric	Methods	Units	Frequency	Performance Criteria
Goal: Habitat Enhancement for Resident and Transient Species				
Density of Selected Species and/or Faunal Groups	Sample density of selected target species/faunal groups using quadrat samples (epifaunal sessile invertebrates), core samples (infaunal invertebrates), substrate baskets (small resident mobile fish and invertebrates), seines, lift nets, etc. (transient crustaceans and juvenile fish), gillnets (transient adult fish), or visual surveys (waterbirds).	ind/m2, wet weight (g/m^2),length (mm), CPUE where applicable. For waterbirds ind/ha	Annually or seasonally at a time that coincides with maximum abundances of target species	Trend of increasing densities of target species/ faunal groups as monitoring progresses, with an ultimate goal of statistically greater densities at the project site than at a control site or pre-construction conditions, or a density that is equal to or exceeds that of a natural reference site.
Goal: Enhancement of Adjacent Habitats				
Shoreline Loss/Gain	Establish permanent transects at project site and at control site. Survey shoreline profile from reef to 10m landward of shoreline edge.	m/year	Pre-construction, within 3 months post-construction, then annually thereafter; after major storm events.	A trend of decreasing shoreline loss, or shoreline gain, with an ultimate goal of having statistically less shoreline loss or greater shoreline gain than pre-construction conditions and at the control or reference site.

Restoration Goal-based Metrics (cont.)

Metric	Methods	Units	Frequency	Performance Criteria
Goal: Enhancement of Adjacent Habitats (cont.)				
Shoreline Profile/ Elevation Change	Establish permanent base stakes and transects on shoreline adjacent to reef and at control site and survey shoreline slope and profile using Total Station, Surveyor's transit and rod, or string and measuring tape.	Profile comparisons, m/year	Pre-construction, within 3 months post-construction, then annually thereafter; after major storm events.	A trend of decreasing slope and increasing mean elevation at the shoreline, with an ultimate goal of having statistically lower shoreline slope or increased mean shoreline elevation than pre-construction conditions and at the control, or a decreased 'step' at the water's edge.
Density and Percent Cover of Marsh/ Mangrove Plants	Quadrat measurements along transects	Live shoots/m^2, Percent cover	Annually at the end of the peak growing season	A trend of increasing mean plant density and mean percent coverage, with an ultimate goal of having statistically greater mean plant density and mean percent coverage than pre-construction conditions and at the control site, or a mean density and mean present coverage that is equal to or exceeds that of the natural reference site.

Restoration Goal-based Metrics (cont.)

Metric	Methods	Units	Frequency	Performance Criteria
Goal: Water Clarity Improvement				
Seston and/or Chlorophyll a Concentration	Take water samples at three locations on reef (midpoint, 0.5 m beyond reef edge up-current and down-current of reef) and test for seston (total particulates and organic content) and/or chlorophyll a concentration.	mg/L (total particulates), % (organic content), µg/L (chlorophyll a)	Quarterly in the year prior to construction and quarterly post-construction.	Trend of decreasing seston and/or cholorphyll a concentrations as monitoring progresses, with an ultimate goal of statistically lower concentrations at the project site than at a control site or pre-construction conditions, or concentrations that are equal to or less that of a natural reference site.
Light Penetration	Measure light penetration at points on reef using instrumentation (such as light intensity sensors), a sechi disc, or a transparency tube.	Will vary for instrumentation, cm (secchi disk and transparency tube).	Quarterly in the year prior to construction and quarterly post-construction.	Trend of increasing light penetration as monitoring progresses, with an ultimate goal of statistically greater penetration at the project site than at a control site or pre-construction conditions, or concentrations that are equal to or less that of a natural reference site.

Appendix II: Historic and Present Oyster Data by Region (adapted from zu Ermgassen et al. (2012a)

Gulf of Maine/Bay of Fundy	State	Historic Reef Area (ha)	Historic Extent Year	Historic Mean Density (ind/m²)	Historic Mean Length (mm)	Historic Biomass Total (kg x 10³)	Current Reef Area (ha)	Current Extent Year	Total Density (ind/m²)	Mean Length (mm)	Present Biomass Total (kg x 10³)	% Historic Extent Remaining	% Historic Biomass Remaining
Great Bay[1,2]	NH	~	~	~	~	~	87	2003-2009	287	55	136	~	~
Virginian													
Barnegat Bay[3-6]	NJ	5261	1889	18*	67*	880	0	0	NA	~	0	0	<1
Bogue Sound[4,5,7]	NC	666	1886-1887	18*	67*	73	~	~	~	~	~	~	~
Delaware Bay[3-5,8]	NJ/DE	25149	1889/1910	18	67*	4206	11471	2009	16	57	1179	46	28
Hudson River/Raritan Bay[4,5,8-11]	NY/NJ	1660	1886-1887	18*	67*	278	402	1983/2010	16*	57*	41	24	15
Ingram/Fleets Bays[12,13]	VA	~	~	~	~	~	83	1980	69	~	17	~	~
James River[4,14]	VA	4467	1910	19	67	535	2410	1998-2006	~	~	200	54	37
Long Island Sound[3-5]	NY/CT	13267	1886-1887	18*	67*	2219	~	~	~	~	~	~	~
Lynnhaven River[15]	VA	~	~	~	~	~	5	2004-2006	~	~	9	~	~
Narragansett Bay[16]	RI	~	~	~	~	~	0	2011	NA	~	0	0	<1
New Jersey Inland Bays[3-5,8,11]	NJ	1619	1889	18*	67*	271	46	1983	16*	57*	5	3	2
Pamlico Sound[4,5,7]	NC	3320	1886-1887	18*	67*	364	~	~	~	~	~	~	~

* = data proxied from nearest estuary

See zu Ermgassen et al. (2012a) for a complete list of references and a key to symbols

	State	Historic Reef Area (ha)	Historic Extent Year	Historic Mean Density (ind/m²)	Historic Mean Length (mm)	Historic Biomass Total (kg x 10³)	Current Reef Area (ha)	Current Extent Year	Total Density (ind/m²)	Mean Length (mm)	Present Biomass Total (kg x 10³)	% Historic Extent Remaining	% Historic Biomass Remaining
Virginian (cont.)													
Piankatank River/ Mobjack Bay[4,12,17,18]	VA	3223†	~1893	18*	67*	494	188	1980	41	~	16§	6	3
Southern Maryland Coastal Bays (Chincoteaque/ Sinepuxent)[4,19,20]	MD	672	1906-1912	18*	67*	37	0	1994	NA	~	0	0	<1
Tangier/Pocomoke Sounds[4,12,21]	MD/ VA	35536	1878	2	82*	657	7126	1980	12	44*	248	20	38
Virginia Eastern Shore[22]	VA	~	~	~	~	~	377	2002-2008	4	33	340	~	~
York River[4,12,17]	VA	698†	~1893	19*	67*	107	161	1980	15*	44*	10	23	10
Carolinian													
Altamaha River[23]	GA	81	1891	~	~	~	~	~	~	~	~	~	~
Broad River[24,25]	SC	51	1890	~	~	~	622	2006-2008	1229	43	968	~	~
Charleston Harbor[24,25]	SC	27	1890	~	~	~	57¶	2006-2008	1229	43	88	~	~
New River[7]	NC	126	1886-1887	~	~	~	~	~	~	~	~	~	~
Ossabaw Sound[23]	GA	71	1891	~	~	~	~	~	~	~	~	~	~
Savannah River[23,25]	GA/SC	62	1890/1891	~	~	~	~	~	~	~	~	~	~

* = data proxied from nearest estuary

See zu Ermgassen et al. (2012a) for a complete list of references and a key to symbols

	State	Historic Reef Area (ha)	Historic Extent Year	Historic Mean Density (ind/m²)	Historic Mean Length (mm)	Historic Biomass Total (kg x 10³)	Current Reef Area (ha)	Current Extent Year	Total Density (ind/m²)	Mean Length (mm)	Present Biomass Total (kg x 10³)	% Historic Extent Remaining	% Historic Biomass Remaining
Carolinian (cont.)													
St. Andrews, St. Simons Sounds[23]	GA	185	1891	~	~	~	~	~	~	~	~	~	~
St. Catherines/Sapelo Sounds[23]	GA	271	1891	~	~	~	~	~	~	~	~	~	~
St. Helena Sound[24,25]	SC	48	1890	~	~	~	401	2006-2008	1229	43	625	~	~
Stono/North Edisto Rivers[24,25]	SC	115	1890	~	~	~	199	2006-2008	1229	43	309	~	~
Floridian													
Biscayne Bay[26]	FL	~	~	~	~	~	0	2005-2006	NA	~	0	~	<1
Caloosahatchee River[27]	FL	~	~	~	~	~	1	2010	~	~	~	~	~
Charlotte Harbor[27]	FL	~	~	~	~	~	97	2010	~	~	~	~	~
Indian River[27,28]	FL	~	~	~	~	~	90	2007/2010	~	~	~	~	~
North Ten Thousand Islands[27]	FL	~	~	~	~	~	28	>2000	~	~	~	~	~
Rookery Bay[27]	FL	~	~	~	~	~	10	>2000	~	~	~	~	~
Sarasota Bay[27,28]	FL	~	~	~	~	~	23	2010	102ŧ*	35*	4	~	~
Tampa Bay[27,28]	FL	~	~	~	~	~	18	2010	102ŧ	35*	3	~	~

* = data proxied from nearest estuary

See zu Ermgassen et al. (2012a) for a complete list of references and a key to symbols

	State	Historic Reef Area (ha)	Historic Extent Year	Historic Mean Density (ind/m²)	Historic Mean Length (mm)	Historic Biomass Total (kg x 10³)	Current Reef Area (ha)	Current Extent Year	Total Density (ind/m²)	Mean Length (mm)	Present Biomass Total (kg x 10³)	% Historic Extent Remaining	% Historic Biomass Remaining
Northern Gulf of Mexico													
Apalachee Bay[27,28]	FL	~	~	~	~	~	1535	2010	158ŧ*	51*	1296	~	~
Apalachicola Bay[28-30]	FL	2695	1915	15	65	465	3491	1990-2010	158ŧ	51	2947	130	634
Aransas Bay[31-34]	TX	3885	1891	58*	81*	4249	482	2007	23	65	134	12	3
Atchafalaya/Vermilion Bays[35,36]	LA	8050	1905	31	71	3545	~	~	~	~	~	~	~
Barataria Bay[37]	LA	~	~	~	~	~	1028	2010	6	57	45	~	~
Breton/Chandeleur Sounds[35,37,38]	LA	5427	1898	31*	69*	1697	4525	2010	6	51	119	83	7
Calcasieu Lake[37]	LA	~	~	~	~	~	1581	2010	18	57	357	~	~
Choctawhatchee Bay[27,28]	FL	~	~	~	~	~	89	2010	68ŧ	57	45	~	~
Corpus Christi Bay[31-34ŧŧ]	TX	3367	1891	58*	81*	3683	290	2007	3	54	6	9	<1
Galveston Bay[31-33,39††]	TX	12950	1891	58*	81*	14165	10795	1995	8	62	1027	83	7
Laguna Madre (Upper and Lower, & Baffin Bay)[31-34††]	TX	777	1891	58*	81*	850	68	2007	1	53	0.5	9	<1
Matagorda Bay[31,33,40††]	TX	16679	1907/1915	58	81	18243	2229	2011	10	54	159	13	1
Mobile Bay[28,41-43]	AL	1151	1894	29	75	498	1045	1968/1995	ŧ47	51*	631	91	127
Pensacola Bay[28]	FL	~	~	~	~	~	3144	2010	68ŧ	57	1585	~	~
Sabine Lake[31-33,37§§]	TX/LA	259	1891	58*	81*	283	480	2010	41	69	301	185	106

* = data proxied from nearest estuary

See zu Ermgassen et al. (2012a) for a complete list of references and a key to symbols

	State	Historic Reef Area (ha)	Historic Extent Year	Historic Mean Density (ind/m²)	Historic Mean Length (mm)	Historic Biomass Total (kg x 10³)	Current Reef Area (ha)	Current Extent Year	Total Density (ind/m²)	Mean Length (mm)	Present Biomass Total (kg x 10³)	% Historic Extent Remaining	% Historic Biomass Remaining
Northern Gulf of Mexico (cont.)													
San Antonio Bay[31,33,34††]	TX	2590	1891	58*	81*	2833	2158	?	8	57	147	83	5
St. Andrew Bay[27,28]	FL	~	~	~	~	~	890	2010	230	44	660	~	~
Suwannee River[27,28]	FL	~	~	~	~	~	2138	2010	68ŧ	57	1078	~	~
Terrebonne/Timbalier Bays[37]	LA	~	~	~	~	~	1177	2010	17	57	80	~	~
West Mississippi Sound[35,37,38]	MS	3391	1898/ 1911	31	69	1061	6490	2010/ 1973	2	57	84	191	8
Southern California Bight													
Newport Bay[44]	CA	~	~	~	~	~	0	2011	NA	~	0	0	~
Northern California													
Elkhorn Slough[45-47]	CA	2	1929	116*	35*	0.5	0	2010	NA	~	0	0	<1
San Francisco Bay[46,48,49]	CA	3251	1893	116*	35*	801	0	2006	NA	~	0	0	<1
O.W.V.C.													
Humboldt Bay[34,46,50]	CA	137	1935	116*	35*	34	0	2007	NA	~	0	0	<1
Netarts Bay	OR	~	~	~	~	~	4	2011	33	34	0.007	~	~
Wilapa Bay[44,46,51,52]	WA	6225†††	~1850	116*	35*	1533	0	2011	NA	~	0	0	0
Yaquina Bay[46,53]	OR	27	1895	116	35	7	~	~	~	~	~	~	~

* = data proxied from nearest estuary

See zu Ermgassen et al. (2012a) for a complete list of references and a key to symbols

Appendix III: NOAA Data Record Sheets

The NOAA Restoration Center has developed monitoring data worksheets to record "universal" or basic parameters for several habitat types, including restored oyster habitat. The oyster monitoring worksheet reflects the monitoring recommendations in this handbook. In order to provide additional monitoring coordination across projects, we encourage the use of this worksheet whether or not your project receives NOAA funding. To access these worksheets see, http://www.habitat.noaa.gov/funding/applicantresources.html under "Monitoring Guidance."

Notes

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Notes

The Nature Conservancy

URI Graduate School of Oceanography
South Ferry Road
Narragansett, RI 02882