Final Report

To

Virginia Coastal Zone Management Program
National Oceanic and Atmospheric Administration

By

The Virginia Institute of Marine Science
College of William and Mary

December 15, 2017

FY2015 Task 10 Eelgrass and Bay Scallop Restoration in the Seaside Bays of Virginia.
(April 1, 2016 to September 30, 2017)

Robert Orth, Kenneth Moore, Richard Snyder, Mark Luckenbach, Albert Curry, Jr., Sean Fate, Frederick Holbert, Betty Neikirk, David Parrish, Paul Richardson, Erin Shields, and David Wilcox

Virginia Institute of Marine Science, School of Marine Science, 1375 Greate Road, P. O. Box 1346, College of William and Mary, Gloucester Point, Virginia 23062

This restoration project was funded, in part, by" the Virginia Coastal Zone Management Program at the Department of Environmental Quality through Grant # NA15NOS4190164 of the U.S. Department of Commerce, National Oceanic and Atmospheric Administration, under the Coastal Zone Management Act of 1972, as amended. Matching funds were provided by the Virginia Institute of Marine Science.

INTRODUCTION

The Virginia coastal bays suffered a catastrophic ecosystem state change in the last century primarily due to a wasting disease that devastated eelgrass beds followed by a significant hurricane in 1933 that likely eliminated the remaining populations (Orth et al. 2006, unpublished data). This state change from eelgrass to an "unvegetated" bottom, dominated by benthic algae, resulted in the loss of critical ecosystem services, including the provision of food and nursery habitat for numerous avian and marine species, including the bay scallop, *Argopecten irradians,* which supported a significant commercial fishery prior to these events and it never recovered following the eelgrass decline (Orth et al. 2006). While eelgrass eventually rebounded from the pandemic decline both in the Chesapeake Bay and in many coastal bays along the eastern seaboard of the United States (Cottam and Munro 1954), there are no records of eelgrass recovery in the VCR coastal bays until the mid-1990s (Orth et al. 2006).

In 1997, the discovery of two small patches of eelgrass in South Bay, one of the Virginia coastal bays, suggested that this bay could support the growth of eelgrass and that the limiting issue for expansion of eelgrass may be the lack of seed input. Based on this we subsequently began an attempt to restore eelgrass to the coastal bays with seeds. In 1999, we initiated large scale (>100 m^2 areas) seed introductions using millions of seeds starting in South Bay and in later years expanding to three additional bays where the relative isolation from the nearest seed-producing beds may have historically resulted in rare, low-density seedling recruitment. The success of this restoration effort has been documented in many final reports and published papers (see papers in Orth and McGlathery 2012) and represents one of the most successful eelgrass restoration efforts in the world today. This success led to the initiation of the program to re-introduce the bay scallop back to these coastal bays with initial attempts showing moderate successes documented in field surveys conducted in 2011 through 2015.

The goal of this project was to continue the enhancement of eelgrass and the bay scallop to these coastal bays. Specific objectives of the FY 2015 funds were: 1. Plant eelgrass using seeds to increase the recovery of the eelgrass beds into the Virginia coastal bays region; 2. Determine seedling establishment rates and evaluate the effectiveness of the seed planting; 3. Assess eelgrass bed growth and expansion; 4. Enumerate the finfish community that may be potential bay scallop predators; 5. Monitor water quality conditions to assess changes that may be associated with the eelgrass recovery and to identify new potential areas for restoration activities; and 6. Continue bay scallop restoration efforts initiated in 2009 with NOAA's American Reinvestment and Recovery Act Funds and supported by FY2009 through FY2014 Coastal Zone Management support.

STUDY SITES

Eelgrass and bay scallop restoration studies were conducted in the four adjacent sub-basins along the lower Delmarva Peninsula in 2012: South Bay, Cobb Bay, Spider Crab Bay and Hog Island Bay (Figure 1). The coastal bays are part of the Virginia Coast Reserve (VCR) Long-Term Ecological Research site. We initiated large scale eelgrass restoration with seeds in South Bay in 1999, Cobb Bay in 2001, Spider Crab Bay in 2003, and Hog Island Bay in 2006 following at least 1-yr survival of test plots in each bay. Spider Crab bay was again identified as the bay to receive seeds in 2016. Water quality was measured in all four bays using DATAFLOW while continuous sensors were located in both South and Spider Crab bays. Bay scallop restoration efforts were concentrated in South and Cobb Bays.

METHODS

Seed collection and distribution
Eelgrass flowering shoots with maturing seeds were harvested either by hand (primarily volunteers organized by The Nature Conservancy) or by mechanical harvester in May, 2016, and stored in aerated, flow-through tanks until seed release following procedures described by Marion & Orth (2010) either at the Gloucester Pt. or Oyster seed curing facilities. Seeds were separated from the senescing shoots and held in recirculating seawater tanks until distribution in October, just prior to the normal period of seed germination in this region (Moore et al. 1993). The proportion of viable seeds was determined just before distribution by individually assessing firmness and fall velocity of in subsamples as detailed in Marion and Orth (2010). Batches of seeds with targeted numbers of viable seeds for individual restoration plots were measured volumetrically, and all seed numbers reported here refer to viable seeds.

In the fall, 2016, eelgrass seeds were hand broadcast from a boat into pre-determined un-vegetated plots in Spider Crab Bay (Figure 2). Plot size was 0.4 ha (one acre) and seed density was 100,000 seeds per plot.

Germination rates of seeds collected in 2016 were estimated by planting replicate batches of 20 seeds at approximately 5-7 mm depth in sandy sediments (generally greater than 95% sand and < 1% organic matter) (Moore et al. 1993) in a re-circulating seawater system inside a greenhouse. Water temperatures were adjusted to follow ambient water temperatures in the field. Germination was considered successful with the emergence of the cotyledon and first leaf.

Field assessment of seedling establishment was made in April and May, 2017, six months after broadcast. Since seeds become rapidly incorporated into the sediment and do not move far from where they settle to the bottom (Orth et al. 1994), we were able to accurately assess establishment rates in seeded plots. Seeds typically germinate in early to late November in this region (Moore et al. 1993) and grow slowly during the winter months when water temperatures range from 0° to 5° C. Divers counted the number of seedlings in 0.5 m belt transects along the two diagonals of designated plots and adjusted to total number of seedlings per 0.4 ha. This number was then divided by the number of seeds broadcast into the plot to determine seedling success.

Eelgrass Assessment - Broad Scale
As in 2014, the atmospheric and water quality conditions over the coastal bays in both spring and fall, 2016 were some of the most difficult we encountered since we began this project. The favorable conditions required to fly an aerial survey mission over these bays were as follows: 1. specified tidal stage (+/- 60 minutes of low tide); 2. plant growth season (peak biomass); 3. sun angle (between 20-40^{o}); 4. atmospheric transparency (cloud cover less than 10%); 5. water turbidity (edge of grassbeds should be visible); and 6. wind (less than 10 kts) (Dobson et al. 1995).

Finfish Sampling

Since 2012 we have conducted monthly nekton (fish) surveys in South Bay using a 4.9-m otter trawl towed from a shallow draft vessel at 2300-rpm for 2-minutes, (n = 6). Using GPS start and stop points we have determined that the average tow length was approximately 150-m. These surveys were conducted monthly from May through October.

Once caught, fish size and abundance were recorded and then specimens were released. Unidentifiable specimens were photographed and released, or euthanized with an ice slurry (IACUC-2015-03-16-jprich) and transported to the laboratory for identification confirmation.

Data storage, manipulation and summary statistics were performed with Microsoft Excel. Statistical analyses and plots were performed in R (R Core Development Team 2017).

Water Quality

Two complementary approaches to documenting water quality conditions were continued during the FY 2015 reporting period (October 1, 2016- Sept 2017). Broad spatial patterns in water quality were documented using continuous underway sampling (DATAFLOW) in 2016 and 2017 as in previous years (this effort commenced in 2003 and has been conducted annually, Orth et al. 2013) (Figure 1). In addition, temporal patterns in water quality were documented through sensor deployments at two fixed stations, South Bay and Spider Crab Bay. The DATAFLOW cruise track traversed restoration areas in all four bays: South Bay, Cobb Bay, Spider Crab Bay, and Hog Island Bay. Cruise tracks were expanded from the initial track in 2003 over South Bay as successive bays were added to the restoration effort. By 2005 the cruise track covered all four major bays and remained similar since. Cruises were generally conducted monthly throughout the eelgrass growing season, from March through November of each year. While the length of cruise tracks in vegetated and unvegetated areas varied annually as the eelgrass beds developed and expanded, the track has encompassed all four bays as it did previously. The DATAFLOW underway sampler recorded 'in vivo' measurements of surface water quality taken at 2-3 second intervals (0.25 m depth below surface; approximately every 50 m) along each cruise track. Measurements included turbidity, chlorophyll fluorescence, temperature, salinity, pH, dissolved oxygen, GPS location and depth using a YSI 6600 V2 sensor array (YSI Inc., Yellow Springs, Ohio that has been synchronized with various models of Garmin GPSMAP Sounders including the 168, the 498 and the 540S (Garmin Ltd., Olathe, KS)). All sensors on the YSI 6600 V2 were both pre-cruise calibrated and post-cruise checked according to YSI standard procedures. In addition to the continuous underway sensor measurements, 5 calibration and verification stations were sampled at discrete locations spaced along each cruise track for total suspended solids, extracted pigment chlorophyll, and light attenuation profiles. Total suspended solids (TSS) were determined by filtration of known volume of seawater (precombusted Gelman, Type A/E), rinsing with freshwater, and drying at 60ºC. Chlorophyll a was collected on Whatman GF/F glass fiber filters, extracted in a solvent mixture of acetone, dimethyl sulfoxide, and 1% diethylamine (45:45:10 by volume) and determined fluorometrically (Shoaf and Lium 1976). Chlorophyll concentrations were uncorrected for phaeopigments. Chlorophyll fluorescence measurements were converted to extracted chlorophyll equivalents reported in this paper by developing a regression between extracted and fluoresced chlorophyll using the extracted chlorophyll and fluoresced samples taken simultaneously at each verification station for the entire study period.

Diffuse downwelling attenuation of photosynthetically available radiation (PAR) was determined by triplicate water column measurements of downwelling photosynthetic photon flux density measured with a LI-COR, LIO-192, underwater cosine corrected sensor (LI-COR Biosciences, Lincoln, Nebraska). Measurements were taken every 25 cm from 10 cm below the surface to a depth of 1.0-m. Similar to the YSI chlorophyll measurements, YSI turbidity measurements were converted to light attenuation equivalents using regression analysis relating turbidity to downwelling light attenuation coefficients (Kd) using all simultaneously measured light profiles and turbidities taken at the verification stations over the course of the study. In order to capture high frequency temporally intensive water quality information, a YSI 6600 EDS was deployed at a fixed monitoring station beginning in South Bay in 2003, and a second station added in July, 2011, in Spider Crab Bay. In 2015 equipment at both stations was updated with YSI EXO2 datasondes and Storm3 telemetry units. Both stations have been monitoring year-round since 2011. Both are equipped with telemetry and real-time data are available through the VECOS web site (www.VECOS.ORG). Dataflow cruises were described in this report were successfully completed at monthly intervals from October 2015 through September 2016.

Scallop Seed Production

Broodstock for hatchery production of seed scallops were maintained in the field nursery and grow out cages kept in the South Bay grass beds. These cages allowed for not only growing scallops for release and maintaining broodstock, but also promote some in situ spawning of scallops in the cages. All of these scallops originated from parental stock of *Argopecten irradians concentricus* collected from Bogue Sound and Core Sound, North Carolina During 2009, 2010, and 2012, but are now fully integrated to serve as a Virginia Broodstock line.

Gametogenesis initiates in the adult scallops held in South Bay with increasing spring water temperatures, and was monitored by the field crew on maintenance visits. Several weeks prior to spawning, broodstock scallops were brought to the Castagna Shellfish Research Hatchery at the VIMS Eastern Shore Laboratory in Wachapreague. Broodstock were held there in tanks of filtered seawater and fed mixed cultured micro algae until gametes were ripe. Adults were then induced to spawn by a thermal shift. Fertilized eggs were collected by sieve from the spawning table and transferred to a conical tank. Hatched larvae were fed cultured micro algae through the free-swimming phase, approximately 9 days.

Larvae competent for settlement were collected by sieve and transferred to flowing seawater nursery tables fed a diet of natural phytoplankton from Wachapreague Channel. Juveniles were grown in this system for 4-6 weeks until >2 mm in size, permitting them to be transferred to 2 mm mesh bags for deployment in field grow out cages maintained in South Bay. Monthly maintenance tracks growth, cleans fouling from mesh and cages, and divides the scallops into larger mesh sized bags as they grew to avoid overcrowding. Scallops were regularly measured for growth and mortality assessed during these population splits and maintenance visits.

Maintenance of Scallop Spawning Stocks in Grass beds

Our scallop restoration strategy is predicated on maintaining spawning stocks from hatchery-produced cohorts in cages within the target seagrass beds. The choice to use caged broodstock is based on the need to maximize survival, especially during summer months when predation rates are high, and on fertilization efficiency by providing for critical densities for *in situ* spawning success. The cages were constructed of vinyl coated wire screening with 1 inch square mesh openings. The design is widely used for bottom culture of caged oysters. Two hundred adult scallops were place in plastic mesh bags (¼ inch to ½ inch mesh) and two bags were placed in each cage. The cages and bags required periodic cleaning by power washing at VIMS ESL, and fouled cages were exchanged with cleaned cages for this purpose.

Assessment of Wild Adult Scallop Populations
As in previous years we utilized diver surveys, targeting adult scallops that reside on the benthos within the eelgrass beds. These surveys were conducted by randomly selecting 320 point locations across the following three coastal bay regions: Cobb Bay, South Bay, and Southern South Bay (Figure 3.). Each of these regions was divided into 4 sub-regions containing the sample locations (Figure 3). At each of these sample locations one of five divers swam one of 5 transects arranged in a stellate pattern about the anchored research skiff. During each swim they randomly placed a 1 m^2 quadrat in 10 locations and they thoroughly search the area for adult scallops by touch as visibility was often poor. Overall, 16000-m^2 quadrats (about 4 acres) of bottom were sampled within the three bays. The number of scallops collected per m^2 was multiplied by the area of the grass bed to obtain an estimate of total scallop numbers for the grassbed. For comparison across the grassbeds, scallops per meter squared were also presented.

RESULTS

Eelgrass Seeding
In 2016, seeds were broadcast into 27 one acre (0.4-ha) plots in Spider Crab Bay (Figure 2) at a density of 100,000 seeds per acre for a total of 2.7 million seeds (Table 1b). A remaining balance of 25,000 seeds were broadcast into a small 0.1-Ha plot at the southern extent of the 2016 plots (Table 1b and Figure 2). Through 2016, over 71 million seeds have been broadcast into 213 ha (526 acres) (Tables 1a, b, Figures 4, 5).

Eelgrass Seedling Establishment
Seeding was successful but seedling establishment rates varied among individual plots, bays, and years. The mean seedling establishment rate for all evaluated plots seeded in 2016 was 9.6% (range of 3.2 – 14.3%). The laboratory mean germination rate of seeds assessed as viable was 88% with a range of 81 – 93% confirming that the seeds that we dispersed were largely viable seeds.

Meadow Expansion and Development
In 2016 areal measurements were unavailable for the seaside grassbeds for the reasons described in the methods section of this report.

Finfish
Over the years of our predator sampling total abundance of fishes was highest in the summer months, most notably in July (Figure 6). Pinfish *Logodon rhomboids*, a historically more tropical species, has been very abundant in some of our samples, most notably in July of 2013 and June of 2015 (Figure 7). The highest abundance recorded since the inception of this trawl survey was in June, 2015 with a mean of over 150 individuals per trawl (Figure 7). Furthermore, there appears to be an asynchronous relationship between pinfish abundances and abundances of the

other dominant species of our samples, silver perch (*Bairdiella chrysoura*) (Figure 7). Regarding pinfish size distribution, we also see a shift in overall length of pinfish from a mean of 5-cm in June to 9-cm in July to 12-cm in August (Figure 8).

Cumulative fish species (β) richness consists of 44 taxa for the sampling years of 2012 – 2016. A full list of these species with the cumulative numbers sampled can be seen in Table 2.

Water Quality

Dataflow cruises were successfully completed at monthly intervals from March 2016 through November 2016. Figure 9 presents box plots (median, 25th and 75th percentile and the minimum and maximum of the lower 99% of the data) for each of the restoration areas during the March-November 2016 SAV growing season. Temperature medians were nearly identical at 20^{o}C among all the sites (Figure 9). Median salinities again were very similar (~31) at all sites, however, salinities occasionally reached lower levels at South and Spider Crab bays (Figure 9) as in previous years. Similarly dissolved oxygen (Figure 9) and pH levels (Figure 9 were very similar across the bays in 2016 although the slightly lower pH observed at Cobb, South and Spider Crab sites were not observed at Hog Island Bay. Turbidity medians ranged between 8 and 10 NTU at all the bays (Figure 9). Although turbidity levels were slightly lower at Hog Island, the upper 99% of concentrations reached nearly 9 NTU higher than the other sites suggesting that higher short term pulses of high turbidity were observed there. This is consistent with previous years' measurements. Chlorophyll concentrations were lowest at South and Cobb Bays, increasing Spider Crab and highest at Hog (Figure 9). These differences among sites have been consistent over recent years.

Figure 10, presents a time series of the yearly March through November integrated, median 25% and 75% quadrille, maximum and minimum of the chlorophyll levels recorded by the DATAFLOW cruises across each of the four restoration areas for the entire 2003-2016 restoration project period. Overall, the pattern of lower median levels of chl-a found in 2016 in Cobb and South Bays were consistent earlier years with typical annual concentrations of 5.0 and 5.2 µg /l. Inter-annual variability was distinctly evident with markedly higher concentrations in 2005 and 2006 as well as 2012 at all the sites. Cobb, Spider Crab and Hog Island had low median yearly turbidities compared to previous years while turbidities in South Bay were more similar compared to previous annual medians.

Figure 11, presents a time series of the yearly March through November integrated, median 25% and 75% quadrille, maximum and minimum of the turbidity levels recorded by the DATAFLOW cruises across each of the four restoration areas for the entire 2003-2016 restoration project period. Turbidities measurements are reflective of suspended particle concentrations. These particles are usually comprised of inorganic and non-living organic particles as well as phytoplankton. All four restoration areas again showed similar patterns in median annual turbidity throughout the project period. Median annual turbidities in Cobb, Spider crab and Hog Island Bays were lower in 2016 that 2015 while those in South Bay were higher. Elevated turbidities were especially evident in 2006 and 2012 when median levels exceeded 11 NTU. 2005 had the highest median turbidity levels ranging from 14 to 17 NTU. This was approximately double the median levels observed between 2003 and 2014, which ranged from 8 to 9.5 NTU. For this coastal bays region we have calculated that the seasonal light requirements for seagrass growth to 1 m MSL in this region are approximately 10 NTU or lower. Therefore over time all the sites typically have suitable seasonal turbidities for plant growth to the seagrass restoration depths being used here (Figures 11). Years 2006, 2009 and 2012 have been above that threshold. Lower median turbidities in 2016 for Cobb, Spider Crab and Hog Island suggest that light availability for SAV growth was better while conditions in South Bay were slightly worse.

Scallop Seed Production, Grow out, and Brood stock

During the spring of 2016 approximately 152,000 juvenile bay scallops were deployed in cages in the South Bay grass bed. These individuals were split into additional bags in the fall of 2016 and will be retained until July 2017 with another split and cleaning in early spring 2017. Broodstock individuals from this age class have been selected and are being retained at lower stocking densities in cages for spring 2017 spawning. In July 2016 approximately 35,000 adult bay scallops were released in the South Bay grass bed.

The first October release was on 10/12/16, with 10,115 fall 2015 scallops. Scallops were released along a transect in South Bay starting N37 14.571 W75 51.263 and ending N37 14.433 W75 51.076). On the 13th of October, 12,500 of the spring 2015 spawn were released along a transect in South Bay starting N37 14.571 W75 51.263 and ending N37 14.433 W75 51.076. Total releases for October were 22,615 adult scallops. 5,000 of the fall 2016 spawn are being retained in cages. The larger growing juveniles from nursery were deployed in one section of a cage. An additional 4,850 more were added to the cage in two spat bags (3mm mesh), 1 bag with ~1400 and another with ~3600 individuals. 56,000 of the spring 2016 scallops are being retained. Scallops held in 7 cages were split to 11 cages. Mortality appeared somewhat high, most likely from crabs. One cage with 400 individuals was transferred to a separate cage moved to the East end of the South line to be held as new broodstock. More scallops from the spring 2016 spawn were be added to the broodstock cages in the spring of 2017.

Assessment of Wild Scallop Population

Diver surveys of 1354 acres within the South Bay grassbed during July 2015 yielded an estimated population of 104,000 scallops. This is down from the 113,000 estimated population size in 2014 (Figure 12 a) but the difference is not statistically significant so there was essentially no change. From 2013 to 2014, however, there was a highly significant increase in scallop abundance from 28,000 in 2013 ($p<0.001$).

In 2015 the sampling effort expanded to include another South Bay grassbed, south of the main bed, designated Southern South Bay, and another north of Sand Shoal Inlet called Cobb Bay (Figure 3). In 2015 we found surprisingly high numbers of scallops with an estimated population abundance of 54,000 in Southern South Bay and 84,000 in Cobb Bay (Figure 12 a) Therefore, on a scallops/m^2 basis, we estimate that there were approximately 0.019 scallops/m^2 in South Bay, 0.022/m^2 in Southern South Bay and 0.027/m^2 in Cobb bay (Figure 12 b). Scallop numbers were lower in 2016 with an estimated abundance of 29,000 in Cobb Bay, 35,000 in South Bay and 15,000 in Southern South Bay (Figure 12a). Therefore we estimate there were approximately 0.009, 0.006 and 0.006 scallops/m^2 respectively (Figure 12b).

We expect that each of these estimates represent an underestimate of the actual population size with the grassbeds since catch efficiency, though unknown, is likely less than 100%.

DISCUSSION

Eelgrass Bed Development

The use of seeds in the recovery of eelgrass in the Virginia coastal bays continued successfully in 2016. The collection process of harvesting flowering shoots for seeds, followed by maintenance of the shoots in our seed curing tanks until seeds were released, removal of seeds from these tanks once seeds were fully released, and storage of seeds in our greenhouse under appropriate environmental conditions of temperature and salinity, yielded a large number of seeds that we were able to use in the restoration process. We were able to broadcast seeds into 27 1 acre plots in 2016. In the spring of 2017, for the subset of acre plots that we checked, we found that establishment rates were much higher (14.3%) than the 6.28% observed in the spring of 2016.

The growth of eelgrass in new areas of these coastal bays as well as the expansion of the existing beds supports the concept that seeds are important is this expansion process and dispersal of flowering fragments with seeds is the primary dispersal mechanism for colonizing areas that are distant from any established beds. We anticipate that seagrass meadows will continue to expand both from our additional restoration effort using seeds but also dispersed seeds from established beds. The expansion of seagrass here in the coast al bays stands in stark contrast to what is being recorded in Chesapeake Bay (Lefcheck, et al., 2017)

Finfish

The high abundance of fishes in the summer months (Figure 6) is most likely a function of seasonal temperature that determines the timing of their arrival to the South Bay SAV beds. These data continue to help us better understand the food web dynamics of the seaside SAV beds, and, more practically, they assist in more informed decisions regarding the timing of scallop releases. Using these data we continue to conduct "smart releases" of scallops, timed to avoid maximum predation by all predators and in particular, pinfish which have been seen in higher abundance in South Bay, most prominently in June of 2015 (Figure 7).

In addition to their high summertime abundance, the ontological shift in the diets of pinfish make them of special concern for juvenile scallops (Figure 8). Small pinfish (<60mm) prefer small crustaceans until they reach a size of 60mm. From 61 – 120 mm they feed on larger benthic invertebrates including bivalve mollusks. Pinfish > 120 mm generally become omnivorous (Livingston 2003).

This ontological shift can be seen in Figure 8 and with the color-shaded factor of month it might also be inferred that this is the same population of pinfish as its mean size grows. From these data, it is apparent that in general, the summer months are probably more dangerous for juvenile scallops due to high fish abundance and pinfish food preference. This is also in line with recent work by (Schmitt et al. 2016), where they found large numbers of pinfish in the predaceous size range, in the month of July, with the samples from the summers of 2013 and 2015 dominated by pinfish (Figure 7). We also find it interesting that we observed an apparent asynchronous relationship between pinfish and silver perch, hinting at the possibility of a competitive interaction between the two dominant fishes (Figure 7). With our improved understanding of the fish community, we hope to not only contribute to the ecological literature in the near future, but we also plan to use this knowledge to continue timing the release of juvenile scallops to avoid high predation intensity by pinfish, silver perch and other predators in the future.

Water Quality in the Virginia Coastal Bays

Water quality monitoring of the four restoration areas in 2016 indicates that, overall, mean water quality remained suitable for eelgrass growth and restoration in all of the coastal lagoon areas studied. Growing season, salinity pH, dissolved oxygen, and temperature were very comparable across all the sites and generally within the ranges necessary for growth and spreading of eelgrass. Lower relative chlorophyll and turbidity levels in Cobb, Spider Crab and Hog Island in 2016 compared to 2015 was positive for those regions while South Bay's slightly higher levels were concerning. The overall continued lower concentrations of chlorophyll and turbidity since 2012 are however positive for eelgrass populations in this region. Because of the positive feedbacks between restored seagrass bed size and abundance and water quality which we have observed here and in other coastal systems, this, now three year overall improvement, is encouraging. Storms or other factors including warming climate, disease or man induced perturbations from aquaculture fishery activities or dredging could change this trajectory and these factors should be carefully monitored and in the case of man-made stressors, minimized as much as possible.

Scallop Restoration

Previous reports (FY11 Task 12, FY12 Task 11, FY13, Task 11, FY14, Task 11) have detailed our restoration strategy for bay scallops and the early success that we have had in (a) developing and maintaining a Virginia brood stock line of bay scallops, (b) spawning, maintaining and out planting scallops in the grass bed, and (c) establishing a wild population in the grass bed. Anecdotal evidence continues to suggest these scallops are spreading beyond the release and in situ spawning sites. One change in our spawning strategy implemented this year was to use the wild scallops recovered during the annual census for broodstock, and thereby incorporation any selective pressures for survival in the system in our next cycle of production from the ESL hatchery.

The quantitative annual census suggests a loss in numbers of the wild population in 2016 (Figures 12a, 12b), which may in fact be due to intense predation by rays, which also created substantial numbers of feeding pits in the SAV beds dislodging *Zostera* plants. Despite this setback, we are confident that ultimately the population can reach sustainable levels. Expanding the seagrass habitat further will help us to create a sustainable population of scallops in the seaside bays of Virginia.

One of the bottlenecks to increasing the production of our scallop output are the high labor and space requirements for the cage rearing of scallops in in the field. In 2016, we established a partnership with The Nature Conservancy to use their tanks in Oyster Harbor, VA to augment scallop juvenile production for direct release into the SAV beds when predator abundance is low. Although initial production levels were not as expected, further development of this partnership will be critical to achieving a critical mass of scallops to release in the coastal bays.

Overall, we remain encouraged by these early successes, however, in our best informed judgment, the standing stock of wild bay scallops has not reached a point at which we expect it will be self-sustaining. With an abundance of 0.01 scallops/m^2 (Figure 12b), we must achieve an order of magnitude higher population to be self-sustaining. Thus, as we move forward we will continue to explore ways to improve our restoration strategy. For example, the State of Florida uses a 1 scallop per m^2 density as the threshold for opening their public recreational harvest.

Acknowledgments
We appreciate the numerous staff and students, who made the success of this project possible, notably Corey Holbert, Rebecca Turner, Darian Kelley, Edward Smith, Jim Goins, Steve Snyder, Paige Ross, Stephanie Boniwell, AJ Johnson, as well as the numerous citizens who volunteered to help collect eelgrass seeds, and summer interns at VIMS Wachapreague who helped in the bay scallop assessment. Additional funding for this project was provided by the grants from the Virginia Recreational Fishing License Fund, The Nature Conservancy, as well as private grants from Norfolk-Southern, and the Keith Campbell Foundation for the Environment.

LITERATURE CITED

de Bello F, Lavergne S, Meynard CN, Leps J, Thuiller W (2010) The partitioning of diversity: showing Theseus a way out of the labyrinth. J. of Veg Science 21:992-1000

Dobson JE, Bright EA, Ferguson RL, Field DW, Wood LL, Haddad KD, Iredale H, III, Jensen JR, Klemas VV, Orth RJ, Thomas JP (1995) NOAA Coastal change analysis program (C-CAP): Guidance for regional implementation. NOAA Tech. Rep. NMFS 123. 92.

Lefcheck JS, Wilcox DJ, Murphy RR, Marion SR, Orth RJ (2017). Multiple stressors threaten a critical foundation species in Chesapeake Bay. Global Biology Change doi: 10.11.11/gcb.13623

Livingston, R. J. (2003) Trophic Organization in Coastal Systems. CRC Press. U.S.A. 408 pages.
Marion SR, Orth RJ (2010) Innovative techniques for large scale collection, processing, storage, and dispersal of eelgrass (*Zostera marina*) seeds. Restor Ecol 18:514-526

McGlathery KJ, Reynolds L Cole LW, Orth RJ, Marion SR, Schwarzchild A (2012**)** Recovery trajectories in a temperate system restored by seeding. Mar Ecol Prog Ser 448:209-221

Moore KA, Orth RJ, Nowak JF (1993) Environmental regulation of seed germination in *Zostera marina* L. (eelgrass) in Chesapeake Bay: Effects of light, oxygen and sediment burial. Aquat Bot 45:79-91

Orth RJ, Luckenbach MW, Moore KA (1994) Seed dispersal in a marine macrophyte: Implications for colonization and restoration. Ecology 75:1927-1939

Orth RJ, Luckenbach ML, Marion SR, Moore KA, Wilcox DJ (2006) Seagrass recovery in the Delmarva Coastal Bays, USA. Aquat Bot 84:26-36

Orth, RJ, McGlathery, KJ. (2012). Eelgrass recovery in the coastal bays of the Virginia Coast Reserve, USA. Mar Ecol Prog Ser 448 173-176

Orth RJ, Moore KA, Marion SR, Wilcox DJ, Parrish DB (2012) Seed addition facilitates eelgrass recovery in a coastal bay system. Mar Ecol Prog Ser 448:177-195

Orth RJ, Wilcox DJ, Nagey LS, Owens AL, Whiting JR, Kenne AK (2010) Distribution of Submerged Aquatic Vegetation in the Chesapeake Bay and Coastal Bays - 2009. VIMS Special Scientific Report Number 149. Final report to U.S. EPA Chesapeake Bay Program, Annapolis, MD

R Development Core Team (2017) *R: A Launguage and Environment for Statistacl Computing, R Foundation for Statistical Computing.* Vienna, Austria. ISBN 3-900051-07-0, URL http://www.R-project.org/.

Sanders, HL (1968) Marine Benthic Diversity: A Comparative Study. The American Naturalist, 102:243-282.

Schmitt EL, Luckenbach MW, Lefcheck JS, Orth RJ (2016) Predator-prey Interactions in a restored eelgrass ecosystem: strategies for maximizing success of a reintroduced species, *Argopecten irradians*. Restor Ecol 24:558-565.

Table 1a. Summary of eelgrass seed distributions for South and Cobb Bays (number of viable seeds distributed, total area seeded, size and number of plots seeded).

	South Bay				Cobb Bay			
Year	Seeds x 10^6	Area (ha)	Plot size (ha)	n plots	Seeds x 10^6	Area (ha)	Plot size (ha)	n plots
1999	0.3	1.2	0.6	2				
2000	0.6	0.1	0.0	9				
2001	3.6	9.7	0.4	24	0.6	1.6	0.4	4
2002	1.8	9.7	0.4	24				
2003					1.1	4.9	0.2	24
2004	0.7	2	2.0	1				
2005	0.5	1.6	0.2	8				
2006								
2007								
2008								
2009					2.3	6.1	0.4	15
2010								
2011					1.1	2.4	0.4	6
2012								
2013								
2014	4.2	11.2	0.4	28				
2015								
2016								
Total	11.7	35.5		96	5.1	15		49

Table 1b. Summary of eelgrass seed distributions for Spider Crab and Hog Island Bays (number of viable seeds distributed, total area seeded, size and number of plots seeded).

	Spider Crab Bay				Hog Island Bay			
Year	Seeds x 10^6	Area (ha)	Plot size (ha)	n plots	Seeds x 10^6	Area (ha)	Plot size (ha)	n plots
1999								
2000								
2001								
2002								
2003	0.500	2.2	0.2	11				
2004	0.600	1.6	0.2	8				
2004	5.900	11.8	0.8 - 2	7				
2005	1.000	2.8	0.2	14				
2006	0.500	2.4	0.2	12	0.6	2.8	0.2	14
2006					1.2	5.7	0.4	14
2007	1.500	6.1	0.2	30	0.5	2.4	0.2	12
2007					0.9	4.9	0.4	12
2008	1.200	4.7	0.2	23	0.6	2.4	0.4	6
2009	6.000	16.2	0.4	40				
2010	5.500	22.3	0.4	55				
2011	2.000	10.9	0.4	27				
2012	7.300	14.2	0.4	35				
2013	6.000	12.1	0.4	30				
2014	3.500	9.2	0.4	23				
2015	6.300	16.8	0.4	42				
2016	0.025	0.1	0.1	1				
2016	2.720	11.0	0.4	28				
Total	50.5	144.4		386.0	3.8	18.2		58.0

Table 2. Species list of fauna collected during the South Bay trawl survey from 2012 – 2016.

Species	Common Name	
Bairdiella chrysoura	Silver Perch	5343
Lagodon rhomboides	Pinfish	2003
Menidia menidia	Atlantic Silverside	538
Sygnathus sp.	Pipefish	500
Callinectes sapidus	Blue crab	452
Leiostomus xanthurus	Spot	281
Orthopristis chrysoptera	Pigfish	263
Anchoa mitchelli	Anchovy	193
Archosargus probatocephalus	Sheepshead	130
Blue Crab	Callinectes sapidus	72
Syngnathus floridae	Dusky Pipefish	53
Centropristis striata	Black Seabass	51
Mycteroperca microlepis	Gag Grouper	36
Anchoa mitchilli	Bay Anchovy	35
Opsanus tau	Toadfish	25
Urophycis regia	Spotted codling	21
Drum unid	Drum unid	18
Chilomycterus schoepfii	Striped Burrfish	13
Eucinostomus gula	Silver Jenny	10
Paralichthys dentatus	Summer Flounder	10
Tautoga onitis	Tautog	9
Cynoscion nebulosus	Spotted seatrout	8
Prionotus	Sea Robin	7
Sphyraena borealis	Northern Sennet	6
Pomatomus saltatrix	Bluefish	6
Sphoeroides maculatus	Northern Puffer	5
Conger oceanicus	Conger eel	4
Eucinostomus argenteus	Silver Mojarra	4
Fistularia tabacaria	Blue Spotted Cornetfish	3
Sciaenops ocellata	Red drum	2
Southern Sting Ray	Dasyatis americana	2
Diplodus holbrookii	Spottail pinfish	2
Synodus foetens	Inshore Lizardfish	2
Chaetodon ocellatus	Spotfin Butterfly Fish	2
Chasmoides bosquianus	Striped blenny	1
Stephanolepis hispida	Planehead filefish	1
Myctoperca	Grouper	1
Carax sp	Carax sp.	1
Menticirrhus saxatilis	Northern Kingfish	1

Gobiosoma bosc	Naked Goby	1
Dasyatis sabina	Atlantic stingray	1
Epinephelus niveatus	Snowy grouper	1
Panopeus sp	Mudcrab	1
Eucinostomus sp.	Eucinostomus sp.	1

FIGURES

Figure 1. Study region in the lower Virginia coastal bays. Hatched polygons represent eelgrass seed distribution regions. The solid line across all four bays represents the boat track for continuous underway water quality sampling (DATAFLOW) cruises. The open circles in South Bay and Spider Crab are the sites of the continuous monitoring stations.

Figure 2. Map showing the 27 0.4-ha and 1 0.1-ha plots of Spider Crab Bay each of which received 100,000 and 25,000 eelgrass seeds respectively in 2016.

Figure 3. Maps showing the sample scheme for the 2016 adult scallop survey for each of the seagrass regions of (a) Cobb Bay, (b) South Bay and (c) Southern South Bay. The purple lines denote the sampling sub-regions. Yellow dots indicate the randomly chosen survey locations.

Figure 4. Cumulative area of seeding (blue) and total area estimate from the aerial mapping (red) for all four seaside bays through 2015. Aerial survey data for all 4 bays were unavailable for 2005, 2013, 2014 and 2016.

Figure 5. Area of seeding in each of four bays (left axis), and area mapped in two density classes by aerial photography each year (right axis). (Note – in Spider Crab Bay and aerial measurements were available for South Bay only in 2013. No aerial data were available in 2014 or 2016).

Figure 6. Mean fish abundance/150-m trawl for the months of May (05) through October (10) for the years of 2012 through 2016. Bars indicate the standard error about the mean.

Figure 7. Time series of the mean abundance of pinfish (red) and silver perch (green) per 150-m trawl over the course of the survey period from 2012 through September 2016. The shaded areas around lines indicate the 95% confidence intervals and the dots indicate individual observations per trawl.

Figure 8. Histogram showing the number of pinfish as a function of fish length, with colors representing month with red = June, green = July, blue = August and purple = September (Note – only the first 10 individuals per trawl were measured). Data are from the period 2012 – 2016.

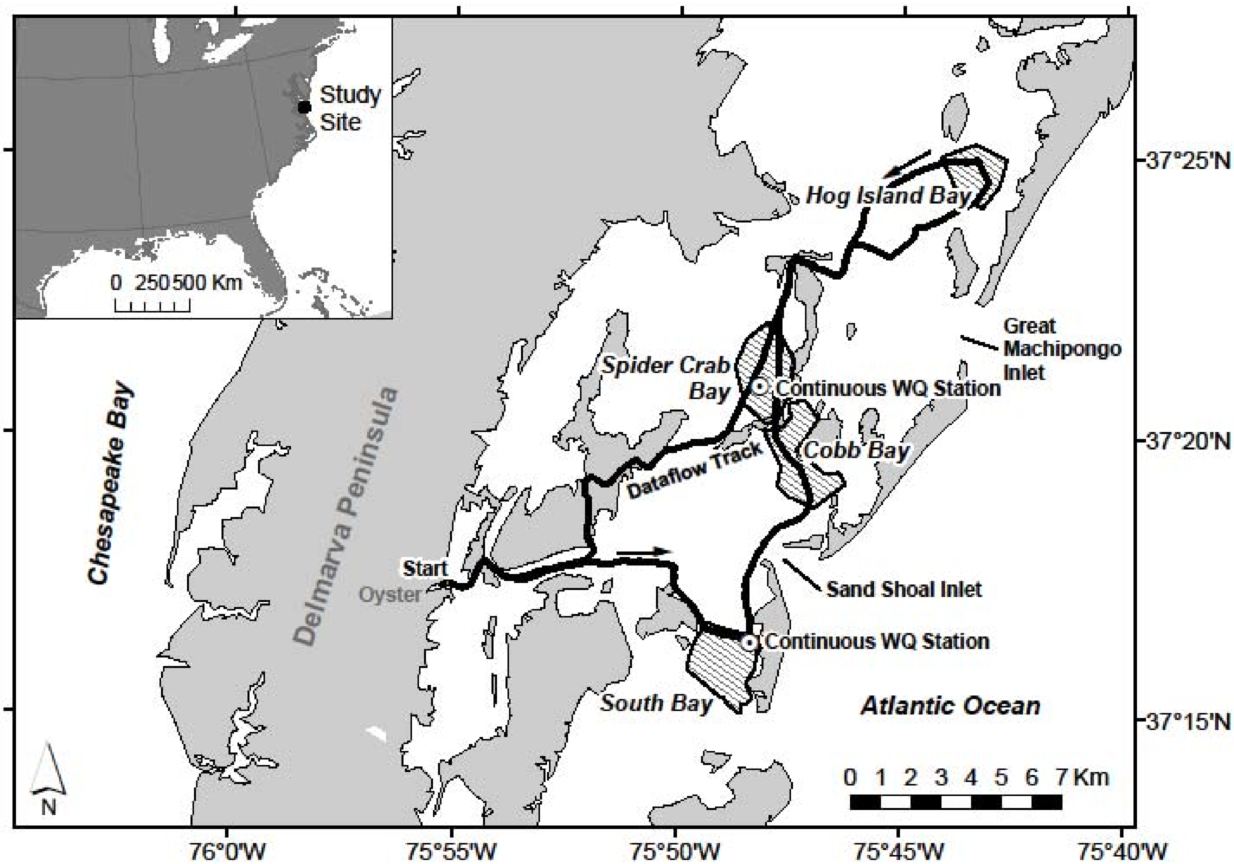
Figure 9. Box plots showing DATAFLOW (a) turbidity concentrations (median, 25th and 75th percentiles, and the minimum and maximum of the lower 99% of the data) from four restoration bay areas for the March-November periods from 2003-2015, and the same for (b) salinity (psu), (c) dissolved oxygen (mg/L), (d) pH, (e) Chlorophyll-a, and (f) Temperature (^{o}C).

Figure 10 a – d. Box plots showing DATAFLOW chlorophyll concentrations (median, 25th and 75th percentiles, and the minimum and maximum of the lower 99% of the data) from four restoration bay areas for the March –November periods from 2003-2015.

Figure 11 a – d. Box plots showing DATAFLOW turbidity concentrations (median, 25th and 75th percentiles, and the minimum and maximum of the lower 99% of the data) from four restoration bay areas for the March –November periods from 2003-2015.

Figure 12. Bar plots showing (a) the population estimate of adult scallops as estimated by the total seagrass bed area for South Bay (green) for the years of 2012 – 2015 and for Cobb Bay (orange) and Southern South Bay (blue) in 2015 – 2016 and (b) the estimated number of scallops per meter squared for the same years and bays as in a.

Figure 1


Study Site
0 250 500 Km
Chesapeake Bay
Delmarva Peninsula
Hog Island Bay
Spider Crab Bay
Continuous WQ Station
Great Machipongo Inlet
Cobb Bay
Dataflow Track
Start
Oyster
Sand Shoal Inlet
Continuous WQ Station
South Bay
Atlantic Ocean
N
0 1 2 3 4 5 6 7 Km
37°25'N
37°20'N
37°15'N
76°0'W
75°55'W
75°50'W
75°45'W
75°40'W

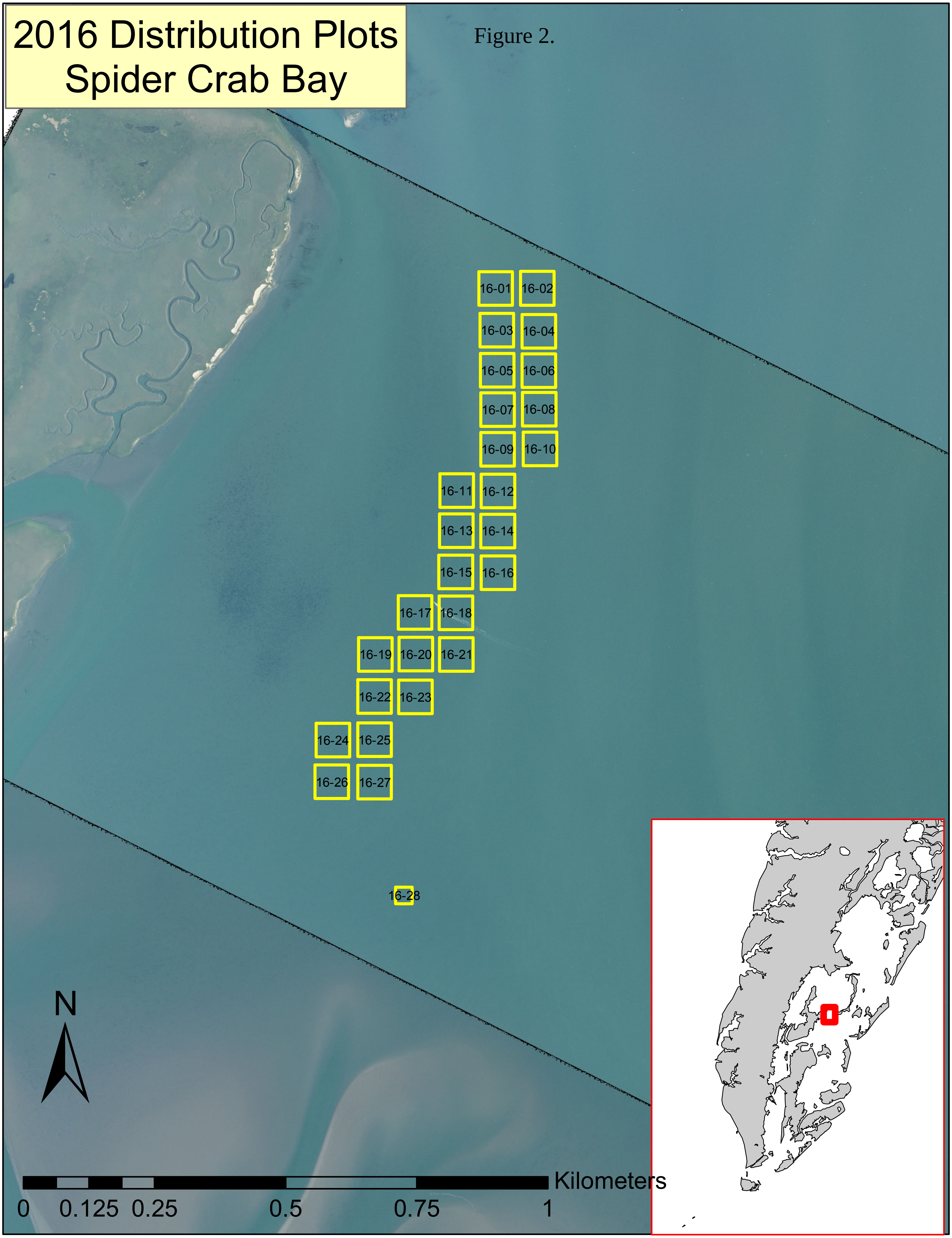

2016 Distribution Plots
Spider Crab Bay
Figure 2.
16-01
16-02
16-03
16-04
16-05
16-06
16-07
16-08
16-09
16-10
16-11
16-12
16-13
16-14
16-15
16-16
16-17
16-18
16-19
16-20
16-21
16-22
16-23
16-24
16-25
16-26
16-27
16-28
N
Kilometers
0
0.125
0.25
0.5
0.75
1

Figure 3a.

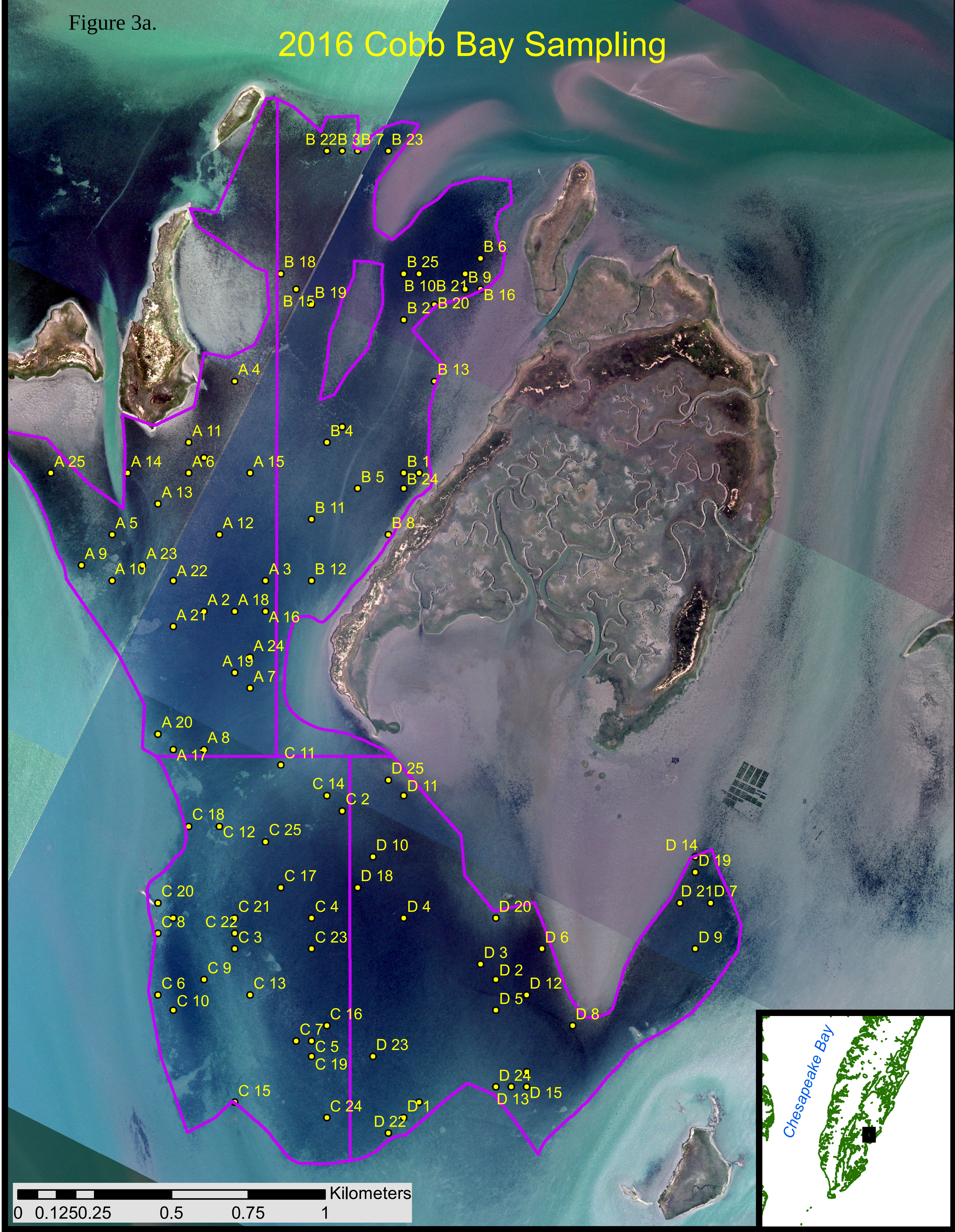
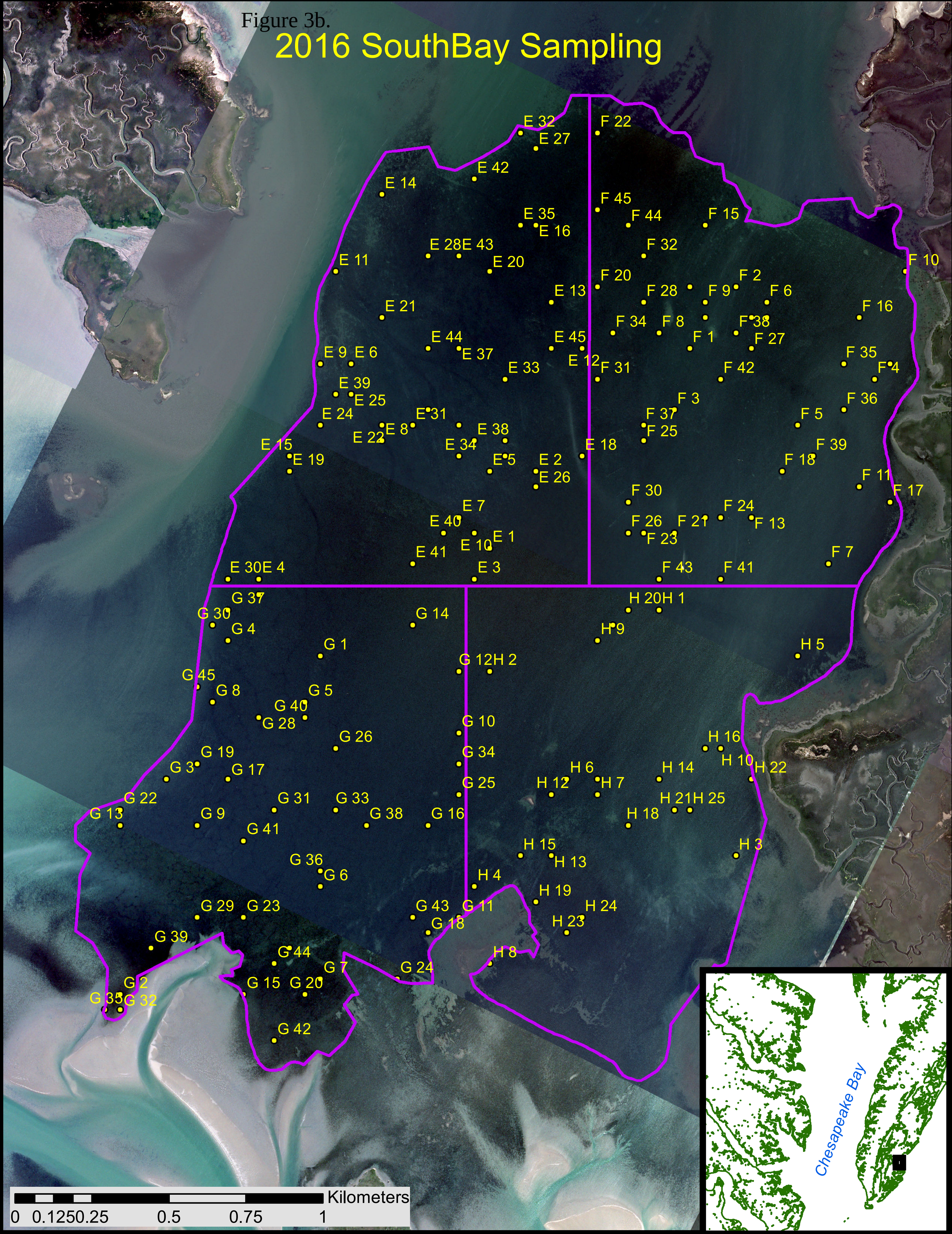
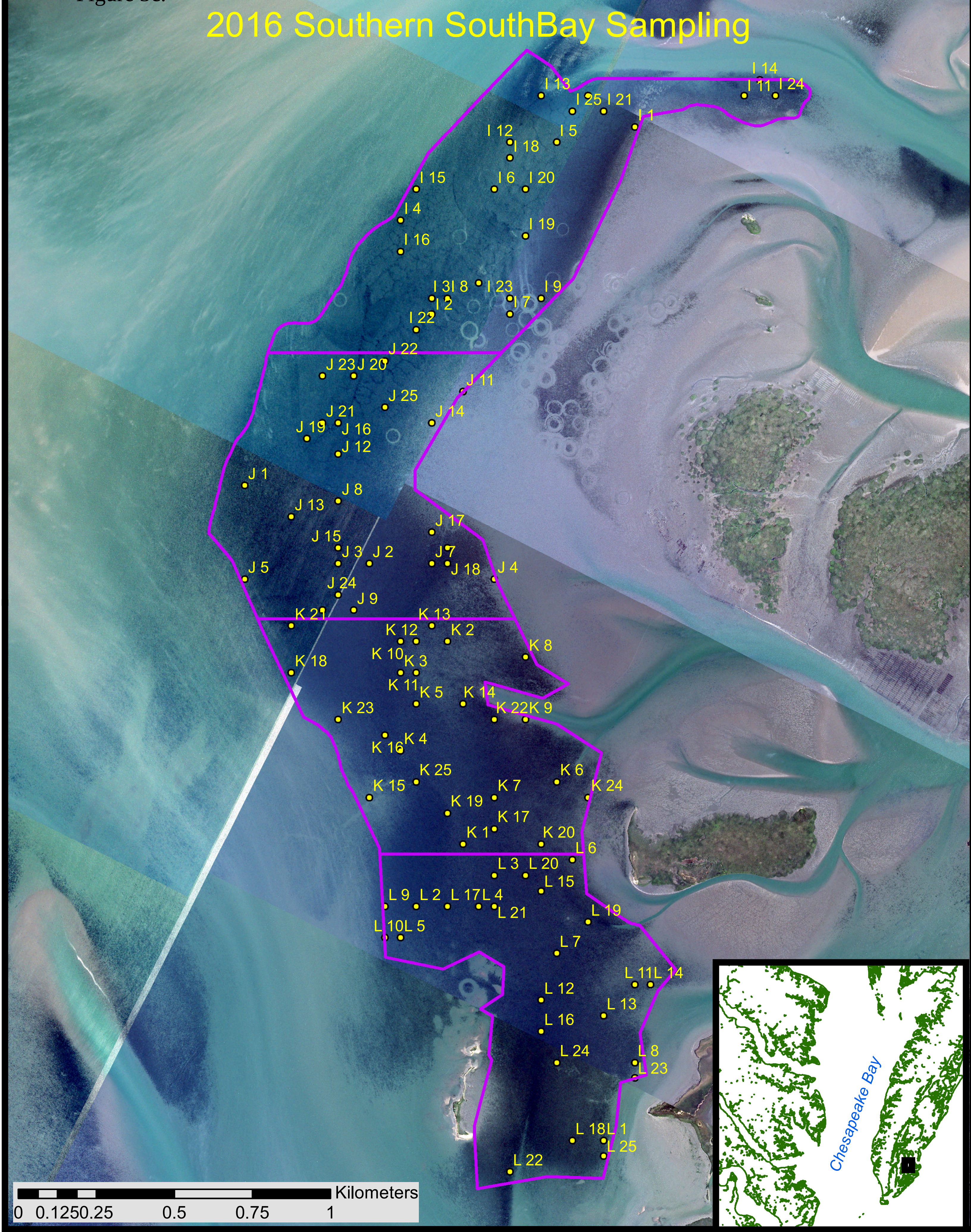

2016 Cobb Bay Sampling
B 22
B 3
B 7
B 23
B 6
B 25
B 9
B 10
B 21
B 16
B 18
B 19
B 15
B 2
B 20
B 13
A 4
B 4
A 11
A 25
A 14
A 6
A 15
B 1
B 24
B 5
A 13
B 11
A 5
A 12
B 8
A 9
A 23
A 10
A 22
A 3
B 12
A 2
A 18
A 21
A 16
A 24
A 19
A 7
A 20
A 8
A 17
C 11
D 25
C 14
D 11
C 2
C 18
C 12
C 25
D 10
D 14
D 19
C 17
D 18
C 20
C 21
C 4
D 4
D 20
D 21
D 7
C 8
C 22
C 3
C 23
D 6
D 9
D 3
C 9
D 2
C 6
C 13
D 12
C 10
D 5
C 16
D 8
C 7
C 5
D 23
C 19
D 24
C 15
D 13
D 15
C 24
D 1
D 22
Chesapeake Bay
Kilometers
0
0.125
0.25
0.5
0.75
1

Figure 3b.



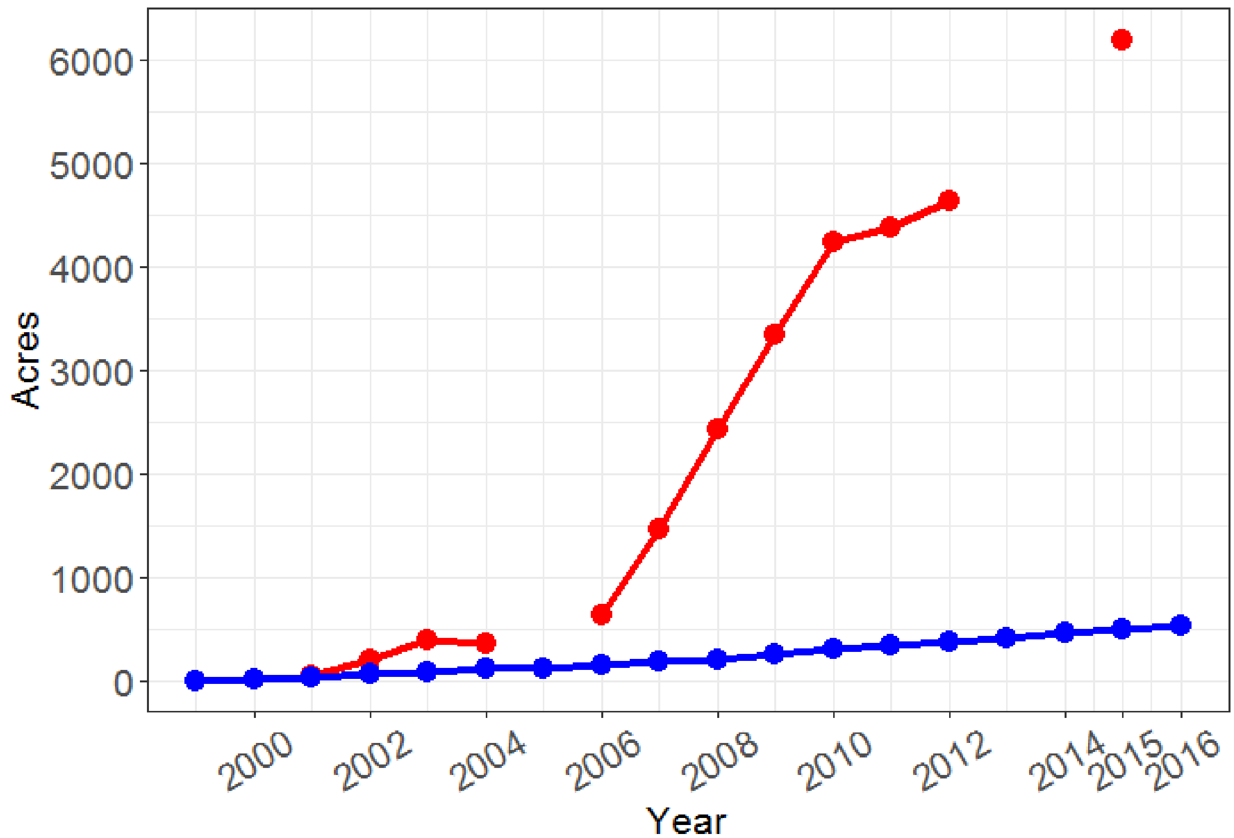
2016 SouthBay Sampling
E 32
E 27
E 42
E 14
E 35
E 16
E 28
E 43
E 20
E 11
E 13
E 21
E 44
E 37
E 45
E 12
E 9
E 6
E 33
E 39
E 25
E 24
E 31
E 22
E 8
E 38
E 34
E 15
E 19
E 5
E 2
E 26
E 18
E 7
E 40
E 1
E 10
E 41
E 3
E 30
E 4
F 22
F 45
F 44
F 15
F 32
F 10
F 20
F 2
F 28
F 9
F 6
F 16
F 34
F 8
F 38
F 1
F 27
F 35
F 31
F 42
F 4
F 3
F 36
F 37
F 25
F 5
F 39
F 18
F 11
F 30
F 17
F 24
F 26
F 21
F 13
F 23
F 7
F 43
F 41
G 37
G 30
G 4
G 14
G 1
G 12
G 45
G 8
G 5
G 40
G 28
G 10
G 26
G 19
G 34
G 3
G 17
G 25
G 22
G 13
G 31
G 33
G 9
G 38
G 16
G 41
G 36
G 6
G 29
G 23
G 43
G 11
G 18
G 39
G 44
G 7
G 24
G 2
G 15
G 20
G 35
G 32
G 42
H 20
H 1
H 9
H 5
H 2
H 16
H 10
H 22
H 6
H 14
H 12
H 7
H 21
H 25
H 18
H 15
H 13
H 3
H 4
H 19
H 24
H 23
H 8
Chesapeake Bay
Kilometers
0 0.1250.25 0.5 0.75 1

Figure 3c.



2016 Southern SouthBay Sampling
Chesapeake Bay
Kilometers
0 0.1250.25 0.5 0.75 1

Figure 4.



6000
5000
4000
3000
2000
1000
0
Acres
2000
2002
2004
2006
2008
2010
2012
2014
2015
2016
Year

Figure 5.

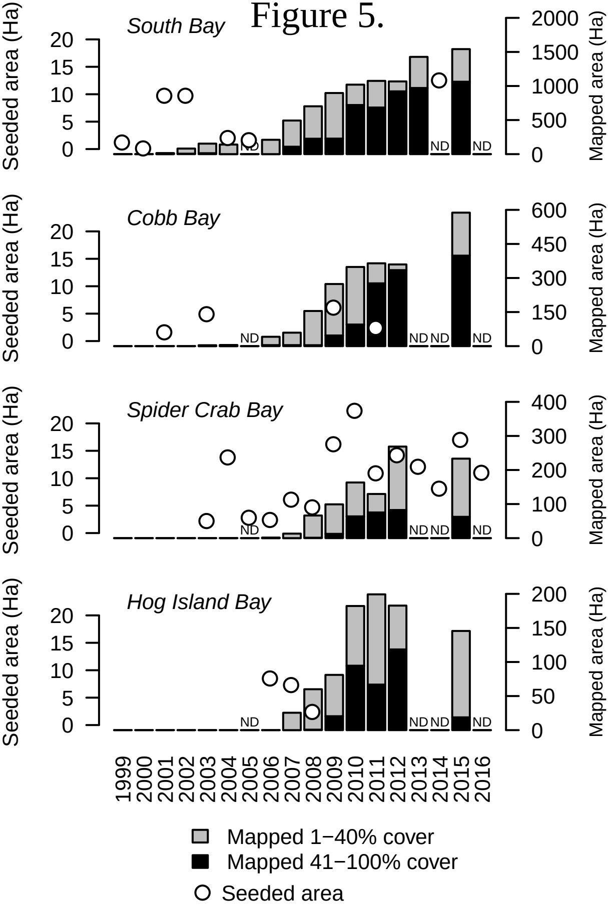

South Bay
Cobb Bay
Spider Crab Bay
Hog Island Bay
Seeded area (Ha)
Mapped area (Ha)
ND
1999
2000
2001
2002
2003
2004
2005
2006
2007
2008
2009
2010
2011
2012
2013
2014
2015
2016
Mapped 1−40% cover
Mapped 41−100% cover
Seeded area

Figure 6.

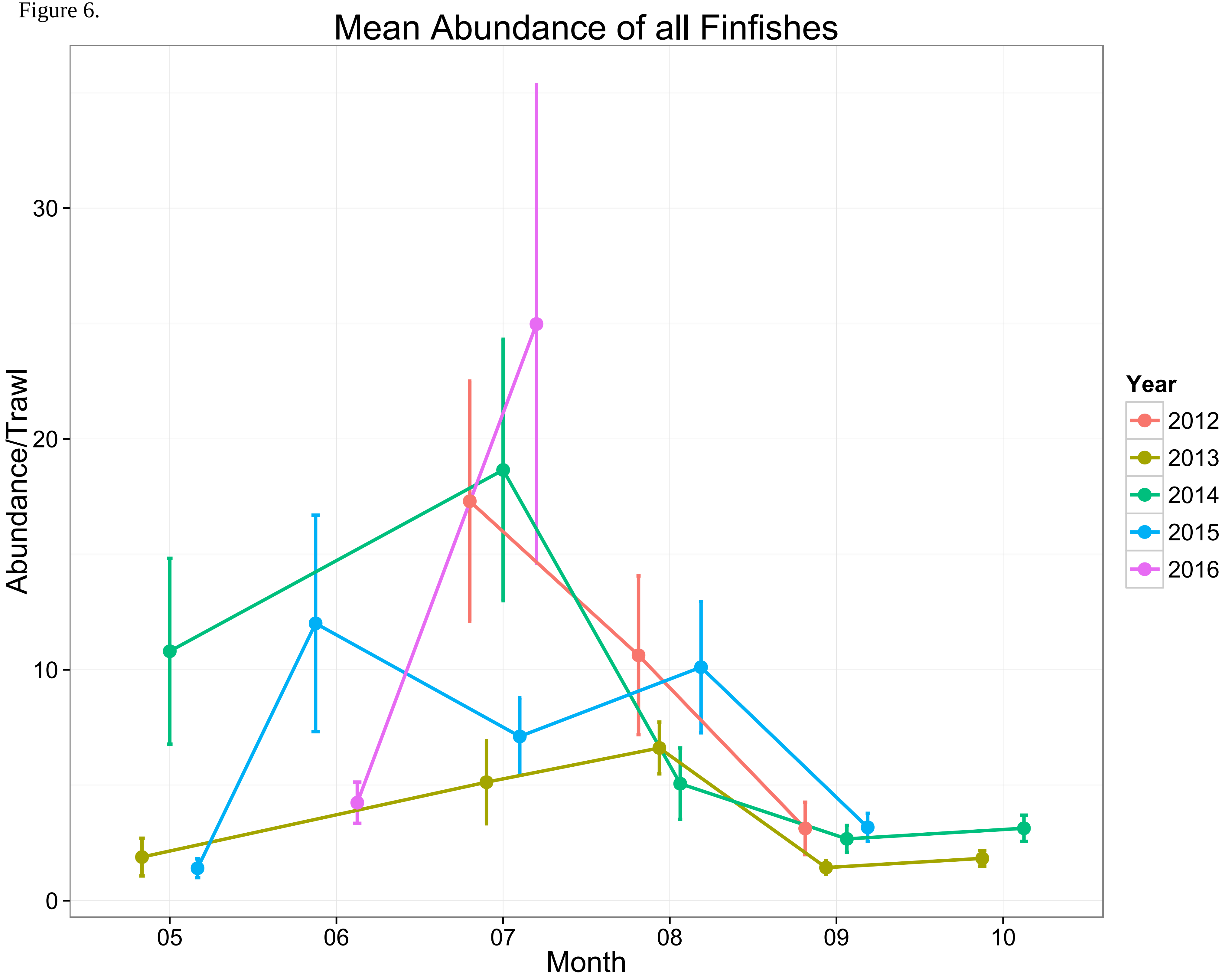
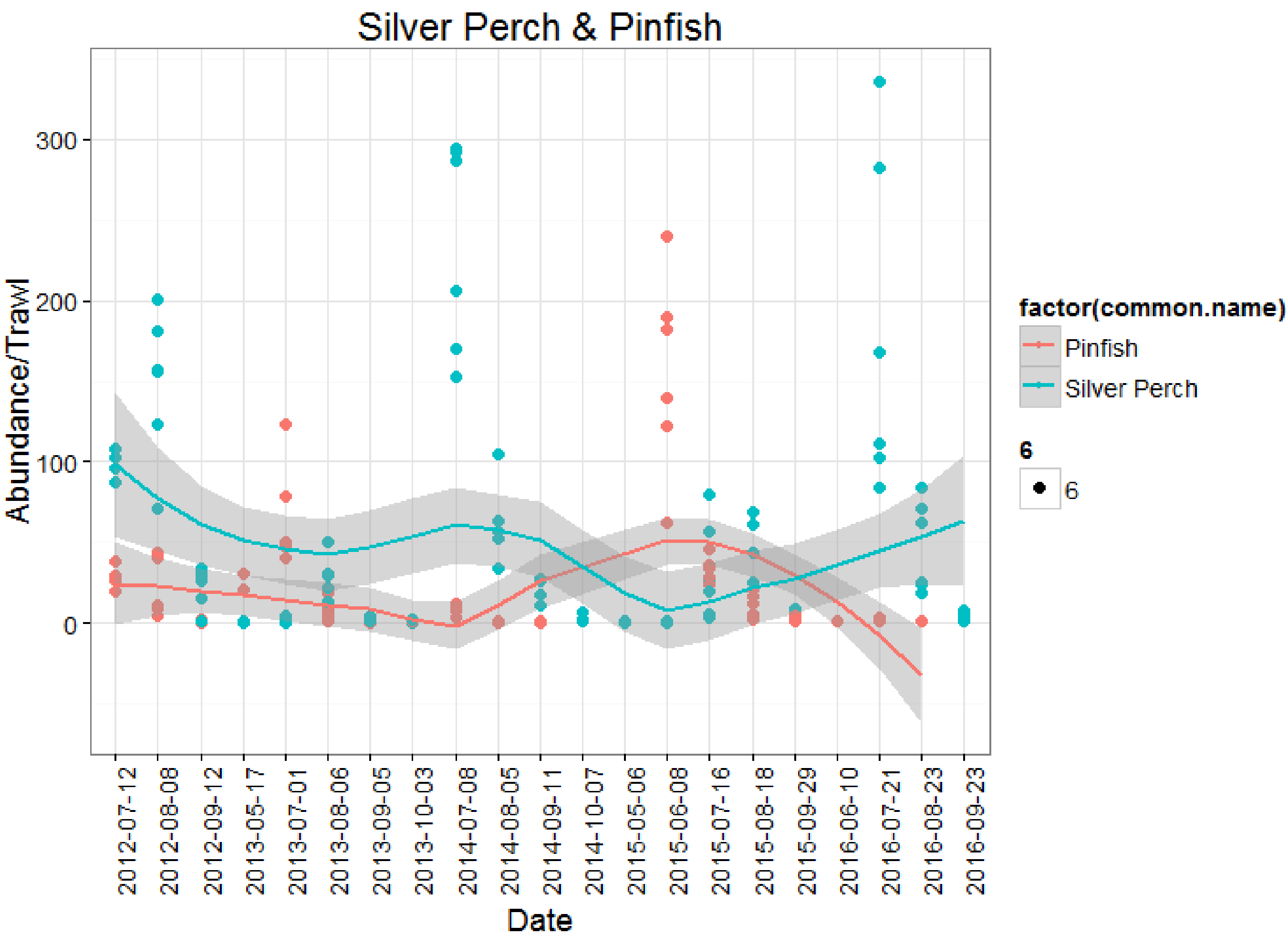

Mean Abundance of all Finfishes
Abundance/Trawl
0
10
20
30
05
06
07
08
09
10
Month
Year
2012
2013
2014
2015
2016

Figure 7.



Silver Perch & Pinfish
Abundance/Trawl
300
200
100
0
2012-07-12
2012-08-08
2012-09-12
2013-05-17
2013-07-01
2013-08-06
2013-09-05
2013-10-03
2014-07-08
2014-08-05
2014-09-11
2014-10-07
2015-05-06
2015-06-08
2015-07-16
2015-08-18
2015-09-29
2016-06-10
2016-07-21
2016-08-23
2016-09-23
Date
factor(common.name)
Pinfish
Silver Perch
6
6

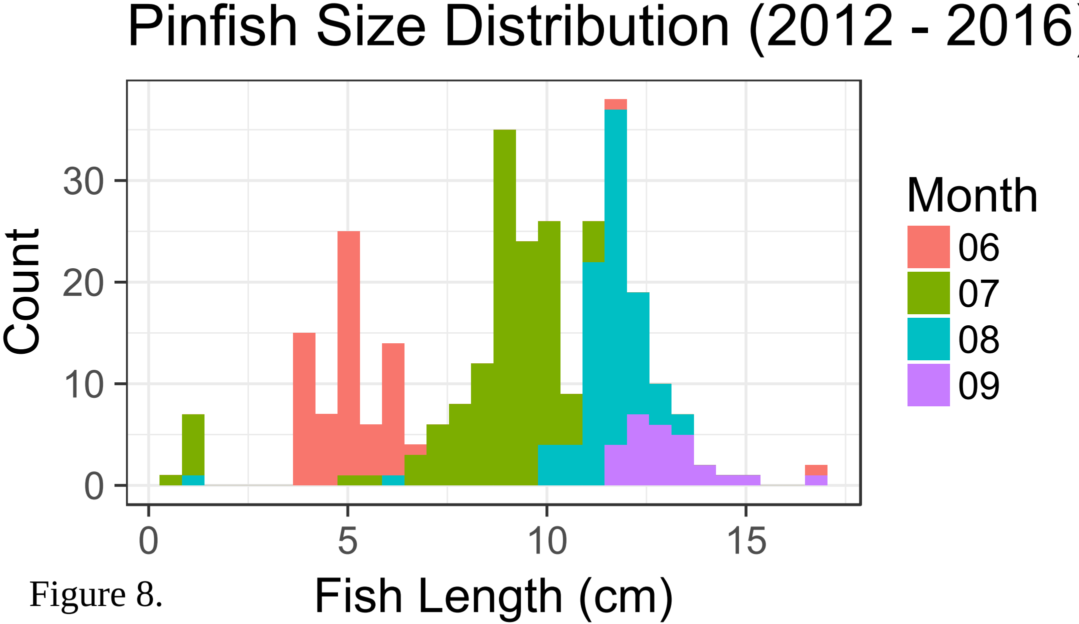

Pinfish Size Distribution (2012 - 2016)
Count
0
10
20
30
0
5
10
15
Fish Length (cm)
Month
06
07
08
09

Figure 8.

Figure 9.

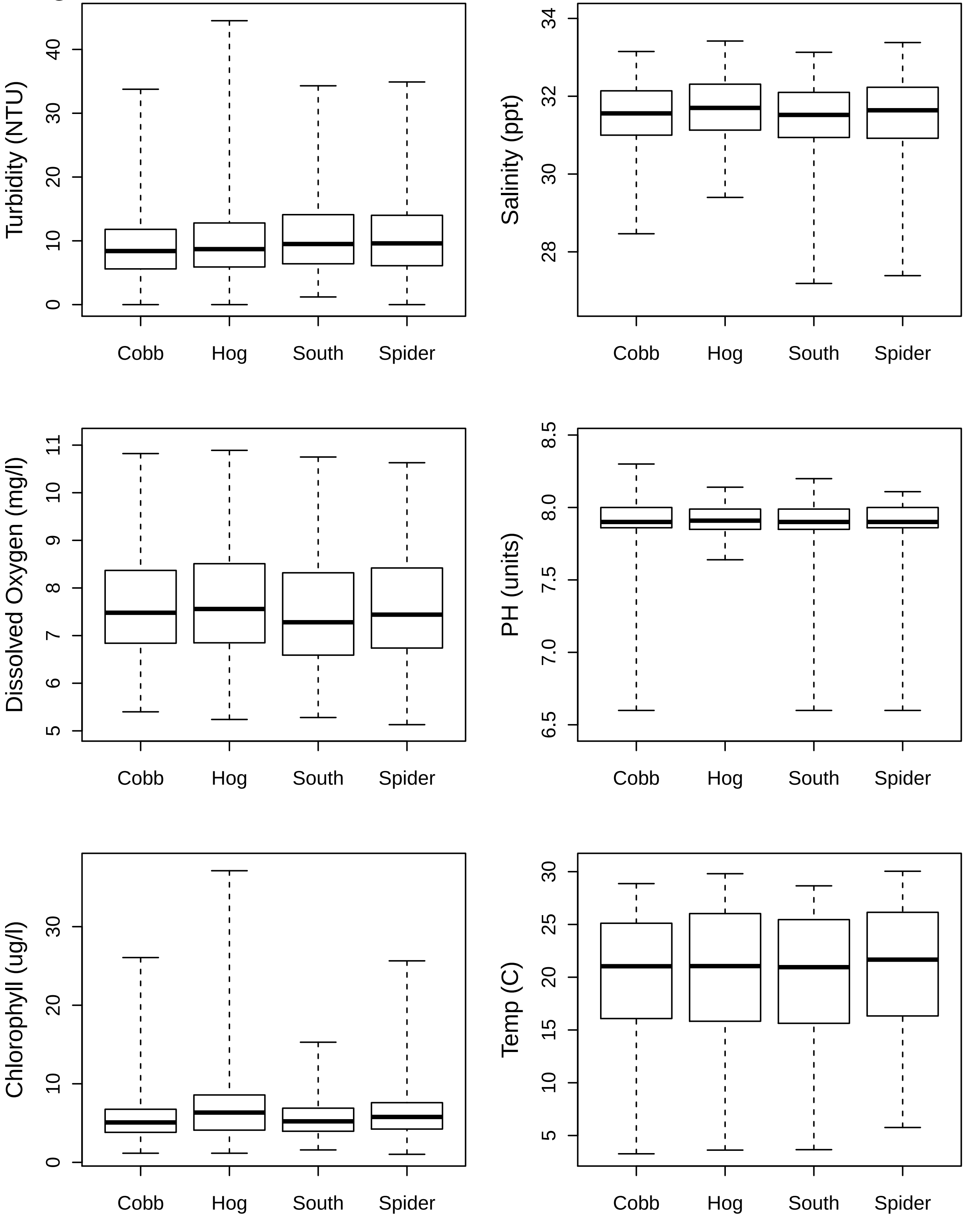
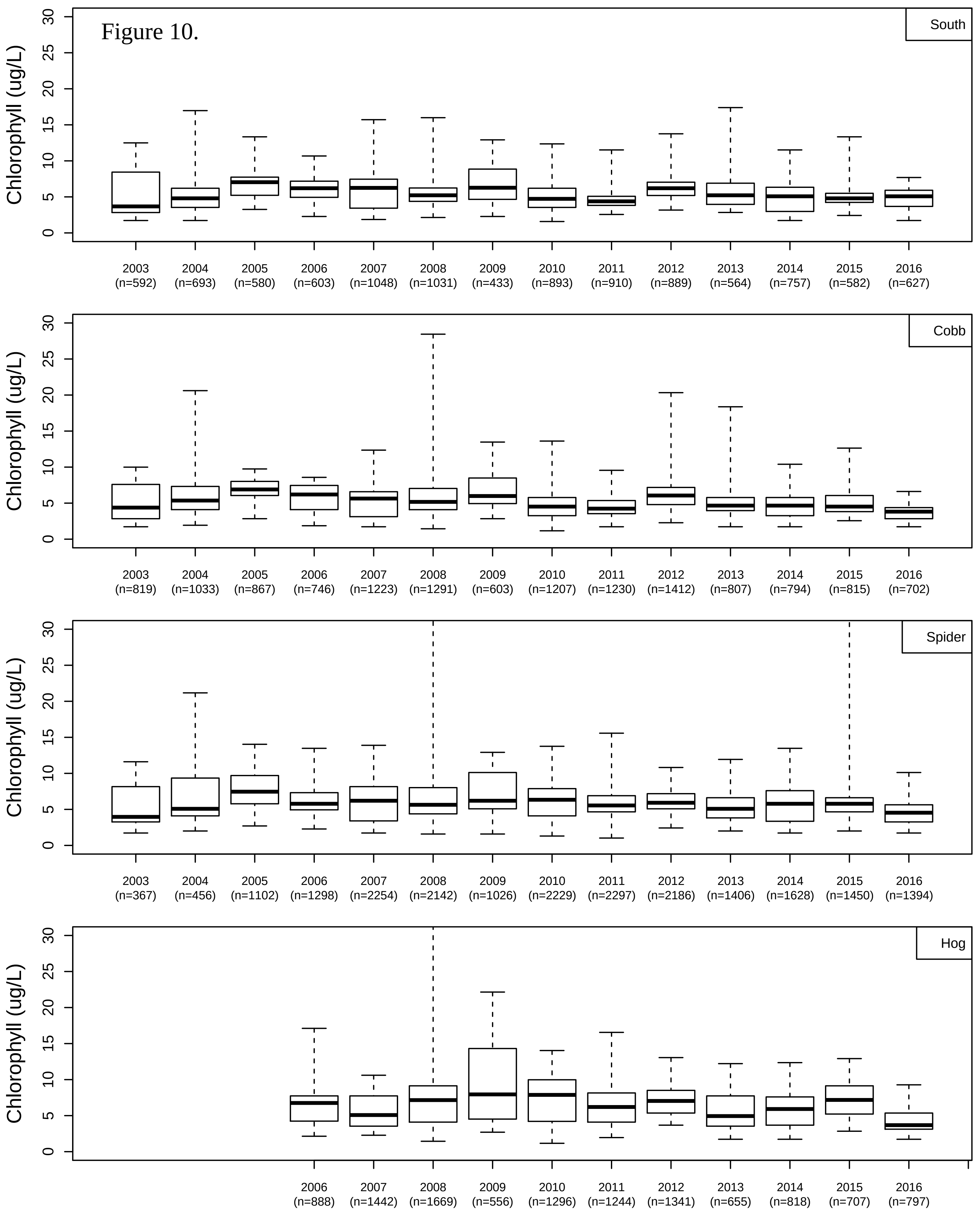

Turbidity (NTU)
0
10
20
30
40
Cobb
Hog
South
Spider
Salinity (ppt)
28
30
32
34
Cobb
Hog
South
Spider
Dissolved Oxygen (mg/l)
5
6
7
8
9
10
11
Cobb
Hog
South
Spider
PH (units)
6.5
7.0
7.5
8.0
8.5
Cobb
Hog
South
Spider
Chlorophyll (ug/l)
0
10
20
30
Cobb
Hog
South
Spider
Temp (C)
5
10
15
20
25
30
Cobb
Hog
South
Spider

Figure 10.


South
Chlorophyll (ug/L)
0
5
10
15
20
25
30
2003 (n=592)
2004 (n=693)
2005 (n=580)
2006 (n=603)
2007 (n=1048)
2008 (n=1031)
2009 (n=433)
2010 (n=893)
2011 (n=910)
2012 (n=889)
2013 (n=564)
2014 (n=757)
2015 (n=582)
2016 (n=627)
Cobb
Chlorophyll (ug/L)
0
5
10
15
20
25
30
2003 (n=819)
2004 (n=1033)
2005 (n=867)
2006 (n=746)
2007 (n=1223)
2008 (n=1291)
2009 (n=603)
2010 (n=1207)
2011 (n=1230)
2012 (n=1412)
2013 (n=807)
2014 (n=794)
2015 (n=815)
2016 (n=702)
Spider
Chlorophyll (ug/L)
0
5
10
15
20
25
30
2003 (n=367)
2004 (n=456)
2005 (n=1102)
2006 (n=1298)
2007 (n=2254)
2008 (n=2142)
2009 (n=1026)
2010 (n=2229)
2011 (n=2297)
2012 (n=2186)
2013 (n=1406)
2014 (n=1628)
2015 (n=1450)
2016 (n=1394)
Hog
Chlorophyll (ug/L)
0
5
10
15
20
25
30
2006 (n=888)
2007 (n=1442)
2008 (n=1669)
2009 (n=556)
2010 (n=1296)
2011 (n=1244)
2012 (n=1341)
2013 (n=655)
2014 (n=818)
2015 (n=707)
2016 (n=797)

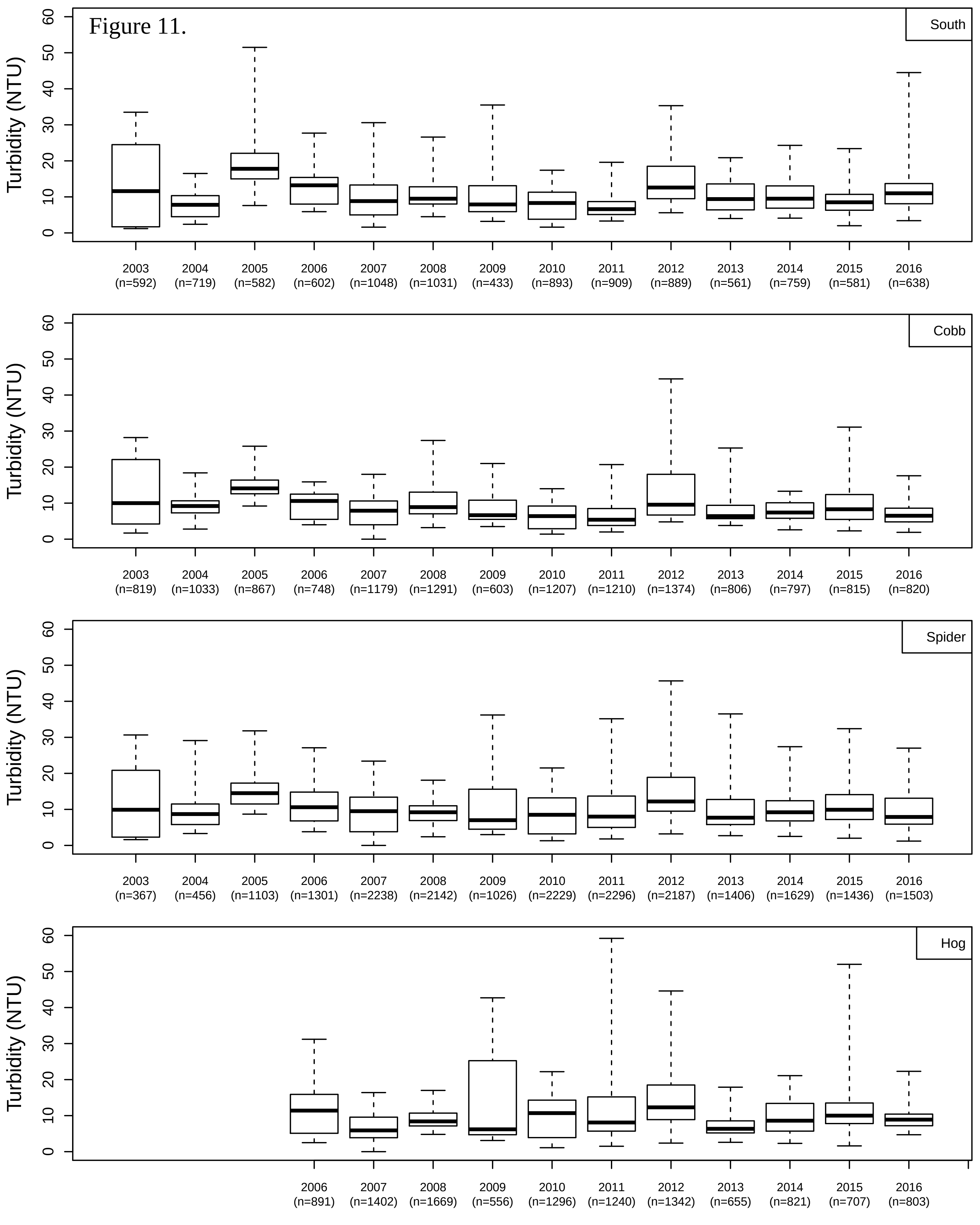
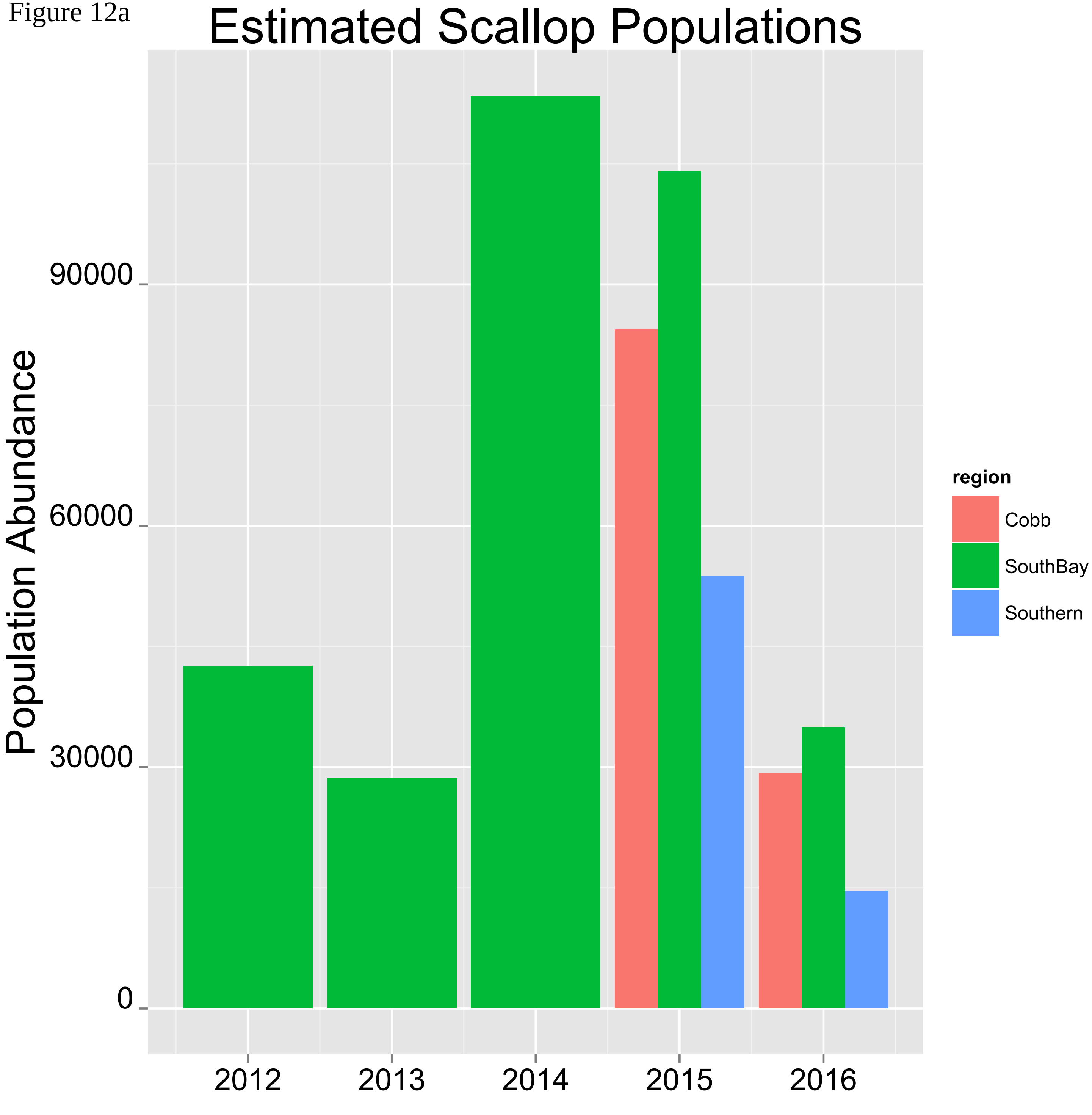


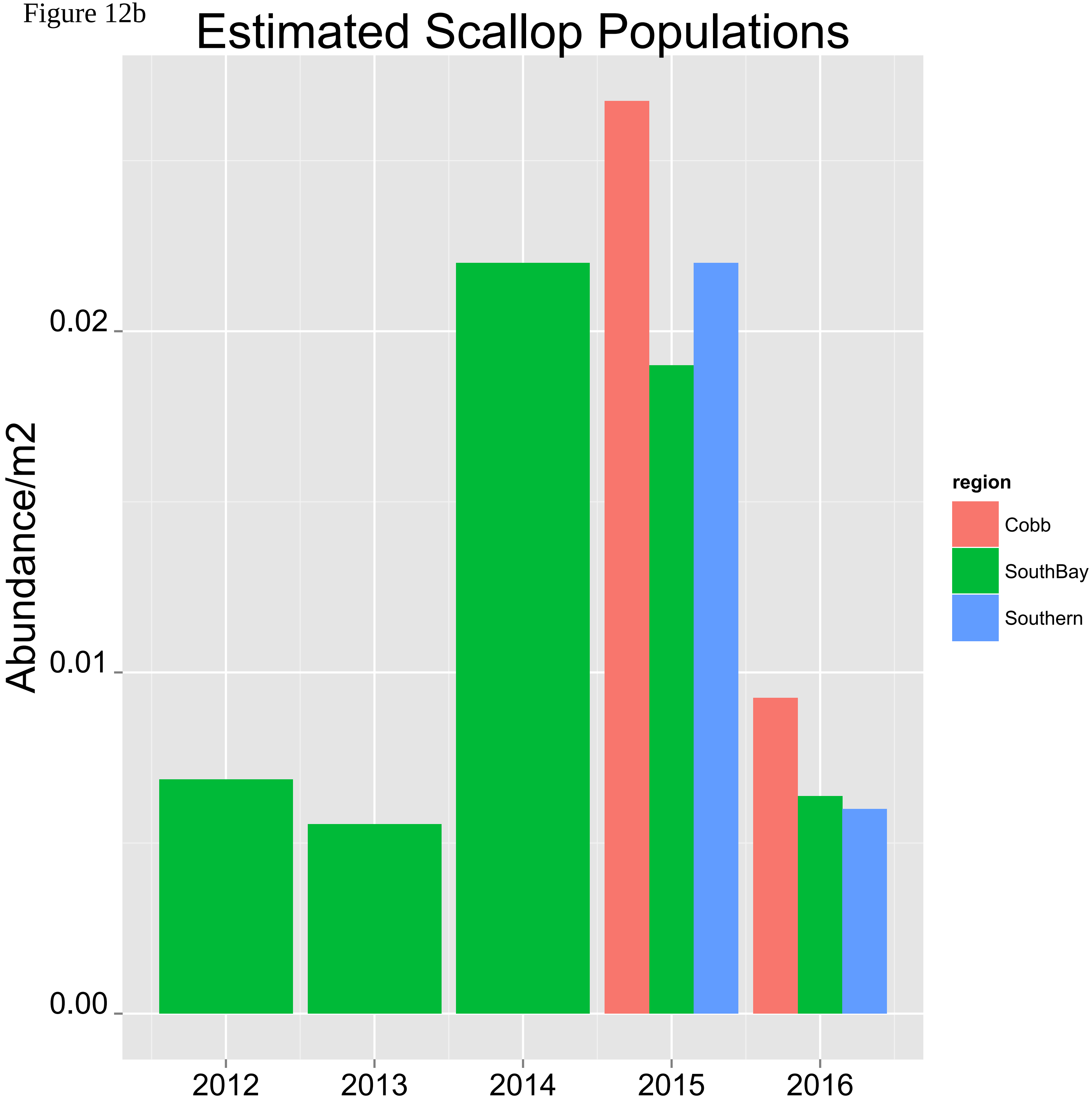
Figure 11.
South
Cobb
Spider
Hog
Turbidity (NTU)
0
10
20
30
40
50
60
2003 (n=592)
2004 (n=719)
2005 (n=582)
2006 (n=602)
2007 (n=1048)
2008 (n=1031)
2009 (n=433)
2010 (n=893)
2011 (n=909)
2012 (n=889)
2013 (n=561)
2014 (n=759)
2015 (n=581)
2016 (n=638)
2003 (n=819)
2004 (n=1033)
2005 (n=867)
2006 (n=748)
2007 (n=1179)
2008 (n=1291)
2009 (n=603)
2010 (n=1207)
2011 (n=1210)
2012 (n=1374)
2013 (n=806)
2014 (n=797)
2015 (n=815)
2016 (n=820)
2003 (n=367)
2004 (n=456)
2005 (n=1103)
2006 (n=1301)
2007 (n=2238)
2008 (n=2142)
2009 (n=1026)
2010 (n=2229)
2011 (n=2296)
2012 (n=2187)
2013 (n=1406)
2014 (n=1629)
2015 (n=1436)
2016 (n=1503)
2006 (n=891)
2007 (n=1402)
2008 (n=1669)
2009 (n=556)
2010 (n=1296)
2011 (n=1240)
2012 (n=1342)
2013 (n=655)
2014 (n=821)
2015 (n=707)
2016 (n=803)

Figure 12a



Estimated Scallop Populations
Population Abundance
90000
60000
30000
0
2012
2013
2014
2015
2016
region
Cobb
SouthBay
Southern

Figure 12b


Estimated Scallop Populations
Abundance/m2
0.00
0.01
0.02
2012
2013
2014
2015
2016
region
Cobb
SouthBay
Southern