2020 Maryland Oyster Monitoring Report

Analysis of Data from the '10 Tributaries' Sanctuary Oyster Restoration Initiative in Maryland

Data collected from October 2020 through January 2021

Produced in partnership with the Maryland Oyster Restoration Interagency Workgroup of the Chesapeake Bay Program's Sustainable Fisheries Goal Implementation Team

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This report, past monitoring reports, tributary-specific oyster restoration plans ('blueprints'), and other oyster restoration technical documents produced by the Maryland Oyster Restoration Interagency Workgroup of the Chesapeake Bay Program's Sustainable Fisheries Goal Implementation Team are available at https://www.chesapeakebay.net/who/publications-archive/maryland_and_virginia_oyster_restoration_interagency_teams.

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Executive Summary

Context for This Report

- The 2014 Chesapeake Bay Watershed Agreement[1] includes a goal to restore oyster populations in 10 Chesapeake Bay tributaries by 2025 (hereafter, the 10 tributaries initiative').

- In Maryland, partners including the National Oceanic and Atmospheric Administration (NOAA), U.S. Army Corps of Engineers' Baltimore District (USACE), Oyster Recovery Partnership (ORP), and the Maryland Department of Natural Resources (DNR) are working to achieve this goal through the Maryland Interagency Oyster Restoration Workgroup (hereafter, the Workgroup). The Workgroup is convened under the Sustainable Fisheries Goal Implementation Team of the Chesapeake Bay Program and is chaired by Stephanie Reynolds Westby (NOAA).

- A set of oyster restoration success criteria, commonly known as the Chesapeake Bay Oyster Metrics[2], was developed prior to implementing restoration work in the 10 tributaries. In past years, the annual versions of this report described the success of each reef monitored relative to the six Oyster Metrics success criteria: oyster density, oyster biomass, multiple year classes, shell budget, reef height, and reef footprint. However, COVID-related restrictions in 2020 and 2021 prohibited data collection on reef height and reef footprint parameters. These data will be collected in fall 2021 to ensure a complete data set, where possible. This report therefore describes reef success relative to the four success criteria for which data was collected: oyster density, oyster biomass, multiple year classes, and shell budget.

- Restored reefs are monitored three years and again six years after initial restoration. A subset of reefs in Harris Creek, Little Choptank River, and Tred Avon River are now either three or six years old, and were due for monitoring in fall 2020.

- Trends observed in previous monitoring years generally continued in 2020, with the wide majority of restored reefs meeting all Oyster Metrics success criteria for which they were monitored.

- Data and analyses in this report can be used by restoration partners to help inform what adaptive management measures, if any, should be taken on each of the monitored reefs. Results may also guide restoration in other tributaries.

Key Fall 2020 Monitoring Results

- In fall 2020, 40 three-year-old restored reefs (103 acres) were monitored in the Little Choptank and Tred Avon rivers combined, and 27 six-year-old restored reefs (119 acres) were monitored in Harris Creek and Little Choptank River combined.

- Overall, the vast majority of reefs monitored in fall 2020 met the Oyster Metrics success criteria.

  - Oyster density: 98% of three-year-old reefs and 100% of six-year-old reefs, met the minimum threshold criteria (see Figure 1).

  - Oyster biomass: Results for this criteria tracked closely with oyster density (see Figure 5).

  - Multiple year class and shell budget: 100% of reefs met these criteria (see Table 3).

  - Reef height and reef footprint: These criteria were not monitored in fall 2020 due to COVID-related restrictions on vessel operations.

- See Section 3.1: Summary of Fall 2020 Monitoring Results, and Appendix A: Table of Summary Data by Reef, for more complete results information.

Key Cumulative Monitoring Results 2015-2020

- From fall 2015 through fall 2020, 203 three-year-old restored reefs (711 acres) were monitored in Harris Creek, Little Choptank River, and Tred Avon River combined, and 70 six-year-old restored reefs (310 acres) were monitored in Harris Creek and Little Choptank River combined.

- The vast majority of reefs monitored during this period met all of the minimum Oyster Metrics success criteria.

  - Oyster density: 96% of three-year-old reefs and 99% of six-year-old reefs, met the minimum threshold criteria (see Figure 1).

  - Oyster biomass: Results for this criteria tracked closely with oyster density throughout the period (see Figure 5).

  - Multiple year classes and shell budget: 100% of reefs met these criteria (see Table 3).

  - Reef height and footprint: 100% of reefs monitored for these parameters met the success criteria.

- See Section Section 3.2: Summary of Cumulative Results, 2015-2020 for more complete results information.
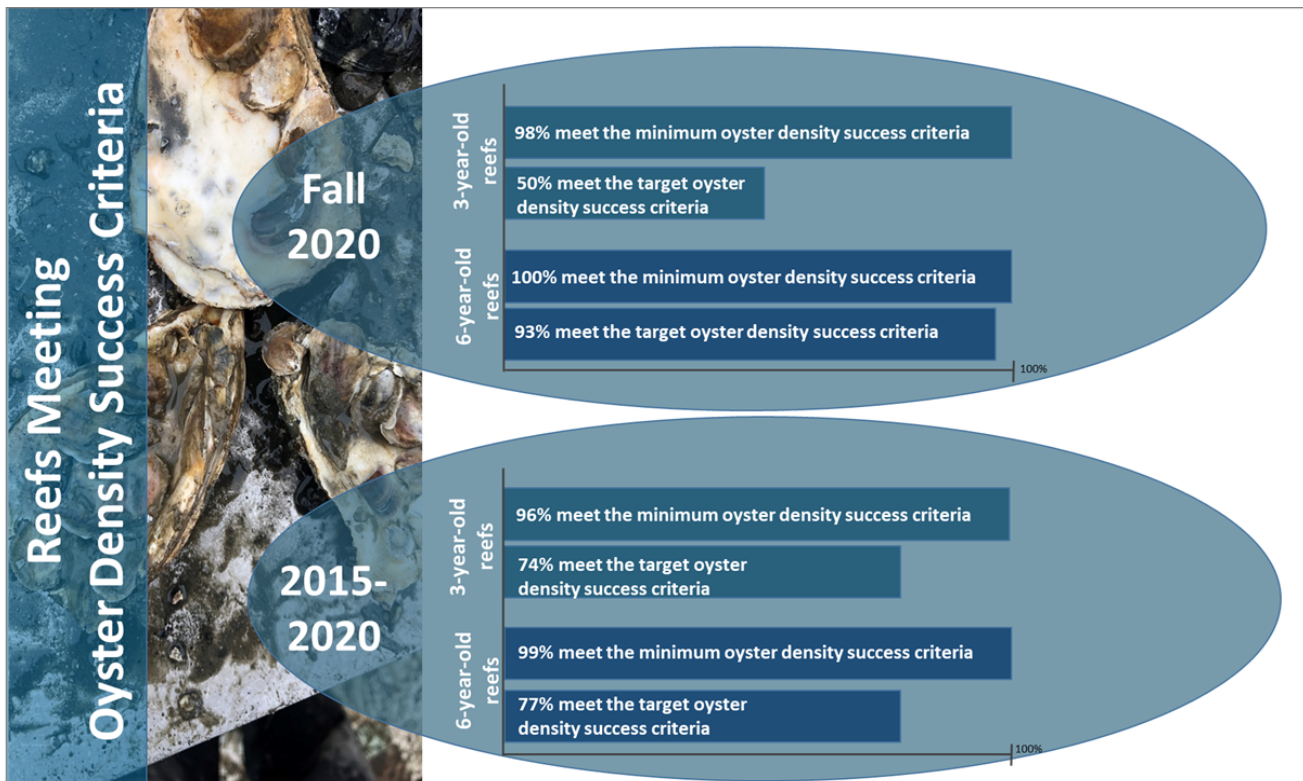
Figure 1: *Graphic showing reefs meeting the oyster density success criteria in 2020 and 2015-2020 (cumulative). Oyster biomass followed a similar trend (see Figure 5).*

Section 1: Background and Overview

1.1: Policy Drivers, Oyster Metrics Success Criteria, and Oyster Restoration Planning

The 2014 Chesapeake Bay Watershed Agreement[1] oyster outcome calls for restoring oyster populations in 10 Chesapeake Bay tributaries by 2025. The Chesapeake Bay Program's Sustainable Fisheries Goal Implementation Team (Fisheries GIT) is charged with working to achieve this goal. Driven by Executive Order 13508 (Chesapeake Bay Protection and Restoration) of 2009, some work toward tributary-scale oyster restoration was under way even before the Chesapeake Bay Watershed Agreement was signed. The Fisheries GIT had convened the Chesapeake Bay Oyster Metrics Workgroup, which, in its 2011 report "Restoration Goals, Quantitative Metrics and Assessment Protocols for Evaluating Success on Restored Oyster Reef Sanctuaries,"[2] (hereafter, 'Oyster Metrics') established Bay-wide, science-based, consensus success criteria for oyster restoration to be tracked three years and six years following restoration efforts (Table 1).

Once these success criteria were adopted, the Fisheries GIT convened interagency workgroups in Maryland and Virginia to plan and coordinate restoration work in each state. In Maryland, the Maryland Oyster Restoration Interagency Workgroup (hereafter, 'the Workgroup') is chaired by the National Oceanic and Atmospheric Administration (NOAA) and includes members from the Maryland Department of Natural Resources (DNR), Oyster Recovery Partnership (ORP), and the U.S. Army Corps of Engineers' Baltimore District (USACE). The Workgroup developed oyster restoration tributary plans (also known as "blueprints") for Harris Creek[3], Little Choptank River[4], Tred Avon River[5], upper St. Marys River[6], and Manokin River[7] in consultation with a group of consulting scientists and the public.

Biological Metrics	Oyster density	Minimum threshold = 15 oysters per m^2 over 30% of the reef area. Target = 50 oysters per m^2 over 30% of the reef area.
	Oyster biomass	Minimum threshold = 15 grams dry weight per m^2 over 30% of the reef area. Target = 50 grams dry weight per m^2 over 30% of the reef area.
	Multiple year classes	Presence of multiple year classes on the reef, as defined by oysters in at least two of the following size classes: market (>76 mm); small (40-75 mm); spat (<40 mm).
	Shell budget	Stable or increasing shell volume on the reef.
Structural Metrics	Reef footprint	Stable or increasing reef footprint compared to baseline.
	Reef height	Stable or increasing reef height compared to baseline.

Table 1: Oyster Metrics reef-level success criteria. Note that in fall 2020, reef height and reef structure were not monitored due to COVID-related restrictions on vessel operations.

1.2: Overview of Report Content

Restored reefs are monitored at three and six years per Oyster Metrics recommendations and each river's tributary plan. Restored reefs in Harris Creek, Little Choptank River, and Tred Avon River have matured to three or six years, and therefore were monitored in October 2020 through January 2021 (referred to as the 'fall 2020' monitoring cycle). Data and analysis for these reefs, plus reference reefs (controls that received no restoration action) and sentinel reefs (restored sites that are monitored annually) are included in this report. Data summaries for each reef individually are in Appendix A: Table of Summary Data by Reef. This report describes success relative to four of the six Oyster Metrics criteria (oyster density, oyster biomass, multiple year classes, and shell budget). Data relating to the remaining two success criteria (reef height and reef footprint) were not collected in fall 2020 due to COVID-related restrictions on vessel operations. Past monitoring reports are available from the Chesapeake Bay Program's Maryland and Virginia Oyster Restoration Interagency Teams Publications page.

In addition to Oyster Metrics success criteria monitoring, oyster disease data is also collected by DNR, and is available in DNR's annual Fall Survey Report.

1.3: Funding and Acknowledgements

- Monitoring data for the biological success metrics (oyster density, oyster biomass, multiple year classes, and shell budget) were collected, managed, and analyzed by ORP, Coastal Marine Sciences Inc., and contracted commercial watermen, with assistance from Workgroup partners. This was accomplished with funding from:

  - A $130,000 award from NOAA to ORP, and

  - A $124,183 programmatic agreement from USACE to ORP.

- This report was drafted by NOAA, with guidance from the Workgroup. Results of these analyses will be used to document the success or failure of restoration work relative to the Oyster Metrics criteria, to guide adaptive management of these reefs, and to inform future oyster restoration efforts. Technical review of this report was provided by technical experts and Workgroup members, per NOAA research communications guidelines.

Section 2: Overview of Methods and Revised Protocols for 2020

2.1: Location of Monitored Reefs
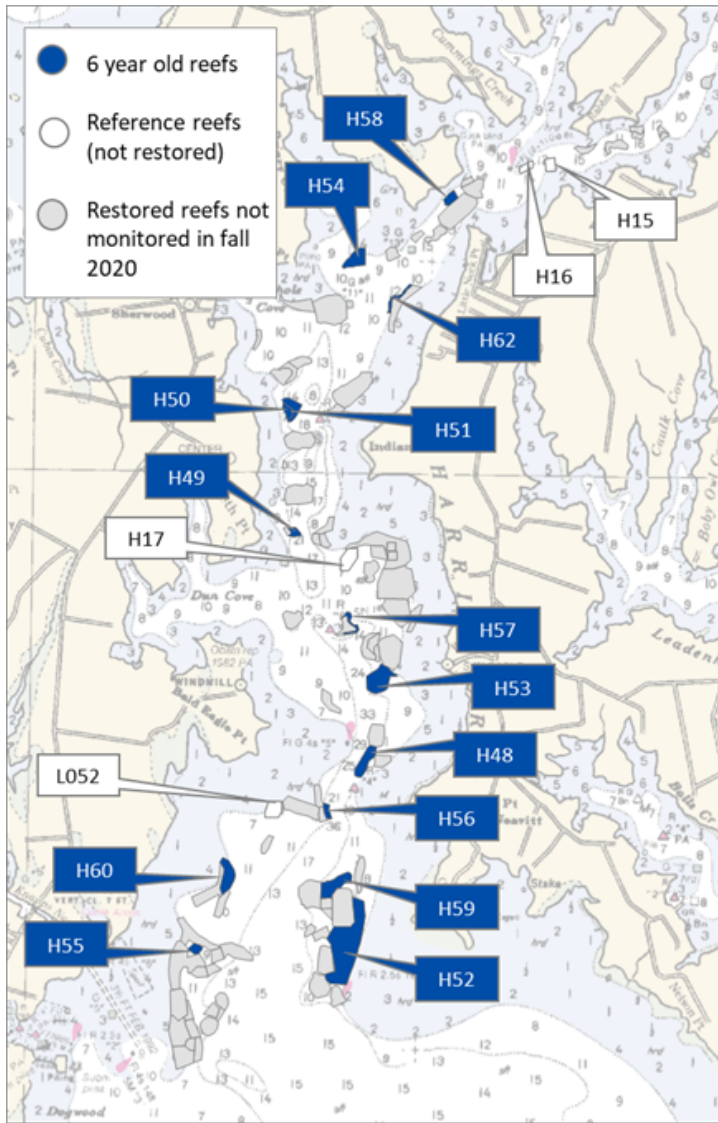
Figure 2: *Locations of Harris Creek reefs monitored in fall 2020. 14 six-year-old reefs were monitored in fall 2020. (Three-year monitoring is complete on all reefs.)*
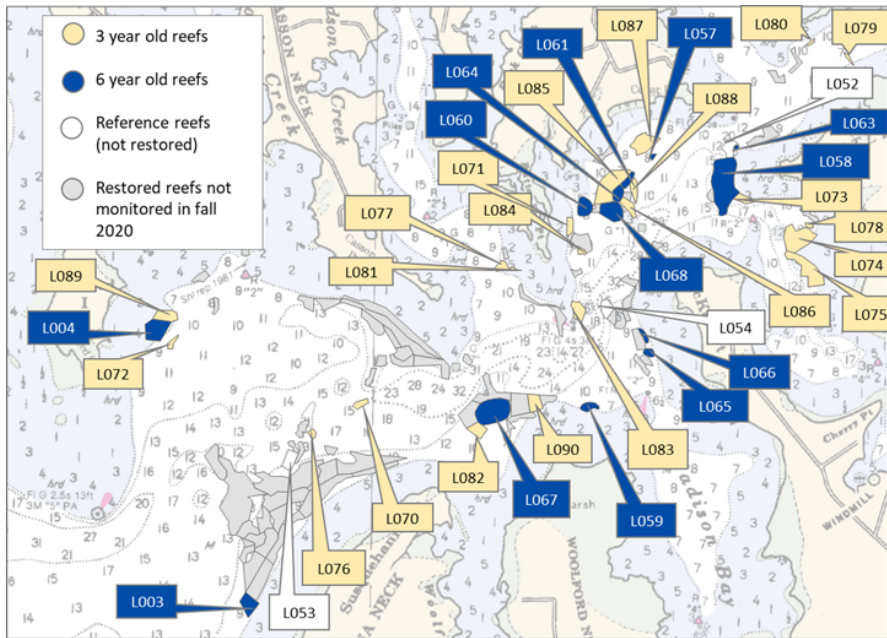
Figure 3: *Locations of Little Choptank River reefs monitored in fall 2020. 21 three-year-old reefs and 13 six-year-old reefs were monitored.*
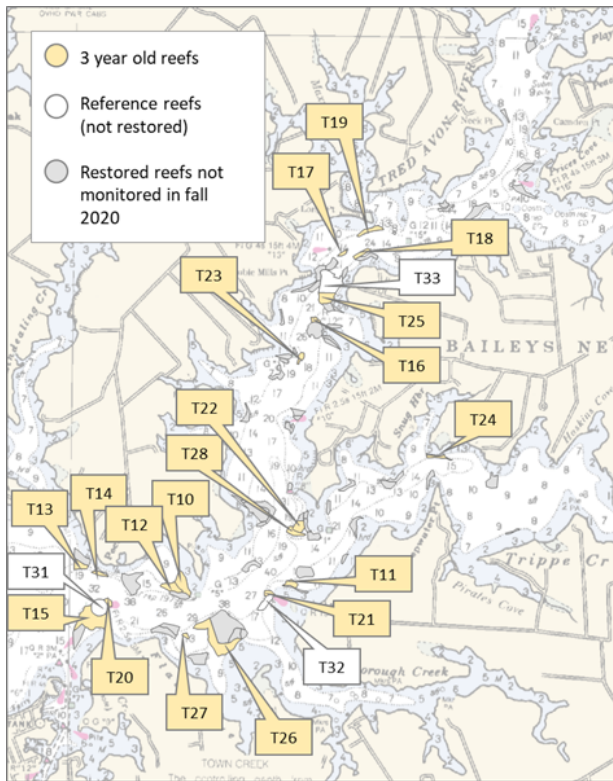
Figure 4: *Locations of the Tred Avon River reefs monitored in fall 2020. 19 three-year-old reefs were monitored. No restored Tred Avon River reefs have yet matured to six years.*

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2.2: Methods Summary

See Appendix C for full methods description.

Data to determine success relative to the four biological metrics (oyster density, oyster biomass, multiple year classes, and shell volume) were collected at the same time, using a stratified random survey design. Methods used to select sampling sites, analyze samples, and assess success relative to each biological were identical for all reefs. Data collection occurred between October 2020 and January 2021.

As in previous years, two different types of gear were used to collect samples, depending on reef substrate type:

- Divers were used to collect samples from reefs with substrate materials that were not amenable to patent tong sampling (stone and fossil shell substrate reefs).

- Patent tongs were used to collect samples from all other reef types (seed only, mixed-shell base, reference, and premet reefs) because it is more cost efficient than using divers.

- See Table 2 for description of the various treatment types, and the gear used to monitor the biological metrics on each.

Previous field comparisons[8] on natural oyster reefs revealed no difference in sampling efficiency between oyster densities estimated using divers and those estimated using patent tongs. A similar field comparison on restored reefs in Harris Creek[9] showed that densities estimated using patent tongs resulted in statistically significantly smaller numbers of oysters than those estimated using divers. In that study[9], the densities estimated by divers were 3.35 times higher than those from hydraulic patent tongs, on average. Monitoring results in this report show oyster densities and biomass relative to the established Oyster Metrics benchmarks (e.g., minimum threshold oyster density of 15 oysters per m^2 to be considered successful). Because two different gear types were used for sampling, and results of research[8,9] on the relative sampling efficiencies of those gears vary, it may not be appropriate to use data in this report to compare relative efficacy among reef treatment types.

For both diver and patent tong data, oyster density and oyster biomass information were standardized based on area sampled. Data was then analyzed to determine success relative to each oyster metric success criteria, per the full protocols detailed in Appendix C.

Treatment Type	Reef-building substrate added?	Substrate Material	Cap Material	Reef initially seeded?	Gear type used to collect biological metrics data
Seed Only	No	None	None	Yes (spat-on-shell)	Patent tongs
Mixed shell	Yes	Mixed shell (clam, conch, and whelk)	None	Yes (spat-on-shell)	Patent tongs
Fossil shell	Yes	Fossil shell	None	Yes (spat-on-shell)	Divers
Oyster gardening reef	No	None	None	Yes (adult oysters)	Patent tongs
Stone	Yes	Amphibolite (stone)	None	Yes (spat-on-shell)	Divers
Stone topped with mixed shell	Yes	Amphibolite (stone)	Mixed shell (clam, conch, and whelk)	Yes (spat-on-shell)	Divers
Stone topped with fossil shell	Yes	Amphibolite (stone)	Fossil shell	Yes (spat-on-shell)	Divers
Reference	No	None	None	No	Patent tongs
Premet	No	None	None	No	Patent tongs

Table 2: Description of treatments used to restore reefs in Harris Creek, Little Choptank River, and Tred Avon River. Also listed is the gear type used to monitor each reef treatment type for the biological metrics (oyster density, oyster biomass, multiple year classes, and shell volume). See Section 4: Definitions for full definitions.

2.3: Revised Protocols for 2020

Overall protocols have remained largely consistent since this monitoring effort started in 2015. However, some adaptation has been required as the effort progresses. Changes from previous years' protocols are highlighted below. Full methods are described in Appendix C.

- Due to COVID-related restrictions, no data was collected on reef height and reef footprint in fall 2020. Data collection for these metrics will resume in fall 2021, assuming COVID protocols allow.

- Oyster biomass metric: As in past years, oyster biomass per m^2 was calculated from the size (shell height) of individual live oysters within each sample. In 2020, the shell height-to-biomass regression developed by Jordan et al.[10] was used for these calculations (see Appendix C for formula and full description). This is a change from past years, where the regression developed by Mann and Evans[11] was used to calculate biomass. The Workgroup determined that the Jordan et al. regression was more appropriate because it was developed using only Maryland oysters. The Mann and Evans regression, by contrast, was developed using oysters on the James River in Virginia, which may grow differently due to different ambient conditions. DNR uses the Jordan et al. regression in its biomass calculations for the annual oyster Fall Oyster Survey, so switching to the Jordan et al. regression brings the biomass calculation methodologies in this report in line with the DNR standard.

- Shell budget metric: In typical years, shell budget is assessed by comparing the current year shell volume with shell volume from three years prior. Sites that do not have significant differences between those

measurements are deemed to have a stable shell budget. Upon examining the data set from three years ago (2017), the Workgroup realized that, on the stone reefs that year, divers had not excavated the entire dive quadrat when collecting this data. This resulted in likely errors in the 2017 shell volume data set. Therefore, instead of comparing *shell volume* between 2017 and 2020, *oyster volume* (clumps and individual oysters) was compared between 2017 and 2020, as it was likely a truer representation of shell budget. Analysis of variance followed by Tukey's HSD was used to determine if changes between years were significant. Sites that did not have significant decreases between oyster volume measurements in 2017 and in 2020 were deemed to have a stable shell budget. This was done only for diver-surveyed reefs (those constructed from stone or fossil shell), as the patent tong reefs in 2017 did not experience this data issue. See Appendix A to see which reef-base material, and which monitoring gear, was used for each reef.

Section 3: Results Summary

Section 3.1: Summary of Fall 2020 Monitoring Results

On reefs monitored in fall 2020 (Table 3):

- For three-year-old reefs (Little Choptank River and Tred Avon River reefs combined; all Harris Creek reefs are older than three years):

  - 98% of restored reefs met the minimum threshold oyster density success criterion, and 50% met the higher, target density.

  - Oyster biomass tracked closely with oyster density (Figure 5).

  - 100% of restored reefs met the multiple year class criterion.

- For six-year-old reefs (Harris Creek and Little Choptank River reefs combined; no Tred Avon River reefs have matured to six years):

  - 100% of restored reefs met the minimum oyster density success criterion and 93% met the higher target density.

  - Oyster biomass tracked closely with oyster density (Figure 5).

  - 100% of restored reefs monitored met the multiple year class and shell budget criteria.

Reef Type	Tributary	# of reefs monitored in fall 2020	Oyster Density		Oyster Biomass		Multiple Year Classes	Shell budget
			% meeting minimum threshold	% meeting target	% meeting minimum threshold	% meeting target	% with multiple year classes present	% with stable/ increasing
3-year-old	All tribs combined	40	98%	50%	100%	43%	100%	TBD @ 6 years*
	Little Choptank	21	100%	95%	100%	76%	100%	TBD @ 6 years*
	Tred Avon	19	95%	0%	100%	5%	100%	TBD @ 6 years*
6-year-old	All tribs combined	27	100%	93%	100%	81%	100%	100%
	Harris Creek	14	100%	93%	100%	86%	100%	100%
	Little Choptank	13	100%	92%	100%	77%	100%	100%
Reference Reefs	Harris Creek	4	75%	25%	75%	25%	100%	NMA**
	Little Choptank	3	100%	100%	100%	100%	100%	NMA**
	Tred Avon	3	67%	0%	67%	0%	100%	NMA**

Reef shell volume at three years will be compared to that at six years to determine success relative to the shell budget metric.
**Not measured annually on reference reefs.

Table 3: *Percent of three-year-old, six-year-old, and reference reefs monitored in fall 2020 that met each Oyster Metrics success criteria. In 2020, only Little Choptank River and Tred Avon River had three-year-old reefs, and only Harris Creek and Little Choptank River had six-year-old reefs.*

Section 3.2: Summary of Cumulative Results, 2015-2020

Looking at all restored reefs monitored from 2015-2020 combined (Table 4):

- For three-year-old reefs, across all tributaries:

  - 96% of restored reefs met the minimum oyster density success criterion and 74% met the higher target oyster density.

  - Oyster biomass tracked closely with oyster density (Figure 5).

  - 100% of restored reefs met the multiple year class success criterion.

- For six-year-old reefs (Harris Creek and Little Choptank River reefs combined; no Tred Avon River reefs have matured to six years):

  - 99% of restored reefs met the minimum oyster density success criterion and 77% met the higher target density.

  - Oyster biomass tracked closely with oyster density (Figure 5).

  - 100% of restored reefs met the multiple year class and shell budget success criteria.

Reef Type	Tributary	# of reefs moitored 2015-2020	Oyster Density		Oyster Biomass		Multiple year classes	Shell budget
			% meeting minimum threshold	% meeting target	% meeting minimum threshold	% meeting target	% with multiple year classes present	% with stable/ increasing
3-year-old	All tribs combined	203 (711)	96%	74%	97%	69%	100%	TBD @ 6 years*
	Harris Creek	90 (348 acres)	98%	80%	98%	81%	100%	TBD @ 6 years*
	Little Choptank	84 (282 acres)	98%	89%	98%	76%	100%	TBD @ 6 years*
	Tred Avon	29 (81 acres)	86%	10%	90%	10%	100%	TBD @ 6 years*
6-year-old	All tribs combined	70 (310 acres)	99%	77%	99%	69%	100%	100%
	Harris Creek	57 (257 acres)	98%	74%	98%	67%	100%	100%
	Little Choptank	13 (53 acres)	100%	92%	100%	77%	100%	100%

Reef shell volume at three years will be compared to that at six years to determine success relative to the shell budget metric.

Table 4: *Percent of three-year-old and six-year-old reefs monitored from 2015-2020 that met each Oyster Metrics success criteria. Only Harris Creek and Little Choptank River have reefs that have matured to six years. See Section 5 for discussion of results.*

Section 3.3: Density-to-biomass fit plot
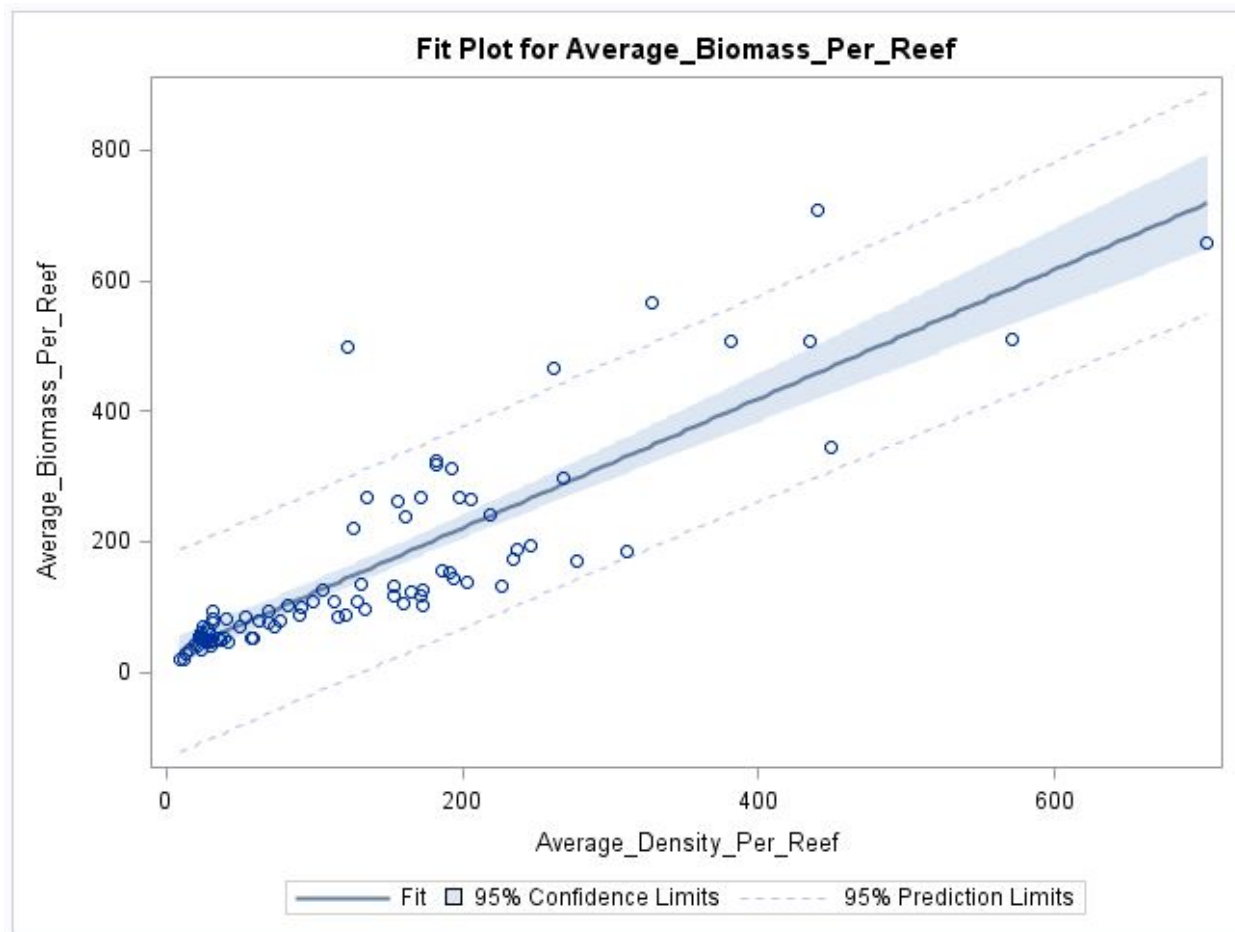
Figure 5: Fit plot describing the relationship between oyster density and oyster biomass on all reefs monitored in fall 2020. $R^2 = 0.74$, $p < 0.0001$, slope= 0.99.

Section 4: Discussion

- Trends observed in previous monitoring years generally continued in 2020, with a large majority of restored reefs meeting the Oyster Metrics success criteria.

- Per Oyster Metrics, a reef is considered successfully restored if, at six years post restoration, it meets the minimum threshold oyster density, biomass, and the other success criteria.

- Data and analysis in this report can be used by restoration partners to understand the success or failure of reefs relative to the six Oyster Metrics criteria, and to inform future restoration and adaptive management. The monitoring undertaken three years post restoration is considered an adaptive management checkpoint. Information from this interval is used by restoration partners to determine whether a reef requires the second-year-class seeding called for in each river's tributary plan, and if unsuccessful reefs should receive other management actions.

- Harris Creek was the first of the planned 10 tributaries to have its oyster restoration work completed. As of fall 2020, 57 of its 90 reefs have matured to six years—the point where, per Oyster Metrics, a reef can be considered truly 'restored' if it meets all of the Oyster Metrics success criteria. Of these 57 reefs, 56 meet the biological Oyster Metrics minimum success criteria (oyster density, oyster biomass, multiple year classes, and shell budget). Forty-two of the 57 reefs meet the higher, target oyster density and biomass success criteria. (See Table 4). Due to COVID-related restrictions, the 14 reefs in the fall 2020 six-year-old reef class were not monitored for the structural metrics (reef height and reef footprint). However, 100% of the six-year-old reefs that have been monitored as of fall 2019 for these metrics were successful[12]. This bodes well for the overall restoration success of Harris Creek. The final 33 Harris Creek reefs will turn six years old in fall 2021, and will be monitored for all Oyster Metrics success criteria.

- The percent of three-year-old reefs meeting the target oyster density is down from 85% in fall 2019 to 50% in fall 2020. This decrease is likely attributable to the number and type of reefs that were monitored in fall 2020 in the Tred Avon River. The earliest reefs built in this river were seed-only or shell-base reefs, which, per the monitoring methods used in this effort (see Appendix C), typically show lower oyster densities across all tributaries than their stone or fossil shell counterparts. These lower-density reefs reduce the cumulative percentage of reefs meeting the target density metric. Additionally, Tred Avon has historically shown lower spat sets than Harris Creek and Little Choptank River, so reefs here may not benefit from robust natural recruitment. This could also affect the ability of the Tred Avon River seed-only and shell reefs to meet the higher, target oyster density.

- 2020 was the first year that restored reefs in the Little Choptank River had matured to six years. It is encouraging to see that 100% of the reefs met the minimum Oyster Metrics density success criterion and 92% met the higher, target criterion. Initial restoration work in this tributary started in 2014 and was completed in 2020.

- In the Tred Avon River, one three-year-old reef (reef T13; 1.95 acres) did not meet the minimum Oyster Metrics density success criterion. The density on this reef in fall 2020 fell just shy of the minimum, at an average density of 13.3 oysters per m^2. The reef did, however, just meet the minimum threshold for oyster biomass, with an average biomass across the reef of 15.3 grams dry tissue weight per m^2. The restoration treatment used on this reef was seed only (see Table 2 for description). This reef will receive a second-year-class seeding in 2021 to help ensure it meets the oyster density success criterion at year six.

- Although the information in this report looks promising for success in Harris Creek, Little Choptank River, and Tred Avon River, several factors could affect continued success. These include future water-quality issues (e.g., low salinity, low river bottom dissolved oxygen levels), oyster disease, funding, and poaching (illegal oyster harvesting).

Section 5: Definitions

Fall 2020 monitoring: Monitoring undertaken on restored reefs that turned three or six years old in fall 2020. Monitoring was also done on reference reefs and sentinel reefs. Actual data collection extended from October 2020 through January 2021.

Fossil shell: Consolidated fossil oyster shell material from Florida used as a base to construct reefs. This is oyster shell cemented into a fossilized limestone, and is a true fossil, mined from 30 to 40 feet under dry land, as opposed to the Chesapeake Bay dredged shell.

Mixed shell: A mixture of scallop, conch, and clam shell from seafood processing plants.

Oyster gardening reef: A reef planted with oysters from various community-based oyster gardening programs, where volunteers grow oysters in cages hanging from docks.

Oyster Metrics: Success criteria for restored oyster reefs targeted for restoration under the 2014 Chesapeake Bay Watershed Agreement. These are defined in the report "Restoration Goals, Quantitative Metrics and Assessment Protocols for Evaluating Success on Restored Oyster Reef Sanctuaries."[2] See Table 1 for description of the six reef-level criteria.

Premet reefs: Reefs that were assumed to have met the Oyster Metrics density target criteria (50+ oysters per m^2) when surveyed prior to commencement of large-scale restoration efforts, and therefore did not initially receive further restoration treatment. However, the prerestoration data on some reefs was at an insufficient resolution to determine definitively whether or not the reefs met the density target. Thus, it is an assumption that the reefs in fact met the density success metric at that time, but it is not certain. These reefs are monitored every three years, as are other reefs, to determine appropriate adaptive management needs.

Reef restoration treatment: The particular method used to restore a reef. See Table 2 for description of reef treatment types.

Reference reefs: Reefs left unrestored (untreated) to serve as comparisons to restored (treated) reefs. Typically, these would be called 'control' reefs, but they are not true controls, as it is not possible to ensure that restoring nearby reefs would not influence these reference reefs. That is, these reefs might receive larvae from nearby restored reefs, so the term 'reference reefs' is used. Per oyster population data collected prior to commencing large-scale restoration work in Harris Creek, the reference reefs did not meet the 50 oysters per m^2 Oyster Metrics target success criterion. See Table 2 for reef treatment type relative to other treatment types.

Second-year-class seeding: A second planting of spat-on-shell some reefs receive approximately four years after initial restoration. This is intended to ensure that each reef has at least two year classes, which is an Oyster Metrics criteria. It can also help ensure that reefs meet the oyster density and biomass criteria. Second-year-class seedings are called for in each river's oyster restoration tributary plan. If a reef shows higher-than-expected oyster density when monitored three years post restoration, and a second year class is present, a second-year-class seeding may not be required.

Seed-only reefs: Reefs treated only with hatchery-produced oyster seed (spat-on-shell). No base reef-building substrate was added prior to seeding. This treatment was generally used on reefs where the prerestoration population was five oysters per m^2 or greater, but fewer than 50 oysters per m^2 (see Harris Creek Tributary Plan[2], Little Choptank Tributary Plan[3], and Tred Avon Tributary Plan[4] for detailed description of how the

Workgroup determined treatment type for each reef). See Table 2 for reef treatment type relative to other treatment types.

Sentinel reefs: A subset of the restored reefs that are monitored annually (rather than only three years and six years after restoration, which is the standard for other restored reefs). See Table 4 for reef treatment type relative to other treatment types.

Six-year-old reef: Reef that received restoration treatment in 2014, and—per Oyster Metrics and tributary plans— was monitored in 2017 (three years post restoration) and again in 2020 (six years post restoration).

Spat-on-shell: Hatchery-produced juvenile oysters attached to the shells of dead oysters. Shell typically comes from shucking houses.

Stone substrate reefs: Reefs constructed using a type of stone that is geologically classified as amphibolite. The stone was graded to fit through a six-inch mesh screen. These reefs were then seeded with spat-on-shell. See Table 2 for reef treatment type relative to other treatment types.

Stone reefs topped with mixed shell: Reefs constructed from a stone base, then capped with mixed shell and seeded with spat-on-shell. See Table 2 for reef treatment type relative to other treatment types.

Stone reefs topped with fossil shell: Reefs constructed from a stone base, then capped with fossil shell and seeded with spat-on-shell. See Table 2 for reef treatment type relative to other treatment types.

Substrate + seed reefs: Reefs treated with reef-building substrate, generally to a height of six inches to one foot above the surrounding soft bottom. Substrate was either mixed shell, fossil shell, stone, or a combination. Substrate placement was followed by planting with hatchery-produced spat-on-shell. Substrate + seed treatment type was typically used where prerestoration oyster populations were below five oysters per m^2, or where sonar surveys found no evidence of shell. See Table 2 for reef treatment type relative to other treatment types.

Three-year-old reef: Reef that received restoration treatment in 2017, and—per Oyster Metrics and tributary plans—was monitored in 2020 (three years post restoration).

References

1. Chesapeake Executive Council, 2014. Chesapeake Bay Watershed Agreement.
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Appendix A: Table of Summary Data by Reef

To access Appendix A, please click on this link to download an Excel file.

Appendix B: Length-Frequency Histogram for Each Reef

Appendix B is broken into two tables:

- B1: Length-frequency histograms for reefs monitored using divers.
- B2: Length-frequency histograms for reefs monitored using patent tongs.

To access Appendix B, please click on this link to download an Excel file.

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Appendix C: Methods for Data Collection and Analysis

This section describes methods for determining success relative to biological Oyster Metrics criteria (oyster density, oyster biomass, multiple year classes, shell budget). No data was collected in 2020 for the remaining two Oyster Metrics success criteria (reef height and reef footprint), due to COVID-related constraints.

Survey Design

A stratified random survey is used to collect biological data on restored reefs. Each reef is its own stratum, and a random number of sample points are assigned based on reef size, reducing relative error among samples. The number of samples collected at each reef is optimized for data precision and accuracy for each gear type used (Slacum et al. 2018).

- For reefs sampled using patent tongs: the number of samples increased with reef size, and averaged 6.1 samples per acre.
- For reefs sampled using divers: five samples were collected per reef, averaging 4.8 samples per acre.

ArcGIS is used to generate sampling points for each reef. All reefs that are due for monitoring are compiled into a shapefile, and samples are generated within the area of the reef that was planted with spat on shell. This ensures that sample points are created within the area that received oysters.

Field Component

Data are typically collected in the fall. The gear used depends on the reef material. Hydraulic patent tongs are used to sample on seed-only reefs, mixed-shell-base reefs, reference reefs, and premet reefs. Divers are used to sample on fossil-shell-base reefs, stone-base reefs topped with mixed shell, and stone-based reefs topped with fossil shell. Because two different gear types are employed, it is not appropriate to directly compare oyster density and biomass on reefs sampled with patent tongs versus divers (see Section 2.2: Methods summary). For both diver and patent tongs data, oyster density and oyster biomass information are standardized based on area sampled.

Sampling is conducted during daylight hours. Navigation to sampling locations and sample coordinate documentation is done using a differential global positioning system (DGPS) attached to a laptop with ArcView 10.2 used as the navigational program. The vessel navigates as closely as possible to the designated random points, and a waypoint (virtual GPS marker) is created at the location of each sample.

Patent Tongs

Hydraulic patent tongs are a specialized commercial fishing gear used to harvest oysters in the Chesapeake Bay. The patent tong design functions much like a benthic grab, collecting oysters and underlying substrate from a known fixed area of the bottom. The tongs used in 2020 sampled an area equal to 1.928 m^2 of the seafloor. The patent tongs are suspended from a boom over one side of the vessel and deployed to the bottom at each sampling location. A DGPS antenna is positioned adjacent to the location where the patent tongs are deployed, and a waypoint with the geographic coordinates of each sample location is documented.

Diver Surveys

Diver surveys are used to collect samples on reefs constructed with either a stone or fossil shell base, and are conducted by navigating the vessel to each sampling location and deploying buoys with anchors to mark each sample location. Divers descend to the bottom at each buoy with a 0.71 m x 0.71 m (0.5041 m^2) quadrat and sample collection crates. The quadrat is placed up current of the buoy anchor.

Before disturbing the reef surface, the diver makes observations on the number of oysters visible and the percent of reef substrate within the quadrat. Any material contained within the quadrat, including loose oysters, loose shell, and any reef substrate, are removed and transported to the vessel for processing.

Sample Processing

In each sample, all oysters are counted and identified as live or dead, and a minimum of 30 live oysters are measured for each sample. Oyster clumps, the number of oysters associated with a clump, and the substrate type that oysters are attached to are documented. The shell height and total count of dead (old box) and recently dead (gapers) oysters are documented from each sample. The percent of the sample covered by tunicates or mussels is documented for each sample. Additionally, field crews record the volume of each sample that is black (anoxic, shell) and measure oyster and shell volume to the nearest half liter using graduated buckets. Surface and bottom water temperature, dissolved oxygen, pH, and salinity are collected during each sampling event using a YSI Pro-Plus water quality sonde (YSI Corporation, Yellow Springs, Ohio). Other environmental and station specific variables collected at each site include sample number, date and time, weather information, depth of water, Yates Bar name, vessel name, and staff conducting the monitoring.

Data Entry and Analysis

All data are entered into a Microsoft Access database. QA/QC protocols are used to review data for nonsensical values and typos. Oyster lengths and counts are used to derive density estimates for each reef. Graphs are made to visually display size class information and proportion of live to dead oysters at the reef level. Additionally, all sample locations are plotted in ArcGIS to ensure that samples are collected on the reef footprint. Methods for analyzing data per each Oyster Metrics success criterion follow.

Oyster Density

- Oyster Metrics success criteria: Minimum threshold = 15 oysters per m^2 over 30% of the reef area; Target = 50 oysters per m^2 over 30% of the reef area.

- Method: Oyster density was calculated as the number of individual live oysters collected in the area of a patent-tong grab or diver quadrat standardized to a square meter. Total counts of live oysters or other variables (e.g., oyster size class, shell volume) were averaged over all samples collected at the individual reef. To meet the Oyster Metrics threshold or target, at least 30% of the samples collected must meet the specified densities. This represents a change from the previous survey design, in which the area of the sampled grid cells meeting the target or threshold must have been equal to or greater than 30% of the reef area. Past years of monitoring data were analyzed using this method to ensure that the methods are comparable.

Oyster Biomass

- Oyster Metrics success criteria: Minimum threshold = 15 grams dry weight per m^2 over 30% of the reef area; Target = 50 grams dry weight per m^2 over 30% of the reef area.

- Method: Oyster biomass per m^2 was calculated from the size of individual live oysters within each sample, using the regression developed by Jordan et al. (2002):

  $W = ((10^{((\log 10(L) * 2.06) - 3.76)}))$, where W = dry tissue weight in g and L = shell height in mm

This formula represents a change from previous years of monitoring, which used the regression developed by Mann and Evans. After some discussion, the Workgroup determined that the Jordan et al. regression was more appropriate since it was developed using only Maryland oysters. Biomass was then summed for the entire sample and standardized to a square meter. The biomass value is scaled based on oysters measured out of total oysters counted. The same approach as oyster density (above) was employed, in which at least 30% of samples collected had to meet the threshold or target to demonstrate restoration success.

Multiple Year Classes

- Oyster Metrics success criterion: Presence of two or more year classes of live oysters.

- Method: Year-class presence was approximated by examining length frequency data of all oyster heights measured at each reef. Sampling teams are trained to measure and record all oysters, regardless of size. For simplicity, a reef was determined to have multiple year classes when oysters from at least two standard size class categories (market: >76 mm; small: 40–75 mm; spat: <40mm) were present.

- There is no differentiation between hatchery-produced oysters and natural oysters.

Shell Budget

- Oyster Metrics success criterion: Neutral or positive shell budget on the reef.

- Method: The volume of sampled shell is measured with graduated buckets and standardized to square meter based on the area sampled by patent tong. Field measurements of shell resources included total shell volume and the percent of black (buried) shell estimated in a sample. Surface shell estimates were calculated as the percent of the total sampled shell volume that was not considered black shell, as shown below:

  Surface shell volume = Total shell volume - (Total shell volume*Percent Black Shell)

Calculating shell volume is conducted similarly for diver sampling. The volume of sampled shell is measured in graduated buckets and standardized to square meter based on the size of the diver quadrat for each sample. Alternative substrates (fossil shell, granite) are not included in this volume measurement. Again, the percent of black (buried) shell is visually estimated. Changes to the shell budget at individual reefs were analyzed by comparing shell volume data from 2017 (baseline data, when reefs were three years old) with shell volume data from 2020 (when reefs were six years old). For the 2020 data, the Workgroup reviewed 2017 shell volume data to determine if the budget was increasing or stable. It was found that 2017 volume estimates for granite sites did not involve excavating the entire dive quadrat. Therefore, members of the Workgroup concluded that oyster volume (which was assessed using the counted clumps and individuals) would be a truer representation of volume. Analysis of variance was used, followed by Tukey HSD post-hoc, to determine significant differences between years. Sites that did not have significant differences between measurements in 2017 and

measurements in 2020 were concluded to have a stable shell budget. Sites with significant increases in shell budget were also concluded to have met the metric.

Appendix C References

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