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planting seed oyster: photo credit Robert Fisher, VIMS.

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VIMS Marine Resource Report No. 2014-7

Project summary and recommendations.

This project was an academic – government – industry – non-profit collaboration wherein all parties began and ended the project with a singular commitment to stewardship of the Chesapeake Bay oyster resource for both ecological and economic purposes. The project addressed a critical issue, the potential loss of oysters to predation by cownose rays, in a controlled experimental design but at a scale commensurate with industry practices and large-scale restoration efforts. This rare and extensive collaboration was a resounding success in terms of participation and information exchange. All participating individuals and the entities they represent should be applauded for a productive project outcome.

The project focused on the value of shell overlays to oyster plantings as a ray predator deterrence mechanism. Typical industry practice of oyster seed planting was followed in an experimental design employing treatment Areas in the 0.5-1.0 acre range. Areas were prepared in the Lower Machodoc River, VA by the initial application of shell to insure a stable substrate under planted seed oysters. Seed oysters were then obtained from the James River, VA and planted using industry methods on each of four Areas. The Areas were located two upstream and two downstream of a constriction in the Lower Machodoc that dictated differing physical environments in the respective locations with downstream locations being more exposed to northeast wind driven stresses and, historically, a greater incidence of ray predation. Once oysters were planted, two of the Areas, one upstream and one downstream of the aforementioned constriction, were additionally treated with a shell overlay as a predation deterrent. The oyster seed were planted in February 2012. Market oysters were harvested in December 2013 and January 2014. In between limited monitoring of the population was accomplished using both diver and patent tong survey methods.

Final harvest data demonstrated that shell overlays do not offer additional protection to planted oyster seed with respect to possible cownose ray predation. Evidence of predation in the form of ray signatures – characteristically broken oyster valves – were recorded on all treatment Areas. Concurrent stomach content analysis of rays captured at the study location and observations of fouling community associated with the cultured oysters taken during the harvest operation indicate broad dietary preferences for rays when such a variety exists in the foraging region. Oysters are not the singular preferred diet item, although localized and intensive feeding on oysters remains an option for rays with a wide foraging range. Additionally, the overlay procedure does not appear to increase local productivity in that the exposed Areas demonstrated higher production than the “protected” Areas with shell overlay. Accordingly we recommend against the use of shell overlays as predator deterrents for cownose rays in large deployments of unprotected oyster seed.

Introduction and identification of the problem.

There are in progress enormous efforts to both restore the oyster (*Crassostrea virginica*) resource of the Chesapeake Bay for ecological purposes (driven by Executive Order 13508 originally proffering the goal of restoring 20 estuaries within the Chesapeake Bay to self sustaining oyster populations by 2025) and to rebuild the commercial oyster industry (landings from the Virginia Chesapeake Bay have increased 12-fold in the past decade). Both goals involve the placement of large numbers of oysters on the bay bottom in essentially unprotected environments that leave them subject to predation loss. Much has been debated on the role of cownose rays (*Rhinoptera bonasus*) in predation losses of oyster thus planted. Rays are opportunistic and their impacts can be locally very destructive leaving few, if any oysters alive. The question is posed as to what options are available to limit losses of oysters to ray predation? Two options exist to control losses to ray predation: reduce the number of predators and/or improve exclusion of predators. These are additive options, one does not exclude the other, but the options have differing data needs to adequately design the optimal approach, differing time courses of implementation, and differing scales of impact. A round table discussion to compare these options and examine their feasibility was hosted by the Chesapeake Bay Foundation (CBF) at the Virginia Institute of Marine Science (VIMS) on October 21, 2010. Represented were CBF, VIMS, the Virginia Marine Resources Commission (VMRC), the Virginia Seafood Council (VSC), Cowart Seafood Corporation (CSC) and Bevans Oyster Company (BOC), NOAA's Chesapeake Bay Office (NCBO), and the Elasmobranch Society (ES). A productive exchange ensued and consensus was reached on an approach that formed the basis of this project. Herein we describe the two options in brief summary, outlining why this project sought to examine the planting deterrence option in preference to the reduction fishery option, and present a description of a two year field study to test a deterrence option (overplanting density) at real world scales as a collaboration between industry, academia, non profit groups, and state and federal agencies.

Setting the stage part 1: Arguments for and against a reduction fishery.

A directed fishery for rays would reduce the number of predators and thus, it is presumed, reduce loss of oysters to predation. It is technically feasible to harvest large numbers of cownose rays, and from these develop attractive products (although the market has yet to develop to full potential and cost stability). However, any fishery is dictated by regulation to be guided by principles of management that are widely applied to established finfish fisheries. The Magnuson Stevens Fishery Conservation and Management Act (Public Law 94-265, currently under reauthorization) embodies these principles and dictates focus on sustainable harvest, which in turn is based on a quantitative knowledge of the life history of the target species and a current stock assessment. There are significant data gaps in appropriate knowledge to develop a comprehensive management plan for cownose rays in the Chesapeake Bay. What we do know is that the cownose ray is a migratory, long-lived species with large body size, low fecundity, relatively late age of maturation and distinct schooling and sex specific behavior. Even with the limited available data we can make the general statement that a sustainable harvest can only remove a modest percentage of the extant stock in any year or the prospect of stock collapse arises. In practical terms for sustainable fishery development there is a need for better life history data and a stock assessment in order to develop a management plan. This will take some time and resources, but time is of the essence in developing an oyster industry wide response to the ray threat. A sustained harvest will have a positive effect on prospects for oyster survival. The goal is worthy of pursuit, but on a relative scale the **overall** impact of harvest on oyster survival will be small, but **locally** its value could be very high. The time frame for implementation will be several years as the management plan evolves. The time and investment requirements for a fishery management plan provides only marginal comfort for both restoration advocates and the industry as the federal government invests millions of dollars and the industry is challenged daily by a suite of environmental and supply threats. A more timely control option is required.

Setting the stage part 2: Arguments for the deterrence in planting approach.

The development of exclusion approaches to guard oyster seed from cownose ray predation is not a new idea, but it has been inadequately examined to date because of scales and cost issues. The current project was designed to provide a large scale, "on the ground", economically justified experimental design that can be applied in both restoration and commercial enterprises, with data available in a reasonable time frame (2 years).

Project objective.

The **overall project objective** was to evaluate the practice of shell overplanting of oyster seed as a predator deterrence option to reduce or eliminate predation loss to cownose rays of those oyster seed when planted for restoration or commercial purposes.

General considerations in experimental design.

There are two sources of seed oyster currently available to both the restoration and commercial community. These are seed oysters from traditional shell planting actions and spat on shell (SOS) from aquaculture. Natural seed can be obtained in abundance in years of good recruitment, but they are unselected in terms of growth rate and disease tolerance, and are available with a "date of birth" that is limited to a short time window (that in turn dictates period of and maximum size at planting). Spat on shell can be both selected for growth and disease tolerance, and produced over a wider time window (this facilitating a desired size at time of planting), but they have a considerable price premium compared to natural seed. Data provided by either seed source is applicable to the other in extensive grow out application.

The present study focused on shell "over-planting", that is covering deployed seed with a light layer of shell, as a deterrent to ray predation. The design required several stages of action including (a) pre-planting preparation of the target area with shell to insure a uniform base on which the experimental study would be performed, (b) planting seed oysters by commercial large scale methods at a density commensurate with commercial practice, (c) presence or absence (control) of post plant overplanting with additional shell (understanding that this could increase subsequent harvest costs), and (d) varying harvest strategies if over-planting resulted in differing growth rates of planted oysters. Careful consideration of each of (a) through (d) was required in that each had modest ranges that, when considered in combination, provided a large matrix of final design options. Important in this design phase is realization that these "experiments" are large and difficult to implement with complete uniformity because the planting and harvest approaches use large barges and dredges – the tools of industry rather than experimental academics. This approach does, however, insure that final recommendations on optimal combinations will be applicable at real world industry or restoration scales. Indeed the impacts of cownose ray predation are typically NOT that of an individual ray, but of a school of rays with a signature in acres.

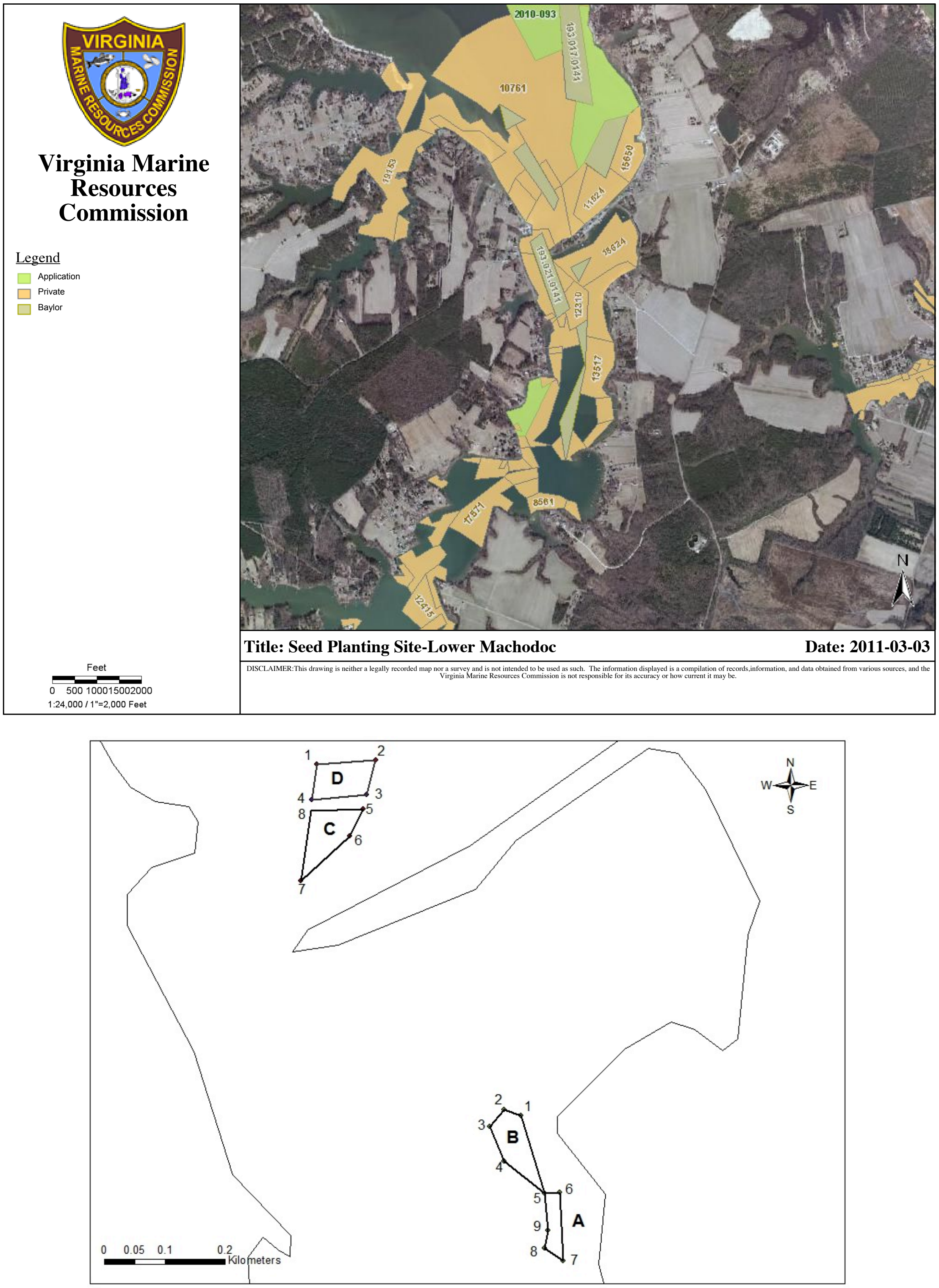
The range of values of each variable (a through d) were developed in concert with the industry partners, CSC and BOC, both of which have long standing experience at these large scales and the participating state agency (VMRC) which also has prior experience with large scale management and restoration efforts. The study was completed on commercial leases where there is history of oyster production and ray predation. We endeavored to minimize variation in lease characteristics beyond experimentally determined variables through location selection. The final design and timeline of activity is presented below in bulleted summary form and then in detail by section.

- February 2012: Completed overall site selection and marked area perimeters in the Lower Machodoc River, VA.
- February 2012: Designated experimental areas: A (low predation, with shell overlay), 0.52 acres; B (low predation, w/o shell), 1.06 acres; C (high predation, w/o shell), 1.13 acres; D (high predation with shell) 1.06 acres.
- February 2012: Completed a pre planting survey that revealed few or no shellfish or prey present.
- February 2012: Planted a shell base on all areas (9027 bushels in total, 2394 bushels /acre, 3978 bushels or 2518 bushels/acre on the low predation areas, 5049 bushels or 2305 bushels/acre on the high predation areas)
- February 15-28, 2012: Seed oysters planted (James River 2010 and 2011 recruits) at 1200 bushels/acre.
- Checked planting seed density: 3 x replicate counts on: 2/17/12 = 1056/bushels, 2/24/12 = 998/bushels, and 2/28/12 = 975/bushels.
- March 5-9, 2012: Selective overplant at 1000-2000 bushels/acre – three loads for each target area, these were large shells on the low predation area.
- April 20, 2012: Diver survey after all planting was completed.
- June 21, 2012: First ray caught.
- September 21, 2012: Diver survey.
- September 16, 2013: Patent tong final survey.
- December 2013 to January 2014: Final harvest.
- March 10, 2014: Resurvey of areas post harvest.

Site selection (February 2012).

The study site was on leased bottom at 1.5-4.0m depth maintained by Cowart Seafood Corporation (CSC) and Bevans Oyster Company (BOC) in the Lower Machodoc River, VA. This is a tributary on the southern shore of the Potomac River (see Figure 1). There is restricted access into this body of water near the mouth of the river. Historically ray predation is low upriver of this feature. Downriver of the feature the occurrence for ray predation has historically been higher. Thus prospective low and high predation sites were

Figure 1: VMRC overview map of the Lower Machodoc River, VA. Experimental areas A – D in lower graphic, see text for details, numbers correspond to Figures 8 and 9.



chosen with respect to this geographical feature. Following the pre experimental site survey, all lease sites were subject to initial shell deployment to insure a uniform base for planting - 9027 bushels in total were planted at the rates given above. The desired one-acre minimum area size was achieved at three of the four areas (A (low predation, with shell overlay), 0.52 acres; B (low predation, w/o shell), 1.06 acres; C (high predation, w/o shell), 1.13 acres; D (high predation with shell) 1.06 acres). The exception, area A, was considered a reasoned choice given that other area options required spatial separation of low predation treatments. The trade off of proximity versus size weighed in favor of the smaller but closer selection of area A.

Lease surveys prior to planting (February 2012)

This initial survey was completed using a hydraulic patent tong (one sq. meter opening) deployed from the VMRC vessel R/V J.B. Baylor using the methods described in Mann et al. (2009) for oyster stock assessment. The surveys focused on resident populations of known ray prey items (infaunal *Mya, Mercenaria, Macoma, Ensis* sp. and epifaunal mussels, crabs, and amphipods). Prior studies by investigator Fisher found stomach contents in rays captured adjacent to commercial oyster grounds were dominated by soft shell clams, mussels, and crabs, not available oysters. The surveys found minimal densities of these shellfish species in the target site areas.

The planting of a shell base on all sites (February 2012)

The shell base planting was completed with deployment of 9027 bushels in total, 2394 bushels /acre, 3978 bushels or 2518 bushels/acre on the low predation area A and B, 5049 bushels or 2305 bushels/acre on the high predation areas C and D.

Natural oyster seed were planted in the period February 15-28, 2012.

Seed oysters were obtained from the James River, Virginia. These were 2010 and 2011 recruits and were planted at a target density of 1281 bushels/acre. The density of seed per bushel (not to be confused with bushels per acre) was checked on three occasions during the seed planting process. Three replicate counts on the following dates provide the seed density estimates as follows: 2/17 = 1056 oysters/bushel, 2/24 = 998 oysters/bushel, and 2/28 = 975 oysters/bushel. A mean value (= (1056 + 998 + 975)/3) of 1010 oysters/bushel was obtained. At a planting density of 1281 bushels/acre this is equivalent to 1.3×10^6 oyster seed/ acre or 320 seed oysters/m^2 all size included. A total of 4828 bushels of seed were planted, at 1010 oyster/bushel this is an estimate total of 4,876,280 seed oysters.

Selective overplanting (March 5-9, 2012)

Shell overplanting of the target areas was completed at a density of 1000-2000 bu/acre. This was three barge loads for each area, and were large shells on the low predation area. The overplanting rate is equivalent to a uniform thickness of approximately 1-2 shells thick.

Periodic monitoring

Monitoring of the prepared areas occurred at three intervals during the study period. A diver-based survey employing quarter meter square quadrats was completed in

Spring 2012 (April, 20, 2012) after the completion of shell overplanting. A second diver-based survey was completed in Fall 2012 (September 21, 2012) at the end of the first growing season. A third and final survey was completed in Fall 2013 (September 16, 2013) prior to harvest, this time using a hydraulic patent tong with a $1m^2$ opening. Absolute density and length demographics were recorded on all occasions. The Fall 2013 survey included examination for signatures of ray feeding on the crushed valves.

A challenge with all such sampling is adequacy of the sampling protocol. For stock assessment using the aforementioned hydraulic patent tong we have previously used the recommendation of Bros and Cowell (1987) by plotting the standard error of the mean (s.e.m.) with increasing sample number (n) within a defined stratum (=area), and considering sampling to be robust (that is increased sampling produces sequentially less useful information) when the slope of the plot decreases. The location of individual samples is set by locating a virtual grid over a plat of the stratum, labeling the intersections of the grid sequentially, and choosing the sampling sequence of these intersecting points using a random number generator. In stock assessment mode our protocols define geo-references for each of the intersecting grid points and thus sampling points. These are located in the field using the GPS on the sampling vessel (usually the R/V J.B. Baylor from the VMRC fleet). For the current study a defined series of geo-referenced points sampled in sequence was not possible because the area to be sampled was very small, and the combination of error in the GPS location in the field combined with the inability to maintain the vessel exactly in the desired location (wind and tide driven) resulted in a series of effective sampling zones with considerable overlap. Thus our approach was to employ a series of haphazardly chosen sample points within the area boundaries as set in the field by the permanent corner markers. Real time plots were made of s.e.m. versus n in the field. To illustrate adequacy of sampling by this approach data plots from the Fall 2013 effort are included in the data presentations below.

Data from the three surveys is summarized in 6 sequential figures (Figures 2-7 inclusive). Raw data is provided in Appendix 1 = April2012, Appendix 2 = September2012, and Appendix 3 = September2013.

Changes in absolute density (all size classes included) for the three monitoring dates are illustrated in Figure 2. The initial survey densities for areas A through C by diver survey exhibit both considerable variance within an area and differences between area A through C and area D, with the latter having higher mean values (90-130 oysters m^{-2} versus ~320 oysters m^{-2}). The latter value is in agreement with the densities based on seed planting (see above section on natural oyster seed planting). The data emphasize the patchy nature of the planting process plus the difficulty of diver based sampling in near zero visibility. Fall 2012 sampling gave comparable mean values for density on areas A, B and D (range 90-120 oysters m^{-2}) but lower density on site C (~30 oysters m^{-2}). By the final sampling, with patent tongs, all densities were reduced to ~20-30 oysters m^{-2}. Table 1 provides the density of oysters for Fall 2013 sampling for the four areas with the associated sample number in the sequence of sampling. Figure 3 provides the associated plot of calculated s.e.m. values (vertical axis) versus sample number (horizontal axis). Note the coherence of the s.e.m. values above sample number 5 and the decreasing trend of the s.e.m. value as n increases from 5 to 10. This is in agreement with the recommendations of adequate sampling based on Bros and Cowell (1987).

Figure 2: Changes in absolute density (all size classes included) for three monitoring dates.

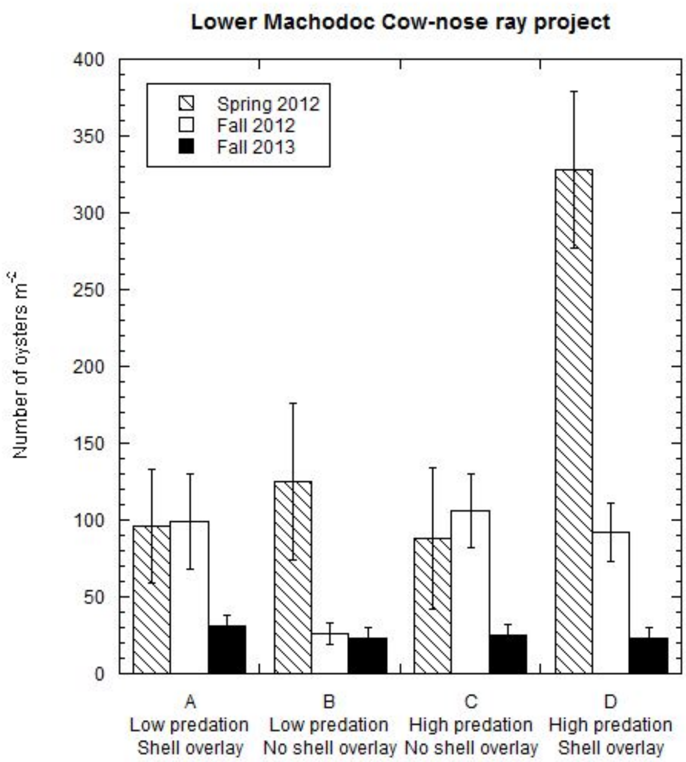
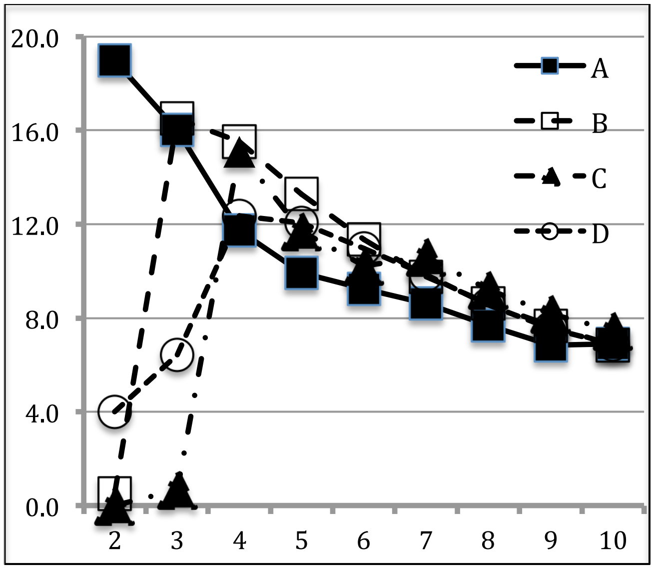


Table 1. Density of oysters, Fall 2013 sampling for areas A-D with sample number.

sample #	SITE A	B	C	D
1	5	5	0	50
2	43	4	0	58
3	59	54	2	36
4	48	62	61	2
5	19	3	13	0
6	8	6	37	0
7	5	9	62	1
8	42	33	20	25
9	22	31	16	22
10	60	25	39	34
mean	31.1	23.2	25	22.8

Figure 3. Data of Table 1 - associated plot of calculated s.e.m. values (vertical axis) versus sample number (horizontal axis) – see text.



Figures 4-7 summarize changes in population demographics (as percentage in size category versus length in 5mm intervals where length is the maximum dimension from the hinge to the growing edge) for the four experimental areas (A through D respectively in sequence) for the three monitoring dates.

A clear progression of growth in the population is evident at area A, with a shell overlay, over the time course of the study (Figure 4). Note again that the initial seed planting contained two-year classes of recruitment, 2010 and 2011, and were planted in early 2012. Thus no growth would be expected in calendar 2012. The 2010 year class is notable as a presence between 56 and 76 mm with a mode at 61-65 mm. This is commensurate with growth expectancy for the James River as described in Mann et al. (2009). The 2011 year class has a presence between 20-55 mm with highest abundance in the 26-45 mm range. By Fall 2012 the demographic spread from 46-100mm with size classes through 85 mm all being well represented. Approximately 32% were in excess of the market size of 76mm. The smallest of the 2010 year class has been "merged" with the largest of the 2011 year class by the time of this sampling. By Fall 2013 essentially all oysters were in excess of 60mm with the major peak occupying broad distribution between 70 and 120 mm – approximately 80% were in excess of the market size of 76mm.

Area B, with no shell overlay, illustrated similar post planting demographics to area A but with additional animals in the 50-70mm size classes suggesting some inclusion of 2009 year class individuals in the James River seed (Figure 5). By Fall 2012 the dominant size classes were between 51 and 90mm, with 28% at market size and another 12% in the 71-75mm size range. By Fall 2013 approximately 80% were at market size with the 86-95mm size classes being the largest by percentage.

Figure 4: population demographics, area A, low predation, with shell overlay

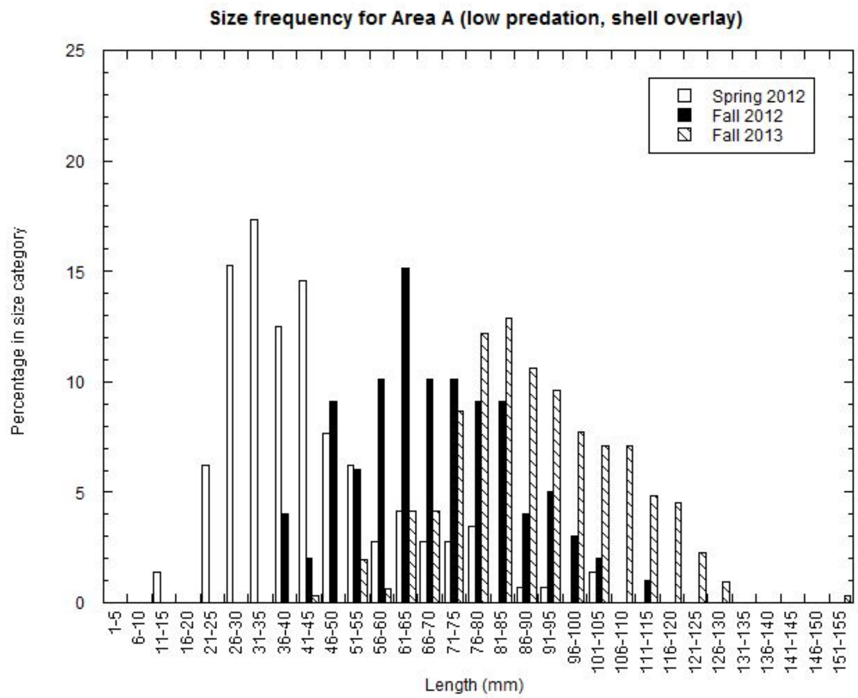
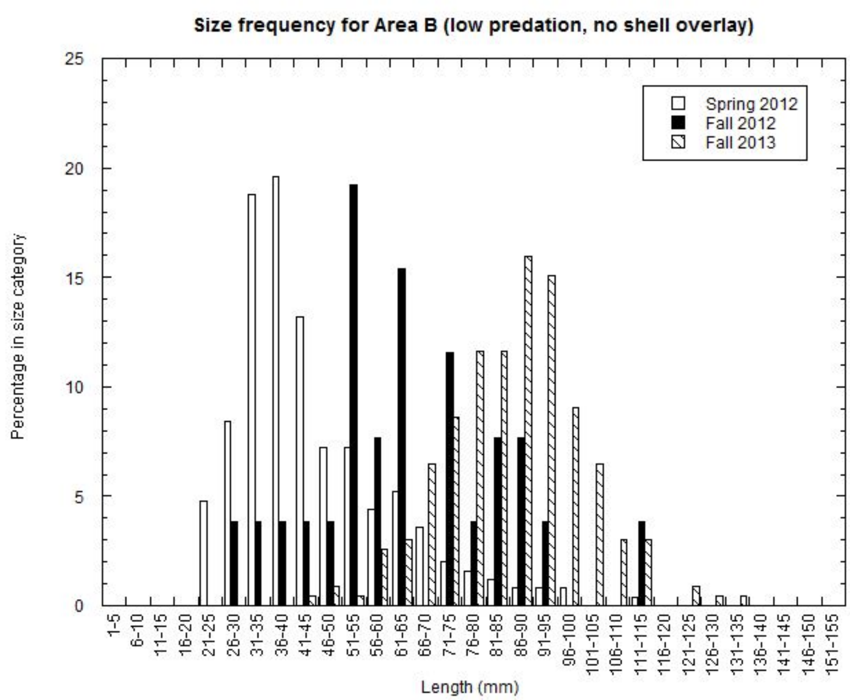
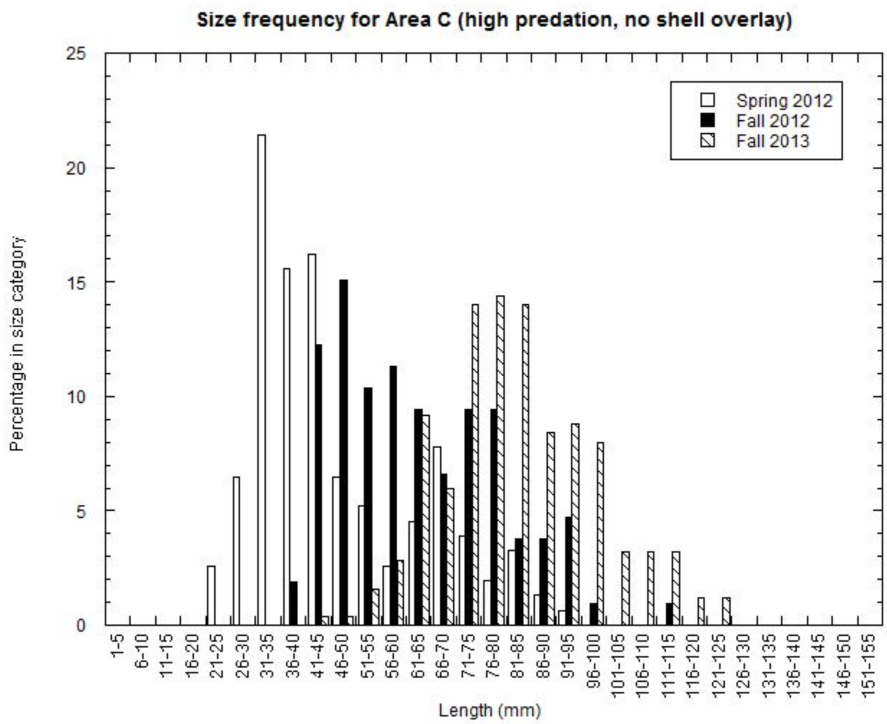


Figure 5: population demographics, area B, low predation, no shell overlay



Area C had similar demographics to area A post planting (Figure 6). By Fall 2012 the demographic had notable representation in all size classes between 41 and 80 mm. 22% of the total were at or above market size. By Fall 2013 35% were still below market size, although 14% were in the 71-75mm size class. The dominant size classes were between 71 and 85 mm, representing 43% of the total. 33% were 86mm or larger, compared to 55% for the same size interval in area B.

Figure 6: population demographics, area C, high predation, no shell overlay



Area D had, again, similar demographics to area A post planting (Figure 7). By Fall 2012 only 14% were above market size with dominant size classes at 51-60 mm and 71-75 mm representing 50% of the total demographic. By Fall 2013 38% remained below market size with the 76-95mm size classes representing 52% of the total.

Growth progression was observed in all areas, although the percentages at or above market size by the Fall 2013 pre harvest sampling was greater in areas A and B (80% in both) than in areas C and D (65% and 62%) respectively.

Figure 7: population demographics, area D, high predation, with shell overlay

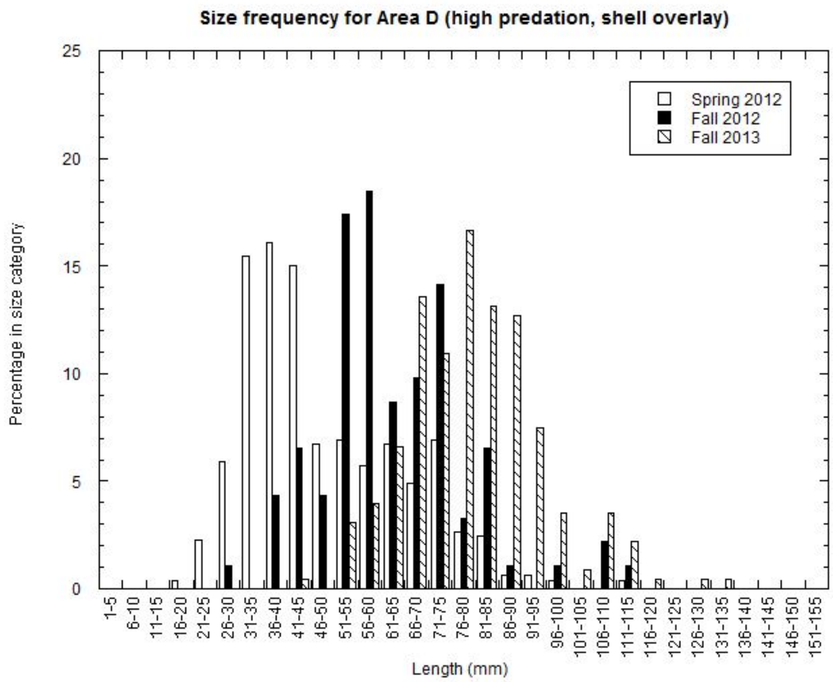


Table 2. Daily oyster harvest, in bushels, for study areas for the period 12/16/2013 through 1/9/2014. See text for methods. Yield data is in pints/bushel. Average yield over all harvest periods included

DATE	A	B	C	D	TOTALS	YIELD
12/16/13	11.0	134.0	119.5	11.0	**275.5**	**6.12**
12/17/13	0.0	139.5	126.0	0.0	**265.5**	**6.12**
12/18/13	136.0	121.0	0.0	0.0	**257.0**	**6.07**
12/19/13	0.0	111.0	135.5	0.0	**246.5**	**6.45**
12/20/13	0.0	121.0	124.0	0.0	**245.0**	**5.87**
12/21/13	0.0	98.0	0.0	112.5	**210.5**	**5.87**
12/23/13	0.0	92.5	79.0	0.0	**171.5**	**6.25**
12/26/13	0.0	73.0	64.0	0.0	**137.0**	**6.33**
12/27/13	0.0	64.0	0.0	53.5	**117.5**	**6.33**
12/30/13	0.0	33.0	37.0	0.0	**70.0**	**6.74**
12/31/13	31.5	0.0	28.5	0.0	**60.0**	**6.74**
1/2/14	27.0	31.5	0.0	0.0	**58.5**	**6.74**
1/9/14	24.0	0.0	0.0	22.0	**46.0**	**7.08**
TOTALS	**229.5**	**1,018.5**	**713.5**	**199.0**	**2,160.5**	**6.36**

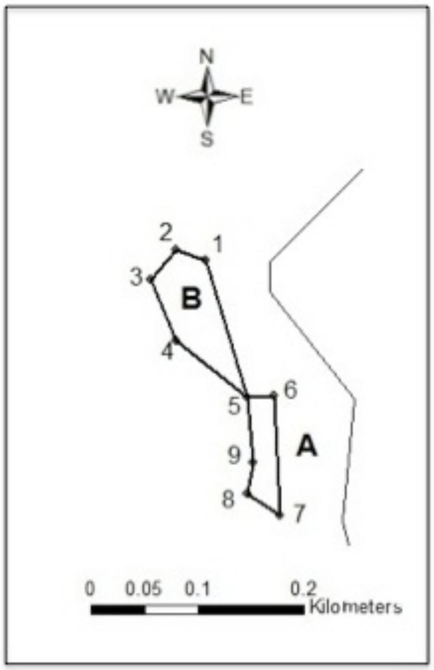
Oyster harvest was completed in late December 2013 and early January 2014 by standard methods used by the industry partners, Cowart Seafood Corporation and Bevans Oyster Company. Their collective experience has shown it prudent to harvest a complete shell plant rather than sort by size and return smaller oysters. The disturbance of mature shell plants is considered to increase losses to ray predation post return of this smaller fraction. Harvesting proceeded between December 16, 2013 and January 9, 2014 (Table 2) using a 24-inch wide oyster dredge operated from a skiff. A total of 2160.5 bushels of oysters were harvested. At an estimated 350 market oysters /bushel this is 756,175 market oysters. Given that 4828 bushels of seed oysters were initially planted the return is 0.45 bushels of market oysters for each bushel of seed planted. This is a typical industry return. Overall survival from seed to harvest is (756,175 /4,876,280) = 0.155, or 15.5%. Investigator Fisher was on board the harvest vessel on December 16 and 17, 2013 to collect data on oysters/bushel and incidence of ray signatures on broken shells included in the harvest material (Table 3).

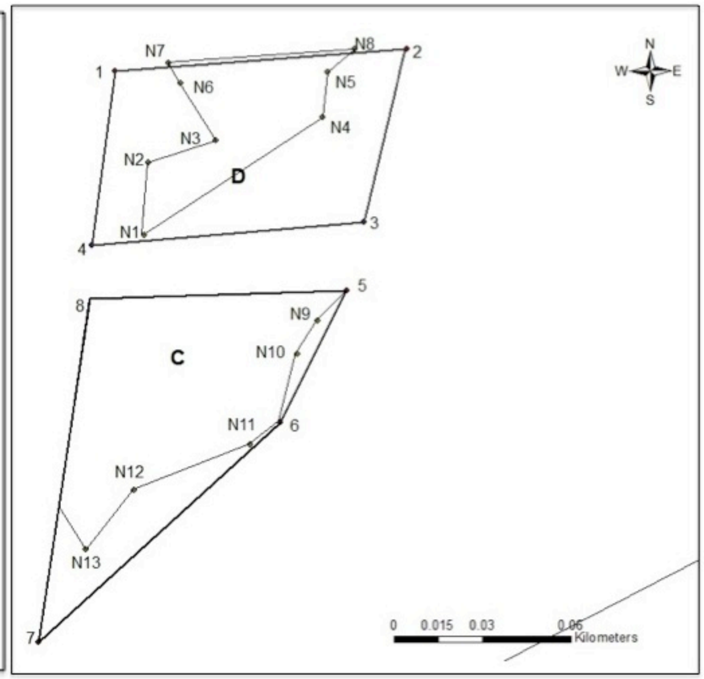
Two features of the data in Table 2 are notable. The first is the decrease in harvest per day (catch per unit effort, CPUE) in successive harvest days when full days of effort were focused on a particular area. This is to be expected. When plotted as CPUE versus day of effort (not shown) these data decline in a linear manner for each area in agreement with the expectation of a Leslie - DeLury estimator (Leslie and Davis 1939, DeLury 1947). The second feature is the marked difference in total harvest among the four areas.

Consideration of the differences in harvest by area is a stepwise process. Initially we examined just the initial planting area, harvest and presence or absence of overlay. The upstream area with shell overlay (A) had an initial area of 0.52 acres and produced 229.5 bushels of oysters. The upstream area without shell overlay (B) had an initial area of 1.06 acres and produced 1018.5 bushels. The downstream area without overlay (C) had an initial area of 1.13 acres and produced 713.5 bushels of oysters. The downstream area with shell overlay (D) had an initial area of 1.06 acres and produced 199 bushels of oysters. When expressed as per acre production values these are as follows: upstream with overlay (A) at 441 bu/acre, upstream without overlay (B) at 961 bu/acre, downstream without overlay (C) at 631 bu/acre, and downstream with overlay (D) at 188 bu/acre. These mean values are strongly indicative that a shell overlay is not conducive to oyster production.

Observations during harvest suggested that oysters were not evenly distributed or even present throughout the entire original area footprints and that actions of sand migration, siltation, burial and/or physical movement of seed by tidal and storm action may have modified the original distributions during the near two years of the study. Thus a post harvest resurveying of the four study areas was completed on March 10, 2014 with the able assistance of VMRC Engineering Surveyor Robert Butler using standard surveying techniques (Leica 1200 Series GPS, connected to Smartnet reference network) that locates to the centimeter level (finer than the ability to function from a small boat given vessel movement). The survey relocated the area perimeter markers, then ran parallel transects at <10m intervals from boundary to boundary within each area until the entire area footprint had been covered. Along the transects the bottom was sampled by sounding pole at 3-5m intervals. Where shell appeared or disappeared locations were digitally marked by the surveyor, and subsequently re-plotted as an overlay on the original survey plots. Areas A and B (Figure 8) had very minor modification as is reported in the location specific notes summarized below; however, this was not the case for areas C and D (Figure 9).

Figures 8 (left) and 9 (right): Resurvey of Areas A through D post harvest. Sequence 1-9 (left) defines original boundaries in areas A and B. Sequence 1-8 (right) defines original boundaries in areas C and D. N1-N13 define post harvest boundaries in areas C and D.





Despite the original addition of shell overlay area A was generally softer than area B Figure 8). The western perimeter, line 8 to 9 to 5, was slightly softer than the inshore eastern perimeter even though shells were still present. The possibility of some thin mud/sand cover was possible but difficult to determine with certainty, thus we have not adjusted the shell coverage area downward in subsequent density estimates (although this is debatably a very conservative stance). Area B was well supplied with shell, even extending slightly over the boundaries on the western (offshore) perimeter. The shell coverage on B was more substantial than A despite the lack of an initial overlay on B.

Area C, initially planted without a shell overlay, was only deficient in shell at its southernmost corner (point 7 in Figure 9 where hard sand prevailed). Overall the bottom is much better than Area D; the bottom is harder and there is more shell, even multiple layers of shell along the northern boundary from points 5 to 8. The depths at points N9 through N13 were 1.5, 1.8, 0.9, 1.2, and 2.1 meters respectively. The exposed shell layer was very thin throughout area D, despite being the location with an initial overlay. We estimate that approximately half of the area had lost its shell cover to burial. The sequence of data points from N1 to N8 is as follows: N1 at 3.66m has shell present. At N2 in 3.05m a sand layer had buried shell to approximately 15 cm below the surface. Transects on an east-west plane originating at locations N3 (3.05m depth), N4 (2.45m), and N5 (2.75m) all found surface shell. The northwest corner of the area had buried shell (approximately 10 cm below the surface) at location N6 (2.75m). The marginal surface shell at N7 (3.35m) had disappeared by N8 (2.75m) as the survey progressed in an easterly direction along the northern perimeter of the area.

The positions of the study areas relative to prevailing winds, tides and channel position must be considered with respect to the post harvest survey results. The revised estimates of coverage for areas C and D are 0.8 and 0.37 acres respectively. This represents 29% and 65% reductions in coverage respectively for areas C and D. Area C and especially area D are subject to northeasterly winds and storm events. Thus some loss of bottom coverage on the northerly flank of area D is explained. Oysters may have moved as part of associated bed transport from the southern flank of area D to area C. Some shell was noted beyond the western boundary of area C as would be expected from such wind stress. Perimeter losses on both the western and eastern flanks of area D and the southern corner of area C appear to be related to sand movement in shallow water. The western (deeper) boundary of area A was not, as mentioned earlier, substantially revised to lower the final coverage for Area A. Nonetheless sedimentation appears to have been prevalent across the entire footprint of area A where shell was much less prevalent in the final survey than for area B. The revised final densities at time of harvest are recalculated as follows. Area A (with initial overlay) maintained at 0.52 acres and produced 229.5 bushels of oysters. Area B (without shell overlay) maintained at 1.06 acres and produced 1018.5 bushels. Area D (with initial shell overlay) decreased to 0.37 acres and produced 199 bushels of oysters. Area C (without overlay) decreased to 0.8 acres and produced 713.5 bushels of oysters. When expressed as per acre production values the final densities are as follows: A at 441bushels /acre, B at 961 bushels/acre, C at 892 bushels/acre, and D at 537 bushels/acre. Again, the data demonstrate higher spatial production in area without initial shell overplanting.

Indirect observations of ray impact through predation signatures.

Ray predation on oysters leaves distinct crushed shell signatures. These have been observed by both industry members in their everyday operations as well as by VMRC-VIMS as part of our annual survey activity. Examples are given in Figure 10 from the Fall 2013 sampling using patent tongs. Note the clean breaks in the shell. As mentioned earlier investigator Fisher accompanied harvesters in the field on December 16 and 17, 2013 to examine the catch for signatures of ray predation. The observations are summarized in Table 3. Critical in this summary is a consideration of the topography of the bottom within the study areas and that the data were purposely collected on the first two days of harvest activity.

Figure 10. Examples of ray predation signatures in shells of consumed oysters collected from the experimental areas in Fall 2013.

During the December 16-17, 2013 period the harvesters towed dredges over the main bodies of the areas to break any crustal formation that had developed in the near two years undisturbed period since seed planting. The nature of the harvest data in Table 3 does not allow comprehensive statistical comparisons - the n values of tows are modest at 3 dredge tows sampled for each area for each of the sampling days. In all instances bushels of oysters per tow retained were in the range 0.9-1.6 inclusive of all areas on both days. Area A had a mean of 262 oysters/bushel (both days) compared to 211 oysters/bushel for area B, indicating larger oysters in area B where overlay was absent. This observation is in agreement with the Fall 2013 patent tong data. Of additional note in the harvest data is that signatures per bushel of collected material were not reduced by the presence of a shell overlay. The mean value (14.4/bushel) for area A was the highest of the four treatments compared to only 3.9/bushel in the adjacent area B. A value of 12.4/bushel was observed on 12/16 on area C, but this was reduced to 3.9 the following day. Similarly the observation for area D decreased from 5.9/bushel to 1.9/bushel from 12/16 to 12/17/2013. In addition to the observations of ray predation signatures investigator Fisher made other observations – these are briefly summarized in the comments column of table 3 and are discussed with respect to ray gut analysis in the following text section.

Table 3. Signatures of day predation observed with final oyster harvest for study areas December 16-17, 2013. All values are the means of three dredge tows for each area on each date. See text for methods.

Date	area	overlay	Oysters (bu/tow)	oyster/bu	Shell (bu)	signatures/bu	Comments
12/16/13	A	Y	1.2	229.6	1.0	18.1	center of plot,
12/17/13	A	y	1.1	293.6	1.0	10.4	shallow, less
							mussels, deeper
all	A	y	1.2	261.6	1.0	14.4	at edge
12/16/13	B		1.5	186.8	0.8	4.1	
12/17/13	B		1.8	235.0	1.0	3.8	center of plot
all	B		1.6	210.9	0.9	3.9	
12/16/13	C		0.9		1.7	12.4	center of plot. >
12/17/13	C		1.1		1.2	3.9	mussels than
							A&B. < mussels
							on 12/17 than
all	C		1.0		1.4	7.7	12/16
12/16/13	D	y	1.6		0.7	5.9	lots of mussels,
12/17/13	D	y	1.4		1.2	1.9	better oysters,
							less shell, deeper
all	D	y	1.5		0.9	4.0	at edge of area

Direct observations of ray impact through gut analysis.

Long lines (appropriately marked – this is a multi-use water body) were deployed in the planted regions to target rays for gut content examination. Long lines have been used previously for such sampling by investigator Wesson. They have relatively low efficiency. Five captured rays were examined for body morphometrics, sex, presence/absence of young in the uterus, and gut content. Investigator Fisher has worked extensively with ray biology for the past 21 years and is custodian of a substantial database on this subject. The stomach contents were considered with respect to depth-fouling continuum. The high sediment load in the Chesapeake Bay subestuaries results in rapid light attenuation with depth, thus even modest depth increments alter fouling community density and structure. These gradients in turn offer differing chemical signatures and exposed prey field to foraging rays. Data analysis must be respective of this fouling community (= alternate prey field) feature.

Cownose rays were sampled for stomach analysis using tended long line fishing gear from May through September 2012, with fishing effort directed when rays were visually observed within the research plot areas. Long lines consisted of two hundred pound monofilament ground line 100 meters in length with 2 meter gangions placed 10 meters apart with 30 size 14/0 circle hooks per line. Long lines were baited with either menhaden (*Brevoortia tyrannus*) or peeler crabs (*Callinectes sapidus*) and fished twice per day (2-4 hr. soak time) to target cownose rays. Upon capture, rays were placed on ice, boxed, and delivered to VIMS for necropsy. Stomachs were removed by severing the esophagus as it entered the peritoneal cavity at the cranial side of digestive tract and where the stomach leads into the spiral intestine on the caudal side. Removed stomachs were placed in plastic whirlpack bags, frozen, and held in freezer cold storage until processed, from 4-6 weeks after collection. Frozen stomachs were thawed in cool water within sealed sample bags for one to four hours depending on size. Once thawed, full stomach wet weights were recorded to the nearest milligram on an electronic analytical balance. The stomach contents were then emptied into a petri dish for sorting and identification and the empty stomach was weighed. The overall stomach contents weight was then calculated by difference.

With the use of field guides and taxonomic keys, prey items were identified to the lowest possible taxon and sorted for collective weights for each food category. Shell fragments of bivalves were identified to lowest possible taxon and sometimes to species if sufficient characteristic attributes were found (e.g., hinges). Enumeration of prey items was not feasible due to the level of mastication of food items. Each food category was weighed to the nearest milligram. The total weight of each food category was expressed as a percentage of the overall weight of the stomach contents. For this report, results of stomach content analysis are reported as occurrence of prey items and percent of observed prey item in stomachs with quantifiable contents. Spiral valves were sampled with stomachs to insure there were no prey items missed, especially hard prey items that are frequently retained longer within the ray digestive system. Examination of spiral valves in conjunction with stomachs provide for better enumeration of hard-bodied prey in cownose ray diet (Fisher 2010).

A total of 5 adult cownose rays, 4 females and 1 male, were collected from experimental plots in June-July 2012; 3 from the outside plots and 2 from the inside plots (Table 4). Two of the females (ray #3 and ray #4) were carrying near-term embryos, both males with disc widths of 39mm and 41.5mm, respectively. The stomachs and spiral valves of each cownose ray were analyzed for prey items. Some teleost remains were present, but were identified as bait (menhaden) and not considered a prey item. Examination of spiral valves in conjunction with stomachs provided better enumeration of hard-bodied prey in cownose ray diet. Most prey flesh remnants found in the spiral valve were beyond recognition due to advanced digestion. Retention of non-digestible hard parts of certain prey in the spiral valve was largely identifiable to at least prey category and some to species level.

Table 4. Summary of ray collections for stomach content analysis

Ray #	Date collected	sex	Disc width (DW)	Inside/outside plot	With embryo Y/N
1	6-21-2012	F	98	Inside	N
2	6-25-2012	M	83.5	Outside	N/A
3	6-25-2012	F	99	Outside	Y
4	6-25-2012	F	98	Outside	Y
5	7-18-2-12	F	101.5	Inside	N

Table 5. Content of ray stomachs and/or spiral valves by weight (g) and percentage (%) of total.

Ray #	Content	Stomach (g/%)	Spiral valve (g/%)
1	Soft shell clam (Unidentified shell)	-	4.03/99
	Detritus (woody)	-	0.04/1
3	Hard clam (*Mercenaria mercenaria*)	2.55/57	-
	Soft shell clam (*Tagelus* spp.)	0.66/15	2.18/12.4
	Fish (*Brevoorita tyrannus*)	0.28/28	-
	Hooked mussel (*Ischadium recurvum*)	-	11.25/64
	Blue crab (*Callinectes sapidus*)	-	0.20/1
	Mud crab (*Eurypanopeus depressus*)	-	0.14/.3
	Mud crab (*Rhithropanopeus harrisii*)	-	0.06/.8
	Detritus (plant matter)	-	0.25/1.4
	Rock (2mm pebbles)	-	0.09/.5
	Unidentified shell and soft tissue	-	3.45/19.6
4	Crab body parts, blue crab	-	2.77/32
	Hooked mussel (*Ischadium recurvum*)	-	1.87/21
	Soft shell clam (*Tagelus* spp.)	-	1.2/14
	Unidentified thin shell and soft tissue	-	2.1/24
	Fish (*Brevoorita tyrannus*)	-	0.82/9

From the five rays collected, stomachs were completely empty in rays 1, 2, and 5, with only a small quantity (0.42 gr) of detritus (woody) found in the stomach of ray 4. The spiral valves of rays 2 and 5 were also completely empty. Prey items were found in the stomach of ray 3, and in the spiral valves of rays 1, 3, and 4 (Table 5). Though the sample size was small in this study, prey items were dominated by thin-shelled clams (*Tagelus sp.*, *Ischadium recurvum* and *Mercenaria mercenaria*) and crustaceans (*Callinectes sapidus*, *Eurypanopeus depressus*, and *Rhithropanopeus harrisii*). Oysters were not observed as a primary prey item. These observations are in agreement with findings from a more comprehensive study of rays feeding near commercial oyster grounds by Fisher (2010).

The weight and volume of stomach and spiral valve content, when present, was relatively small with little to no soft tissue from prey items remaining (Figures 11-13). Hard structure of prey items (bivalve shell, crustacean exoskeleton) dominated content. These observations, together with those of stomachs and spiral valves void of content, suggest that rays may have been harvested between feeding periods. In such situations, soft-bodied prey items would have already been digested. This appears to be the case in each of our samples in that no benthic worms were represented. A comparison of prey diversity between areas is limited because rays 1 and 5 (collected from inside plots) were devoid of prey items in their stomachs; however, *Tagelus* sp. was recorded in both the spiral valve of ray 5 and in rays 3 and 4 collected from outside plots. Menhaden and blue crab items found in rays in this study are most likely the result of bait ingested upon capture.

Figure11. Content from cownose ray (Ray #1) spiral valve with only soft clam (*Tagelus* sp.) represented.

Figure 12. Content found in cownose ray (Ray #3) stomach and spiral valve. cr-Crustacean (mud crab, blue crab); f-fish (menhaden, bait); hc-hard clam; sc-soft clam (*Tagelus* sp.); m-mussel (hooked mussel); ui-unidentified shell material; r-rock.

Figure 13. Content from cownose ray (Ray #4) spiral valve. cr-Crustacean (blue crab); f-fish (menhaden, bait); hc-hard clam; sc-soft clam (*Tagelus* sp.); m-mussel (hooked mussel); ui-unidentified shell material; r-rock.

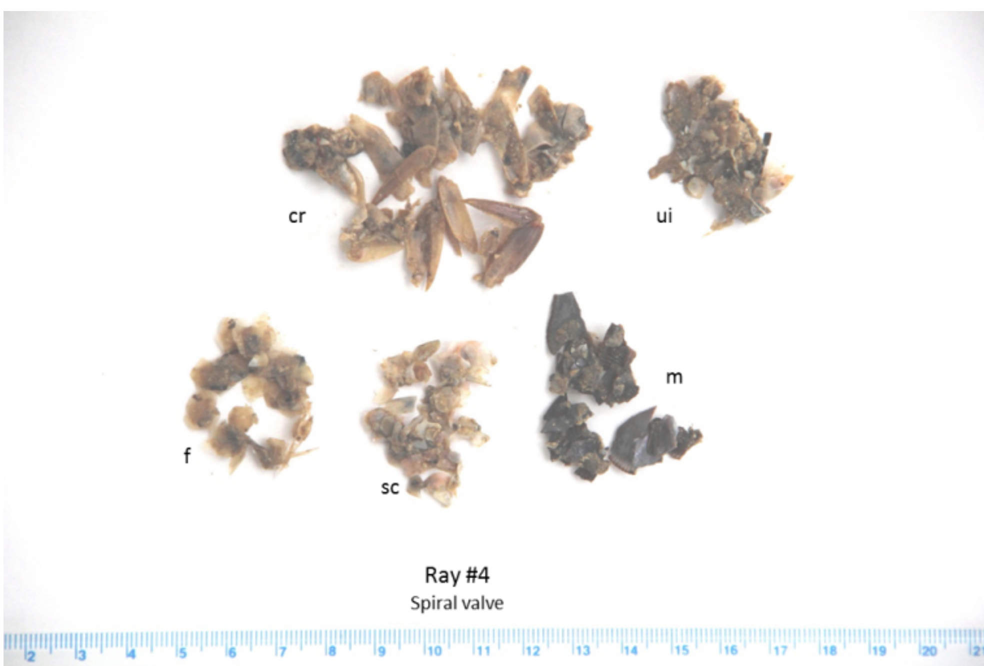


Figure 14. Oysters at harvest (12-17-2013): left, research Areas A/B with small amount of mussel fouling observed; right, research Areas C/D with heavy mussel fouling observed in shallower areas within Area.

Figure 15. Oysters at harvest (12-17-2013) showing heavy mussel fouling. Inset showing a cluster of 4 oysters

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Appendices

Field data, oyster density by demographics, 5 mm intervals of shell length (SL). Data given with date of collection, method (diver or patent tong), sample size (0.25 m^2 or 1.0 m^2 respectively), and sample number of the date. Values are presented for mean, standard error of the mean (sem) for each size interval, and as a percentage of the total (%).

Appendix 1. April2012
Appendix 2. September2012
Appendix 3. September2013

Appendix 1. April 2012, diver sample, 0.25m^2, Area A

live oyster	sample number								
SL (mm)	1	2	3	4	5	6	mean	sem	%
1-5	0	0	0	0	0	0	0.0	0.0	0
6-10	0	0	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0	2	0.3	0.3	1
16-20	0	0	0	0	0	0	0.0	0.0	0
21-25	1	0	0	4	1	3	1.5	0.7	6
26-30	4	0	0	9	0	9	3.7	1.8	15
31-35	7	0	0	4	1	13	4.2	2.1	17
36-40	6	0	0	6	1	5	3.0	1.2	13
41-45	10	0	0	7	1	3	3.5	1.7	15
46-50	1	1	0	3	2	4	1.8	0.6	8
51-55	2	0	0	3	1	3	1.5	0.6	6
56-60	3	0	0	0	0	1	0.7	0.5	3
61-65	1	0	0	3	0	2	1.0	0.5	4
66-70	0	0	0	3	0	1	0.7	0.5	3
71-75	1	0	0	0	1	2	0.7	0.3	3
76-80	3	0	0	0	1	1	0.8	0.5	3
81-85	0	0	0	0	0	0	0.0	0.0	0
86-90	0	0	0	0	0	1	0.2	0.2	1
91-95	0	0	0	0	0	1	0.2	0.2	1
96-100	0	0	0	0	0	0	0.0	0.0	0
101-105	0	0	0	1	1	0	0.3	0.2	1
106-110	0	0	0	0	0	0	0.0	0.0	0
111-115	0	0	0	0	0	0	0.0	0.0	0
116-120	0	0	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0	0	0.0	0.0	0
total	39	1	0	43	10	51	24	9.3	
live shell	2	0.1	0.1	2	2	3	1.53	0.48	
blank shell	3	7	10	5	7	5	6.17	0.98	
oysters/m^2	156	4	0	172	40	204	96	37.35	

Appendix 1. April 2012, diver sample, 0.25m², Area B

live oyster	sample number									
SL (mm)	1	2	3	4	5	6	7	8	mean	sem	%
1-5	0	0	0	0	0	0	0	0	0	0	0
6-10	0	0	0	0	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0	0	0	0	0.0	0.0	0
16-20	0	0	0	0	0	0	0	0	0.0	0.0	0
21-25	2	0	0	0	0	1	3	6	1.5	0.8	5
26-30	0	0	3	0	0	5	9	4	2.6	1.2	8
31-35	4	0	1	0	0	11	20	11	5.9	2.6	19
36-40	0	0	4	0	0	10	20	15	6.1	2.8	20
41-45	3	0	3	1	0	7	14	5	4.1	1.7	13
46-50	1	0	2	0	0	2	6	7	2.3	1.0	7
51-55	0	0	1	1	0	5	6	5	2.3	0.9	7
56-60	1	0	0	0	0	3	4	3	1.4	0.6	4
61-65	0	0	0	1	0	0	5	7	1.6	1.0	5
66-70	0	0	0	0	0	2	4	3	1.1	0.6	4
71-75	0	0	3	0	0	1	1	0	0.6	0.4	2
76-80	0	0	1	0	0	1	1	1	0.5	0.2	2
81-85	0	0	0	0	0	1	1	1	0.4	0.2	1
86-90	0	0	0	0	0	2	0	0	0.3	0.3	1
91-95	0	0	0	0	0	2	0	0	0.3	0.3	1
96-100	0	0	0	0	0	0	0	2	0.3	0.3	1
101-105	0	0	0	0	0	0	0	0	0.0	0.0	0
106-110	0	0	0	0	0	0	0	0	0.0	0.0	0
111-115	0	0	1	0	0	0	0	0	0.1	0.1	0
116-120	0	0	0	0	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0	0	0	0	0.0	0.0	0
total	11	0	19	3	0	53	94	70	31.25	12.83	
live shell	0.1	0.25	0.5	0.1	0	2	3	3	1.12	0.47	
blank shell	2	3	9.5	5	3	2	3	0.5	3.50	0.97	
oysters/m²	44	0	76	12	0	212	376	280	125	51.34	

Appendix 1. April 2012, diver sample, 0.25m^2, Area C

live oyster	sample number									
SL (mm)	1	2	3	4	5	6	7	mean	sem	%
1-5	0	0	0	0	0	0	0	0.0	0.0	0
6-10	0	0	0	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0	0	0	0.0	0.0	0
16-20	0	0	0	0	0	0	0	0.0	0.0	0
21-25	0	0	0	0	0	0	4	0.6	0.6	3
26-30	4	1	0	0	0	0	5	1.4	0.8	6
31-35	18	0	1	0	0	0	14	4.7	2.9	21
36-40	9	0	2	0	0	1	12	3.4	1.9	16
41-45	12	0	4	0	1	0	8	3.6	1.8	16
46-50	6	0	0	0	0	0	4	1.4	0.9	6
51-55	3	0	2	0	0	0	3	1.1	0.6	5
56-60	0	0	0	0	1	0	3	0.6	0.4	3
61-65	3	0	2	0	0	0	2	1.0	0.5	5
66-70	4	0	2	0	0	1	5	1.7	0.8	8
71-75	2	1	1	0	0	0	2	0.9	0.3	4
76-80	0	0	0	0	0	0	3	0.4	0.4	2
81-85	0	0	2	0	0	0	3	0.7	0.5	3
86-90	0	0	0	0	0	1	1	0.3	0.2	1
91-95	0	0	0	0	0	0	1	0.1	0.1	1
96-100	0	0	0	0	0	0	0	0.0	0.0	0
101-105	0	0	0	0	0	0	0	0.0	0.0	0
106-110	0	0	0	0	0	0	0	0.0	0.0	0
111-115	0	0	0	0	0	0	0	0.0	0.0	0
116-120	0	0	0	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0	0	0	0.0	0.0	0
total	61	2	16	0	2	3	70	22	11.45	
live shell	0.1	0.3	1.0	0.1	1.0	1.0	4.0	1.1	0.5	
blank shell	3	2	3	5	6	3	0	3	1	
oysters/m^2	244	8	64	0	8	12	280	88	46	

Appendix 1. April 2012, diver sample, 0.25m^2, Area D

live oyster	sample number								
SL (mm)	1	2	3	4	5	6	mean	sem	%
1-5	0	0	0	0	0	0	0.0	0.0	0
6-10	0	0	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0	0	0.0	0.0	0
16-20	0	1	0	0	1	0	0.3	0.2	0
21-25	1	2	2	2	2	2	1.8	0.2	2
26-30	7	6	0	7	2	7	4.8	1.2	6
31-35	14	19	6	6	15	16	12.7	2.2	15
36-40	13	18	5	15	13	15	13.2	1.8	16
41-45	19	10	7	20	6	12	12.3	2.4	15
46-50	8	11	1	2	4	7	5.5	1.6	7
51-55	9	10	1	7	1	6	5.7	1.6	7
56-60	5	4	5	6	2	6	4.7	0.6	6
61-65	9	6	2	6	1	9	5.5	1.4	7
66-70	10	6	1	4	1	2	4.0	1.4	5
71-75	12	6	1	9	2	4	5.7	1.7	7
76-80	1	4	2	3	2	1	2.2	0.5	3
81-85	1	4	2	3	2	0	2.0	0.6	2
86-90	1	2	0	0	0	0	0.5	0.3	1
91-95	0	1	1	1	0	0	0.5	0.2	1
96-100	1	1	0	0	0	0	0.3	0.2	0
101-105	0	0	0	0	0	0	0.0	0.0	0
106-110	0	0	0	0	0	0	0.0	0.0	0
111-115	2	0	0	0	0	0	0.3	0.3	0
116-120	0	0	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0	0	0.0	0.0	0
total	113	111	36	91	54	87	82	13	100
live shell	7	5	2	7	2	3	4	1	
blank shell	2	2	5	3	3	1	3	1	
oysters/m^2	452	444	144	364	216	348	328	51	

Appendix 2. September 2012, diver sample, 0.25m^2, Area A, only samples number 1-4 included in estimating mean and sem values

live oyster	sample number							
SL (mm)	1	2	3	4	5	mean	sem	%
1-5	0	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0	0.0	0
26-30	0	0	0	0	0	0	0.0	0
31-35	0	0	0	0	0	0	0.0	0
36-40	1	1	0	2	0	0.8	0.4	3
41-45	1	1	0	0	0	0.4	0.2	2
46-50	4	4	0	1	0	1.8	0.9	7
51-55	4	2	0	0	0	1.2	0.8	5
56-60	3	4	0	3	0	2	0.8	8
61-65	3	7	3	2	0	3	1.1	12
66-70	3	7	0	0	0	2	1.4	8
71-75	3	5	0	2	0	2	0.9	8
76-80	3	2	1	3	0	1.8	0.6	7
81-85	4	1	0	4	0	1.8	0.9	7
86-90	1	1	0	2	0	0.8	0.4	3
91-95	2	1	0	2	0	1	0.4	4
96-100	1	1	0	1	0	0.6	0.2	2
101-105	2	0	0	0	0	0.4	0.4	2
106-110	0	0	0	0	0	0	0.0	0
111-115	0	0	0	1	0	0.2	0.2	1
116-120	0	0	0	0	0	0	0.0	0
121-125	0	0	0	0	0	0	0.0	0
126-130	0	0	0	0	0	0	0.0	0
131-135	0	0	0	0	0	0	0.0	0
136-140	0	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0	0.0	0
total	35	37	4	23	0	24.75	7.68	
live shell	4	4	1	3	0	2.22	0.80	
blank shell	2	0	4	1	3	2.00	0.69	
oysters/m^2	140	148	16	92	0	99.00	30.71	

Appendix 2. September 2012, diver sample, 0.25m², Area B

live oyster	sample number						
SL (mm)	1	2	3	4	mean	sem	%
1-5	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0.0	0
26-30	0	0	0	1	0.25	0.3	4
31-35	0	0	1	0	0.25	0.3	4
36-40	0	0	1	0	0.25	0.3	4
41-45	1	0	0	0	0.25	0.3	4
46-50	0	0	0	1	0.25	0.3	4
51-55	2	0	0	3	1.25	0.8	19
56-60	0	0	0	2	0.5	0.5	8
61-65	0	1	1	2	1	0.4	15
66-70	0	0	0	0	0	0.0	0
71-75	1	0	1	1	0.75	0.3	12
76-80	0	1	0	0	0.25	0.3	4
81-85	1	0	0	1	0.5	0.3	8
86-90	0	2	0	0	0.5	0.5	8
91-95	0	1	0	0	0.25	0.3	4
96-100	0	0	0	0	0	0.0	0
101-105	0	0	0	0	0	0.0	0
106-110	0	0	0	0	0	0.0	0
111-115	0	0	0	1	0.25	0.3	4
116-120	0	0	0	0	0	0.0	0
121-125	0	0	0	0	0	0.0	0
126-130	0	0	0	0	0	0.0	0
131-135	0	0	0	0	0	0.0	0
136-140	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0.0	0
total	5	5	4	12	6.5	1.8	
live shell	0.5	0.5	1	1	0.8	0.1	
blank shell	3	4	6	1	3.5	1.0	
oysters/m²	20	20	16	48	26.0	7.4	

Appendix 2. September 2012, diver sample, 0.25m², Area C

live oyster	sample number						
SL (mm)	1	2	3	4	mean	sem	%
1-5	0	0	0	0	0.0	0.0	0
6-10	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0.0	0.0	0
16-20	0	0	0	0	0.0	0.0	0
21-25	0	0	0	0	0.0	0.0	0
26-30	0	0	0	0	0.0	0.0	0
31-35	0	0	0	0	0.0	0.0	0
36-40	1	1	0	0	0.5	0.3	2
41-45	8	1	2	2	3.3	1.6	12
46-50	6	2	4	4	4.0	0.8	15
51-55	5	1	5	0	2.8	1.3	10
56-60	4	4	1	3	3.0	0.7	11
61-65	5	3	2	0	2.5	1.0	9
66-70	4	0	1	2	1.8	0.9	7
71-75	4	1	3	2	2.5	0.6	9
76-80	3	4	0	3	2.5	0.9	9
81-85	0	3	1	0	1.0	0.7	4
86-90	0	1	2	1	1.0	0.4	4
91-95	2	3	0	0	1.3	0.8	5
96-100	1	0	0	0	0.3	0.3	1
101-105	0	0	0	0	0.0	0.0	0
106-110	0	0	0	0	0.0	0.0	0
111-115	1	0	0	0	0.3	0.3	1
116-120	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0.0	0.0	0
total	44	24	21	17	26.5	6.0	
live shell	4	3	4	2	3.3	0.5	
blank shell	0.75	7	2	8	4.4	1.8	
oysters/m²	176	96	84	68	106.0	24.0	

Appendix 2. September 2012, diver sample, 0.25m^2, Area D

live oyster	sample number								
SL (mm)	1	2	3	4	5	6	7	mean	sem	%
1-5	0	0	0	0	0	0	0	0.0	0.0	0
6-10	0	0	0	0	0	0	0	0.0	0.0	0
11-15	0	0	0	0	0	0	0	0.0	0.0	0
16-20	0	0	0	0	0	0	0	0.0	0.0	0
21-25	0	0	0	0	0	0	0	0.0	0.0	0
26-30	0	0	0	0	1	0	0	0.1	0.1	1
31-35	0	0	0	0	0	0	0	0.0	0.0	0
36-40	0	0	0	0	2	1	1	0.6	0.3	4
41-45	0	0	0	0	4	0	2	0.9	0.6	7
46-50	0	0	0	0	1	2	1	0.6	0.3	4
51-55	0	0	0	2	3	5	6	2.3	0.9	17
56-60	0	0	0	2	5	4	6	2.4	1.0	18
61-65	0	0	0	1	5	1	1	1.1	0.7	9
66-70	0	0	0	0	2	1	6	1.3	0.8	10
71-75	0	0	0	3	4	3	3	1.9	0.7	14
76-80	0	0	0	1	1	0	1	0.4	0.2	3
81-85	0	0	0	1	3	2	0	0.9	0.5	7
86-90	0	0	0	0	1	0	0	0.1	0.1	1
91-95	0	0	0	0	0	0	0	0.0	0.0	0
96-100	0	0	0	0	0	1	0	0.1	0.1	1
101-105	0	0	0	0	0	0	0	0.0	0.0	0
106-110	0	0	0	0	0	1	1	0.3	0.2	2
111-115	0	0	0	0	0	1	0	0.1	0.1	1
116-120	0	0	0	0	0	0	0	0.0	0.0	0
121-125	0	0	0	0	0	0	0	0.0	0.0	0
126-130	0	0	0	0	0	0	0	0.0	0.0	0
131-135	0	0	0	0	0	0	0	0.0	0.0	0
136-140	0	0	0	0	0	0	0	0.0	0.0	0
141-145	0	0	0	0	0	0	0	0.0	0.0	0
146-150	0	0	0	0	0	0	0	0.0	0.0	0
total	0	0	0	10	32	22	28	13.1	7.0	
live shell	0	0	0	2	2	3	4	1.6	0.6	
blank shell	4	4.5	6	3	7	6	3	4.8	0.6	
oysters/m^2	0	0	0	40	128	88	112	52.6	28.1	

Appendix 3. September 2013, patent tong sample, 1.0 m^2, Area A

live oyster	sample number											
SL (mm)	1	2	3	4	5	6	7	8	9	10	mean	sem	%
1-5	0	0	0	0	0	0	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0	0	0	0	0	0	0.0	0
26-30	0	0	0	0	0	0	0	0	0	0	0	0.0	0
31-35	0	0	0	0	0	0	0	0	0	0	0	0.0	0
36-40	0	0	0	0	0	0	0	0	0	0	0	0.0	0
41-45	0	0	0	1	0	0	0	0	0	0	0.1	0.1	0
46-50	0	0	0	0	0	0	0	0	0	0	0	0.0	0
51-55	0	0	0	2	1	0	1	0	0	2	0.6	0.3	2
56-60	0	1	0	0	0	0	0	1	0	0	0.2	0.1	1
61-65	1	2	3	2	0	1	0	2	0	2	1.3	0.3	4
66-70	0	2	4	1	2	0	0	1	2	1	1.3	0.4	4
71-75	0	5	5	6	1	0	0	1	3	6	2.7	0.8	9
76-80	1	4	8	5	1	1	2	5	2	9	3.8	0.9	12
81-85	0	4	4	8	1	1	0	10	5	7	4	1.1	13
86-90	1	10	3	4	1	0	1	3	2	8	3.3	1.0	11
91-95	0	2	10	5	0	0	1	4	1	7	3	1.1	10
96-100	1	3	3	5	2	0	0	6	1	3	2.4	0.6	8
101-105	1	3	4	2	2	1	0	1	2	6	2.2	0.6	7
106-110	0	2	4	2	3	3	0	4	2	2	2.2	0.4	7
111-115	0	1	6	1	2	0	0	1	0	4	1.5	0.6	5
116-120	0	2	1	4	3	0	0	1	1	2	1.4	0.4	5
121-125	0	1	4	0	0	0	0	1	1	0	0.7	0.4	2
126-130	0	1	0	0	0	1	0	1	0	0	0.3	0.2	1
131-135	0	0	0	0	0	0	0	0	0	0	0	0.0	0
136-140	0	0	0	0	0	0	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0	0	0	0	0	0	0.0	0
151-155	0	0	0	0	0	0	0	0	0	1	0.1	0.1	0
oysters/m^2	5	43	59	48	19	8	5	42	22	60	31.1	6.9	100
live vol (L)	1	10	12	10	5	2	0	10	3	10	6.3	1.4	
blank vol (L)	5	8	8	16	11	8	6	8	4	15	8.9	1.3	
ray boxes	1	3	2	6	3	0	3	0	3	2	2.3	0.6	

Appendix 3. September 2013, patent tong sample, 1.0 m², Area B

live oyster	sample number											
SL (mm)	1	2	3	4	5	6	7	8	9	10	mean	sem	%
1-5	0	0	0	0	0	0	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0	0	0	0	0	0	0.0	0
26-30	0	0	0	0	0	0	0	0	0	0	0	0.0	0
31-35	0	0	0	0	0	0	0	0	0	0	0	0.0	0
36-40	0	0	0	0	0	0	0	0	0	0	0	0.0	0
41-45	0	0	0	0	0	0	0	1	0	0	0.1	0.1	0
46-50	0	0	0	1	0	0	0	0	1	0	0.2	0.1	1
51-55	0	0	0	0	0	0	0	1	0	0	0.1	0.1	0
56-60	0	0	0	2	0	0	1	1	1	1	0.6	0.2	3
61-65	0	0	1	4	0	0	0	1	0	1	0.7	0.4	3
66-70	1	0	4	3	0	0	2	2	2	1	1.5	0.4	6
71-75	1	0	4	5	0	1	1	3	3	2	2	0.5	9
76-80	1	1	2	6	0	0	0	5	8	4	2.7	0.9	12
81-85	0	1	5	6	0	1	1	3	5	5	2.7	0.7	12
86-90	0	0	12	13	0	0	0	5	4	3	3.7	1.6	16
91-95	0	0	12	11	1	1	2	2	4	2	3.5	1.4	15
96-100	0	1	2	6	2	1	1	5	1	2	2.1	0.6	9
101-105	2	0	5	1	0	1	1	0	1	4	1.5	0.5	6
106-110	0	1	2	2	0	1	0	1	0	0	0.7	0.3	3
111-115	0	0	4	2	0	0	0	1	0	0	0.7	0.4	3
116-120	0	0	0	0	0	0	0	0	0	0	0	0.0	0
121-125	0	0	1	0	0	0	0	1	0	0	0.2	0.1	1
126-130	0	0	0	0	0	0	0	1	0	0	0.1	0.1	0
131-135	0	0	0	0	0	0	0	0	1	0	0.1	0.1	0
136-140	0	0	0	0	0	0	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0	0	0	0	0	0	0.0	0
151-155	0	0	0	0	0	0	0	0	0	0	0	0.0	0
oysters/m²	5	4	54	62	3	6	9	33	31	25	23.2	6.9	100
live vol (L)	1	0.25	10	10	0.1	0.75	0.5	7	6	6	4.2	1.3	
blank vol (L)	4	16	11	21	6	7	9	11	12	8	10.5	1.6	
ray boxes	0	0	0	2	0	2	0	0	3	2	0.9	0.4	

Appendix 3. September 2013, patent tong sample, 1.0 m², Area C

live oyster					sample number								
SL (mm)	1	2	3	4	5	6	7	8	9	10	mean	sem	%
1-5	0	0	0	0	0	0	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0	0	0	0	0	0	0.0	0
26-30	0	0	0	0	0	0	0	0	0	0	0	0.0	0
31-35	0	0	0	0	0	0	0	0	0	0	0	0.0	0
36-40	0	0	0	0	0	0	0	0	0	0	0	0.0	0
41-45	0	0	0	1	0	0	0	0	0	0	0.1	0.1	0
46-50	0	0	0	0	0	0	1	0	0	0	0.1	0.1	0
51-55	0	0	1	2	0	0	0	1	0	0	0.4	0.2	2
56-60	0	0	0	3	0	0	2	1	1	0	0.7	0.3	3
61-65	0	0	0	6	2	2	4	3	1	5	2.3	0.7	9
66-70	0	0	0	4	0	2	4	1	0	4	1.5	0.6	6
71-75	0	0	1	12	3	6	6	5	0	2	3.5	1.2	14
76-80	0	0	0	8	2	6	13	2	2	3	3.6	1.3	14
81-85	0	0	0	6	2	7	8	1	3	8	3.5	1.1	14
86-90	0	0	0	4	0	4	1	3	2	7	2.1	0.8	8
91-95	0	0	0	2	2	2	7	0	4	5	2.2	0.8	9
96-100	0	0	0	5	0	4	6	2	1	2	2	0.7	8
101-105	0	0	0	3	0	0	3	0	1	1	0.8	0.4	3
106-110	0	0	0	2	1	1	2	1	1	0	0.8	0.2	3
111-115	0	0	0	2	1	3	2	0	0	0	0.8	0.4	3
116-120	0	0	0	1	0	0	1	0	0	1	0.3	0.2	1
121-125	0	0	0	0	0	0	2	0	0	1	0.3	0.2	1
126-130	0	0	0	0	0	0	0	0	0	0	0	0.0	0
131-135	0	0	0	0	0	0	0	0	0	0	0	0.0	0
136-140	0	0	0	0	0	0	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0	0	0	0	0	0	0.0	0
151-155	0	0	0	0	0	0	0	0	0	0	0	0.0	0
oysters/m²	0	0	2	61	13	37	62	20	16	39	25	7.5	100
live vol (L)	0	0	0	11	3	8	13	5	2	8	5.0	1.5	
blank vol (L)	2	16	12	17	16	11	8	7	8	12	10.9	1.5	
ray boxes	0	0	1	2	0	0	4	1	0	1	0.9	0.4	

Appendix 3. September 2013, patent tong sample, 1.0 m², Area D

live oyster	sample number											
SL (mm)	1	2	3	4	5	6	7	8	9	10	mean	sem	%
1-5	0	0	0	0	0	0	0	0	0	0	0	0.0	0
6-10	0	0	0	0	0	0	0	0	0	0	0	0.0	0
11-15	0	0	0	0	0	0	0	0	0	0	0	0.0	0
16-20	0	0	0	0	0	0	0	0	0	0	0	0.0	0
21-25	0	0	0	0	0	0	0	0	0	0	0	0.0	0
26-30	0	0	0	0	0	0	0	0	0	0	0	0.0	0
31-35	0	0	0	0	0	0	0	0	0	0	0	0.0	0
36-40	0	0	0	0	0	0	0	0	0	0	0	0.0	0
41-45	0	1	0	0	0	0	0	0	0	0	0.1	0.1	0
46-50	0	0	0	0	0	0	0	0	0	0	0	0.0	0
51-55	3	1	0	0	0	0	0	1	0	2	0.7	0.3	3
56-60	1	5	2	0	0	0	0	0	1	0	0.9	0.5	4
61-65	8	2	3	0	0	0	0	2	0	0	1.5	0.8	7
66-70	6	4	5	0	0	0	1	5	2	8	3.1	0.9	14
71-75	4	6	2	0	0	0	0	1	8	4	2.5	0.9	11
76-80	6	10	8	0	0	0	0	4	6	4	3.8	1.2	17
81-85	13	7	4	0	0	0	0	1	1	4	3	1.3	13
86-90	3	9	5	0	0	0	0	4	2	6	2.9	1.0	13
91-95	3	5	1	1	0	0	0	3	1	3	1.7	0.5	7
96-100	1	2	2	0	0	0	0	1	1	1	0.8	0.2	4
101-105	0	1	0	0	0	0	0	0	0	1	0.2	0.1	1
106-110	1	3	2	0	0	0	0	2	0	0	0.8	0.4	4
111-115	1	0	2	1	0	0	0	0	0	1	0.5	0.2	2
116-120	0	0	0	0	0	0	0	1	0	0	0.1	0.1	0
121-125	0	0	0	0	0	0	0	0	0	0	0	0.0	0
126-130	0	1	0	0	0	0	0	0	0	0	0.1	0.1	0
131-135	0	1	0	0	0	0	0	0	0	0	0.1	0.1	0
136-140	0	0	0	0	0	0	0	0	0	0	0	0.0	0
141-145	0	0	0	0	0	0	0	0	0	0	0	0.0	0
146-150	0	0	0	0	0	0	0	0	0	0	0	0.0	0
151-155	0	0	0	0	0	0	0	0	0	0	0	0.0	0
oysters/m²	50	58	36	2	0	0	1	25	22	34	22.8	6.9	100
live vol (L)	10	12	9	1	0	0	0	5	3	8	4.8	1.5	
blank vol (L)	11	8	7	18	5	3	8	10	11	8	8.9	1.3	
ray boxes	6	4	2	3	0	0	0	0	0	5	2.0	0.7	