Nitrogen and Phosphorus Reduction Associated with Harvest of Hatchery-Produced Oysters and Reef Restoration: Assimilation and Enhanced Denitrification

PANEL RECOMMENDATIONS

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Oyster BMP Expert Panel Second Incremental Report

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Approved by Water Quality Goal Implementation Team (WQGIT), in coordination with the Sustainable Fisheries and Habitat GITs: ______________

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The novelty and complexity of oyster BMP development, as well as changes in Panel and Support personnel and COVID-related interruptions, at times have made the oyster BMP Panel process more difficult and lengthy than usual. The Panel is grateful for all the assistance we received and the contributions of data and data analysis by many individuals. In particular, the default denitrification rates were a product of both fieldwork and data analysis by Dr. Lisa Kellogg at the Virginia Institute of Marine Science. Her contributions of field and lab-derived data, data analysis, and report writing are infused throughout the report and the Panel is grateful for all her efforts.

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Contents

Acronyms in Document

BMP- Best Management Practice

CBF- Chesapeake Bay Foundation

CBP- Chesapeake Bay Program

DNF- Denitrification

DW- Dry Weight

EPA- Environmental Protection Agency

GIS- Geographic Information System

GIT- Goal Implementation Team

HPO- Hatchery Produced Oyster

LOH- Licensed Oyster Harvest

MD- Maryland

MD DNR – Maryland Department of Natural Resources

MDORIW- Maryland Oyster Restoration Interagency Workgroup

N- Nitrogen

NOAA- National Oceanographic and Atmospheric Administration

ORP- Oyster Recovery Partnership

ORR- Oyster Reef Restoration

P- Phosphorus

POA- Private Oyster Aquaculture

SH- Shell Height

SRP- Soluble Reactive Phosphorus

TMDL- Total Maximum Daily Load

TNC- The Nature Conservancy

UMCES- University of Maryland Center for Environmental Science

VIMS- Virginia Institute of Marine Science

WQGIT- Water Quality Goal Implementation Team

WTWG- Watershed Technical Workgroup

Key Definitions

Alternate substrates: All materials suitable for oyster settlement and survival not composed solely of oyster shells

Ancillary benefits: Potential positive effects of the oyster practice beyond its impacts on nitrogen and phosphorus.

Assimilation: Incorporation of nitrogen and phosphorus from digested food into oyster tissues and shells.

Batch incubation: Benthic time course incubation without continuous exchange of water

Biodeposit: Organic matter and associated inorganic materials (e.g., feces and pseudofeces from oysters) deposited on the bottom (i.e., sediment surface).

Biodeposition: The process through which biodeposits are deposited on the bottom.

BMP Site: The location in which enhancement activities occur and which is potentially eligible for nitrogen and phosphorus reduction credit if all qualifying conditions are met. The BMP site is an area.

BMP site area: Area (e.g., acres) of the actual enhancement activities at the BMP site. The BMP site area may be smaller than the BMP site.

Burial: The process in which nutrients are trapped in the bottom sediment for long timescales (i.e., below the active zone where active decomposition of organic and inorganic material and reworking of sediments occurs).

Denitrification: The process that reduces nitrates or nitrites to nitrogen gas, commonly by bacteria in the bottom sediment. Nitrogen gas ultimately escapes into the atmosphere and is a form of nitrogen that is not readily available for phytoplankton growth. For the purposes of this document, we include all processes that produce nitrogen gas (N2) as denitrification.

Diploid oyster: Wild or hatchery-produced oysters containing two complete sets of chromosomes, one from each parent and capable of sexual reproduction.

Engineered structure: Suitable substrate for oyster larvae that has been manufactured (e.g., ReefBalls™, Oyster Castles®).

Flow-through incubation: An approach to measuring biogeochemical fluxes that assesses the difference in concentration of analytes of interest (e.g., oxygen, nitrate, nitrite, ammonium, di-nitrogen gas) between water flowing into and out of an incubation chamber and uses this information along with the flow rate to calculate a flux.

Flux: Change in concentration of analyte per unit time, generally expressed on a areal basis.

Hatchery-produced oyster: Diploid or triploid oysters that originated from oyster larvae propagated outside their natural environment.

Incubation: A method for measuring biogeochemical fluxes that involves sealing a sample of the bottom component of interest (e.g. sediments, oyster reef) in a chamber and collecting samples from the overlying water column either as a time series of measurements.

Incubation chamber: An enclosure containing a sample of the bottom component of interest (e.g. sediments, oyster reef) used for measuring biogeochemical fluxes.

Large substrate: Suitable substrate characterized by (a) <90% of the material by volume has a maximum diameter of ≤12 inches (304.8 mm) or (b) a uniform, regular structure when deployed.

Licensed oyster harvest: Oyster harvest from a State-managed fishery area by individuals holding the proper licenses.

Live oyster shell biomass: The total dry weight of shell from living oysters.

Live oyster tissue biomass: The total dry weight of soft tissue from living oysters.

No-harvest area: Designated areas where oysters are permanently protected from harvest (e.g., oyster sanctuaries or areas closed due to water quality concerns).

Oyster associated processes/ Oyster processes: Ecological, biological, chemical, or physical mechanisms that occur within individual oysters or on oyster reefs.

Oyster hatchery: Facility that produces diploid and/or triploid oyster larvae outside their natural environment for research, restoration, educational, and/or commercial uses.

Oyster practice: Oyster management approaches that can be implemented as best management practices to assess progress of water quality goals established by the Chesapeake Bay Total Maximum Daily Load.

Oyster protocol: Oyster-associated processes that reduce nitrogen, phosphorus, and/or suspended sediment as defined by this document

Oyster reef restoration: Activities aimed at increasing oyster populations/biomass in no-harvest areas.

Oyster shell height: The longest distance (parallel to the long axis) between the hinge and lip of the oyster.

Oyster spat: Typically refers to oysters that have settled (attached) onto substrate and are less than one year old.

Ploidy: The number of sets of chromosomes in a cell affecting reproductive capabilities (e.g., infertile triploid oysters have three sets of chromosomes, while fertile diploid oysters have two complete sets).

Practice-protocol combination: Individual reduction effectiveness crediting protocols that can quantify the reduction of nitrogen, phosphorus, and/or suspended sediment for different oyster practices.

Private oyster aquaculture: Growing and harvesting diploid or triploid oysters in areas designated for oyster aquaculture where public harvest is not allowed (e.g., State-permitted oyster aquaculture leases to private oyster aquaculturists).

Quantile regression: Type of regression analysis that estimates the conditional median or other quantiles of the response variable.

Recruitment: The number of individuals surviving to a certain size, age, or life stage (e.g., spat, reproductive maturity, etc.).

Seston: All organic and inorganic, living and non-living material suspended in the water column.

Small substrate: Suitable substrate that is characterized by (a) ≥90% of the material by volume has a maximum diameter of ≤12 inches (304.8 mm) and (b) a non-uniform or irregular structure when deployed.

Spat-on-shell planting: Oyster larvae that have settled (attached) onto shell and have been placed on the bottom.

Substrate addition: The act of placing suitable substrate (e.g., shell, stone, etc.) on the sediment surface to enhance the potential recruitment of wild oyster larvae or to serve as a base for planting spat on shell.

Sufficient Science: In the Panel's best professional judgment, data of sufficient quality and scope exist and can be used to generate a reasonably constrained estimate of the reduction associated with a particular oyster practice.

Suitable substrate: Material on which oysters can settle and survive to adulthood. Can include, but are not limited to: non-oyster shell, fossilized shell, limestone, granite, crushed concrete, engineered structures, etc.

Suspended sediment: Very fine soil particles that remain in suspension in water for a considerable period of time.

Triploid oyster: Hatchery-produced oysters containing three sets of chromosomes and generally incapable of sexual reproduction.

Unintended Consequence: Potential negative effects resulting from the oyster practice.

Verifiable: In the Panel's best professional judgment, a practical method exists, or could be created, to track reduction effectiveness if the BMP is implemented.

Wild oyster: Diploid oysters produced in their natural environment without human involvement.

1.0 Introduction

In 2010, the U.S. Environmental Protection Agency (EPA) established the Chesapeake Bay Total Maximum Daily Load (TMDL), a set of accountability measures implemented towards improving water quality in Chesapeake Bay (EPA 2010). Since the TMDL was established, the Chesapeake Bay Program (CBP) has convened expert panels to assist in identifying best management practices (BMPs) and quantifying the degree to which those practices reduce inputs of nutrients (i.e., nitrogen and phosphorus) and suspended sediments to Chesapeake Bay. Once BMPs are established, their nutrient and suspended sediment reduction effectiveness is credited and progress towards achieving water quality goals is tracked using the CBP Partnership's model framework (CBP 2022).

The Oyster Best Management Practice Expert Panel (hereafter, "Panel") was convened in September 2015 because of interest in evaluating and implementing ***oyster practices*** as best management practices. Oysters can contribute to the reduction of nutrients (i.e., nitrogen and phosphorus) and suspended sediment by filtering organic and inorganic particulates, such as algae and suspended sediment, from the water column. The nitrogen and phosphorus from these particulates can be (1) assimilated in the oyster tissue or shell or (2) incorporated into oyster biodeposits (i.e., feces, pseudofeces) deposited on the seafloor along with ingested sediment where they can be denitrified or buried (Kellogg et al. 2013, 2014, Grizzle et al. 2018). Relative to other benthic environments, oyster reefs are hotspots for denitrification (Caffrey et al. 2016, Arfken et al. 2017, Jackson et al. 2018). Denitrification is the final step in a set of transformations that converts organic nitrogen to di-nitrogen gas (N_2), a form of nitrogen that cannot be used for growth by most phytoplankton and algae. Burial occurs when oyster waste is deposited onto the sediment surface and becomes trapped in the benthic sediment for long timescales (i.e., below the active zone where decomposition occurs) making the nitrogen, phosphorus, and sediment it contains unavailable to the water column (Newell et al. 2005).

The Panel was charged with developing (1) a decision framework to determine the nutrient and suspended sediment reduction effectiveness of oyster practices as BMPs for application in the CBP Partnership's model framework and (2) recommendations on the nitrogen, phosphorus, and suspended sediment reduction effectiveness of oyster practices based on existing science. As part of this process, the Panel worked with the EPA to verify that oyster practices could be applied as in-water BMPs under the Clean Water Act. The EPA agreed that CBP Partnership-approved oyster BMPs can qualify for nutrient and sediment pollutant reductions under the Clean Water Act (Appendix C).

The Panel's first set of recommendations were approved by the Chesapeake Bay Program (CBP) Partnership's Water Quality Goal Implementation Team (WQGIT), in coordination with the Habitat Goal Implementation Team (Habitat GIT) and Sustainable Fisheries Goal Implementation Team (Fisheries GIT), on December 19, 2016 and can be found in their first report (Reichert-Nguyen et al. 2016). This first report includes the Panel's decision framework and recommended estimates for nitrogen and phosphorus assimilated in tissue of oysters harvested from private oyster aquaculture practices. The approved decision framework allows for the incremental determination, approval, and implementation of individual reduction effectiveness crediting ***protocols*** that can quantify the reduction of nitrogen, phosphorus, and/or suspended sediment for different oyster practice categories (hereafter, ***practice-protocol combination***).

Overall, the Panel identified 96 potential practice-protocol combinations in which oysters could reduce nutrients and suspended sediments in Chesapeake Bay (Table 1.1). A total of six practice-protocol combinations were approved of the 10 reviewed in the first report (Reichert-Nguyen et al. 2016). In this report, the Panel reviewed 45 total practice-protocol combinations and is endorsing 12 combinations for approval by the CBP Partnership. Oyster biomass is an important parameter in developing the reduction effectiveness estimates. The Panel's decision concerning which oyster practices should undergo BMP consideration was based on whether the practices include an enhancement activity that could result in the overall production of new oysters, and consequently, oyster biomass.

The details and rationale for the practice-protocol combinations are described in the Panel's first report (Reichert-Nguyen et al. 2016). The practice-protocol combinations are based on twelve oyster practices and eight reduction effectiveness protocols. The oyster practices were determined after considering the oyster's fate (i.e., removed or remains in the waterbody), fisheries management approach (i.e., private oyster aquaculture, licensed oyster harvest [previously called public fishery], and oyster reef restoration), oyster type (i.e., diploid or triploid hatchery-produced oysters or wild diploid oysters), and activity/culture method (i.e., oysters grown off or on the bottom, transplanted oysters, substrate addition, or no activity). The Panel also added information on who has access to the oysters in connection with ownership (i.e., private oyster aquaculture leaseholders who grow and harvest oysters from leased State bottom, license-holders who fish on public harvest areas, state resource management agencies who plant and monitor oysters/substrate in no harvest areas) since it can influence the development of implementation and verification procedures (Table 1.2; further described in Chapter 5.0).

The Panel opted to change the overarching fisheries management approach title of "public fishery" to "licensed oyster harvest" to better reflect the Panel's intent to only put forward BMP recommendations for practices where activities occur that enhance oyster populations. ***Licensed oyster harvest*** specifically refers to oyster harvest from a State-managed fishery area by individuals holding the proper harvest licenses; BMP crediting occurs only in areas that are supplemented with hatchery produced oysters (e.g., spat on shell or single oysters). In addition to changes to the public fishery titles, the titles for the oyster reef restoration practices and descriptions have been slightly modified from the first report for clarity (Table 1.2).

The reduction effectiveness of the oyster practices endorsed for BMP use is based on biological processes that can reduce nitrogen, phosphorus, and suspended sediment (Figure 1.1). These were divided into the following eight reduction crediting protocols:

Protocol 1. Nitrogen assimilation in oyster tissue
Protocol 2. Nitrogen assimilation in oyster shell
Protocol 3. Enhanced denitrification associated with oysters
Protocol 4. Phosphorus assimilation in oyster tissue
Protocol 5. Phosphorus assimilation in oyster shell
Protocol 6. Suspended sediment reduction associated with oysters
Protocol 7. Enhanced nitrogen burial associated with oysters
Protocol 8. Enhanced phosphorus burial associated with oysters

Protocols 1, 2, 4, & 5 involve oysters consuming and assimilating nitrogen and phosphorus from filtered organic particles (e.g., algae, phytoplankton) into their tissue and shell. The filtration of particles out of the water column can also lead to enhanced burial of suspended sediments (**Protocol 6**) containing nitrogen and phosphorus (**Protocol 7 & 8**).

Protocol 3 involves the biogeochemical process of denitrification, which is dependent on the microbial communities living within the reef and surrounding sediment. While oysters do not perform denitrification directly, they can enhance the process by depositing excess organic matter to the sediment or by increasing the amount of habitat per-unit-area for nitrifying and denitrifying organisms.

Differences among the reviewed oyster practices (Table 1.2) affect the determination and verification of the nutrient reduction effectiveness. For assimilation (**Protocol 1, 2, 4, & 5**), harvest (via private oyster aquaculture or licensed oyster harvest) continually removes nitrogen and phosphorus assimilated into the tissue and shell of harvested oysters as long as new oysters are added and harvest occurs. In contrast, oysters that remain in the water (via oyster reef restoration in no-harvest areas) vary in the amount of nitrogen and phosphorus sequestered as the reef's standing stock biomass varies. Requirements could also differ between practices involving harvest or restoration for denitrification (**Protocol 3**), due to differences in the fate of oyster biodeposits and reef community structure on harvested vs. un-harvested reefs. Additional research is required to determine how oyster harvest, movement, and replacement impact denitrification in these different settings.

Differences in the context of oyster harvest can also affect the determination and verification of reduction effectiveness. Private oyster aquaculture occurs when the State permits water column or bottom leases to private entities who then grow oysters in those areas. In accordance with Virginia and Maryland State policies, private oyster aquaculture leases are typically permitted in areas with few to no pre-existing wild oysters (Code of Maryland Regulations COMAR 08.02.04.17; Code of Virginia §28.2-603). Licensed oyster harvest can occur on public, State-designated areas by any individual holding the proper licenses and harvesting in accordance with established rules and regulations. Licensed oyster harvest can occur in areas where there are existing or historically productive oyster bars. Compared to private oyster aquaculture practices, licensed oyster harvest practices require additional assessment of baseline conditions to verify whether nutrient reduction occurs.

Table 1.1. The Panel's review progress for the 96 oyster practice-protocol combinations. "Approved" indicates combinations that have been approved for BMP use by the CBP Partnership. "Complete" indicates combinations that are endorsed by the Panel in this report and ready for BMP use pending approval. "Research Gap" and "Policy Issue" identify combinations that have been endorsed by the Panel for BMP use but lack certain information to complete the recommendations. "Not Endorsed" are combinations that the Panel agreed should not undergo BMP consideration since they do not result in enhanced oyster populations. Practice-protocol combinations highlighted in dark gray were discussed in the Panel's first report (Reichert-Nguyen et al. 2016). Practice-protocol combinations highlighted in light gray are discussed in this report.

	Private Oyster Aquaculture					**Licensed Oyster Harvest**				**Oyster Reef Restoration**		
Oyster Practice Category x Crediting Protocol	A. Off-bottom private aquaculture using hatchery-produced oysters	B. On-bottom private aquaculture using hatchery-produced oysters	C. On-bottom private aquaculture using transplanted wild oysters	D. On-bottom private aquaculture using substrate addition	E. Private oyster aquaculture with no activity	F. Licensed harvest using hatchery-produced oysters	G. Licensed harvest using transplanted wild oysters	H. Licensed harvest using substrate addition	I. Licensed harvest with no activity	J. Reef restoration using hatchery-produced oysters	K. Reef restoration using substrate addition	L. Reef restoration using no harvest area designation only
1. Nitrogen assimilation in tissue	**1st Approved**	**1st Approved**	1st Not Endorsed	**1st Approved**	1st Not Endorsed	**2nd Complete**	2nd Not Endorsed	Later	2nd Not Endorsed	**2nd Complete**	**2nd Complete**	2nd Policy Issue
2. Nitrogen assimilation in shell	2nd Research Gap	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	Later	2nd Not Endorsed	**2nd Complete**	**2nd Complete**	2nd Policy Issue
3. Enhanced denitrification	2nd Research Gap	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	Later	2nd Not Endorsed	**2nd Complete**	**2nd Complete**	2nd Policy Issue
4. Phosphorus assimilation in tissue	**1st Approved**	**1st Approved**	1st Not Endorsed	**1st Approved**	1st Not Endorsed	**2nd Complete**	2nd Not Endorsed	Later	2nd Not Endorsed	**2nd Complete**	**2nd Complete**	2nd Policy Issue
5. Phosphorus assimilation in shell	2nd Research Gap	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	2nd Research Gap	2nd Not Endorsed	Later	2nd Not Endorsed	**2nd Complete**	**2nd Complete**	2nd Policy Issue
6. Suspended sediment reduction	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later
7. Enhanced nitrogen burial	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later
8. Enhanced phosphorus burial	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later	Later

Table 1.2. Chesapeake Bay oyster practice categories identified by the Panel. HPO = Hatchery-produced oysters, POA = Private oyster aquaculture, LOH = Licensed oyster harvest, ORR = Oyster reef restoration.

Oyster Fate	Oysters removed (harvested) from Bay									Oysters remain in Bay		
Fisheries Management Approach	Oyster cultivation									Conservation		
	Private oyster aquaculture (POA)					Licensed oyster harvest (LOH)				Oyster reef restoration (ORR)		
Description	Oyster harvest from State-issued water column and bottom leases					Oyster harvest from State-managed fishing areas				No harvest allowed		
Access to Oysters	Lease-holder					License-holder				State resource management agency		
Oyster Type	Hatchery-produced oysters (HPO)		Wild oysters			HPO	Wild oysters			HPO	Wild oysters	
Activity	HPO grown off the bottom using gear	HPO grown on the bottom using no gear	Moving wild oysters from one location to another	Addition of substrate to enhance recruitment of wild oyster larvae	None	Addition of HPO	Moving wild oysters from one location to another	Addition of substrate to enhance recruitment of wild oyster larvae	None	Designate no-harvest area followed by addition of HPO	Designate no-harvest area followed by addition of substrate	Designate no-harvest area with no additional activity
Oyster Practice	**A. Off-bottom POA using HPO**	**B. On-bottom POA using HPO**	C. On-bottom POA using transplanted wild oysters	**D. On-bottom POA using substrate addition**	E. POA with no activity	**F. LOH using HPO**	G. LOH using transplanted wild oysters	H. LOH using substrate addition	I. LOH with no activity	**J. ORR using HPO**	**K. ORR using substrate addition**	L. ORR using no-harvest area designation only
Recommended for BMP?	**Yes**	**Yes**	No	**Yes**	No	**Yes**	No	Later	No	**Yes**	**Yes**	Later

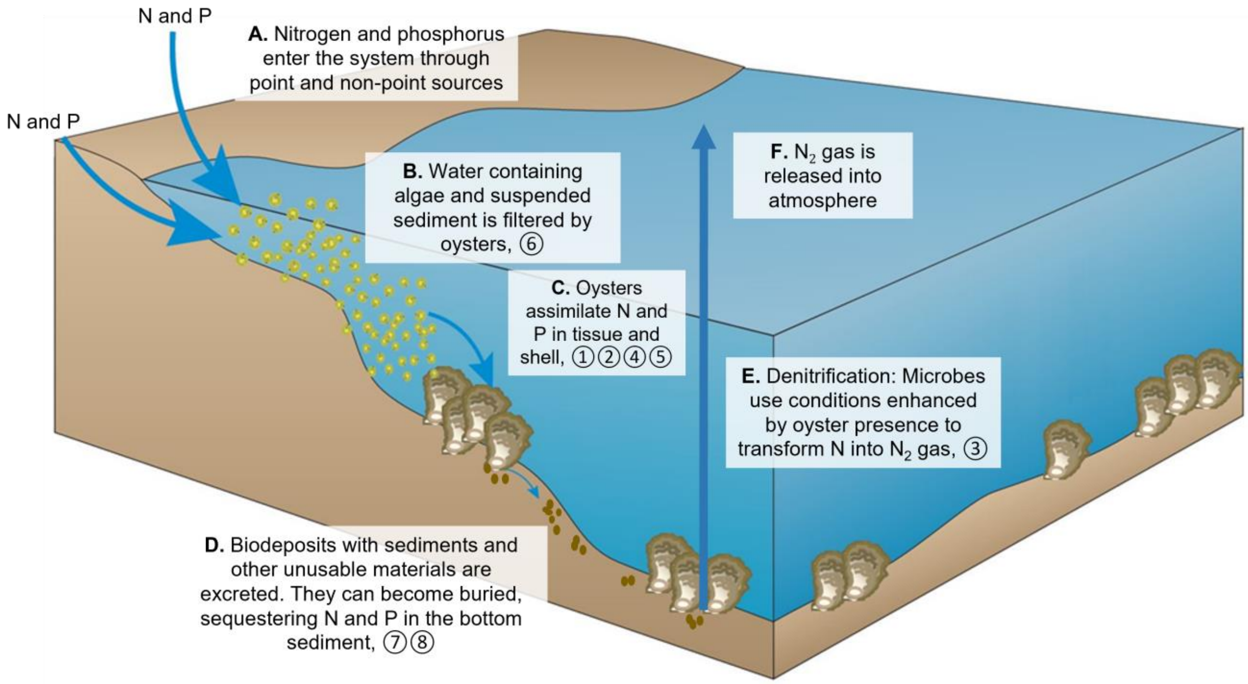


Figure 1.1. Oyster-associated processes that reduce nitrogen (N) and phosphorus (P) and suspended sediment. The numbers in circles correspond with the reduction effectiveness crediting protocols identified in Table 1.1.

This report includes recommendations for assessing and implementing nitrogen and phosphorus reduction effectiveness BMPs for twelve oyster practice-protocol combinations (Table 1.1). The recommended practice-protocol combinations are as follows:

- **Practice F**. Licensed oyster harvest using hatchery-produced oysters
  - **Protocol 1**. Nitrogen assimilation in oyster tissue
  - **Protocol 4**. Phosphorus assimilation in oyster tissue
- **Practice J**. Oyster reef restoration using hatchery-produced oysters
  - **Protocol 1**. Nitrogen assimilation in oyster tissue
  - **Protocol 2.** Nitrogen assimilation in oyster shell
  - **Protocol 3.** Enhanced denitrification associated with oysters
  - **Protocol 4.** Phosphorus assimilation in oyster tissue
  - **Protocol 5.** Phosphorus assimilation in oyster shell
- **Practice K.** Oyster reef restoration using substrate addition
  - **Protocol 1.** Nitrogen assimilation in oyster tissue
  - **Protocol 2.** Nitrogen assimilation in oyster shell
  - **Protocol 3.** Enhanced denitrification associated with oysters
  - **Protocol 4.** Phosphorus assimilation in oyster tissue
  - **Protocol 5.** Phosphorus assimilation in oyster shell

The Panel is asking the WQGIT, in coordination with the CBP Partnership and Fisheries and Habitat GITs, to review and approve the recommendations found in this second incremental report for BMP implementation.

This report covers two unique fishery management approaches and two distinct nutrient-reducing, oyster-associated processes. The Panel supports the approval of these separately, if needed, and is requesting that issues preventing approval of one set of recommendations not affect the approval of another. Once approved, the Panel expects that the practice-protocol combinations will be BMPs available to the implementing programs to help meet their TMDL goals. The incremental approach reviewed in this report is in line with the approved *Oyster BMP Nutrient and Suspended Sediment Reduction Effectiveness Determination Decision Framework* from the Panel's first report (Reichert-Nguyen et al. 2016). This framework allows for a practical and adaptive strategy to implement oyster practices as BMPs that accounts for the variety of practices and processes in which oysters can reduce nutrients and suspended sediment.

The Panel provides informational recommendations for practice-protocol combinations where there was not enough information to develop complete recommendations in Appendix H & I. This information can be used later to formulate complete recommendations once research gaps are addressed.

1.1 Report Structure

The report structure is as follows:

Chapter 2.0 provides a summary of the Panel's recommendations for oyster practice-protocol combinations for BMP approval.
Chapter 3.0 provides a summary of the Panel's membership and charge.
Chapter 4.0 describes the Panel's *Oyster BMP Nutrient and Suspended Sediment Reduction Effectiveness Determination Decision Framework* (Reichert-Nguyen et al. 2016). Once a reduction effectiveness crediting protocol is approved for a given oyster practice, it should be implemented in this framework.
Chapter 5.0 defines and provides examples of the oyster practices evaluated for BMP consideration in this report (Table 1.2).

The Panel's complete recommendations for oyster practice-protocol combinations involved extensive review of the scientific literature and addresses all requested items in the CBP Partnership's BMP Review Protocol (CBP 2015). The complete recommendations are described in the main body of this report and in the corresponding appendices for each chapter:

Chapter 6.0 describes the Panel's recommendations on the reduction effectiveness of nitrogen and phosphorus assimilated in tissue of hatchery-produced oysters removed by licensed oyster harvest.

- **Practice F.** Licensed oyster harvest using hatchery-produced oysters
    - Protocol 1. Nitrogen assimilation in oyster tissue
    - Protocol 4. Phosphorus assimilation in oyster tissue

Chapter 7.0 describes the Panel's recommendations on the reduction effectiveness of nitrogen and phosphorus assimilated in live oysters on restored oyster reefs.

- **Practices J & K.** Oyster reef restoration using hatchery-produced oysters and substrate addition
    - Protocol 1**.** Nitrogen assimilation in oyster tissue
    - Protocol 2. Nitrogen assimilation in oyster shell
    - Protocol 4. Phosphorus assimilation in oyster tissue
    - Protocol 5. Phosphorus assimilation in oyster shell

Chapter 8.0 describes the Panel's recommendations on the reduction effectiveness of nitrogen removed by enhanced denitrification associated with oysters on restored oyster reefs.

- **Practice J & K.** Oyster reef restoration using hatchery-produced oysters and substrate addition
    - Protocol 3. Enhanced denitrification associated with oysters

Chapter 9.0 describes the ancillary benefits of oyster reef restoration.

Chapter 10.0 provides a summary of the recommendations and progress described in detail in this report.

Chapter 11.0 provides a list of documents and sources referenced in this report.

Appendices associated with this report provide supplemental information on the Panel's approach to estimating reduction effectiveness, informational recommendations for future studies or adjusting the recommendations provided in this report, Panel activities, and technical reporting requirements for integrating the oyster BMPs into the CBP's TMDL model.

Appendix A provides a summary of the Panel's activities leading to the recommendations found in this report.

Appendix B describes this report's conformity with the CBP Partnership's BMP Review Protocol (CBP 2015).

Appendix C contains the legal opinion from the U.S. Environmental Protection Agency (EPA) concluding that the removal of pollutants from the water column by in-water BMPs is legal through the Clean Water Act.

Appendix D describes analyses used to develop the default reduction estimates for licensed oyster harvest (*Supplemental to Chapter 6*).

Appendix E contains supporting information and analyses used to develop the reduction estimates for oyster restoration practices (*Supplemental to Chapters 7 and 8*).

Appendix F provides the Panel's criteria while quantifying denitrification rates on restored oyster reefs (*Supplemental to Chapter 8*).

Appendix G provides the full denitrification lookup table created by the Panel to estimate annual denitrification enhancement in oyster tissue (*Supplemental to Chapter 8*).

Appendix H and I include informational recommendations for practice-protocol combinations and other considerations for which approval is not being sought at this time due to knowledge gaps.

- *Appendix H* discusses information relevant to future evaluations of nitrogen and phosphorus assimilated in oyster shell (**Protocols 2 and 5**) for harvested oysters (**Practice A, B, D, F**).
- *Appendix H* also includes a preliminary analysis that could inform updates to the estimates of nitrogen and phosphorus assimilated in tissue (**Protocols 1 and 4**) of harvested diploid oysters. These analyses are based on new data that became available after December 2016 for diploid oysters grown in gear.
- *Appendix I* discusses information relevant to future evaluations of enhanced denitrification (**Protocol 3**) for harvested oysters (**Practice A, B, D, F**) and for oyster restoration using large substrates.

Appendix J describes how oysters influence Chesapeake Bay water quality in the CBP Modeling framework.

Appendix K describes the Watershed Technical Workgroup's requirements for reporting and implementing the recommended oyster BMPs in the Phase 6 Watershed Model. [Still in development]

Appendix L contains the minutes from all Panel meetings throughout the oyster BMP recommendation process.

1.2 Policy Issues

There were two policy issues identified by the Panel or CBP membership when reviewing oyster practices that could be recommended for BMP approval.

The Panel identified a policy issue for the CBP Management Board to review concerning the oyster reef restoration **Practice L** (oyster reef restoration using no-harvest area designation only). This practice designates an area where oysters are not allowed to be harvested but where there are no additional oyster plantings or addition of substrate to enhance oyster populations. Most of the Panel members agreed to endorse this practice for BMP consideration. One Panelist was not in support of endorsing this practice. The concern was that this practice was outside the scope of traditional land-based BMPs since no physical activity is occurring. Since the rationale for not endorsing this practice was based on a policy issue, the Panel recommends that the CBP Partnership Management Board review the issue and decide on whether this practice can be applied as a BMP. From a scientific perspective, the activity of removing harvest pressure on a natural oyster reef by designating it as a no-harvest area could result in increased oyster biomass, which would reduce nutrient concentrations via enhanced denitrification and sequestration in live oysters. The Panel agreed that the process for estimating nutrient reduction via enhanced denitrification and nitrogen and phosphorus assimilation recommended for other oyster restoration practices could also be applied for this practice if approved for BMP consideration.

A CBP representative on the Panel raised an additional policy concern related to the **Practice F** (licensed oyster harvest using hatchery-produced oysters). Since this practice applies to a public fishery, it is unclear if it would set a precedent that may have unintended management consequences for other fisheries. There were no issues concerning the technical merit of the Panel's recommendations for this practice. Therefore, further discussion within the CBP Partnership may be needed to evaluate policy implications.

The WQGIT determined that policy issues raised by the Panel and stakeholders were outside the purview of the Panel's charge and should be evaluated by the CBP Partnership Management Board. The CBP Partnership Management Board is working on resolving these policy issues as the Oyster BMP Expert Panel refines recommendations on the nutrient reduction effectiveness based on existing science. It is the Panel's understanding that unresolved policy issues will not prevent a decision on the Panel's report since the Panel's recommendations focus on the scientific and technical aspects concerning the nutrient reduction effectiveness of oyster practices.

2.0 Summary of Recommendations Covered in this Report

This report covers the Panel's recommendations for 45 of the 96 practice-protocol combinations identified as possible oyster BMPs (Table 1.1). Of the 45 combinations reviewed in this iteration, the Panel is putting forward complete recommendations for 12 combinations for BMP approval (labeled as "Complete" in Table 1.1). Two of these combinations fall under the "licensed oyster harvest" fishery management approach and involve nitrogen and phosphorus assimilation in tissue of harvested oysters (Chapter 6). Ten of these combinations fall under the "oyster reef restoration" fishery management approach and involve either nitrogen and phosphorus assimilation in tissue and shell (8 combinations; Chapter 7) or nitrogen removal from enhanced denitrification (2 combinations; Chapter 8).

Of the remaining 33 practice-protocol combinations not recommended for BMP approval at this time, 16 were not endorsed, 12 were identified as containing research gaps and therefore the reduction effectiveness cannot be estimated at this time, and 5 were not agreed upon by the Panel for possible policy issues (Table 1.1). Practices that involved transplanting wild oysters from one location to another (**Practice C & G**) or oyster harvest from areas receiving no enhancement activity (**Practice E & I**) were not endorsed (following Reichert-Nguyen et al. 2016).

Research gaps existed for the following practice-protocol combinations:

- **Practice F**. Licensed oyster harvest using hatchery-produced oysters
    - **Protocol 2 & 5**. Nitrogen and phosphorus assimilation in oyster shell
    - **Protocol 3**. Enhanced denitrification associated with oysters
- **Practice A, B, & D**. Private oyster aquaculture
    - **Protocol 2 & 5**. Nitrogen and phosphorus assimilation in oyster shell
    - **Protocol 3**. Enhanced denitrification associated with oysters

A policy issue was identified for **Practice L** (oyster reef restoration using no-harvest area designation) since no physical activity occurs to enhance oyster biomass (described in Subchapter 1.2). If the CBP Management Board decides that this practice can be considered a BMP, then the Panel's complete recommendations in Chapter 7 (restoration-assimilation) and 8 (restoration-denitrification) could also be applied to this practice.

To date, the Panel has reviewed a total of 55 practice-protocol combinations (10 from the first report; 45 in this report) of the 96 that were identified as possible oyster BMPs. Of the remaining 41 combinations, 36 involve the protocols on nitrogen and phosphorus reduction from enhanced burial (**Protocol 6 & 7**) and suspended sediment reduction associated with oysters (**Protocol 8**). The Panel felt it best to evaluate these in a future report since limited data are available to conduct a thorough review of these mechanisms. The Panel also decided to wait on evaluating the remaining 5 combinations because of conflicting information associated with Protocols 1-5 for **Practice H** (licensed oyster harvest using substrate addition; see Chapter 5.0). The Panel recommends waiting to evaluate these practice-protocol combinations until more relevant data become available.

2.1 Informational Recommendations to Support Future Evaluations

The Panel is not looking for approval on the informational recommendations included in this report (Appendix H & I). These Appendices outline the following considerations to update existing and/or to support future evaluation of additional oyster practices for BMP implementation.

- Appendix H – New data (after December 2016) can be used to update the estimates for percent nitrogen and phosphorus assimilated in oyster tissue (**Protocol 1 & 4**) for harvested diploid oysters grown in gear (**Practice A & B**).
- Appendix H – Recommendations for creating a decision framework to determine the reduction effectiveness estimates for nitrogen and phosphorus assimilated in shell (**Protocol 2 & 5**) of harvested oysters (**Practice A, B, D & F**).
- Appendix I – Existing data and knowledge gaps that need to be addressed to evaluate site-specific estimates of enhanced denitrification (**Protocol 3**) for practices where oysters are harvested (**Practice A, B, D, & F**) and for oysters growing on large substrates, such as engineered structures.

3.0 Expert Panel Membership, Charge, and Effort

3.1 Panel Membership

The Panel includes oyster scientists and practitioners from the US East Coast region. Panel members represent academia, non-profit organizations, and county, state, and federal agencies. Panel members are experts in oyster biology/ecology, water quality, biogeochemical processes, fishery management, and/or oyster practice implementation.

Panel membership changed slightly after November 2016. Matt Johnston assisted Jeff Sweeny as the Panel's Watershed Technical Workgroup (WTWG) representative from November 2016 to July 2018 and Ralph Spagnolo took over for Ed Ambrogio after his retirement from the EPA around March 2016. Ralph has since retired. Karen Hudson stepped down from the Panel after completion of the first report in December 2016. Frank Marenghi stepped in for Lynn Fegley around June 2018. Lisa Kellogg stepped down from the Panel in August 2022. Table 3.1 shows the current and past Panel membership.

Table 3.1. Current and past Oyster BMP Expert Panel membership.

Panelists	Status	Affiliation	Expertise
Jeff Cornwell (Panel Chair)	Active	U. of Maryland Center for Environmental Science	Oyster filter-feeding; nutrient cycling dynamics; modeling; sediment biogeochemistry; oyster ecology; population dynamics
Suzanne Bricker	Active	NOAA, National Centers for Coastal Ocean Science	Nutrient-related water quality research; oyster and nutrient cycling modeling
Lynn Fegley	Inactive	Maryland Department of Natural Resources	Fisheries management
Karen Hudson	Inactive	Virginia Institute of Marine Science	Shellfish aquaculture
Lisa Kellogg	Inactive	Virginia Institute of Marine Science	Oyster reef ecology and restoration; oyster filter-feeding and nutrient cycling dynamics
Andy Lacatell	Active	The Nature Conservancy	Oyster restoration; oyster aquaculture
Mark Luckenbach	Active	Virginia Institute of Marine Science	Oyster ecology and restoration; interactions between shellfish aquaculture and the environment; land-use practices and water quality in tidal water environments
Frank Marenghi	Active	Maryland Department of Natural Resources	Fisheries management
Chris Moore	Active	Chesapeake Bay Foundation	Fisheries and oyster restoration; oyster aquaculture; water quality; implementation of Chesapeake Bay TMDL; BMP review
Matt Parker	Active	Sea Grant at U. of Maryland, Prince George's County Office	Oyster aquaculture; business planning
Ken Paynter	Active	U. of Maryland Marine, Estuarine, Environmental Sciences/Chesapeake Bay Laboratory	Oyster restoration; oyster biology and population dynamics
Julie Rose	Active	NOAA Northeast Fisheries Science Center, Milford Lab	Nutrient bioextraction; marine spatial planning for shellfish activities; aquaculture-environment interactions

Larry Sanford	Active	U. of Maryland Center for Environmental Science	Coastal physical oceanography; sediment transport; oceanographic instrumentation
Bill Wolinski	Active	Talbot County Department of Public Works (Retired July 2019)	Watershed Implementation Plans; BMP implementation; water quality
Advisors	**Status**	**Affiliation**	**Expertise**
Ed Ambrogio	Inactive	U.S. EPA Chesapeake Bay Program Office	EPA Region 3 Representative
Rich Batiuk	Inactive	U.S. EPA Chesapeake Bay Program Office	BMP Verification Representative
Matt Johnson	Inactive	U.S. EPA Chesapeake Bay Program Office	Watershed Technical Workgroup (WTWG) Representative
Lew Linker	Active	U.S. EPA Chesapeake Bay Program Office	Chesapeake Bay Modeling Team Representative
Lucinda Power	Active	U.S. EPA Chesapeake Bay Program Office	Water Quality Goal Implementation Team Representative
Jeremy Hanson	Active	U.S. EPA Chesapeake Bay Program Office (previously Virginia Tech)	Watershed Technical Workgroup (WTWG) Representative, Water Quality Goal Implementation Team (WQGIT) Coordinator
Jeff Sweeney	Active	U.S. EPA Chesapeake Bay Program Office	Watershed Technical Workgroup (WTWG) Representative
Ralph Spagnolo	Inactive	U.S. EPA Region III	EPA Region 3 Representative
Coordinators	**Status**	**Affiliation**	**Expertise**
Olivia Caretti	Active	Oyster Recovery Partnership	Oyster restoration and monitoring; coastal ecology
Emily French	Inactive	Oyster Recovery Partnership	Seagrass ecology; water quality monitoring; oyster restoration
Julie Reichert-Nguyen	Inactive	NOAA, Chesapeake Bay Office (previously Oyster Recovery Partnership)	Coordination and facilitation; Clean Water Act; TMDL program; water quality; fisheries science; eutrophication; climate change; ocean acidification
Ward Slacum	Active	Oyster Recovery Partnership	Program management; oyster restoration; environmental monitoring; fisheries ecology

3.2 Panel Charge

The Oyster BMP Expert Panel was charged with fulfilling three goals based on the Chesapeake Bay Program Partnership's Expert BMP Panel Review Protocol for nutrient (nitrogen and phosphorus) and sediment controls (CBP 2015):

Goal 1. Reach a consensus on acceptable nutrient and suspended sediment reduction effectiveness estimates for oyster practices in Chesapeake Bay based on existing science.

Goal 2. Determine a methodology to update these estimates when new science becomes available.

Goal 3. Establish reduction effectiveness crediting and verification guidelines that can be incorporated in the CBP Partnership's model framework used to inform the Chesapeake Bay TMDL.

To meet these goals, the Oyster BMP Expert Panel focused on the following three charges:

Charge 1. Identify and define oyster practices, including aquaculture and restoration activities, for nutrient reduction BMP consideration. Evaluate whether existing science supports the evaluation of sediment reduction effectiveness.

Charge 2. Develop a decision framework that will allow for incremental approval of recommended oyster BMPs.

Charge 3. Propose recommendations for estimating nitrogen, phosphorus, and suspended sediment reduction effectiveness for various oyster practices and oyster-associated processes to help inform the Chesapeake Bay TMDL.

3.2.1 Key changes from the Oyster BMP Expert Panel Charge

- In the Panel Charge (ORP 2015) the decision framework was referred to as the *Pollutant Removal Crediting Decision Framework*. The Panel has decided to refer to it as the *Oyster BMP Nutrient and Suspended Sediment Reduction Effectiveness Determination Decision Framework*, (hereafter, "decision framework"). This change was executed to make it clear that the framework is for determining the nitrogen, phosphorus, and suspended sediment reduction effectiveness of oyster practices and not decisions concerning other pollutants or how to implement nutrient trading credits.
- Initially, the Panel Charge included an incremental approval step in the timeline solely for the decision framework. This framework was instead presented and approved in the Panel's first report (Reichert-Nguyen et al. 2016).
- The oyster practices presented in this report have been refined from what was presented in the Panel Charge and the first report. Table 1.1 and 1.2 show the updated oyster practices.

3.3 Panel Effort

The Panel began developing the recommendations outlined in this report in December 2016. They have held 30 meetings to date and have had numerous e-mail and phone conversations to develop the recommendations found in this report. Panel meeting minutes are listed in Appendix L.

Public and stakeholder engagement and outreach was conducted while the Panel was convened. These efforts included open public stakeholder meetings, presentations at GIT meetings, and written updates to the GITs. Details of Panel engagement and communication with stakeholders are listed in Appendix A.

This report was open for a 40-day review period for the CBP Partnership and interested parties from January 30, 2023 to March 10, 2023.

4.0 Oyster BMP Nutrient and Suspended Sediment Reduction Effectiveness Determination Framework

The Panel created a decision framework to provide a consistent and science-based approach for designing oyster BMPs. The full decision framework was approved and is described in more detail in Reichert-Nguyen et al. (2016). This framework was implemented to (1) identify oyster practices that could lead to nutrient and suspended sediment reduction and (2) estimate reduction effectiveness of endorsed practices to inform the Chesapeake Bay TMDL. Figure 4.1 displays the main steps of the Panel's decision framework and Table 4.1 summarizes the main decision considerations for each step.

The Panel's decision framework allows for the incremental determination, approval, and implementation of nitrogen, phosphorus, and suspended sediment reduction effectiveness estimates based on available science for various oyster practices. The decision framework consists of individual reduction effectiveness crediting protocols based on oyster-associated processes that can reduce nitrogen, phosphorus, or suspended sediment. Developing protocols for individual practices allows these protocols to be applied in a step-wise manner when sufficient data becomes available to evaluate each practice-protocol combination for BMP use. Once approved, the practice-protocol combination is added as a BMP option along with any other previously approved oyster BMPs.

Multiple protocols can be paired with each practice as long as the qualifying conditions for each protocol are fulfilled. For multiple protocols that address the same nutrient (e.g., nitrogen), the reduction estimates can be added together to generate a total reduction estimate for a specific practice. The decision framework also allows for opportunities to identify knowledge gaps that should be addressed to determine nutrient reduction. This includes an option to re-evaluate estimates when new data become available.

Any policy questions that were raised by the Panel were shared with the CBP Partnership Management Board for resolution. Addressing policy issues (e.g., nutrient trading) is beyond the purview of the Panel and not included in the decision framework. The decision framework is specific for oyster practices, but the Panel acknowledges that a similar framework could be developed for other filter-feeding organisms found in the Chesapeake Bay and its tributaries.

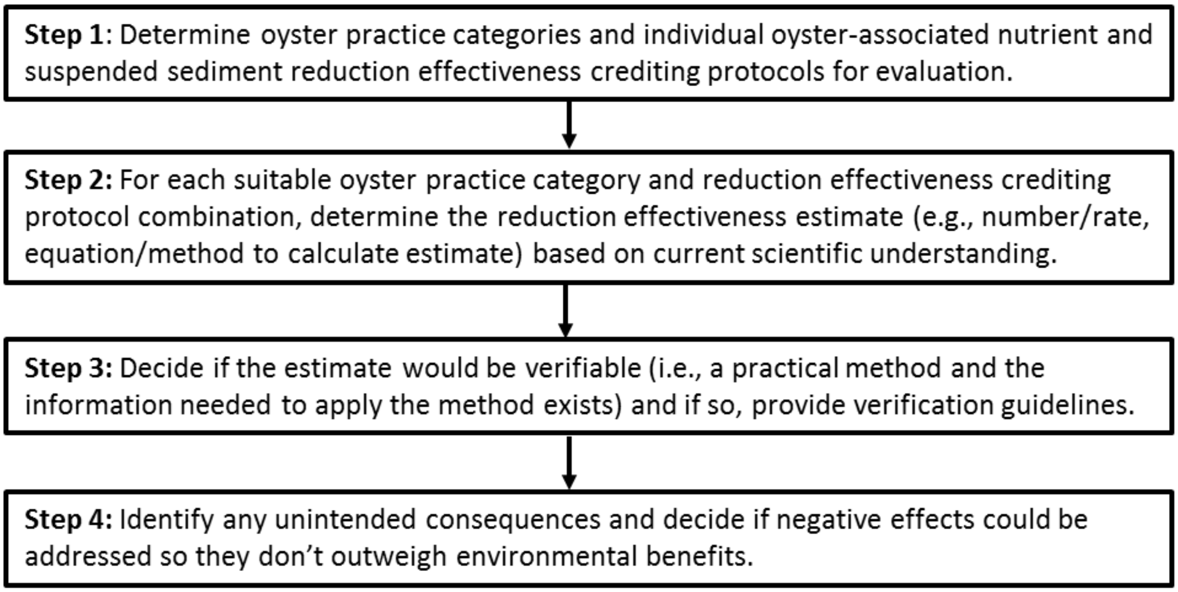


Figure 4.1. Main steps of the Panel's decision framework for providing nutrient reduction effectiveness estimates (Reichert-Nguyen et al. 2016).

Table 4.1. Key decision points for each step in the Panel's decision framework (Reichert-Nguyen et al. 2016). Decisions were made based on the Panel's best professional judgement.

Step	Decision Consideration	Description
1	Suitable for reduction consideration	The nutrient or suspended sediment reduction process should occur in association with a particular oyster practice. The reduction process involves an enhancement activity that could result in the production of new oysters.
2	Sufficient science	Data of sufficient quality and scope exist and can be used to generate a reasonable estimate of the reduction associated with a particular oyster practice.
3	Verifiable	A practical method exists, or could be created, to track reduction effectiveness if the BMP is implemented.
4	Unintended consequences	Identify potential negative effects on the environment resulting from the practice.

5.0 Oyster Practices Evaluated for BMP Consideration

Various oyster practices exist in the Chesapeake Bay that locally enhance oyster populations. The goals of these practices range from increasing oyster production for harvest to meeting oyster population and/or ecosystem-level restoration or conservation goals. Regardless of the goals or management approach of a particular practice, any practice that increases oyster biomass could potentially lead to water quality benefits.

The specific oyster practices identified and reviewed by the Panel encompass the following considerations (also provided in Table 1.2):

- Oyster type
  - Diploid vs. triploid
  - Hatchery-produced vs. wild
- Culture Method
  - Grown on- vs. off-bottom
  - Grown within vs. without gear
  - Planting hatchery-produced oysters vs. transplanting wild juvenile oysters
- Fisheries management approach
  - Private oyster aquaculture – oysters removed (harvested) from water
  - Licensed oyster harvest – oysters removed (harvested) from water
  - Oyster reef restoration – oysters remain in water

The oyster type varies based on the individual practice, fisheries management goal, and limitations of specific locations where enhancement is planned. Wild oysters are diploid and capable of sexual reproduction. Hatchery-produced diploid oysters are similar to wild oysters, but can also be selectively bred to exhibit faster growth and/or be resistant to common diseases (Rawson et al. 2010, Dégremont et al. 2015). Hatchery-produced triploid oysters are created by manipulating chromosomes of reproductively viable adults to produce offspring incapable of sexual reproduction. Triploid oysters usually grow faster than diploid oysters (Allen & Downing 1986) and may exhibit greater disease resistance (Dégremont et al. 2015).

The remainder of this chapter discusses each oyster practice (summarized in Table 1.1 and 1.2), its definition, and relevant examples in detail. The Panel endorsed an oyster practice for BMP consideration if the practice includes an enhancement activity that could result in the production of new oysters and/or increases in oyster biomass. The reduction effectiveness must be attributed to the practice.

5.1. Private Oyster Aquaculture

Private oyster aquaculture practices occur in State-permitted areas where licensed oyster harvesting is not allowed and use either hatchery-produced diploid or triploid oysters, wild oysters, or a combination of these (Table 5.1). These practices involve growing oysters on or off the bottom in protective gear (e.g., floating rafts near the surface or cages near the bottom) or directly on the bottom without gear. Oysters grown off-bottom are usually grown as single oysters where the initial shell substrate is indistinguishable from the rest of the shell (“cultchless”). Aquaculturists growing oysters on the bottom without gear typically enhance the bottom by reclaiming existing shell or adding hard substrate (e.g., shell, stone, etc.; hereafter, “substrate addition)” to facilitate recruitment of naturally-occurring oyster larvae. There are also on-bottom aquaculture operations that will plant hatchery-produced spat-on-shell (oyster larvae that have settled on a shell base) on their leased areas. Private oyster aquaculture operations may also move wild juvenile oysters (which occur naturally on pre-existing reefs) from one location to another to enhance areas that do not receive high densities of larvae. In some instances, lease holders do not carry out any enhancement activities and instead harvest wild oysters within their leased area. From the Panel's understanding, this is not the intended use of oyster aquaculture leases, since state policies typically issue leases in areas that are unlikely to have pre-existing oysters or viable reef habitat (Code of Maryland Regulations COMAR 08.02.04.17, Code of Virginia §28.2-603).

In it's first report, the Panel endorsed three out of five private oyster aquaculture practices for BMP consideration (Table 5.1):

- **Practice A**. Off-bottom private oyster aquaculture using hatchery-produced oysters
- **Practice B**. On-bottom private oyster aquaculture using hatchery-produced oysters
- **Practice D**. On-bottom private oyster aquaculture using substrate addition

The definitions and examples for these three practices are presented in Reichert-Nguyen et al. (2016). Currently, the Panel is not recommending any additional reduction effectiveness estimates for approval for these practices. The Panel has included additional considerations for these practices in Appendices E and F.

Table 5.1. Private oyster aquaculture practices and endorsement decisions approved in Reichert-Nguyen et al. 2016. HPO = Hatchery-produced oysters, POA = Private oyster aquaculture.

Oyster Fate	Oysters removed (harvested) from Bay				
Fisheries Management Approach	Private oyster aquaculture (POA)				
Description	Oyster harvest from State-issued water column and bottom leases				
Access to Oysters	Lease-holder				
Oyster Type	Hatchery-produced oysters (HPO: diploid or triploid)		Wild oysters (diploid)		
Activity	HPO grown off the bottom using gear	HPO grown on the bottom using no gear	Moving wild oysters from one bottom location to another	Addition of substrate to enhance recruitment of wild oyster larvae	None
Oyster Practice	**A. Off-bottom POA using HPO**	**B. On-bottom POA using HPO**	C. On-bottom POA using transplanted wild oysters	**D. On-bottom POA using substrate addition**	E. POA with no activity
Recommended for BMP?	**Yes**	**Yes**	No	**Yes**	No

5.2 Licensed Oyster Harvest

The Panel defined ***licensed oyster harvest*** as oyster harvest from a State-managed fishery area by individuals holding the proper licenses. The Panel reviewed four licensed oyster harvest practices (Table 5.2) but determined that crediting for licensed oyster harvest can only occur on areas that are supplemented with hatchery produced oysters (e.g., spat-on-shell or single oysters). The addition of hatchery-produced oysters is common in areas of the Chesapeake Bay where natural recruitment is low (e.g., Upper Bay). In other areas, natural larval supply is higher, and practices that increase the abundance of suitable substrate are common (i.e., substrate addition). In some cases, juvenile oysters are moved from areas of high density to areas of low oyster density to supplement local populations for harvest. All of these practices aim to enhance the number of harvestable oysters on public reefs. Licensed oyster harvest can also occur in areas where no additional enhancement activity takes place.

The Panel endorsed one out of four licensed oyster harvest practices for BMP consideration (Table 5.2):

- **Practice F**. Licensed oyster harvest using hatchery-produced oysters
  Definition: Planting oysters (e.g., spat-on-shell, single oysters) produced from hatchery techniques directly on the bottom to enhance the stock in State-designated fishing areas (e.g., public shellfish fishing grounds) for eventual removal (harvest) from the water by individuals holding the proper licenses.

This practice is similar to private oyster aquaculture **Practice B** (On-bottom private oyster aquaculture using hatchery-produced oysters), but there is a fundamental difference in where the oysters are planted and who has access to the oysters. Leases are typically issued in areas that are unlikely to have pre-existing oysters or viable reef habitat. For example, Virginia law currently does not allow leasing of “public oyster beds, rocks, or shoals, as defined by law and included in the Baylor survey” (Code of Virginia §28.2-603) and Maryland will only consider permitting a new lease “If the results of a biological survey conducted by the Department or a designated agent show that the average density of oysters per square meter is equal to or below the maximum threshold of one oyster that is 1 inch or greater per square meter” (Code of Maryland Regulations COMAR 08.02.04.17). **Practice F** explicitly applies only to areas where hatchery-produced oysters are planted on public harvest areas.

The location of enhancement and who has access to oysters could also affect verification procedures. Private oyster aquaculture offers a controlled setting as all oyster-related activities are generally implemented by a specific entity (single individual or small group of individuals). Planting oysters in a public harvest area results in oyster-related activities implemented by a much larger group of people. Oysters added to public harvest areas can also be harvested by any harvester that has the proper licenses. For this practice, accurate and timely harvest reporting and verification that the reported data are accurate will be crucial. Therefore, while the Panel is endorsing **Practice F** (Licensed oyster harvest using hatchery-produced oysters) for BMP consideration, they agreed that additional requirements are needed to ensure that these confounding factors are addressed. These recommendations are described in Chapter 6.0.

Similar to the private oyster aquaculture **Practice C & E**, the Panel agreed that licensed oyster harvest **Practice G & I** (transplant wild oysters & no enhancement activity) should not undergo BMP consideration. Transferring live animals from one location to another (**Practice G**) does not result in a net reduction of nitrogen or phosphorus because there is no increase in oyster production. Moreover, practices that do not include any enhancement activity (**Practice I**) are not endorsed by the Panel. These oysters are already present in the water and are better suited to be incorporated as an ecological component in the CBP Partnership’s TMDL model.

The Panel is not currently endorsing licensed oyster harvest using substrate addition alone (**Practice H**) because of the lack of information and confounding results on whether the addition of substrate alone increases oyster production in productive areas where harvesting occurs. This pending practice is discussed in Subchapter 5.4.

Table 5.2. Licensed oyster harvest practices and endorsement decisions seeking approval in this report. HPO = Hatchery-produced oysters, LOH = Licensed oyster harvest.

Oyster Fate	Oysters removed (harvested) from Bay			
Fisheries Management Approach	Licensed oyster harvest (LOH)			
Description	Oyster harvest from State-managed fishing areas			
Access to Oysters	License-holder			
Oyster Type	Hatchery-produced oysters (HPO: diploid or triploid)	Wild oysters (diploid)		
Activity	Addition of HPO	Moving wild oysters from one location to another	Addition of substrate to enhance recruitment of wild oyster larvae	None
Oyster Practice	**F. LOH using HPO**	G. LOH using transplanted wild oysters	H. LOH using substrate addition	I. LOH with no activity
Recommended for BMP?	**Yes**	No	Later	No

5.2.1 Example of **Practice F**: Licensed Oyster Harvest using Hatchery-Produced Oysters

Licensed oyster harvest using hatchery-produced oysters (**Practice F**) consists of planting hatchery-produced diploid or triploid oysters directly on the bottom. The goal is for these oysters to be removed once they reach market size (3 inches in MD and VA) and can be harvested for consumption. Oysters require approximately two to three years to reach minimum market size, but oysters may be left in the water beyond this size. In some cases, the fishery area may be temporarily closed to harvest to allow oysters to grow. These may be referred to as “reserves” or incorporated into a rotational harvest design.

To the best of the Panel’s knowledge, licensed oyster harvest using hatchery-produced oysters has only occurred in Maryland and spans less than 180 acres (F. Marenghi, MD DNR, pers. comm.). This practice is executed through a program where County Oyster Committees or local Waterman Associations work with the Maryland Department of Natural Resources (MD DNR) to (1) identify locations needing replenishment and (2) plant hatchery-produced oysters in public harvest areas (MD DNR 2016). MD DNR reviews the reefs selected for replenishment to ensure that policy guidance is being met. Plantings typically use diploid, hatchery-produced spat-on-shell and occur on portions of historically productive oyster bars where pre-existing oysters could be present. In rare cases, hatchery-produced, triploid oysters are used to reach minimum legal harvest size faster than would be possible if using diploid oysters (F. Marenghi, MD DNR, pers. comm.). The Oyster Recovery Partnership coordinates and assists with plantings and tracks the locations and amounts of oysters that are planted (ORP 2021). Although this practice is not currently used in Virginia, the Panel agreed it could be applied as a BMP in Virginia if occurs in the future.

5.3 Oyster Reef Restoration

In the Chesapeake Bay, oyster reef restoration in designated no-harvest areas aims to increase the number of oysters that will remain in the water. Oysters form 3-dimensional structures which provide additional surfaces available for oyster larvae to settle, grow, and reproduce. Adding diploid oysters to the water will also increase the number of oyster larvae that could settle, grow, and reproduce, thus contributing even more oyster larvae to the system through positive feedback mechanisms. The primary goal of enhancing oyster habitat is to create self-sustaining local oyster populations. Other goals include increasing disease resilience and providing additional ecosystem services, such as increasing habitat for other organisms, improving water quality, etc.

The oyster reef restoration practices evaluated by the Panel include two active approaches (**Practices J & K**) involving the addition of oysters and/or substrate and one passive approach (**Practice L**) in which neither oysters nor substrate are added as part of the restoration effort. In this case, the enhancement activity is the elimination of harvest pressure to allow the oyster population to naturally recover. The Panel is not endorsing **Practice L** at this time due to a policy issue but endorsement is pending (Subchapter 5.4).

The Panel endorsed two out of three oyster restoration practices for BMP consideration in this report (Table 5.3):

- **Practice J**. Oyster reef restoration using hatchery-produced oysters
  Definition: Planting oysters (e.g., spat-on-shell, single oysters) produced from hatchery techniques directly on the bottom or on suitable substrate to enhance oyster biomass in areas where removal is not permitted.
- **Practice K**. Oyster reef restoration using substrate addition
  Definition: Planting oyster shells and/or alternate substrate directly on the bottom to attract recruitment of naturally occurring (wild) oyster larvae to enhance oyster biomass in areas where removal is not permitted.

Enhancement activities associated with these restoration practices include planting hatchery-produced oysters as spat-on-shell and/or substrate (e.g., shell, stone, concrete, etc.) on the bottom to increase oyster density on a per-acre basis. There are instances where substrate is added to improve the bottom conditions for oyster survival before planting spat-on-shell. When only substrate is added, recruitment is based solely on wild oyster larvae settling on the added substrate.

Given that oyster shell is a limited resource in the Chesapeake Bay, there has been a need to use alternate substrate for restoration. The type of substrate used and how it is incorporated into restoration designs vary from project to project. The size and configuration of alternate substrates used for restoration should be considered if implementers seek to apply certain practice-protocol combinations for BMP use, as not all substrates are eligible for all restoration practice-protocol combinations endorsed by the Panel (see Subchapter 7.2.3, 8.2.3, and 8.5).

Table 5.3. Oyster reef restoration practices and endorsement decisions seeking approval in this report. HPO = Hatchery-produced oysters, ORR = Oyster reef restoration.

Oyster Fate	Oysters remain in Bay		
Fisheries Management Approach	Oyster Reef Restoration (ORR)		
Description	No harvest allowed		
Access to Oysters	State resource management agency		
Oyster Type	Hatchery-produced oysters (HPO: diploid)	Wild oysters (diploid)	
Activity	Designate no-harvest area followed by addition of HPO	Designate no-harvest area followed by addition of substrate	Designate no-harvest area with no additional activity
Oyster Practice	**J. ORR using HPO**	**K. ORR using substrate addition**	L. ORR using no harvest area designation only
Recommended for BMP?	**Yes**	**Yes**	Later

5.3.1 Example of **Practice J**: Oyster Reef Restoration using Hatchery-Produced Oysters

Oyster reef restoration using hatchery-produced oysters is most commonly used when natural oyster recruitment is a significant factor limiting local oyster populations. In the Chesapeake Bay, the addition of hatchery-produced oysters as part of a restoration effort is frequently achieved by planting spat-on-shell. Spat-on-shell consists of juvenile oysters, known as "spat", growing on adult oyster shell. Spat-on-shell is generally produced by placing hatchery-produced oyster larvae that are ready to settle in a tank with aged, clean oyster shell and allowing the larvae to settle on the shell and grow until they are deemed large enough for transport to the planting location. Spat-on-shell can either be planted directly on the bottom if it is suitable for oyster survival and growth, or the bottom may be amended prior to planting though the addition of substrate to improve conditions for oyster survival and growth. To date, spat-on-shell planting has been used in a wide variety of restoration efforts, including but not limited to:

- The "10 Tributaries by 2025" effort in Maryland and Virginia (ORIW 2022) established by the 2014 Chesapeake Bay Watershed Agreement in response to Executive Order 13508 (2009) entitled "Chesapeake Bay Protection and Restoration"
- MD DNR's Nanticoke (MD DNR 2022c) and Severn River (MD DNR 2022d) restoration projects.
- Chesapeake Bay community initiatives such as Marylanders Grow Oysters (MD DNR 2022a) and the Virginia Oyster Gardening (CBF 2022) programs.

5.3.2 Example of **Practice K**: Oyster Reef Restoration using Substrate Addition

Oyster reef restoration using substrate addition is most commonly used when a lack of hard substrate for oyster larvae to settle on is the primary factor limiting local oyster populations. Conventional oyster reef restoration methods using substrate addition involve (1) securing suitable substrate (e.g., shell, fossilized shell, granite, crushed concrete, concrete engineered structures, etc.) and (2) deploying this material to create habitat to support natural oyster recruitment. The substrate is deployed in areas where the bottom is suitable for oyster survival and typically in areas that have demonstrated relatively high natural oyster recruitment. This method has been practiced in both Maryland and Virginia but is more common in Virginia. Representative examples of this method can be found in several river systems in Virginia and Maryland that are part of the "10 Tributaries by 2025" restoration program (ORIW 2022). In many cases where suitable habitat is limited and larval supply is low, restoration practitioners employ both the addition of substrate followed by planting of spat-on-shell.

5.4 Oyster Practices with Pending Endorsement

The Panel is currently unable to fully endorse two of the 12 practices reviewed in this report due to lack of information or policy issues:

- **Practice H**. Licensed oyster harvest using substrate addition
  Definition: Planting oyster shells or alternative substrate directly on the bottom to attract recruitment of naturally occurring (wild) oyster larvae to enhance the stock in State-designated fishing areas (e.g., public shellfish fishing grounds) for eventual removal (harvest) from the water by individuals holding the proper licenses.

- **Practice L**. Oyster reef restoration using no-harvest area designation only
  Definition: Designating an area where the removal of oysters is not permitted to enhance the current oyster population with no additional activity (e.g., planting oysters and/or substrate).

The Panel is currently not endorsing licensed oyster harvest using substrate addition (**Practice H**) because of the lack of information and confounding results on whether the addition of shell alone consistently increases oyster production in productive areas where harvesting occurs (Judy 2017, Marenghi et al. 2012, R. Mann, VIMS, unpubl. data). This practice typically aims to restore a positive shell budget (i.e., replace shell removed by harvest) by adding shell to historically productive oyster reefs rather than enhancing oyster biomass directly through the addition of hatchery produced oysters. There is currently not enough information in the existing scientific literature to support the claim that this practice consistently enhances oyster production (leads to increases in oyster biomass) on already productive public harvest areas. Moreover, the Panel determined that it could be difficult to estimate nitrogen and phosphorus reduction effectiveness associated with adding substrate to areas where there is already abundant shell and/or oysters present. Future considerations should investigate whether this practice could be a useful BMP in areas that have relatively high natural recruitment but where substrate for oyster settlement is limited. An example of this pending practice is provided in Subchapter 5.4.1.

The majority of the Panel was in agreement to endorse **Practice L:** (oyster reef restoration using no harvest area designation only) for BMP consideration. This practice designates an area where oysters are not allowed to be harvested but this is not followed by any additional effort to enhance oyster populations (e.g., addition of substrate, addition of spat-on-shell). One panelist was not in support of endorsing this practice since no physical enhancement activity is occurring. Since the rationale for not endorsing this practice was based on a policy concern, the Panel recommends that the CBP Partnership Management Board review this issue and decide on whether this practice can be applied as a BMP (outlined in detail in Subchapter 1.2). From a scientific perspective, the activity of removing harvest pressure on a natural oyster reef by designating it as a no-harvest area could result in increased oyster biomass and consequently increased nitrogen and phosphorus removal via enhanced denitrification and assimilation. The Panel agreed that their recommendations to estimate the reduction effectiveness of nitrogen and phosphorus assimilated in live oysters (Chapter 7.0) and enhanced denitrification (Chapter 8.0) could be applied to this practice, if approved for BMP use. An example of this pending practice is provided in Subchapter 5.4.2.

5.4.1 Example of **Practice H**: Licensed Oyster Harvest using Substrate Addition

Licensed harvest using substrate addition involves (1) securing suitable substrate and (2) deploying this material to create habitat to support natural oyster recruitment. This practice typically occurs in areas where availability of hard substrate for settlement, rather than supply of naturally occurring oyster larvae, is the primary factor limiting local oyster populations. When oysters reach market size (3 inches) they are harvested for consumption. Oysters typically require two to three years to reach minimum market size, but oysters may be left in the water beyond this size. In some cases, the area may be temporarily closed to harvest to allow oysters to settle and grow. In other cases, the area is left open to harvest immediately after deploying the substrate.

Substrate addition to public harvest areas is a common practice in Virginia and Maryland. Locations where this practice occurs in Virginia are documented in the Virginia Oyster Stock Assessment and Replenishment Archive (VIMS 2022). In Maryland, this practice was used as part of the MD DNR Dredged Shell Program that ran from 1960-2006 (Judy 2017). This program dredged fossilized shell from other unproductive locations within the Chesapeake Bay and deployed it in areas where it was thought that the addition of shell substrate would improve oyster production.

5.4.2 Example of **Practice L**: Oyster Reef Restoration using No-Harvest Area Designation Only

No-harvest oyster areas are closed to harvest to protect or allow oyster populations to recover passively through naturally occurring processes (e.g., recruitment, growth, reproduction of wild oysters). This practice does not include any additional enhancement activity (e.g., planting oysters and/or substrate). No-harvest areas are assigned through policy, legislative, and/or regulatory actions. Both Virginia and Maryland have established several oyster sanctuaries where no additional enhancement activities occur (Code of Maryland Regulations COMAR 08.02.04.15, Virginia Administrative Code 4VAC20-650-10). In 2010, Maryland expanded oyster sanctuaries from 9% to 24% (~9,000 acres) of the remaining Maryland oyster reef habitat over a broad geographical range. Virginia has incorporated the preservation of brood-stock sanctuaries in their oyster restoration and rotational harvest into their oyster plan (VMRC 2021).

6.0 Recommendations on Reduction Effectiveness of Nitrogen and Phosphorus Assimilated in Tissue of Hatchery-Produced Oysters Removed by Licensed Oyster Harvest

In its first report (Reichert-Nguyen et al. 2016) the Panel identified two primary oyster harvest approaches used in the Chesapeake Bay: private oyster aquaculture and licensed oyster harvest. The harvest of nitrogen and phosphorus (hereafter "nutrients") assimilated in oyster tissue from private oyster aquaculture is now an approved best management practice. Licensed oyster harvest from public harvest areas also removes the nutrients contained in oyster tissue from the Chesapeake Bay. However, as noted in Chapter 5, the Panel is not endorsing harvest from areas where no enhancement activity has occurred (**Practice I**) or from areas where wild oysters have been transplanted to enhance oyster populations (**Practice G**). After considering whether to endorse licensed oyster harvest from areas where suitable substrate has been added (**Practice H**), the Panel concluded that data were insufficient at this time to support endorsement of this practice. Therefore, the Panel is only endorsing licensed harvest of hatchery-produced oysters (**Practice F**).

This chapter describes the Panel's recommendations for two practice-protocol combinations for licensed oyster harvest:

Practice F. Licensed harvest of hatchery-produced oysters

Protocol 1. Nitrogen assimilation in oyster tissue

Protocol 4. Phosphorus assimilation in oyster tissue

Hereafter, these practice-protocol combinations are referred to collectively as "harvest-assimilation" protocols.

As for private oyster aquaculture (Practices A, B, D), the Panel is recommending that reduction effectiveness be based on harvested oysters. Unlike a newly permitted private oyster aquaculture lease, public harvest areas can have robust oyster populations prior to and long after the enhancement activity occurs (i.e, the planting of hatchery-produced oysters). To account for this and to prevent over crediting for the enhancement activity, the Panel recommends:

- Designating the ***BMP site area*** before enhancement begins
- Using a ***default tissue nutrient content*** based on a diploid shell height to biomass quantile regression.
- Using ***site-specific tissue nutrient contents*** calculated from site-specific shell height to biomass regressions to estimate assimilation only using diploid, not triploid, hatchery-produced oysters.
- Setting a ***maximum harvest allowance*** based on the number of hatchery-produced oysters planted and either a default survival rate (3%) or a site-specific survival rate.
- Applying a ***crediting time lag*** based on either the default time it takes oysters to grow to harvest size (2 years) or a site-specific amount of time.
- Setting a ***maximum crediting timeframe*** of five years after the enhancement activity.
- Requiring calculation of credits based on the minimum legal harvestable oyster size for any harvest reported in units that include mixed sized classes of oysters.

A summary of the Panel's recommended reduction effectiveness determination and qualifying conditions for harvest-assimilation protocols is provided in Table 6.1. The literature and data reviewed by the Panel in developing their recommendations are documented in Subchapter 6.1. The Panel's rationale for its recommended approach to reduction effectiveness determination is described in Subchapter 6.2 followed by stepwise guidance for reduction effectiveness determination in Subchapter 6.3. TMDL baseline considerations and qualifying conditions are described in Subchapter 6.4 and Subchapter 6.5, respectively. Subchapter 6.6 provides guidelines for application and verification. Unintended consequences and ancillary benefits are described in Subchapter 6.7 and Subchapter 6.8, respectively. The Panel's recommendations for future research are provided in Subchapter 6.9.

Table 6.1. Summary of the nitrogen and phosphorus reduction effectiveness strategy for the harvest-assimilation protocols.

Fisheries management approach	Licensed oyster harvest
Oyster practice	**Practice F:** Licensed oyster harvest of hatchery-produced oysters
Practice definition	**Practice F:** Planting oysters (e.g., spat-on-shell, single oysters) produced from hatchery techniques directly on the bottom to enhance the stock in State-designated fishing areas (e.g., public shellfish fishing grounds) for eventual removal (harvest) from the water by individuals holding the proper licenses.
Protocols	**Protocols 1 & 4**: Nitrogen and phosphorus assimilation in oyster tissue
Abbreviated name for practice-protocols	Harvest-assimilation protocols
Reduction effectiveness determination (Subchapter 6.3)	Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 6.2.1) Step 2: Document the qualifying enhancement activity and date it occurred (Subchapter 6.2.2) Step 3: Determine the maximum harvest allowance using either the default (3%) or an approved site-specific survival rate (Subchapter 6.2.3) Step 4: Determine the harvest crediting timeframe (Subchapter 6.2.4) Step 5: Determine the total amount of nitrogen and phosphorus harvested from the BMP site during the harvest crediting timeframe based on the numbers and sizes of oysters harvested and either the default (Subchapter 6.2.5.1) or a site-specific (Subchapter 6.2.5.2) estimate of tissue nutrient content per oyster.
Qualifying Conditions (Subchapter 6.5)	• A qualifying enhancement activity using hatchery-produced oysters (Subchapter 6.2.2) must have occurred throughout the BMP site area (Subchapter 6.2.1). • The BMP site area must lie within an area open to licensed oyster harvest (Subchapter 6.2.2). • At the time of planting, the shell height of hatchery-produced oysters must be <2.0 inches (<50.8 mm; Subchapter 6.2.2). • At the time of harvest, all oysters must be live (Subchapter 6.2.2), of legal harvest size (Subchapter 6.2.5.1), and harvested from within the BMP site (Subchapter 6.2.1). • All oysters must be harvested within the harvest crediting timeframe (Subchapter 6.2.4).

6.1 Literature and Data Review

The Panel agreed that the ***default tissue nutrient contents*** for harvest-assimilation protocols could be based on those developed for private oyster aquaculture practices and approved in the Panel's first report. Detailed descriptions of the literature and data used to develop these estimates can be found in that report (Reichert-Nguyen et al. 2016).

After considering both the diploid and triploid default estimates for oyster tissue nutrient contents, the Panel decided to base default tissue nutrient contents solely on the diploid, not the triploid, nutrient contents from the first report. Diploid oysters are typically used for enhancement of public harvest areas with hatchery-produced oysters. The Panel identified only one case where hatchery-produced, triploid oysters were deployed. In this case, the goal was for oysters to reach market size (3 inches) as quickly as possible (F. Marenghi, MD DNR, pers. comm.). Moreover, the enhancement activity for the harvest-assimilation protocols occur in areas where licensed oyster harvest occurs. These areas are likely to have a pre-existing population of diploid oysters that it would make it difficult to distinguish planted triploids from naturally occurring diploids at the time of harvest. For this reason, if triploid oysters are used, only the default diploid estimates can be applied. Since the diploid tissue estimates are lower than the triploid tissue estimates, this leads to a conservative estimate of nitrogen and phosphorus reduction. If diploid oysters are used, either the default diploid estimates or the site-specific reduction effectiveness estimates can be applied.

There were initial concerns that, because the dataset used to develop the Chesapeake Bay-wide diploid shell height-to-tissue dry weight regression equation for oyster aquaculture practices included oysters grown in gear, it would not be representative of oysters from licensed harvest because the oysters are not grown in gear. However, re-evaluating the diploid data by culture method demonstrated that the majority of the tissue biomass estimates were unchanged from the diploid estimates from the first report (Appendix H). There was only one shell height size class (≥ 5.5 with midpoint of 6 inches) where the nitrogen estimate slightly decreased by 0.01 g per oyster, which equated to 22 lbs per one million oysters (Appendix H, Table H-2). The Panel felt this was an insignificant change, especially since this is a size class that is rarely found in areas open to harvest. Therefore, the Panel agreed that the use of the existing diploid estimates from the first report is sufficient for the harvest-assimilation protocols with some modification to how they are used in this context (see Subchapter 6.2.5.1).

6.2 Reduction Effectiveness: Panel Recommendations and Rationale

The following sections identify the information needed to determine reduction effectiveness, outline the Panel's recommendations, and explain the rationale underlying each of those recommendations.

6.2.1 BMP Site Location and Area

As noted in Chapter 1, for an oyster practice to be eligible for consideration as a BMP, the Panel required that the practice include an enhancement activity that could result in the overall production of new oysters, and consequently, an increase in oyster biomass. For the purposes of crediting, the Panel considers the ***BMP site*** to be the location in which enhancement activities occur and which is potentially eligible for nitrogen and phosphorus reduction credit if all qualifying conditions are met. Recognizing that that the planned location of enhancement activities may not always match the actual location of enhancement activities, the Panel specifically recommends that the ***BMP site*** be determined based upon the actual location of enhancement activities (Fig. 6.1). The BMP site should be described by a series of points (with latitudes and longitudes) which can be connected to form a polygon that encompasses only the area in which the enhancement activities occurred. The area of the polygon defined by these points is the ***BMP site area***. All oysters eligible for crediting must be harvested from within the BMP site.

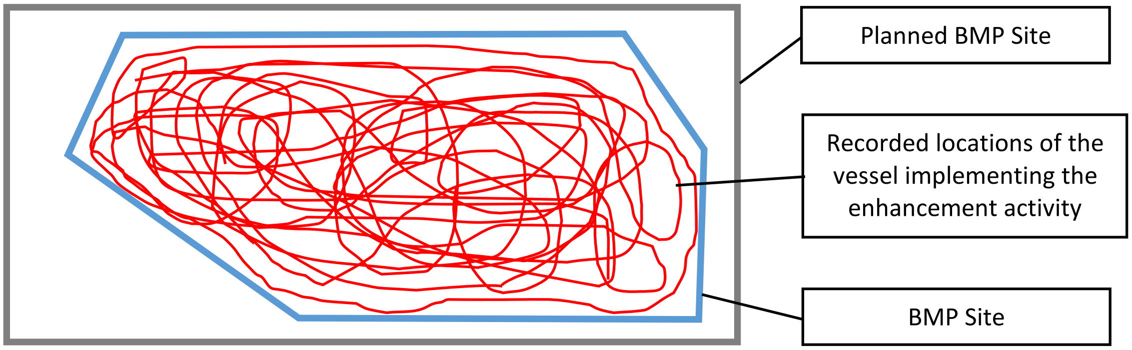


Figure 6.1. Hypothetical map of a BMP site. The area inside the rectangle outlined in gray represents the originally planned location of enhancement activity. The red line represents the recorded locations of the vessel implementing enhancement activity (e.g., the planting “tracklines”). Because the vessel did not implement the restoration activity throughout the entire planned BMP site, the actual BMP site area must be reduced from the original rectangle outlined in gray to the polygon outlined in blue.

6.2.2 Qualifying Enhancement Activities

The Panel considered the types of enhancement activities that could qualify as a BMP under this practice-protocol combination. The Panel found that planting hatchery-produced oysters set on oyster shell (i.e., “spat-on-shell”) in areas open to licensed oyster harvest was the most common practice and that both diploid and triploid hatchery-produced oysters have been used. The Panel agreed that this practice and other similar practices that meet the following conditions could qualify for BMP crediting if:

- The enhancement activity includes the planting of hatchery-produced oysters (diploid or triploid) in an area open to licensed oyster harvest.
- The majority of nutrient assimilation into oyster tissue occurs at the BMP site (i.e., shell height of oysters at time of enhancement activity <2.0 inches shell height).

6.2.3 Maximum Oyster Harvest Allowance

Because enhancement activities for harvest-assimilation protocols occur in areas that likely have pre-existing oyster populations, the Panel deemed it important to try to prevent crediting of oysters unlikely to have resulted from the enhancement activity. As a first step, the Panel compiled reports that included data on the percent survival of oyster spat over time from sites within Chesapeake Bay (Table 6.2). The Panel found that reported survival rates were highly variable and that no studies assessed percent survival to harvest size/age.

Table 6.2. Studies reporting oyster spat survival rates that were reviewed by the Panel. All samples were collected from Chesapeake Bay. Time since planting refers to the amount of time that has elapsed between the time oysters were planted (i.e., deployed in the field) and the time samples were collected and assessed to estimate oyster survival. Lit. = Literature.

Source	Lit. Type	Sampling Locations		Year Sampled	Time since Planting	Survival (%)	
		State	***Sites***			***Mean***	***Range***
Congrove 2008	Thesis	VA	Yecomico, Coan, Great Wicomico, Rappahannock, and Piankatank Rivers	2006	1-2 weeks	63	1-100
Paynter et al. 2012	Report	MD	Chester, Choptank, Little Choptank, Nanticoke Rivers, Harris Creek, and Eastern Bay	2011	4-8 weeks	27	0.4-89.4
Paynter et al. 2013	Report	MD	Harris Creek, Upper Bay	2012	4-8 weeks	36.8	18-61
Paynter et al. 2014	Report	MD	Harris Creek, Severn River	2013	4-8 weeks	37	4-88
Congrove 2008	Thesis	VA	Yecomico, Coan, Great Wicomico, Rappahannock, and Piankatank Rivers	2007	2.5 years	21	8-33

The Panel considered other data that might be available to estimate survival from planting to harvest size and agreed that a subset of the oyster reef restoration monitoring data collected from Harris Creek, MD three years after planting would be most informative (NOAA 2016, 2017). These data were deemed suitable because:

- Monitoring data came from areas in Maryland relatively close to where licensed oyster harvest using hatchery produced oysters currently occurs
- The materials and methods used for restoration in the subset of data analyzed were extremely similar to the materials and methods commonly used for licensed oyster harvest using hatchery produced oysters
- Monitoring data were collected three years after planting. Harvest from areas supplemented with hatchery produced oysters typically occurs two to three years after planting. Use of a spat survivorship from three years after planting should lead to a relatively conservative survival estimate, thereby preventing over crediting.

Analyses found that oyster survival from planting of spat-on-shell to three years ranged from 0.07% to 4.49% with an average of 2.58% (Table 6.3). Weighting of data to account for the acreage of each reef resulted in a weighted average survivorship of 2.91%. Based on these data and rounding the value for the sake of simplicity, the Panel agreed that 3% is reasonable ***default survival rate*** for hatchery produced oysters from the time of planting to the time of harvest. Thus, the ***default maximum harvest allowance*** from a BMP site is 3% of the number of hatchery-produced oysters planted at that site.

Table 6.3. Results of Panel analysis of oyster survivorship from planting of spat-on-shell until three years after planting for select reefs in Harris Creek, MD. See Appendix D for details of analyses.

Reef name	Year planted	Year monitored	Area (acres)	% Survival
Reef #03	2012	2015	6.56	2.75%
Reef #04	2012	2015	11.24	3.48%
Reef #05	2012	2015	15.65	4.22%
Reef #07	2012	2015	10.95	2.52%
Reef #08	2012	2015	7.34	1.38%
Reef #09	2012	2015	12.29	3.02%
Reef #10	2012	2015	10.88	4.32%
Reef #11	2012	2015	6.53	1.80%
Reef #12	2012	2015	7.83	1.63%
Reef H42	2013	2016	5.63	1.81%
Reef H43	2013	2016	4.52	3.53%
Reef H44	2013	2016	2.58	1.95%
Reef H45	2013	2016	3.08	0.07%
Reef H46	2013	2016	7.95	1.69%
Reef H47	2013	2016	9.21	4.49%
			Average:	***2.58%***
		Average weighted by reef area:		***2.91%***

Because the rate of oyster survival to harvest size in some areas may consistently exceed 3%, the Panel agreed that a ***site-specific survival rate*** could be developed and used to create a ***site-specific maximum harvest allowance*** for reduction credit. At a minimum, developing a site-specific mortality estimate would require two surveys of the oyster population at the BMP site: one prior to the enhancement activity and another prior to oyster harvest. Because multiple survey and statistical approaches can be used to evaluate oyster populations and the best approach can depend on a variety of factors, the Panel did not feel it was appropriate to recommend a specific sampling approach. Instead, the Panel recommends that BMP implementers consult with expert(s) knowledgeable in oyster sampling and have their sampling designs endorsed by the state reporting agency and the CBP prior to implementation. The Panel also recommends that site-specific survival rates and the resulting site-specific maximum oyster harvest allowance be reviewed and approved using an approach similar to that described by CBP for re-evaluation of existing estimates (CBP 2015). If approved, revised values can be used only for the BMP site from which data were collected for use in developing site-specific values and cannot be used for other BMP sites.

Regardless of the approach used to set the maximum harvest allowance, the Panel agreed that crediting must be based on the actual numbers and sizes of oysters harvested. The Panel also agreed that all oysters must be alive at the time of harvest to be eligible for crediting.

6.2.4 Timing of Harvest Relative to Enhancement Activity

The Panel also considered the timing of harvest and crediting relative to the timing of the enhancement activity (i.e., planting of spat-on-shell). In Chesapeake Bay, hatchery-produced oysters are typically planted from May to September. Licensed oyster harvest is permitted from October to March. It is expected that most of the planted oysters that survive will reach the currently allowed harvest size of three inches (76 mm) within two years and will be eligible for harvest in the third harvest season after the enhancement activity. Specifically, to ensure that the majority of the harvested oysters eligible for nitrogen and phosphorus reduction credit are likely attributable to the enhancement activity, the Panel recommends a ***default credit time lag*** of two years after the date when hatchery-produced oysters were placed in the field (i.e., "planted").

Because oyster growth rates vary widely, the Panel agreed that a ***site-specific credit time lag*** can be developed if implementing programs collect sufficient oyster growth data from the BMP site to demonstrate that, on average, oysters reach harvest size in less than two years. If a site-specific credit time lag is sought, the Panel recommends that the plan for associated sampling and data analysis be developed in consultation with expert(s) knowledgeable in oyster sampling and endorsed by the state reporting agency and the CBP prior to implementation. Any adjustments to credit time lag should be reviewed and approved using an approach similar to that described by CBP for re-evaluation of existing estimates (CBP 2015). If approved, the revised time lag can be used only for the BMP site from which data were collected for use in developing site-specific values and cannot be used for other BMP sites.

Again, to ensure that the majority of the harvested oysters eligible for credit are likely attributable to the enhancement activity, the Panel considered the maximum amount of time after planting that harvest from a BMP site should be eligible. Based on their expert opinion, the Panel concluded that most of the hatchery produced oysters planted at the BMP site would be harvested within five years. Because planting activities generally occur in summer months, oysters will reach five years old between the fifth and six harvest season after planting. Thus, the Panel is recommending five years as the ***maximum harvest timeframe*** meaning that credits will expire after the fifth and before the sixth harvest season following the enhancement activity. The ***harvest crediting timeframe*** is the time window during which the nitrogen and phosphorus reduction from the oyster harvest allowance can be credited. If using the default reduction credit time lag, this window begins two years after the enhancement activity and ends three years later when the 5-year oyster harvest timeframe ends. If using the Panel's default timeframes, oysters harvested in the third, fourth and fifth harvest seasons after planting are eligible for crediting. For the BMP to remain continuously active, qualifying enhancement activities must occur at the BMP site at a minimum of once every three years.

The Panel agreed that, if the default oyster harvest allowance and default harvest crediting timeframe are used, pre-planting and pre-harvest oyster population assessments are not needed.

6.2.5 Amount of Tissue Harvested and Associated Nutrient Content

The Panel is recommending two options for determining the amount of nitrogen and phosphorus in the tissue of harvested oysters for this practice-protocol combination:

- Default approach
- Site-specific approach

The default estimates for nitrogen and phosphorus assimilation represent typical conditions across the entire Bay and the entire suite of environmental conditions that influence oyster growth. In contrast, site-specific estimates represent the nitrogen and phosphorus contained in the tissue of oysters at a single BMP site. Site-specific estimates can potentially be higher than the default estimate but require collection of considerably more data from the BMP site. Regardless of the approach used, the first step is determining the numbers and sizes of oysters harvested.

6.2.5.1 Numbers and Sizes of Harvested Oysters

The Panel agreed that to be eligible for credit the number and sizes of harvested oysters must be directly assessed. In considering methods that might be used to determine the number of oysters harvested, the Panel explored current oyster harvest reporting requirements. In the Chesapeake Bay, commercial fishermen are required to quantify and report monthly oyster harvest to their state management agency. Harvest is typically reported in the same units in which they are sold (e.g., bushels) and these units can vary across jurisdictions. The Panel agreed that a variety of reporting units should be allowed, but also agreed that there must be a clear and defensible method for converting these units into the number of individual oysters.

At present, a variety of approaches are used to convert the units in which oysters are reported to jurisdictions into numbers of individual oysters. For example, the Maryland Department of Natural Resources (MD DNR) defines a bushel by volume. In 2018, they used 228 oysters per bushel for their stock assessment based on data collected at an oyster dealer that year (F. Marenghi, MD DNR, pers. comm.). Another approach is to use independent third-party verifiers to randomly spot check oyster quantities in the container-type being used (Slacum et al. 2013). This approach is being implemented through the MD DNR's pilot project that is testing daily electronic reporting by watermen for oyster harvest (MD DNR 2022b).

Because a variety of reporting units may be used and these units may change over time, the Panel did not think it was appropriate to recommend a specific method for converting harvest reporting units into the number of individual oysters. Instead, the Panel recommends that an implementer seeking credit work in conjunction with their local jurisdiction and the CBP to develop a scientifically defensible method for converting reporting units into numbers of individual oysters. This method should be reviewed and approved by the reporting jurisdiction and CBP to ensure that it can be applied to meet TMDL requirements. It should also be clearly documented in the implementation plan.

Another challenge in determining the appropriate nutrient credits for licensed oyster harvest is that the majority of harvest units reported include mixed oyster size classes. During harvest, oysters are typically culled to remove oysters smaller than the minimum legal size and then all oysters above that size limit are stored in the same container. The Panel agreed that when oysters of mixed size classes are combined within the reporting unit, the nitrogen and phosphorus content of the tissues of those oysters should be based on oysters of the minimum legal harvest size. For example, if three inches is the legal harvest size, then the 3-inch diploid estimate should be used to calculate the nitrogen and phosphorus reduction even though some larger oysters were likely harvested. This approach results in a conservative estimate of the amount of nitrogen and phosphorus assimilated in oyster tissue.

Although most commercially harvested oysters are not reported in terms of size class, the Panel agreed that, if procedures are in place that require the sorting of oysters by uniform BMP sizes along with statistically supported verification approaches, then crediting oysters by their respective size classes could be feasible. The Panel recommends that any approaches used to generate nutrient reduction credits above the minimum legal harvest size be incorporated into the BMP implementation plan and reviewed and approved by the reporting agency and CBP.

6.2.5.2 Default Tissue Nutrient Content

As noted in Subchapter 6.1, the Panel agreed that the default diploid oyster tissue nutrient contents developed for private oyster aquaculture could serve as the basis for default tissue nutrient content estimates for the harvest-assimilation protocols. The private oyster aquaculture default reduction effectiveness calculations allow crediting of oysters less than three inches in shell height (Fig. 6.2). However, current state regulations do not allow harvest of oysters less than three inches in shell height from public harvest areas (Code of Maryland Regulations COMAR 08.02.04.11, Virginia Administrative Code 4VAC20-260-30). The Panel agreed that only oysters of legal harvest size are eligible for crediting. For this reason, the Panel decided to remove the 2.00 – 2.49 inch size class. The Panel also decided to change the name of the 2.50 – 3.49 inch size class to a 3.00 – 3.49 inch size class but to retain the original estimated nutrient content for the 2.50 – 3.49 inch size class. This results in an intentionally conservative estimate of the nutrient content of oysters in this size class. The resulting ***default tissue nutrient contents*** for the harvest-assimilation protocols are given in Table 6.4 in terms of grams of nitrogen and phosphorus per oyster. In Table 6.5, these same values are given in terms of pounds of nitrogen and phosphorus per million oysters. Details of the approaches and data used to determine the default diploid tissue nutrient content can be found in the Panel's first report.

Figure 6.2. The measurement location for shell height. Shell height is the longest distance (parallel to the long axis) between the hinge and lip of the oyster. Note that shell height is also referred to as oyster shell length in some studies.

Table 6.4. Recommended default nitrogen and phosphorus content of diploid oyster tissue. Oyster size class based on shell height measurements. Mean percent nitrogen content = 8.2%; Mean percent phosphorus content = 0.9%; Regression equation: $y=0.0004x^{1.82}$ (Reichert-Nguyen et al. 2016).

Oyster size class (in)	Midpoint (in)	Midpoint (mm)	Tissue dry weight (g oyster^{-1})	Content in oyster tissue (g oyster^{-1})	
				Nitrogen	Phosphorus
3.00-3.49*	3	76	1.06	0.09	0.01
3.50-4.49	4	102	1.81	0.15	0.02
4.50-5.49	5	127	2.70	0.22	0.02
≥ 5.50	6	152	3.74	0.31	0.03

* Adjusted from 2.50-3.49. See text for details.

Table 6.5. Default nutrient reductions in pounds per one million harvested hatchery-produced oysters. Oyster size class based on shell height measurements.

BMP Name	Oyster size class (in)	Nitrogen (lbs./million oysters)	Phosphorus (lbs./million oysters)
Diploid Licensed Oyster Harvest, Hatchery Produced 3.0 Inches	3.00-3.49*	198	22
Diploid Licensed Oyster Harvest, Hatchery Produced 4.0 Inches	3.50-4.49	331	44
Diploid Licensed Oyster Harvest, Hatchery Produced 5.0 Inches	4.50-5.49	485	44
Diploid Licensed Oyster Harvest, Hatchery Produced >5.0 Inches	≥ 5.50**	683	66

* Adjusted from 2.50-3.49. See text for details.
** Based on midpoint of 6.0 inches

6.2.5.3 Site-specific Nitrogen and Phosphorus Reduction Estimates

The Panel also recommends allowing development of ***site-specific tissue nutrient contents*** for the harvest-assimilation protocols for harvested diploid oysters. The Panel is not recommending this option for areas where hatchery-produced triploid oysters are planted in public harvest areas (see Subchapter 6.1). The site-specific method recommended by the Panel relies on measured oyster tissue biomass and default values for the percentage of nitrogen and phosphorus in oyster tissue. The Panel decided this was a reasonable, scientifically defensible approach because variance in the relationships between oyster shell height and tissue dry weight is far greater than variance in the relationships between tissue dry weight and nutrient content.

To establish site-specific tissue nutrient contents, the Panel recommends the implementer work with the reporting jurisdiction, CBP Partnership, and expert(s) in oyster sampling and sample processing to:

- Define specific oyster size classes if they differ from the size classes used for default tissue nutrient contents
- Identify at least two evenly distributed sampling periods to ensure sampling reflects seasonal differences within the allowed harvesting timeframe set by state regulations.
- Assess the average tissue dry weight for each size class based on 50 randomly selected oysters per size class and sampling period. Oyster samples must be processed at a lab that uses standardized methods to acquire the tissue dry weight in grams (e.g., tissue heated at 60°C until samples reach constant weight; Mo & Neilson 1994, Carmichael et al. 2012).
- Multiply the average tissue dry weight for each size class by the default nitrogen percentage (8.2%) and phosphorus percentage (0.9%) in oyster tissue to determine the site-specific nitrogen and phosphorus content per oyster.

The Panel recommends that site-specific tissue nutrient content be reviewed and approved using an approach similar to that described by CBP for re-evaluation of existing estimates (CBP 2015). If approved, revised values can be used only for the BMP site from which data were collected for use in developing site-specific values and cannot be used for other BMP sites.

6.3 Reduction Effectiveness: Stepwise Determination

To calculate the reduction effectiveness for the harvest-assimilation protocols, the Panel recommends the following:

Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 6.2.1)

Step 2: Document the qualifying enhancement activity and the date it occurred (Subchapter 6.2.2)

Step 3: Determine the maximum harvest allowance using either the default (3%) or an approved site-specific survival rate (Subchapter 6.2.3)

Step 4: Determine the harvest crediting timeframe (Subchapter 6.2.4)

Step 5: Determine the total amount of nitrogen and phosphorus harvested from the BMP site during the harvest crediting timeframe based on the numbers and sizes of oysters harvested and either the default (Subchapter 6.2.5.1) or an approved site-specific (Subchapter 6.2.5.2) estimate of tissue nutrient content per oyster.

6.4 TMDL Baseline Considerations

The CBP Management Board defined the baseline for oyster practices that remove (harvest) oysters to only include oysters that are removed after the BMP is approved/implemented for reduction effectiveness credit in the TMDL. They also established that credit will be counted when oysters are removed (not planted) and that past harvest cannot be credited.

6.5 Qualifying Conditions

The Panel recommends the following qualifying conditions, which account for both the CBP Management Board's defined baseline and the Panel's criteria:

- A qualifying enhancement activity using hatchery-produced oysters (Subchapter 6.2.2) must have occurred throughout the BMP site area (Subchapter 6.2.1).
- The BMP site area must lie within an area open to licensed oyster harvest (Subchapter 6.2.2).
- At the time of planting, the shell height of hatchery-produced oysters must be <2.0 inches (<50.8 mm; Subchapter 6.2.2).
- At the time of harvest, all oysters must be live (Subchapter 6.2.2), of legal harvest size (Subchapter 6.2.5.1) and harvested from within the BMP site (Subchapter 6.2.1).
- All oysters must be harvested within the harvest crediting timeframe (Subchapter 6.2.4).

6.6 Recommended Application and Verification Guidelines

6.6.1 Reporting Guidelines

To assist with application of its recommendations, the Panel developed guidelines for the information to be reported by anyone seeking credit for this practice-protocol combination. The required information is listed below under the associated determination step.

Step 1: Document the BMP site location (Subchapter 6.2.1)

- Geospatial information documenting the vertices of a polygon representing the BMP site
- Name of the licensed oyster harvest area within which the BMP site lies or geospatial information documenting location of the licensed oyster harvest area

Step 2: Document the qualifying enhancement activity and date it occurred (Subchapter 6.2.2)

- Brief description of enhancement activity

- Date of enhancement activity
- Ploidy of hatchery-produced oysters
- Number and size of hatchery-produced oysters at the time of planting

Step 3: Determine the maximum harvest allowance using either the default (3%) or an approved site-specific survival rate (Subchapter 6.2.3)

- Method used to determine maximum harvest allowance (default or site-specific)
    - If site-specific survival rate is used, provide documentation of approval from CBP that site-specific approach has been reviewed and approved.
- Maximum harvest allowance
- Number of oysters previously credited from the same enhancement activity

Step 4: Determine the harvest crediting timeframe (Subchapter 6.2.4)

- Method used to determine the time lag (default or site-specific)
    - If site-specific time lag is used, provide documentation of approval from CBP that site-specific approach has been reviewed and approved.
- Length of time lag
- Start and end dates of harvest crediting timeframe

Step 5: Determine the total amount of nitrogen and phosphorus harvested from the BMP site during the harvest crediting timeframe based on the numbers and sizes of oysters harvested and either the default (Subchapter 6.2.5.1) or a site-specific (Subchapter 6.2.5.2) estimate of tissue nutrient content per oyster.

- Harvest month(s)
- Harvest season
- Units used for reporting oysters
    - If units are not individual oysters, provide documentation of the approved method used to convert units into numbers of individual oysters
- Oysters per reporting unit
- Total number of oysters harvested from the BMP site
    - If size classes are mixed or if all oysters harvested fall into same size class as the minimum legal harvest size class, report total number of individual oysters
    - If BMP size classes greater than the minimum legal harvest size class are used, report number of oysters harvested for each size class and provide documentation of approved size class sorting method and verification.
- Approach used to convert oysters to amounts of nitrogen and phosphorus (default or site-specific). If site-specific, then also include documentation on how the estimates were derived.
- Total amount of nitrogen and phosphorus eligible for credit

The Panel noted that optional information could be provided that could assist with verification of both the enhancement activity and the harvest. This information could include documentation of the purchase of oysters from a hatchery, documentation of costs associated with planting, and additional harvest information such as the name of the harvester and who the oysters were sold to (e.g., dealer, public, etc.).

6.6.2 Example

To give an example of the information needed for crediting, Table 6.6 provides a list of the information along with a hypothetical example of that information for the first year of harvest from a BMP site. Although this example assumes that credit is applied for at the end of the harvest season, credits can be applied for whenever oysters are harvested. Table 6.6 assumes that the values given in monthly reports have been added together and the resulting total number of oysters harvested was used in determining the annual reduction effectiveness credit.

Table 6.6. Information types required for the harvest-assimilation protocols along with an example of each. See text and Tables 6.7, 6.8, and 6.9 for details of calculations used to provide example information below. For this example, the report would also need to include documentation of the approved method used to convert the number of bushels to the number of individual oysters.

Step #	Information Type	Example
1	BMP site location	GIS layer with polygon for BMP site and latitude and longitude for all vertices provided as a separate file
	Name of licensed oyster harvest area	Cedar Point, Broad Creek, MD
2	Enhancement activity	Spat-on-shell planting
	Date(s) of activity (mm/dd/yy)	05/14/19, 06/02/19, 07/17/19
	Ploidy	Diploid
	Number of hatchery-produced oysters planted	50,000,000
	Size of hatchery-produced oysters at time of planting (mm)	10
3	Method used to determine maximum harvest allowance	Default
	Maximum oyster harvest allowance	1,500,000
	Number of oysters previously credited	0
4	Method used to determine time lag	Default
	Length of time lag	2 years
	Oyster harvest crediting timeframe start date	05/14/21
	Oyster harvest crediting timeframe end date	05/14/24
5	Months when harvest occurred	October - March
	Harvest season	2021/2022
	Oyster reporting units	Bushels
	Oysters per reporting unit	300
	Harvest reported	1,000 bushels
	Total number of oysters harvested	300,000
	Number of harvested oysters eligible for credit	300,000
	Method used to convert oyster tissue to nutrients	Default
	Nitrogen removed (lbs)	59.4
	Phosphorus removed (lbs)	6.6

Because oysters attributable to the enhancement activity can be harvested and credited from the start of the harvest crediting timeframe to up to five years after planting, implementers must track both planting and harvest at the BMP site across multiple years. Table 6.7 provides an example of tracking oyster plantings over a 10-year period using the information from Table 6.6 for the first planting. For a BMP to remain continuously active, enhancement activities must occur at least every three years. However, the number of oysters eligible for harvest during a particular harvest season will depend on when oysters are planted, how many are planted, and how many have already been harvested. Table 6.8 uses the planting information from Table 6.7 to provide an example of how all of these interact to determine how much of the harvest allowance is available for crediting at a particular point in time. Following Table 6.8 is a stepwise explanation of the calculations used to develop and fill the table.

Table 6.7. Example of tracking plantings starting with the scenario from Table 6.6.

Year:	2019	2020	2021	2022	2023	2024	2025	2026	2027	2028	Total
Spat Planted (millions)	50.00	25.00	0		25.00	0	0	0	0	0	100.00
Maximum oyster harvest allowance[1] (millions)	1.50	0.75	0	0	0.75	0	0	0	0	0	3.00

[1] Based on 3% default value; could vary if site-specific assessment of spat survivorship is available.

Table 6.8. Example of tracking harvests for the plantings shown in Table 6.7.

Harvest Season:	2019/ 2020	2020/ 2021	2021/ 2022	2022/ 2023	2023/ 2024	2024/ 2025	2025/ 2026	2026/ 2027	2027/ 2028	2028/ 2029	Total
Harvest season after first planting	1st	2nd	3rd	4th	5th	6th	7th	8th	9th	10th	
Harvest of 2019 planting eligible for BMP credit?			Yes	Yes	Yes						
Harvest of 2020 planting eligible for BMP credit?				Yes	Yes	Yes					
Harvest of 2023 planting eligible for BMP credit?							Yes	Yes	Yes		
BMP active based on timeframe?	No[1]	No[1]	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No[2]	
2019 planting harvest allowance remaining (millions)			1.50	1.20	0.75	0	0	0	0	0	
2020 planting harvest allowance remaining (millions)				0.75	0.75	0.75	0	0	0	0	
2023 planting harvest allowance remaining (millions)							0.75	0.15	0	0	
Oysters harvested (millions)			0.30	0.45	0	0.60	0.60	0.75	0.90	0.75	4.35
Oysters credited (millions)			0.30	0.45	0	0.60	0.60	0.15	0	0	2.1
Expiring harvest allowance[3] (millions)					0.75	0.15					0.9

[1] Less than 2 years have passed since 2019 planting
[2] Oysters from most recent planting are more than 5 years old
[3] Eligible harvest allowance not used in final year

Stepwise explanation of calculations in Table 6.8:

- The harvest allowance 1.5 million oysters for the 2019 planting (3% of the 50 million oysters planted) becomes eligible for crediting in the 2021/2022 harvest season and 0.3 million oysters are harvested and credited. This leaves a remaining harvest allowance of 1.2 million oysters at the start of the 2022/2023 harvest season.
- In the 2022/2023 harvest season, the 2020 planting becomes eligible for harvest, adding 0.75 million oysters to the total allowable harvest. To maximize crediting, the 2022/2023 harvest of 0.45 million oysters is subtracted from the harvest allowance for the 2019 planting resulting a remaining harvest allowance of 1.5 million oysters (0.75 million from 2019 planting and 0.75 million from 2020 planting) at the start of the 2023/2024 harvest season.
- In the 2023/2024 harvest season, no oysters are harvested or credited. Because the oysters from the 2019 planting have reached the end of their harvest crediting timeframe, the remaining harvest allowance of 0.75 million oysters expires, leaving 0.75 million oysters from the 2020 planting eligible for crediting at the start of the 2024/2025 harvest season.
- In the 2024/2025 harvest season, 0.6 million oysters are harvested and credited. Because the oysters from the 2020 planting have reached the end of their harvest crediting time frame, the remaining harvest allowance for the 2020 planting of 0.15 million oysters expires.
- In the 2025/2026 harvest season, the 2023 planting becomes eligible for harvest with a maximum harvest allowance of 0.75 million oysters. A total of 0.6 million oysters are harvested and credited leaving a harvest allowance of 0.15 million oysters at the start of the 2026/2027 harvest season.
- In the 2026/2027 harvest season, 0.75 million oysters are harvested. However, only 0.15 million can be credited before reaching the total harvest allowance for the 2023 planting. No credit is given for the remaining 0.60 million oysters.
- In the 2027/2028 harvest season, oysters are harvested but no credits are given because the harvest allowance for the 2023 planting has already been reached. Also, after the 2027/2028 harvest season, the 2023 planting reaches the end of its crediting timeframe.
- In the 2028/2029 harvest season, there is no harvest allowance because no qualifying enhancement activities have taken place since 2023, so the BMP is no longer active.

Out of the 4.35 million oysters harvested in the example given in Table 6.8, 2.1 million of the harvested oysters could be claimed for nitrogen and phosphorus reduction credit. As shown in Table 6.9, the credited harvest equates to a total of 415.8 lbs. of nitrogen and 46.3 lbs. of phosphorus removed.

Table 6.9. Example nitrogen and phosphorus reduction (lbs. removed) based on total harvest claimed for reduction credit (Table 6.7). Default nitrogen and phosphorus contents are for 3.0-inch oysters taken from Table 6.4.

Harvest Season	2019/ 2020	2020/ 2021	2021/ 2022	2022/ 2023	2023/ 2024	2024/ 2025	2025/ 2026	2026/ 2027	2027/ 2028	2028/ 2029	*Total*
Nitrogen (lbs.)	0	0	59.4	89.1	0	118.8	118.8	29.7	0	0	***415.8***
Phosphorus (lbs.)	0	0	6.6	9.9	0	13.2	13.2	3.3	0	0	***46.3***

6.7 Unintended Consequences

The Panel identified no unintended consequences for the harvest-assimilation protocols.

6.8 Ancillary Benefits

The Panel identified several ancillary benefits for the harvest-assimilation protocols. Planting diploid oysters can potentially increase natural recruitment if the oysters spawn and produce viable larvae prior to harvest. In addition, plantings that use spat-on-shell can help maintain a positive shell budget. A positive shell budget increases the possibility of natural spat sets due to the increased availability of suitable substrate. Prior to harvest, oysters will also provide a variety of other ecosystem services including, but not limited to, improving water quality by filtering suspended organic matter and sediments from the water column and providing habitat for other organisms.

6.9 Future Research

The Panel identified the following research gap when developing the nitrogen and phosphorus reduction effectiveness recommendations for the harvest assimilation protocols:

- The default eligible harvest crediting cap was set at 3% of hatchery produced oysters planted based on analyses of data from oyster reef restoration sites in Harris Creek, MD. The Panel suggests that a more robust default harvest allowance and default crediting timeframe could be developed using long-term (>5 year) monitoring of the survival and growth of hatchery-produced oysters planted in areas open to harvest.

7.0 Recommendations on Reduction Effectiveness of Nitrogen and Phosphorus Assimilated in Live Oysters on Restored Oyster Reefs

Oyster reef restoration aims to enhance oyster populations and re-establish self-sustaining reefs. Numerous local, state, and federal partners have been restoring oyster reefs in the Chesapeake Bay since the mid-1990s. These efforts became more focused with the signing of Executive Order 13508 (2009). In 2014, the Chesapeake Bay Watershed Agreement established the goal to restore 10 Chesapeake Bay tributaries by 2025 to achieve increased habitat and water quality benefits (CBP 2014). The "10 Tributaries by 2025" program has been ongoing since 2014.

One of the many benefits of oyster reef restoration is the capacity of restored oyster reefs to sequester nitrogen and phosphorus in the tissues and shells of oysters. When oysters feed on phytoplankton and other organic matter, a portion of the nitrogen and phosphorus contained in that organic matter is assimilated into their tissues and shells. As long as this assimilated nitrogen and phosphorus is retained in the tissues and shells of oysters, water quality is improved because these nutrients cannot be used to fuel excess growth of phytoplankton.

In its first report (Reichert-Nguyen et al. 2016), the Panel identified 12 practice-protocol combinations that could remove nitrogen and phosphorus through assimilation into oysters on restored reefs (Table 1.1). This chapter describes the Panel's recommendations on the following eight oyster practice-protocol combinations seeking BMP approval:

Practice J. Oyster reef restoration using hatchery-produced oysters

- **Protocol 1**. Nitrogen assimilation in oyster tissue
- **Protocol 2.** Nitrogen assimilation in oyster shell
- **Protocol 4.** Phosphorus assimilation in oyster tissue
- **Protocol 5**. Phosphorus assimilation in oyster shell

Practice K. Oyster reef restoration using substrate addition

- **Protocol 1**. Nitrogen assimilation in oyster tissue
- **Protocol 2.** Nitrogen assimilation in oyster shell
- **Protocol 4.** Phosphorus assimilation in oyster tissue
- **Protocol 5.** Phosphorus assimilation in oyster shell

Hereafter, these practice-protocol combinations are referred to collectively as "restoration-assimilation" protocols.

The Panel did not reach consensus on whether to endorse **Practice L** (oyster reef restoration using no harvest area designation only) because of one dissenting opinion due to a policy issue. However, they agreed that the recommendations presented in this chapter could be applied to this practice if endorsed for future BMP use.

To prevent over crediting, the Panel is recommending that the restoration-assimilation protocols be based on appreciated oyster biomass associated with restoration activities at the designated BMP site. Appreciated biomass is defined as the increase in oyster biomass over the maximum biomass previously determined for the site. For the first post-restoration biomass measurement, appreciated biomass will be equal to the post-restoration biomass minus the baseline (i.e., pre-restoration) biomass and is equivalent to biomass enhancement resulting from the restoration activity. For subsequent time periods, appreciated biomass is the amount by which biomass has increased beyond the maximum biomass previously measured at the site. As for other practice-protocol combinations, the Panel is recommending that only the nitrogen and phosphorus assimilated in live native oysters (*Crassostrea virginica)* be eligible for reduction credit.

In contrast to the harvest-assimilation and aquaculture-assimilation protocols, the Panel agreed that it was appropriate to credit nutrients assimilated into both tissue and shell because the shell is not removed from the water as part of reef restoration as it is for these other oyster practices. The Panel also agreed that the reduction effectiveness using restoration-assimilation protocols can be determined for oysters growing in all natural habitats (i.e., subtidal and intertidal) and on all suitable substrate types. For the purposes of this report, ***suitable substrate*** is defined as material on which oysters can settle and survive to adulthood and includes both oyster shell and alternate substrates. ***Alternate substrates*** are all materials not composed solely of oyster shells.

A summary of the Panel's recommended reduction effectiveness determination and qualifying conditions for the restoration-assimilation protocols is provided in Table 7.1. The literature and data reviewed by the Panel in developing their recommendations are documented in Subchapter 7.1. The Panel's rationale for its recommended approach to reduction effectiveness determination is described in Subchapter 7.2 followed by stepwise guidance for reduction effectiveness determination in Subchapter 7.3. TMDL baseline considerations and qualifying conditions are described in Subchapter 7.4 and Subchapter 7.5, respectively. Subchapter 7.6 provides guidelines for application and verification. Unintended consequences and ancillary benefits are described in Subchapter 7.7 and Subchapter 7.8, respectively. The Panel's recommendations for future research are provided in Subchapter 7.9.

The baseline and post-restoration biomass data needed for the restoration-assimilation protocols described in this chapter can also be used for the restoration-denitrification practice protocol combinations described in Chapter 8. The restoration-denitrification protocols include the qualifying condition that the post-restoration oyster tissue biomass from the BMP site has been assessed within the past three years. Therefore, if oyster biomass data are collected at least every three years, then the same data can be used to credit nitrogen reduction for both the restoration-assimilation and restoration-denitrification protocols, assuming all qualifying conditions are met.

Table 7.1. Summary of the nitrogen and phosphorus reduction effectiveness strategy for the restoration-assimilation protocols. Restoration-assimilation protocols can be applied to both subtidal and intertidal reefs, and reefs restored using all suitable substrates.

Fisheries Management Approach	Oyster Reef Restoration
Oyster Practice	**Practice J:** Oyster reef restoration using hatchery-produced oysters **Practice K**: Oyster reef restoration using substrate addition
Practice Definitions	**Practice J:** Planting oysters (e.g., spat-on-shell, single oysters) produced from hatchery techniques directly on the bottom or on suitable substrate to enhance oyster biomass in areas where removal is not permitted. **Practice K:** Planting oyster shells and/or alternate substrate directly on the bottom to attract recruitment of naturally occurring (wild) oyster larvae to enhance oyster biomass in areas where removal is not permitted.
Protocols	**Protocols 1 & 2**: Nitrogen assimilation in oyster tissue and shell **Protocols 4 & 5**: Phosphorus assimilation in oyster tissue and shell
Abbreviated name for practice-protocols	Restoration-assimilation protocols
Reduction Effectiveness Determination Strategy (Subchapter 7.3)	Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 7.2.1). Step 2: Document the qualifying enhancement activity and its date (Subchapter 7.2.2), the type(s) of substrate used for restoration (Subchapter 7.2.3), and the baseline approach (Subchapter 7.2.4). Step 3: Assess baseline and post-restoration tissue and shell biomass (Subchapters 7.2.5) and extrapolate it to determine total tissue and shell biomass estimates for the BMP site (Subchapter 7.2.6). Step 4: Determine the eligible appreciated tissue and shell biomass at the BMP site (Subchapter 7.2.7). Step 5: Convert eligible appreciated tissue and shell biomass to total nitrogen and phosphorus removed (Subchapter 7.2.8).
Qualifying Conditions (Subchapter 7.5)	• A qualifying enhancement activity (Subchapter 7.2.2) must have occurred throughout the BMP site area (Subchapter 7.2.1). • The BMP site area must lie within an area protected from harvest • Baseline oyster biomass must be determined using the appropriate approach and adhere to baseline conditions (Subchapter 7.2.4) • All biomass estimates (Subchapter 7.2.5) must be based on field data collected within 12 months of crediting using a survey design that ensures estimates are representative of the entire BMP site. • Only nutrients associated with eligible appreciated biomass (Subchapter 7.2.7) may be credited.

7.1 Literature Review

By definition, successful oyster reef restoration projects lead to an increase in the abundance and/or biomass of oysters. Numerous studies of oyster reef restoration success in areas where harvesting is not permitted have demonstrated that restoration activities, such as planting oyster shell, hatchery-produced oyster spat-on-shell, and/or alternate substrate, can lead to increases in oyster biomass (e.g., Powers et al. 2009). Within Chesapeake Bay, recent monitoring data from sites in Maryland demonstrate that restoration activities can lead to increases in oyster abundance and biomass (MORIW 2022). Because nitrogen and phosphorus are assimilated into the tissues and shells of oysters, an increase in the biomass per unit area on a reef represents a decrease in the nitrogen and phosphorus available to phytoplankton for growth.

As found for the aquaculture-assimilation protocols (Reichert-Nguyen et al. 2016) and harvest-assimilation protocols (Subchapter 6.1), the Panel's literature review revealed that the nitrogen and phosphorus contents measured in individual *C. virginica* tissue and shell were well constrained in estuaries along the Atlantic Coast of the United States (Appendix E). However, the Panel's review of shell height-to-oyster biomass regressions for sites along the Atlantic and Gulf coasts suggested that relationships were highly variable among sites. Therefore, the Panel decided it was most appropriate to develop default Chesapeake Bay-wide shell height-to-biomass equations based solely on data from the Chesapeake Bay region (Appendix E). Because establishing self-sustaining oyster populations via natural recruitment is almost always one of the goals of oyster reef restoration, the Panel agreed that only data from diploid oysters should be included in the dataset used to develop default tissue and shell nutrient contents for the restoration-assimilation protocols.

7.2 Reduction Effectiveness: Panel Recommendations and Rationale

The following sections identify the information needed to determine reduction effectiveness, outline the Panel's recommendations, and explain the rationale underlying each of those recommendations.

7.2.1 BMP Site Location and Area

As for the harvest-assimilation protocols, the ***BMP site*** is the location in which enhancement activities occur and which is potentially eligible for nitrogen and phosphorus reduction credit if all qualifying conditions are met. The ***BMP site area*** is equal to the area within which enhancement activities occur (i.e., the "footprint" of the restoration project). For additional information on how the BMP site and its area are determined, see Subchapter 6.2.1. Only oysters found within the BMP site are eligible for crediting.

7.2.2 Qualifying Enhancement Activities

The Panel considered the types of enhancement activities that could qualify as a BMP under this practice-protocol combination. In reviewing practices in Chesapeake Bay and elsewhere, the Panel found that a wide variety of materials are used as part of oyster reef restoration efforts. Ultimately the Panel decided that any reef restoration activity that included the addition of hatchery-produced oysters and/or suitable settlement substrate could potentially qualify for crediting under the restoration-assimilation protocol. In this report, ***suitable substrate*** is defined as material on which oysters can settle and survive to adulthood and includes both oyster shell and alternate substrates. To ensure that the majority of the assimilation occurs within the BMP site, the Panel recommends that the shell height of oysters at the time of the restoration activity be <1.0 inch. For oysters larger than 1.0 inch, only incremental growth beyond the planting size can be credited.

7.2.3 Substrate Category

The data available for developing a default regression equation consisted almost entirely of data from projects using oyster shell, spat-on-shell and/or small alternate substrates. Therefore, the Panel decided to categorize substrates for which the default regression should and should not be used. For the purposes of this report, ***alternate substrates*** are all materials not composed solely of oyster shells. Two categories of substrate that are commonly used as part of oyster reef restoration efforts in Chesapeake Bay include:

1. Small, non-engineered substrate (hereafter "***small substrate***"):
   - Suitable substrate characterized by:

        - ≥90% of the material by volume has a maximum diameter of ≤12 inches (304.8 mm) <u>AND</u>
        - a non-uniform or irregular structure when deployed
    - Examples: Oyster shell, spat-on-shell, fossil oyster shell, clam and other types of shell, small concrete, small limestone, small granite or other non-calcium stone, and shell cemented into small irregular clumps.
2. Large or engineered alternate substrate (hereafter "***large substrate***"):
    - Suitable substrate characterized by:
        - <90% of the material by volume has a maximum diameter of ≤12 inches (304.8 mm) <u>OR</u>
        - a uniform, regular structure when deployed.
    - Examples: Large limestone, large granite or other non-calcium stone, and engineered structures including but not limited to ReefBalls™ and Oyster Castles©

The Panel chose to distinguish between large and small substrates because large substrates were not included in the dataset used to develop the default regression equations. The cutoff of 12 inches (304.8 mm) for distinguishing between small and large substrates was chosen to be representative of the maximum size of individual oyster clumps commonly found on reefs restored using oyster shell or spat-on-shell. As seen in Figure E-2 (Appendix E), oysters larger than 6 inches (152.4 mm) in shell height are relatively rare. Thus, two oysters growing in opposite directions outward from the same piece of suitable substrate will rarely result in a clump of oysters larger than 12 inches (304.8 mm) in diameter. The requirement that small structures create non-uniform, irregular structures when deployed was included to address the potential deployment of small uniform structures (e.g., individual Oyster Castles©) in an array that effectively builds a large structure from smaller subunits (e.g., a reef structure composed of many individual Oyster Castles©).

The Panel recommends that implementers restoring reefs with a mix of large and small substrates credit each substrate separately. If a substrate is not eligible for default rates or the default regression, then oysters growing on those substrates would not receive credit unless biomass was measured directly, or a site-specific estimate was generated (Subchapter 7.2.5). For example, an implementer restoring a reef with ReefBalls™ and spat-on-shell would sample each substrate separately to measure appreciated biomass. The biomass of oysters growing as spat-on-shell could be estimated using the Bay-wide default regression. The biomass of oysters growing on ReefBalls™ could be estimated using the direct measurement or site-specific approach.

7.2.4 Baseline Approach

The Panel identified two approaches for determining baseline oyster biomass depending on the oyster biomass data available for the BMP site prior to restoration and when the restoration activities occur relative to the approval of the restoration-assimilation protocols as BMPs. The two approaches are:

1. **Pre-restoration:** Using this approach, live oyster biomass is measured at the BMP site within two years prior to the start of restoration activities. This approach:
    - Must be used for all restoration projects with a start date after approval of the restoration-assimilation protocols as BMPs.
    - Should be used for restoration projects with a start date prior to approval of the restoration-assimilation protocols as BMPs if pre-restoration data are available and of sufficient quality.
2. **Representative site:** Using this approach, a non-restored representative site within the same basin as the BMP site and with conditions similar to those at the BMP site prior to restoration is identified and oyster biomass at the site is surveyed. The selection of the non-restored representative site should include review by expert(s) knowledgeable in reef sampling. This approach:

  - Can only be used for restoration projects with a start date prior to approval of the restoration-assimilation protocols as BMPs for which pre-restoration data either do not exist or are of insufficient quality.
  - Requires that biomass data at the non-restored representative site be collected in the same year and season as the first post-restoration oyster biomass survey at the BMP site.

Regardless of the approach used to determine baseline biomass, all sampling plans should be reviewed and approved by the state reporting agency and the CBP prior to implementation.

7.2.5 Oyster Biomass Assessment

Because nutrient content is based on oyster tissue shell and biomass, accurate assessment of oyster biomass is crucial to accurate assessment of nutrient assimilation. The Panel agreed that all information used to determine baseline and post-restoration biomass must be based on field surveys of oysters per square meter or per large substrate unit, oyster shell heights, and/or direct measurements of oyster shell and tissue biomass per unit area. Because multiple survey and statistical approaches can be used to evaluate oyster populations and biomass on reefs and the best approach can depend on factors such as reef size, patchiness, and sampling gear (patent tongs, dredge, divers), the Panel did not feel it was appropriate to recommend a specific sampling approach. Instead, the Panel recommends that BMP implementers consult with expert(s) knowledgeable in oyster reef sampling and have their baseline and post-restoration sampling designs endorsed by the state reporting agency and the CBP prior to implementation.

Although the Panel did not think it was appropriate to recommend specific sampling designs, they did agree on a set of minimum requirements intended to ensure that biomass surveys collect sufficient data of sufficient quality to accurately assess oyster biomass. These requirements are:

- All biomass estimates must be based on data collected using a survey design that ensures estimates are representative of the entire BMP site.
- If subsampling is used, the methods used need to be documented and must be taken to ensure that each subsample is collected without bias and is extrapolated appropriately.
- A sufficient number of data points must be collected to allow calculation of both mean biomass and its variance. If multiple strata are included in the sampling design (e.g., a stratified random sampling design), data must be sufficient to calculate means and variances for all strata.
- All post-restoration survey data must be collected within 12 months prior to crediting.

After considering methods for determining oyster biomass per unit area based on survey data, the Panel identified three commonly used approaches to determining oyster biomass that are suitable for use for the assimilation-restoration protocols:

- Default regression (small substrate only)
- Direct measurement
- Site-specific regression

The ***default regression*** approach estimates oyster tissue and shell dry weight based on Chesapeake Bay-wide quantile regressions of oyster shell height to oyster tissue and shell dry weight (Appendix E). As such, this regression represents typical conditions across the entire Bay and the full suite of environmental conditions that influence oyster growth. Because all data for developing the default regression equations came from small substrates, the Panel does not recommend use of the default regression equations for large substrates.

The ***direct measurement*** approach does not rely on regressions of oyster shell height to oyster biomass. Instead, for each oyster collected as part of sampling, the tissue and shell are separated, and their dry weights are then measured separately.

The ***site-specific regression*** approach uses samples from the BMP site to develop a site-specific regression of oyster shell height to oyster biomass.

7.2.5.1 Default Regression

The Panel agreed that the approach used to develop default regressions for the aquaculture-assimilation (Reichert-Nguyen et al. 2016) and harvest-assimilation protocols (Chapter 6) could be used to develop default oyster shell height to oyster tissue and shell biomass for the restoration-assimilation protocols. The primary differences between the default regressions developed for the restoration-assimilation protocols and those developed for other protocols is that the data used were taken only from sites the Panel thought to be representative of oyster reef restoration in Chesapeake Bay (e.g., data from triploids and oysters grown in cages were removed) and regressions were developed for both oyster tissue and oyster shell (Appendix E). Because assessments of nutrient removal for the restoration-assimilation protocol are not based on oyster harvest records, the default regression is used with shell height measurements from samples collected during field surveys to determine oyster biomass rather than binning oysters by size class based on harvest records to estimate nutrient removal.

To develop default regressions for oyster shell height to oyster tissue dry weight (g tissue DW) and oyster shell dry weight (g shell DW), the Panel compiled a dataset of oyster shell heights and tissue and shell dry weights from multiple seasons, locations, and habitats across the Chesapeake Bay (Appendix E).

The Panel chose to apply 50th quantile regressions to the compiled datasets. Quantile regression uses the median of the data and is thus less influenced by extremes, making it an appropriate statistical approach to use with highly variable data (such as oyster height-weight relationships). All data and analyses to generate the quantile regression equations are outlined in Appendix E. The resulting default regression equations are presented in Table 7.2.

Table 7.2. Chesapeake Bay-wide quantile regression equations for oyster reef restoration practices, where x = oyster shell height (mm) and y = biomass (g DW).

Parameter	# of Oysters	50th Quantile Regression Equation	Error a	Error b
Tissue	6888	$y = 0.00037x^{1.83359}$	0.00005	0.02896
Shell	4296	$y = 0.00147x^{2.3964}$	0.00035	0.0557

To ensure that default regressions did not result in over crediting, the Panel also evaluated how sensitive the regression equations were to factors that could influence oyster growth and morphology (season, habitat, and salinity regime) (Appendix E). Sensitivity analyses indicated that any variance in the regression fit were within the standard error associated with the full dataset, and that the 50th quantile regression appropriately summarized the relationship between oyster shell height and tissue and shell biomass (Appendix E).

The Panel agreed that the regression equations could be used to estimate oyster biomass for oysters on both subtidal and intertidal reefs but could not be used for oysters growing on large substrates. Although intertidal oysters from only one study were included in the regression analyses, the range of oyster sizes present and the shell height-to-biomass relationships for this intertidal dataset were similar to those in the full dataset.

7.2.5.2 Direct Measurement

The Panel also agreed that direct measurement of oyster tissue and shell dry weights from reef samples can be used. The Panel recommends that implementers use standardized methods to measure the tissue and shell dry weight (Mo & Neilson 1994, Charmichael et al. 2012). The Panel recommends (but does not require) that implementers also measure individual shell heights of sampled oysters as these measurements could be used to develop site-specific shell height-to-dry weight regression equations for future use. The Panel agreed that this approach may be used for both subtidal and intertidal habitats and both large and small substrate categories.

7.2.5.3 Site-specific Regressions

Site-specific oyster shell height to tissue dry weight and oyster shell height to oyster shell dry weight regressions can be developed. If site-specific equations are developed, the Panel recommends that the review and approval of the new equations follow a similar approach as the re-evaluation procedure of existing estimates described in the CBP Partnership BMP Expert Review Protocol (CBP 2015). An expert panel does not need to be re-convened since it is expected that an expert in oyster reef restoration would be consulted during the development of the site-specific estimates. If approved, then the site-specific regression equations can be used to determine the mean live oyster tissue and shell biomass at that specific BMP site assuming conditions at the site remain similar to those present when data were collected for development of the regression equations. The Panel agreed that this approach may be used for both subtidal and intertidal habitats and both large and small substrate categories.

7.2.6 Extrapolating Oyster Biomass from Samples to BMP Site Area

In considering how assessment of oyster biomass differs between large and small substrates, the Panel identified differences in how biomass surveys should be designed and implemented and in how the resulting data should be extrapolated to the scale of the entire BMP site. Specifically, the Panel recommends that:

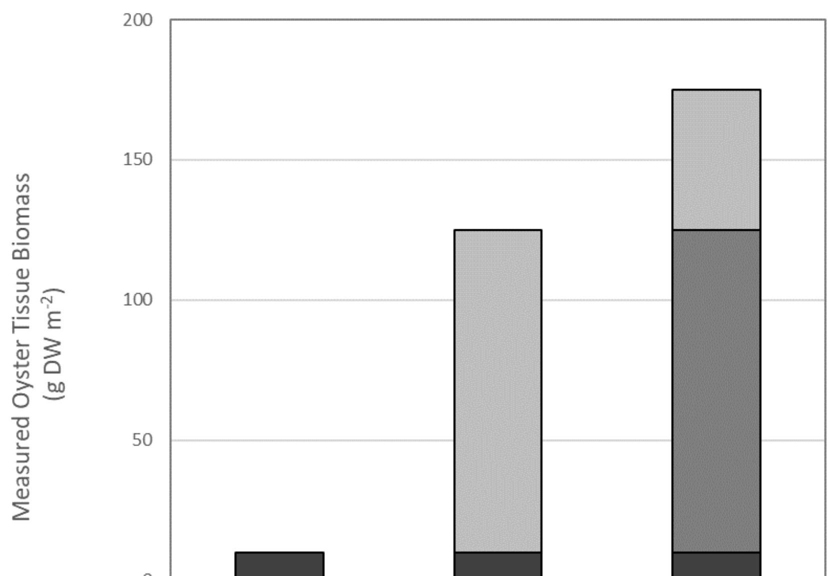
- For restoration using small substrate, oyster biomass should be assessed per unit area of substratum.
    - In general, this involves assessing all oysters within a standardized, defined area of substratum at each sampling point.
    - Examples of this type of sampling include diver quadrat surveys and patent tong surveys.
- For restoration using large substrate, oyster biomass should be assessed per structure.
    - In general, this involves either assessing all oysters on a structural unit or, for larger substrates, subsampling each structural unit and using those data to estimate the biomass of oysters per structural unit. If using subsamples, careful sampling design is needed to ensure that the subsamples are representative of the structural unit as a whole.
    - Examples of these types of surveys include assessing all oysters on an individual Oyster Castle© or divers using small quadrats to subsample the surface of a ReefBall™.

Again, because multiple survey and statistical approaches can be used to evaluate oyster populations and survey designs are often highly site-specific, the Panel did not feel it was appropriate to recommend a specific sampling or extrapolation approaches. Instead, the Panel recommends that BMP implementers consult with expert(s) knowledgeable in oyster reef sampling and have their baseline and post-restoration sampling designs and proposed means of extrapolation to the scale of the BMP site endorsed by the state reporting agency and the CBP prior to implementation. Examples of how extrapolation of measured oyster biomass to the BMP site area can differ between small substrates and large substrates and between different types of large substrates are provided in Subchapter 7.6.

7.2.7 Eligible Appreciated Biomass

A primary goal of most, if not all, oyster reef restoration efforts is to establish a self-sustaining population of oysters on the reef. Over time, individual oysters on the reef will die and new oysters will recruit. Although some nutrients are returned to the water column when an individual oysters dies, there is a net removal of nutrients at the scale of the reef as long as the live oyster biomass per unit area on the restored reef is greater than that prior to restoration, and as long as the amount of biomass per unit area is either stable or increasing. By crediting only increases in the amount of live oyster biomass and not crediting nutrients retained in the shell of dead oysters, the Panel chose an intentionally conservative approach to crediting nutrient assimilation associated with restored oyster reefs.

As shown in Figure 7.1, for the first post-restoration biomass measurement, ***eligible appreciated biomass*** is equal to the post-restoration biomass minus the baseline (i.e., pre-restoration) biomass and is equivalent to biomass enhancement resulting from the restoration activity. For subsequent time periods, eligible appreciated biomass is the amount by which biomass has increased beyond the maximum biomass previously measured at the site. If measured oyster biomass does not exceed previously credited biomass, no credit is given. Eligible appreciated biomass can be measured and credited over the lifetime of the BMP site.



	T0 (Baseline)	T1	T2
Eligible Appreciated		115	50
Non-eligible Appreciated		0	115
Baseline	10	10	10

Figure 7.1. Generalized example of tissue biomass crediting for the restoration-assimilation protocols. T0 represents the measured baseline biomass and T1 and T2 represent the measured biomass based on the first and second post-restoration biomass surveys, respectively. Note that biomass credited based on T1 surveys is no longer eligible for credit at T2.

Because different oyster reef restoration projects will have differing goals and resources available, the Panel is not recommending a specific timeline for measuring biomass. However, to ensure that measured biomass accurately reflects the condition of the BMP site at the time at crediting, the Panel is recommending that biomass surveys used for determining crediting be conducted within the 12 months prior to the time of crediting. Although not required, if the implementing program also plans to seek restoration-denitrification credits (Chapter 8.0), the Panel suggests that post-restoration biomass monitoring every three years would allow the same data to be used to support crediting under both the restoration-assimilation and the restoration-denitrification protocols.

7.2.8 Converting Eligible Appreciated Biomass to Assimilated Nutrients

As for the aquaculture-assimilation and the harvest-assimilation protocols, the Panel agreed that oyster tissue and oyster shell biomass can be converted to amounts of nutrients by multiplying by the appropriate percent content of nutrients. For oyster tissue, the Panel agreed that the diploid oyster tissue nutrient percentages developed previously and already approved by the CBP Partnership could be used for the restoration-assimilation protocols (see Reichert-Nguyen et al. 2016 for details and supporting data). The recommended percentages for use in converting oyster tissue biomass to nutrient content for the restoration-assimilation protocols are:

Mean Percent Nitrogen Content in Oyster Tissue Dry Weight = 8.2%

Mean Percent Phosphorus Content in Oyster Tissue Dry Weight= 0.9%

The percent nitrogen and phosphorus content in oyster shell for use in BMP crediting was not established in the first report (Reichert-Nguyen et al. 2016) but the Panel agreed that the same approach could be used for determining appropriate percentages for converting oyster shell biomass to oyster shell nutrient content. The Panel compiled and reviewed available relevant literature (Appendix E). Nitrogen data included two studies (three sites) from within Chesapeake Bay and three studies conducted outside Chesapeake Bay. Although studies from outside Chesapeake Bay were more variable than those from within the Bay, the mean for all studies and for Chesapeake Bay studies were the same (0.20%). Phosphorus data came from two studies (three sites) in Chesapeake Bay and all reported values were the same (0.04%). Thus, the recommended percentages for use in converting oyster shell biomass to nutrient content for the restoration-assimilation protocols are:

Mean Percent Nitrogen Content in Oyster Shell Dry Weight = 0.2%

Mean Percent Phosphorus Content in Oyster Shell Dry Weight = 0.04%

7.3 Reduction Effectiveness: Stepwise Determination

To calculate the reduction effectiveness for the restoration-assimilation protocols, the Panel recommends the following:

Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 7.2.1).

Step 2: Document the qualifying enhancement activity and its date (Subchapter 7.2.2), the type(s) of substrate used for restoration (Subchapter 7.2.3), and the baseline approach (Subchapter 7.2.4).

Step 3: Assess baseline and post-restoration tissue and shell biomass (Subchapters 7.2.5) and extrapolate it to determine total tissue and shell biomass estimates for the BMP site (Subchapter 7.2.6).

Step 4: Determine the eligible appreciated tissue and shell biomass at the BMP site (Subchapter 7.2.7).

Step 5: Convert eligible appreciated tissue and shell biomass to total nitrogen and phosphorus removed (Subchapter 7.2.8).

7.4 TMDL Baseline Considerations

The TMDL for Chesapeake Bay was created in 2009. Based on this, the CBP Management Board agreed that reduction crediting under the nitrogen and phosphorus restoration-assimilation protocols for oysters can only be given for oyster reef restoration projects that were initiated after the creation of the TMDL in 2009 (CBP 2016).

The Panel coordinated with the WTWG to devise a strategy to credit nitrogen and phosphorus reduction estimated from field data using the current TMDL modeling tools. The WTWG plans to credit appreciated nitrogen and phosphorus in live oyster tissue and shell when monitored oyster biomass demonstrates an increase that hasn't been credited previously. Given potential differences in the BMP and oyster monitoring schedules, application for the credit may or may not occur within the same calendar or fiscal year that the data were collected. To address this discrepancy in timeline, the Panel recommends that application for credit be accepted on a rolling basis, as long as data has been collected within the previous 12 months (Subchapter 7.2.2). No appreciation of live oyster biomass or a lapse in monitoring would result in no reduction credit.

7.5 Qualifying Conditions

The Panel recommends the following qualifying conditions, which account for both the CBP Management Board's defined baseline and the Panel's criteria:

- A qualifying enhancement activity (Subchapter 7.2.2) must have occurred throughout the BMP site area (Subchapter 7.2.1).
- BMP site must lie within an area protected from harvest
- At the time of planting, the shell height of any hatchery-produced oysters should be <1.0 inch (<25.4 mm; Subchapter 7.2.2). For oysters larger than 1.0 inch, only incremental growth beyond the planting size can be credited.
- Baseline oyster biomass must be determined using an appropriate approach and adhere to baseline conditions (Subchapter 7.2.4)
    - For projects using the representative site approach for determining baseline oyster biomass (Subchapter 7.2.4):
        - The representative site must be within the same basin as the BMP site and be representative of conditions at the BMP site before restoration occurred.
        - Data from a non-restored representative site must be collected within the same year and season as the first post-restoration biomass measurement at the BMP site.
    - For projects using the pre-restoration approach for determining baseline oyster biomass (Subchapter 7.2.4):
        - Pre-restoration biomass data must have been collected within two years prior to the start of restoration.
        - For baseline surveys using the pre-restoration approach and for all post-restoration surveys, all data used to estimate oyster biomass must be collected from within the BMP site.
- All biomass estimates (Subchapter 7.2.5) must:
    - Be based on field surveys of live *Crassostrea virginica*

  - Be based on data collected using a survey design that ensures estimates are representative of the entire BMP site.
  - Include enough data points to allow calculation of mean biomass and its variance. If multiple strata are included in the sampling design, data must be sufficient to calculate means and variances for all strata.
  - Be collected within 12 months prior to application for crediting.
- Biomass must be extrapolated appropriately to the scale of the BMP site (Subchapter 7.2.6)
- Only nutrients associated with eligible appreciated biomass (Subchapter 7.2.7) may be credited.

7.6 Recommended Application and Verification Guidelines

Prior to restoration activities, the Panel recommends that applicants submit a sampling plan that includes a detailed oyster biomass survey design for both baseline and post-restoration surveys. All survey designs should be developed in collaboration with experts knowledgeable in oyster reef sampling and should be endorsed by both the State reporting agency and the CBP. At a minimum, the sampling plan should include:

- A description of the type of survey design to be implemented (e.g., random, stratified random) and the rationale for the chosen survey design
- The number of samples to be collected per unit area and the rationale for that number.
- A map and GPS coordinates for proposed sampling points.

The reporting guidelines and examples below assume that a sampling plan has already been submitted and approved and that no significant deviations are made from the approved sampling plan. If significant changes are made after initial approval of the sampling plan, a revised plan should be submitted and approved prior to sampling.

7.6.1 Reporting Guidelines

To assist with application of its recommendations, the Panel developed guidelines for the information to be reported by anyone seeking credit for this practice-protocol combination. The required information is listed below under the associated determination step.

Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 7.2.1)

- Geospatial information documenting the vertices of a polygon representing the BMP site
- Area of the polygon representing the BMP site

Step 2: Document the qualifying enhancement activity and its date (Subchapter 7.2.2), the type(s) of substrate used for restoration (Subchapter 7.2.3), and the baseline approach (Subchapter 7.2.4).

- Date of enhancement activity
- Type(s) of substrate
- Substrate category (small or large)
- Amount of substrate used
- If using hatchery-produced oysters:
  - Ploidy of oysters
  - Number of oysters planted
  - Size of oysters at time of planting
- Baseline approach used (pre-restoration or representative site)

Step 3: Assess baseline and post-restoration biomass (Subchapters 7.2.5) and extrapolate it to determine total biomass estimates for the BMP site (Subchapter 7.2.6).

- For first survey after reef restoration, provide a brief description of biomass survey sampling design for both the baseline and post-restoration biomass surveys. For subsequent surveys, only information on the post-restoration survey design is needed.
  - Sampling date(s)
  - Method used to collect samples (e.g., patent tongs, divers with quadrats, etc.)
  - Spatial scale of sample (e.g., 1.0 m^2, one Reef Ball™, etc.)
  - Number of samples collected
  - If subsampling is used, a description of the subsampling methods, number of subsamples per sample, and method of scaling
  - Methods used to assess oyster tissue and shell biomass per sample (default regression, direct measurement, or site-specific regression)
  - Method used to calculate mean sample biomass
  - Method used to extrapolate mean sample biomass to total biomass per unit area for the BMP site.
  - Mean tissue and shell biomass

Step 4: Determine the eligible appreciated biomass at the BMP site (Subchapter 7.2.7).

- Appreciated tissue and shell biomass

Step 5: Convert eligible appreciated biomass to total nitrogen and phosphorus removed (Subchapter 7.2.8).

- Total nitrogen in eligible appreciated biomass
- Total phosphorus in eligible appreciated biomass

7.6.2 Application Examples

This subchapter provides three hypothetical examples to demonstrate the information and calculations needed to estimate nutrient removal using the restoration-assimilation protocols. The first example focuses on a restoration effort that uses small substrate, the second example focuses on a restoration effort that uses individual large substrate units distributed throughout the BMP site, and the final example focuses on a restoration effort in which individual structural units are used to construct larger substrate units that are distributed throughout the restoration site.

7.6.2.1 Example #1: Small substrate

The following example assumes a restoration project in Maryland which restores one acre of reef using spat-on-shell planted directly on the bottom. After initial planting, oyster biomass surveys were conducted every three years for 12 years using a random sampling design. Samples were collected using patent tongs that sampled a 1m^2 area of the substratum per sample. Table 7.3 provides a list of the information that needs to be reported for this scenario along with data submitted after the first post-restoration survey. For subsequent surveys, only the post-restoration biomass needs to be reported assuming previous biomass measurements are already on record.

<u>Steps 1 and 2:</u> These steps simply record information about the restoration project. The example presented here is a simple one. If more than one type of material is used, all types of materials, amounts and their categories should be recorded. If the BMP site is restored in sections and different methods and/or planting

densities are used for different sections, this needs to be accounted for in the sampling design (e.g., use of a stratified random design).

Step 3: This step uses data from oyster biomass surveys to estimate oyster tissue and shell biomass for the entire BMP site for both the baseline and post-restoration surveys. In the example in Table 7.4, oyster biomass is estimated based on oyster shell height using the default regression equations given in Table 7.2.

Once the total sample biomass for each sampling point has been calculated, these data are used to calculate the mean biomass per unit area for the BMP site by taking their average to get the mean biomass for all samples collected (Table 7.5). Note that if the area sampled is not equal to 1 m^2 then this will need to be accounted for in converting biomass per sample to biomass per unit area. To calculate the total pounds of biomass at the BMP site, the mean biomass per unit area (g DW m^{-2}) is multiplied by the total area of the BMP site (m^2) and divided by 453.59 to convert from grams to pounds dry weight.

Step 4: This step calculates the post-restoration biomass that is eligible for crediting. As shown in Table 7.6 and Figure 7.2, this is accomplished by subtracting the baseline biomass and the previously credited biomass from the measured post-restoration biomass for the BMP site. Calculations are shown for all four post-restoration surveys at the site. In cases where calculations result in negative values, a zero is entered because there is no biomass eligible for credit for that sampling period.

Step 5: This step consists of taking the eligible appreciated biomass and converting to pounds of nitrogen and phosphorus. This is done by multiplying the tissue and shell dry weights by the appropriate percent nutrient content (tissue = 8.2% N and 0.9% P; shell = 0.2% N and 0.04% P). As shown in Table 7.7, the total removal for each nutrient is the sum of the amount in oyster tissue and in oyster shell.

Table 7.3. Information types required for the restoration-assimilation protocols along with an example of each. See text for details of calculations used to provide example information below.

Step #	Information Type	Example
1	BMP site location	See appended map and GIS file
	Area of the BMP site	1 acre
2	Date(s) of activity (mm/dd/yy)	09/21/21
	Type(s) of substrate	Diploid spat-on-shell
	Substrate category	Small
	Amount of substrate	1,000 Maryland bushels of spat-on-shell
	Number of hatchery-produced oysters planted	9,500,000
	Size of oysters at time of planting (mm)	10
	Baseline approach	Pre-restoration
3	Baseline biomass	
	Sampling points	See appended map and GIS file
	Sampling date(s)	07/15/20
	Sampling method	Patent tong
	Spatial scale of sample with units	1 m^2
	Number of samples collected	5
	Method used to assess biomass	Default regression
	Method used to calculate mean biomass	Average of all samples
	Mean biomass: Tissue	14 g DW m^{-2}
	Mean biomass: Shell	631 g DW m^{-2}
	Method for extrapolating to entire BMP site	Multiply by total m^2 and convert to lbs
	Total biomass for the BMP site: Tissue	125 lbs DW
	Total biomass for the BMP site: Shell	5,630 lbs DW
	Post-restoration biomass	
	Sampling date(s)	08/01/24
	Sampling method	Patent tong
	Spatial scale of sample with units	1 m^2
	Number of samples collected	5
	Method used to assess biomass	Default regression
	Method used to calculate mean biomass	Average of all samples
	Mean biomass: Tissue	119 g DW m^{-2}
	Mean biomass: Shell	5,960 g DW m^{-2}
	Extrapolation method	Multiply by total m^2 and convert to lbs
	Total biomass for the BMP site: Tissue	1,062 lbs DW
	Total biomass for the BMP site: Shell	53,178 lbs DW
4	Appreciated biomass: Tissue	937 lbs DW
	Appreciated biomass: Shell	47,548 lbs DW
	Non-eligible appreciated biomass: Tissue	0 lbs DW
	Non-eligible appreciated biomass: Shell	0 lbs DW
	Eligible appreciated biomass: Tissue	937 lbs DW
	Eligible appreciated biomass: Shell	47,548 lbs DW
5	Total nitrogen: Tissue	76.82 lbs N
	Total nitrogen: Shell	95.10 lbs N
	Total creditable nitrogen at BMP site	***171.91 lbs N***
	Total phosphorus: Tissue	8.43 lbs P
	Total phosphorus: Shell	19.02 lbs P
	Total creditable phosphorus at BMP site	***27.45 lbs P***

Table 7.4. Example of data collected for a single survey sample point. Oyster shell heights are converted to oyster tissue and shell biomass using the equations given in Table 7.2. Total sample biomass is the sum of the biomass of all individual oysters in the sample.

					Oyster Biomass		**Total Sample Biomass**	
Survey Type	**Year**	**Sampling Point ID**	**Oyster ID**	**Shell Height (mm)**	**Tissue (g DW)**	**Shell (g DW)**	**Tissue (g DW)**	**Shell (g DW)**
Baseline	2020	B1	1	97	1.63	84.80	17.66	933.08
Baseline	2020	B1	2	101	1.75	93.43		
Baseline	2020	B1	3	62	0.72	29.01		
Baseline	2020	B1	4	87	1.33	65.34		
Baseline	2020	B1	5	101	1.75	93.43		
Baseline	2020	B1	6	89	1.39	69.00		
Baseline	2020	B1	7	126	2.63	158.72		
Baseline	2020	B1	8	108	1.98	109.70		
Baseline	2020	B1	9	81	1.17	55.06		
Baseline	2020	B1	10	103	1.82	97.92		
Baseline	2020	B1	11	93	1.51	76.66		

Table 7.5. Example of baseline data collected for a BMP site and the resulting mean biomass per unit area and total BMP site biomass. Total BMP Site Biomass = Mean Biomass per Unit Area *x* total BMP site area (1 acre in this example). See text for details of calculations.

				Total Sample Biomass		**Mean Biomass per Unit Area**		**Total BMP Site Biomass**	
Survey type	**Year**	**Sampling Point ID**	**# of Oysters**	**Tissue (g DW)**	**Shell (g DW)**	**Tissue (g DW m^{-2})**	**Shell (g DW m^{-2})**	**Tissue (lbs DW)**	**Shell (lbs DW)**
Baseline	2020	B1	11	18	933	14	631	125	5,630
Baseline	2020	B2	13	15	695				
Baseline	2020	B3	8	3	102				
Baseline	2020	B4	25	19	783				
Baseline	2020	B5	16	15	642				

Table 7.6. Example of post-restoration survey data for oyster tissue biomass for a 12-year period. "Total Appreciated Biomass" is the portion of the total biomass that represents an increase over the baseline biomass level. See text for details of calculations. "Appreciated – Not Eligible" is the portion of the appreciated biomass that is not eligible for crediting because it has previously been credited. "Appreciated – Eligible" is the portion of the appreciated biomass that is eligible for crediting and represents an increase over the maximum previously recorded biomass for the BMP site.

Post-Restoration Survey	**1st**		**2nd**		**3rd**		**4th**	
Year	**2024**		**2027**		**2030**		**2033**	
Biomass Component	**Tissue (lbs)**	**Shell (lbs)**	**Tissue (lbs)**	**Shell (lbs)**	**Tissue (lbs)**	**Shell (lbs)**	**Tissue (lbs)**	**Shell (lbs)**
Measured Post-restoration Biomass	1,062	53,178	940	48,858	1,542	94,903	528	28,234
Baseline Biomass (2020)	125	5,630	125	5,630	125	5,630	125	5,630
Total Appreciated Biomass	937	47,548	815	43,228	1,417	89,273	403	22,604
Appreciated - Not Eligible	0	0	815	43,228	937	47,548	403	22,604
Appreciated - Eligible	***937***	***47,548***	***0***	***0***	***480***	***41,725***	***0***	***0***

Figure 7.2. Graph of oyster tissue biomass crediting for example given in Table 7.6

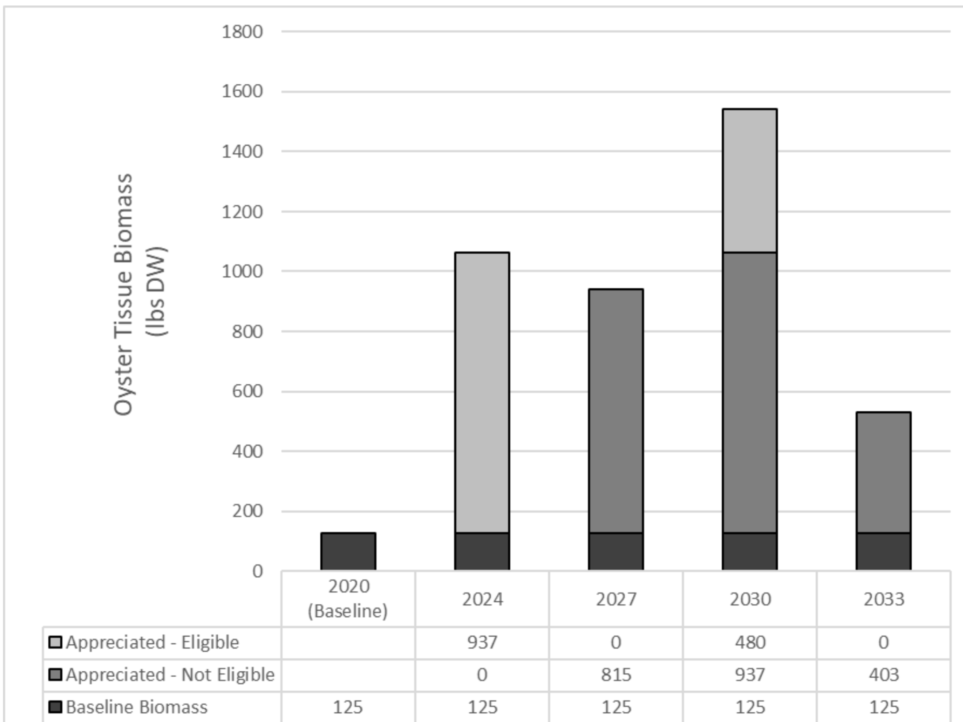


Table 7.7. Example of conversion of eligible biomass to nitrogen and phosphorus credits generated over a 12-year period.

Post-Restoration Survey	1st		2nd		3rd		4th		
Year	2024		2027		2030		2033		
Biomass Component	Tissue (lbs)	Shell (lbs)	Tissue (lbs)	Shell (lbs)	Tissue (lbs)	Shell (lbs)	Tissue (lbs)	Shell (lbs)	Total (lbs)
Appreciated - Eligible Biomass	937	47,548	0	0	480	41,725	0	0	
Creditable Nitrogen	77	95	0	0	39	83	0	0	295
Creditable Phosphorus	8	19	0	0	4	17	0	0	48

7.6.2.2 Example #2: Individual large substrate units

As noted in Section 7.2.5, restoration using large substrates requires a different approach to surveying and extrapolating oyster biomass to the scale of the BMP site. This example assumes that restoration practitioners deploy 25 Reef Balls™ (one Reef Ball™ = one large structural unit) evenly distributed across a 0.5-acre area. Because only the oysters growing on the Reef Balls™ will be considered for crediting and there are no oysters growing on the structures at the time of deployment, the baseline biomass for this restoration project is zero. After consulting with experts, the survey design implemented consists of randomly selecting five structural units, counting and measuring all oysters on the inside and outside of each, and collecting a subsample of oysters from each unit to be used in developing a site-specific shell height to biomass regression. When selecting oysters for sampling, care is taken to capture the full range of oyster sizes found on the units. Table 7.8 shows some of the required data that differ from those for the small substrate restoration project data provided in Table 7.3. Once total post-restoration biomass for the BMP site has been calculated, all other steps are the same as those for restoration projects using small substrates (Subchapter 7.6.2.1)

Table 7.8. Example of how some of data reported for large substrates differs from that for small substrates shown in Table 7.3.

Step #	Information Type	Example
2	Type(s) of substrate	Reef Balls™
	Substrate category	Large
	Amount of substrate	25 individual Reef Balls™
	Number of hatchery-produced oysters planted	0
	Baseline approach	Pre-restoration
3	Baseline biomass: Tissue	0 lbs DW
	Baseline biomass: Shell	0 lbs DW
	Post-restoration biomass	
	Sampling method	Retrieve randomly selected units for assessment
	Spatial scale of sample with units	1 Reef Ball™
	Number of samples collected	5
	Percent of each structural unit sampled	100%
	Method used to assess biomass	Site-specific regression
	Method used to calculate mean biomass	Average of all samples
	Mean biomass: Tissue	12 g DW structural unit^{-1}
	Mean biomass: Shell	52 g DW structural unit^{-1}
	Extrapolation method	Multiply by total # of structural units and convert to lbs
	Total biomass for the BMP site: Tissue	0.66 lbs DW
	Total biomass for the BMP site: Shell	2.87 lbs DW

7.6.2.3 Example #3: Individual structural units used to build larger structural units

This example assumes that restoration practitioners build four Oyster Castle® reefs on a BMP site. Each reef is constructed using 14 individual blocks and, by the time of post-restoration sampling, the blocks have become cemented together with oysters, requiring that each reef be sampled in place. As can be seen in Figure 7.3, some blocks are more accessible for oyster recruitment and biomass sampling than others. This needs to be accounted for in the sampling design and in the extrapolation of the data collected. One approach to sampling these reefs would be to use the interior and exterior of an exposed side (i.e., a side of the block that is on the outermost surface of the reef) as the sampling unit (equal to $1/4^{th}$ of one individual block). Once the biomass data are collected and biomass is calculated as a biomass per exposed side, this number needs to be scaled up to the biomass per reef and then multiplied by the four reefs included in the restoration effort. The simplest way to calculate biomass per reef would be to multiply the mean biomass per exposed side by the total number of exposed sides. In this case, there are four exposed sides on the top layer, eight exposed sides on the middle layer, and 12 exposed sides on the bottom layer for a total of 24 exposed sides. However, this is not the only approach that could be used. The potential complexity of the sampling design and extrapolation of data collected from these types of structures highlights the need to consult with experts in oyster reef sampling to develop a sampling plan and appropriately extrapolate the resulting data to the scale of the BMP site.

Figure 7.3. Example of a large structural unit composed of smaller units. Photo courtesy of P. Kingsley-Smith, South Carolina DNR.

Appreciated oyster biomass and the resulting BMP credits for restoration projects using other large substrates could be assessed using either, or a combination of, the approaches described in Subchapter 7.6.2.2 and 7.6.2.3. The most appropriate sampling approach will depend on the size and configuration of the reef materials. The Panel recommends that BMP implementers consult with expert(s) knowledgeable in oyster reef sampling and have their sampling designs endorsed by the state reporting agency and the CBP prior to implementation.

7.7 Unintended Consequences

There was one unintended consequence identified for the restoration-assimilation protocols. Since credits are given for permanent removal of nutrients, it is possible for over crediting to occur if biomass drops below the previous level of post-restoration biomass for which the BMP area was given nutrient reduction credit. For example, if a monitored reef accrues biomass between years 1 and 2 but then loses biomass between years 2 and 3, the permanent removal in year 3 is less than that which was credited during year 2. However, because both the shells of dead oysters on the reef and the tissues and shells of other reef organism (e.g. mussels, barnacles, etc.) contain significant amounts of nitrogen and phosphorus but are not eligible for credit, the Panel agreed that the risk of attributing more nitrogen and phosphorus to reef restoration than actually occurs was minimal.

7.8 Ancillary Benefits

The Panel identified several ancillary benefits for the restoration-assimilation protocols. Benefits of oyster restoration are described in Chapter 9.

7.9 Future Research

The Panel identified the following research gaps while reviewing the literature to develop the restoration-assimilation BMP recommendations:

- More research is needed on factors that influence spat survivorship and changes in oyster biomass over time. This could lead to the development of predictive relationships to estimate oyster biomass and reduce the need for field surveys.
- More data from intertidal reefs are needed to determine whether separate default shell height to dry weight regressions are warranted for intertidal reefs.
- Additional research is needed to investigate how best to estimate oyster biomass on large substrates and engineered structures. Specifically, measurements of the oyster dry weights and nutrient contents for oysters growing on engineered structures need to be taken to determine whether established relationships hold or whether new equations must be generated.

8.0 Recommendations on Reduction Effectiveness of Nitrogen Removed by Enhanced Denitrification Associated with Oysters on Restored Oyster Reefs

Live oysters and associated animal and bacterial communities on oyster reefs can enhance denitrification at the reef scale. Denitrification is an anaerobic process, usually facilitated by bacteria, that reduces oxidized forms of dissolved inorganic nitrogen, such as dissolved nitrate or nitrite, to nitrogen gas (N_2). This process converts bioavailable forms of nitrogen that can fuel phytoplankton growth in estuaries into N_2 that does not. High denitrification rates in aquatic habitats generally require a supply of organic material to aerobic benthic habitats that support communities of nitrifying bacteria adjacent to anoxic habitats that support communities of denitrifying bacteria. On oyster reefs, the filter-feeding activities of oysters and associated reef organisms supply reactive organic matter to the reef surface in the form of biodeposits (i.e., feces and pseudofeces). The complex three-dimensional structure of oyster reefs provides abundant surface area for the growth of nitrifying bacteria. Oyster reefs also provide an abundance of habitats for the growth of denitrifying bacteria both within anoxic areas found within the reef matrix and in reef sediments. Therefore, increased oyster tissue biomass from reef restoration activities can enhance the deposition of organic matter and the amount of suitable habitat for nitrifying and denitrifying bacteria and result in enhanced denitrification rates.

In its first report (Reichert-Nguyen et al. 2016), the Panel identified three practice-protocol combinations that could remove nitrogen through enhanced denitrification associated with oysters on restored reefs (Table 1.1). This chapter describes the Panel's recommendations on the following two restoration-denitrification protocols seeking BMP approval:

Practice J. Oyster reef restoration using hatchery-produced oysters

Protocol 3. Enhanced denitrification associated with oysters

Practice K. Oyster reef restoration using substrate addition

Protocol 3. Enhanced denitrification associated with oysters

Hereafter, these practice-protocol combinations are referred to collectively as "restoration-denitrification" protocols.

The Panel did not reach consensus on whether to endorse **Practice L** (oyster reef restoration using no-harvest area designation only); however, they agreed that the recommendations presented in this chapter could be applied to this practice if endorsed later for future BMP use. The Panel also decided that existing data were insufficient to determine the nitrogen reduction effectiveness of enhanced denitrification for oyster practices where harvesting occurs (Appendix I).

The Panel recommends that the nitrogen reduction effectiveness of the restoration-denitrification protocols be based on the oyster **tissue** biomass at the BMP site in combination with seasonal patterns in reef denitrification rates. Enhanced denitrification estimates are calculated by subtracting the baseline denitrification rate (determined based on baseline oyster tissue biomass) from the post-restoration denitrification rate (determined based on post-restoration oyster tissue biomass). For reef restoration projects using small substrate in subtidal habitats, the Panel developed a lookup table (Subchapter 8.2.5.1 and Appendix G) that provides annual enhanced denitrification estimates for expected ranges of baseline and post-restoration oyster tissue biomass. Because the data required to develop robust estimates for reef restoration projects in the intertidal and/or using large substrates are not currently available, the Panel recommends site-specific measurements be required for crediting of these projects.

A summary of the Panel's recommended reduction effectiveness determination and qualifying conditions for the restoration-denitrification protocols is outlined in Table 8.1. The literature and data reviewed by the Panel to inform development of their recommendations are documented in Subchapter 8.1. The Panel's approach and recommended steps for determining nitrogen reduction effectiveness by enhanced denitrification are outlined in Subchapter 8.2 and Subchapter 8.3. Enhanced denitrification is a form of permanent nitrogen removal when several qualifying conditions described in Subchapter 8.5 are met. Verification guidelines and recommended applications are described in Subchapter 8.6. Unintended consequences (Subchapter 8.7), ancillary benefits (Subchapter 8.8), and recommendations for future research (Subchapter 8.9) are also discussed for these practice-protocol combinations and reduction effectiveness approach.

Table 8.1. Summary of the nitrogen reduction effectiveness strategy for the restoration-denitrification protocols.

Fisheries Management Approach	Oyster Reef Restoration
Oyster Practice	**Practice J:** Oyster reef restoration using hatchery-produced oysters **Practice K**: Oyster reef restoration using substrate addition
Practice Definitions	**Practice J:** Planting oysters (e.g., spat-on-shell, single oysters) produced from hatchery techniques directly on the bottom or on suitable substrate to enhance oyster biomass in areas where removal is not permitted. **Practice K:** Planting oyster shells and/or alternative substrate directly on the bottom to attract recruitment of naturally occurring (wild) oyster larvae to enhance oyster biomass in areas where removal is not permitted.
Protocols	**Protocol 3.** Enhanced Denitrification Associated with Oysters
Abbreviated name for practice-protocols	Restoration-denitrification
Reduction Effectiveness Determination (Subchapter 8.3)	**Step 1.** Identify the BMP site location and determine the BMP site area (Subchapter 8.2.1) **Step 2.** Document the qualifying enhancement activity and the date it occurred (Subchapter 8.2.2) **Step 3**. Determine the appropriate baseline approach (Subchapter 8.2.4) **Step 4**. Assess baseline and post-restoration tissue biomass (Subchapter 8.2.5) **Step 5.** Determine denitrification enhancement per unit area (Subchapter 8.2.6) **Step 6.** Determine the total nitrogen removal attributable to enhanced denitrification using the estimated denitrification enhancement per unit area and the BMP site area (Subchapter 8.2.7)
Qualifying Conditions (Subchapter 8.5)	• A qualifying enhancement activity (Subchapter 8.2.2) must have occurred throughout the BMP site area (Subchapter 8.2.1). • The BMP site area must lie within an area protected from harvest. • If using the default approach to estimating enhanced denitrification, the reef must be in a subtidal habitat and restoration activities must have utilized only small substrates (Subchapter 8.2.2). • Only live oyster tissue biomass is eligible for credit. • The post-restoration oyster tissue biomass must be greater than the baseline oyster tissue biomass.

8.1 Literature and Data Review

For the purposes of BMP consideration, denitrification is best measured as the net flux of nitrogen gas from the benthos to the water column. Because nitrogen gas (N_2) is the most abundant component (78%) of air, accurate measurement of denitrification rates in aquatic environments is challenging. Methods for direct measurement of N_2 flux in oyster-associated environments are a relatively recent development, with the first laboratory study of the impacts of simulated oyster biodeposits on nitrogen cycling published in 2002 (Newell et al. 2002). Since then, methods for assessing N_2 fluxes from oyster-associated environments have continued to evolve.

In their review of the potential for using oysters to mitigate eutrophication in coastal waters, Kellogg et al. (2014c) found that, of the studies conducted to date, there was more evidence to support enhanced denitrification rates associated with oyster reefs than for oyster aquaculture. The Panel reached a similar conclusion through their independent literature review (Subchapter 8.1, Appendix I). Based on this, the Panel agreed that data were insufficient at present to develop reduction effectiveness estimates for enhanced denitrification associated with either oyster aquaculture or harvest of hatchery-produced oysters and focused their efforts on developing reduction effectiveness estimates for enhanced denitrification associated with restored oyster reefs.

The Panel conducted a thorough review of existing peer-reviewed literature to determine the most appropriate approach to (1) quantify the enhanced denitrification and to (2) estimate the nitrogen reduction effectiveness for the restoration-denitrification protocols. Enhanced denitrification was most commonly calculated as the denitrification rate at the reef site minus the denitrification rate at a reference site. Reference sites were nearby but outside the zone of influence of the restoration site (Figure 8.1) and were similar to the restored site in all respects except that the reference sites had very few or no oysters. The Panel chose to use this approach for the restoration-denitrification protocols with the amendment that the reference denitrification rate could also be calculated from the restoration site before restoration occurred.

Some published estimates for enhanced denitrification were based on methods that measure potential denitrification rates rather than actual denitrification rates (e.g., Seitzinger et al. 1993, Joye et al. 1996, Cornwell et al. 1999). The Panel agreed that actual denitrification rates were most appropriate to determine the nitrogen reduction effectiveness of oyster reef restoration practices.

Reported denitrification rates varied widely and the causes of variation were not easy to identify because of differences in the types of samples collected (e.g., some included oysters and others did not), the types of reefs sampled (e.g., subtidal versus intertidal), and the geographic locations of the studies (Table 8.2). After careful consideration of peer-reviewed published data, project reports, and unpublished data, the Panel decided that the most scientifically defensible nitrogen reduction effectiveness estimates for the restoration-denitrification protocols would come from a meta-analysis of data collected in the Chesapeake Bay. The results of the Panel's meta-analysis and the resulting recommendations are described in detail in Subchapter 8.2.6.1.

Figure 8.1. Diagram of restored reef BMP site and the surrounding area potentially affected by biodeposits from the reef. To accurately assess denitrification enhancement by direct measurement of denitrification rates, restored reef samples should be collected from within the BMP site and baseline measurements should be collected outside the area potentially affected by reef biodeposition.

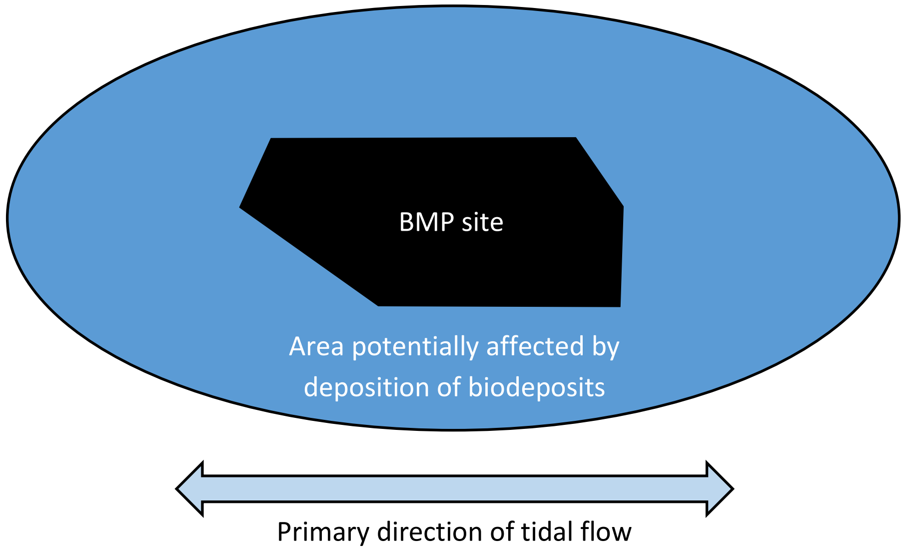


Table 8.2. Maximum denitrification enhancement (reef minus reference) observed in published oyster reef studies. Sediment-only samples contain sediments collected from within or adjacent to oyster reefs and may contain macrofauna but do not include oysters or sessile macrofauna commonly attached to oyster shells. Maximum enhancement is calculated as the greatest difference measured within a season between reef and reference samples. Mean values were used to calculate maximum enhancement unless noted otherwise. "NS" = no significant difference between mean fluxes at reef and reference sites (α = 0.05). DNF = denitrification.

Sample Type	**Reef Type**	**Location**	**Max DNF Enhancement** (µmol N_2-N m^{-2} h^{-1})	**Source**
Reef	Subtidal	Choptank River, MD	1,486	Kellogg et al. (2013)
		Ninigret Pond, RI	~1,100	Humphries et al. (2016)ꟸ
		Harris Creek, MD	~600	Jackson et al. (2018)
Sediment-only	Intertidal	Bogue Sound, NC	~160	Piehler & Smyth (2011)*
			~250	Smyth et al. (2013)
			102	Smyth et al. (2016)
			NS	Onorevole et al. (2018)
		Middle Marsh, NC	~160	Smyth et al. (2015)
		Smith Island Bay, VA	~14	Smyth et al. (2018)
	Subtidal	Lake Fortuna and Sister Lake, LA	NS	Westbrook et al. (2019)
		Great Bay Estuary, NH	~16	Hoellein et al. (2015)

ꟸ Based on median values

*Means not given; calculated by subtracting minimum reference rate from maximum reef rate

8.2 Reduction Effectiveness: Panel Recommendations and Rationale

The following sections identify the information needed to determine reduction effectiveness, outline the Panel's recommendations, and explain the rationale underlying each of those recommendations for the restoration-denitrification protocols.

8.2.1 BMP Site Location and Area

As for the harvest-assimilation and the restoration-assimilation protocols, the ***BMP site*** is the location in which enhancement activities occur and which is potentially eligible for nitrogen reduction credit if all qualifying conditions are met. The ***BMP site area*** is equal to the area within which enhancement activities occur (i.e., the "footprint" of the restoration project). For additional information on how the BMP site and its area are determined, see Subchapter 6.2.1. Only denitrification associated with oysters found within the BMP site are eligible for crediting.

The Panel considered restoration projects in both subtidal and intertidal habitats and found that far more data were available for subtidal habitats. Data from intertidal reefs came from only two areas in Virginia. Variance in these data was high and the sample size was low. The Panel agreed that it was not feasible to develop default relationships between oyster biomass per unit area and denitrification for intertidal reefs at this time.

8.2.2 Qualifying Enhancement Activities

The qualifying enhancement activities for the restoration-denitrification protocols are the same as those for the restoration-assimilation protocols (Subchapter 7.2.2). Specifically, any reef restoration activity that includes the addition of hatchery-produced oysters and/or suitable settlement substrate throughout the BMP site can potentially qualify for crediting. However, the type of substrate used for restoration (large or small) and its location (subtidal or intertidal) will determine the approaches that can be used to estimate denitrification enhancement. Oysters of any size can enhance denitrification, and therefore the Panel does not recommend a maximum size of hatchery-produced oysters at the time of planting.

8.2.3 Substrate Category

The Panel agreed that same substrate categories (large and small) used for the restoration-assimilation protocols should be used for the restoration-denitrification protocols (Subchapter 7.2.3). While reviewing the available data, the Panel found that denitrification rates have rarely been measured for restoration projects using large substrate. The few data that have been collected suggest that rates may be structure-specific (e.g., Reef Balls™ and Oyster Castles® with similar oyster biomass may not have similar denitrification rates; J. Cornwell, UMCES, pers. Comm.). Appendix I includes preliminary data on denitrification rates measured on large substrates. The Panel also noted that the size and distribution of large structures likely influences the extent to which oyster biodeposits are retained within the BMP site (e.g., increasing vertical height and decreasing density of structures per unit area likely decrease retention of biodeposits within the BMP site).

Because of the paucity of data for reef restoration using large structures, the Panel concluded that it was not feasible to develop default denitrification enhancement estimates for large structures at this time. However, the Panel noted that the recommendations presented in this chapter could be applied to reefs restored using large substrates if sufficient data become available to develop robust relationships between denitrification rates and oyster biomass on large substrates, recognizing that these relationships may need to be substrate-specific (e.g., one relationship for Reef Balls™ and a different relationship for Oyster Castles®).

8.2.4 Baseline Approach

The Panel agreed that both the ***pre-restoration*** and the ***representative site*** approaches used for the restoration-assimilation protocols can be used for the restoration-denitrification protocols and that the same conditions and restrictions apply. As for the restoration-assimilation protocols, all sampling plans should be reviewed and approved by the state reporting agency and the CBP prior to implementation regardless of the approach used to determine baseline values. See Subchapter 7.2.4 for a full description of these two baseline approaches.

8.2.5 Oyster Biomass Assessment

The Panel agreed that the same recommendations for assessment of oyster tissue biomass for the restoration-assimilation protocols can be used for the restoration-denitrification protocols and that all the same restrictions and conditions apply for biomass sampling and estimation of biomass per unit area. Because estimates of denitrification enhancement rely solely on oyster tissue dry weight per unit area, data on oyster shell dry weight per unit area are not needed. See Subchapter 7.2.5 for a full description of recommended approaches to oyster tissue biomass assessment.

8.2.6 Denitrification Enhancement per Unit Area

The Panel recommends two approaches for estimating nitrogen removal associated with the restoration-denitrification protocols: default and site-specific. As noted in Subchapters 8.2.1 and 8.2.3, the Panel's review of available data led them to conclude that data are currently insufficient for development of default denitrification estimates for either intertidal reefs or for reefs using large substrate. Therefore, the Panel agreed that the default approach to estimating denitrification enhancement can only be used for reefs restored in subtidal habitats using small substrate (which includes hatchery produced oysters). All other types of restoration must use site-specific estimates of denitrification enhancement. However, the Panel agreed that, if sufficient data become available, the same approach presented in this chapter for developing default relationships between oyster biomass and denitrification enhancement could be applied to restored intertidal oyster reefs and/or reefs using large substrate.

8.2.6.1 Default Approach

An initial review of the literature indicated that peer-reviewed published data from the Chesapeake Bay were sparse, and that denitrification enhancement varied by orders of magnitude among these studies (e.g., Kellogg et al. 2013, Smyth et al. 2018). The Panel noted that, while some studies focused on restored oyster reefs, others were from natural oyster reefs and/or the origin (i.e., natural vs. restored) was not clearly stated. Because there was no clear evidence for differences in denitrification for natural and restored oyster reefs with similar biomass per unit area, the Panel agreed that studies focusing on both natural and restored oyster reefs were suitable for inclusion in the meta-analyses. To increase the pool of data used to determine enhanced denitrification rates related to oyster tissue biomass, the Panel decided to include additional data from project reports and unpublished data that met criteria for inclusion and the Panel's standards for scientific rigor.

To identify data suitable for inclusion in the Panel's meta-analyses, the Panel decided that the studies needed to have:

- Measured denitrification using representative samples from the restored reef that included oysters, shell, sediments, and associated organisms

- Assessed the biomass of oysters within the sample used for denitrification measurements and included a sufficient description of reef characteristics
- Measured denitrification using either the N_2:Ar gas ratio or ^{15}N isotope pairing technique
- Used a batch approach to incubation
- If the study reef occurred in the euphotic zone (where light reaches the seafloor), included both light and dark incubations.

The Panel also preferred studies that reported fluxes of other nitrogen compounds and oxygen but did not exclude data if these measurements were not taken. The rationale for selecting these criteria is described in Appendix F. Many studies excluded from meta-analyses failed to include a representative sample of the oyster reef (e.g., sampling included only sediments collected from an oyster reef). In addition to these types of samples not being representative of the reef-level impacts on denitrification rates, many presented challenges in identifying relationships between oyster biomass per unit area and denitrification rates.

The full set of evaluated studies spanned from mid to lower Chesapeake Bay (Figure 8.2), occurred in all four seasons, and included a wide range of oyster biomass (Table 8.3).

Initial examination of the resulting set of denitrification data for subtidal reefs suggested that, as expected, denitrification rates varied with season, leading the Panel to focus their meta-analyses on identifying seasonal relationships between oyster biomass and denitrification. In examining the distribution of available data across seasons, the Panel found only one study at a single location that sampled denitrification rates during the winter. Based on this, the Panel decided that data were insufficient to estimate winter denitrification rates and that, until more data become available, annual estimates of enhanced denitrification should assume that no denitrification occurs in winter. In total, seven studies from the Chesapeake Bay were used in the meta-analyses to develop season-specific relationships between oyster biomass per unit area and denitrification rates (Table 8.3).

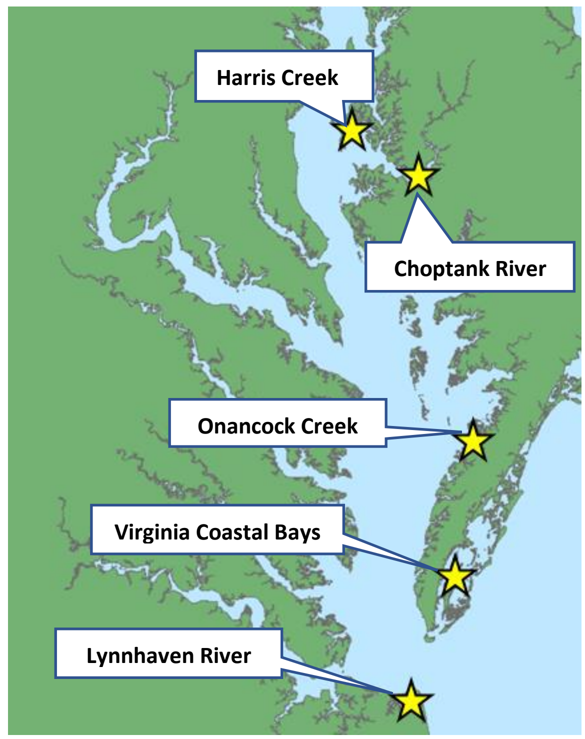


Figure 8.2. Map of Chesapeake Bay showing locations (yellow stars) where oyster reef denitrification rates included in the Panel's meta-analysis were measured. Some of the stars represent multiple sites in close proximity.

Table 8.3 Summary of the data included in analyses to determine if a default rate for enhanced denitrification associated with oyster reef restoration could be generated. Data from intertidal reefs were not included and some data from subtidal reefs were excluded from analyses (see text for details). NA = not applicable.

Reef Setting	Season	State	Water Body	Source	# of Samples Collected	# of Samples Included in Meta-Analyses
Subtidal	Spring	MD	Choptank River	Kellogg et al. (2013)	8	8
			Harris Creek	Cornwell et al. (2016 and 2019)	8	6
		VA	Lynnhaven River	Kellogg et al. (unpublished)	4	3
			Onancock Creek	Kellogg et al. (2014a,b)	6	5
	Summer	MD	Choptank River	Kellogg et al. (2013)	15	12
			Harris Creek	Cornwell et al. (2016 and 2019)	93	84
		VA	Lynnhaven River	Kellogg et al. (unpublished)	8	5
			Onancock Creek	Kellogg et al. (2014a,b)	7	7
	Fall	MD	Choptank River	Kellogg et al. (2013)	8	8
			Harris Creek	Cornwell et al. (2016 and 2019)	26	23
		VA	Lynnhaven River	Kellogg et al. (unpublished)	4	3
				Sisson et al. (2011)	2	2
	Winter	MD	Harris Creek	Cornwell et al. (2016 and 2019)	8	NA
Intertidal	Spring	VA	Virginia Coastal Bays	Kellogg et al. (2014a,b)	7	NA
	Summer	VA	Virginia Coastal Bays	Kellogg et al. (2014a,b)	22	NA
	Fall	VA	Lynnhaven River	Sisson et al. (2011)	6	NA
			Virginia Coastal Bays	Kellogg et al. (2014a,b)	7	NA

The first steps the Panel took in conducting meta-analyses was to normalize all data and screen it for outliers. All studies included in the meta-analyses reported denitrification rates in terms of nitrogen flux per unit area (µmol N m^{-2} h^{-1}). However, some of the studies included in the dataset were conducted in the euphotic zone (i.e., sufficient light reached the substratum to stimulate photosynthesis) while others were conducted below the euphotic zone. Because denitrification rates can be influenced by the presence of light, studies conducted in the euphotic zone measured denitrification rates for the same samples incubated in both the presence and absence of light. Because the number of daylight hours varies between seasons, the data from each study were used to calculate a mean hourly denitrification rate that accounted for the number of daylight hours in each season. For samples from euphotic reefs, there were some instances in which data were successfully collected only under light or only under dark conditions. Because a mean hourly rate could not be calculated for these samples, they were removed from the meta-analyses. This reduced the total number of data points from 26 to 24 for spring, 123 to 111 for summer, and 40 to 36 for fall.

Using the resulting dataset, the Panel then calculated mean hourly denitrification rates for 171 samples. To do this, the Panel first determined the number of daytime and nighttime hours in each season (Table 8.4) using NOAA's Solar Calculator (https://gml.noaa.gov/grad/solcalc/index.html). After considering differences in day length throughout Chesapeake Bay, the Panel decided its impact was minimal and decided to use the day and night hours from the geographic midpoint of the Chesapeake Bay for all locations.

Table 8.4. Number of months, days, hours, daytime hours, and nighttime hours for each season at the midpoint of the Chesapeake Bay.

Season	Months in Season	# Days	# Hours	# Daytime Hours	# Nighttime Hours
Spring	March, April, May	92	2208	1209.8	998.2
Summer	June, July, August	92	2208	1311.3	896.7
Fall	Sept., Oct., Nov.	91	2184	1022.1	1161.9
Winter	Dec., Jan., Feb.	90	2166	909.2	1256.8

The Panel used the resulting daytime and nighttime hours for each season to calculate the mean hourly denitrification (DNF) rate (µmol N_2-N m^{-2} h^{-1}) for each sample included in the meta-analysis:

$$Mean\ Hourly\ DNF\ Rate = \frac{(DNF_L\ x\ h_L) + (DNF_D\ x\ h_D)}{h_T}$$

where: DNF_L = denitrification rate measured in the presence of light
DNF_D = denitrification rate measured in the absence of light
h_L = total number of daytime hours in season
h_D = total number of nighttime hours in season
h_T = total number of hours in season

For samples from sites where light reaching the bottom was insufficient to stimulate photosynthesis, the formula was simplified to:

$$Mean\ Hourly\ DNF\ Rate = DNF_D\ x\ h_T$$

The mean hourly denitrification rates were plotted as a function of oyster tissue biomass density (g DW oyster tissue m^{-2}) for each sample and assessed for outliers. To ensure that denitrification rate estimates based on oyster tissue biomass were conservative, the Panel removed three data points from the spring dataset and three data points from the fall dataset that had unusually high denitrification rates relative to the remainder of these data. After outliers were removed, regression lines were plotted, and the significance of each regression was tested (Figure 8.3).

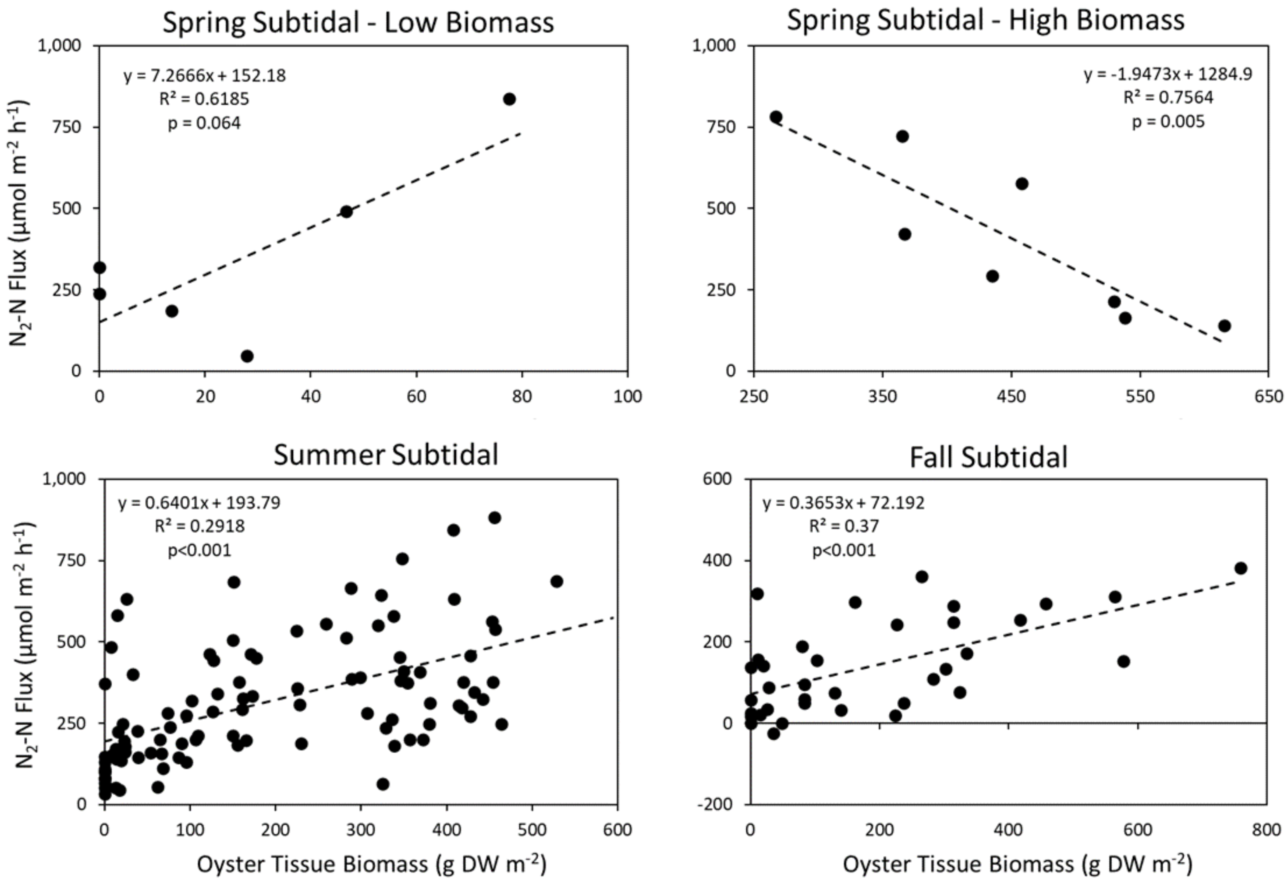


Figure 8.3. Final linear regressions of spring, summer and fall data oyster reef denitrification rates plotted as a function of oyster tissue biomass.

Mean hourly denitrification rates were related to oyster tissue biomass in spring, summer, and fall but that the relationship differed among seasons. In summer and fall, there was a significant ($p < 0.001$) linear increase in denitrification rate with oyster tissue biomass. The pattern for spring was more complicated. For samples with relatively low oyster tissue biomass (≤78 g DW oyster tissue m^{-2}), there was a marginally significant ($p = 0.064$) increase in denitrification rates with oyster tissue biomass. However, for samples with high oyster tissue biomass (≥267 g DW m^{-2}), denitrification decreased significantly ($p = 0.005$) with oyster tissue biomass. The spring dataset included no data for oyster tissue biomass between 78 and 267 g DW m^{-2}. Based on these results, the Panel agreed that default denitrification enhancement estimates based on oyster biomass per unit area could be developed for subtidal oyster reefs in Chesapeake Bay restored using small substrate. Specifically, the Panel agreed that:

- Season-specific linear regressions could be used to estimate denitrification rates based on oyster tissue biomass.
- Summer and fall linear regressions could be used without modification to estimate denitrification rates based on oyster tissue biomass.
- Spring estimates of denitrification rates would be based on two separate linear regressions for reefs with low and high oyster tissue biomass and that values for the region between these two regressions would be derived by connecting the ends of the two regressions with a straight line (a more conservative approach than fitting a polynomial or other type of regression to the gap in data).

- The marginal significance ($p = 0.064$) of the spring low oyster biomass regression was acceptable to estimate denitrification rates for the purposes of this BMP.

The resulting equations are provided in Table 8.5.

Table 8.5. Equations used to estimate seasonal denitrification rates for oyster reef habitats, where x = oyster tissue biomass (g DW m^{-2}) and y = denitrification rate (µmol N_2-N m^{-2} h^{-1}).

Season	Biomass Density	Equation	R^2	p-value
Spring	Low	$y=7.2666x + 152.18$	0.6185	0.064
	Moderate	$y=0.2602x + 695.46$	NA	NA
	High	$y=-1.9473x + 1284.9$	0.7564	0.005
Summer	All	$y=0.6401x + 193.79$	0.2918	<0.001
Fall	All	$y=0.3653x + 72.192$	0.3700	<0.001

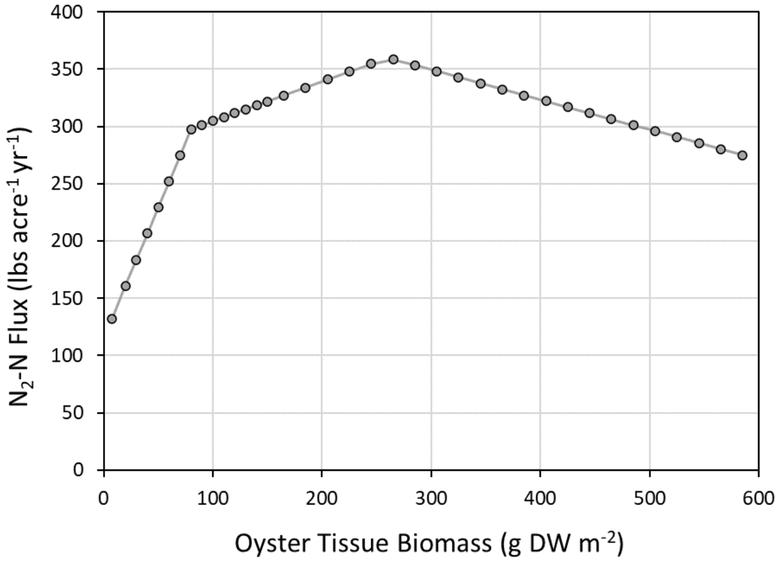
Recognizing the complexities of calculating annual denitrification rates from these regressions, the Panel developed a lookup table that allows implementers to estimate annual enhanced denitrification rates based on baseline and post-restoration oyster tissue biomass. The first step in developing the lookup table was to determine appropriate bins for the levels of oyster biomass per unit area. The Panel decided that the first oyster tissue biomass bin should range from 0-14.9 g DW m^2 because an oyster tissue biomass of 15 g DW m^{-2} is the minimum threshold value for restored oyster reefs to be considered "marginally successful" in Chesapeake Bay (Oyster Metrics Workgroup 2011). For reefs with biomass per unit area ranging from 15-154.9 g DW m^{-2}, the Panel decided to divide oyster tissue biomass into 10 g DW m^{-2} bins based on the following considerations:

- Slope of the curve for each oyster tissue biomass range (denitrification increased more quickly at lower oyster tissue biomass ranges)
- Frequency with which each oyster tissue biomass range was observed in the field (most restoration projects to date have seen oyster tissue biomass range from 15-154.9 g DW m^{-2})
- Number of bins that would be practical for implementation

For sites with oyster tissue biomass ≥155 g DW m^{-2}, the Panel agreed that 20-g bins were the best tradeoff between accurate estimation of denitrification rates and complexity of implementation. Oyster tissue biomass ranges and midpoints were not identified for sites with greater than 594.9 g DW m^{-2} because the regressions did not extend beyond this value for all three seasons and the Panel did not want to extrapolate beyond the range of the data included in analyses. Few, if any, restoration efforts achieve this level of mean oyster tissue biomass.

Once biomass bins were established, the annual denitrification rate for each biomass bin was determined. To do this, the total nitrogen removal per season for each oyster tissue biomass range was calculated by entering the midpoint biomass for each bin into each of the seasonal regression equations (Table 8.5) to get the corresponding denitrification rate per unit area (µmol m^{-2} h^{-1}). This was then multiplied by the total number of hours per season (Table 8.4) to get the total nitrogen removal per season (µmol N_2-N m^{-2}) and the units converted to pounds per acre. The annual denitrification rate (lbs. N $acre^{-1}$ yr^{-1}) was then calculated as the sum of the individual seasonal nitrogen removals.

Figure 8.4. Plot of estimated annual denitrification rates in relation to oyster tissue biomass per unit area for reefs restored using small substrate in subtidal habitats.



The annual nitrogen removal rates were then used to construct a lookup table with baseline oyster tissue biomass (g DW m^{-2}) ranges listed in the rows and post-restoration oyster tissue biomass (g DW m^{-2}) ranges listed in the columns. The cells in the lookup table contain estimates of the enhanced denitrification (lbs acre^{-1} yr^{-1}), which was calculated by subtracting the annual denitrification rate for the baseline oyster tissue biomass range in each row from the annual denitrification rate for the post-restoration oyster tissue biomass range in each column. A partial lookup table is shown in Table 8.6 and the full lookup table is provided in Appendix G. In cases where post-restoration oyster tissue biomass was equal to or less than the baseline oyster tissue biomass, or where enhanced denitrification did not occur (e.g., in cases where biomass >275 g DW m^{-1}; Figure 8.4, Appendix G), the cell was left blank.

Table 8.6. Partial lookup table for use in determining the annual enhanced denitrification rates. For full lookup table, see Appendix G.

Enhanced Nitrogen Removal (lbs acre^{-1} yr^{-1})		***Post-restoration Oyster Biomass Range (g DW m^{-2})***												
		15 - 24.9	***25 - 34.9***	***35 - 44.9***	***45 - 54.9***	***55 - 64.9***	***65 - 74.9***	***75 - 84.9***	***85 - 94.9***	***95 - 104.9***	***105 - 114.9***	***115 - 124.9***	***125 - 134.9***	***135 - 144.9***
Baseline Oyster Biomass Range (g DW m^{-2})	***0 - 14.9***	29	51	74	97	120	143	165	169	172	176	179	183	186
	15 - 24.9		23	46	68	91	114	137	140	144	147	151	154	158
	25 - 34.9			23	46	68	91	114	118	121	124	128	131	135
	35 - 44.9				23	46	68	91	95	98	102	105	109	112
	45 - 54.9					23	46	68	72	75	79	82	86	89
	55 - 64.9						23	46	49	53	56	59	63	66
	65 - 74.9							23	26	30	33	37	40	44
	75 - 84.9								3	7	10	14	17	21
	85 - 94.9									3	7	10	14	17
	95 - 104.9										3	7	10	14
	105 - 114.9											3	7	10
	115 - 124.9												3	7
	125 - 134.9													3

8.2.6.2 Site-specific Approach

For restoration efforts that utilize large substrate and/or reefs in intertidal habitats, denitrification rates must be based on site-specific measurements of denitrification. The Panel recommends that implementers develop season-specific regression equations in consultation with expert(s) knowledgeable in oyster reef sampling and the measurement of oyster-associated denitrification rates. Considerations for measuring denitrification rates on restored reefs are outlined in Appendix F.

8.2.7 Total Annual Denitrification Enhancement

The total amount of nitrogen removed annually from the BMP site through enhanced denitrification is determined by multiplying the enhanced denitrification estimate (lbs acre^{-1}) by the total BMP site area.

8.2.8 Crediting Timeframe

Since monitoring timeframes differ between oyster reef restoration projects and because oyster tissue biomass on a restored reef can fluctuate from year to year, the Panel considered how frequently oyster biomass needs to be measured to verify the persistence of oyster biomass levels at the restoration site and prevent over crediting. The Panel decided that nitrogen reduction effectiveness should be valid for a maximum of three years after a post-restoration biomass survey. After three years, no credit is given until the post-restoration oyster tissue biomass has been determined again from field measurements at the BMP site and the enhanced denitrification estimate is re-evaluated using the baseline oyster tissue biomass and the newly determined post-restoration oyster tissue biomass. If crediting is warranted after a gap in monitoring, crediting can begin again for the year the reef was surveyed but cannot be given retroactively. If biomass is assessed more frequently than once every three years, the most recent post-restoration biomass should be used for determining the appropriate level of credit. Post-restoration oyster tissue biomass can be measured and enhanced denitrification can be credited over the lifetime of the BMP site.

8.3 Reduction Effectiveness: Stepwise Determination

To calculate the reduction effectiveness for the restoration-denitrification protocols, the Panel recommends the following:

Step 1. Identify the BMP site location and determine the BMP site area (Subchapter 8.2.1)

Step 2. Document the qualifying enhancement activity and the date it occurred (Subchapter 8.2.2)

Step 3. Determine the appropriate baseline approach (Subchapter 8.2.4)

Step 4. Assess baseline and post-restoration tissue biomass (Subchapter 8.2.5)

Step 5. Determine denitrification enhancement per unit area using either the biomass-based default denitrification rates per unit area (Subchapter 8.2.6.1) or site-specific measured denitrification rates (Subchapter 8.2.6.2)

Step 6. Determine the total nitrogen removal attributable to enhanced denitrification using the estimated denitrification enhancement per unit area and the BMP site area (Subchapter 8.2.7)

8.4 TMDL Baseline Considerations

The TMDL for Chesapeake Bay was created in 2009. Based on this, the CBP Management Board agreed that reduction crediting under the restoration-denitrification protocols for oysters can only be given for oyster reef restoration projects that were initiated after the creation of the TMDL in 2009.

8.5 Qualifying Conditions

The Panel recommends that the following qualifying conditions be met for applying the restoration-denitrification protocols to receive nitrogen reduction credit. These account for both the CBP Management Board's defined baseline and the Panel's criteria. Most of the qualifying conditions for the restoration-denitrification protocols are identical to those for the restoration-assimilation protocols (highlighted in gray).

- A qualifying enhancement activity (Subchapter 8.2.2) must have occurred throughout the BMP site area (Subchapter 8.2.1).
- BMP site must lie within an area protected from harvest
- Baseline oyster biomass must be determined using an appropriate approach and adhere to baseline conditions (Subchapter 8.2.4)
- All biomass estimates (Subchapter 7.2.5) must:
    - Be based on field surveys of live *Crassostrea virginica*
    - Be based on data collected using a survey design that ensures estimates are representative of the entire BMP site.
    - Include enough data points to allow calculation of mean biomass and its variance. If multiple strata are included in the sampling design, data must be sufficient to calculate means and variances for all strata.
- If using the default approach to estimating enhanced denitrification, the reef must be in a subtidal habitat and restoration activities must have utilized only small substrates (Subchapter 8.2.2).
- Only live oyster tissue biomass is eligible for credit.
- The post-restoration oyster tissue biomass must be greater than the baseline oyster tissue biomass

8.6 Recommended Application and Verification Guidelines

8.6.1 Reporting Guidelines

To assist with application of its recommendations, the Panel developed guidelines for the information to be reported by anyone seeking credit for this practice-protocol combination. The required information is listed below under the associated determination step. The majority of this information is identical to the information that needs to be reported for the restoration-assimilation protocols (highlighted in gray).

Step 1: Identify the BMP site location and determine the BMP site area (Subchapter 7.2.1)

- Geospatial information documenting the vertices of a polygon representing the BMP site
- Area of the polygon representing the BMP site

Step 2: Document the qualifying enhancement activity and its date (Subchapter 7.2.2), the type(s) of substrate used for restoration (Subchapter 7.2.3), and the baseline approach (Subchapter 7.2.4).

- Date of enhancement activity
- Type(s) of substrate

- Substrate category (small or large)
- Amount of substrate used
- If using hatchery-produced oysters:
    - Ploidy of oysters
    - Number of oysters planted
    - Size of oysters at time of planting
- Baseline approach used (pre-restoration or representative site)

Step 3: Assess baseline and post-restoration tissue biomass (Subchapters 7.2.5) and determine the mean tissue biomass per meter square for each.

- For first survey after reef restoration, provide a brief description of biomass survey sampling design for both the baseline and post-restoration biomass surveys. For subsequent surveys, only information on the post-restoration survey design is needed.
    - Sampling date(s)
    - Method used to collect samples (e.g., patent tongs, divers with quadrats, etc.)
    - Spatial scale of sample (e.g., 1.0 m^2, one Reef Ball™, etc.)
    - Number of samples collected
    - If subsampling is used, a description of the subsampling methods, number of subsamples per sample, and method of scaling
    - Methods used to assess oyster tissue biomass per sample (default regression, direct measurement, or site-specific regression)
    - Method used to calculate mean sample biomass
    - Mean tissue biomass in grams dry weight per square meter

Step 4: Determine annual denitrification enhancement per acre (Subchapter 8.2.5).

- Document approach used to estimate denitrification enhancement (default or site-specific)
- Denitrification enhancement per acre
    - If using the default approach, determine denitrification enhancement using the lookup table. No additional documentation is required.
    - If using the site-specific approach, a full report of the methods used to measure denitrification, estimate enhancement, and extrapolate enhancement per unit area is required.

Step 5: Determine total annual denitrification enhancement (Subchapter 8.2.6).

- Total nitrogen removed annually by restoration at the BMP site.

8.6.2 Example

To give an example of the information needed for crediting, Table 8.7 provides a list of the information along with a hypothetical example of that information for the first year of harvest from a BMP site. To emphasize the similarity between the information required for the restoration-assimilation and restoration-denitrification protocols, the example below uses the same scenario as that given for small substrate for the restoration-assimilation protocol (Subchapter 7.6.2.1) and the items below that are identical for both examples are highlighted in gray. The primary difference between the restoration-denitrification and restoration-assimilation calculations is that for restoration-denitrification protocols, the most recently measured oyster biomass per square meter is always compared to the baseline data to determine denitrification enhancement. In contrast, for the restoration-assimilation protocol, the data from the most recent post-restoration biomass survey is compared the highest previously measured biomass per unit area.

If the default approach to estimating denitrification is used, the annual nitrogen removal per acre can be determined by simply finding the cell that corresponds to the appropriate baseline and post-restoration biomass levels and using the value from the lookup table. In this example, the pre-restoration biomass was 14 g DW oyster tissue m^{-2} and the post-restoration biomass was 119 g DW oyster tissue m^{-2}, corresponding to an annual removal of 179 pounds of nitrogen per acre per year. Because the BMP site area is one acre, the annual nitrogen removal for this restoration effort is 179 lbs of nitrogen per year. In this example, biomass is not measured again until 2027, so the value of 179 lbs of nitrogen removal is credited for the years 2024, 2025, and 2026 (Table 8.8). In 2027, the biomass level dropped to 105 g DW m^{-2}. As a result, the estimated denitrification enhancement is reduced to 176 lbs $acre^{-1}$ yr^{-1}. In 2030, a biomass survey finds that biomass has increased to 173 g DW m^{-2} resulting in an increase in annual nitrogen removal to 195 lbs $acre^{-1}$ yr^{-1}. The final reef survey conducted in 2033 finds that biomass had declined to 59 g DW m^{-2} resulting in a decrease in annual crediting to 120 lbs $acre^{-1}$ yr^{-1}. Over the 12-year period during which crediting occurs, the one-acre reef results in a total enhanced nitrogen removal via denitrification of 2,010 lbs (Table 8.8).

Table 8.7. Information types required for the restoration-denitrification protocols along with an example of each. See text for details of calculations used to provide example information below. Items that are identical for both the restoration-assimilation and restoration-denitrification protocols are highlighted in gray.

Step #	Information Type	Example
1	BMP site location	See appended map and GIS file
	Area of the BMP site	1 acre
2	Date(s) of activity (mm/dd/yy)	09/21/21
	Type(s) of substrate	Diploid spat-on-shell
	Substrate category	Small
	Amount of substrate	1,000 Maryland bushels of spat-on-shell
	Number of hatchery-produced oysters planted	9,500,000
	Size of oysters at time of planting (mm)	10
	Baseline approach	Pre-restoration
3	Baseline biomass	
	Sampling points	See appended map and GIS file
	Sampling date(s)	07/15/20
	Sampling method	Patent tong
	Spatial scale of sample with units	1 m^2
	Number of samples collected	5
	Method used to assess biomass	Default regression
	Method used to calculate mean biomass	Average of all samples
	Mean biomass: Tissue	14 g DW m^{-2}
	Post-restoration biomass	
	Sampling date(s)	08/01/24
	Sampling method	Patent tong
	Spatial scale of sample with units	1 m^2
	Number of samples collected	5
	Method used to assess biomass	Default regression
	Method used to calculate mean biomass	Average of all samples
	Mean biomass: Tissue	119 g DW m^{-2}
4	Approach used to estimate denitrification enhancement	Default
	Annual enhanced denitrification per acre	179 lbs $acre^{-1}$ $year^{-1}$
5	Total annual denitrification enhancement	179 lbs $year^{-1}$

Table 8.8. Example of crediting for the restoration-denitrification protocol based on biomass surveys conducted at three year intervals.

Year	Survey Type	Measured Biomass (g DW m^{-2})	Annual Enhanced Nitrogen Removal (lbs $acre^{-1}$)
2020	Baseline	14	NA
2021			NA
2022			NA
2023			NA
2024	Post-restoration	119	179
2025			179
2026			179
2027	Post-restoration	105	176
2028			176
2029			176
2030	Post-restoration	173	195
2031			195
2032			195
2033	Post-restoration	59	120
2034			120
2035			120
		Total	***2,010***

8.7 Unintended Consequences

The Panel's review of published data found no instances where the restoration of subtidal oyster reefs using small substrates resulted in a decrease in net denitrification at the restoration site. However, the approach taken by the Panel to estimate denitrification enhancement focuses on local effects on biogeochemical cycling and does not consider effects at the landscape scale. Post-restoration denitrification rates can be orders of magnitude higher than baseline denitrification rates, but denitrification efficiency may be lower after restoration than before. Denitrification efficiency is a measure of the amount of total nitrogen that is denitrified. It is calculated by dividing the flux of N_2-N by the sum of all nitrogen fluxes and is reported as a percentage. When considered at a landscape scale, it is feasible that the total amount of nitrogen removed from the system could be lower after reef restoration if the restored reef had a low denitrification rate and if phytoplankton would have been remineralized in an environment with higher denitrification efficiency had it not been filtered from the water column by oysters.

However, these conditions are unlikely in Chesapeake Bay. The filtering actions of oysters greatly increase deposition of organic matter to the benthos. In the absence of oyster filtration, much of that phytoplankton would have been remineralized in the water column where denitrification does not occur. The portion of the phytoplankton that did fall to the benthos in the absence of oysters would have to be deposited on oxic sediments for denitrification to occur. Given the eutrophic condition of Chesapeake Bay and that large portions of the Bay are subject to anoxic conditions each year, only a portion of that phytoplankton is likely to fall onto habitats that support high-efficiency denitrification. Because reef restoration also supports assimilation of nitrogen in the tissues and shells of oysters (Chapter 7), the Panel concluded that situations in which reef restoration led to a net decrease in nitrogen removal at the landscape scale would be rare if they occur at all.

Oyster filtration of phytoplankton leads to increased deposition of organic matter to the benthos. Although increasing the supply of organic matter to the seafloor is part of why restored reefs can enhance denitrification rates, too much organic matter can have negative impacts on water quality. The microbial processes that break down organic matter consume oxygen. If bottom waters become anoxic, nitrification (a microbial process that requires oxygen and a precursor to denitrification) can no longer supply the substrates needed for denitrification to occur. The Panel found no reports of situations where reef restoration drove local anoxia or led to a decrease in denitrification rates. However, because these studies focused on restoration efforts using small substrates and few studies have examined the biogeochemical changes associated with the use of large substrates, it is unclear whether this could occur in association with the use of large (e.g., engineered structures) for reef restoration. However, since the Panel is recommending that separate measurements are made to evaluate denitrification on large substrates for crediting purposes, any excessive loading of organic matter would be observed and allow for adaptive management of the practice. Consideration of the potential for negative consequences should be explicitly considered as part of any effort to develop reduction effectiveness estimates for reefs restored using large substrates.

In addition to altering nitrogen dynamics, reef restoration can also alter the phosphorus dynamics on the seafloor. Some of the studies used in the Panel's meta-analysis included data on soluble reactive phosphorus (SRP) fluxes. Results showed both increasing and decreasing SRP flux with increasing oyster biomass. Factors driving differences between studies were unclear. High variance in SRP flux data is not unexpected because phosphorus dynamics can be altered by local sediment composition, specifically whether they contain significant amounts of iron oxides. Under anoxic conditions, iron oxides that bind phosphorus can be converted to iron sulfides leading to the release of phosphorus (O'Keefe 2007, Jordan et al. 2008). Reefs may be on a trajectory of increasing phosphorus flux as the sulfide in sediments increases as the reef matures. Given the limited amount of data available and its high degree of variability, the Panel suggested that the effects of reef restoration on local phosphorus dynamics should be a topic of future study.

8.8 Ancillary Benefits

The Panel identified several ancillary benefits for the restoration-denitrification protocols. Benefits of oyster restoration are described in Chapter 9.

8.9 Future Research

Although the Panel found sufficient data to develop reduction effectiveness estimates for nitrogen removed by enhanced denitrification on subtidal oyster reefs using small substrates, additional research is needed to refine these estimates, to develop default estimates for intertidal reefs and reefs restored using large substrates, and to develop estimates for practices not covered in the current set of recommendations. The Panel suggests that the following future research:

- Determine whether relationships between denitrification rates and oyster tissue biomass are influenced by salinity and/or nutrient gradients. Measurement in more sites would be beneficial.
- Prioritize denitrification measurements for oysters within the 75-270 g DW m^{-2} tissue biomass range. These data will refine the relationship between denitrification rates and oyster tissue biomass and refine the estimates for nitrogen removal.
- Determine whether significant relationships exist between denitrification and oyster tissue biomass on intertidal reefs and on reefs built using large substrates. If these relationships exist, explore whether they are sufficiently robust to develop default nitrogen reduction effectiveness estimates for these practices.

- Develop new methods for measuring denitrification associated with oyster reef restoration that are more cost-effective, allowing efficient development of better seasonal and spatial denitrification measurements.
- Develop denitrification techniques to assess the nutrient removal values for oysters attached to large substrates such as ReefBalls™ and Oyster Castles©.
- Determine whether significant relationships between denitrification rates and oyster tissue biomass for restored oyster reefs exist in winter. If these relationships exist, explore whether they should be incorporated into current nitrogen reduction effectiveness estimates.
- Improve understanding of how restored oyster reefs alter nitrogen dynamics at spatial scales greater than the footprint of the restoration project.
    - A significant portion of reef biodeposits may not be retained within the reef footprint. If these biodeposits are remineralized in oxic sediments outside the footprint of the reef and subsequently denitrified, restored reefs may remove more nitrogen than accounted for in the current reduction effectiveness estimates.
    - The current reduction effectiveness estimates focus on local impacts on denitrification rates. However, reef restoration takes place within a broader landscape. A true accounting of the net effect of oyster reef restoration would evaluate reef impacts at the landscape scale and would account for the fate of phytoplankton had the reefs not been constructed.
- Explore effects of reef restoration on phosphorus dynamics.
- Examine whether on-bottom aquaculture has similar value as restored reefs for enhancing denitrification.

9.0 Ancillary Benefits of Oyster Reef Restoration

The ancillary benefits of oyster reef restoration are numerous (e.g., Coen et al. 1999, 2007). Perhaps the most well-documented benefit of oyster reef restoration is the provision of habitat for other organisms. Oyster reefs provide habitat for an abundance of marine species. Numerous studies have recorded a greater abundance and diversity of marine fauna on natural and restored oyster reefs relative to areas with no reef structure (e.g., Tolley & Volety 2005, Rodney & Paynter 2006, Kellog et al. 2013). Many reef-associated species serve as prey for commercially and recreationally important species. Thus, reef restoration has the potential to enhance the production of recreational and commercially significant species in the Chesapeake Bay, which would have additional economic benefits in the region (e.g., Grabowski et al. 2012, Knoche et al. 2020, Bruce et al. 2021).

Because many reef-associated organisms are also filter feeders and also assimilate nitrogen and phosphorus, enhancing their populations through reef restoration may increase water quality benefits beyond those provided by oysters. For example, hooked mussels are filter feeders commonly found on oyster reefs in Chesapeake Bay. A study by Gedan et al. (2014) found that hooked mussel biomass can sometimes exceed that of oysters on restored reefs and that they can more than double the filtration capacity of the reef. In their assessment of reef nutrient assimilation, Kellogg et al. (2013) found that 34% of the total assimilated nitrogen and 33% of the total assimilated phosphorus in oyster reef samples was in organisms other than oysters. This suggests that increases in biomass of other reef organisms can assimilate greater portions of nutrients than oysters alone and therefore the estimates generated in this report are likely conservative.

The filtration capacity of oysters and reef communities has the potential to increase light penetration through the water column which may benefit seagrasses. A modeling study by Newell & Koch (2004) suggested that increasing oyster populations in Chesapeake Bay would reduce turbidity and facilitate seagrass growth and the expansion of seagrass beds to deeper water. Another modeling study by Cerco & Noel (2007) suggested that a 10% increase in oyster populations in Chesapeake Bay should increase the biomass of submerged aquatic vegetation (SAV). Because seagrasses are a refuge for marine animals and provide other water quality benefits (Orth et al. 2006), the relationship between oyster restoration and seagrass establishment could be synergistic. However, more recent field studies on the effects of increased oyster density and/or oyster reef restoration on SAV habitats suggest that these effects may not be observed under field conditions, that their spatial scale may be very limited, and that interactions may be positive or negative (Booth & Heck 2009, Plutchak et al. 2010, Grizzle et al. 2018).

Restored oyster reefs can also provide protection for shorelines vulnerable to wave energy and sea level rise. Some such breakwater reefs have increased sedimentation to adjacent marsh edges and reduce erosion and marsh loss (Stricklin et al. 2010) while creating habitat for commercially important species like blue crab and red drum (Scyphers et al. 2011). The potential economic impact of combined benefits from shoreline protection and increased fish production can be significant (e.g., Kroeger & Guannel 2014).

10.0 Conclusion

In this report, the Panel provides complete recommendations for implementing twelve oyster practice-protocol combinations for BMP use (Table 1.1) in addition to the six combinations approved in the Panel's first oyster BMP report (Reichert-Nguyen et al. 2016). For these twelve combinations, the Panel verified that sufficient data and information were available to address all items outlined in the CBP Partnership's BMP Review Protocol. The Panel concluded that certain licensed oyster harvest (**Practice F**) and oyster restoration (**Practices J & K**) practices could permanently remove nitrogen and phosphorus through assimilation in oyster tissue (**Protocols 1 & 4**) and shell (**Protocols 2 & 5**), and/or through the biogeochemical process of denitrification (**Protocol 3**).

For all practice-protocol combinations, the Panel concluded that oyster biomass data are required to estimate reduction effectiveness and to verify that the enhancement activity led to an increase in oyster production. The Panel used common statistical approaches (e.g., 50th quantile regression) to determine relationships between oyster tissue and/or shell biomass and the amount of nitrogen and/or phosphorus removed from the water column. The Panel developed tools (e.g., default equations and/or lookup tables) for implementers to use to estimate reduction effectiveness and developed a comprehensive set of verification guidelines and qualifying conditions to minimize nutrient over crediting for each oyster practice.

The Panel identified a few potential unintended consequences associated with crediting oyster restoration practices; however, the Panel agreed that significant over crediting is unlikely because the recommended crediting approaches are intentionally conservative and significant nitrogen and phosphorus are removed by processes that are not captured by the recommended practice-protocol combinations (e.g. nutrients retained in dead oyster shells, nutrients assimilated in reef-associated organisms, enhanced denitrification associated with oysters prior to harvest).

The Panel also identified several next steps and research needs required to more accurately quantify nutrient reduction through oyster-related processes in the Chesapeake Bay. In general, these include gaining a better understanding of:

- Nitrogen and phosphorus assimilation in shell on harvested oyster reefs
- Spat survivorship estimates on both harvested and restored reefs
- Oyster biomass estimates and denitrification rates for intertidal reefs
- Oyster biomass estimates and denitrification rates for large structures and other suitable substrates
- How abiotic factors impact denitrification rates at the reef scale
- Seasonal and spatial variability in denitrification measurements (e.g., in winter)
- How restored reefs alter nitrogen dynamics at the landscape scale
- Denitrification rates associated with other oyster practices (harvest and/or aquaculture)
- Phosphorus cycling processes associated with oysters

The Panel was unable to provide recommendations for suspended sediment reduction (**Protocol 6**) or enhanced nitrogen and phosphorus burial associated with oysters (**Protocols 7 & 8**). Quantifying permanent removal through these processes is challenging because sediment can be resuspended and deposited several times within a basin. Sediment transport patterns also vary within and among basins as a function of several abiotic factors. The Panel agreed that more research is needed to minimize uncertainty in sediment deposition rates and recommends that STAC convene a workshop to re-evaluate whether data are available or can be collected. The CBP Management Board will need to work closely with a future oyster BMP Panel to reach consensus on which metrics can most accurately quantify sediment and nutrient removal through deposition and burial.

If approved by the CBP Partnership, the BMP process and recommendations described here can contribute measurable positive progress towards the water quality goals established by the Chesapeake Bay Total Maximum Daily Load framework.

11.0 References

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