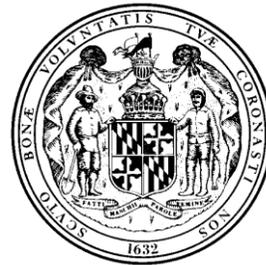


**PERFORMANCE REPORT FOR FEDERAL AID GRANT F-63-R, SEGMENT 6**

**2015**

**MARINE AND ESTUARINE FINFISH ECOLOGICAL AND HABITAT  
INVESTIGATIONS**



Maryland Department of Natural Resources

Fisheries Service

Tawes State Office Building B-2

Annapolis, Maryland 21401

April, 2016

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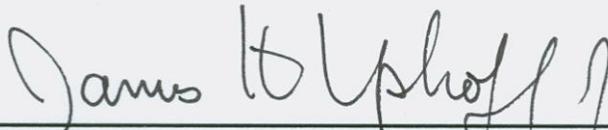


**Approval**



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**Sarah Widman, Director**  
**Policy and Planning Division**  
**Maryland Fisheries Service**  
**Department of Natural Resources**



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**James H. Uphoff, Jr.**  
**Fisheries Habitat and Ecosystem Program**  
**Policy and Planning Division**  
**Maryland Fisheries Service**  
**Department of Natural Resources**

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| Sampling          | Volunteer       | Sampling   | Volunteer        |
|-------------------|-----------------|------------|------------------|
| Mattawoman Creek  | Bob Boxwell     | Bush River | Sharyn Spray     |
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|                   | Ken Hastings    |            | Bryon Bodt       |
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|                   | Yvonne Irvin    |            | Alex Krach       |
|                   | C.J. Schlick    |            | Jeniece Suddoth  |
|                   | Kyle Schlick    |            | Jenn Raulin      |
|                   |                 |            | Andrew Mandovi   |
|                   | Bob Shakeshaft  |            |                  |
|                   | Jackie Fisher   |            |                  |
|                   | Jenn Raulin     |            |                  |
| Summer Surveys    | Nestina Jackson |            | Ellen O'Hanlon   |
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|                   |                 |            | Alex Krach       |
|                   |                 |            | Hope Gerstler    |
|                   |                 |            | Jeniece Suddoth  |
|                   |                 |            | Rebecca          |
| Striped Bass Diet | Jim Price       |            | Mastripolito     |
|                   | Henrietta Price |            | Trey Orman       |
|                   | Joe Boone       |            |                  |
|                   | Chuck Prah      | Deer Creek | Jim Thompson     |
|                   | Ed Kilduff      |            |                  |

**Project Staff**  
 Jim Uphoff  
 Margaret McGinty  
 Alexis Park  
 Carrie Hoover  
 Shaun Miller  
 Charles Johnson  
 Patrick Donovan

**Report Organization**

This report was completed during April, 2016. It consists of summaries of activities for Jobs 1–4 under this grant cycle. All pages are numbered sequentially; there are no separate page numbering systems for each Job. Job 1 activities are reported in separate numbered sections. For example, Job 1, section 1 would cover development reference points (Job 1) for stream spawning habitat of anadromous fish (Section 1). Tables in a Job are numbered as section number – table number (1-1, 1-2, etc). Figures are numbered in the same fashion. Throughout the report, multiple references to past annual report analyses are referred to. The complete PDF versions of many past annual reports can be found under the Publications and Report link on the Fisheries Habitat and Ecosystem (FHEP) website page on the Maryland DNR website. The website address is <http://www.dnr.maryland.gov/fisheries/fhep/>. Table 1 provides the page number for each job and section.

Table 1. Job and section number, topic covered, and page number.

| Job | Section | Topic                                      | Pages   |
|-----|---------|--|---------|
| 1   | 1       | Anadromous fish stream spawning            | 6-38    |
| 1   | 2       | Yellow Perch larval dynamics               | 39-72   |
| 1   | 3       | Summer fish community and habitat dynamics | 73-119  |
| 2   |         | Supporting activities                      | 120-124 |
| 3   |         | Spatial data for prioritizing habitat      | 125-154 |
| 4   |         | Striped Bass forage benchmarks             | 155-197 |

**SURVEY TITLE: MARINE AND ESTUARINE FINFISH ECOLOGICAL AND HABITAT INVESTIGATIONS**  
**PROJECT 1: FINFISH HABITAT AND MANAGEMENT**

**Job 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern: development targets and thresholds**

Carrie Hoover, Alexis Park, Jim Uphoff, Margaret McGinty, Charles Johnson, and Patrick Donovan

**Executive Summary**

*Section 1: Stream Ichthyoplankton* – Proportions of stream drift net samples with an anadromous fish group's eggs or larvae were compared to level of development indicated by structures per hectare or C/ha ( $C/ha \leq 0.27$  is considered a target and  $\geq 0.83$ , a threshold) and conductivity, an indicator of water quality strongly associated with development. Anadromous fish groups were White Perch, Yellow Perch or "Herring" (Blueback Herring, Alewife, American Shad, and Hickory Shad). Surveys were conducted in Mattawoman Creek ( $C/ha = 0.46$  in 1991 and 0.87-0.91 in 2008-2015), Piscataway Creek ( $C/ha = 1.41-1.49$  in 2008-2009 and 2012-2014), Bush River ( $C/ha = 1.37-1.51$  in 2005-2008 and 2014) and Deer Creek ( $C/ha = 0.24$  in 2012-2015). Most of these surveys were conducted by citizen-scientist volunteers.

Proportion of samples with Herring eggs and-or larvae ( $P_{herr}$ ) provided reasonably precise annual estimates of relative abundance based on encounter rate. Regression analyses indicated a negative relationships of  $P_{herr}$  with C/ha and conductivity, and a positive relationship of C/ha with conductivity. These associations were consistent with the hypothesis that urbanization was detrimental to stream spawning. Changes in  $P_{herr}$  from year to year may have indicated how much habitat was available or how attractive it was rather than fluctuations in abundance of spawners since  $P_{herr}$  was stable in the watershed at the target level of development and more variable in those at or past the development threshold. In developed watersheds, a combination of urban and natural stream processes may create varying amounts of ephemeral Herring spawning habitat annually and dampen spawning migrations through changes in water chemistry (indicated by conductivity). Herring make more extensive use of stream spawning than the other two anadromous groups studied.

We pooled Mattawoman Creek data across years to overcome the effect of their limited spatial distribution on annual sample size in order to estimate proportions of samples with White Perch eggs and larvae or Yellow Perch larvae. This allowed us to compare for 1989-1991 collections ( $C / ha = 0.43-0.47$ ) with 2008-2010 ( $C / ha = 0.87-0.90$ ), and 2011-2015 ( $C / ha = 0.90-0.91$ ) at the same combinations of downstream sites. These estimates did not detect a loss in stream spawning for Yellow Perch. Less White Perch stream spawning was detected during 2008-2010 than the other time periods. Proportions of stream samples with White Perch eggs or larvae were similar for 1989-1991 and 2011-2015. Yellow Perch stream spawning did not extend as far upstream as white perch spawning.

*Section 2: Estuarine Yellow Perch Larval Sampling* - Presence-absence sampling for Yellow Perch larvae in 2015 was conducted with towed, conical 0.5-m ichthyoplankton nets in the upper tidal reaches of the Choptank, Nanticoke, and Patuxent Rivers, and in Mattawoman Creek during the month of April and through the first week of May. Annual  $L_p$ , the proportion of tows with Yellow Perch larvae during a standard time period and where larvae would be

expected, provided a cost-effective measure of the product of egg production and survival through the early post-larval stage.

Estimates of  $L_p$  declined perceptibly once watershed development exceeded the suburban threshold (0.83 structures per hectare, C/ha, equivalent to 10% impervious surface, IS). Interpretation of the influence of subestuary salinity class (tidal-fresh or brackish) or agricultural and forest land cover on  $L_p$  was hindered because existing patterns of development did not represent all possible combinations.

Correlation and regression analyses indicated that development influenced the percentage of wetlands present within a watershed, which could in turn influence organic matter (OM) and larval Yellow Perch feeding dynamics. Analysis indicated an optimum level of OM for first-feeding larvae existed for the watersheds studied. Too much OM may have functioned as a predation refuge and too little OM may have limited zooplankton production. Comparisons of RNA/DNA ratios of first-feeding larvae between two watersheds below the development target with two watersheds with development above, but near, the threshold have not supported differences indicated by correlation and regression comparisons of watershed development, OM levels and feeding success during 2010-2015 (N = 31).

*Section 3: Estuarine Fish Community Summer Sampling* – In 2015, we continued to evaluate summer nursery and adult habitat for recreationally important finfish in tidal-fresh, oligohaline (low salinity), and mesohaline (brackish) subestuaries of Chesapeake Bay. Correlation analyses suggested that DO responded to temperature at depth and C/ha differently depending on subestuary salinity classification. Negative associations of surface or bottom DO with corresponding mean water temperatures at depth in oligohaline subestuaries suggested respiration was a major consideration in this class of subestuary. Associations of temperature and DO were not detected in mesohaline or tidal-fresh subestuaries. The strongest and only negative association between bottom DO and C/ha was found in mesohaline subestuaries, where strongest stratification was expected. Positive associations of surface DO with development were suggested for fresh-tidal and oligohaline subestuaries and in fresh-tidal subestuaries. A positive association was suggested for bottom DO and C/ha in fresh-tidal subestuaries; a relationship was not suggested for oligohaline subestuaries; and a negative relationship was suggested for mesohaline subestuaries. Plots of species richness in seine and trawl samples and C/ha did not suggest a relationship in tidal-fresh or oligohaline subestuaries, but a decline was evident with development in mesohaline subestuaries.

Tidal-fresh Mattawoman Creek's finfish abundance during 1989-2015 appeared to exhibit boom and bust dynamics after 2001 as the C/ha threshold was approached and breached. "Busts" were concurrent with spikes (2002) or plateaus (2007-2009) of total ammonia nitrogen (TAN). Recovery of fish abundance since 2011 has coincided with moderate values of median TAN. Shifts in ecosystem status observed in Mattawoman Creek may represent ecological shifts to a different unstable or stable state. Approaching and breaching the development threshold in Mattawoman Creek's watershed has been concurrent with changes in stream hydrology and water quality, increased sediment and nutrient loading from stream erosion and construction, decreased chlorophyll a and DO. Water clarity has increased, as has TAN and SAV while finfish abundance has become more variable and less diverse (particularly planktivores) in the subestuary.

In 2015, we explored DO trends and adult White Perch size structure (proportional stock densities or PSD) in adjacent mesohaline subestuaries of the lower Choptank River: Broad Creek (C/ha ~ 0.30 during 2012-2015), Harris Creek (C/ha ~ 0.39 during 2012-2015), and Tred Avon

River ( $C/ha = 0.71-0.75$  during 2007-2015). During 2015, below threshold and target DO readings ( $DO < 3.0$  mg / L and  $5.0$  mg / L, respectively) were more frequent in more developed Tred Avon River. An ANOVA that compared bottom DO by station in Tred Avon indicated that bottom DO at the station at Easton was significantly lower than downstream stations 2, 3, and 4. Deterioration of DO with upstream distance indicated that watershed development emanating from Easton was the source of poor water local quality rather than water intruding from downstream. ANOVAs of Harris Creek or Broad Creek stations and bottom DO concentrations did not indicate significant differences among stations. A higher proportion of White Perch adults in Harris and Broad Creeks were of a size of interest to anglers than more developed Tred Avon River. Size quality of White Perch directly aligned with the percentage of all DO measurements below the target level; however, sample sizes indicated higher abundance in Tred Avon River, so diminished size quality may reflect density-dependent dynamics.

### **Section 1: Stream Ichthyoplankton Sampling**

Carrie Hoover, Alexis Park, Margaret McGinty, Jim Uphoff, Patrick Donovan, and Charles Johnson

#### **Introduction**

Surveys to identify spawning habitat of White Perch, Yellow Perch and “Herring” (Blueback Herring, Alewife, American Shad, and Hickory Shad) were conducted in Maryland during 1970-1986. These data were used to develop statewide maps depicting anadromous fish spawning habitat (O’Dell et al. 1970; 1975; 1980; Mowrer and McGinty 2002). Many of these watersheds have undergone considerable development and recreating these surveys provided an opportunity to explore whether spawning habitat declined in response to urbanization. Surveys based on the sites and methods of O’Dell et al. (1975) were used to sample Mattawoman Creek (2008-2015), Piscataway Creek (2008-2009 and 2012-2014), Bush River (2005-2008 and 2014) and Deer Creek (2012-2015; Figure 1-1).

Mattawoman and Piscataway Creeks are adjacent Coastal Plain watersheds along an urban gradient emanating from Washington, DC (Figure 1-1). Piscataway Creek’s watershed is both smaller than Mattawoman Creek’s and closer to Washington, DC. Bush River is located in the urban gradient originating from Baltimore, Maryland, and is located in both the Coastal Plain and Piedmont physiographic provinces. Deer Creek is entirely located in the Piedmont north of Baltimore, near the Pennsylvania border (Clearwater et al. 2000). Bush River and Deer Creek are adjacent to each other (Figure 1-1).

We developed two indicators of anadromous fish spawning in a watershed based on presence-absence of eggs and larvae: occurrence at a site (a spatial indicator) and proportion of samples with eggs and larvae (a spatial and temporal indicator). Occurrence of eggs or larvae of an anadromous fish group (White Perch, Yellow Perch, or Herring) at a site, recreated the indicator developed by O’Dell et al. (1975; 1980). This spatial indicator was compared to the extent of development in the watershed (counts of structures per hectare or  $C/ha$ ) between the 1970s and the present (Topolski 2015). An indicator of habitat occupation in space and time from collections in the 2000s was estimated as proportion of samples with eggs and-or larvae of anadromous fish groups. Proportion of samples with an anadromous fish group was compared to level of development ( $C/ha$ ) and an indicator of water quality strongly associated with development (conductivity; Wang and Yin 1997; Paul and Meyer 2001; Wenner et al. 2003; Morgan et al. 2007; Carlisle et al. 2010; Morgan et al. 2012).

## Methods

Stream sites sampled for the anadromous fish eggs and larvae during 2005-2015 were typically at road crossings that O'Dell et al. (1975) determined were anadromous fish spawning sites during the 1970s. O'Dell et al. (1975) summarized spawning activity as the presence of any species group egg, larva, or adult at a site. O'Dell et al. (1975) sampled eggs and larvae with stream drift ichthyoplankton nets and adults were sampled by wire traps.

All collections during 2005-2015, with the exception of Deer Creek during 2012-2015, were made by citizen volunteers who were trained and monitored by program biologists. During March to May, 2008-2015, ichthyoplankton samples were collected in Mattawoman Creek from three tributary sites (MUT3-MUT5) and four mainstem sites (MC1-MC4; Figure 1-2; Table 1-1). Tributary site (MUT4) was selected based on volunteer interest and added in 2010, while tributary site (MUTX) was added in 2014. Piscataway Creek stations were sampled during 2008-2009 and 2012-2014 (Figure 1-3; Uphoff et al. 2010). Bush River stations were sampled during 2005-2008 and 2014 (Figure 1-4; McGinty et al. 2009). Deer Creek sites SU01-SU04 were added to sampling in 2012 and sampling continued in 2013-2015 with the addition of site SU05 (Figure 1-5). Table 1-1 summarizes sites, dates, and sample sizes in Mattawoman, Piscataway and Deer Creeks, and Bush River during 2005-2015.

Ichthyoplankton samples were collected at each site using stream drift nets constructed of 360-micron mesh. Nets were attached to a square frame with a 300 • 460 mm opening. The stream drift net configuration and techniques were the same as those used by O'Dell et al. (1975). The frame was connected to a handle so that the net could be held stationary in the stream. A threaded collar on the end of the net connected a mason jar to the net. Nets were placed in the stream for five minutes with the opening facing upstream. Nets were retrieved and rinsed in the stream by repeatedly dipping the lower part of the net and splashing water through the outside of the net to avoid sample contamination. The jar was removed from the net and an identification label describing site, date, time, and collectors was placed in the jar. The jar was sealed and placed in a cooler with ice for transport when collections were made by volunteers. Preservative was not added by volunteers at a site because of safety and liability concerns. After a team finished sampling for the day, the samples were preserved with 10% buffered formalin on site by DNR personnel. Water temperature (°C), conductivity ( $\mu\text{S}/\text{cm}$ ), and dissolved oxygen (DO, mg/L) were recorded at each site using either a hand-held YSI Model 85 meter or a YSI Pro2030 meter. Meters were calibrated for DO each day prior to use. All data were recorded on standard field data forms and verified at the site by a volunteer. Approximately 2-ml of rose bengal dye was added in order to stain the organisms red to aid sorting.

Ichthyoplankton samples were sorted in the laboratory by project personnel. All samples were rinsed with water to remove formalin and placed into a white sorting pan. Samples were sorted systematically (from one end of the pan to another) under a 10x bench magnifier. All eggs and-or larvae were removed and were retained in a small vial with a label (site, date, and time) and stored with 20% ethanol for later identification under a microscope. Each sample was sorted systematically a second time for quality assurance (QA). Any additional eggs and-or larvae found were removed and placed in a vial with a label (site, date, time, and QA) and stored with 20% ethanol for identification under a microscope. All eggs and larvae found during sorting (both in original and QA vials) were identified as either Herring (Blueback Herring, Alewife, and Hickory Shad), Yellow Perch, White Perch, unknown (eggs and-or larvae that were too damaged to identify) or other (indicating another fish species) and a total count (combining both original

and QA vials) for each site was recorded, as well as the presence and absence of each of the above species. The three Herring species' eggs and larvae are very similar (Lippson and Moran 1974) and identification to species can be problematic. American Shad eggs and larvae would be larger at the same stages of development than those identified as Herring (Lippson and Moran 1974) and none have been detected in our surveys. Quality assurance vials only contained additional eggs and-or larvae of target species already present in the original vials. No new target species were detected during the assessment of the QA vials.

We used property tax map based counts of structures in a watershed, standardized to hectares (C/ha), as our indicator of development (Uphoff et al. 2012; Topolski 2015). This indicator has been provided to us by M. Topolski (MD DNR). Tax maps are graphic representations of individual property boundaries and existing structures that help State tax assessors locate properties (Maryland Department of Planning or MDP 2013). All tax data were organized by county. Since watersheds straddle political boundaries, one statewide tax map was created for each year of available tax data, and then subdivided into watersheds. Maryland's tax maps are updated and maintained electronically as part of MDP's Geographic Information System's (GIS) database. Files were managed and geoprocessed in ArcGIS 9.3.1 from Environmental Systems Research Institute (ESRI 2009). All feature datasets, feature classes, and shapefiles were spatially referenced using the NAD\_1983\_StatePlane\_Maryland\_FIPS\_1900 projection to ensure accurate feature overlays and data extraction. ArcGIS geoprocessing models were developed using Model Builder to automate assembly of statewide tax maps, query tax map data, and assemble summary data. MdProperty View tax data are annually updated by each Maryland jurisdiction to monitor the type of parcel development for tax assessment purposes. To create watershed land tax maps, each year's statewide tax map was clipped using the MD 8-digit watershed boundary file; estuarine waters were excluded. These watershed tax maps were queried for all parcels having a structure built from 1700 to the tax data year. A large portion of parcels did not have any record of year built for structures, but consistent undercounts should not have presented a problem since we were interested in the trend and not absolute magnitude (Uphoff et al. 2012). Mattawoman Creek C/ha declined between 2011 and 2012 and then returned to a higher level in 2013. We replaced the 2012 estimate of C / ha for Mattawoman Creek with the average of 2011 and 2013.

Uphoff et al. (2012) developed an equation to convert annual estimates of C/ha to estimates of impervious surface (IS) calculated by Towson University from 1999-2000 satellite imagery. Estimates of C/ha that were equivalent to 5% IS (target level of development for fisheries; a rural watershed), 10% IS (development threshold for a suburban watershed), and 15% IS (highly developed suburban watershed) were estimated as 0.27, 0.83, and 1.59 C/ha, respectively (Uphoff et al. 2012).

Mattawoman Creek's watershed equaled 25,168 ha and estimated C/ha was 0.87-0.91 during 2008-2015; Piscataway Creek's watershed equaled 17,999 ha and estimated C/ha was 1.41-1.49 during 2008-2015; and Bush River's watershed equaled 39,644 ha and estimated C/ha was 1.37-1.51 during 2005-2015; (M. Topolski, MD DNR, personal communication). Deer Creek (Figure 1-1), a tributary of the Susquehanna River, was added in 2012 as a spawning stream with low watershed development (watershed area = 37,702 ha and development level = 0.24 C/ha). It was sampled in 2012-2015 by DNR biologists from the Fishery Management Planning and Fish Passage Program at no charge to this grant.

Conductivity measurements collected for each date and stream site (mainstem and tributaries) during 2008-2015 from Mattawoman Creek were plotted and mainstem

measurements were summarized for each year. Mainstem sites would be influenced by development in Waldorf, while the monitored tributaries would not. Unnamed tributaries were excluded from calculation of summary statistics to capture conditions in the largest portion of habitat. Comparisons were made with conductivity minimum and maximum reported for Mattawoman Creek during 1991 by Hall et al. (1992). Conductivity data were similarly summarized for Piscataway Creek mainstem stations during 2008-2009 and 2012-2014. A subset of Bush River stations that were sampled each year during 2005-2008 and 2014 (i.e., stations in common) were summarized; stations within largely undeveloped Aberdeen Proving Grounds were excluded because they were not sampled every year. Conductivity was measured with each sample in Deer Creek in 2012-2015.

A water quality database maintained by DNR's Tidewater Ecosystem Assessment (TEA) Division (S. Garrison, MD DNR, personal communication) provided conductivity measurements for Mattawoman Creek during 1970-1989. These historical measurements were compared with those collected in 2008-2015 to examine changes in conductivity over time. Monitoring was irregular for many of the historical stations. Table 1-2 summarizes site location, month sampled, total measurements at a site, and what years were sampled. Historical stations and those sampled in 2008-2015 were assigned river kilometers (RKM) using a GIS ruler tool that measured a transect approximating the center of the creek from the mouth of the subestuary to each station location. Stations were categorized as tidal or non-tidal. Conductivity measurements from eight non-tidal sites sampled during 1970-1989 were summarized as monthly medians. These sites bounded Mattawoman Creek from its junction with the estuary to the city of Waldorf (Route 301 crossing), the major urban influence on the watershed. Historical monthly median conductivities at each mainstem Mattawoman Creek non-tidal site were plotted with 2008-2015 spawning season median conductivities.

Presence of White Perch, Yellow Perch, and Herring eggs and-or larvae at each station in 2015 was compared to past surveys to determine which sites still supported spawning. We used the criterion of detection of eggs and-or larvae at a site (O'Dell et al. 1975) as evidence of spawning. Raw data from early 1970s collections were not available to formulate other metrics.

Four Mattawoman Creek mainstem stations sampled in 1971 by O'Dell et al. (1975) were sampled by Hall et al. (1992) during 1989-1991 for water quality and ichthyoplankton. Count data were available for 1991 ( $C/ha = 0.46$ ) in a tabular summary at the sample level and these data were converted to presence-absence. Hall et al. (1992) collected ichthyoplankton with 0.5 m diameter plankton nets (3:1 length to opening ratio and  $363\mu$  mesh set for 2-5 minutes, depending on flow) suspended in the stream channel between two posts instead of stream drift nets. Changes in spawning site occupation among the current study (2008-2015), 1971 (O'Dell et al. 1975) and 1991 (Hall et al. 1992) were compared to  $C/ha$  in Mattawoman Creek. Historical and recent  $C/ha$  were compared to site occupation for Piscataway Creek 1971 (O'Dell et al. 1975), 2008-2009, and 2012-2014; Bush River 1973 (O'Dell et al. 1975), 2005-2008 (McGinty et al. 2009; Uphoff et al. 2010) and 2014; and Deer Creek 1972 (O'Dell et al. 1975) and 2012-2015.

The proportion of samples where Herring eggs and-or larvae were present ( $P_{herr}$ ) was estimated for Mattawoman Creek mainstem stations (MC1-MC4) during 1991 and 2008-2015. Volunteer sampling of ichthyoplankton in Piscataway Creek (2008-2009 and 2012-2014), Bush River (2005-2008 and 2014; McGinty et al. 2009), and Deer Creek (2012-2015) also provided sufficient sample sizes to estimate  $P_{herr}$ . Herring was the only species group represented with adequate sample sizes for annual estimates with reasonable precision. Mainstem stations (PC1-

PC3) and Tinkers Creek (PTC1) were used in Piscataway Creek (Figure 1-3). Only sites in streams that were sampled in all years (sites in common) in the Bush River drainage were analyzed (Figure 1-4; see Uphoff et al. 2014 for sites sampled in other years). Deer Creek stations SU01, SU04, and SU05 correspond to O'Dell et al. (1975) sites 1, 2, and 3 respectively (Figure 1-5). Two additional sites, SU02 and SU03 are sampled in this system as well.

For the rivers and stations described above, the proportion of samples with Herring eggs and-or larvae present was estimated as:

$$(1) P_{herr} = N_{present} / N_{total};$$

where  $N_{present}$  equaled the number of samples with Herring eggs and-or larvae present and  $N_{total}$  equaled the total number of samples taken. The SD of each  $P_{herr}$  was estimated as:

$$(2) SD = [(P_{herr} \cdot (1 - P_{herr})) / N_{total}]^{0.5} \text{ (Ott 1977).}$$

The 90% confidence intervals were constructed as:

$$(3) P_{herr} \pm (1.44 \cdot SD).$$

White Perch and Yellow Perch have been present in samples at the downstream-most one or two stations in Mattawoman Creek during 1989-1991 (Hall et al. 1992) and 2008-2015. We pooled two to three years (1989-1991, 2008-2010, 2011-2013, and 2014-2015) to estimate the proportion of samples with White or Yellow Perch eggs and larvae in order to gain enough precision to separate these estimates from zero. Formulae for estimating proportions were the same as for estimating  $P_{herr}$  and its SD and 90% CI's (see above). White Perch spawning occurred at MC1 and MC2. Yellow Perch spawning was only detected at Station MC1.

Regression analyses examined relationships of development (C/ha) with standardized conductivity measurements (median conductivity adjusted for Coastal Plain or Piedmont background level; see below), C/ha and Herring spawning intensity ( $P_{herr}$ ), and standardized conductivity with  $P_{herr}$ . Data were from Bush River and Mattawoman, Piscataway, and Deer Creeks. Twenty-three estimates of C/ha and  $P_{herr}$  were available (1991 estimates for Mattawoman Creek could be included), while twenty-two estimates were available for standardized conductivity (Mattawoman Creek data were not available for 1991). Examination of scatter plots suggested that a linear relationship was the obvious choice for C/ha and  $P_{herr}$ , but that either linear or curvilinear relationships might be applicable to C/ha with standardized conductivity and standardized conductivity with  $P_{herr}$ . Power functions were used to fit curvilinear models:

$$(4) Y = a \cdot X^b;$$

where Y = dependent variable (standardized conductivity or  $P_{herr}$ ), X = independent variable (standardized conductivity or C/ha), a is a scaling coefficient and b is a shape parameter. Linear regressions were analyzed in Excel, while the non-linear regression analysis used Proc NLIN in SAS (Freund and Littell 2006). A linear or nonlinear model was considered the best description if it was significant at  $\alpha < 0.05$  (both were two parameter models), it explained more variability than the other ( $r^2$  for linear or approximate  $r^2$  for nonlinear) and examination of residuals did not suggest a problem. We expected negative relationships of  $P_{herr}$  with C/ha and standardized conductivity, while standardized conductivity and C/ha were expected to be positively related.

Conductivity was summarized as the median for the same stations that were used to estimate  $P_{herr}$  and was standardized by dividing by an estimate of the background expected from a stream absent anthropogenic influence (Morgan et al. 2012; see below). Piedmont and Coastal Plain streams in Maryland have different background levels of conductivity (Morgan et al. 2012). Morgan et al. (2012) provided two sets of methods of estimating spring base flow background conductivity for two different sets of Maryland ecoregions, for a total set of four

potential background estimates. We chose the option featuring Maryland Biological Stream Survey (MBSS) Coastal Plain and Piedmont regions and the 25<sup>th</sup> percentile background level for conductivity. These regions had larger sample sizes than the other options and background conductivity in the Coastal Plain fell much closer to the observed range estimated for Mattawoman Creek in 1991 (61-114  $\mu\text{S}/\text{cm}$ ) when development was relatively low (Hall et al. 1992). Background conductivity used to adjust median conductivities was 109  $\mu\text{S}/\text{cm}$  in Coastal Plain streams and 150  $\mu\text{S}/\text{cm}$  in Piedmont streams.

## Results

Development level of the watersheds of Piscataway, Mattawoman, and Deer Creeks and Bush River started at approximately 0.05 C/ha in 1950, (Figure 1-6). Surveys conducted by O'Dell et al. (1975, 1980) in the 1970s, sampled largely rural watersheds (C/ha < 0.27) except for Piscataway Creek (C/ha = 0.47). By 1991, C/ha in Mattawoman Creek was similar to that of Piscataway in 1971. By the mid-2000s Bush River and Piscataway Creek were at higher suburban levels of development (~1.30 C/ha) than Mattawoman Creek (~0.80 C/ha). Deer Creek, zoned for agriculture and preservation, remained rural through 2015 (0.24 C/ha; Figure 1-6).

Conductivity measurements in mainstem Mattawoman Creek during 2008-2015 never fell within the range observed during 1991 (Figure 1-7). Conductivity in Mattawoman Creek tributaries sampled during 2008-2015 often fell within the range observed during 1991.

In 2015, conductivity measurements in mainstem Mattawoman Creek were highly elevated in March (> 200  $\mu\text{S}/\text{cm}$ ) and declined over the next two months, but never fell to the 1991 maximum (114  $\mu\text{S}/\text{cm}$ ; Figure 1-7). Conductivity in tributary MUT3 was elevated above the 1991 maximum for three of 11 measurements, similar to what was observed in 2014. Elevations like these were not observed in the tributaries during 2009-2013 (Figure 1-7). Conductivity values and samples in tributaries MUT4 and MUT5 were only sampled on two occasions in 2015 due to the discovery of beaver dams that appeared to be blocking spawning access, but those measurements fell within or below the range reported by Hall et al. (1992) for the mainstem. Conductivities in Mattawoman Creek's mainstem stations in 2009 were highly elevated in early March following application of road salt in response to a significant snowfall that occurred just prior to the start of the survey (Uphoff et al. 2010). Measurements during 2009 steadily declined for nearly a month before leveling off slightly above the 1989-1991 maximum. 2015 had extremely cold temperatures and higher snowfall than previous years, with the exception of 2014, and a conductivity pattern similar to 2009 (Figure 1-7). Higher conductivity at the most upstream mainstem site (MC4) followed by declining conductivity downstream to the site on the tidal border is a general pattern in all years. This, along with low conductivities typically seen at the unnamed tributaries, indicates that development at and above MC4 associated with Waldorf affected water quality (Figure 1-7).

Conductivity levels in Piscataway Creek and Bush River have been elevated when compared to Mattawoman Creek (Table 1-3). With the exception of Piscataway Creek in 2012 (median = 195  $\mu\text{S}/\text{cm}$ ), median conductivity estimates during spawning surveys were always greater than 200  $\mu\text{S}/\text{cm}$  in Piscataway Creek and Bush River during the 2000s. Median conductivity in Mattawoman Creek was greater than 200  $\mu\text{S}/\text{cm}$  during 2009, but was less than 155  $\mu\text{S}/\text{cm}$  during the next four years. With increased snowfalls seen in 2014 and 2015, median conductivity rose to 166 and 173  $\mu\text{S}/\text{cm}$ , respectively (Table 1-3).

During 1970-1989, 73% of monthly median conductivity estimates in Mattawoman Creek were at or below the background level for Coastal Plain streams; C/ha in the watershed

increased from 0.25 to 0.41. Higher monthly median conductivities in the non-tidal stream were more frequent nearest the confluence with Mattawoman Creek's estuary and in the vicinity of Waldorf (RKM 35) (Figure 1-8). Conductivity medians were highly variable at the upstream station nearest Waldorf during 1970-1989. During 2008-2015 ( $C/ha = 0.85-0.91$ ), median spawning survey conductivities at mainstem stations MC2 to MC4, above the confluence of Mattawoman Creek's stream and estuary (MC1), were elevated beyond nearly all 1979-1989 monthly medians and increased with upstream distance toward Waldorf. Most measurements at MC1 fell within the upper half of the range observed during 1970-1989 (Figure 1-8). None of the non-tidal conductivity medians estimated at any mainstem site during 2008-2015 were at or below the Coastal Plain stream background criterion.

Herring spawning was detected at all mainstem stations in Mattawoman Creek during 1971 and 1991 (Table 1-4). Herring spawning in fluvial Mattawoman Creek was detected at two mainstem sites during 2008-9 and all four mainstem stations during 2010-2015. Herring spawning was not detected at MUT 3 during 2008-2009, but was consistently present afterwards. Spawning was intermittent at MUT 4 and MUT 5 in sampling during the 2000s. During 1971 and 1989-1991, White Perch spawning occurred annually at MC1 and intermittently at MC2. Stream spawning of White Perch in Mattawoman Creek was not detected during 2009, 2011, and 2012, but spawning was detected at MC1 during 2008, 2010 and 2013-2015, and at MC2 during 2013 and 2014. Spawning was detected at MC3 during 1971, but has not been detected since then. Station MC1 was the only stream station in Mattawoman Creek where Yellow Perch spawning has been detected in surveys conducted since 1971. Yellow Perch spawning occurred at station MC1 every year except 2009 and 2012. (Table 1-4).

Herring spawning was detected at all mainstem sites in Piscataway Creek in 2012-2014. Stream spawning of anadromous fish had nearly ceased in Piscataway Creek between 1971 and 2008-2009 (Table 1-5). Herring spawning was not detected at any site in the Piscataway Creek drainage during 2008 and was only detected on one date and location (one Herring larvae on April 28 at PC2) in 2009. Stream spawning of White Perch was detected at PC1 and PC2 in 1971, was not detected during 2008-2009 and 2012-2013, but was detected at PC1 in 2014 (Table 1-5).

Changes in stream site spawning of Herring, White Perch, and Yellow Perch in the Bush River stations during 1973, 2005-2008, and 2014 were not obvious (Table 1-6). Herring eggs and larvae were present at three to five stations (not necessarily the same ones) in any given year sampled. Occurrences of White and Yellow Perch eggs and larvae were far less frequent during 2005-2008 than 1973 and 2014 (Table 1-6).

O'Dell et al. (1975) reported that Herring, White Perch, and Yellow Perch spawned in Deer Creek during 1972 (Table 1-7). Three sites were sampled during 1972 in Deer Creek and one was located upstream of an impassable dam near Darlington (a fish passage was installed there in 1999). During 1972, Herring spawning was detected at both sites below the dam (SU01 and SU03), while White and Yellow Perch spawning were detected at the mouth (SU01). During 2012-2015, Herring spawning was detected at all sites sampled in each year. White Perch spawning was not detected in Deer Creek in 2012 but was detected at three sites each in 2013 and 2014, and two sites in 2015. Yellow Perch spawning detection has been intermittent, with two, zero, three, and zero sites showing evidence of spawning in 2012, 2013, 2014, and 2015 respectively (Table 1-7).

The 90% confidence intervals of  $P_{herr}$  (Figure 1-9) provided sufficient precision for us to categorize four levels of stream spawning: very low levels at or indistinguishable from zero

based on confidence interval overlap (level 0); a low level of spawning that could be distinguished from zero (level 1); a mid-level of spawning that could usually be separated from the low levels (level 2); and a high level (3) of spawning likely to be higher than the mid-level. Stream spawning of Herring in Mattawoman Creek was categorized at levels 1 (2008-2009), 2 (2010 and 2012), and level 3 (1991, 2011, and 2013-2015). Spawning in Piscataway Creek was at level 0 during 2008-2009, at level 2 during 2012, and at level 1 during 2013-2014. Bush River Herring spawning was characterized by levels 0 (2006), 1 (2005 and 2007-2008), and 2 (2014). Deer Creek, the least developed watershed, was characterized by the highest level of Herring spawning (level 3) during 2012-2015 (Figure 1-9).

The 90% CI's of proportions of samples with White Perch eggs and larvae at Mattawoman Creek's stations MC1 and MC2, pooled in 2-to-3-year intervals, indicated less stream spawning occurred during 2008-2010 than during 1989-1991 (Figure 1-10). Status of White Perch spawning in Mattawoman Creek during 2011-2015 was not clear since 90% CI's of the proportion of samples with White Perch eggs and larvae during 2011-2015 overlapped both 1989-1991 and 2008-2010. The 90% CI's for stream spawning of Yellow Perch (at MC1 only) overlapped for 1989-1991, 2008-2010, 2011-2013, and 2014-2015 indicating significant change in stream spawning had not been detected (Figure 1-10).

Standardized conductivity increased with development, while  $P_{herr}$  declined with both development and standardized conductivity. Regression analyses indicated significant and logical relationships among  $P_{herr}$ , C/ha, and standardized median conductivity (Table 1-8). The relationship of C/ha with standardized median conductivity was linear, significant, and positive ( $r^2 = 0.44$ ,  $P = 0.0007$ ,  $N = 22$ ; Figure 1-11). Estimates of  $P_{herr}$  were linearly, significantly, and negatively related to C/ha ( $r^2 = 0.54$ ,  $P = <.0001$ ,  $N = 23$ ). A negative curvilinear regression (power function) best described the relationship of  $P_{herr}$  and standardized median conductivity (approximate  $r^2 = 0.33$ ,  $P < 0.0001$ ,  $N = 22$ ; Figure 1-12). Low estimates of  $P_{herr}$  were much more frequent beyond the C/ha threshold (0.83 C/ha) or when standardized conductivity was 1.5-times or more than the baseline level (Figure 1-12).

## Discussion

Proportion of samples with Herring eggs and-or larvae ( $P_{herr}$ ) provided a reasonably precise estimate of habitat occupation based on encounter rate. Regression analyses indicated significant and logical relationships among  $P_{herr}$ , C/ha, and conductivity consistent with the hypothesis that urbanization was detrimental to stream spawning. Conductivity was positively related with C/ha in our analysis and with urbanization in other studies (Wang and Yin 1997; Paul and Meyer 2001; Wenner et al. 2003; Morgan et al. 2007; Carlisle et al. 2010; Morgan et al. 2012). Limburg and Schmidt (1990) found a highly nonlinear relationship of densities of anadromous fish (mostly Alewife) eggs and larvae to urbanization in Hudson River tributaries, reflecting a strong, negative threshold at low levels of development.

An unavoidable assumption of regression analyses of  $P_{herr}$ , C/ha, and summarized conductivity was that watersheds at different levels of development were a substitute for time-series. Extended time-series of watershed specific data were not available. Mixing physiographic provinces in this analysis had the potential to increase scatter of points, but standardizing median conductivity to background conductivity moderated the province effect in analyses with that variable. Differential changes in physical stream habitat and flow with urbanization due to differences in geographic provinces could also have affected fits of regressions. Estimates of C/ha may have indexed these physical changes as well as water chemistry changes, while

standardized conductivity would only have represented changes in water chemistry. Estimates of C/ha explained more variation in  $P_{herr}$  (54%) than standardized conductivity (33%).

Elevated conductivity, related primarily to chloride from road salt (but including most inorganic acids and bases; APHA 1979), has emerged as an indicator of watershed development (Wenner et al. 2003; Kaushal 2005; Morgan et al. 2007; Morgan et al. 2012). Use of salt as a deicer may lead to both “shock loads” of salt that may be acutely toxic to freshwater biota and elevated baselines (increased average concentrations) of chloride that have been associated with decreased fish and benthic diversity (Kaushal et al. 2005; Wheeler et al. 2005; Morgan et al. 2007; 2012). Commonly used anti-clumping agents for road salt (ferro- and ferricyanide) that are not thought to be directly toxic are of concern because they can break down into toxic cyanide under exposure to ultraviolet light. Although the degree of breakdown into cyanide in nature is unclear (Pablo et al. 1996; Transportation Research Board 2007), these compounds have been implicated in fish kills (Burdick and Lipschuetz 1950; Pablo et al. 1996; Transportation Research Board 2007). Heavy metals and phosphorous may also be associated with road salt (Transportation Research Board 2007).

At least two hypotheses can be formed to relate decreased anadromous fish spawning to conductivity and road salt use. First, eggs and larvae may die in response to sudden changes in salinity and potentially toxic amounts of associated contaminants and additives. Second, changing stream chemistry may cause disorientation and disrupted upstream migration.

Levels of salinity associated with our conductivity measurements are very low (maximum 0.2 ppt) and anadromous fish spawn successfully in brackish water (Klauda et al. 1991; Piavis et al. 1991; Setzler-Hamilton 1991). A rapid increase might result in osmotic stress and lower survival since salinity represents osmotic cost for fish eggs and larvae (Research Council of Norway 2009).

Elevated stream conductivity may prevent anadromous fish from recognizing and ascending streams. Alewife and Herring are thought to home to natal rivers to spawn (ASMFC 2009a; ASMFC 2009b), while Yellow and White Perch populations are generally tributary-specific (Setzler-Hamilton 1991; Yellow Perch Workgroup 2002). Physiological details of spawning migration are not well described for our target species, but homing migrations in anadromous American Shad and Salmon have been connected with chemical composition, smell, and pH of spawning streams (Royce-Malmgren and Watson 1987; Dittman and Quinn 1996; Carruth et al. 2002; Leggett 2004). Conductivity is related to total dissolved solids in water (Cole 1975) which reflects chemical composition.

Processes such as flooding, riverbank erosion, and landslides vary by geographic province (Cleaves 2003) and influence physical characteristics of streams. Unconsolidated layers of sand, silt, and clay underlie the Coastal Plain and broad plains of low relief and wetlands characterize the natural terrain (Cleaves 2003). Coastal Plain streams have low flows and sand or gravel bottoms (Boward et al. 1999). The Piedmont is underlain by metamorphic rocks and characterized by narrow valleys and steep slopes, with regions of higher land between streams in the same drainage. Most Piedmont streams are of moderate slope with rock or bedrock bottoms (Boward et al. 1999). The Piedmont is an area of higher gradient change and more diverse and larger substrates than the Coastal Plain (Harris and Hightower 2011) that may offer greater variety of Herring spawning habitats.

Urbanization and physiographic province both affect discharge and sediment supply of streams (Paul and Meyer 2001; Cleaves 2003) that, in turn, could affect location, substrate composition, and extent and success of spawning. Alewife spawn in sluggish flows, while

Blueback Herring spawn in sluggish to swift flows (Pardue 1983). American Shad select spawning habitat based on macrohabitat features (Harris and Hightower 2011) and spawn in moderate to swift flows (Hightower and Sparks 2003). Spawning substrates for Herring include gravel, sand, and detritus (Pardue 1983). Detritus loads in subestuaries are strongly associated with development (see Section 2) and urbanization affects the quality and quantity of organic matter in streams (Paul and Meyer 2001) that feed into subestuaries.

Herring spawning became more variable in streams as watersheds developed. The surveys from watersheds with C/ha of 0.46 or less had high  $P_{herr}$ . Estimates of  $P_{herr}$  from Mattawoman Creek during 2008-2015 (C/ha was 0.85-0.91) varied from barely different from zero to high. Eggs and larvae were nearly absent from fluvial Piscataway Creek during 2008-2009, but  $P_{herr}$  rebounded to 0.45 in 2012 and then dropped again to 0.2 in 2013-2014 (C/ha was 1.39-1.49). The rebound in Herring spawning in Piscataway Creek during 2012 was concurrent with the lowest mean and median conductivities encountered there in the four years sampled. Variability of Herring spawning in Bush River during 2005-2008 and 2014 involved “colonization” of new sites as well as absence from sites of historical spawning (Uphoff et al. 2014). Estimates of  $P_{herr}$  were consistently high in the least developed watershed, Deer Creek.

Ranges of  $P_{herr}$  in study streams may have indicated variability in available spawning habitat or its attractiveness rather than abundance of spawners. In developed watersheds, a combination of urban and natural stream processes may create varying amounts of ephemeral spawning habitat annually and dampen spawning migrations through increased conductivity. Observed variation in  $P_{herr}$  would indicate wide annual and regional fluctuations in population size. However, stock assessments of Alewife and Blueback Herring along the Atlantic coast, including those in Maryland, indicate they are in decline or are at depressed stable levels (ASMFC 2009a; 2009b; Limburg and Waldman 2009; Maryland Fisheries Service 2012) rather than fluctuating.

Application of presence-absence data in management needs to consider whether absence reflects a disappearance from suitable habitat or whether habitat sampled is not really habitat for the species in question (MacKenzie 2005). Our site occupation comparisons were based on the assumption that spawning sites detected in the 1970s were indicative of the extent of habitat. O’Dell et al. (1975) summarized spawning activity as the presence of any species group’s egg, larva, or adult (latter from wire fish trap sampling) at a site and we used this criterion (spawning detected at a site or not) for a set of comparisons. Raw data for the 1970s were not available to formulate other metrics. This site-specific presence-absence approach did not detect permanent site occupation changes or an absence of change since only a small number of sites could be sampled (limited by road crossings) and the positive statistical effect of repeated visits (Strayer 1999) was lost by summarizing all samples into a single record of occurrence in a sampling season. A single year’s record was available for each of the watersheds in the 1970s and we were left assuming this distribution applied over multiple years of low development.

Proportion of positive samples ( $P_{herr}$  for example) incorporated spatial and temporal presence-absence and provided an economical and precise alternative estimate of habitat occupation based on encounter rate. Encounter rate is readily related to the probability of detecting a population (Strayer 1999). Proportions of positive or zero catch indices were found to be robust indicators of abundance of Yellowtail Snapper *Ocyurus chrysurus* (Bannerot and Austin 1983), age-0 White Sturgeon *Acipenser transmontanus* (Counihan et al. 1999), Pacific Sardine *Sardinops sagax* eggs (Mangel and Smith 1990), Chesapeake Bay Striped Bass eggs (Uphoff 1997), and Longfin Inshore Squid *Loligo pealeii* fishery performance (Lange 1991).

Unfortunately, estimating reasonably precise proportions of stream samples with White or Yellow Perch eggs annually was not be logistically feasible without major changes in sampling priorities. Estimates for Yellow or White Perch stream spawning would require more frequent sampling to obtain precision similar to that attained by  $P_{herr}$  since spawning occurred at fewer sites. Given staff and volunteer time limitations, this would not be possible within our current scope of operations. In Mattawoman Creek, it was possible to pool data across years to increase precision of estimates of proportions of samples with White Perch eggs and larvae (sites MC1 and MC2) or Yellow Perch larvae (MC1) for 1989-1991 collections to compare with 2008-2015 collections at the same combinations of sites. These estimates did not indicate a loss in stream spawning in these downstream sites.

Volunteer-based sampling of stream spawning during 2005-2015 used only stream drift nets, while O'Dell et al. (1975) and Hall et al. (1992) determined spawning activity with ichthyoplankton nets and wire traps for adults. Tabular summaries of egg, larval, and adult catches in Hall et al. (1992) allowed for a comparison of how site use in Mattawoman Creek might have varied in 1991 with and without adult wire trap sampling. Sites estimated when eggs and-or larvae were present in one or more samples were identical to those when adults present in wire traps were included with the ichthyoplankton data (Hall et al. 1992). Similar results were obtained from the Bush River during 2006 at sites where ichthyoplankton drift nets and wire traps were used; adults were captured by traps at one site and eggs and-or larvae at nine sites with ichthyoplankton nets (Uphoff et al. 2007). Wire traps set in the Bush River during 2007 did not indicate different results than ichthyoplankton sampling for Herring and Yellow Perch, but White Perch adults were observed in two trap samples and not in plankton drift nets (Uphoff et al. 2008). These comparisons of trap and ichthyoplankton sampling indicated it was unlikely that an absence of adult wire trap sampling would impact interpretation of spawning sites when multiple years of data were available.

The different method used to collect ichthyoplankton in Mattawoman Creek during 1991 could bias that estimate of  $P_{herr}$ , although presence-absence data tend to be robust to errors and biases in sampling (Green 1979). Removal of 1991 data lowered the fit between  $C/ha$  and  $P_{herr}$  (from  $r^2 = 0.54$ ,  $P = <.0001$  to  $r^2 = 0.51$ ,  $P = 0.0002$ ), but did not alter the negative relationship (95% CI's of slopes and intercepts of both models overlapped).

Absence of detectable stream spawning does not necessarily indicate an absence of spawning in the estuarine portion of these systems. Estuarine Yellow Perch presence-absence surveys in Mattawoman and Piscataway Creeks, and Bush River did not indicate that lack of detectable stream spawning corresponded to their elimination from these subestuaries. Yellow Perch larvae were present in upper reaches of both subestuaries, (see Section 2). Yellow Perch do not appear to be dependent on non-tidal stream spawning, but their use may confer benefit to the population through expanded spawning habitat diversity. Stream spawning is very important to Yellow Perch anglers since it provides access for shore fisherman and most recreational harvest probably occurs during spawning season (Yellow Perch Workgroup 2002).

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Table 1-1. Summary of subestuaries, years sampled, number of sites, first and last dates of sampling, and stream ichthyoplankton sample sizes (N).

| Subestuary | Year | Number of Sites | 1st Sampling Date | Last Sampling Date | Number of Dates | N   |
|------------|------|-----------------|-------------------|--------------------|-----------------|-----|
| Bush       | 2005 | 13              | 18-Mar            | 15-May             | 16              | 99  |
| Bush       | 2006 | 13              | 18-Mar            | 15-May             | 20              | 114 |
| Bush       | 2007 | 14              | 21-Mar            | 13-May             | 17              | 83  |
| Bush       | 2008 | 12              | 22-Mar            | 26-Apr             | 17              | 77  |
| Bush       | 2014 | 6               | 22-Mar            | 1-Jun              | 10              | 60  |
| Deer       | 2012 | 4               | 20-Mar            | 7-May              | 11              | 44  |
| Deer       | 2013 | 5               | 19-Mar            | 23-May             | 19              | 87  |
| Deer       | 2014 | 5               | 2-Apr             | 28-May             | 12              | 60  |
| Deer       | 2015 | 5               | 23-Mar            | 26-May             | 15              | 75  |
| Mattawoman | 2008 | 9               | 8-Mar             | 9-May              | 10              | 90  |
| Mattawoman | 2009 | 9               | 8-Mar             | 11-May             | 10              | 70  |
| Mattawoman | 2010 | 7               | 7-Mar             | 15-May             | 11              | 75  |
| Mattawoman | 2011 | 7               | 5-Mar             | 15-May             | 14              | 73  |
| Mattawoman | 2012 | 7               | 4-Mar             | 13-May             | 11              | 75  |
| Mattawoman | 2013 | 7               | 10-Mar            | 25-May             | 12              | 80  |
| Mattawoman | 2014 | 8               | 9-Mar             | 25-May             | 12              | 87  |
| Mattawoman | 2015 | 7               | 15-Mar            | 24-May             | 11              | 60  |
| Piscataway | 2008 | 5               | 17-Mar            | 4-May              | 8               | 39  |
| Piscataway | 2009 | 6               | 9-Mar             | 14-May             | 11              | 60  |
| Piscataway | 2012 | 5               | 5-Mar             | 16-May             | 11              | 55  |
| Piscataway | 2013 | 5               | 11-Mar            | 28-May             | 11              | 55  |
| Piscataway | 2014 | 5               | 10-Mar            | 1-Jun              | 9               | 45  |

Table 1-2. Summary of historical conductivity sampling in non-tidal Mattawoman Creek. RKM = site location in river kilometers from the mouth; Months = months when samples were drawn; Sum = sum of samples for all years.

| RKM  | Months  | Sum | Years Sampled   |
|------|---------|-----|-----------------|
| 12.4 | 1 to 12 | 218 | 1971, 1974-1989 |
| 18.1 | 4 to 9  | 8   | 1974            |
| 27   | 4 to 9  | 9   | 1970, 1974      |
| 30   | 8 and 9 | 2   | 1970            |
| 34.9 | 4 to 9  | 9   | 1970, 1974      |
| 38.8 | 8 and 9 | 2   | 1970            |

Table 1-3. Summary statistics of conductivity ( $\mu\text{S}/\text{cm}$ ) for mainstem stations in Mattawoman, Piscataway, and Deer Creeks, and Bush River during 2005-2014. Unnamed tributaries were excluded from analysis. Tinkers Creek was included with mainstem stations in Piscataway Creek.

| Conductivity   | Year |      |      |       |       |       |       |       |       |       |       |
|----------------|------|------|------|-------|-------|-------|-------|-------|-------|-------|-------|
|                | 2005 | 2006 | 2007 | 2008  | 2009  | 2010  | 2011  | 2012  | 2013  | 2014  | 2015  |
| Bush           |      |      |      |       |       |       |       |       |       |       |       |
| Mean           | 269  | 206  | 263  | 237   |       |       |       |       |       | 276.7 |       |
| Standard Error | 25   | 5    | 16   | 6     |       |       |       |       |       | 15    |       |
| Median         | 230  | 208  | 219  | 234   |       |       |       |       |       | 253.4 |       |
| Kurtosis       | 38   | 2    | 22   | 7     |       |       |       |       |       | 3.16  |       |
| Skewness       | 6    | -1   | 4    | 0     |       |       |       |       |       | 1.56  |       |
| Range          | 1861 | 321  | 1083 | 425   |       |       |       |       |       | 606   |       |
| Minimum        | 79   | 0    | 105  | 10    |       |       |       |       |       | 107   |       |
| Maximum        | 1940 | 321  | 1187 | 435   |       |       |       |       |       | 713   |       |
| Count          | 81   | 106  | 79   | 77    |       |       |       |       |       | 60    |       |
| Deer           |      |      |      |       |       |       |       |       |       |       |       |
| Mean           |      |      |      |       |       |       |       | 174.9 | 175.6 | 170.3 | 191.8 |
| Standard Error |      |      |      |       |       |       |       | 1.02  | 1.5   | 1.4   | 0.9   |
| Median         |      |      |      |       |       |       |       | 176.8 | 177.7 | 171.7 | 193.5 |
| Kurtosis       |      |      |      |       |       |       |       | 17.22 | 13.88 | 9.21  | 7.43  |
| Skewness       |      |      |      |       |       |       |       | -3.78 | -2.25 | -2.42 | -1.97 |
| Range          |      |      |      |       |       |       |       | 39.3  | 122   | 66    | 51    |
| Minimum        |      |      |      |       |       |       |       | 140.2 | 93    | 116   | 156   |
| Maximum        |      |      |      |       |       |       |       | 179.5 | 215   | 183   | 207   |
| Count          |      |      |      |       |       |       |       | 44    | 87    | 60    | 75    |
| Mattawoman     |      |      |      |       |       |       |       |       |       |       |       |
| Mean           |      |      |      | 120.1 | 244.5 | 153.7 | 147.5 | 128.9 | 126.1 | 179.4 | 181.8 |
| Standard Error |      |      |      | 3.8   | 19.2  | 38    | 2.8   | 1.9   | 2.4   | 9.1   | 6.5   |
| Median         |      |      |      | 124.6 | 211   | 152.3 | 147.3 | 130.9 | 126.5 | 165.8 | 172.5 |
| Kurtosis       |      |      |      | 2.1   | 1.41  | 1.3   | 8.29  | -0.26 | 5.01  | 0.33  | 1.49  |
| Skewness       |      |      |      | -1.41 | 1.37  | 0.03  | 1.72  | -0.67 | -1.70 | 1.00  | 1.33  |
| Range          |      |      |      | 102   | 495   | 111   | 117   | 49    | 96    | 261   | 185   |
| Minimum        |      |      |      | 47    | 115   | 99    | 109   | 102   | 63    | 88    | 130   |
| Maximum        |      |      |      | 148   | 610   | 210   | 225   | 151   | 158   | 350   | 315   |
| Count          |      |      |      | 39    | 40    | 43    | 44    | 44    | 48    | 48    | 44    |
| Piscataway     |      |      |      |       |       |       |       |       |       |       |       |
| Mean           |      |      |      | 218.4 | 305.4 |       |       | 211.4 | 245   | 249.4 |       |
| Standard Error |      |      |      | 7.4   | 19.4  |       |       | 5.9   | 6.9   | 11.1  |       |
| Median         |      |      |      | 210.4 | 260.6 |       |       | 195.1 | 238.4 | 230   |       |
| Kurtosis       |      |      |      | -0.38 | 1.85  |       |       | 0.11  | -0.29 | 2.56  |       |
| Skewness       |      |      |      | 0.75  | 1.32  |       |       | 0.92  | 0.73  | 1.50  |       |
| Range          |      |      |      | 138   | 641   |       |       | 163   | 173   | 274   |       |
| Minimum        |      |      |      | 163   | 97    |       |       | 145   | 181   | 174   |       |
| Maximum        |      |      |      | 301   | 737   |       |       | 308   | 354   | 449   |       |
| Count          |      |      |      | 29    | 50    |       |       | 44    | 44    | 36    |       |

Table 1-4. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Mattawoman Creek during 1971, 1989-1991, and 2008-2015. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-2.

| Station      | Year |      |      |      |      |      |      |      |      |      |      |      |
|--------------|------|------|------|------|------|------|------|------|------|------|------|------|
|              | 1971 | 1989 | 1990 | 1991 | 2008 | 2009 | 2010 | 2011 | 2012 | 2013 | 2014 | 2015 |
| Herring      |      |      |      |      |      |      |      |      |      |      |      |      |
| MC1          | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    |
| MC2          | 1    | 1    | 1    | 1    | 0    | 0    | 1    | 1    | 1    | 1    | 1    | 1    |
| MC3          | 1    |      |      | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    | 1    |
| MC4          | 1    |      |      | 1    | 0    | 0    | 1    | 1    | 1    | 1    | 1    | 1    |
| MUT3         | 1    |      |      |      | 0    | 0    | 0    | 1    | 1    | 1    | 1    | 1    |
| MUT4         |      |      |      |      |      |      | 0    | 0    | 1    | 0    | 0    | 0    |
| MUT5         | 1    |      |      |      | 1    | 0    | 0    | 0    | 0    | 0    | 1    | 0    |
| White Perch  |      |      |      |      |      |      |      |      |      |      |      |      |
| MC1          | 1    | 1    | 1    | 1    | 1    | 0    | 1    | 0    | 0    | 1    | 1    | 1    |
| MC2          | 0    | 0    | 1    | 0    | 0    | 0    | 0    | 0    | 0    | 1    | 1    | 0    |
| MC3          | 1    |      |      | 0    | 0    | 0    | 0    | 0    | 0    | 0    | 0    | 0    |
| Yellow Perch |      |      |      |      |      |      |      |      |      |      |      |      |
| MC1          | 1    | 1    | 1    | 1    | 1    | 0    | 1    | 1    | 0    | 1    | 1    | 1    |

Table 1-5. Site-specific presence-absence of Herring (Blueback Herring, Hickory and American Shad, and Alewife) and White Perch spawning in Piscataway Creek during 1971, 2008-2009, and 2012-2014. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-3.

| Station     | Year |      |      |      |      |      |
|-------------|------|------|------|------|------|------|
|             | 1971 | 2008 | 2009 | 2012 | 2013 | 2014 |
| Herring     |      |      |      |      |      |      |
| PC1         | 1    | 0    | 0    | 1    | 1    | 1    |
| PC2         | 1    | 0    | 1    | 1    | 1    | 1    |
| PC3         | 1    | 0    | 0    | 1    | 1    | 1    |
| PTC1        | 1    | 0    | 0    | 1    | 1    | 0    |
| PUT4        | 1    |      | 0    | 0    | 0    | 0    |
| White Perch |      |      |      |      |      |      |
| PC1         | 1    | 0    | 0    | 0    | 0    | 1    |
| PC2         | 1    | 0    | 0    | 0    | 0    | 0    |

Table 1-6. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch spawning in Bush River streams during 1973, 2005-2008, and 2014. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-4.

| Station      | Year |      |      |      |      |      |
|--------------|------|------|------|------|------|------|
|              | 1973 | 2005 | 2006 | 2007 | 2008 | 2014 |
| Herring      |      |      |      |      |      |      |
| BBR1         | 0    | 1    | 1    | 1    | 1    | 1    |
| BCR1         | 1    | 0    | 0    | 1    | 0    | 1    |
| BHH1         | 0    | 0    | 1    | 1    | 1    | 1    |
| BJR1         | 0    | 1    | 1    | 1    | 0    | 1    |
| BOP1         | 1    | 1    | 1    | 1    | 1    | 1    |
| BWR1         | 1    | 0    | 0    | 1    | 0    | 1    |
| White Perch  |      |      |      |      |      |      |
| BBR1         | 1    | 0    | 0    | 0    | 0    | 1    |
| BCR1         | 1    | 0    | 0    | 0    | 0    | 1    |
| BHH1         | 0    | 0    | 0    | 0    | 0    | 0    |
| BJR1         | 0    | 0    | 0    | 0    | 0    | 0    |
| BOP1         | 1    | 0    | 0    | 1    | 0    | 1    |
| BWR1         | 1    | 0    | 0    | 0    | 0    | 0    |
| Yellow Perch |      |      |      |      |      |      |
| BBR1         | 1    | 0    | 0    | 0    | 0    | 0    |
| BCR1         | 0    | 0    | 0    | 0    | 0    | 1    |
| BHH1         | 0    | 0    | 0    | 0    | 0    | 1    |
| BJR1         | 1    | 0    | 0    | 0    | 0    | 1    |
| BOP1         | 0    | 0    | 0    | 0    | 0    | 0    |
| BWR1         | 1    | 0    | 1    | 0    | 0    | 0    |

Table 1-7. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Deer Creek during 1972 and 2012-2015. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-5.

| Station      | Year |      |      |      |      |
|--------------|------|------|------|------|------|
|              | 1972 | 2012 | 2013 | 2014 | 2015 |
| Herring      |      |      |      |      |      |
| SU01         | 1    | 1    | 1    | 1    | 1    |
| SU02         |      | 1    | 1    | 1    | 1    |
| SU03         |      | 1    | 1    | 1    | 1    |
| SU04         | 1    | 1    | 1    | 1    | 1    |
| SU05         | 0    |      | 1    | 1    | 1    |
| White Perch  |      |      |      |      |      |
| SU01         | 1    | 0    | 1    | 1    | 1    |
| SU02         |      | 0    | 1    | 0    | 1    |
| SU03         |      | 0    | 0    | 1    | 0    |
| SU04         | 0    | 0    | 1    | 1    | 0    |
| SU05         | 0    |      | 0    | 0    | 0    |
| Yellow Perch |      |      |      |      |      |
| SU01         | 1    | 1    | 0    | 1    | 0    |
| SU02         |      | 1    | 0    | 1    | 0    |
| SU03         |      | 0    | 0    | 1    | 0    |
| SU04         | 0    | 0    | 0    | 0    | 0    |
| SU05         | 0    |      | 0    | 0    | 0    |

Table 1-8. Summary of best regression models for standardized conductivity (annual median/province background) versus development level (C/ha), proportion of samples with herring eggs or larvae ( $P_{herr}$ ) versus C/ha, and  $P_{herr}$  versus standardized conductivity.

| Linear Model   |          | Standardized conductivity = Structure density (C/ha) |         |         |                |           |
|----------------|----------|--|---------|---------|----------------|-----------|
| ANOVA          | df       | SS   | MS      | F       | Significance F |           |
| Regression     | 1        | 1.26482  | 1.26482 | 16.07   | 0.0007         |           |
| Residual       | 20       | 1.57438  | 0.07872 |         |                |           |
| Total          | 21       | 2.8392   |         |         |                |           |
| $r^2 = 0.4455$ |          |  |         |         |                |           |
|                | Estimate | SE   | t Stat  | P-value | Lower 95%      | Upper 95% |
| Intercept      | 0.99925  | 0.15015  | 6.66    | <.0001  | 0.68604        | 1.31245   |
| C / ha         | 0.53714  | 0.134  | 4.01    | 0.0007  | 0.25762        | 0.81667   |

| Linear Model   |          | Proportion of samples with herring eggs or larvae ( $P_{herr}$ ) = Structure density (C/ha) |         |         |                |           |
|----------------|----------|---|---------|---------|----------------|-----------|
| ANOVA          | df       | SS  | MS      | F       | Significance F |           |
| Regression     | 1        | 0.83983   | 0.83983 | 24.58   | <.0001         |           |
| Residual       | 21       | 0.71751   | 0.03416 |         |                |           |
| Total          | 22       | 1.55728   |         |         |                |           |
| $r^2 = 0.5393$ |          |   |         |         |                |           |
|                | Estimate | SE  | t Stat  | P-value | Lower 95%      | Upper 95% |
| Intercept      | 0.81050  | 0.09392   | 8.63    | <.0001  | 0.61512        | 1.00581   |
| C / ha         | 0.42316  | 0.08535   | -4.96   | <.0001  | -0.60065       | -0.24567  |

| Nonlinear Model            |          | Proportion of samples with herring eggs or larvae ( $P_{herr}$ ) = Standardized Conductivity |           |           |        |
|----------------------------|----------|--|-----------|-----------|--------|
| Source                     | df       | SS   | MS        | F         | P      |
| Model                      | 2        | 3.5208   | 1.7604    | 35.75     | <.0001 |
| Error Uncorrected          | 20       | 0.985  | 0.0492    |           |        |
| Total                      | 22       | 4.5058   |           |           |        |
| Approximate $r^2 = 0.3268$ |          |  |           |           |        |
| Parameter                  | Estimate | Approximate SE   | Lower 95% | Upper 95% |        |
| a                          | 0.753    | 0.1763   | 0.3852    | 1.1208    |        |
| b                          | -1.8815  | 0.7391   | -3.4233   | -0.3397   |        |

Figure 1-1. Watersheds sampled for stream spawning anadromous fish eggs and larvae during 2005-2015. Coastal Plain and Piedmont Regions are indicated.

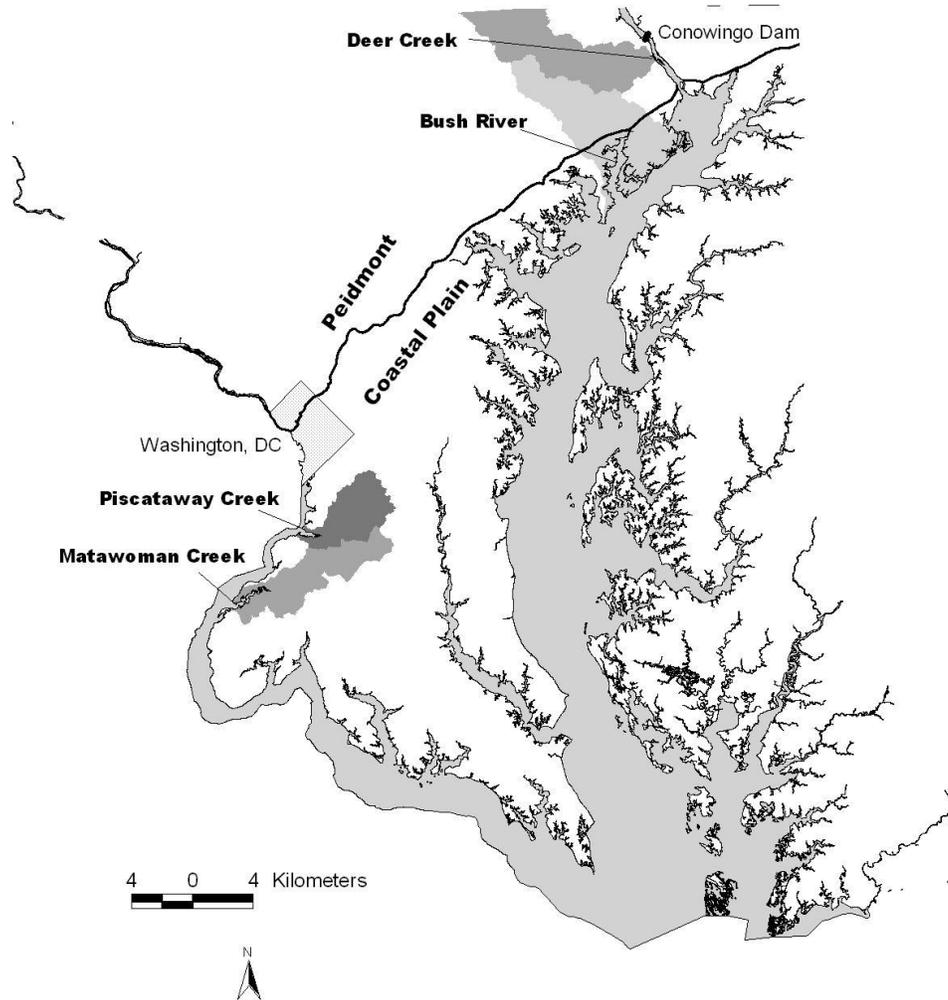


Figure 1-2. Mattawoman Creek's 1971 and 2008-2015 sampling stations. Bar approximates lower limit of development associated with the town of Waldorf.

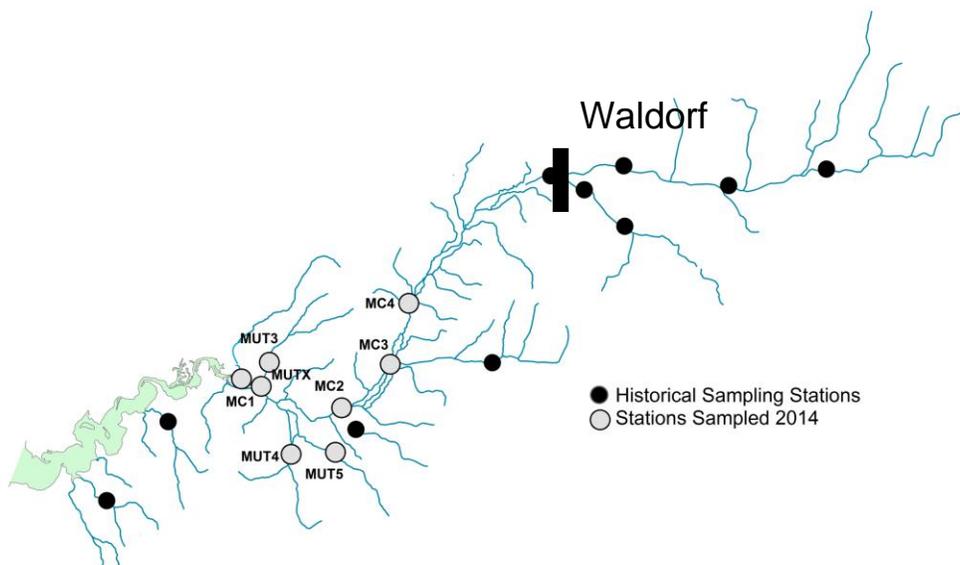


Figure 1-3. Piscataway Creek's 1971, 2008-2009, and 2012-2014 sampling stations.

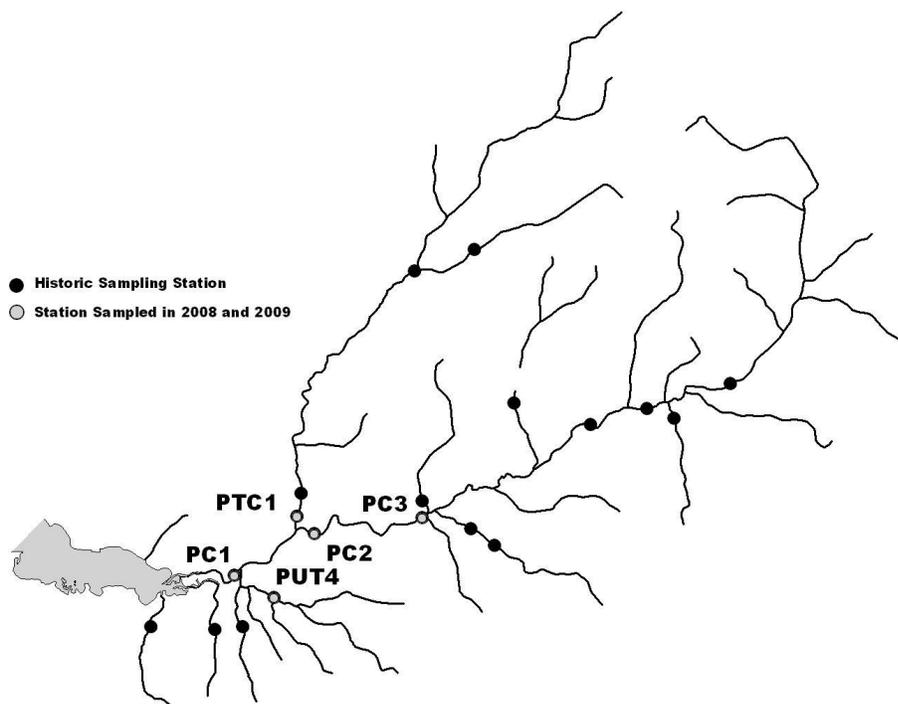


Figure 1-4. Bush River's 1973, 2005-2008, and 2014 sampling stations. Stations in Aberdeen Proving Grounds (APG) have been separated from other Bush River stations. Line delineates APG streams that were excluded.

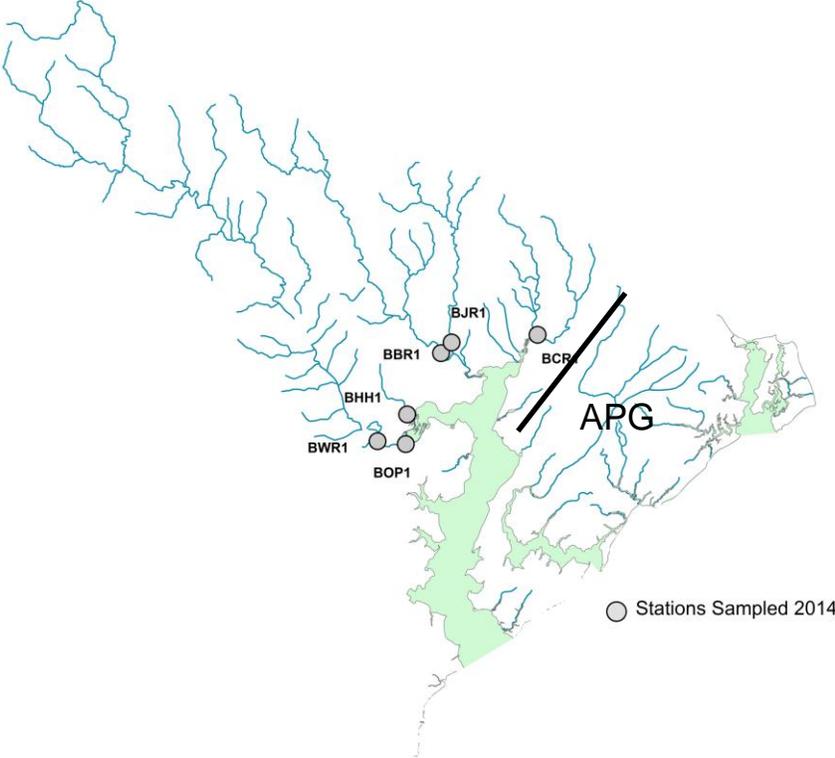


Figure 1-5. Deer Creek's 1972 and 2012-2015 sampling stations.

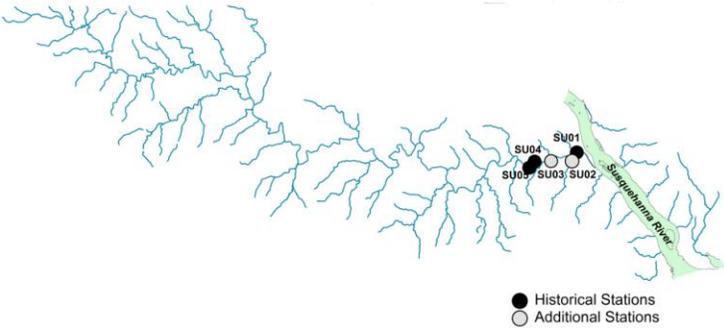


Figure 1-7. Stream conductivity measurements ( $\mu\text{S} / \text{cm}$ ), by station and date, in Mattawoman Creek during (A) 2009, (B) 2010, (C) 2011, (D) 2012, (E) 2013, (F) 2014, and (G) 2015. Lines indicate conductivity range measured at mainstem sites (MC1 – MC4) during 1991 by Hall et al. (1992).

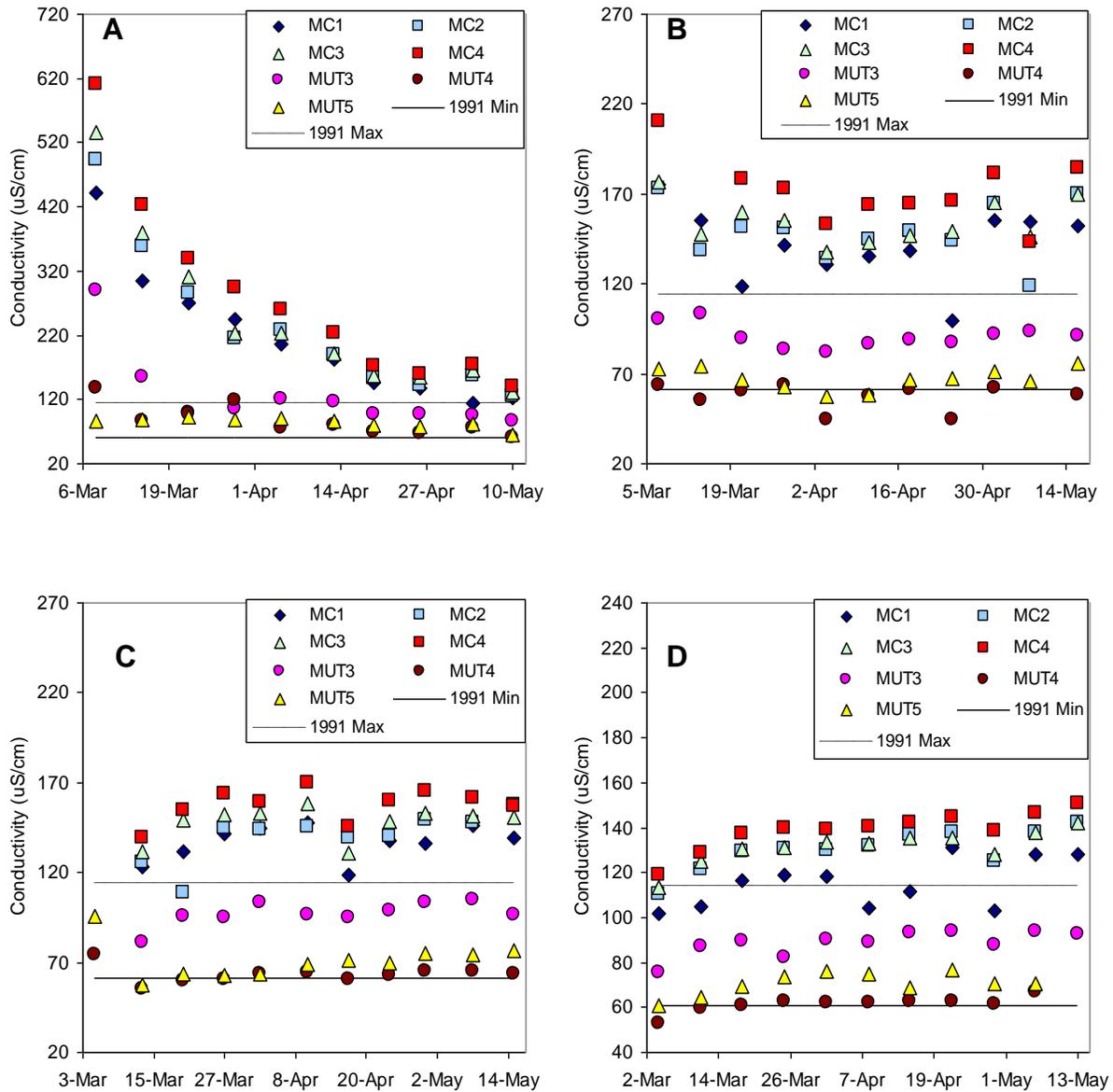


Figure 1-7 cont.

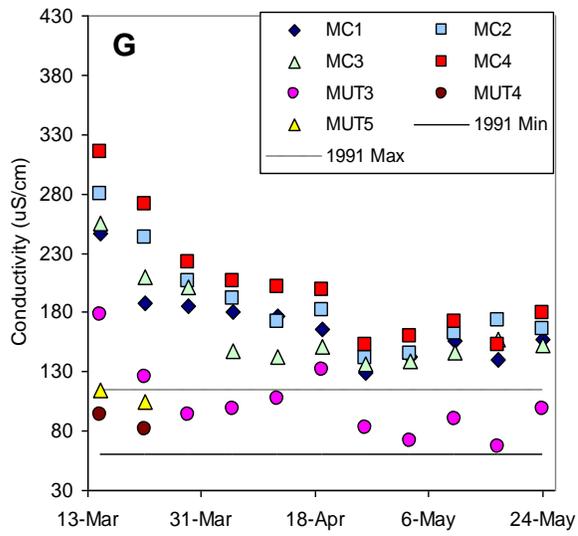
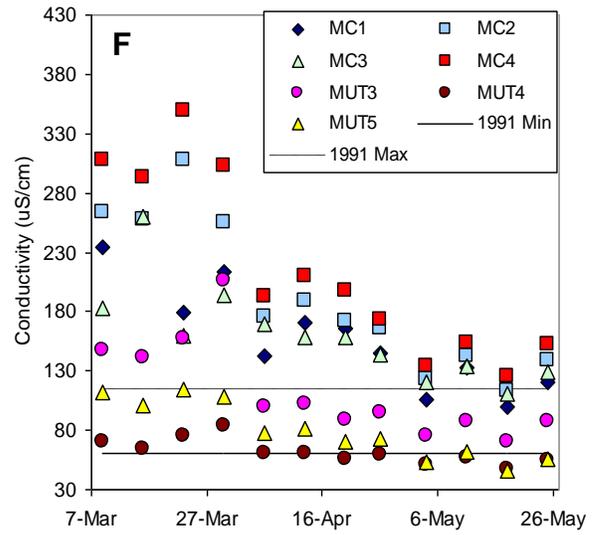
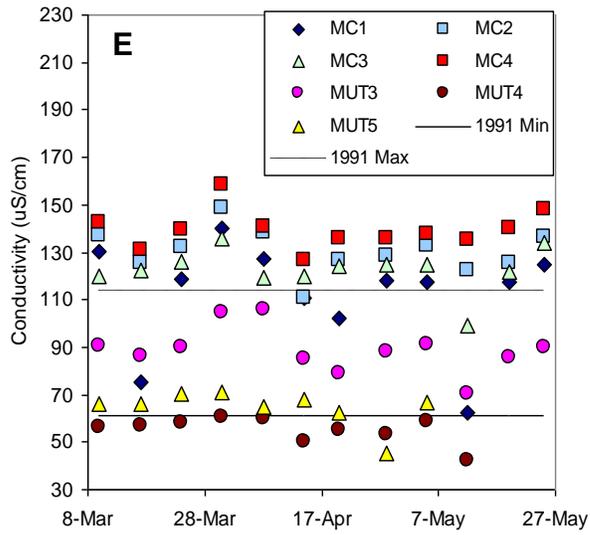


Figure 1-8. Historical (1970-1989) median conductivity measurements and current (2008-2015) anadromous spawning survey median conductivity in non-tidal Mattawoman Creek (between the junction with the subestuary and Waldorf) plotted against distance from the mouth. The two stations furthest upstream are nearest Waldorf. Median conductivity was measured during March-May, 2008-2015, and varying time periods (see Table 1-2) during 1970-1989.

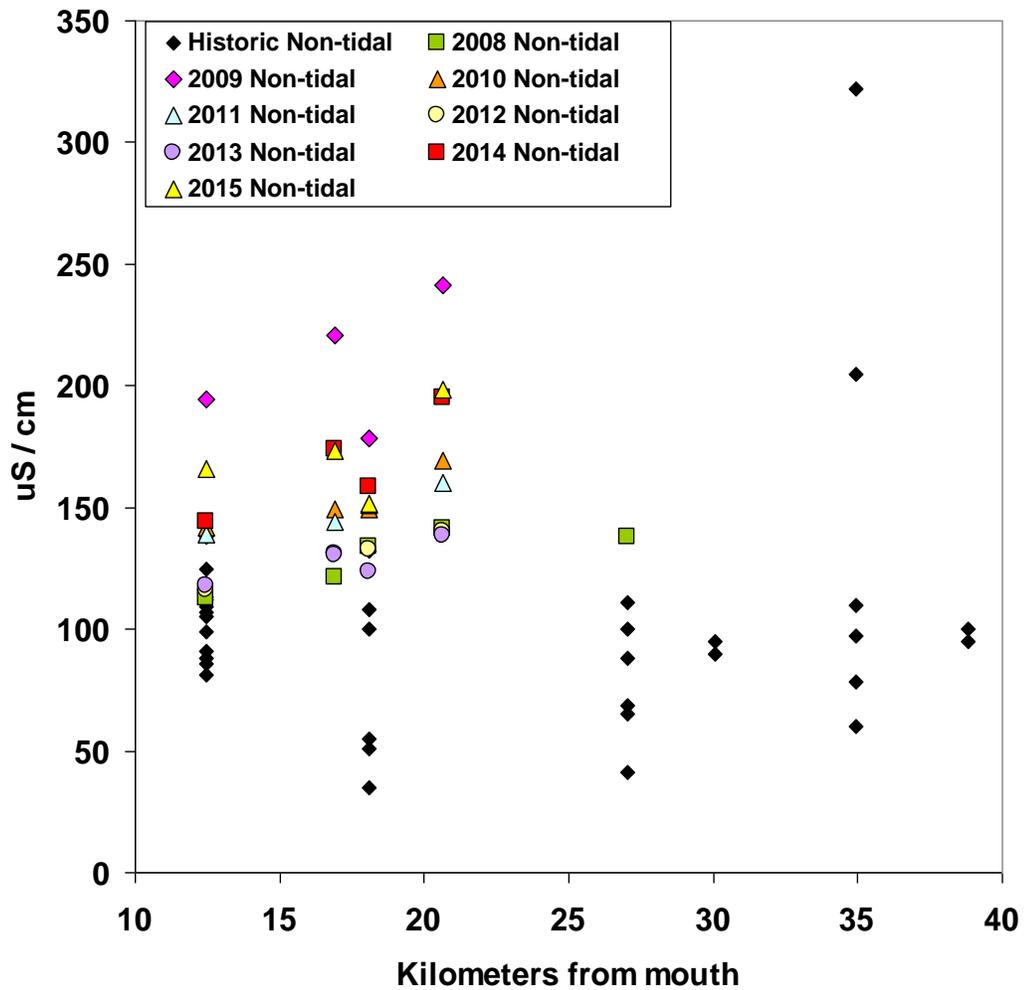


Figure 1-9. Proportion of samples ( $P_{herr}$ ) with Herring and its 90% confidence interval for stream ichthyoplankton surveys in Mattawoman, Piscataway and Deer Creeks, and Bush River.

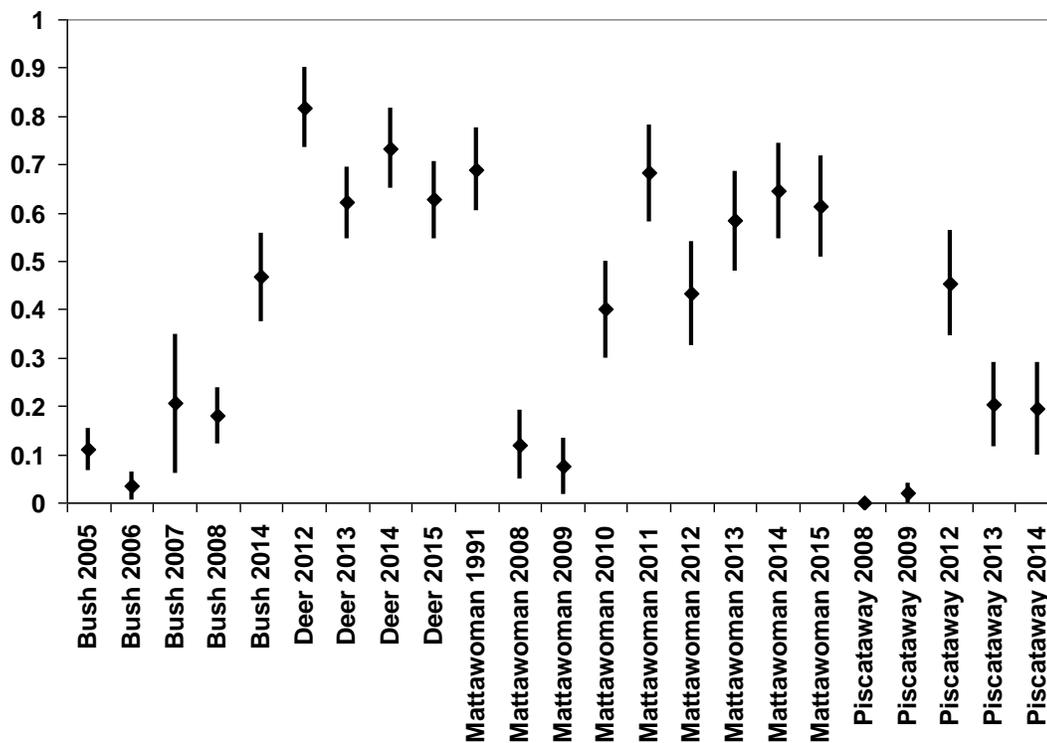


Figure 1-10. Mattawoman data pooled across years to form estimates of proportions of samples with White Perch (WP) eggs and-or larvae (sites MC1 and MC2) or Yellow Perch (YP) eggs and-or larvae (MC1) for 1989-1991 collections compared to 2008-2010, 2011-2013, and 2014-2015 collections at the same combination of sites.

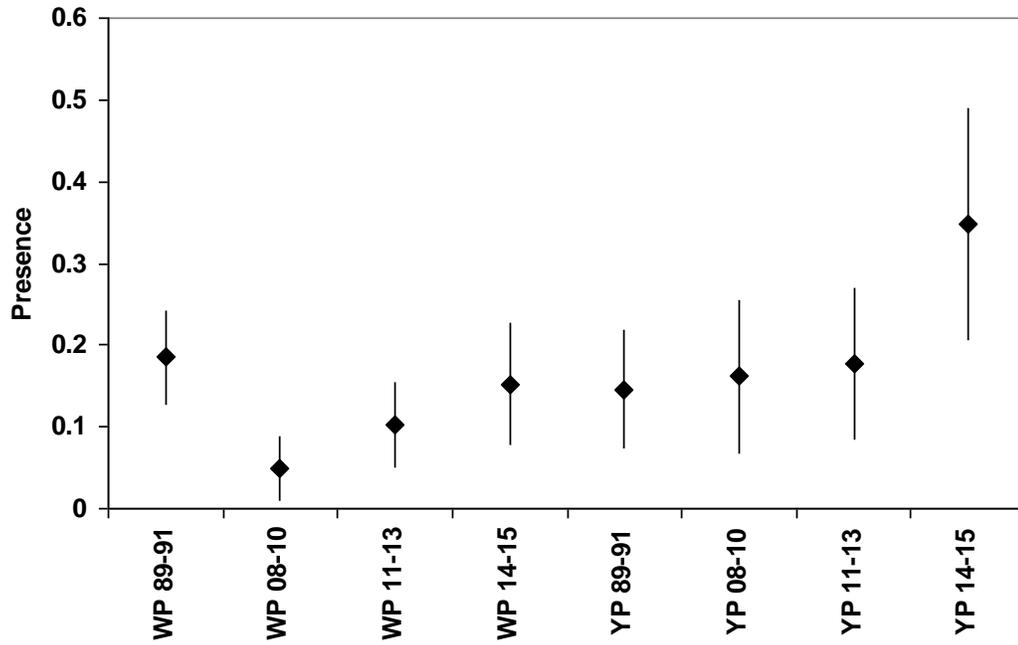


Figure 1-11. Standardized median conductivity during spring spawning surveys and level of development (C / ha). Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).

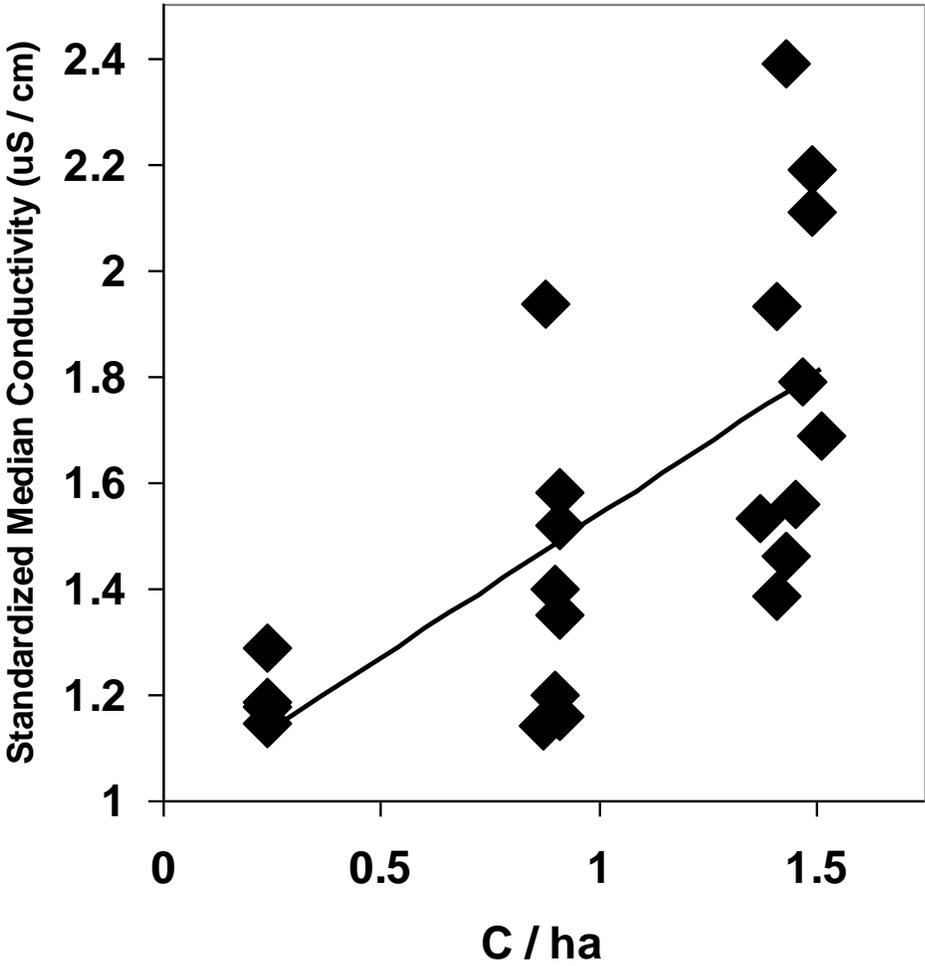
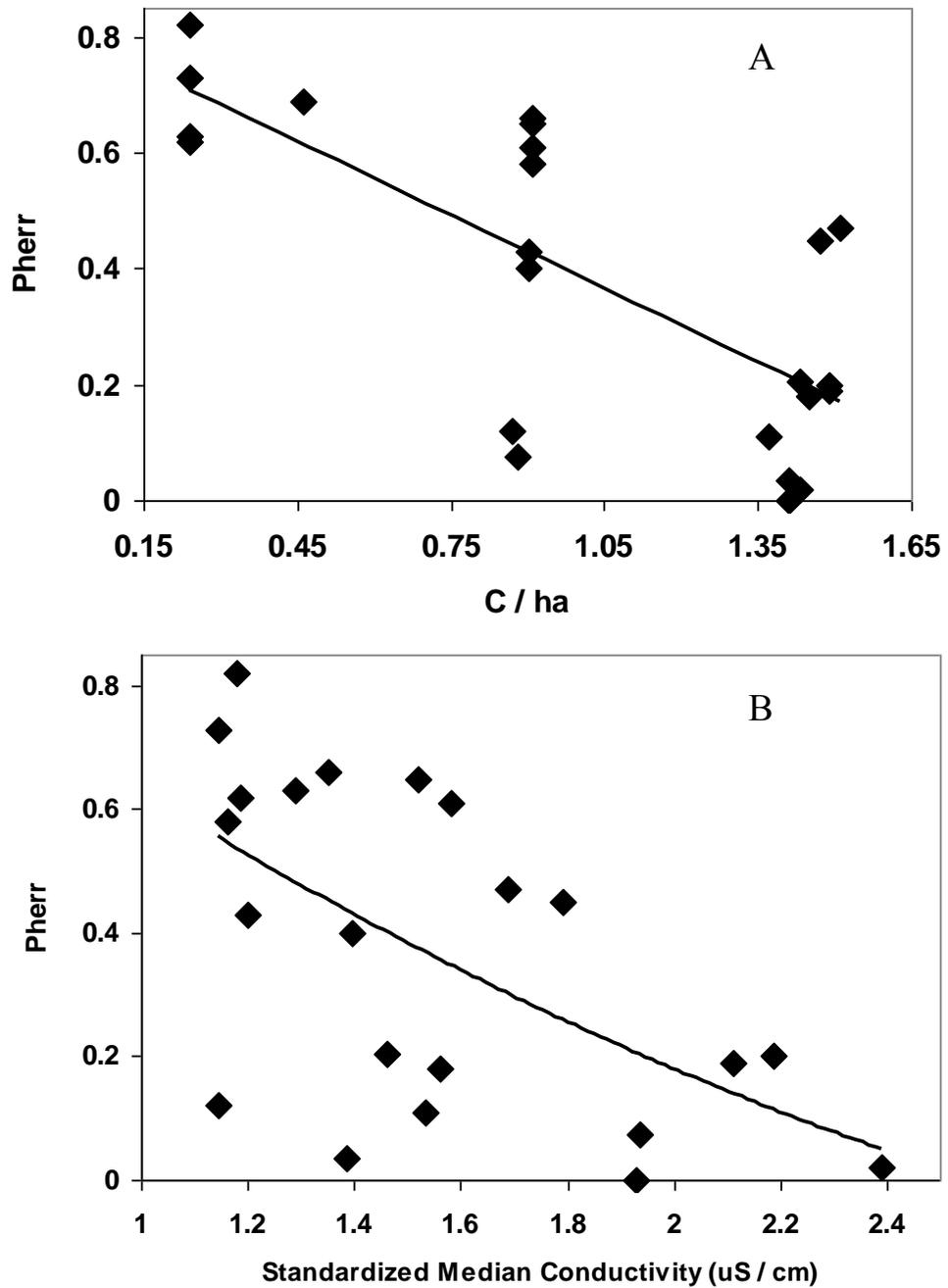


Figure 1-12. (A) Proportion of stream samples with Herring eggs or larvae ( $P_{herr}$ ) and level of development ( $C / ha$ ). (B)  $P_{herr}$  and standardized median spawning survey conductivity ( $\mu S / cm$ ). Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).



## Section 2: Estuarine Yellow Perch Larval Presence-Absence Sampling

Carrie Hoover, Alexis Park, Jim Uphoff, Margaret McGinty, Patrick Donovan, and Charles Johnson

### Introduction

Presence-absence sampling for Yellow Perch larvae in 2015 was conducted in the upper tidal reaches of the Choptank, Nanticoke, and Patuxent Rivers, and in Mattawoman Creek during the month of April and through the first week of May (Figure 2-1). Annual  $L_p$ , the proportion of tows with Yellow Perch larvae during a standard time period and where larvae would be expected, provides a cost-effective measure of the product of egg production and survival through the early post-larval stage. In 2015 we used regression and correlation analyses to examine associations and relationships of structures per hectare (C/ha; an indicator of development; Topolski 2015),  $L_p$ , feeding success, diet composition, and relative detritus levels collected during spring surveys, along with Maryland Department of Planning estimates of wetland percentages for each watershed.

We examined a hypothesis that watershed development and wetland coverage impacted related organic matter (OM) dynamics, altering zooplankton production important for Yellow Perch larval feeding success and survival (the OM hypothesis). Urbanization was expected to negatively impact Yellow Perch larval feeding success because it affects quality and quantity of OM in streams (Paul and Meyer 2001) and was negatively associated with extent of wetlands in many subestuary watersheds evaluated by Uphoff et al. (2011). Riparian zones and floodplains that are sources of OM become disconnected from stream channels by stormwater management in suburban and urban watersheds (Craig et al. 2008; Kaushal et al. 2008; Elmore and Kaushal 2008; Brush 2009; NRC 2009), altering quantity and transport of OM (Paul and Meyer 2001; McClain et al. 2003; Stanley et al. 2012). We used the empirical-statistical approach recommended by Austin and Ingham (1978) and Crecco and Savoy (1984) for resolving the effects of environment on fish recruitment. This approach offers a working hypothesis that is tested for validity with empirical data and a thorough statistical analysis. Shortage of appropriate food has been frequently hypothesized to cause high mortality of fish larvae (Martin et al. 1985; Miller et al. 1988; Heath 1992). Years of high spring discharge favor anadromous fish recruitment in Chesapeake Bay (Hoffman et al. 2007; Martino and Houde 2010) and may represent episodes of hydrologic transport of accumulated OM from watersheds that fuel zooplankton production and feeding success (McClain et al. 2003). Under natural conditions, riparian marshes and forests would provide OM subsidies in high discharge years (Hoffman et al. 2007), while phytoplankton would be the primary source of OM in years of lesser flow.

During 2012-2015, Yellow Perch were collected for analysis of the ratio of ribonucleic acid (RNA) concentration to deoxyribonucleic acid (DNA) concentration in body tissue (RNA/DNA ratio) to further explore the OM hypothesis. Samples were gathered from Choptank River and Patuxent River during 2015, two subestuaries of Chesapeake Bay with watersheds exhibiting rural (C/ha = 0.16) and suburban levels of development (C/ha = 1.21), respectively. We expected RNA/DNA ratios to decline with increased development. The quantity of DNA within a cell is constant within a species while the quantity of RNA varies with protein synthesis (Tardiff et al. 2005). Since

growth is a function of protein synthesis, RNA/DNA ratios provide a sensitive indicator of recent growth at any given time (Buckley 1984). This ratio is a useful indicator of nutritional status and somatic growth in larval fish (Buckley 1984) that provides a method for examining connections of feeding success and larval condition (Buckley 1984; Martin et al. 1985; Wright and Martin 1985; Clemmesen 1994; Blom et al. 1997) without requiring extensive sampling and sample processing needed to measure mortality directly. Tardif et al. (2005) used RNA/DNA ratios of Yellow Perch larvae and juveniles to determine differences in productivity of managed and natural wetlands of Lake St. Pierre, Canada.

## Methods

Conical plankton nets were towed from boats in upper portions of subestuaries to collect Yellow Perch larvae. Nets were 0.5-m in diameter, 1.0-m long, and constructed of 0.5 mm mesh. Nets were towed with the current for two minutes at a speed that maintained the net near the surface (approximately 2.8 km per hour). Temperature, dissolved oxygen, conductivity, and salinity were measured at each site on each sample date.

Ten sites were sampled twice weekly in the Choptank, Patuxent, and Nanticoke rivers, and weekly in Mattawoman Creek (Figure 2-1). Boundaries of areas sampled were determined from Yellow Perch larval presence in estuarine surveys conducted during the 1970s and 1980s (O'Dell 1987). Larval sampling usually occurs during late March through mid-to-late April, but due to late winter conditions in 2015, sampling occurred during April (continuing through the first week of May in Mattawoman Creek).

Each sample was emptied into a glass jar and checked for larvae. Yellow Perch larvae can be readily identified in the field since they are larger and more developed than Striped Bass and White Perch larvae with which they could be confused (Lippson and Moran 1974). Contents of the jar were allowed to settle and then the amount of settled OM was assigned a rank: 0 = a defined layer was absent; 1 = defined layer on bottom; 2 = more than defined layer and up to ¼ full; 3 = more than ¼ to ½ and; 4 = more than ½ full. If a jar contained enough OM to obscure seeing larvae, it was emptied into a pan with a dark background and observed through a 5X magnifying lens. Organic matter was moved with a probe or forceps to free larvae for observation. If OM loads, wave action, or collector uncertainty prevented positive identification, samples were preserved and taken back to the lab for sorting.

Choptank and Patuxent Rivers were sampled by program personnel in 2015, while Nanticoke River was voluntarily sampled by another Maryland Fisheries Service project during its normal operations without charge to this grant. Mattawoman Creek was sampled by citizen scientist volunteers from the Mattawoman Watershed Society trained by our program biologists.

The proportion of tows with Yellow Perch larvae ( $L_p$ ) for each subestuary was determined annually for dates spanning the first catch through the last date that larvae were consistently present as:

$$^{(1)} L_p = N_{present} / N_{total};$$

where  $N_{present}$  equaled the number of samples with Yellow Perch larvae present and  $N_{total}$  equaled the total number of samples. The SD of  $L_p$  was estimated as:

$$^{(2)} SD = [(L_p \cdot (1 - L_p)) / N_{total}]^{0.5} \text{ (Ott 1977).}$$

The 95% confidence intervals were constructed as:

<sup>(3)</sup>  $L_p + 1.96 \text{ SD}$ ; (Ott 1977).

In general, sampling to determine  $L_p$  began during the last days of March or first days of April and ended after larvae were absent (or nearly so) for two consecutive sampling rounds. In years where larvae disappeared quickly, sampling rounds into the third week of April were included in analysis even if larvae were not collected. Inclusion of these zeros reflected expectation (based on previous years) that larvae would be available to the sampling gear had they been there. This sampling schedule has been maintained for tributaries sampled by program personnel since 2006. Sampling by other Fisheries Service projects and volunteers sometimes did not adhere as strictly to this schedule.

Historical collections in the Choptank and Nanticoke Rivers targeted striped bass eggs and larvae (Uphoff 1997), but Yellow Perch larvae were also common (Uphoff 1991). Uphoff et al. (2005) reviewed presence-absence of Yellow Perch larvae in past Choptank and Nanticoke River collections and found that starting dates during the first week of April or early in the second week were typical and end dates occurred during the last week of April through the first week of May. Larval presence-absence was calculated from data sheets (reflecting lab sorting) for surveys through 1990. During 1998-2004,  $L_p$  in the Choptank River was determined directly in the field and recorded on data sheets (P. Piavis, MD DNR, personal communication). All tows were made for two minutes. Standard 0.5 m diameter nets were used in the Nanticoke River during 1965-1971 (1.0 • 0.5 mm mesh) and after 1998 in the Choptank River (0.5 mm mesh). Trawls with 0.5 m nets (0.5 mm mesh) mounted in the cod-end were used in the Choptank River during 1980-1990 (Uphoff 1997; Uphoff et al. 2005). Survey designs for the Choptank and Nanticoke Rivers were described in Uphoff (1997).

We used property tax map-based counts of structures per hectare (C/ha) in a watershed as our indicator of development (Uphoff et al. 2012; Topolski 2015). Tax maps are graphic representations of individual property boundaries and existing structures that help State tax assessors locate properties (Maryland Department of Planning or MDP 2010; Topolski 2015). All tax data were organized by county. Since watersheds straddle political boundaries, one statewide tax map was created for each year of available tax data, and then subdivided into watersheds. Maryland's tax maps are updated and maintained electronically as part of MDP's Geographic Information System's (GIS) database. Files were managed and geoprocessed in ArcGIS 9.3.1 from Environmental Systems Research Institute (ESRI 2009). All feature datasets, feature classes, and shapefiles were spatially referenced using the NAD\_1983\_StatePlane\_Maryland\_FIPS\_1900 projection to ensure accurate feature overlays and data extraction. ArcGIS geoprocessing models were developed using Model Builder to automate assembly of statewide tax maps, query tax map data, and assemble summary data. Each year's statewide tax map was clipped using the MD 8-digit watershed boundary file, and modified to exclude estuarine waters, to create watershed land tax maps. These watershed tax maps were queried for all parcels having a structure built from 1700 to the tax data year (Topolski 2015). A large portion of parcels did not have any record of year built for structures but consistent undercounts should not present a problem since we are interested in the trend and not absolute magnitude (Uphoff et al. 2012).

Estimates of C/ha were used as a measure of watershed development intensity for analysis with  $L_p$ . Whole watershed estimates were used with the following exceptions: Nanticoke, Choptank, and Patuxent River watersheds were truncated at the lower boundaries of their striped bass spawning areas and at the Delaware border (latter due to lack of comparable data). Estimates of C/ha were available from 1950 through 2013 (M. Topolski, MD DNR, personal communication). Estimates of C/ha for 2013 were used to represent 2014 and 2015 for all systems.

Estimates of C/ha for the IS target and limit were estimated from a power function that converts C/ha to IS based on Towson University satellite data interpretation (Uphoff et al. 2012). The target proposed in Uphoff et al. (2011), 5.5% IS, was reduced to 5% to meet IS guidelines being developed by Maryland's Department of Natural Resources (MD DNR 2012). The IS threshold of 10% in Uphoff et al. (2011) remained unchanged. An estimate equivalent to 15% IS was also made to designate suburban watersheds that were developed well beyond the threshold. Estimates of C/ha that were equivalent to 5% IS, 10% IS, and 15% IS were estimated as 0.27, 0.83, and 1.59 C/ha, respectively by Uphoff et al. (2012).

Uphoff et al. (2012) developed  $L_p$  thresholds for brackish and tidal-fresh systems. Three brackish subestuaries with C/ha > 1.59 (10 estimates from Severn, South, and Magothy Rivers) exhibited chronically depressed  $L_p$  and their maximum  $L_p$  (0.33) was chosen as a threshold indicating serious deterioration of brackish subestuary larval nursery habitat. Similarly, tidal-fresh Piscataway Creek's four estimates of  $L_p$  (2008-2011) consistently ranked low when compared to other tidal-fresh subestuaries sampled (13<sup>th</sup> to 17<sup>th</sup> out of 17 estimates). The maximum for Piscataway Creek's four estimates,  $L_p$  = 0.65, was chosen as a threshold indicating serious deterioration of tidal-fresh larval habitat. Estimates of  $L_p$  would need to be consistently at or below this level to be considered "abnormal" as opposed to occasional depressions (Uphoff et al. 2012).

Two regression approaches were used to examine the relationship between C/ha and  $L_p$ . First, separate linear regressions of C/ha against  $L_p$  were estimated for brackish and tidal-fresh subestuaries. If 95% CIs of slopes overlapped and 95% CIs of the intercepts did not overlap, we used the multiple regression of C/ha and salinity class against  $L_p$ . This latter approach assumed slopes were equal for two subestuary salinity categories, but intercepts were different (Freund and Littell 2006). Salinity was modeled as an indicator variable in the multiple regression with 0 indicating tidal-fresh conditions and 1 indicating brackish conditions. High salinity has been implicated in contributing to low  $L_p$  in Severn River (Uphoff et al. 2005). The association of mean salinity and IS can be significant and strong (Uphoff et al. 2010), and salinity is important to formation of stressful DO conditions in summer in mesohaline tributaries (see Section 3). Ricker (1975) warned against using well correlated variables in multiple regressions, so categorizing salinity for multiple or separate regressions of C/ha against  $L_p$  minimized confounding salinity with level of development. Level of significance was set at  $P < 0.05$ . Residuals were inspected for trends, non-normality, and need for additional terms.

We used Akaike information criteria adjusted for small sample size,  $AIC_c$ , to evaluate the models that describe hypotheses that related changes in  $L_p$  to C/ha for each salinity category (separate slopes) or to C/ha and salinity category (common slopes, separate intercepts; Burnham and Anderson 2001):

$$^{(4)} AIC_c = -2(\log\text{-likelihood}) + 2K + [(2K \cdot (K+1)) / (n-K-1)];$$

where  $n$  is sample size and  $K$  is the number of model parameters. Model parameters for the least squares regressions consisted of their mean square error estimates (variance), intercepts, slopes, and salinity category in the case of the multiple regression. We rescaled  $AIC_c$  values to  $\Delta_i$  ( $AIC_{c_i} - \text{minimum } AIC_c$ ), where  $i$  is an individual model, for the tidal-fresh or brackish regression compared to the multiple regression. The  $\Delta_i$  values provided a quick “strength of evidence” comparison and ranking of models and hypotheses. Values of  $\Delta_i \leq 2$  have substantial support, while those  $> 10$  have essentially no support (Burnham and Anderson 2001).

An additional view of the relationship of  $L_p$  and  $C/ha$  was developed by considering dominant land use classification when interpreting salinity classification (brackish or tidal-fresh),  $C/ha$ , and  $L_p$  regressions. Primary land use (forest, agriculture, or urban) was determined from Maryland Department of Planning estimates for 1973, 1994, 1997, 2002, or 2010 that fell closest to a sampling year. These latter categories were not used in regression analyses, but were considered in the interpretation of results. Urban land consisted of high and low density residential, commercial, and institutional acreages (MD DNR 1999).

Composite samples of larvae were collected for feeding analyses from sites in Mattawoman Creek, and Choptank, Nanticoke, and Patuxent Rivers during several sample trips in 2015. Subsamples of postlarvae 12 mm TL or less were examined for gut contents from each day’s samples of each subestuary, although only larvae 6-9 mm were used for analyses. These 6-9 mm larvae represented first-feeding and early postlarvae that absorbed their yolk and began active feeding (Hardy 1978). Larvae were measured to the nearest 0.5 millimeter. Gut fullness was judged visually and assigned a rank: 0 = empty; 1 = up to  $\frac{1}{4}$  full; 2 = up to  $\frac{1}{2}$  full; 3 = up to  $\frac{3}{4}$  full; and 4 = full. Major food items were classified as copepods, cladocerans, or other, and the presence (coded 1) or absence (coded 0) of each group was noted.

The mean of feeding success rank was calculated annually for each subestuary sampled in 2010-2015, as was mean total length (TL in mm) of larvae. The proportion of guts without food ( $P_0$ ) was estimated for each subestuary as was the proportion of larvae with copepods ( $P_{cope}$ ), cladocerans ( $P_{clad}$ ), or other ( $P_{other}$ ) food items. The latter three proportions were not additive.

We used OM0 (proportion of samples without organic material, i.e., rank = 0) as our indicator of detritus availability. Proportions of samples without OM were only estimated during 2011-2015, so fewer observations were available for analyses. The distribution of OM ranks assigned to samples were highly skewed towards zero, and few ranks greater than one were reported. We regressed OM0 against  $C/ha$ , and associations of OM0 with mean feeding rank,  $P_0$ ,  $P_{cope}$ ,  $P_{clad}$ , and  $P_{other}$  were tested with regression analysis as well. An additional set of regression analyses examined associations among mean feeding success rank and  $P_{cope}$ ,  $P_{clad}$ , and  $P_{other}$ .

We were specifically interested in the relationships of the amount of organic matter to development and larval feeding success. Examination of the plot of OM0 and  $C/ha$  suggested that the relationship could be nonlinear, with OM0 increasing at a decreasing rate with  $C/ha$ . We fit a power and logistic growth function to these data using Proc NLIN in SAS (Freund and Littell 2006). The power function was described by the equation:

$$^{(5)} OM0 = a \cdot (C/ha)^b;$$

where  $a$  is a scaling coefficient and  $b$  is a shape parameter. The logistic growth function was described by the equation:

$$^{(6)}\text{OM0} = b / ((1 + ((b - c) / c) \cdot (\exp(-a \cdot C/\text{ha})));$$

where  $a$  is the growth rate of OM0 with  $C/\text{ha}$ ,  $b$  is maximum OM0, and  $c$  is OM0 at  $C/\text{ha} = 0$  (Prager et al. 1989).

We used linear and quadratic regressions to explore relationships of feeding success (mean of feeding ranks) with OM0 (Freund and Littel 2006). Linear and quadratic regressions explored this relationship for all data, with the linear regression describing a hypothesis about steady change, while the dome-shaped quadratic relationship would indicate an optimum value of OM0 for feeding success.

We were interested in links among OM0, percent wetlands in a watershed, and  $C/\text{ha}$ . Examination of the plot of percent wetlands and  $C/\text{ha}$  suggested that the relationship was nonlinear, with percentage of wetlands decreasing at a decreasing rate with  $C/\text{ha}$ , and appeared to be a mirror image of the plot of OM0 and  $C/\text{ha}$ . Examination of the plot of OM0 and percent wetlands suggested a linear relationship, with proportion of samples without organic material decreasing as percent wetlands per watershed increased. We fit power, logistic growth, or a linear function to these data sets, respectively.

During 2015, we collected Yellow Perch larvae for RNA/DNA analysis from two tributaries of the Chesapeake Bay with watersheds exhibiting different levels of development; the rural, agricultural Choptank River watershed ( $C/\text{ha} = 0.16$ ) and the suburban Patuxent River watershed ( $C/\text{ha} = 1.21$ ). Based on previous years' collections, we anticipated that sampling from these two rivers on three occasions would provide 30 larvae per system, per date, for a minimum of 180. During 2013 and 2014, we had collected Yellow Perch Larvae from Nanjemoy Creek ( $C/\text{ha} = 0.09$ ) and Mattawoman Creek ( $C/\text{ha} = 0.91$ ; Uphoff et al. 2014; 2015) and these data were available for comparisons with 2015.

Samples for RNA/DNA analysis were collected when larvae were gathered for analysis of gut contents. In the field, Yellow Perch larvae were composited from several stations (where possible) that bracketed where larvae are abundant. Once a candidate jar had been checked for larvae and OM, the sample was poured through a  $500 \mu$  screen and larvae were transferred to a large tube with special preservative (RNAlater®). The vial was labeled with the subestuary name and sample date. Larvae from other sites from one subestuary were composited into the vial on the same date.

In the lab, larvae for each date were processed for both gut contents and RNA/DNA ratios. Yellow Perch larvae 12 mm TL or less were examined for gut contents from each sample, although only larvae 6-9 mm in size (sizes in common) were used for analyses. These larvae represented first-feeding and early postlarvae, larvae that absorbed their yolk and began active feeding. Generally, 6 mm larvae were the smallest that contained food. Larvae were removed from the composite sample and placed in a Petri dish of water, examined for gut contents and then the guts were removed. The RNA/DNA ratio estimate did not contain food items. If a larva had not fed, the guts were teased away to be safe. Each processed larva was placed in a small individual vial of RNAlater® preservative. The vial was coded on the outside as follows: letter designating which river, number designating which sample date, and number designating which individual larva was placed in the vial.

RNA/DNA ratios were estimated by science staff at the Cooperative Oxford Laboratory and partners from the University of Maryland Eastern Shore. Protocols for estimating RNA/DNA generally followed Kaplan et al. (2001). Larvae were stored at 4°C in RNAlater® for up to three weeks until processing. Whole body samples, minus gut contents, were digested with 1% sodium dodecylsulfate, proteinase K digestion buffer (66ug/ml), and 0.1M NaCl at 55°C for several hours until completely digested. Samples were centrifuged at 11,000 rpm for 10 minutes, and the supernatant containing the nucleic acids was removed and stored at -80°C until ready for processing.

A 400 µL portion of the supernatant was removed for digestion of DNA prior to analysis of RNA. Removal of DNA was accomplished by treating this portion of supernatant with DNase digestion buffer (0.2M Tris-HCl pH=7.5, 0.1M MgCl and 0.02M CaCl, and 10 U RNase-free DNase I). Samples incubated at 37 °C for 45 minutes in a dry bath. Samples were centrifuged for five minutes at 8,000 rpm. The supernatant was removed and stored at -80 °C until ready for processing.

Samples were analyzed for DNA and RNA using Quant-it™ PicoGreen® and Quant-it™ RiboGreen® (Molecular Probes, Oregon), respectively, according to the manufacturer's protocol. Samples were plated in triplicate on solid black 96-well microplates and fluorescence was measured at 480 nm excitation and 520 nm.

During sample processing in 2014 it was discovered that a dilution had been missed in the instructions used to estimate ratios for 2013 and that year. Samples had been retained for both years and it was hoped these could be used to develop adjustments for the missed dilution. In 2014, to quantify nucleic acids, sample fluorescent readings were compared to DNA and RNA standard curves. These curves were developed by creating eight separate solutions of tissue digestion buffer and nucleic acid standard solutions. Lambda phage DNA and E. coli ribosomal 16S and 23S RNA (Molecular Probes, Oregon) were used as DNA and RNA standards, respectively. Serial dilutions of the 16 standard solutions (eight solutions per nucleic acid) were plated on 96-well microplates followed by the addition of PicoGreen® for DNA and RiboGreen® for RNA. Fluorescence was read at 480 nm excitation and 520 nm. The natural log-transformed fluorescent measures from each standard solution (F) were plotted against their respective nucleic acid concentration (C). Polynomial linear regression was used to determine the coefficients (Table 2-1) for each curve. The regression model used was

$${}^{(7)} \text{Log}_e F = (a \cdot C) + (b \cdot C^2) + d;$$

where F and C are as defined previously, a and b are coefficients and d is the intercept. These coefficients were used to determine sample concentrations of DNA and RNA after back-transformation. This same process was supposed to be followed, and adjustments made, for 2013 samples as well. The freezer they were being stored in failed, however, and samples were lost. An attempt to make standards using the original erroneous methodology was undertaken, and correction curves were developed and used to create new “outputs” for samples that contained fluorescence in 2013. This data set was judged to be unreliable, with a huge range of values that do not resemble any other year of this work, or anything found so far in the literature.

Ratios of RNA/DNA were expected to increase as larval fish grow (Rooker et al. 1997). We used linear regressions to explore the relationship of RNA/DNA ratios and larval lengths by system in 2014 and 2015. We used t-tests to compare slopes or intercepts for differences between systems within each year. RNA/DNA ratios for each

subestuary were also plotted against larval TL, and reference lines indicating starving (RNA/DNA < 2; Blom et al. 1997) and fed larvae (RNA/DNA > 3; Buckley 1984; Wright and Martin 1985) based on values from larvae of several marine species and Striped Bass were added to the plots.

The proportions of larvae with RNA/DNA ratios less than 2 (proportion starved or  $P_s$ ) were estimated for each subestuary as

$$^{(8)} P_s = N_{<2} / N_{total};$$

where  $N_{<2}$  equaled the number of samples with RNA/DNA ratios less than 2 and  $N_{total}$  equaled the total number of RNA/DNA samples. Proportions of larvae with RNA/DNA ratios greater than 3 (proportion fed or  $P_f$ ) were estimated as in equation 8, but  $P_f$  was estimated with the number of larvae with RNA/DNA ratios greater than 3 ( $N_{>3}$ ) in the numerator of equation 8.

## Results

During 2015, sampling on Choptank River lasted from April 7 to May 6, while sampling on Patuxent River lasted from April 8 to May 8. Samples through April 24 and April 30 were used to estimate  $L_p$  in Choptank and Patuxent Rivers, respectively. Sampling began on April 2 in the Nanticoke River and ended on April 28; dates between April 7 and 28 were used for estimating  $L_p$ . Mattawoman Creek was sampled between April 14 and May 6, and all dates were used to estimate  $L_p$ .

Based on 95% CIs, estimates of  $L_p$  during 2015 were sufficiently precise to separate them from the thresholds (Figure 2-2). Brackish subestuaries (Choptank, Nanticoke, and Patuxent Rivers) were all similar to each other ( $L_p$  range 0.65 to 0.82) based on 95% CI overlap. Mattawoman Creek ( $L_p = 1.0$ ) was not overlapped by 95% CI's of the three brackish subestuaries (Figure 2-2).

Comparisons of  $L_p$  during 2015 with historical estimates for brackish subestuaries is plotted in Figure 2-3 and for fresh-tidal values in Figure 2-4. The range of C/ha values available for analysis with  $L_p$  was 0.05-2.73 for brackish subestuaries and 0.46-3.33 for tidal-fresh (Table 2-2). Estimates of  $L_p$  in 2015 were among the top historical values for Choptank and Nanticoke rivers and Mattawoman Creek; historical values were not available for Patuxent River.

Separate linear regressions of C/ha and  $L_p$  by salinity category were significant at  $P \leq 0.0005$ ; Table 2-3). These analyses indicated that C/ha was negatively related to  $L_p$  and  $L_p$  was, on average, higher in tidal-fresh subestuaries than in brackish subestuaries.

Estimates of C/ha accounted for 28% of variation of  $L_p$  in brackish subestuaries and 34% in tidal-fresh subestuaries. Based on 95% CI overlap, intercepts were significantly different between tidal-fresh (mean = 0.94, SE = 0.09) and brackish (mean = 0.58, SE = 0.04) subestuaries. Mean slope for C/ha estimated for tidal-fresh subestuaries (mean = -0.29, SE = 0.07) were steeper, but 95% CI's overlapped CI's estimated for the slope of brackish subestuaries (mean = -0.17, SE = 0.04; Table 2-3). Both regressions indicated that  $L_p$  would be extinguished between 3.0 and 3.5 C/ha (Figure 2-5).

Overall, the multiple regression approach offered a similar fit ( $R^2 = 0.34$ ; Table 2-3) to separate regressions for each type of subestuary. Intercepts of tidal-fresh and brackish subestuaries equaled 0.94 and 0.57, respectively; the common slope was -0.19. Predicted  $L_p$  over the observed ranges of C/ha would decline from 0.58 to 0.12 in brackish subestuaries and from 0.81 to 0 in tidal-fresh subestuaries (Figure 2-5).

Akaike's Information Criteria values equaled 9.5 for the regression of  $C/ha$  and  $L_p$  for brackish subestuaries, 9.9 for tidal-fresh estuaries, and 11.5 for the multiple regression that included salinity category. Calculations of  $\Delta_i$  for brackish or tidal-fresh versus multiple regressions were approximately 2.01 and 1.59 (respectively), indicating that either hypothesis (different intercepts for tidal-fresh and brackish subestuaries with different or common slopes describing the decline of  $L_p$  with  $C/ha$ ) were plausible.

Although we have analyzed these data in terms of tidal-fresh and brackish subestuaries, inspection of Table 2-2 indicated an alternative interpretation based on primary land use estimated by MDP. Predominant land use at lowest levels of development may be influencing the intercept estimates. Rural watersheds were absent for tidal-fresh subestuaries analyzed and the lowest levels of development were dominated by forest (Figure 2-6). Nearly all rural land in brackish tributaries was dominated by agriculture. Dominant land cover estimated by MD DOP for watersheds of tidal-fresh subestuaries was equally split between forest ( $C/ha = 0.46-0.91$ ; 17 observations) and urban ( $C/ha > 1.17$ ; 14 observations). Brackish subestuary watershed rural lands were dominated by agriculture ( $C/ha < 0.27$ ; 34 observations), while forest land cover ( $C/ha \sim 0.09$ ) was represented by six observations. The range of  $L_p$  was similar in brackish subestuaries with forest and agricultural cover, but the distribution seemed shifted towards higher  $L_p$  in the limited sample from the forested watershed (Nanjemoy Creek). Urban land cover predominated in 11 observations of brackish subestuaries ( $C/ha > 1.21$ ; Table 2-2; Figure 2-6). Tidal-fresh subestuary intercepts may have represented the intercept for forest cover and brackish subestuary intercepts may have represented agricultural influence. If this is the case, then forest cover provides for higher  $L_p$  than agriculture. Increasing suburban land cover leads to a significant decline in  $L_p$  regardless of rural land cover type.

We examined 156 larval guts of 6-9 mm TL Yellow Perch larvae during 2010, 337 in 2011, 442 in 2012, 618 in 2013, 1495 in 2014, and 1417 in 2015 (Table 2-4). Samples were drawn primarily from tidal-fresh subestuaries (19 of 31 subestuary and year combinations). A smaller sample size was available for correlations with OM0 ( $N = 26$ ) than other variables ( $N = 31$ ) because observations of OM did not start until 2011.

Larval Yellow Perch guts contained food in all years and subestuaries except Piscataway Creek during 2011 (Table 2-4). Copepods were not found in larval stomachs in Elk River in 2012 and Northeast River in 2013, but were the most prevalent food item in 16 out of 30 system-year combinations (Piscataway Creek in 2011 is excluded since guts there did not contain any food), and were found in 35-100% of stomachs sampled. In 2015, copepods were present 73-96% of the time. Cladocerans were not found in larval stomachs in six of the 30 system-year combinations, and were the most prevalent food item 13% of the time. In 2015 cladocerans were present in 11-87% of guts sampled. "Other" food items were present in a higher fraction of samples in all system-year combinations than cladocerans, and were the most prevalent category 34% of the time. This category was predominant in larval gut samples from all five subestuaries during 2012 (69-100%; Table 2-4), but it should be noted that most gut contents in that year were already too digested to be identifiable and could not be categorized any other way. Gut content identification was more straightforward in other years, and except for 2014 when large amounts of digested material again could not be identified, "other" food items

were present in 0-42% of samples. In 2015 “other” food items were present in 22-36% of stomachs sampled.

During 2010-2015, percentage of guts without food (P0) ranged from 0 to 52% in all subestuary and year combinations except Piscataway Creek during 2011 (100%). Mean fullness rank ranged between 0.50 and 3.23 in all subestuary and year combinations except Piscataway Creek during 2011 (it was 0; Table 2-4).

Estimates of C/ha and OM0 were significantly related. A non-linear power function fit the data (approximate  $r^2 = 0.48$ ,  $P < 0.0001$ ;  $N = 25$ ), depicting OM0 increasing towards 1.0 at a decreasing rate as C/ha approached 1.50 (Figure 2-7). The relationship was described by the equation:

$$^{(9)} \text{OM0} = 0.87 \cdot ((\text{C/ha})^{0.14});$$

Approximate standard errors were 0.05 and 0.06 for parameters a and b, respectively. The logistic growth function (equation 6) fit these data similarly, but term a was not significantly different from zero.

Regression analyses suggested that the amount and type of food present in larval Yellow Perch stomachs was related to OM, although not all relationships were linear (Table 2-5). Quadratic relationships were indicated for some comparisons, suggesting there might be too much OM (acting as a prey refuge), too little (not enough to support zooplankton), and an optimum amount. Estimates of mean fullness rank exhibited a dome-shaped quadratic relationship with OM0 ( $r^2 = 0.37$ ,  $P = 0.006$ ; Table 2-5; Figure 2-8).  $P_{\text{cope}}$  also had a significant dome-shaped relationship with OM0 ( $r^2 = 0.30$ ,  $P = 0.02$ ). The relationship of  $P_{\text{other}}$  to OM0 was linear and increasing ( $r^2 = 0.16$ ,  $P = 0.05$ ), although this relationship could be biased since there were years when stomach contents were too digested to identify and this was only way they could be categorized. Relationships of OM with  $P_{\text{clad}}$  or P0 were not detected (Table 2-5).

The amount of food present in larval guts was significantly related with to presence of copepods. Both mean fullness rank and P0 were linearly related with  $P_{\text{cope}}$  ( $r^2 = 0.71$ ,  $P = <0.0001$  and  $r^2 = 0.41$ ,  $P = <0.0001$ , respectively). Estimates of  $P_{\text{clad}}$  were linearly and significantly related to P0 ( $r^2 = 0.15$ ,  $P = 0.03$ ). Copepods represent a much larger food item than cladocerans typically encountered.

Percent wetlands per watershed (determined from the most recent MD Department of Planning estimates in 2010) and development were negatively related. An inverse power function fit the relationship of C/ha and percent wetland well (approximate  $r^2 = 0.68$ ,  $P = 0.004$ ,  $N = 8$ ; Figure 2-9). This relationship suggested that wetlands could be the main source of organic material in our study areas. We do not know whether lower wetland percentages were normal for more developed watersheds or if wetlands were drained and filled during development prior to wetland conservation regulations.

Yellow Perch larvae were collected for RNA/DNA analysis from Patuxent ( $N = 167$ ;  $\text{C/ha} = 1.21$ ) and Choptank Rivers ( $N = 136$ ;  $\text{C/ha} = 0.16$ ) from April 13 - 30, 2015 (Table 2-6). Collections had larvae less than 6 mm, and exceeding 12 mm in length, but analysis was restricted to sizes in common for all years (6-9 mm) and typical of that of first-feeding larvae (Figure 2-10). Choptank River’s watershed was below the threshold development level, while Patuxent River has passed the suburban threshold. Estimates of OM0 were 0.56 and 0.54 in Patuxent and Choptank Rivers, respectively.

During 2015, all 6-9 mm TL Yellow Perch larvae examined from Patuxent River had RNA/DNA ratios above the starvation threshold (2; Figure 2-10) and 97% had a ratio

greater than 3, indicating well fed larvae. Fifteen percent of Choptank River had RNA/DNA ratios below 2 and 67% had ratios greater than 3 (Figure 2-10). In 2014, few larvae in either Mattawoman or Nanjemoy creeks had RNA/DNA ratios greater than 3 ( $P_f = 0.08$ ,  $N = 217$  and  $P_f = 0.05$ ,  $N = 255$ , respectively). The majority of larvae collected were in starved condition ( $P_s = 0.57$  and  $0.66$ , respectively; Figure 2-9).

Differences in relationships of RNA/DNA ratios with length were not detected between systems within the same year (slopes and intercepts were not different), but slopes were different between 2014 (declining) and 2015 (not different from 0; Table 2-7; Figure 2-10). T-tests of slopes and intercepts from linear regressions of RNA/DNA ratios versus larval TL (6-9 mm, in 0.5 mm increments) by year and system (2014 = Mattawoman and Nanjemoy, and 2015 = Choptank and Patuxent) did not indicate significantly different relationships between systems within each year (Table 2-7;  $P = 0.60$  and  $P = 0.71$ , respectively).

### Discussion

Estimates of  $L_p$  declined perceptibly once development exceeded the threshold (0.83 C/ha or 10% IS). A forest cover classification in a watershed was associated with higher  $L_p$  (median  $L_p = 0.78$ ) than agriculture (median  $L_p = 0.53$ ) or development (median  $L_p = 0.30$ ), but these differences may have also reflected dynamics unique to brackish or fresh-tidal subestuaries. Correlation and regression analyses indicated that development influenced the percentage of wetlands present within a watershed, which could in turn influence organic matter (OM) and larval Yellow Perch feeding dynamics. A “Goldilocks Hypothesis” indicated an optimum level of OM for first-feeding larvae existed. Too much OM may function as a predation refuge and too little OM may limit zooplankton production. Comparisons of RNA/DNA ratios of first-feeding larvae between two watersheds below the development target with two watersheds with development above, but near, the threshold have not supported differences indicated by comparisons of watershed development, OM levels and feeding success during 2010-2015 ( $N = 31$ ).

Interpretation of the influence of salinity class or primary land cover on  $L_p$  needs to consider that our survey design was limited to existing patterns of development. All estimates of  $L_p$  at or below target levels of development (forested and agricultural watersheds) or at the threshold or beyond high levels of development (except for one sample) were from brackish subestuaries; estimates of  $L_p$  for development between these levels were from tidal-fresh subestuaries with forested watersheds. Larval dynamics below the target level of development primarily reflected Eastern Shore agricultural watersheds. Two types of land use would be needed to balance analyses: (1) agricultural, tidal-fresh watersheds with below target development and (2) forested, brackish watersheds with development between the target and threshold. We do not believe that these combinations exist where Yellow Perch spawning occurs in Maryland’s portion of Chesapeake Bay.

We have relied on correlation and regression analyses to judge the effects of watershed development on yellow perch larval dynamics. Ideally, manipulative experiments and formal adaptive management should be employed (Hilborn 2016). In large-scale aquatic ecosystems these opportunities are limited and are not a possibility for us. Correlations are often not causal, but may be all the evidence available. Correlative evidence is strongest when (1) correlation is high, (2) it is found consistently across multiple situations, (3) there are not competing explanations, and (4) the correlation is

consistent with mechanistic explanations that can be supported by experimental evidence (Hilborn 2016).

Salinity may restrict  $L_p$  in brackish subestuaries by limiting the amount of available low salinity habitat over that of tidal-fresh subestuaries. Uphoff (1991) found that 90% of larvae collected in Choptank River during 1980-1985 were from 1‰ or less. Mortality of Yellow Perch eggs and prolarvae in experiments generally increased with salinity and was complete by 12‰ (Sanderson 1950; Victoria et al. 1992). The range of suitable salinities for prolarvae was lower than that for eggs (Victoria et al. 1992).

Yellow Perch egg viability declined in highly developed suburban watersheds of Chesapeake Bay (C/ha above threshold level; Uphoff et al. 2005; Blazer et al. 2013). Abnormalities in ovaries and testes of adult Yellow Perch during spawning season were found most frequently in subestuaries with suburban watersheds and these abnormalities were consistent with contaminant effects (Blazer et al. 2013). Blazer et al. (2013) explained the biology behind low egg viability observed by Uphoff et al. (2005) in Severn River during 2001-2003 and persistently low  $L_p$  detected in three western shore subestuaries with highly developed suburban watersheds (C/ha > 1.59; Severn, South, and Magothy Rivers). Endocrine disrupting chemicals were more likely to cause observed egg hatching failure in well developed tributaries than hypoxia and increased salinity (Blazer et al. 2013), factors identified as potential contributors to poor egg hatching success in Severn River (Uphoff et al. 2005). Low  $L_p$  occurs sporadically in subestuaries with rural watersheds and appears linked to high temperatures (Uphoff et al. 2013).

It is unlikely that low  $L_p$  has always existed in well developed Magothy, Severn, and South rivers since all supported well known recreational fisheries into the 1970s (the C/ha thresholds were met during the late 1960s - 1970s). Severn River supported a state hatchery through the first half of the twentieth century and hatching rates of eggs in the hatchery were high up to 1955, when records ended (Muncy 1962). News accounts described concerns about fishery declines in these rivers during the 1980s and recreational fisheries were closed in 1989 (commercial fisheries had been banned many years earlier; Uphoff et al. 2005). Egg hatching success of Severn River Yellow Perch declined drastically by the early 2000s when estimates of  $L_p$  were persistently low (Uphoff et al. 2005). Yellow Perch egg per recruit (EPR) analyses incorporating Severn River egg hatch ratios or relative declines in  $L_p$  with C/ha indicated that recovery of Yellow Perch EPR in Severn River (and other developed tributaries) by managing the fishery alone would not be possible (Uphoff et al. 2014). Angler reports indicated that viable recreational fisheries for Yellow Perch returned to these areas in the mid-to-late 1990s. These reconstituted fisheries were likely supported by juvenile Yellow Perch that migrate from the upper Bay nursery rather than internal production (Uphoff et al. 2005). Trends in volunteer angler catch per trip in Magothy River matched upper Bay estimates of stock abundance during 2008-2014 (P. Piavis, MD DNR, personal communication). Recreational fisheries in these three subestuaries were reopened to harvest in 2009 to allow for some recreational benefit of fish that migrated in and provided a natural “put-and-take” fishery.

High estimates of  $L_p$  that were equal to or approaching 1.0 have been routinely encountered in the past, and it is likely that counts would be needed to measure relative abundance if greater resolution was desired. Mangel and Smith (1990) indicated that

presence-absence sampling of eggs would be more useful for indicating the status of depleted stocks and count-based indices would be more accurate for recovered stocks. Larval indices based on counts have been used as a measure of year-class strength of fishes generally (Sammons and Bettoli 1998) and specifically for Yellow Perch (Anderson et al. 1998). Tighter budgets necessitate development of less costly indicators of larval survival and relative abundance in order to pursue ecosystem-based fisheries management. Characterizations of larval survival and relative abundance normally are derived from counts requiring labor-intensive sorting and processing. Estimates of  $L_p$  were largely derived in the field and only gut contents and RNA/DNA required laboratory analysis. These latter two analyses represented separate studies rather than a requirement for estimating  $L_p$ .

There appears to be some potential for development to influence the percentage of wetlands present within a watershed, which, in turn, influences organic matter (OM) and larval Yellow Perch feeding dynamics. A “Goldilocks” hypothesis explains the dome-shaped relationship of OM and larval feeding we detected; too much OM might function as a predation refuge and too little OM limits zooplankton production (Hoffman et al. 2007). Foraging arena theory suggests that high levels of OM may moderate predation by Yellow Perch larvae by providing zooplankton more places to hide and forage safely (Walters and Martell 2004) or OM could physically hinder larval attack success.

Years of high spring discharge favor anadromous fish recruitment in Chesapeake Bay (Hoffman et al. 2007; Martino and Houde 2010) and may represent episodes of hydrologic transport of accumulated OM from watersheds (McClain et al. 2003) that fuel zooplankton production and feeding success. Under natural conditions in York River, Virginia, riparian marshes and forests would provide OM subsidies in high discharge years (Hoffman et al. 2007), while phytoplankton would be the primary source of OM in years of lesser flow. Stable isotope signatures of York River American shad larvae and zooplankton indicated that terrestrial OM largely supported one of its most successful year-classes. Lesser year-classes of American shad on the York River were associated with low flows, OM based on phytoplankton, and lesser zooplankton production (Hoffman et al. 2007). The York River watershed, with large riparian marshes and forest, was largely intact relative to other Chesapeake Bay tributaries (Hoffman et al. 2007). Multiple regression models provided evidence that widespread climate factors (March precipitation as a proxy for OM transport and March air temperature) influenced survival of Yellow Perch eggs and larvae in Chesapeake Bay (Uphoff et al. 2013).

Zooplankton supply (cladocerans and copepods) for first-feeding Yellow Perch larvae has been identified as an influence on survival in Lake Michigan (Dettmers et al. 2003; Redman et al. 2011; Weber et al. 2011) and Canadian boreal lakes (Leclerc et al. 2011), and survival of European perch *Perca fluviatilis* in the Baltic Sea (Ljunggren et al. 2003). In a two-year study in Lake Saint Pierre, Canada, Tardif et al. (2005) attributed larval Yellow Perch RNA/DNA response to wetland types, cumulative degree days, and feeding conditions. The importance of adequate zooplankton supply and factors influencing zooplankton dynamics have been established for survival of Chesapeake Bay Striped Bass, White Perch, and American Shad larvae (North and Houde 2001; 2003; Hoffman et al. 2007; Martino and Houde 2010). Yellow Perch larvae share habitat in Chesapeake Bay subestuaries with these species, but little has been published on larval Yellow Perch dynamics and feeding ecology in Chesapeake Bay (Uphoff 1991).

Urbanization reduces quantity and quality of OM in streams (Paul and Meyer 2001; Gücker et al. 2011; Stanley et al. 2012). Riparian zones and floodplains that are sources of OM become disconnected from stream channels by stormwater management in suburban and urban watersheds (Craig et al. 2008; Kaushal et al. 2008; Brush 2009; NRC 2009). Small headwater streams in the Gunpowder and Patapsco Rivers watersheds (tributaries of Chesapeake Bay) were sometimes buried in culverts and pipes, or were paved over (Elmore and Kaushal 2008). Decay of leaves occurred much faster in urban streams, apparently due to greater fragmentation from higher stormflow rather than biological activity (Paul and Meyer 2001). Altered flowpaths associated with urbanization affect timing and delivery of OM to streams (McClain et al. 2003). Organic matter was transported further and retained less in urban streams (Paul and Meyer 2001). Uphoff et al. (2011) and our current analysis found that the percentage of Maryland's Chesapeake Bay subestuary watersheds in wetlands declined hyperbolically as IS increased, so this source of OM diminishes with development.

Management for organic carbon is nearly non-existent despite its role as a great modifier of the influence and consequence of other chemicals and processes in aquatic systems (Stanley et al. 2012). It is unmentioned in the Chesapeake Bay region as reductions in nutrients (N and P) and sediment are pursued for ecological restoration ([http://www.epa.gov/reg3wapd/pdf/pdf\\_chesbay/BayTMDLFactSheet8\\_6.pdf](http://www.epa.gov/reg3wapd/pdf/pdf_chesbay/BayTMDLFactSheet8_6.pdf)). However, most watershed management and restoration practices have the potential to increase OM delivery and processing, although it is unclear how ecologically meaningful these changes may be. Stanley et al. (2012) recommended beginning with riparian protection or re-establishment and expand outward as opportunities permit. Wetland management represents an expansion of effort beyond the riparian zone (Stanley et al. 2012).

Agriculture also has the potential to alter OM dynamics within a watershed (Stanley et al. 2012) and the effect of this major land use on fish habitat warrants further study. Agriculture has been associated with increased, decreased, and undetectable changes in OM that may reflect diversity of farming practices (Stanley et al. 2012). As indicated earlier, extensive forest cover in a watershed may be linked to higher  $L_p$  than agriculture. However, Uphoff et al (2011) noted that agricultural watersheds had more area in wetlands than urban watersheds and this could buffer loss of OM from decreased forest cover. Streams in agricultural watersheds were unlikely to become disconnected since urban stormwater controls would not be in use (Uphoff et al. 2011).

In our analyses, we assumed that mainstem Potomac or Susquehanna River water was not a major influence on subestuary water quantity, water quality, and zooplankton supply. Sampling for Yellow Perch larvae occurred in the upper portions of subestuaries and this should have minimized the influence of mainstem waters, although some intrusion would have been possible at the most downstream sites in the smallest systems closest to the major Rivers (i.e., Piscataway Creek for the Potomac). Strong correlations of  $C/ha$ ,  $L_p$ , and  $OM_0$  indicated that local conditions prevailed.

Comparisons of RNA/DNA ratio of first-feeding larvae between two watersheds below the development target and two watersheds with development above (but near) the threshold during 2014 and 2015 have not supported differences indicated by comparisons of watershed development, OM levels and feeding success during 2010-2015. RNA/DNA ratios for Yellow Perch larvae from Patuxent and Choptank Rivers during 2015 indicated that most were in the well fed category, while ratios in Mattawoman and Nanjemoy

Creeks in 2014 indicated that most were in the starved category. Differences in RNA/DNA ratios between systems within a year were not apparent, but differences between years were. The RNA/DNA ratio reflects the condition of larvae during the few days prior to sampling (Kimura et al. 2000), and the response time of RNA/DNA ratios of larval fishes characterizes the feeding environment within a week of sampling (Tardif et al. 2005). Ratios of RNA/DNA of fed larvae were expected to increase with body size (Clemmensen 1994), but did not in 2014 or 2015 samples. Instead of an expected increase in RNA/DNA with body size (Rooker et al. 1997), a significant decline was observed in 2014 and no change was indicated in 2015.

Laboratory studies of RNA/DNA ratios of fed and starved larval Yellow Perch have not been conducted and we have relied on general guidelines from other species (Blom et al. 1997). Tardif et al. (2005) determined that RNA/DNA ratios of Yellow Perch in Lake Saint Pierre, Canada, averaged below 2, but did not provide indication of nutritional state of these larvae. Surveys of larval Striped Bass RNA/DNA in 1981 in the Potomac River estuary exhibited a similar declining pattern, but ratios stabilized above starvation values (Martin et al. 1985). Blom et al. (1997) detected a decline in RNA/DNA ratios of Atlantic herring *Clupea harengus*; but few herring larvae were observed with ratios indicating starvation. A significant part of variation in early life history traits of fish can be attributed to the parental origin of the individual larvae (Bang et al. 2006), and at hatch, parental origin is the cause of large variation among individual larvae in important traits such as length and weight (Bang et al. 2006). However, 95% confidence intervals of our four sets of intercepts overlapped, suggesting that initial RNA/DNA levels (6 mm larvae) were not different. Low RNA/DNA ratios exhibited by larger Yellow Perch in 2014 were likely to have reflected external nutrition. RNA/DNA ratios of Atlantic Herring larvae fed shortly after hatching were in the same range as those found for starved larvae and were thought to result from problems in changing from internal to external nutrition (Clemmensen 1994). There was no difference in RNA/DNA ratios for starved and fed Atlantic Herring larvae up to an age of 10 days, but after 10 days deprivation of food lead to a significant decrease in RNA/DNA ratios in comparison to fed Atlantic Herring larvae (Clemmensen 1994). Low RNA/DNA ratios of larger, and presumably older, Yellow Perch larvae sampled from our subestuaries may be more indicative of poor feeding conditions, although it is possible that bias may have resulted from starving, weaker, poorly-growing larvae being more vulnerable to our plankton nets than fed larvae. Poor nutritional condition of larvae not only contributes to an increase in mortality rates through starvation, but can lead to an increase in predation mortality through diminished escape responses or increased duration of larval stage (Caldarone et al. 2003).

Development was an important influence on Yellow Perch egg and larval dynamics and negative changes generally conformed to impervious surface reference points developed from distributions of dissolved oxygen, and juvenile and adult target fish in mesohaline subestuaries (Uphoff et al. 2011). Hilborn and Stokes (2010) advocated setting reference points related to harvest for fisheries (stressor) based on historical stock performance (outcome) because they were based on experience, easily understood, and not based on modeling. We believe applying IS or C/ha watershed development reference points (stressor) based on  $L_p$  (outcome) conforms to the approach advocated by Hilborn and Stokes (2010).

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Table 2-1. Regression coefficients for DNA and RNA standard curves used for quantification of sample concentrations of nucleic acids in 2014.

| Model | Intercept | Slope ([DNA or RNA]) | Slope ([DNA or RNA] <sup>2</sup> ) | p-value | R <sup>2</sup> |
|-------|-----------|----------------------|------------------------------------|---------|----------------|
| DNA   | 2.111     | 4.15E-03             | -2.71E-06                          | <0.0001 | 0.90           |
| RNA   | 5.802     | 6.85E-03             | -4.16E-06                          | <0.0001 | 0.95           |

Table 2-2. Estimates of proportions of ichthyoplankton net tows with Yellow Perch larvae ( $L_p$ ) during 1965-2015 and data used for regression with counts of structures per hectare (C/ha). Salinity class 0 = tidal-fresh ( $\leq 2.0$  ‰) and 1 = brackish ( $> 2.0$  ‰). Year is the year a subestuary was sampled. Primary land use was determined from Maryland Department of Planning estimates for 1973, 1994, 1997, 2002, or 2010 that were closest to a sampling year. These latter categories were not used in regression analysis.

| River      | Year | C / ha | Primary Land Use | Salinity | $L_p$ | River      | Year | C / ha | Primary Land Use | Salinity | $L_p$ |
|------------|------|--------|------------------|----------|-------|------------|------|--------|------------------|----------|-------|
| Bush       | 2006 | 1.17   | Urban            | 0        | 0.79  | Nanjemoy   | 2011 | 0.09   | Forest           | 1        | 0.99  |
| Bush       | 2007 | 1.19   | Urban            | 0        | 0.92  | Nanjemoy   | 2012 | 0.09   | Forest           | 1        | 0.03  |
| Bush       | 2008 | 1.20   | Urban            | 0        | 0.55  | Nanjemoy   | 2013 | 0.09   | Forest           | 1        | 0.46  |
| Bush       | 2009 | 1.21   | Urban            | 0        | 0.86  | Nanjemoy   | 2014 | 0.09   | Forest           | 1        | 0.82  |
| Bush       | 2011 | 1.23   | Urban            | 0        | 0.96  | Nanticoke  | 1965 | 0.05   | Agriculture      | 1        | 0.50  |
| Bush       | 2012 | 1.24   | Urban            | 0        | 0.28  | Nanticoke  | 1967 | 0.05   | Agriculture      | 1        | 0.43  |
| Bush       | 2013 | 1.25   | Urban            | 0        | 0.15  | Nanticoke  | 1968 | 0.05   | Agriculture      | 1        | 1.00  |
| Choptank   | 1986 | 0.09   | Agriculture      | 1        | 0.53  | Nanticoke  | 1970 | 0.06   | Agriculture      | 1        | 0.81  |
| Choptank   | 1987 | 0.09   | Agriculture      | 1        | 0.73  | Nanticoke  | 1971 | 0.06   | Agriculture      | 1        | 0.33  |
| Choptank   | 1988 | 0.10   | Agriculture      | 1        | 0.80  | Nanticoke  | 2004 | 0.11   | Agriculture      | 1        | 0.49  |
| Choptank   | 1989 | 0.10   | Agriculture      | 1        | 0.71  | Nanticoke  | 2005 | 0.11   | Agriculture      | 1        | 0.67  |
| Choptank   | 1990 | 0.10   | Agriculture      | 1        | 0.66  | Nanticoke  | 2006 | 0.11   | Agriculture      | 1        | 0.35  |
| Choptank   | 1998 | 0.13   | Agriculture      | 1        | 0.60  | Nanticoke  | 2007 | 0.11   | Agriculture      | 1        | 0.55  |
| Choptank   | 1999 | 0.13   | Agriculture      | 1        | 0.76  | Nanticoke  | 2008 | 0.11   | Agriculture      | 1        | 0.19  |
| Choptank   | 2000 | 0.13   | Agriculture      | 1        | 0.25  | Nanticoke  | 2009 | 0.11   | Agriculture      | 1        | 0.41  |
| Choptank   | 2001 | 0.13   | Agriculture      | 1        | 0.21  | Nanticoke  | 2011 | 0.11   | Agriculture      | 1        | 0.55  |
| Choptank   | 2002 | 0.14   | Agriculture      | 1        | 0.38  | Nanticoke  | 2012 | 0.11   | Agriculture      | 1        | 0.04  |
| Choptank   | 2003 | 0.14   | Agriculture      | 1        | 0.52  | Nanticoke  | 2013 | 0.11   | Agriculture      | 1        | 0.43  |
| Choptank   | 2004 | 0.15   | Agriculture      | 1        | 0.41  | Nanticoke  | 2014 | 0.11   | Agriculture      | 1        | 0.35  |
| Choptank   | 2013 | 0.16   | Agriculture      | 1        | 0.47  | Nanticoke  | 2015 | 0.11   | Agriculture      | 1        | 0.64  |
| Choptank   | 2014 | 0.16   | Agriculture      | 1        | 0.68  | Northeast  | 2010 | 0.46   | Forest           | 0        | 0.68  |
| Choptank   | 2015 | 0.16   | Agriculture      | 1        | 0.82  | Northeast  | 2011 | 0.46   | Forest           | 0        | 1.00  |
| Corsica    | 2006 | 0.21   | Agriculture      | 1        | 0.47  | Northeast  | 2012 | 0.47   | Forest           | 0        | 0.77  |
| Corsica    | 2007 | 0.22   | Agriculture      | 1        | 0.83  | Northeast  | 2013 | 0.47   | Forest           | 0        | 0.72  |
| Elk        | 2010 | 0.59   | Forest           | 0        | 0.75  | Northeast  | 2014 | 0.47   | Forest           | 0        | 0.77  |
| Elk        | 2011 | 0.59   | Forest           | 0        | 0.79  | Patuxent   | 2015 | 1.21   | Urban            | 1        | 0.72  |
| Elk        | 2012 | 0.59   | Forest           | 0        | 0.55  | Piscataway | 2008 | 1.41   | Urban            | 0        | 0.47  |
| Langford   | 2007 | 0.07   | Agriculture      | 1        | 0.83  | Piscataway | 2009 | 1.43   | Urban            | 0        | 0.39  |
| Magothy    | 2009 | 2.73   | Urban            | 1        | 0.17  | Piscataway | 2010 | 1.45   | Urban            | 0        | 0.54  |
| Mattawoman | 1990 | 0.46   | Forest           | 0        | 0.81  | Piscataway | 2011 | 1.46   | Urban            | 0        | 0.65  |
| Mattawoman | 2008 | 0.87   | Forest           | 0        | 0.66  | Piscataway | 2012 | 1.47   | Urban            | 0        | 0.16  |
| Mattawoman | 2009 | 0.88   | Forest           | 0        | 0.92  | Piscataway | 2013 | 1.49   | Urban            | 0        | 0.50  |
| Mattawoman | 2010 | 0.90   | Forest           | 0        | 0.82  | Severn     | 2002 | 2.02   | Urban            | 1        | 0.16  |
| Mattawoman | 2011 | 0.91   | Forest           | 0        | 0.99  | Severn     | 2004 | 2.09   | Urban            | 1        | 0.29  |
| Mattawoman | 2012 | 0.90   | Forest           | 0        | 0.20  | Severn     | 2005 | 2.15   | Urban            | 1        | 0.33  |
| Mattawoman | 2013 | 0.91   | Forest           | 0        | 0.47  | Severn     | 2006 | 2.18   | Urban            | 1        | 0.27  |
| Mattawoman | 2014 | 0.91   | Forest           | 0        | 0.78  | Severn     | 2007 | 2.21   | Urban            | 1        | 0.30  |
| Mattawoman | 2015 | 0.91   | Forest           | 0        | 1.00  | Severn     | 2008 | 2.24   | Urban            | 1        | 0.08  |
| Middle     | 2012 | 3.33   | Urban            | 0        | 0.00  | Severn     | 2009 | 2.25   | Urban            | 1        | 0.15  |
| Nanjemoy   | 2009 | 0.09   | Forest           | 1        | 0.83  | Severn     | 2010 | 2.26   | Urban            | 1        | 0.03  |
| Nanjemoy   | 2010 | 0.09   | Forest           | 1        | 0.96  | South      | 2008 | 1.61   | Urban            | 1        | 0.14  |

Table 2-3. Summary of results of regressions of proportions of tows with Yellow Perch larvae ( $L_p$ ) and counts of structures per hectare (C/ha). Separate regressions by salinity (tidal-fresh  $\leq 2.0$  ‰ and brackish  $> 2.0$  ‰) and a multiple regression using salinity as a class variable (tidal-fresh = 0 and brackish = 1) are presented.

| ANOVA     |              | Brackish |         |         |           |           |
|-----------|--------------|----------|---------|---------|-----------|-----------|
| Source    | df           | SS       | MS      | F       | P         |           |
| Model     | 1            | 1.02214  | 1.02214 | 19.55   | <.0001    |           |
| Error     | 49           | 2.56148  | 0.05228 |         |           |           |
| Total     | 50           | 3.58363  |         |         |           |           |
| $r^2$     | 0.2852       |          |         |         |           |           |
|           | Coefficients | SE       | t Stat  | P-value | Lower 95% | Upper 95% |
| Intercept | 0.58434      | 0.03811  | 15.33   | <.0001  | 0.50775   | 0.66092   |
| C / ha    | -0.17028     | 0.03851  | -4.42   | <.0001  | -0.24767  | -0.0929   |

| ANOVA     |              | Tidal-Fresh |         |         |           |           |
|-----------|--------------|-------------|---------|---------|-----------|-----------|
| Source    | df           | SS          | MS      | F       | P         |           |
| Model     | 1            | 0.76581     | 0.76581 | 15.21   | 0.0005    |           |
| Error     | 29           | 1.46024     | 0.05035 |         |           |           |
| Total     | 30           | 2.22605     |         |         |           |           |
| $r^2$     | 0.344        |             |         |         |           |           |
|           | Coefficients | SE          | t Stat  | P-value | Lower 95% | Upper 95% |
| Intercept | 0.94173      | 0.08682     | 10.85   | <.0001  | 0.76416   | 1.11931   |
| C / ha    | -0.28802     | 0.07385     | -3.9    | 0.0005  | -0.43907  | -0.13697  |

| ANOVA     |              | Multiple Regression |         |         |           |           |
|-----------|--------------|---------------------|---------|---------|-----------|-----------|
| Source    | df           | SS                  | MS      | F       | P         |           |
| Model     | 2            | 2.11405             | 1.05703 | 20.25   | <.0001    |           |
| Error     | 79           | 4.12314             | 0.05219 |         |           |           |
| Total     | 81           | 6.23719             |         |         |           |           |
| $r^2$     | 0.3389       |                     |         |         |           |           |
|           | Coefficients | SE                  | t Stat  | P-value | Lower 95% | Upper 95% |
| Intercept | 0.84458      | 0.05437             | 15.53   | <.0001  | 0.73637   | 0.9528    |
| C / ha    | -0.19472     | 0.03425             | -5.68   | <.0001  | -0.2629   | -0.12654  |
| Salinity  | -0.24713     | 0.05482             | -4.51   | <.0001  | -0.35625  | -0.138    |

Table 2-4. Summary of estimates used in regression analyses of Yellow Perch larval feeding success. C/ha = counts of structures per hectare. Mean full = mean of fullness ranks assigned to larval guts. OM0 = proportion of samples without organic matter (detritus). P0 = proportion of guts without food. Pclad = proportion of guts with cladocerans. Pcope = proportion of guts with copepods. Pother = proportion of guts with “other” food items. Mean TL = mean TL of larvae in mm. N = number of Yellow Perch larvae examined. 2015 analyses are restricted to sizes-in-common of 6-9 mm for all years.

| River      | Year | C / ha | Mean full | OM0  | P0    | Pclad | Pcope | Pother | Mean TL | N   |
|------------|------|--------|-----------|------|-------|-------|-------|--------|---------|-----|
| Elk        | 2010 | 0.59   | 2.43      |      | 0.00  | 0.00  | 1.00  | 0.14   | 8.7     | 7   |
| Mattawoman | 2010 | 0.90   | 1.42      |      | 0.15  | 0.15  | 0.67  | 0.09   | 8.4     | 33  |
| Nanjemoy   | 2010 | 0.09   | 2.89      |      | 0.00  | 0.08  | 1.00  | 0.19   | 8.6     | 36  |
| Northeast  | 2010 | 0.46   | 1.89      |      | 0.28  | 0.30  | 0.57  | 0.32   | 7.5     | 47  |
| Piscataway | 2010 | 1.45   | 2.24      |      | 0.03  | 0.00  | 0.52  | 0.67   | 8.4     | 33  |
| Elk        | 2011 | 0.59   | 2.98      | 0.76 | 0.07  | 0.00  | 0.93  | 0.00   | 8.1     | 60  |
| Mattawoman | 2011 | 0.91   | 0.59      | 0.78 | 0.48  | 0.00  | 0.41  | 0.11   | 8.2     | 54  |
| Nanjemoy   | 2011 | 0.09   | 2.00      | 0.56 | 0.07  | 0.05  | 0.77  | 0.13   | 7.5     | 84  |
| Nanticoke  | 2011 | 0.11   | 3.23      | 0.55 | 0.08  | 0.70  | 0.93  | 0.13   | 7.9     | 40  |
| Northeast  | 2011 | 0.46   | 2.43      | 0.58 | 0.10  | 0.00  | 0.89  | 0.08   | 8.0     | 72  |
| Piscataway | 2011 | 1.46   | 0.00      | 1.00 | 1.00  | 0.00  | 0.00  | 0.00   | 8.2     | 27  |
| Bush       | 2012 | 1.24   | 2.33      |      | 0.00  | 0.52  | 0.52  | 1.00   | 8.2     | 33  |
| Elk        | 2012 | 0.59   | 0.76      | 0.77 | 0.25  | 0.02  | 0.00  | 0.69   | 7.6     | 190 |
| Mattawoman | 2012 | 0.90   | 1.69      | 1.00 | 0.00  | 0.38  | 0.85  | 1.00   | 8.3     | 13  |
| Northeast  | 2012 | 0.47   | 1.17      | 0.99 | 0.02  | 0.05  | 0.08  | 0.98   | 7.5     | 198 |
| Piscataway | 2012 | 1.47   | 1.63      | 0.98 | 0.00  | 0.50  | 0.75  | 1.00   | 8.5     | 8   |
| Choptank   | 2013 | 0.16   | 1.00      | 0.33 | 0.22  | 0.34  | 0.35  | 0.32   | 7.5     | 302 |
| Mattawoman | 2013 | 0.91   | 1.66      | 0.79 | 0.00  | 0.81  | 0.66  | 0.05   | 7.2     | 80  |
| Nanjemoy   | 2013 | 0.09   | 1.60      | 0.65 | 0.00  | 0.60  | 0.44  | 0.23   | 7.3     | 62  |
| Nanticoke  | 2013 | 0.11   | 0.97      | 0.13 | 0.37  | 0.35  | 0.23  | 0.24   | 7.8     | 116 |
| Northeast  | 2013 | 0.47   | 0.50      | 1.00 | 0.52  | 0.13  | 0.00  | 0.41   | 7.9     | 46  |
| Piscataway | 2013 | 1.49   | 2.33      | 0.74 | 0.00  | 0.33  | 0.75  | 0.17   | 7.6     | 12  |
| Choptank   | 2014 | 0.16   | 1.56      | 0.60 | 0.006 | 0.87  | 0.54  | 0.88   | 7.8     | 539 |
| Mattawoman | 2014 | 0.91   | 1.88      | 0.72 | 0.00  | 0.95  | 0.87  | 1.00   | 7.1     | 241 |
| Nanjemoy   | 2014 | 0.09   | 2.43      | 0.53 | 0.00  | 0.34  | 0.73  | 0.42   | 7.7     | 292 |
| Nanticoke  | 2014 | 0.11   | 1.36      | 0.11 | 0.00  | 0.61  | 0.43  | 0.04   | 8.1     | 28  |
| Northeast  | 2014 | 0.47   | 1.40      | 0.86 | 0.05  | 0.65  | 0.53  | 0.69   | 7.9     | 395 |
| Choptank   | 2015 | 0.16   | 2.89      | 0.54 | 0.00  | 0.34  | 0.93  | 0.29   | 7.2     | 227 |
| Mattawoman | 2015 | 0.91   | 1.58      | 0.74 | 0.06  | 0.11  | 0.73  | 0.22   | 7.5     | 738 |
| Nanticoke  | 2015 | 0.11   | 1.51      | 0.25 | 0.09  | 0.34  | 0.76  | 0.27   | 7.8     | 237 |
| Patuxent   | 2015 | 1.21   | 2.33      | 0.56 | 0.00  | 0.87  | 0.90  | 0.36   | 7.4     | 215 |

Table 2-5. Summary of regression results for Yellow Perch larval feeding success. Mean full = average feeding rank of larvae. OM0 = proportion of samples without organic matter (detritus). P0 = proportion of guts without food. Pclad = proportion of guts with cladocerans. Pcope = proportion of guts with copepods. Pother = proportion of guts with “other” food items. Independent/dependent = how variables were assigned in regression analyses, shape = shape of plotted data with ND indicating none detected,  $r^2$  = coefficient of determination, and P = level of significance. Gray shading indicates significance of interest at  $P \leq 0.05$ .

| Independent | Dependent | Relationship | Shape    | $r^2$ | P       | Intercept | SE     | Parameter a | SE     | Parameter b | SE     |
|-------------|-----------|--------------|----------|-------|---------|-----------|--------|-------------|--------|-------------|--------|
| OM0         | P0        | Quadratic    | U        | 0.18  | 0.11    | 0.3299    | 0.2063 | -1.1601     | 0.7195 | 1.1425      | 0.5921 |
| OM0         | Pclad     | Quadratic    | Dome     | 0.07  | 0.46    | 0.3736    | 0.2923 | 0.4646      | 1.0196 | -0.6130     | 0.8391 |
| OM0         | Pcope     | Quadratic    | Dome     | 0.30  | 0.02    | 0.1094    | 0.2563 | 2.3502      | 0.8937 | -2.1649     | 0.7356 |
| OM0         | Pother    | Linear       | Increase | 0.16  | 0.05    | 0.0336    | 0.1818 | 0.5370      | 0.2569 | ---         | ---    |
| OM0         | Mean full | Quadratic    | Dome     | 0.37  | 0.006   | 0.3873    | 0.6137 | 6.4901      | 2.1402 | -6.0246     | 1.7615 |
| Pclad       | P0        | Linear       | Decline  | 0.15  | 0.03    | 0.2181    | 0.0550 | -0.2804     | 0.1227 | ---         | ---    |
| Pcope       | P0        | Linear       | Decline  | 0.41  | <0.0001 | 0.4010    | 0.0688 | -0.4581     | 0.1021 | ---         | ---    |
| Pother      | P0        | Linear       | Decline  | 0.09  | 0.11    | 0.1968    | 0.0580 | -0.1859     | 0.1122 | ---         | ---    |
| P0          | Mean full | Linear       | Decline  | 0.47  | <0.0001 | 2.0691    | 0.1188 | -2.4602     | 0.4810 | ---         | ---    |
| Pclad       | Mean full | Linear       | ND       | 0.02  | 0.44    | 1.6404    | 0.2111 | 0.3685      | 0.4710 | ---         | ---    |
| Pcope       | Mean full | Linear       | Increase | 0.71  | <0.0001 | 0.4656    | 0.1737 | 2.1476      | 0.2577 | ---         | ---    |
| Pother      | Mean full | Linear       | ND       | 0.007 | 0.66    | 1.8372    | 0.2159 | -0.1876     | 0.4177 | ---         | ---    |

Table 2-6. Summary of feeding success, larval length, sample size, and RNA/DNA characteristics, by subestuary and sample date. Data only for dates with feeding information and with RNA/DNA analysis are summarized. Mean fullness = mean feeding rank. Mean TL is in mm. N = the sample size of larvae processed for gut contents and with RNA/DNA ratios available for the date. Mean RNA/DNA is the average for the date. SE RNA/DNA is the standard error for the date. N RNA/DNA > 3 is the number of ratios above the fed criterion. N RNA/DNA < 2 is the number of ratios below the starvation criterion.

| Subestuary | Variable                | 13-Apr | 15-Apr | 22-Apr | 24-Apr | 29-Apr |        |
|------------|-------------------------|--------|--------|--------|--------|--------|--------|
| Choptank   | Mean Fullness           | 2.7    | 3.0    | 3.2    | 2.7    | 3.0    |        |
|            | Mean TL                 | 6.9    | 7.4    | 9.6    | 11.1   | 11.8   |        |
|            | N                       | 50     | 60     | 13     | 7      | 6      |        |
|            | Mean RNA/DNA            | 3.89   | 4.00   | 4.29   | 4.73   | 5.36   |        |
|            | SE RNA/DNA              | 0.29   | 0.18   | 0.35   | 0.31   | 0.72   |        |
|            | <i>Ps</i> (RNA/DNA ≥ 3) | 0.50   | 0.82   | 0.77   | 1      | 1      |        |
|            | <i>Pf</i> (RNA/DNA ≤ 2) | 0.38   | 0.10   | 0.08   | 0      | 0      |        |
|            |                         | 14-Apr | 16-Apr | 21-Apr | 23-Apr | 28-Apr | 30-Apr |
| Patuxent   | Mean Fullness           | 2.2    | 2.4    | 2.1    | 2.2    | 2.6    | 3.3    |
|            | Mean TL                 | 7.2    | 7.4    | 9.8    | 10.8   | 12.3   | 13.3   |
|            | N                       | 50     | 49     | 45     | 10     | 9      | 4      |
|            | Mean RNA/DNA            | 4.68   | 5.10   | 4.07   | 5.61   | 5.35   | 5.75   |
|            | SE RNA/DNA              | 0.15   | 0.11   | 0.29   | 0.18   | 0.29   | 0.43   |
|            | <i>Ps</i> (RNA/DNA ≥ 3) | 1      | 1      | 0.60   | 1      | 1      | 1      |
|            | <i>Pf</i> (RNA/DNA ≤ 2) | 0      | 0      | 0.18   | 0      | 0      | 0      |

Table 2-7. Summary of data and t-test results comparing intercepts, slopes, and their standard errors (SE) of RNA/DNA ratios versus length in 2014 (Mattawoman and Nanjemoy Creeks) and 2015 (Patuxent and Choptank Rivers).

| 2014 RNA/DNA vs Length | Mattawoman | Nanjemoy | 2015 RNA/DNA vs Length | Patuxent | Choptank |
|------------------------|------------|----------|------------------------|----------|----------|
| RNA/DNA Intercept      | 4.82718    | 4.86381  | RNA/DNA Intercept      | 6.16724  | 3.98931  |
| RNA/DNA Intercept SE   | 0.36144    | 0.35112  | RNA/DNA Intercept SE   | 1.05537  | 1.8828   |
| RNA/DNA Slope          | -0.40391   | -0.39967 | RNA/DNA Slope          | -0.17545 | -0.00652 |
| RNA/DNA Slope SE       | 0.05101    | 0.04551  | RNA/DNA Slope SE       | 0.14235  | 0.25824  |

| <i>2014 RNA/DNA vs Length</i> | <i>Variable 1</i> | <i>Variable 2</i> | <i>2015 RNA/DNA vs Length</i> | <i>Variable 1</i> | <i>Variable 2</i> |
|-------------------------------|-------------------|-------------------|-------------------------------|-------------------|-------------------|
| Mean                          | 1.20893           | 1.2151925         | Mean                          | 1.7973775         | 1.5309575         |
| Variance                      | 5.917334652       | 6.01165633        | Variance                      | 8.759145923       | 3.383645686       |
| Observations                  | 4                 | 4                 | Observations                  | 4                 | 4                 |
| Pearson Correlation           | 0.999993827       |                   | Pearson Correlation           | 0.956463739       |                   |
| Hypothesized Mean Difference  | 0                 |                   | Hypothesized Mean Difference  | 0                 |                   |
| df                            | 3                 |                   | df                            | 3                 |                   |
| t Stat                        | -0.592719158      |                   | t Stat                        | 0.405264262       |                   |
| P(T<=t) one-tail              | 0.297539508       |                   | P(T<=t) one-tail              | 0.356225993       |                   |
| t Critical one-tail           | 2.353363435       |                   | t Critical one-tail           | 2.353363435       |                   |
| P(T<=t) two-tail              | 0.595079016       |                   | P(T<=t) two-tail              | 0.712451986       |                   |
| t Critical two-tail           | 3.182446305       |                   | t Critical two-tail           | 3.182446305       |                   |

Figure 2-1. Areas sampled Yellow Perch larval presence-absence studies, 2006-2015. Areas sampled in 2015 are highlighted in green. Nanticoke River watershed delineation was unavailable for Delaware and Northeast and was unavailable for Pennsylvania.

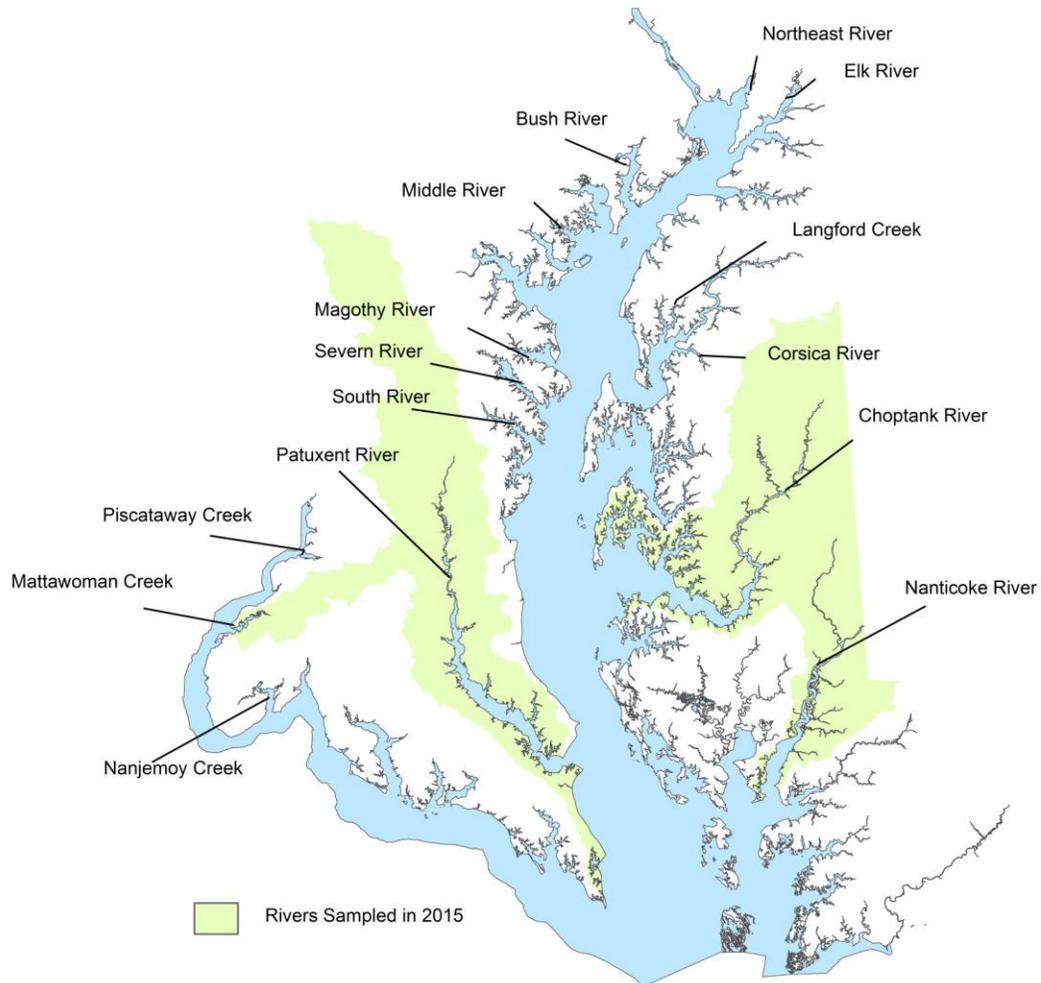


Figure 2-2. Proportion of tows with larval Yellow Perch (*Lp*) and its 95% confidence interval in systems studied during 2015. Mean *Lp* of brackish tributaries indicated by diamond and tidal-fresh mean indicated by dash.

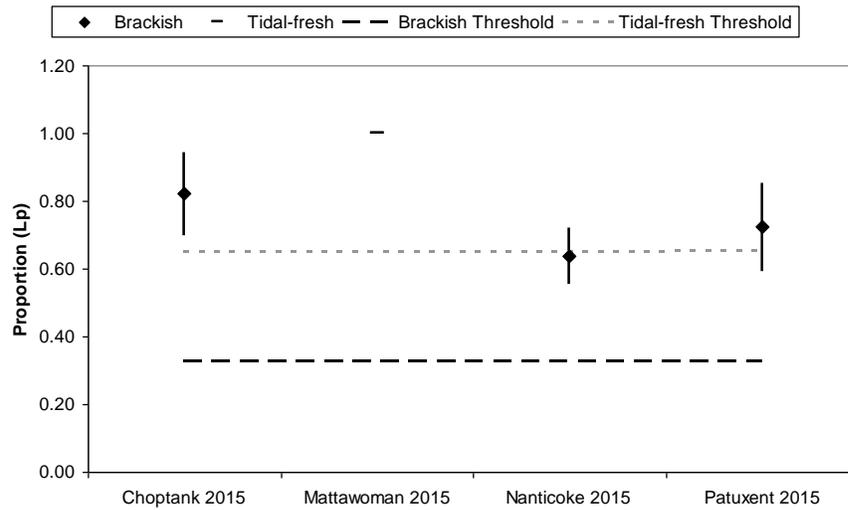


Figure 2-3. Proportion of tows with Yellow Perch larvae (*Lp*) for brackish subestuaries, during 1965-2015. Dotted line provides reference for persistent poor *Lp* exhibited in developed brackish subestuaries.

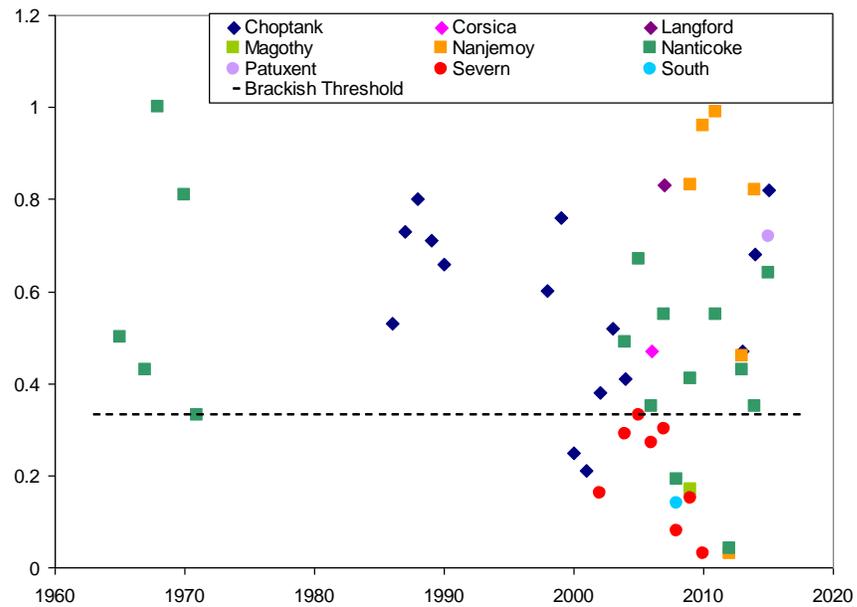


Figure 2-4. Proportion of tows with Yellow Perch larvae (*Lp*) for fresh-tidal subestuaries, during 1990-2015. Dotted line provides reference for consistent poor *Lp* exhibited in a more developed fresh-tidal subestuary (Piscataway Creek).

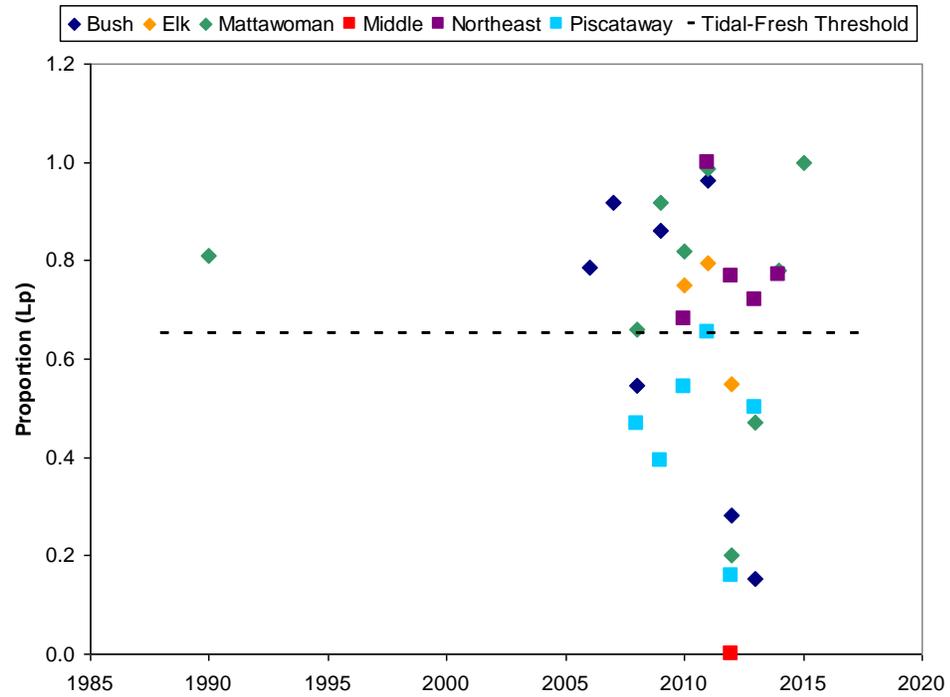


Figure 2-5. Relationship of proportion of plankton tows with Yellow Perch larvae and development (structures per hectare or C/ha) indicated by multiple regression of fresh and brackish subestuaries combined (prediction = MR) and separate linear regressions for both (prediction = LR).

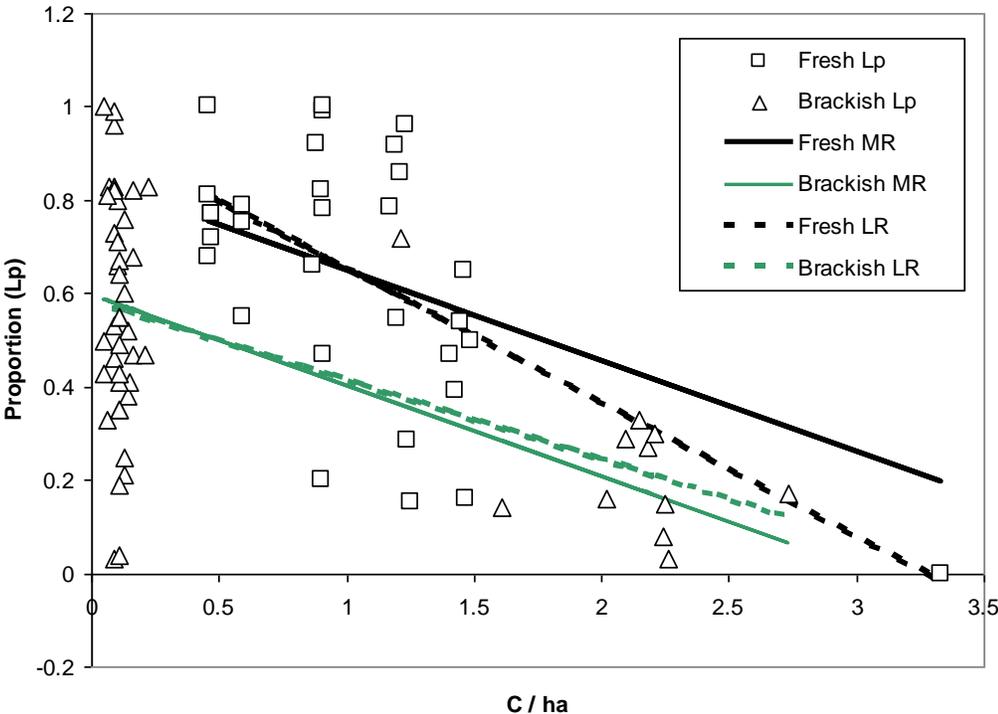


Figure 2-6. Proportion of plankton tows with Yellow Perch larvae plotted against development (C/ha) with Department of Planning land use designations and salinity class indicated by symbols. Diamonds and a “1” behind land use in the key indicate brackish subestuaries, while squares and a “0” indicate tidal-fresh.

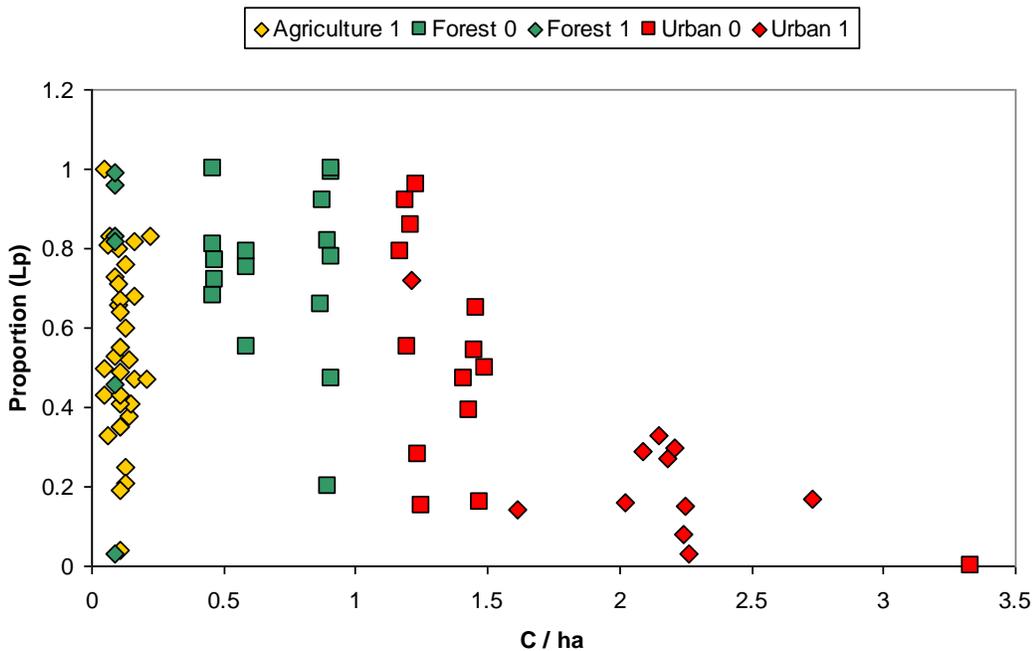


Figure 2-7. Relationship of proportion of plankton tows without detritus (OM0) and development (structures per hectare or C/ha).

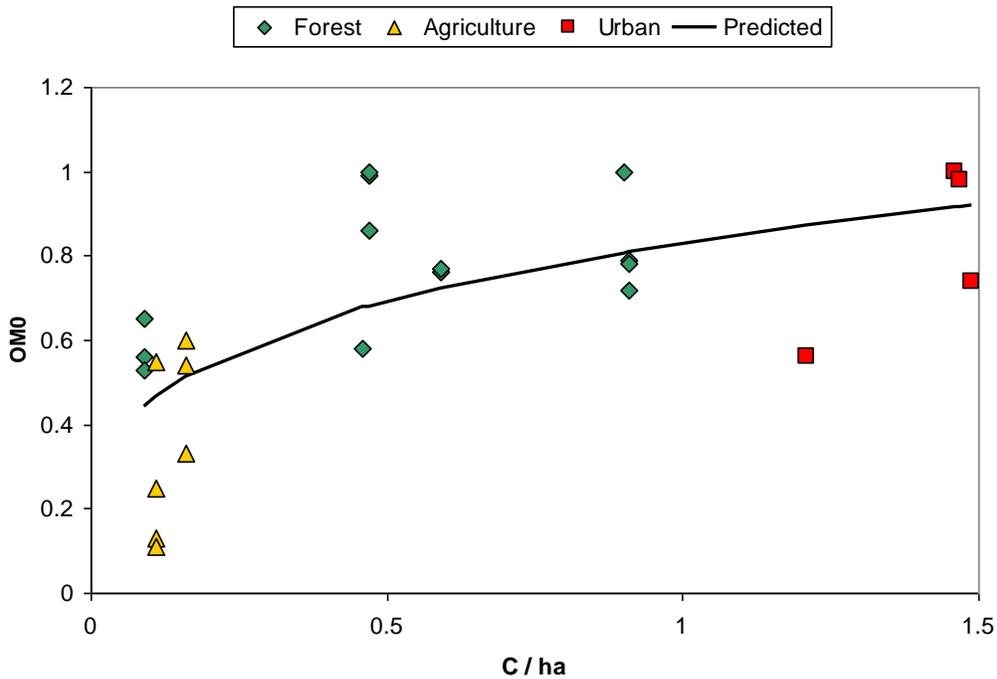


Figure 2-8. Suggested relationship of mean fullness rank of larval Yellow Perch and proportion of plankton tows without detritus (OM0) during 2011-2015.

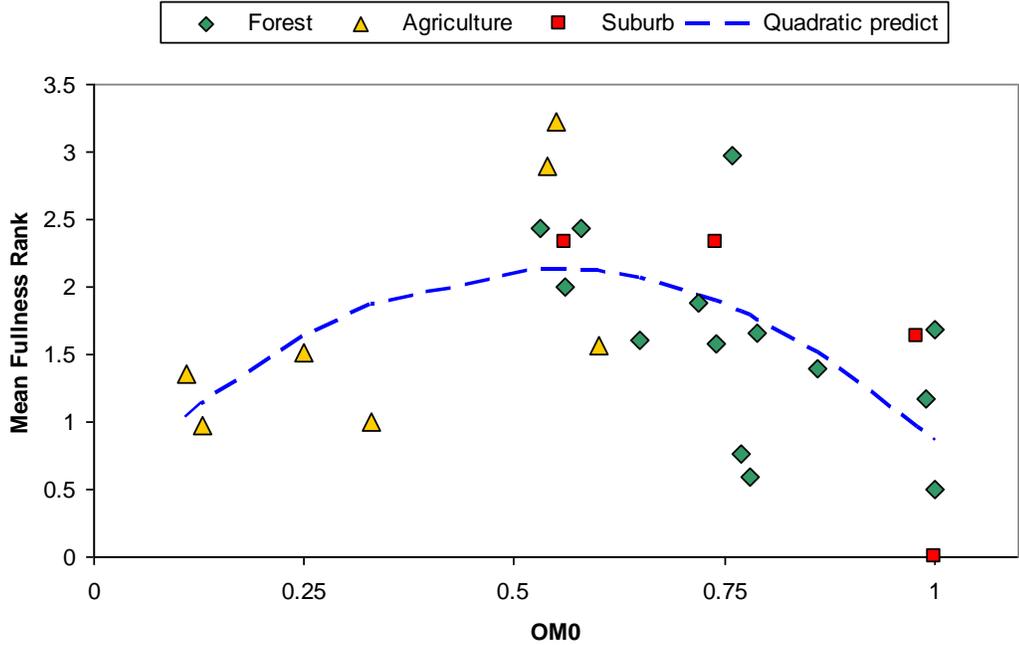


Figure 2-9. (A) Relationship of percent wetlands per watershed obtained from 2010 Department of Planning estimations and level of development (C/ha). (B) Proportion of samples without organic material (OM0) and percent wetlands per watershed.

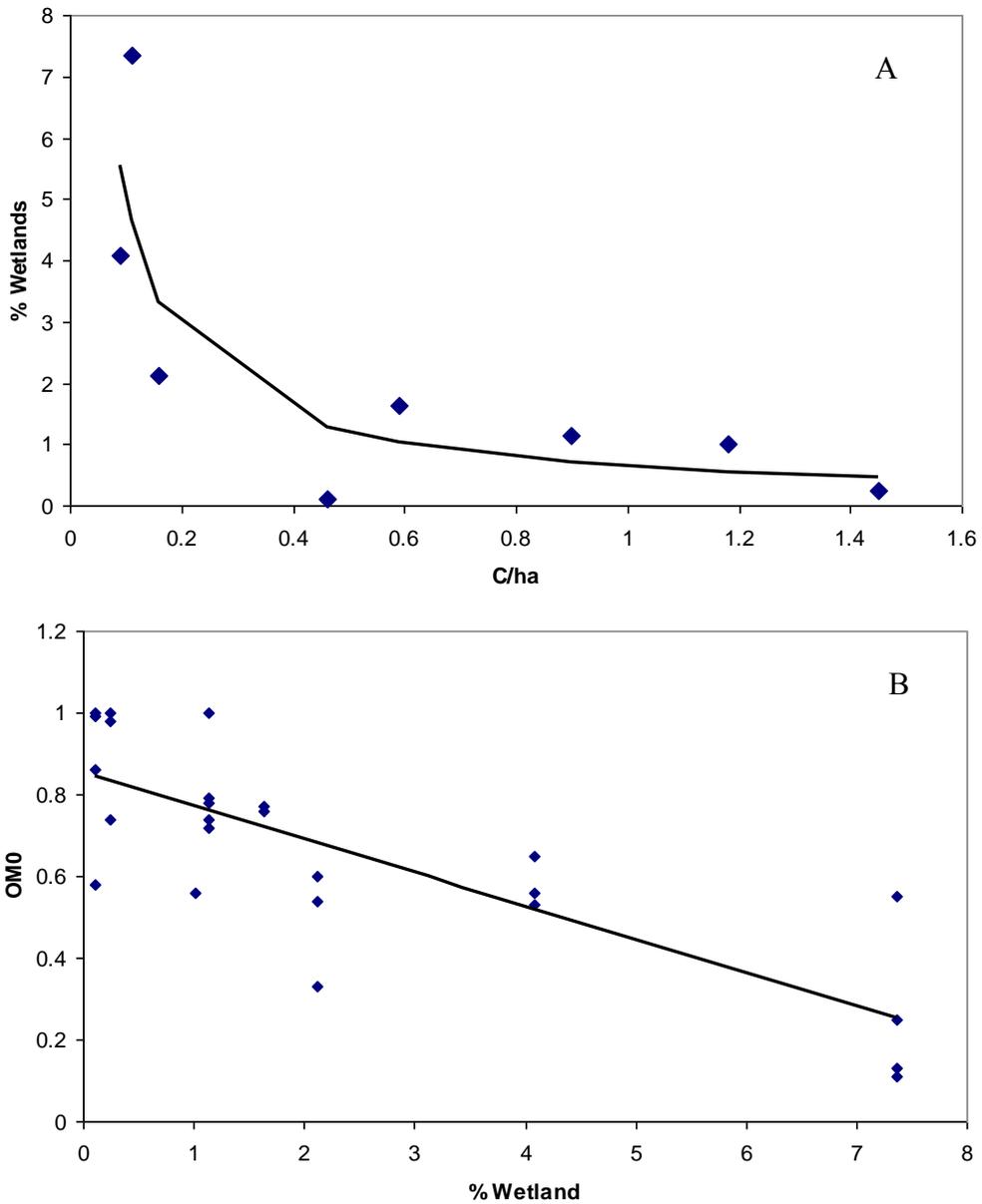
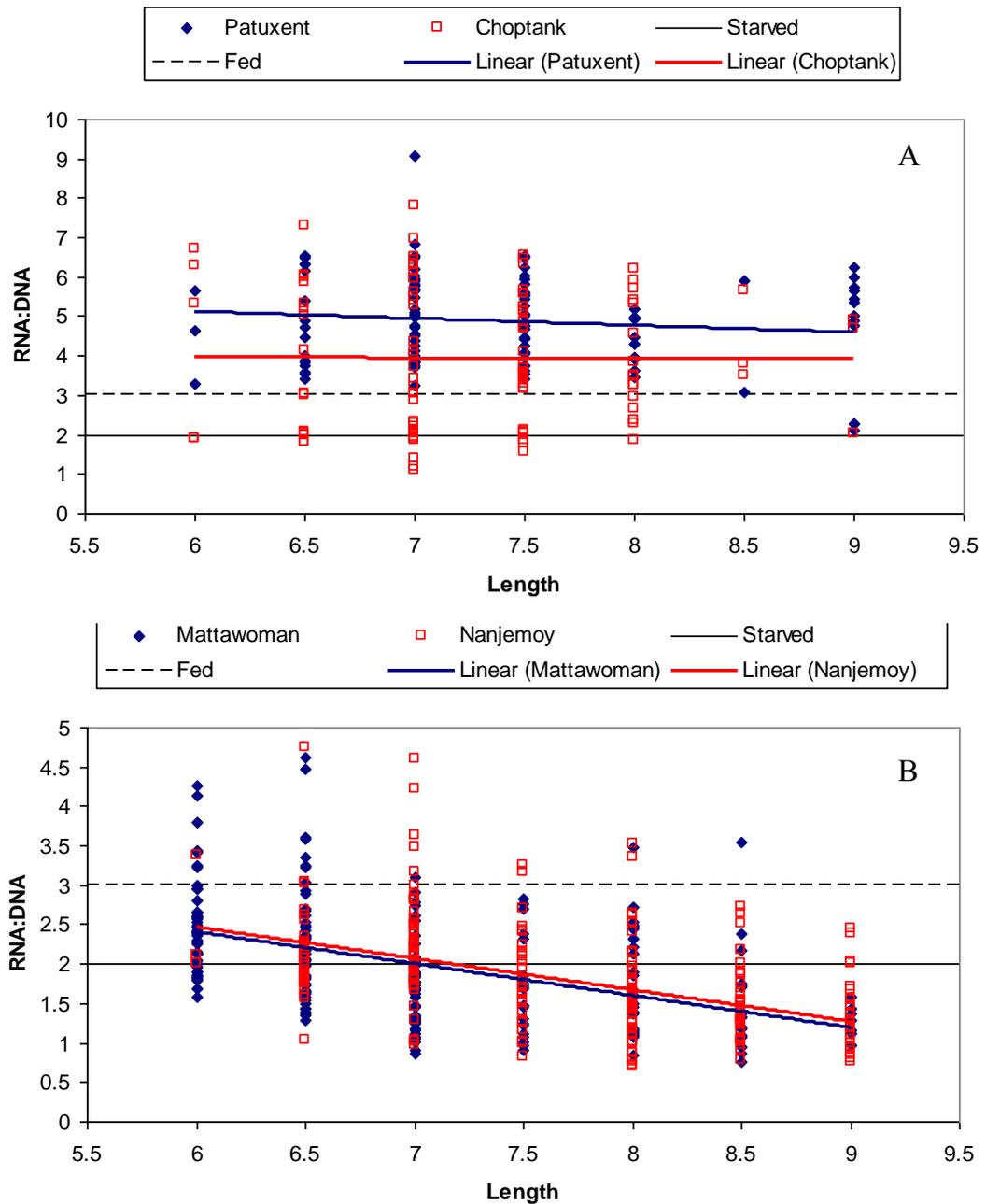


Figure 2-10. RNA/DNA ratios for Yellow Perch larvae by total length. (A) Larvae collected from Patuxent and Choptank Rivers during 2015. (B) Larvae collected from Mattawoman and Nanjemoy Creeks during 2014. Subestuaries are indicated by symbols. Reference lines are provided for ratios indicative of starved and fed conditions.



### **Section 3 - Estuarine Fish Community Sampling**

Alexis Park, Carrie Hoover, Patrick Donovan, Charles Johnson, Margaret McGinty, and Jim Uphoff

#### **Introduction**

Human population growth in the Chesapeake Bay (or Bay) watershed since the 1950s added a suburban landscape layer to the Chesapeake Bay watershed (Brush 2009) that has been identified as a threat to the Bay (Chesapeake Bay Program or CBP 1999). Development converts land use typical of rural areas (farms, wetlands, and forests) to residential and industrial uses (Wheeler et al. 2005; National Research Council or NRC 2009; Brush 2009). These are the basic trade-off in land use facing Maryland as its population grows (Maryland Department of Planning 2015) and they have ecological, economic, and societal consequences (Szaro et al. 1999).

Water quality and aquatic habitat is altered by agricultural activity and urbanization within watersheds. Both land-uses include pesticide and fertilizer application. Agriculturally derived nutrients have been identified as the primary driver of hypoxia and anoxia in the mainstem Chesapeake Bay (or Bay; Hagy et al. 2004; Kemp et al. 2005; Fisher et al. 2006; Brush 2009). Land in agriculture has been relatively stable, but farming itself has become much more intensive (fertilizer and pesticide use has increased) in order to support population growth (Fisher et al. 2006; Brush 2009).

Urbanization may introduce additional industrial wastes, contaminants, stormwater runoff and road salt (Brown 2000; NRC 2009; Benejam et al. 2010) that act as ecological stressors. Extended exposure to biological and environmental stressors affect fish condition and survival (Rice 2002; Barton et al. 2002; Benejam et al. 2008; Benejam et al. 2010). Reviews by Wheeler et al. (2005), the National Research Council (NRC 2009) and Hughes et al. (2014a; 2014b) documented deterioration of non-tidal stream habitat with urbanization.

Development of the Bay watershed brings with it ecologically stressful factors that conflict with demand for fish production and recreational fishing opportunities from its estuary (Uphoff et al. 2011; Uphoff et al 2015). Uphoff et al. (2011a) estimated target and limit impervious surface reference points (ISRPs) for productive juvenile and adult fish habitat in brackish (mesohaline) Chesapeake Bay subestuaries based on dissolved oxygen (DO) criteria, and associations and relationships of watershed impervious surface (IS), summer DO, and presence-absence of recreationally important finfish in bottom waters. Watersheds at a target of 5.5% IS (based on Towson University IS estimates for 1989-2013) or less (rural watershed) maintained mean bottom DO above 3.0 mg / L (threshold DO), but mean bottom DO was only occasionally at or above 5 mg / L (target DO). Mean bottom DO seldom exceeded 3.0 mg / L above 10% IS (suburban threshold; Uphoff et al. 2011a). Although bottom DO concentrations were influenced by development (indicated by IS) in brackish subestuaries, Uphoff et al. (2011b; 2012; 2013; 2014; 2015) have found adequate concentrations of DO in bottom channel habitat of tidal-fresh and oligohaline subestuaries with watersheds at suburban and urban levels of development. They suggested these bottom channel waters were not succumbing to low

oxygen because stratification due to salinity was weak or not present, allowing them to become well mixed.

In 2015, we continued to evaluate summer nursery and adult habitat for recreationally important finfish in tidal-fresh, oligohaline, and mesohaline subestuaries of Chesapeake Bay. In this report, we evaluated the influence of watershed development on target species presence-absence and abundance, total abundance of finfish, and finfish species richness. We evaluated development's effect on three Choptank River subestuaries: Tred Avon River, Harris Creek and Broad Creek. We continued to emphasize Mattawoman Creek in this report as part of Maryland DNRs' effort to assist Charles County with its comprehensive growth plan in order to conserve natural resources of its watershed, including its recreational fisheries (MDDNR 2013).

### **Methods**

We sampled eight subestuaries in Chesapeake Bay during 2015: Broad Creek, Harris Creek, and Tred Avon River, mesohaline tributaries of the Choptank River; Mattawoman Creek and Nanjemoy Creek, fresh-tidal and oligohaline tributaries of the Potomac River; and Northeast River, Middle River, and Gunpowder River, fresh-tidal and oligohaline tributaries located in the upper Chesapeake Bay (Table 3-1; Figure 3-1). This is the fourth year of sampling of Broad Creek and Harris Creek. These watersheds, downstream of Tred Avon River (sampled since 2006), represented rural to near suburban development within a single major watershed (Choptank River; Table 3-1). Harris Creek underwent an extensive Oyster restoration effort in 2014 – 2015 (MD DNR 2014). Two Potomac River tributaries were sampled in 2015; Mattawoman Creek has been sampled since 1989 and Nanjemoy Creek since 2008. Three subestuaries were sampled in upper Chesapeake Bay in 2015: Northeast River (sampled since 2007), Middle River (since 2009), and Gunpowder River (since 2009; Table 3-1).

We obtained compatible data from Bush River monitoring by citizen volunteers and staff from the Anita C. Leight Estuary Center (Bush River; Table 3-1; Figure 3-1). The Bush River has been sampled since 2006; the Estuary Center and its citizen volunteers, trained in 2011 by the Fisheries Service staff, have taken over sampling. We included their data in this report.

Housing density (C/ha) and impervious surface (IS) were estimated for each watershed (Table 3-1). We used property tax map based counts of structures in a watershed, standardized to hectares (C/ha), as our indicator of development (Uphoff et al. 2012; Topolski 2015). This indicator has been provided to us by M. Topolski (MD DNR).

Tax maps are graphic representations of individual property boundaries and existing structures that help State tax assessors locate properties (Maryland Department of Planning or MDP 2013; Topolski 2015). All tax data were organized by county. Since watersheds straddle political boundaries, one statewide tax map was created for each year of available tax data, and then subdivided into watersheds. Maryland's tax maps are updated and maintained electronically as part of MDP's Geographic Information System's (GIS) database. Files were managed and geoprocessed in ArcGIS from Environmental Systems Research Institute (ESRI 2009). All feature datasets, feature classes, and shapefiles were spatially referenced using the

NAD\_1983\_StatePlane\_Maryland\_FIPS\_1900 projection to ensure accurate feature overlays and data extraction. ArcGIS geoprocessing models were developed using ArcGIS Model Builder to automate assembly of statewide tax maps, query tax map data, and assemble summary data. Each year's statewide tax map was clipped using the Maryland 8-digit watershed boundary file to create watershed land tax maps. Watershed area estimates excluded estuarine waters. These watershed tax maps were queried for all parcels having a structure built from 1700 to the tax data year. A large portion of parcels did not have any record of year built for structures but consistent undercounts should not have presented a problem since we were interested in the trend and not absolute magnitude (Uphoff et al. 2012; Topolski 2015).

Uphoff et al. (2012) developed a nonlinear regression equation to convert annual estimates of C/ha to IS calculated by Towson University based on 1999-2000 (years in common) satellite imagery. The relationship of C/ha and IS was well described by the equation:

$$IS = 10.98 (C/ha)^{0.63}, (r^2 = 0.96; P < 0.0001).$$

Estimates of C/ha that were equivalent to 5% IS (target level of development for fisheries; a rural watershed), 10% IS (development threshold for a suburban watershed), and 15% IS (highly developed suburban watershed) were estimated as 0.27, 0.83, and 1.59 C/ha, respectively (Uphoff et al. 2011a; Uphoff et al. 2012).

Property tax data are annually updated by each Maryland jurisdiction to monitor the type of parcel development for tax assessment purposes (Topolski 2015). Detailed records of each structures composition, including the foundation's square footage, are included. Therefore, the tax data can be used to estimate increasing development within a given area: total number of structures (C/ha) and total structure square feet (SQFT / ha). Several watersheds have exhibited a one year downward trend in C/ha: Broad Creek and Mattawoman Creek (2011-2012; shifts of -0.3% and -0.9%, respectively), and Harris Creek (2000-2001; a shift of -2.19%), indicating some annual variability is possible that may be due to duplication or omission of records during annual database development. Determination of the exact cause of the trend shifts requires verification of database records and comparison of specific tax records with corresponding parcel maps within suspect sub-watersheds. The time frame for completion of this analysis exceeds that available for completion of this 2015 Federal Aid Report.

Tidal water surface area of each subestuary was estimated using the planimeter function on MDMerlin satellite photographs and maps ([www.mdmerlin.net](http://www.mdmerlin.net); Table 3-1). Shorelines were traced five-times for each system, and an average area was calculated. The lower limit of each water body was arbitrarily determined by drawing a straight line between the lowest downriver points on opposite shores (the mouth of each system) and the upper limits were to include all waters influenced by tides.

Surveys focused on eleven target species of finfish that fell within four broad life history groups: anadromous (American Shad, Alewife, Blueback Herring, Striped Bass), estuarine residents (White Perch, Yellow Perch), marine migrants (Atlantic Menhaden and Spot), and tidal-fresh forage (Spottail Shiner, Silvery Minnow, Gizzard Shad). With the exception of White Perch, adults of the target species were rare and juveniles were common. Use of target species is widespread in studies of pollution and environmental

conditions (Rice 2003). These species are widespread and support important recreational fisheries in Chesapeake Bay (directly or as forage); they are sampled well by commonly applied seine and-or trawl techniques (Bonzek et al. 2007); and the Bay serves as an important nursery for them (Lippson 1973; Funderburk et al. 1991). Gear specifications and techniques were selected to be compatible with other Fisheries Service surveys.

Ideally, four evenly spaced haul seine and bottom trawl sample sites were located in the upper two-thirds of each subestuary. Nanjemoy Creek was covered sufficiently by three sites. However, during 2011 and 2012, NOAA, who was assisting with sampling, added an additional site in Nanjemoy Creek upstream of our three sites; the data collected during those years were added into all analyses for 2011 and 2012. Broad and Harris creeks lacked shoreline for a fourth comparable seine site, both systems have four trawl sites and three seine sites. Sites were not located near a subestuary's mouth to reduce influence of mainstem waters on fish habitat. We used GPS to record latitude and longitude at the middle of the trawl site, while latitude and longitude at seining sites were taken at the seine starting point on the beach.

Sites were sampled once every two weeks during July-September. All sites on one river were sampled on the same day, usually during morning through mid-afternoon. Sites were numbered from upstream (site 1) to downstream (site 4). The crew leader flipped a coin each day to determine whether to start upstream or downstream. This coin-flip somewhat randomized potential effects of location and time of day on catches and DO. However, sites located in the middle would not be as influenced by the random start location as much as sites on the extremes because of the bus-route nature of the sampling design. If certain sites needed to be sampled on a given tide then the crew leader deviated from the sample route to accommodate this need. Trawl sites were generally in the channel, adjacent to seine sites. At some sites, seine hauls could not be made because of permanent obstructions, SAV beds, or lack of beaches.

Water quality parameters were recorded at all sites. Temperature ( $^{\circ}\text{C}$ ), DO ( $\text{mg} / \text{L}$ ), conductivity ( $\text{mS} / \text{cm}$ ), salinity ( $\text{‰}$ ), and pH were recorded at the surface, middle, and bottom of the water column at the trawl sites and at the surface of the seine site. Mid-depth measurements were omitted at sites with less than 1.0 m difference between surface and bottom. Secchi depth was measured to the nearest 0.1 m at each trawl site. Weather, tide state (flood, ebb, high or low slack), date, and start time were recorded for all sites.

Dissolved oxygen concentrations were evaluated against a target of  $5.0 \text{ mg} / \text{L}$  and a threshold of  $3.0 \text{ mg} / \text{L}$  (Batiuk et al. 2009; Uphoff et al. 2011a). This target DO is considered sufficient to support aquatic life needs in Chesapeake Bay (Batiuk et al. 2009) and has been used in a regulatory framework to determine if a water body is meeting its designated aquatic life uses. The target criterion was associated with asymptotically high presence of target species in bottom channel habitat in brackish subestuaries (Uphoff et al. 2011a). Presence of target species declined sharply when bottom DO fell below the  $3.0 \text{ mg} / \text{L}$  threshold (Uphoff et al. 2011a). In each subestuary, we estimated the percentages of DO samples that did not meet the target or threshold for all samples (surface to bottom) and for bottom waters alone. The percentages of DO measurements that met or fell below the  $5 \text{ mg} / \text{L}$  target ( $V_{\text{target}}$ ) or fell at or below the  $3 \text{ mg} / \text{L}$  threshold ( $V_{\text{threshold}}$ ) were estimated as

$$V_{\text{target}} = (N_{\text{target}} / N_{\text{total}}) * 100;$$

and

$$V_{\text{threshold}} = (N_{\text{threshold}} / N_{\text{total}}) * 100;$$

where  $N_{\text{target}}$  was the number of measurements meeting or falling below 5 mg / L,  $N_{\text{threshold}}$  was the number of measurements falling at or below 3 mg / L, and  $N_{\text{total}}$  was total sample size.

Separate correlation analyses were conducted for surface or bottom temperature or C/ha with surface or bottom DO for all subestuaries sample since 2003. This analysis explored multiple hypotheses related to DO conditions. Structure per hectare estimates were considered proxies for nutrient loading and processing due to development (Uphoff et al. 2011a) in the subestuaries in this analysis. Water temperature would indicate system respiration and stratification influences (Kemp et al. 2005; Murphy et al. 2011). Conducting correlation analyses by salinity classification provided a means of isolating the increasing influence of salinity on stratification from temperature. Our primary interest was in associations of C/ha to DO in surface and bottom channel waters. Temperature and salinity were potential influences on DO because of their relationships with DO saturation and stratification (Kemp et al. 2005; Murphy et al. 2011). We correlated mean surface temperature with mean surface DO, mean bottom temperature with mean bottom DO, and C/ha with surface and bottom DO for each salinity class. We chose annual means of surface or bottom DO and water temperature in summer at all sites within a subestuary for analyses to match the geographic scale of C/ha estimates (whole watershed) and characterize chronic conditions.

Conductivity measurements were collected at each site in every system from July to September. Conductivity measurements recorded in 2012-2013 were recorded incorrectly. The raw conductivity was recorded instead of the specific conductivity, which compensates for temperature. An equation was used to correct the error and convert the raw conductivity measurements that were recorded to specific conductivity (Fofonoff and Millard 1983):

$$\text{Specific Conductivity} = \text{Conductivity} / (1 + ((0.02 \cdot T) - 25));$$

for each °C change in water temperature (T) there was a 2% change in conductivity.

Each subestuary was classified into a salinity category based on the Venice System for Classification of Marine Waters (Oertli, 1964). Salinity influences distribution and abundance of fish (Hopkins and Cech, 2003; Cyrus and Blaber, 1992; Allen, 1982) and DO (Kemp et al. 2005). Uphoff et al. (2012) calculated an arithmetic mean of all bottom salinity measurements over all years available to determine salinity class for each subestuary. Tidal-fresh ranged from 0-0.5 ‰; oligohaline, 0.5-5.0 ‰; and mesohaline, 5.0-18.0 ‰ (Oertli, 1964). Mattawoman Creek, Bush River, and Northeast River were classified as tidal-fresh subestuaries (Table 3-1). Gunpowder River, Middle River, and Nanjemoy Creek were considered oligohaline. Broad Creek, Harris Creek, and Tred Avon River, were mesohaline subestuaries (Table 3-1). We grouped data by these classifications when examining effects of development.

A 4.9 m headrope semi-balloon otter trawl was used to sample fish in mid-channel bottom habitat. The trawl was constructed of treated nylon mesh netting

measuring 38 mm stretch-mesh in the body and 33 mm stretch-mesh in the codend, with an untreated 12 mm stretch-mesh knotless mesh liner. The headrope was equipped with floats and the footrope was equipped with a 3.2 mm chain. The net used 0.61 m long by 0.30 m high trawl doors attached to a 6.1 m bridle leading to a 24.4 m towrope. Trawls were towed in the same direction as the tide. The trawl was set up tide to pass the site halfway through the tow, allowing the same general area to be sampled regardless of tide direction. A single tow was made for six minutes at 3.2 km / hr (2.0 miles / hr) per site on each visit. The contents of the trawl were emptied into a tub for processing.

During 2009-2015, a 3.1 m box trawl made of 12.7 mm stretch-mesh nylon towed for five minutes was used on the same day sampling was conducted with a 4.9 m trawl in Mattawoman Creek to create a catch-effort time-series directly comparable to monitoring conducted during 1989-2002 (Carmichael et al. 1992). The net to start with on each day in Mattawoman Creek alternated between sampling days.

An untreated 30.5 m 1.2 m bagless knotted 6.4 mm stretch mesh beach seine, the standard gear for Bay inshore fish surveys (Carmichael et al. 1992; Durell 2007), was used to sample inshore habitat. The float-line was rigged with 38.1 mm by 66 mm floats spaced at 0.61 m intervals and the lead-line rigged with 57 gm lead weights spaced evenly at 0.55 m intervals. One end of the seine was held on shore, while the other was stretched perpendicular to shore as far as depth permitted and then pulled with the tide in a quarter-arc. The open end of the net was moved towards shore once the net was stretched to its maximum. When both ends of the net were on shore, the net was retrieved by hand in a diminishing arc until the net was entirely pursed. The section of the net containing the fish was then placed in a washtub for processing. The distance the net was stretched from shore, maximum depth of the seine haul, primary and secondary bottom type, and percent of seine area containing aquatic vegetation were recorded.

All fish captured were identified to species and counted. Striped Bass and Yellow Perch were separated into juveniles and adults. White Perch were separated into three categories (juvenile, small adults and harvestable size) based on size and life stage. The small adult White Perch category consisted of ages-1+ White Perch smaller than 200 mm. White Perch greater than or equal to 200 mm were considered to be of harvestable size and all captured were measured to the nearest millimeter. White Perch of this size or larger corresponded to the quality length category minimum (36-41% of the world record TL) proposed by Anderson (1980) for proportional stock density (PSD) indices; 200 mm TL is used as the length cut-off for White Perch in Chesapeake Bay assessments of White Perch (Piavis and Webb 2013). Small adult and harvestable White Perch were combined for adult counts. Catch data were summarized and catch statistics were reported for both gears combined and each gear separately.

Three basic metrics of community composition were estimated for subestuaries sampled: geometric mean catch of all species, total number of species (species richness), and species comprising 90% of the catch. The geometric mean (GM) was estimated as the back-transformed mean of  $\log_e$ -transformed catches (Ricker 1975; Hubert and Fabrizio 2007). The GM is a more precise estimate of central tendency of fish catches than the arithmetic mean, but is on a different scale (Ricker 1975; Hubert and Fabrizio 2007). We noted which target species were within the group that comprised 90% of fish collected. We summarized these metrics by salinity type since some important ecological

attributes (DO and high or low SAV densities) appeared to reflect salinity class (Uphoff et al. 2012).

We plotted species richness collected by seine and by 4.9 m trawl against C/ha by salinity class. A greater range of years (1989-2015) was available for seine samples than the 4.9 m trawl (2004-2015) due to a change from the 3.1 m trawl used during 1989-2002 (Carmichael et al. 1992). We set a minimum number of samples (15) for a subestuary in a year to include estimates of species richness based on analyses in Uphoff et al. (2014). This eliminated years where sampling in a subestuary had to be ended due to site losses, typically from SAV growth that did not permit sampling throughout a season.

We discovered an error in Nanjemoy Creek data during 2011-2012 due to station identifications being switched; stations were correctly identified using the latitudes-longitudes recorded at the time of sampling and matched to the correct sites. Errors were also discovered in Mattawoman 2009-2011 trawl data, the 3.1 m box trawls and 4.9 m headrope semi-balloon otter trawls were not correctly identified in entered data; trawls were correctly identified from field data sheets.

We continued to track bottom DO, SAV area, finfish abundance and finfish species richness in 3.1 m and 4.9 m trawl samples from Mattawoman Creek and compared them to changes in C/ha. Estimates for SAV area are one year delayed due to the availability of the data, the data is not available till after the submission of this report.

We obtained measurements of total ammonia nitrogen (TAN;  $\text{NH}_3$  plus  $\text{NH}_4$ ; US EPA 2013), chlorophyll a, and pH in Mattawoman Creek during the SAV growing season (April-October) from Chesapeake Bay Program (CBP; 2016) monitoring site MAT0016, located in the channel between our stations 3 and 4 (W. Romano, MD DNR, personal communication). The TAN estimates were available for 1986-2015, but we eliminated 1986-1990 from analysis because of methodology differences. During 1991-2009, TAN samples were collected twice a month, only the first TAN sample of each month was selected for analysis (except in 1991, the first sample during May was not available so the second sample was selected) to correspond equally with the number of samples in the following years 2010-2015 (N=7). In 2014, only 6 TAN samples were used in analysis because samples were not collected in July. Measurements of growing season TAN were annually summarized as minimum, median, and maximum and compared to US EPA ambient water quality criteria for TAN (US EPA 2013) to capture the potential for acute and chronic toxicity.

Sampling with 3.1 m trawls was conducted during 1989-2002 and 2009-2015 and 4.9 m trawls have been used since 2003. Geometric means of total fish abundance and their 95% CI's were estimated for the 3.1 m and 4.9 m trawls for samples from Mattawoman Creek. We compared trends of GMs of total fish abundance in the years in common for the 3.1 m and 4.9 m trawls in Mattawoman Creek using linear regression.

Estimates of species richness in Mattawoman Creek (number of species encountered) were made for 3.1 m trawl samples during 1989-2002 and 2009-2015. Sampling during 1989-2002 was based on monthly sampling of five stations (Carmichael et al. 1992). Station 5, the furthest downstream station sampled during 1989-2002, was dropped because it was outside the range of stations 1-4 sampled during 2009-2015. Remaining stations were the same throughout the time-series, but were sampled monthly

during 1989-2002 (annual N = 12) and bi-monthly during 2009-2015 (annual N = 24). In order to match the annual sample sizes of 1989-2002, we made two sets of estimates for each sample year during 2009-2015: one for the first round of the month and one for the second. As a result, all comparisons of species richness in Mattawoman Creek were based on the same annual sample size.

We analyzed the trends in DO among the Choptank tributaries, Broad Creek, Harris Creek, and Tred Avon River, in 2015. We obtained land use data from the Maryland Department of Planning (MD DOP 2010); the most recent land cover data available by MD DOP is based on 2010 data. Five categories of land use were presented for all three Choptank tributaries, agriculture, forest, urban, wetlands, and water. The first four categories contain only land portion of the watershed, the water category is the amount of water in each watershed. The land use categories of barren and roads were not available. The trajectory of C/ha for each of the three tributaries since 1950 was plotted. A scatter plot depicted the effect of C/ha intensity on bottom DO. The percentage of target and threshold violations were estimated using all DO measurements (surface, middle, and bottom) and bottom DO measurements. Annual mean bottom DO at each site during 2006–2015 summer sampling was estimated and plotted. These annual, site-specific means were compared to the median bottom DO during 2006 – 2015. One-way ANOVAs were used to analyze for differences in bottom DO among stations in the three Choptank River tributaries. ANOVA's were run separately for Harris Creek, Broad Creek, and Tred Avon River. If the ANOVA indicated significant differences ( $P \leq 0.05$ ), a Tukey Studentized Range or Tukey Honestly Significant Difference (HSD) Test (SAS Enterprise 5.1) was used to detect differences in bottom DO readings among sites.

Individual total lengths (TL) of White Perch (>200 mm TL) that should be of interest to anglers have been collected during trawl and seine sampling since 2004. White Perch of this size or larger corresponded to the quality length category minimum (36-41% of the world record TL) proposed by Anderson (1980) for proportional stock density (PSD) indices; 200 mm TL is used as the length cut-off for White Perch in Chesapeake Bay stock density indices (Piavis and Webb 2013). These data collected in the three Choptank River tributaries provided an opportunity to evaluate the influence of development on the availability of fish for anglers to harvest. Uphoff et al. (2014) found that White Perch of a size of interest to anglers were more likely to be found in mesohaline subestuaries with rural or transition watersheds and least likely to be found in subestuaries with suburban-urban subestuaries.

Proportions of White Perch of quality length or greater ( $\geq 200$  mm) and their 95% CIs were calculated for each of the three Choptank River subestuaries and compared to their watershed development and DO conditions. Trawl and seine samples were pooled, as were years in common (2012-2015). The proportion of samples with quality length or greater White Perch was estimated as  $P_{Qwp} = N_{Quality} / N_{total}$ ; where  $N_{Quality}$  equaled the number of samples with quality length or greater White Perch present and  $N_{total}$  equaled the total number of White Perch in the ages 1+ and harvestable categories. The SD of each  $P_{Qwp}$  was estimated as

$$SD = [(P_{Qwp} \cdot (1 - P_{Qwp})) / N_{total}]^{0.5} \text{ (Ott 1977).}$$

The 95% confidence intervals were constructed as  $P_{Qwp} \pm (1.96 \cdot SD)$ .

## Results and Discussion

Broad Creek, Harris Creek, Gunpowder River, and Mattawoman Creek did not have DO readings less than the target level (5.0 mg / L) during 2015 (Table 3-2). During 2015, a mix of fresh-tidal, oligohaline, and mesohaline subestuaries had non-zero estimates of  $V_{target}$  in surface and bottom waters. Eight percent of all DO measurements from Tred Avon River were below the target ( $V_{target} = 8.3\%$ ); Middle River, 7.5%; Nanjemoy Creek, 3.9%; Bush River, 4.0%; and Northeast River, 4.7%. When we evaluated  $V_{target}$  in bottom channel waters, Middle River had 20.8%; Nanjemoy Creek, 20.0%; Northeast River, 16.7%; and Tred Avon, 33.3%. All other subestuaries had  $V_{target}$  estimates of zero. In 2015, two subestuaries had measurements of bottom DO below the 3 mg / L threshold (Table 3-2): Tred Avon River at 12.5% and Northeast River at 4.2%.

Data used for correlation analyses of DO with temperature at depth and C/ha are presented in Table 3-3. Overall, these correlation analyses suggested that DO responded to temperature at depth and C/ha differently depending on salinity classification. Negative associations of surface or bottom DO with corresponding mean water temperatures at depth were detected for oligohaline subestuaries by correlation analyses (surface,  $r = -0.34$ ,  $P < 0.05$  and bottom,  $r = -0.52$ ,  $P < 0.002$ ), suggesting respiration was a major consideration in this class of subestuary (Table 3-4). Associations of temperature and DO were not detected in mesohaline or tidal-fresh subestuaries. The strongest and only negative association between bottom DO and C/ha was found in mesohaline subestuaries ( $r = -0.58$ ,  $P < 0.0001$ ), where strongest stratification was expected. Positive associations of surface DO with development were suggested for fresh-tidal and oligohaline subestuaries ( $r = 0.35$ ,  $P = 0.05$  and  $r = 0.37$  and  $P = 0.03$ , respectively). A positive association of was found for bottom DO and C/ha in fresh-tidal subestuaries as well ( $r = 0.42$ ,  $P < 0.02$ ); an association was not suggested for bottom DO and C/ha in oligohaline subestuaries. Given that multiple comparisons were made, these positive correlations that were significant at  $P \leq 0.02$  for might be considered spurious if one rigorously adheres to significance testing (Nakagawa 2004). However, these two classifications were less likely to stratify because of low or absent salinity and the biological consequences of no or positive relationships would be similar (i.e., a negative impact on habitat would be absent). Sample sizes of mesohaline subestuaries ( $N = 59$ ) were over twice as high as oligohaline ( $N = 34$ ) or tidal-fresh subestuaries ( $N = 31$ ), so ability to detect significant associations in mesohaline subestuaries was greater (Table 3-4).

During 2015, dense SAV prevented seining in Mattawoman Creek. Seining in Middle River was very restricted because of high tides that limited beach availability and dense SAV in seine sites; only two seine sites were available. Additional seine sites sampled in Middle River and Nanjemoy Creek for NOAA's Integrated Assessment were dropped since NOAA terminated field collections. In Gunpowder River, one seine site (Site 2) was not sampled at all after it was roped off for swimming.

Geometric means of seine hauls ranged from 147 to 442 fish during 2015, with little indication that salinity class or development level exerted an influence (Table 3-4). Subestuaries had 17-24 samples, except for Middle River with only 3 samples. Seining results are summarized for Middle River, but too few samples were made for meaningful comparisons with other systems. Nanjemoy Creek, an oligohaline tributary, had the greatest number of species (29) during 2015. The tidal fresh systems had the 2<sup>nd</sup> and 3<sup>rd</sup>

highest number of species, 24 and 25, respectively. The remaining oligohaline subestuary had 22 species. The three mesohaline subestuaries had 23, 22, and 20 species (Table 3-5).

A total of 47,264 fish representing 47 species were captured by beach seine in 2015 (Table 3-5). Nine species comprised 90% of the total fish caught in 2015, including Atlantic Silverside, White Perch, Atlantic Menhaden, Striped Killifish, Blueback Herring, Spottail Shiner, Gizzard Shad, Banded Killifish, Mummichog, and Alewife. White Perch, Gizzard Shad, Blueback Herring, Spottail Shiner, Alewife, and Atlantic Menhaden represented target species among the species comprising 90% of the total catch (Table 3-5). Eight target species were present among species comprising 90% of the seine catch (dominant species) when viewed by subestuary; White Perch were present in six of the eight subestuaries seined; Blueback Herring in four; Spottail Shiner, Gizzard Shad, and Alewife in three; Yellow Perch, American Shad, and Atlantic Menhaden in one (Table 3-5). White perch were present in the top 90% of species for all systems except Broad Creek. Target species comprised 90% of the catch in Bush River. However, Bush River volunteers were unable to identify some of Blueback Herring or Alewife sampled to species and classified them as “Herrings Unidentified”. A larger number of Alosid species were sampled during 2015 and were present in all the top 90% of species in all subestuaries except Middle and Tred Avon Rivers (Table 3-4).

Bottom trawling with a 4.9 m headrope trawl was conducted in all nine subestuaries in 2015. A total of 56,338 fish and 42 fish species were captured (Table 3-6). Five species comprised 90% of the total catch for 2015, White Perch (juveniles and adults), Bay Anchovy, Spottail Shiner, Alewife and Pumpkinseed. White Perch, Alewife, and Spottail Shiner were target species (Table 3-6). Blueback Herring was in the top 90% in two subestuaries and Alewife in one subestuary.

Geometric mean trawl catches during 2015 were between 40 and 416 (Table 3-6). Subestuaries had 18-24 samples; except Bush River, which had 14. Number of species captured by trawl in subestuaries sampled during 2015 (14-23) overlapped for all three salinity classifications (Table 3-5).

White Perch (juveniles) were among species comprising 90% of 4.9 m trawl catches in 6 of the 9 subestuaries (Table 3-6). Bay Anchovy were the most frequent species comprising 90% in 8 subestuaries. Nanjemoy Creek had the highest GM (416) and the Harris Creek had the lowest GM (40; Table 3-6).

Plots of species richness in seine samples and C/ha did not suggest a relationship in tidal-fresh or oligohaline, or mesohaline subestuaries (Figure 3-2); tidal-fresh subestuary watersheds were represented by a limited range of C/ha (0.43 - 0.72). Oligohaline subestuary watersheds were represented by the widest range of C/ha (0.09 - 3.33, rural to urban) of the three salinity classes.

Plots of species richness in trawl samples and C/ha did not suggest a relationship for tidal-fresh or oligohaline subestuaries (Figure 3-3). Species richness declined in mesohaline subestuaries as C/ha advanced beyond the threshold ( $C/ha = 0.83$ ). Eleven or less species were present in mesohaline subestuaries when C/ha was over the threshold. Species richness fell to this level less frequently at lower levels of development (Figure 3-3).

In general these exploratory analyses of species richness and development supported trends found in analyses of development and DO (Figures 3-4 and 3-5). Levels of bottom DO were not negatively influenced by development in tidal-fresh or oligohaline subestuaries, but were in mesohaline subestuaries (Table 3-4).

Depletion of bottom DO in mesohaline subestuaries to hypoxic or anoxic levels represented a direct loss of habitat to be occupied. Uphoff et al. (2011a) determined that the odds of adult and juvenile White Perch, juvenile Striped Bass, Spot, and Blue Crabs being present in seine samples from mesohaline subestuaries were not influenced by development, but odds of target species being present in bottom channel trawl samples were negatively influenced by development.

The extent of bottom channel habitat that can be occupied does not appear to diminish with development in tidal-fresh and oligohaline subestuaries due to low DO. Sampling of DO in dense SAV beds in tidal-fresh Mattawoman Creek in 2011 indicated that shallow water habitat could be negatively impacted by low DO within the beds (Uphoff et al. 2012; 2013; 2014; 2015); it was not feasible for us to sample fish within the beds so the impact on target finfish could not be estimated. The summer fish community of tidal-fresh Mattawoman Creek underwent drastic changes in abundance and species richness as development threshold was approached that were unrelated to adequacy of DO in channel waters, indicating other stressors (see below) were important (Uphoff et al. 2009; 2012; 2013; 2014; 2015). During November, 2015, the oligohaline Middle River subestuary (the most heavily developed watershed in our study) experienced an extensive fish kill attributable to harmful algal blooms (MDE 2016). Middle River has exhibited a diverse and abundant fish community over the course of our monitoring.

The level of development in Mattawoman Creek's watershed more than doubled between 1989 (0.43 C/ha) and 2011 (0.91 C/ha; Figure 3-6). This watershed reached the threshold for suburban development (C/ha = 0.83) in 2006.

There appeared to be two periods of bottom DO in the Mattawoman Creek time-series (Figure 3-7). Mean bottom DO was near or above the median for the time-series (8.4 mg / L) during 1989-2000 (C/ha  $\leq$  0.67) and then fell below the median afterward (with the exceptions of 2003, 2013 and 2014). Mean bottom DO in 2015 was 7.7 mg / L. Mean bottom DO during summer sampling has never fallen below the target of 5.0 mg / L (Figure 3-7) and excursions below this level have been rare. These shifts in bottom DO corresponded to changes in Mattawoman Creek's subestuary chlorophyll a levels ( $r^2 = 0.4869$ ,  $P < 0.0001$ ,  $N = 27$ ) from high (16-40  $\mu\text{g} / \text{L}$ ) to low ( $< 15 \mu\text{g} / \text{L}$ ; Figure 3-8) and shift in SAV acreage from low (coverage  $\leq 10\%$  of water area) to high (coverage  $> 30\%$ ; Figure 3-9; Uphoff et al. 2011b; 2012; 2013; 2014; 2015).

The TAN measurements collected by Chesapeake Bay Program (2016) at MAT0016 ranged from 0.005 mg / L to 0.049 mg / L and had a median of 0.009 mg / L during 2015 (Figure 3-10). The median TAN (mg/L) readings from 1990 - 2014 significantly correspond to the percentage of SAV coverage in Mattawoman Creek ( $r^2 = 0.448$ ,  $P = 0.0003$ ,  $N = 24$ ; Figure 3-11). Data points for 2001 were omitted due to incomplete mapping of SAV. Data for SAV coverage in Mattawoman Creek for 2015 was not available at the time of this report. Measurements of pH at MAT0016 during

April – October dropped and became more variable after the late 1990s (Figure 3-12). Median pH levels have fluctuated between 7.70 and 8.24 since 2006. During 2007 and 2010, pH reached the highest levels observed (9.88 and 9.84, respectively; Figure 3-12). Levels of pH greater than 9 promotes ammonium ( $\text{NH}_4^+$ ) to change to toxic ammonia nitrogen ( $\text{NH}_3$ ), causing fish kills (Randall and Tsui 2002; US EPA 2013).

Geometric mean catches for 3.1 m and 4.9 m trawls in Mattawoman Creek are presented in Figure 3-13. The linear regression of GM catches of 4.9 m and 3.1 m trawls during 2009-2015 indicated that their trends were closely and linearly related ( $r^2 = 0.99$ ,  $P < 0.0001$ ,  $N = 7$ ). The slope estimate and its SE were 0.354 and 0.018, respectively. The intercept estimate and its SE were -5.20515 and 4.78617 (not different from 0 based on its 95% CI). We predicted the missing portion of the 3.1 m trawl GM time-series from the slope alone (Figure 3-13). The span of GMs in the regression was similar to those that were predicted, so values did not have to be extrapolated beyond limits of data. The full 3.1 m GM time-series (observations and predictions) suggested abundance of all species became much more variable after 2001. During 1989-2002, minimum, maximum, and median GM catches of 3.1 m for all species were 12, 108, 50, respectively; during 2003-2015, minimum, maximum, and median GM catches of all species (predictions for missing years included) in 3.1 m trawls were 0, 200, 46, respectively (Figure 3-13).

During 1989-2002, minimum, maximum, and median number of species collected annually in 3.1-m trawls were 8, 19, and 14 respectively; during 2009-2015, minimum, maximum, and median were 5, 20, and 11, respectively (Figure 3-14). Species comprising 90% of the catch (dominant species) changed between 1989-2002 and 2009-2015 (Figure 3-15a). Young-of-year White Perch were usually a significant fraction of catch during both periods, but adult White Perch were not a dominant species after 2001. Planktivorous Blueback Herring, Alewife, Gizzard Shad, and Bay Anchovy, dominant during 1989-2002, were replaced by Spottail Shiners. The percentage contribution of YOY White Perch and Spottail Shiners were similar during 2013 – 2015 (Figure 3-15a).

White Perch (YOY) and Spottail Shiners became the only target species in Mattawoman Creek to qualify as dominant in 4.9 m trawls after 2011 (Figure 3-15b). Since 2003, planktivores have been uncommon and adult White Perch have dropped out of the dominant species category. Species comprising of 90% of catch since 2011 have primarily been YOY White Perch and Spottail Shiners. This past year, 2015, was the first year since 2012 that a planktivore, Blueback Herring, was in the top 90% and that more than two species comprised 90% of species collected (Figure 3-15b).

Mattawoman Creek's finfish abundance appeared to exhibit boom and bust dynamics after 2001. "Busts" were concurrent with spikes (2002) or plateaus (2007-2009) of TAN (Figure 3-10). Collapses of the magnitude exhibited during 2002 and 2008-2009 were not detected previously (Figure 3-13). Uphoff et al. (2010) determined that the collapse of abundance in 2008-2009 was local to Mattawoman Creek and not widespread in the Potomac River. Recovery of fish abundance since 2011 has coincided with moderate values of median TAN.

Shifts in ecosystem status observed in Mattawoman Creek may represent shifts to different unstable or stable states (shifting baselines or regime shifts, respectively) of ecological systems rather than steady declines (Steele and Henderson 1984; Duarte et al.

2009). The term “regime shift” has been used to suggest jumps between alternative equilibrium states are nonlinear, causally connected, and linked to other changes in an ecosystem (Steele 1996; Duarte et al. 2009). The regime shift concept implies that different regimes have inherent stability, so that significant forcing is required to flip the system into alternative states (Steele 1996). Eutrophication is one of these forcing mechanisms (Duarte et al. 2009), while urbanization creates a set of stream conditions (urban stream syndrome; Hughes et al. 2014a; 2014b) that qualifies as a shift as well. Both of these processes (eutrophication and urban stream syndrome) are inter-related products of development in Mattawoman Creek’s watershed. Sediment loads in Mattawoman Creek from construction and stream bank erosion were high (Gellis et al. 2009) and increased nutrient loading there was strongly associated with sediment level increases that occurred after 2003 (J. Uphoff, MDDNR, unpublished analysis of USGS data obtained by W. Romano, MDDNR). Approaching and breaching the development threshold in Mattawoman Creek’s watershed has been concurrent with changes in stream hydrology and water quality, increased sediment and nutrient loading from stream erosion and construction, decreased chlorophyll a (a powerful indicator of ecosystem response to nutrients; Duarte et al. 2009) and DO. Water clarity has increased, as has TAN and SAV while finfish abundance has become more variable and less diverse (particularly planktivores) in the subestuary (Gellis et al. 2009; Uphoff et al. 2009; 2010; 2011b; 2012; 2013; 2014; 2015). When evaluated in the context of Chesapeake Bay Program’s habitat goals, Mattawoman Creek superficially resembles a restored system with reduced nutrient loads, i.e., increased clarity, reduced chlorophyll a, and increased SAV. Together, these factors were expected to increase habitat for fish (Chesapeake Bay Program 2014). However, Chanat et al. (2102) reported that nutrient and sediment loads in Mattawoman Creek were nearly twice those of the Choptank River, an agriculturally dominated watershed twice the size of Mattawoman Creek. Boyton et al. (2012) modeled nutrient inputs and outputs in Mattawoman Creek and found that nutrients were not exported out of the subestuary, suggesting that wetlands, emergent vegetation, and SAV in Mattawoman Creek were efficiently metabolizing and sequestering nutrients. The fish community has become highly variable and less diverse under these conditions. Duarte et al. (2009) analyzed responses of phytoplankton of four coastal ecosystems to eutrophication and oligotrophication and found diverse, idiosyncratic responses. An expectation that ecosystems would revert to an expected reference condition was unsupported (Duarte et al. 2009). During 2014, we further explored a hypothesis that water quality dynamics in Mattawoman Creek’s extensive SAV beds (low DO, high pH, and high organic matter) may be creating episodes of ammonia toxicity for fish with a 24-hour study in a single SAV bed (Uphoff et al. 2014). This study suggested that fish could be caught in a habitat squeeze in SAV from high ammonia at the surface and low DO at the bottom (Uphoff et al. 2014).

In 2015, we explored DO trends in mesohaline Broad Creek, Harris Creek, and Tred Avon River, subestuaries of the lower Choptank River. These adjacent watersheds have undergone development at different levels, with two having just passed the target level of development and one approaching the threshold (Figure 3-16). Our past surveys (Uphoff et al. 2011a) have, by necessity, substituted comparisons of rural and well developed watersheds for time-series of development related changes. We have monitored the Tred Avon in anticipation of measuring DO and fish community changes

in a mesohaline subestuary as its watershed develops over time and contrast it with less developed watersheds in the same region.

Similar in percentages of land in agriculture (43-45%), forest (20-25%), and urban (30-34%) categories were estimated in these three Choptank River tributaries by the Maryland Department of Planning (MD DOP) (Table 3-7; Figure 3-16; MD DOP 2010). Wetlands comprised 0.4% of Broad Creek's watershed, 5.6% of Harris Creek's, and 0.8% of Tred Avon's watershed. Water comprised a larger fraction of the area considered by MD DOP (2010) in Broad and Harris Creeks (57% and 61%, respectively) than Tred Avon River (27%), i.e., water to watershed ratios were higher in the former.

Tax map estimates of C/ha indicated that the Tred Avon River watershed has been subject to more development than Harris Creek or Broad Creek watersheds and more than indicated by the MD DOP urban category (Table 3-8; Figure 3-17). Time-series for all three watersheds started at a rural level of development ( $C/ha < 0.27$ ) in 1950. Broad Creek has been subject to the least growth ( $C/ha = 0.30$  in 2013), followed by Harris Creek By 2013 ( $C/ha = 0.39$  in 2013), and Tred Avon ( $C/ha = 0.75$ ). Development accelerated noticeably in the Tred Avon watershed during 1999-2007 and then slowed. Tred Avon River's watershed has been approaching the suburban threshold, ( $C/ha > 0.87$ ). The other two watersheds have just passed the rural target (Figure 3-17).

During 2015, bottom DO readings below the threshold ( $DO < 3.0$  mg / L) were more frequent in more developed Tred Avon River than the other two subestuaries (Table 3-8; Figure 3-18). Seven percent of DO measurements over a ten year period in Tred Avon River were below the threshold. Harris Creek did not exhibit any measurements in DO measurements below the threshold during 2012-2015, while 1% were below the threshold in Broad Creek. Below target DO conditions ( $\leq 5.0$  mg / L) were most consistently present in Tred Avon River (14%), followed by Broad Creek (6%), and Harris Creek (0.03%; Table 3-8; Figure 3-18) over all sampling years. A linear regression detected a significant negative relationship between bottom DO readings and the intensity of development in these three subestuaries ( $r^2 = 0.08$ ,  $P < 0.0001$ ,  $N = 423$ ).

An ANOVA of Tred Avon River stations and bottom DO was significant ( $P < 0.0001$ ,  $N = 235$ ). Tukey Studentized Range or Tukey Honestly Significant Difference (HSD) tests indicated that bottom DO at station 1 (station at Easton, Maryland) was significantly lower than downstream stations 2, 3, and 4 (critical value of studentized range = 3.65974). This result was consistent with other mesohaline tributaries with high impervious surface - DO declines as you approach the head of tide (Uphoff et al. 2011a). The mean and SE for bottom DO at all stations in Tred Avon River were 5.380 mg / L and 0.0912. Means and SE for station 1 were 4.146 mg / L and 0.214; station 2 means and SE were 5.745 mg / L and 0.122; station 3 means and SE were 5.809 mg / L and 0.142; and station 4 means and SE were 5.854 mg / L and 0.129. Deterioration of DO at the uppermost station (Figure 3-19) indicated that watershed development in Easton was the source of poor water quality than water intruding from downstream. ANOVAs of Harris Creek or Broad Creek stations and bottom DO concentrations did not indicate significant differences among stations ( $P = 0.3418$  and  $P = 0.2185$ , respectively;  $N = 94$  in both systems). The overall means and SE for bottom DO in Harris Creek and Broad Creek were 6.264 mg / L and 0.076 and 6.029 mg / L and 0.123, respectively.

Proportional stock densities for White Perch ( $P_{Qwp}$ ) during 2012-2015 were higher in less developed Harris and Broad Creeks ( $P_{Qwp} = 19.6\%$ ,  $SD = 1.1\%$ ,  $N = 1,205$  and  $P_{Qwp} = 11.4\%$ ,  $SD = 1.1\%$ ,  $N = 1,600$ , respectively) than more developed Tred Avon River ( $P_{Qwp} = 4.8\%$ ,  $SD = 0.3\%$ ,  $N = 4,884$ ). Size quality of White Perch directly aligned with the percentage of all DO measurements below the target level (5.0 mg / L). However, sample sizes indicate that White Perch were more abundant in Tred Avon River, so diminished size quality may reflect density-dependent dynamics. Abundance of adult White Perch in trawl samples was negatively influenced by development and distance from their spawning area (Uphoff et al. 2011a). Tred Avon River is both the most developed watershed of the three Choptank River subestuaries and is closer to the Choptank River spawning area.

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Table 3-1. Percent impervious cover (IS), structures per hectare (C/ha), watershed area, area of tidal water, and salinity class for the subestuaries sampled.

| Area      | Watershed       | Subestuary       | IS   | C/ha | Total Hectares | Water Hectares | Salinity Class |
|-----------|-----------------|------------------|------|------|----------------|----------------|----------------|
| Mid-Bay   | Lower Choptank  | Broad Creek      | 5.1  | 0.29 | 4,730          | 3,148          | Mesohaline     |
| Mid-Bay   | Lower Choptank  | Harris Creek     | 6.0  | 0.39 | 3696           | 2,919          | Mesohaline     |
| Mid-Bay   | Gunpowder River | Middle River     | 23.5 | 3.34 | 2,753          | 982            | Oligohaline    |
| Mid-Bay   | Lower Choptank  | Tred Avon River  | 9.2  | 0.75 | 9,563          | 2,429          | Mesohaline     |
| Potomac   | Lower Potomac   | Mattawoman Creek | 10.4 | 0.91 | 24,441         | 729            | Tidal Fresh    |
| Potomac   | Lower Potomac   | Nanjemoy Creek   | 2.4  | 0.09 | 18,893         | 1,131          | Oligohaline    |
| Upper-Bay | Bush River      | Bush River       | 14.2 | 1.51 | 36,038         | 2,962          | Tidal Fresh    |
| Upper-Bay | Gunpowder River | Gunpowder River  | 9.0  | 0.73 | 113,760        | 4,108          | Oligohaline    |
| Upper-Bay | Elk River       | Northeast River  | 6.9  | 0.47 | 16,342         | 1,579          | Tidal Fresh    |

Table 3-2. Percentages of all DO measurements and bottom DO measurements that did not meet target (= 5.0 mg/L) and threshold (= 3.0 mg/L) conditions during July-September, 2015, for each subestuary. C/ha = structures per hectare.

| Subestuary       | Salinity Class | C/ha | N  | All DO       |    | Bottom DO    |              |
|------------------|----------------|------|----|--------------|----|--------------|--------------|
|                  |                |      |    | % < 5.0 mg/L | N  | % < 5.0 mg/L | % < 3.0 mg/L |
| Broad Creek      | Mesohaline     | 0.29 | 82 | 1            | 23 | 0            | 0            |
| Harris Creek     | Mesohaline     | 0.39 | 85 | 0            | 24 | 0            | 0            |
| Tred Avon River  | Mesohaline     | 0.75 | 96 | 8.3          | 24 | 33.3         | 12.5         |
| Middle River     | Oligohaline    | 3.34 | 67 | 7.5          | 24 | 20.8         | 0            |
| Gunpowder River  | Oligohaline    | 0.73 | 54 | 0            | 13 | 0            | 0            |
| Nanjemoy Creek   | Oligohaline    | 0.09 | 52 | 3.9          | 10 | 20           | 0            |
| Bush River       | Tidal Fresh    | 1.51 | 47 | 4            | 14 | 7            | 0            |
| Mattawoman Creek | Tidal Fresh    | 0.91 | 96 | 0            | 43 | 0            | 0            |
| Northeast River  | Tidal Fresh    | 0.47 | 85 | 4.7          | 24 | 16.7         | 4.2          |

Table 3-3. Subestuaries sampled during 2003 – 2015 divided by salinity class with mean annual surface and bottom temperatures, mean summer dissolved oxygen (mg/L), and C / ha.

| River              | Year | C / ha   | Temperature |          | Dissolved Oxygen |          |
|--------------------|------|----------|-------------|----------|------------------|----------|
|                    |      |          | Surface     | Bottom   | Surface          | Bottom   |
| Mesohaline         |      |          |             |          |                  |          |
| Blackwater River   | 2006 | 0.037667 | 28.1444444  | 27.98333 | 5.266667         | 4.116667 |
| Breton Bay         | 2003 | 0.265021 | 26.4        | 25.6875  | 8.1              | 3.745833 |
|                    | 2004 | 0.281742 | 27.00625    | 25.95417 | 7.360417         | 3.725    |
|                    | 2005 | 0.298533 | 28.6214286  | 27.50833 | 6.978571         | 3.990417 |
| Broad Creek        | 2012 | 0.293475 | 27.4953659  | 26.60167 | 8.30439          | 5.965417 |
|                    | 2013 | 0.2958   | 27.2995238  | 26.48913 | 7.257857         | 5.756957 |
|                    | 2014 | 0.2958   | 27.6229167  | 26.64167 | 7.64625          | 5.77625  |
|                    | 2015 | 0.2958   | 28.0490244  | 27.0487  | 7.930244         | 6.631304 |
| Corsica River      | 2003 | 0.171949 | 25.9        | 26.13043 | 6.5              | 4.669565 |
|                    | 2004 | 0.184452 | 27.1756098  | 26.875   | 5.568293         | 4.571    |
|                    | 2005 | 0.193959 | 28.5380952  | 28.14286 | 6.483333         | 3.08     |
|                    | 2006 | 0.211423 | 27.3857143  | 26.84118 | 7.548571         | 4.047059 |
|                    | 2007 | 0.224649 | 25.9414634  | 25.81818 | 6.2425           | 4.218182 |
|                    | 2008 | 0.23705  | 26.204878   | 25.21538 | 7.319512         | 4.207692 |
|                    | 2011 | 0.250586 | 27.0023256  | 27.01    | 5.295455         | 3.280556 |
|                    | 2012 | 0.250586 | 27.7928571  | 27.46875 | 4.7125           | 3.403333 |
| Fishing Bay        | 2006 | 0.033932 | 26.2296296  | 25.27857 | 7.240741         | 6.792857 |
| Harris Creek       | 2012 | 0.387979 | 26.5542857  | 26.41783 | 7.438571         | 6.354348 |
|                    | 2013 | 0.387708 | 26.3914634  | 26.05292 | 7.015366         | 6.01     |
|                    | 2014 | 0.387708 | 26.8294779  | 26.15404 | 6.766543         | 4.502407 |
|                    | 2015 | 0.387708 | 26.6163415  | 26.62125 | 7.193902         | 6.564583 |
| Langford Creek     | 2006 | 0.072884 | 27.0511111  | 26.52083 | 6.946667         | 5.675    |
|                    | 2007 | 0.073608 | 26.2326087  | 25.47895 | 6.691892         | 5.684615 |
|                    | 2008 | 0.073504 | 27.0676799  | 26.62511 | 6.652126         | 4.773951 |
| Magothy River      | 2003 | 2.678242 | 25.7        | 25.31429 | 7.3              | 2.035714 |
| Miles River        | 2003 | 0.23851  | 25.5        | 25.6     | 6.5              | 4.092    |
|                    | 2004 | 0.243382 | 25.7525     | 25.63913 | 6.0825           | 5.466087 |
|                    | 2005 | 0.244374 | 28.0333333  | 27.44167 | 5.961905         | 3.308333 |
| Rhode River        | 2003 | 0.466475 | 25          | 24.69286 | 7.1              | 4.8      |
|                    | 2004 | 0.473539 | 27          | 26.94545 | 6.578261         | 5.389091 |
|                    | 2005 | 0.476647 | 27.7791667  | 27.15833 | 6.5              | 4.025833 |
| Severn River       | 2003 | 2.058995 | 26.3        | 24.75185 | 7.6              | 1.574074 |
|                    | 2004 | 2.09118  | 27.4166667  | 26.17917 | 7.05             | 2.636667 |
|                    | 2005 | 2.148981 | 28.0148936  | 26.22917 | 7.07234          | 0.96375  |
| South River        | 2003 | 1.234149 | 25.4        | 24.56429 | 7.6              | 2.610714 |
|                    | 2004 | 1.2497   | 25.7875     | 25.47917 | 6.45625          | 3.77375  |
|                    | 2005 | 1.26471  | 27.5708333  | 26.67083 | 6.016667         | 2.49125  |
| St. Clements River | 2003 | 0.192976 | 26          | 25.28519 | 8.2              | 3.481481 |
|                    | 2004 | 0.19621  | 26.0791667  | 25.775   | 6.8375           | 4.608333 |
|                    | 2005 | 0.198532 | 26.8848434  | 26.36662 | 6.712767         | 4.333602 |
| Transquaking River | 2006 | 0.028893 | 26.675      | 22.75    | 5.75             | 5.85     |

(Cont. Table 3-3)

|                 |                 |          |            |            |          |          |          |
|-----------------|-----------------|----------|------------|------------|----------|----------|----------|
| Tred Avon River | 2006            | 0.691286 | 27.1159091 | 26.72083   | 6.181818 | 5.341667 |          |
|                 | 2007            | 0.713035 | 26.85      | 26.59167   | 6.485106 | 5.391304 |          |
|                 | 2008            | 0.724433 | 26.2770833 | 25.6087    | 6.895833 | 4.833333 |          |
|                 | 2009            | 0.736144 | 26.1541667 | 26.03333   | 7.370833 | 6.305833 |          |
|                 | 2010            | 0.74681  | 27.4695652 | 26.92917   | 7.084783 | 5.258333 |          |
|                 | 2011            | 0.750993 | 28.4840909 | 28.18095   | 6.815909 | 5.10619  |          |
|                 | 2012            | 0.75298  | 27.2710638 | 27.15833   | 7.022292 | 5.46625  |          |
|                 | 2013            | 0.754025 | 26.7880769 | 26.39038   | 7.15     | 4.998077 |          |
|                 | 2014            | 0.754025 | 26.65875   | 26.50542   | 6.116667 | 5.902917 |          |
|                 | 2015            | 0.754025 | 27.996875  | 27.59542   | 6.924792 | 5.537083 |          |
|                 | West River      | 2003     | 0.642769   | 24.9       | 24.31429 | 7.4      | 4.835714 |
|                 |                 | 2004     | 0.64863    | 26.8333333 | 26.59167 | 7.366667 | 5.583333 |
|                 |                 | 2005     | 0.658398   | 27.9611111 | 27.15    | 6.722222 | 3.986667 |
|                 | Wicomico River  | 2003     | 0.193906   | 25.4       | 23.83043 | 7        | 5.852174 |
|                 |                 | 2011     | 0.212462   | 27.0782609 | 26.8913  | 5.567609 | 4.301739 |
| 2012            |                 | 0.212462 | 27.5683333 | 27.382     | 6.585417 | 5.444667 |          |
| Wye River       | 2007            | 0.095131 | 26.7541667 | 26.45      | 7.075    | 5.7      |          |
|                 | 2008            | 0.095424 | 26.9844444 | 26.21875   | 5.702222 | 5.113333 |          |
| Oligohaline     |                 |          |            |            |          |          |          |
| Bohemia River   | 2006            | 0.111575 | 26.7880952 | 26.02      | 7.009524 | 6.41     |          |
| Bush River      | 2006            | 1.406067 | 25.4763158 | 24.28      | 7.957895 | 7.472727 |          |
|                 | 2007            | 1.429209 | 27.0222222 | 26.41818   | 7.677778 | 6.536364 |          |
|                 | 2008            | 1.447218 | 26.5857143 | 24.2       | 9        | 5.433333 |          |
|                 | 2009            | 1.461508 | 25.8809524 | 24.3375    | 9.409524 | 8.54     |          |
|                 | 2010            | 1.470554 | 27.7194444 | 23.8       | 7.791667 | 7.04     |          |
|                 | 2011            | 1.479684 | 26.9820513 | 26.94      | 6.465641 | 5.496667 |          |
|                 | 2012            | 1.487564 | 26.79      | 26.16667   | 6.6275   | 5.200833 |          |
|                 | 2013            | 1.506516 | 25.1107143 | 24.725     | 9.98     | 6.7275   |          |
|                 | 2014            | 1.506516 | 26.6976631 | 25.79921   | 7.225839 | 5.063915 |          |
|                 | 2015            | 1.506516 | 26.7185891 | 25.78887   | 7.25893  | 5.195288 |          |
|                 | Gunpowder River | 2009     | 0.720501   | 25.7093023 | 26.05    | 7.390698 | 6.789444 |
|                 |                 | 2010     | 0.722787   | 25.1688889 | 25.90769 | 7.893182 | 7.130769 |
|                 |                 | 2011     | 0.724747   | 25.0858065 | 25.55556 | 8.283871 | 7.144444 |
|                 |                 | 2012     | 0.726953   | 26.4844444 | 25.93133 | 8.193778 | 6.708667 |
|                 |                 | 2013     | 0.729485   | 25.8528205 | 27.45667 | 8.047949 | 6.1      |
| 2014            |                 | 0.729485 | 26.5861753 | 25.83227   | 7.220582 | 5.179648 |          |
| 2015            |                 | 0.729485 | 27.5102439 | 27.65      | 8.016341 | 6.631538 |          |
| Middle River    | 2009            | 3.300754 | 26.4962963 | 25.78182   | 7.266667 | 6.067727 |          |
|                 | 2010            | 3.320004 | 24.6486486 | 24.2       | 8.437838 | 7.113636 |          |
|                 | 2011            | 3.329084 | 27.1309524 | 26.41667   | 8.354286 | 7.333333 |          |
|                 | 2012            | 3.333079 | 28.05      | 26.59565   | 8.817105 | 5.209167 |          |
|                 | 2013            | 3.335258 | 27.120303  | 26.45545   | 7.58303  | 5.786364 |          |
|                 | 2014            | 3.335258 | 26.5318235 | 25.90089   | 7.817535 | 6.276438 |          |
|                 | 2015            | 3.335258 | 28.4655556 | 27.2       | 8.195556 | 6.232083 |          |
| Nanjemoy Creek  | 2003            | 0.084899 | 25.9       | 28.8       | 7.3      | 4.96     |          |
|                 | 2008            | 0.091092 | 27.5257143 | 26.575     | 7.851429 | 6.65     |          |
|                 | 2009            | 0.091197 | 26.3055556 | 24.6375    | 7.05     | 7.49375  |          |
|                 | 2010            | 0.091568 | 26.4965789 | 24.79857   | 7.664474 | 7.015714 |          |
|                 | 2011            | 0.091568 | 29.33625   | 28.54962   | 6.1275   | 5.303077 |          |

(Cont. Table 3-3)

|                  |      |          |            |          |          |          |
|------------------|------|----------|------------|----------|----------|----------|
|                  | 2012 | 0.091727 | 26.17875   | 25.9245  | 6.73075  | 5.978    |
|                  | 2013 | 0.091885 | 26.8794444 | 26.295   | 6.759722 | 5.86     |
|                  | 2014 | 0.091885 | 26.6877126 | 26.32321 | 7.68058  | 6.308718 |
|                  | 2015 | 0.091885 | 27.3988889 | 27.098   | 7.159722 | 6.324    |
| <hr/>            |      |          |            |          |          |          |
| Tidal Fresh      |      |          |            |          |          |          |
| Mattawoman Creek | 2003 | 0.762374 | 26         | 25.74545 | 9        | 8.813636 |
|                  | 2004 | 0.786923 | 27.3258065 | 27.14348 | 8.341935 | 7.95087  |
|                  | 2005 | 0.807012 | 28.772     | 28.0875  | 7.736    | 7.266875 |
|                  | 2006 | 0.83238  | 27.047619  | 26.44    | 7.095    | 6.495455 |
|                  | 2007 | 0.858197 | 26.8875    | 26.85417 | 6.704167 | 6.475    |
|                  | 2008 | 0.871208 | 26.3958333 | 24.51538 | 7.968182 | 6.325    |
|                  | 2009 | 0.883196 | 26.2047826 | 26.63846 | 7.920217 | 7.858846 |
|                  | 2010 | 0.897762 | 26.20625   | 26.09524 | 6.945833 | 6.62381  |
|                  | 2011 | 0.910569 | 27.079375  | 27.45794 | 6.327083 | 6.511765 |
|                  | 2012 | 0.902877 | 26.695625  | 26.81977 | 7.39875  | 6.999535 |
|                  | 2013 | 0.914415 | 26.35125   | 25.93733 | 9.218958 | 8.404    |
|                  | 2014 | 0.914415 | 26.7315222 | 26.18579 | 7.587535 | 6.298986 |
|                  | 2015 | 0.914415 | 27.9139583 | 26.84419 | 8.6575   | 7.742791 |
| Northeast River  | 2007 | 0.43979  | 26.8270833 | 26.42727 | 9.733333 | 7.747826 |
|                  | 2008 | 0.443095 | 25.3479167 | 24.98421 | 8.429167 | 7.7      |
|                  | 2009 | 0.449642 | 26.3306122 | 25.54783 | 9.35102  | 7.361304 |
|                  | 2010 | 0.459127 | 25.9042553 | 26.20588 | 7.761702 | 6.782353 |
|                  | 2011 | 0.464818 | 25.9673913 | 25.7125  | 6.872826 | 5.792083 |
|                  | 2012 | 0.467816 | 27.7833333 | 27.59167 | 7.877083 | 6.033333 |
|                  | 2013 | 0.473507 | 26.6136957 | 26.10957 | 9.333696 | 7.055217 |
|                  | 2014 | 0.473507 | 26.693027  | 26.31995 | 7.803857 | 6.736735 |
|                  | 2015 | 0.473507 | 26.6552083 | 26.23    | 7.835833 | 6.17375  |
| Piscataway Creek | 2003 | 1.300181 | 25.6       | 24.63333 | 10.2     | 8.333333 |
|                  | 2006 | 1.38186  | 28.155     | 24.96667 | 8.7      | 6.85     |
|                  | 2007 | 1.401642 | 27.4666667 | 26       | 8.566667 | 7.6      |
|                  | 2009 | 1.433215 | 26.7166667 | 27.06667 | 8.555556 | 6.622857 |
|                  | 2010 | 1.448746 | 27.0666667 | 25.075   | 9.355556 | 7.625    |
|                  | 2011 | 1.462066 | 28.2461111 | 30.06667 | 9.05     | 9.466667 |
|                  | 2012 | 1.472495 | 27.92      | 25.50875 | 9.532105 | 9.3425   |
|                  | 2013 | 1.49035  | 27.1870588 | 26.22111 | 9.87     | 7.648889 |
|                  | 2014 | 1.49035  | 26.8738712 | 26.18172 | 8.71462  | 7.365881 |

Table 3-4. Correlations of 2003-2015 arithmetic mean annual surface and bottom DO (mg/L) with mean water temperatures at depth (surface or bottom) or watershed development (C/ha = structures per hectare), by salinity class. Bold numbers indicate a significant relationship.

| DO Depth    | Statistics | Temperature Depth | C / ha           |
|-------------|------------|-------------------|------------------|
| Mesohaline  |            |                   |                  |
| Surface     | r          | -0.20387          | 0.20181          |
|             | $\alpha$   | 0.1214            | 0.1253           |
|             | N          | 59                | 59               |
| Bottom      | r          | 0.01402           | <b>-0.57778</b>  |
|             | $\alpha$   | 0.9161            | <b>&lt;.0001</b> |
|             | N          | 59                | <b>59</b>        |
| Oligohaline |            |                   |                  |
| Surface     | r          | <b>-0.34345</b>   | <b>0.36646</b>   |
|             | $\alpha$   | <b>0.0467</b>     | <b>0.033</b>     |
|             | N          | <b>34</b>         | <b>34</b>        |
| Bottom      | r          | <b>-0.51708</b>   | -0.01405         |
|             | $\alpha$   | <b>0.0017</b>     | 0.9372           |
|             | N          | <b>34</b>         | 34               |
| Tidal Fresh |            |                   |                  |
| Surface     | r          | 0.03811           | 0.35077          |
|             | $\alpha$   | 0.8387            | 0.053            |
|             | N          | 31                | 31               |
| Bottom      | r          | 0.10861           | <b>0.42039</b>   |
|             | $\alpha$   | 0.5608            | <b>0.0185</b>    |
|             | N          | 31                | <b>31</b>        |

Table 3-5. Beach seine catch summary, 2015. C/ha = structures per hectare. GM is the geometric mean catch of all fish per seine. Italicized species names = target species.

| River           | Stations Sampled | Number of Samples | Number of Species | Species Comprising 90% of Catch  | C / ha   | Total Catch | GM CPU E |
|-----------------|------------------|-------------------|-------------------|--|----------|-------------|----------|
| Broad Creek     | 3                | 17                | 22                | <i>ATLANTIC MENHADEN</i><br>ATLANTIC SILVERSIDE<br>STRIPED KILLIFISH<br>BANDED KILLIFISH<br>MUMMICHOG<br>FOUR SPINE<br>STICKLEBACK   | 0.2<br>9 | 9525        | 442      |
| Bush River      | 4                | 20                | 24                | <i>WHITE PERCH (JUV)</i><br><i>BLUEBACK HERRING</i><br><i>SPOTTAIL SHINER</i><br><i>GIZZARD SHAD</i><br><i>HERRINGS (UNIDENTIFIED)</i><br><i>ALEWIFE</i>   | 1.5<br>1 | 6085        | 233      |
| Gunpowder River | 3                | 17                | 22                | <i>SPOTTAIL SHINER</i><br><i>WHITE PERCH (JUV)</i><br><i>BLUEBACK HERRING</i><br><i>WHITE PERCH</i><br>BANDED KILLIFISH<br><i>YELLOW PERCH (JUV)</i><br><i>ALEWIFE</i>   | 0.7<br>3 | 2586        | 152      |
| Harris Creek    | 3                | 17                | 23                | INLAND SILVERSIDE<br>ATLANTIC SILVERSIDE<br>STRIPED KILLIFISH<br>MUMMICHOG<br><i>WHITE PERCH (JUV)</i><br><i>ALEWIFE</i>   | 0.3<br>9 | 7165        | 402      |
| Middle River    | 1                | 3                 | 13                | <i>WHITE PERCH (JUV)</i><br>PUMPKINSEED<br>BANDED KILLIFISH<br>INLAND SILVERSIDE<br>BLUEGILL   | 3.3<br>4 | 872         | 426      |
| Nanjemoy Creek  | 3                | 18                | 29                | <i>WHITE PERCH (JUV)</i><br>ATLANTIC SILVERSIDE<br>MUMMICHOG<br>BAY ANCHOVY<br>BANDED KILLIFISH<br><i>BLUEBACK HERRING</i><br><i>GIZZARD SHAD</i><br><i>WHITE PERCH</i><br>PUMPKINSEED<br>INLAND SILVERSIDE<br><i>AMERICAN SHAD</i><br><i>GIZZARD SHAD</i> | 0.0<br>9 | 3888        | 165      |
| Northeast       | 4                | 24                | 25                |  | 0.4      | 4318        | 147      |

|                 |    |     |    |                            |     |      |     |
|-----------------|----|-----|----|----------------------------|-----|------|-----|
| River           |    |     |    | <i>BLUEBACK HERRING</i>    | 7   |      |     |
|                 |    |     |    | <i>WHITE PERCH (JUV)</i>   |     |      |     |
|                 |    |     |    | <i>ALEWIFE</i>             |     |      |     |
|                 |    |     |    | <i>WHITE PERCH</i>         |     |      |     |
|                 |    |     |    | <i>SPOTTAIL SHINER</i>     |     |      |     |
|                 |    |     |    | <i>PUMPKINSEED</i>         |     |      |     |
|                 |    |     |    | <i>BAY ANCHOVY</i>         |     |      |     |
| Tred Avon River | 4  | 24  | 20 | <i>ATLANTIC SILVERSIDE</i> | 0.7 | 1282 | 414 |
|                 |    |     |    | <i>STRIPED KILLIFISH</i>   | 5   | 5    |     |
|                 |    |     |    | <i>WHITE PERCH (JUV)</i>   |     |      |     |
|                 |    |     |    | <i>MUMMICHOG</i>           |     |      |     |
|                 |    |     |    | <i>BANDED KILLIFISH</i>    |     |      |     |
| Grand Total     | 25 | 140 | 47 | <i>ATLANTIC SILVERSIDE</i> |     | 4726 |     |
|                 |    |     |    | <i>WHITE PERCH (JUV)</i>   |     | 4    |     |
|                 |    |     |    | <i>ATLANTIC MENHADEN</i>   |     |      |     |
|                 |    |     |    | <i>STRIPED KILLIFISH</i>   |     |      |     |
|                 |    |     |    | <i>BLUEBACK HERRING</i>    |     |      |     |
|                 |    |     |    | <i>SPOTTAIL SHINER</i>     |     |      |     |
|                 |    |     |    | <i>GIZZARD SHAD</i>        |     |      |     |
|                 |    |     |    | <i>BANDED KILLIFISH</i>    |     |      |     |
|                 |    |     |    | <i>MUMMICHOG</i>           |     |      |     |
|                 |    |     |    | <i>WHITE PERCH</i>         |     |      |     |
|                 |    |     |    | <i>ALEWIFE</i>             |     |      |     |

Table 3-6. Bottom trawl (4.9m) catch summary, 2015. C/ha = structures per hectare. GM is the geometric mean catch of all fish per seine. Italicized species names = target species.

| River            | Stations Sampled | Number of Samples | Number of Species | Species Comprising 90% of Catch  | C / ha | Total Catch | GM CPU E |
|------------------|------------------|-------------------|-------------------|--|--------|-------------|----------|
| Broad Creek      | 4                | 24                | 18                | BAY ANCHOVY<br>WEAKFISH<br>ATLANTIC SILVERSIDE   | 0.29   | 5226        | 103      |
| Bush River       | 3                | 14                | 14                | <i>WHITE PERCH (JUV)</i><br><i>WHITE PERCH</i><br><i>ALEWIFE</i><br>BROWN BULLHEAD<br><i>BLUEBACK HERRING</i><br>BAY ANCHOVY | 1.51   | 5424        | 321      |
| Gunpowder River  | 4                | 24                | 23                | <i>WHITE PERCH (JUV)</i><br><i>SPOTTAIL SHINER</i><br>BAY ANCHOVY<br><i>WHITE PERCH</i>                                      | 0.73   | 6744        | 218      |
| Harris Creek     | 4                | 24                | 15                | BAY ANCHOVY<br>ATLANTIC SILVERSIDE<br><i>STRIPED BASS (JUV)</i><br>WEAKFISH  | 0.39   | 1318        | 40       |
| Mattawoman Creek | 4                | 24                | 19                | <i>WHITE PERCH (JUV)</i><br><i>SPOTTAIL SHINER</i><br><i>WHITE PERCH</i><br><i>BLUEBACK HERRING</i>                          | 0.91   | 9651        | 217      |
| Middle River     | 4                | 24                | 19                | <i>WHITE PERCH (JUV)</i><br>BAY ANCHOVY<br>PUMPKINSEED   | 3.34   | 9968        | 286      |
| Nanjemoy Creek   | 3                | 18                | 19                | <i>WHITE PERCH (JUV)</i><br>BAY ANCHOVY  | 0.09   | 9376        | 416      |
| Northeast River  | 4                | 24                | 18                | <i>WHITE PERCH</i><br><i>WHITE PERCH (JUV)</i><br>BAY ANCHOVY  | 0.47   | 4763        | 150      |
| Tred Avon River  | 4                | 24                | 18                | BAY ANCHOVY<br>BAY ANCHOVY<br>WEAKFISH<br><i>STRIPED BASS (JUV)</i><br><i>WHITE PERCH (JUV)</i>                              | 0.75   | 3868        | 80       |
| Grand Total      | 34               | 200               | 42                | <i>WHITE PERCH (JUV)</i><br>BAY ANCHOVY<br><i>WHITE PERCH</i><br><i>SPOTTAIL SHINER</i>                                      |        | 56338       |          |

Table 3-7. Maryland Department of Planning (2010) estimates of percentage of each broad land use category for each Choptank River subestuary watershed. The top four land use categories are estimated from the land portion of the watershed. Water category is the percent of are occupied by water in each watershed (water area / [water + land area]).

| Land Use Category | Subestuary  |              |                 |
|-------------------|-------------|--------------|-----------------|
|                   | Broad Creek | Harris Creek | Tred Avon River |
| Agriculture       | 42.55       | 44.87        | 43.2            |
| Forest            | 25.39       | 19.72        | 21.63           |
| Urban             | 31.47       | 29.8         | 33.57           |
| Wetlands          | 0.36        | 5.61         | 0.85            |
| Water             | 57.28       | 61.18        | 24.4            |

Table 3-8. Percentages of all DO measurements (surface, middle, and bottom) and bottom DO measurements that did not meet target (= 5.0 mg/L) and threshold (= 3.0 mg/L) conditions during July - September for all sampling years, for each Choptank River subestuary.

| Subestuary      | Year | C / ha | N   | All DO       |    | Bottom DO    |              |
|-----------------|------|--------|-----|--------------|----|--------------|--------------|
|                 |      |        |     | % < 5.0 mg/L | N  | % < 5.0 mg/L | % < 3.0 mg/L |
| Broad Creek     | 2012 | 0.29   | 83  | 7            | 24 | 17           | 4            |
|                 | 2013 | 0.30   | 78  | 10           | 23 | 30           | 0            |
|                 | 2014 | 0.30   | 81  | 6            | 24 | 21           | 0            |
|                 | 2015 | 0.30   | 82  | 1            | 23 | 0            | 0            |
| Harris Creek    | 2012 | 0.39   | 82  | 0            | 23 | 0            | 0            |
|                 | 2013 | 0.39   | 83  | 0            | 24 | 0            | 0            |
|                 | 2014 | 0.39   | 84  | 1            | 23 | 4            | 0            |
|                 | 2015 | 0.39   | 85  | 0            | 24 | 0            | 0            |
| Tred Avon River | 2006 | 0.69   | 91  | 19           | 24 | 38           | 0            |
|                 | 2007 | 0.71   | 93  | 11           | 23 | 26           | 4            |
|                 | 2008 | 0.72   | 89  | 24           | 21 | 48           | 14           |
|                 | 2009 | 0.74   | 95  | 6            | 24 | 13           | 0            |
|                 | 2010 | 0.75   | 89  | 20           | 24 | 38           | 13           |
|                 | 2011 | 0.75   | 82  | 22           | 21 | 48           | 10           |
|                 | 2012 | 0.75   | 94  | 10           | 24 | 29           | 0            |
|                 | 2013 | 0.75   | 103 | 15           | 26 | 31           | 15           |
|                 | 2014 | 0.75   | 96  | 11           | 24 | 21           | 0            |
| 2015            | 0.75 | 96     | 8   | 24           | 21 | 13           |              |



Figure 3-1. Subestuaries sampled during summer, 2015.



Figure 3-2. Number of finfish species collected by seining in fresh-tidal, oligohaline, and mesohaline subestuaries versus intensity of watershed development (C/ha = structures per hectare). Points were omitted if seine effort < 15.

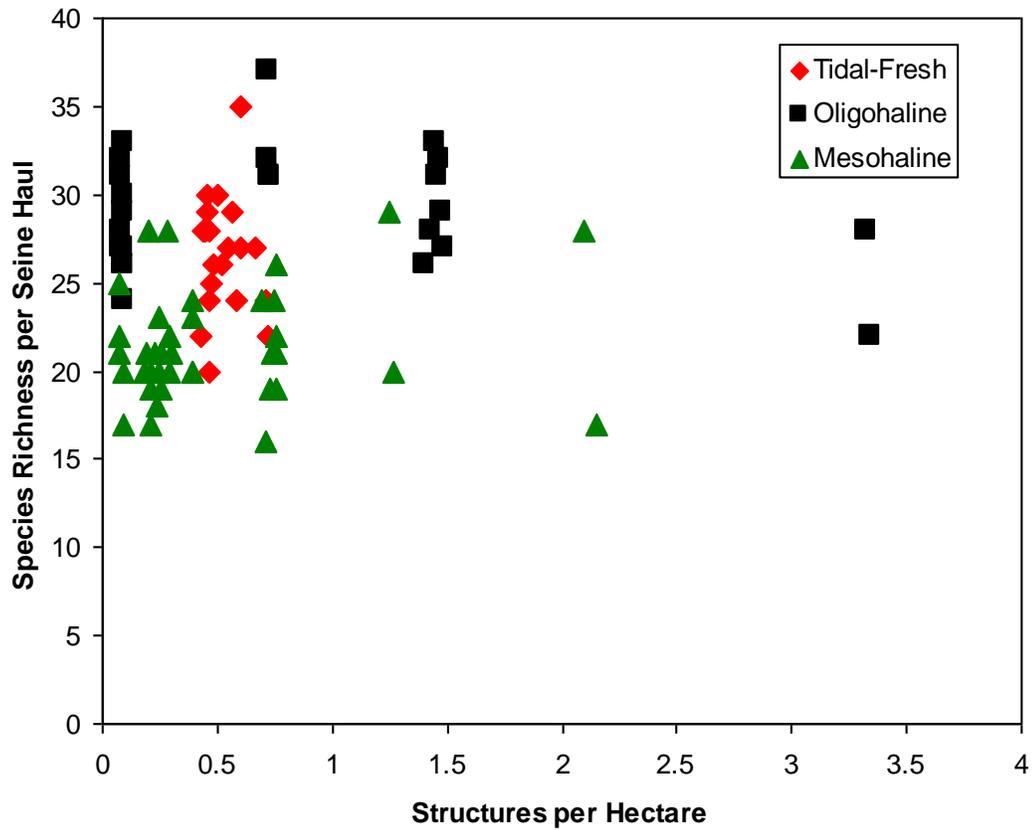


Figure 3-3. Number of finfish species collected by trawling in fresh-tidal, oligohaline, and mesohaline subestuaries versus intensity of watershed development (C/ha = structures per hectare). Points were omitted if seine effort < 15.

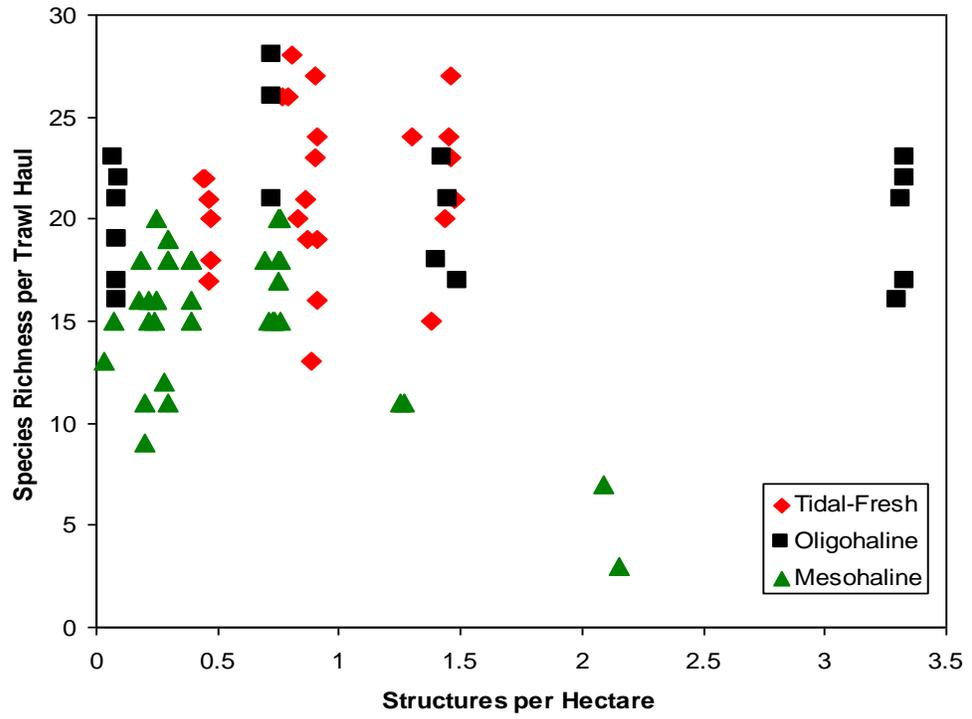


Figure 3- 4. Mean subestuary bottom DO during summer sampling, 2003-2015, plotted against structures per hectare (C / ha).

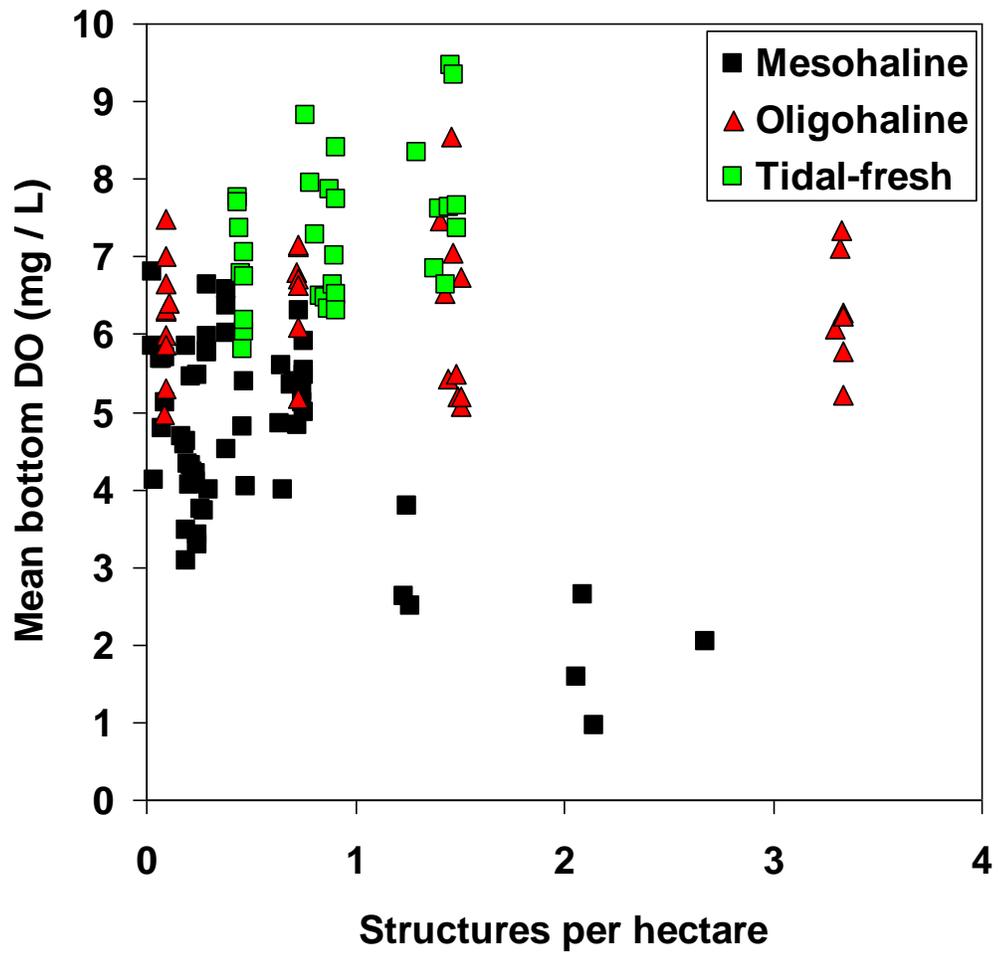


Figure 3-5. Mean subestuary surface DO during summer sampling, 2003-2015, plotted against structures per hectare (C / ha).

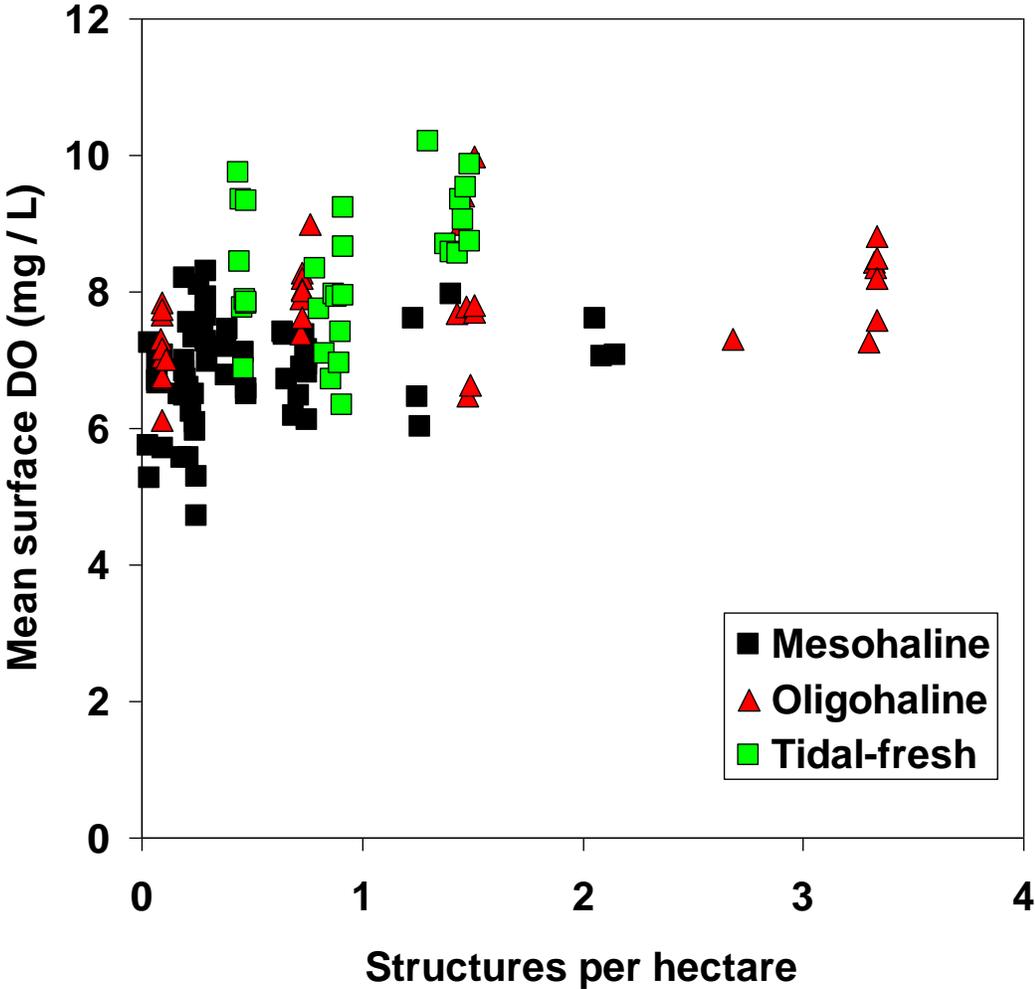


Figure 3-6. Trend in development (structures per hectare or C / ha) of Mattawoman Creek's watershed during 1989-2015. Black square indicates values that are at or beyond the threshold for a suburban watershed.

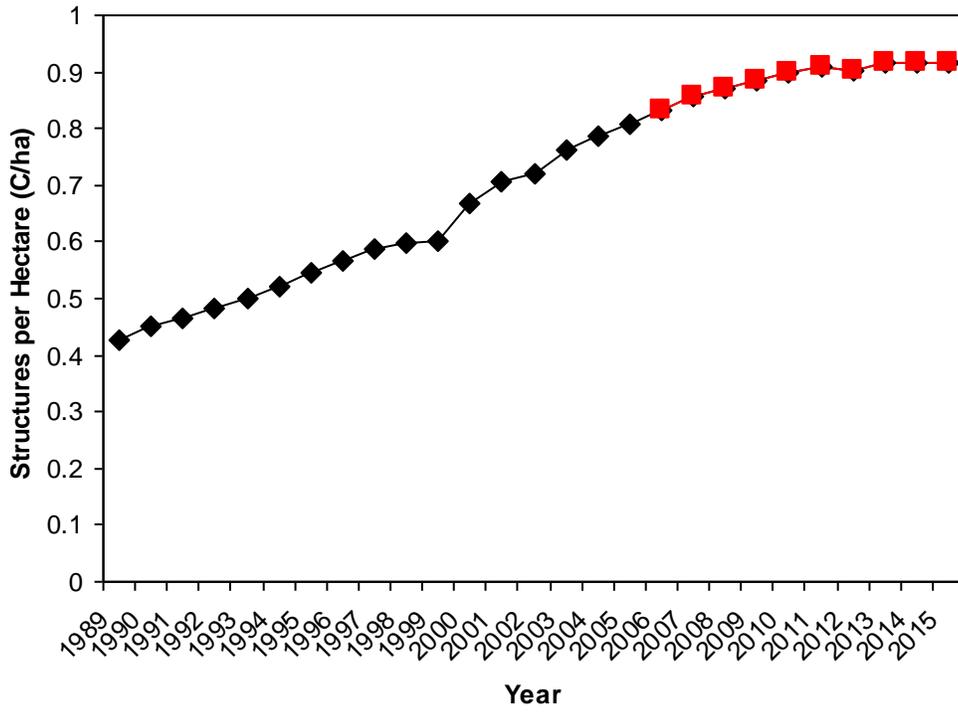


Figure 3-7. Mean bottom dissolved oxygen (DO) during July-September in Mattawoman Creek's subestuary, 1989-2015. Dashed line indicates median for the time-series of annual means.

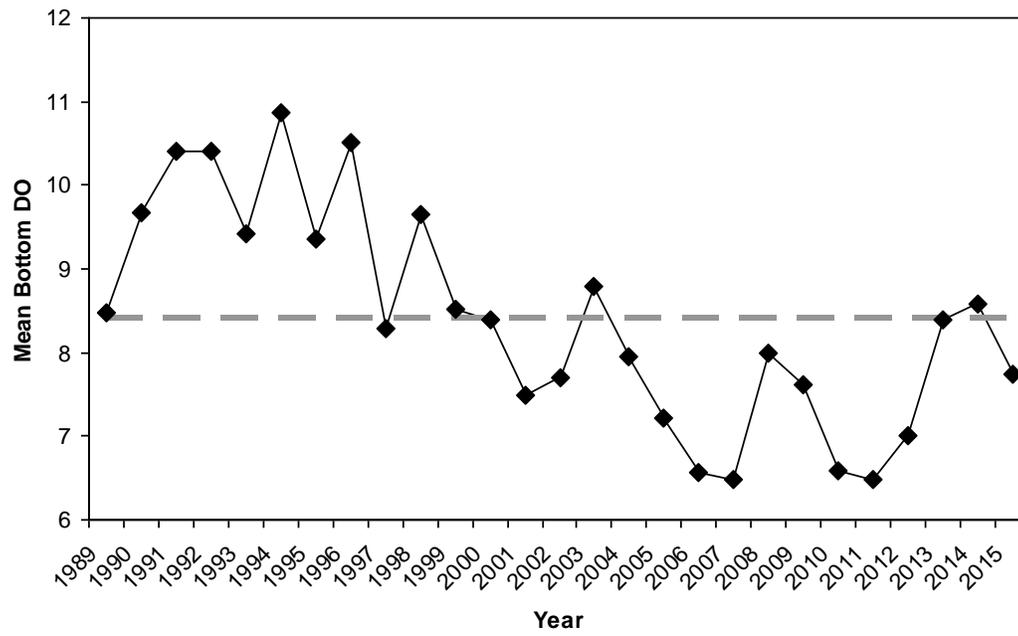


Figure 3-8. Range (vertical solid gray line) and the median chlorophyll a (ug/L) (solid black line) at a Chesapeake Bay Program monitoring station in Mattawoman Creek (MAT0016) during SAV growing season (April - October).

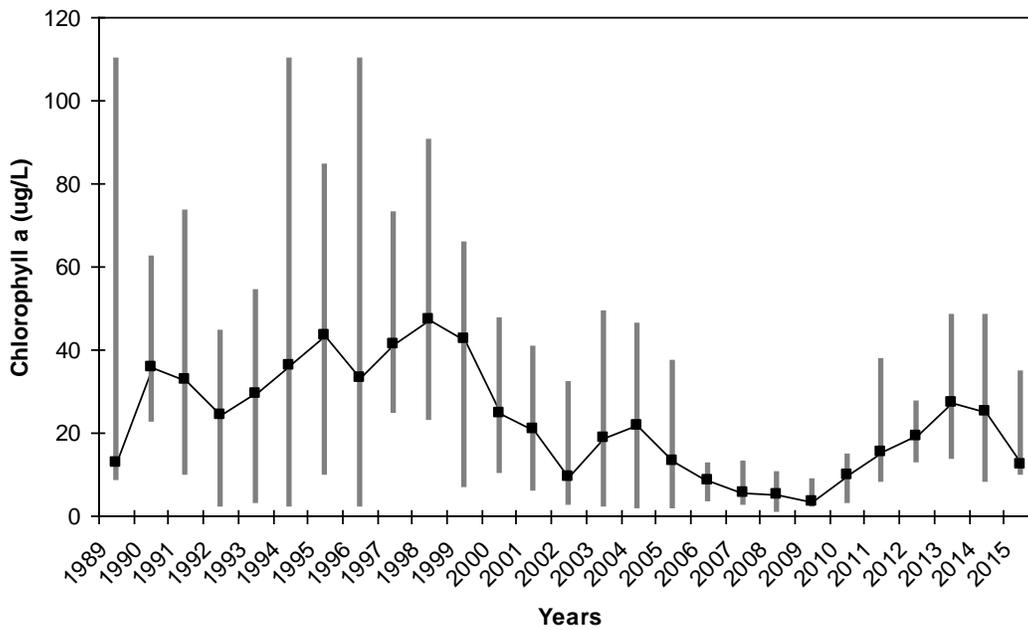


Figure 3-9. Percent of Mattawoman Creek's subestuary covered by SAV during 1989-2014 (2001 was only partially mapped).

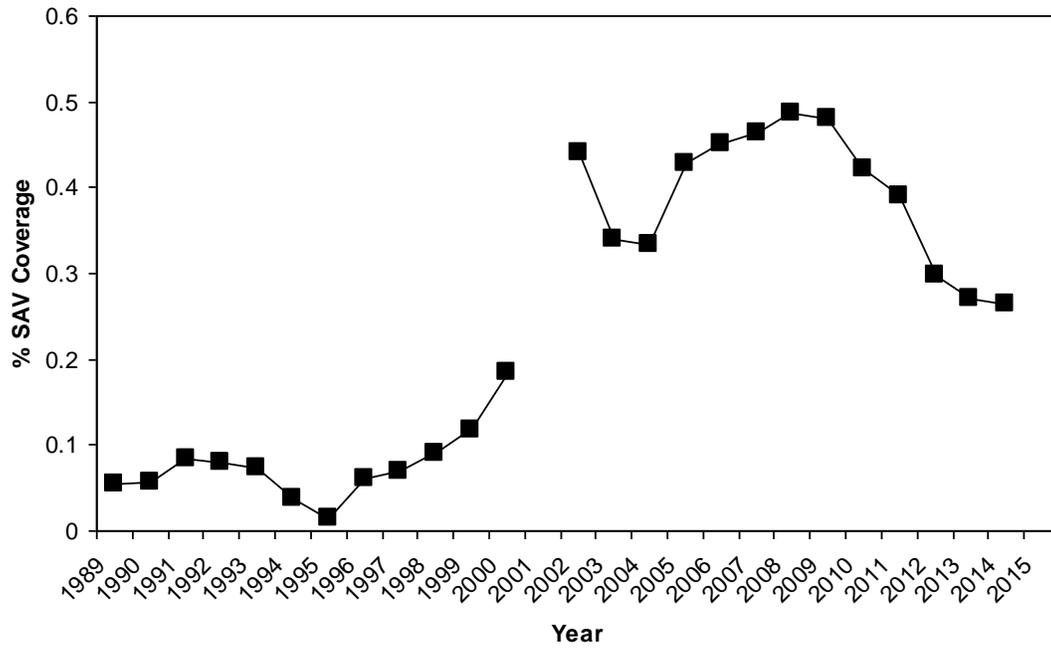


Figure 3-10. Range (vertical solid gray line) and the median total ammonia nitrogen (TAN; mg/L) (solid black line) at a Chesapeake Bay program monitoring station in Mattawoman Creek (MAT0016) during SAV growing season (April - October).

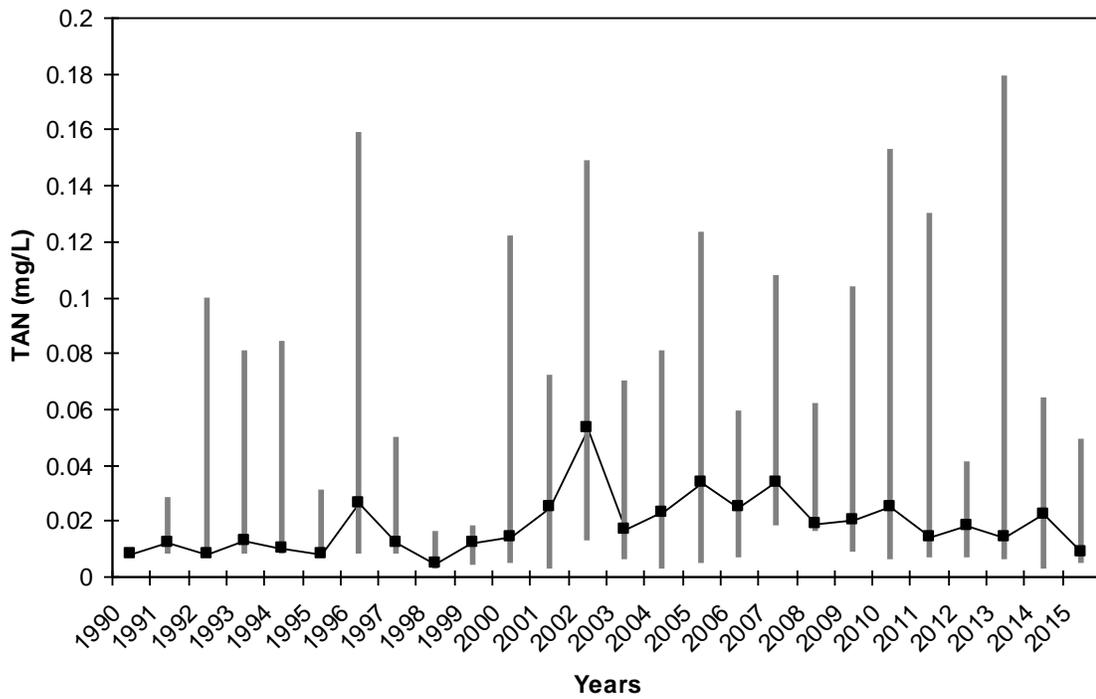


Figure 3-11. Median TAN (mg/L) versus the percentage of SAV coverage 1990-2014 in Mattawoman Creek (2001 omitted due to incomplete mapping).

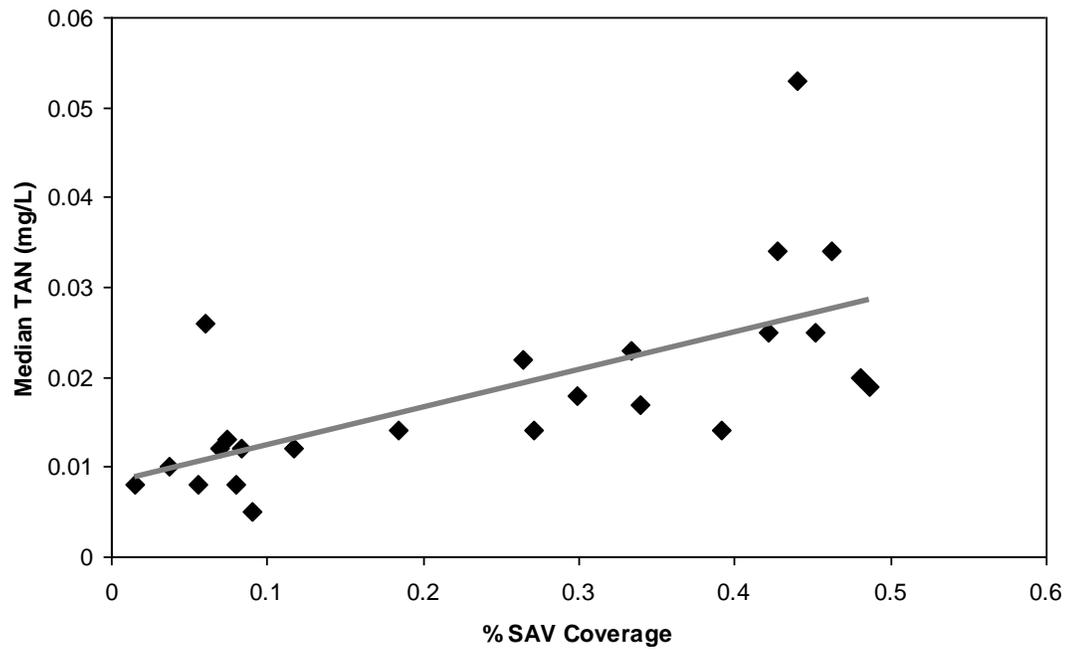


Figure 3-12. Median pH (solid black line) and its range (vertical solid grey line) at a Chesapeake Bay program monitoring station in Mattawoman Creek (MAT0016) during SAV growing season (April - October).

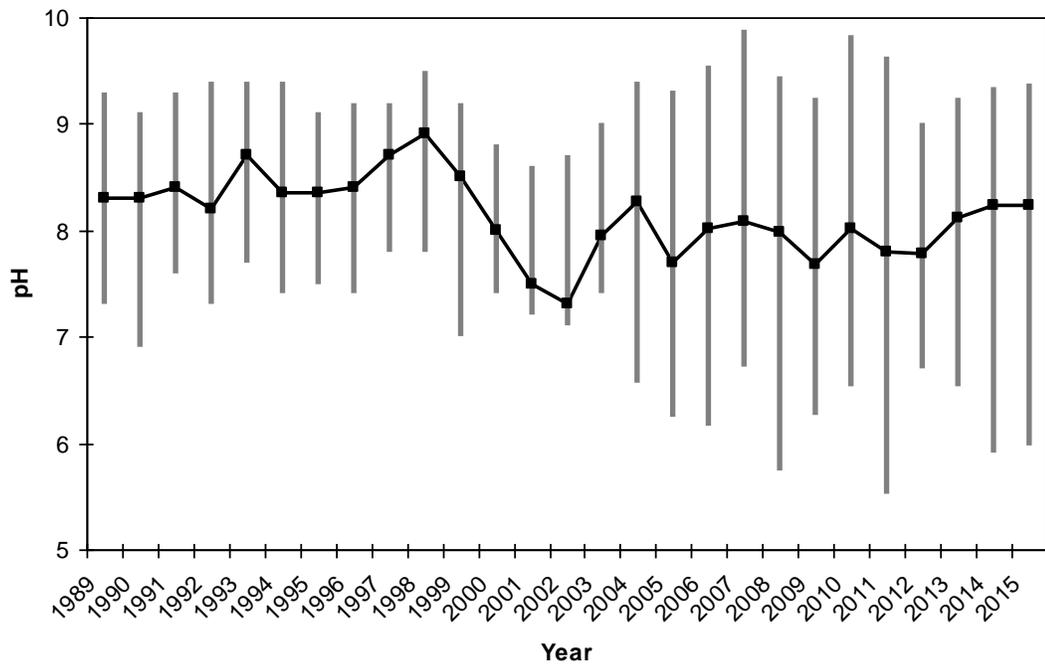


Figure 3-13. Geometric mean (GM) catches per trawl of all species of finfish in Mattawoman Creek during 1989-2015. Note dual axes for 3.1 m and 4.9 m trawls. Predicted 3.1 m GM is based on a linear regression of 3.1 m and 4.9 m trawl GMs during 2009-2015. Dotted horizontal lines indicate median GM of 3.1 m trawl samples for 1989-2001 (red dotted line) and 2002-2014 (blue dotted line).

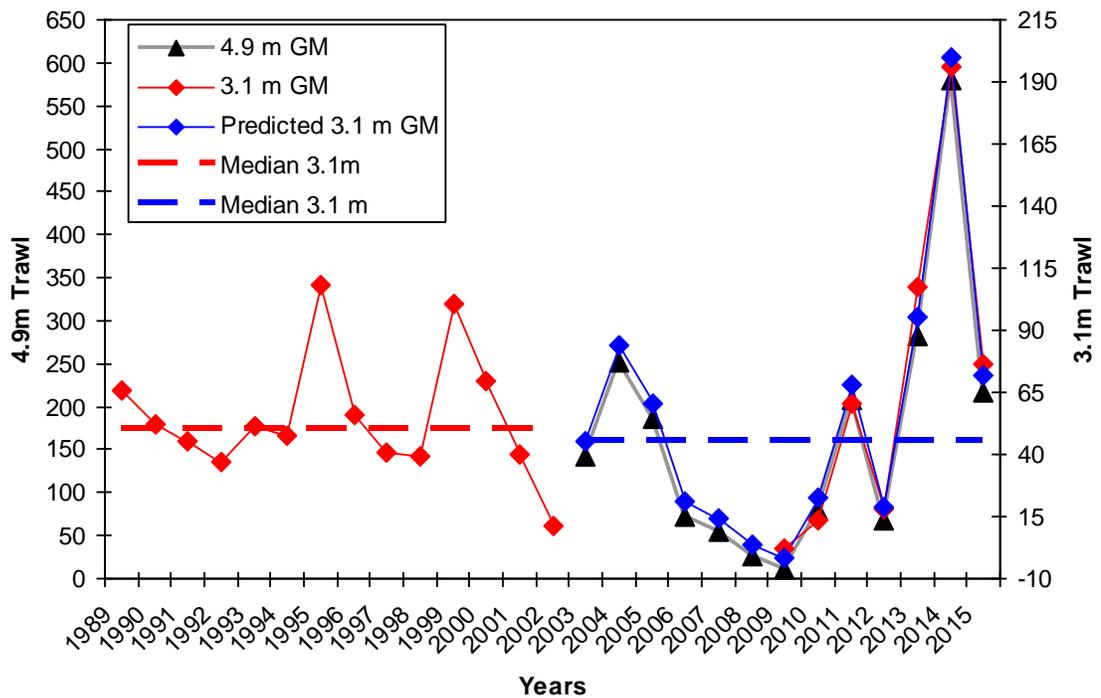


Figure 3-14. Species richness (number of species) in Mattawoman Creek 3.1 m trawl samples during summer sampling. N = 12 for all points. Bimonthly sampling during 2009-2015 allowed for two estimates of N = 12 per year. Median number of species during 1989-2002 is indicated by the green line; median number of species during 2009-2015 is indicated by the red line.

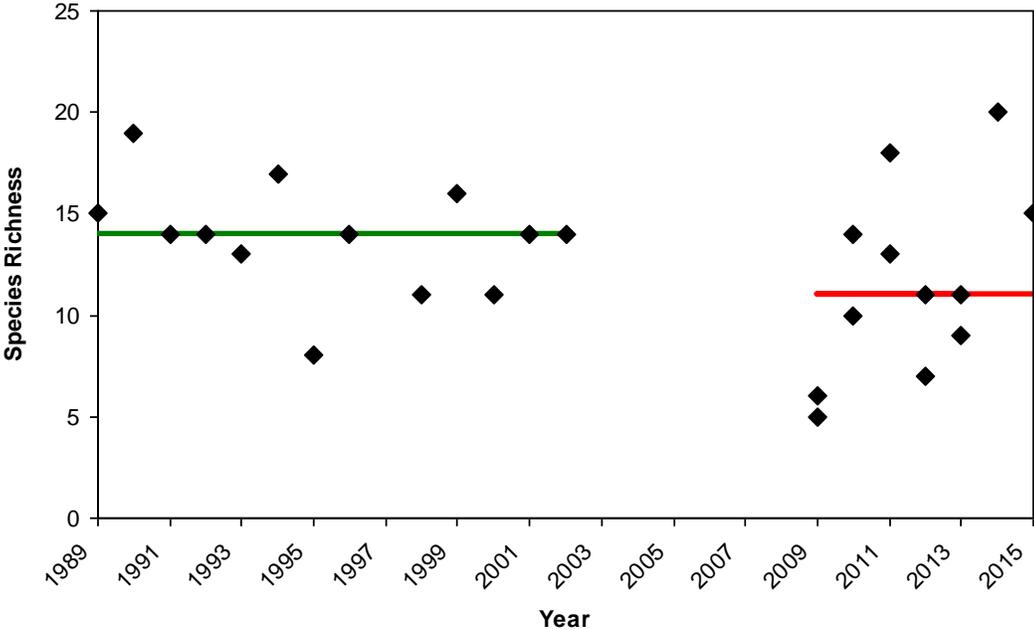


Figure 3-16. Map illustrating Maryland Department of Planning land use categories (2010) for the lower Choptank River subestuaries, Harris Creek, Broad Creek, and Tred Avon River.

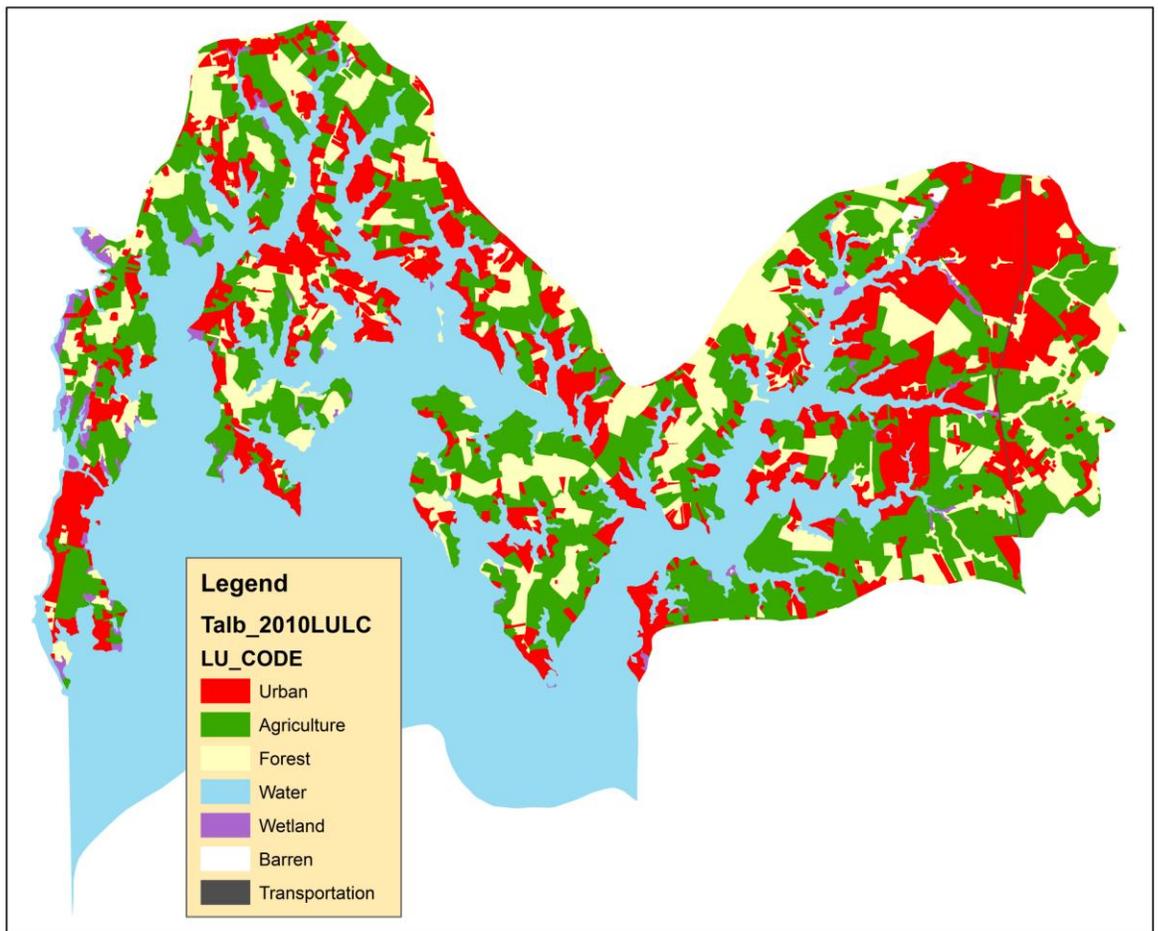


Figure 3-17. Trends in development in watersheds of three adjacent subestuaries in the Choptank River drainage, 1950-2013.

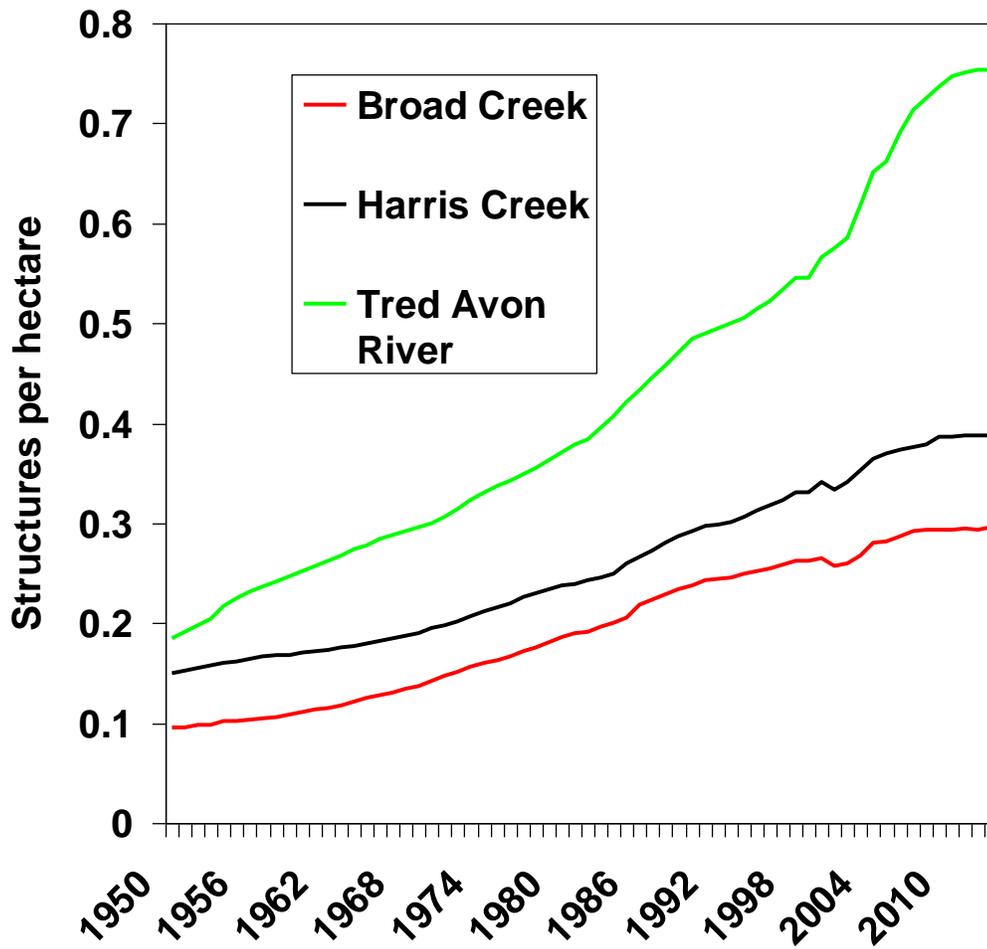


Figure 3-18. Bottom dissolved oxygen (mg/L) readings versus intensity of development (C / ha = structures per hectare) for Tred Avon River 2006 – 2015 and Harris and Broad Creeks 2012 – 2015.

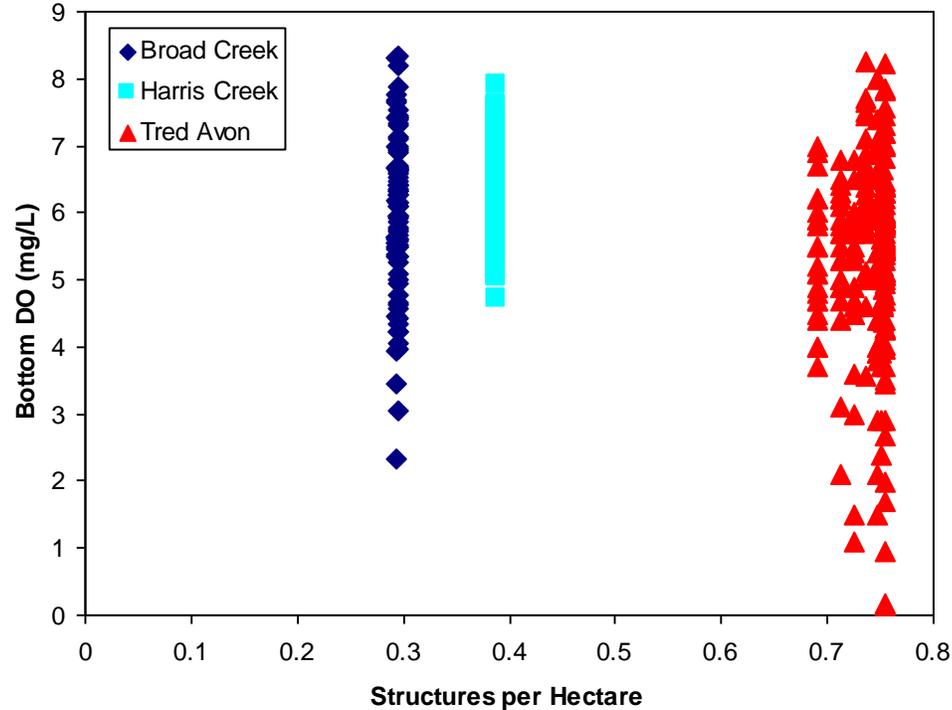
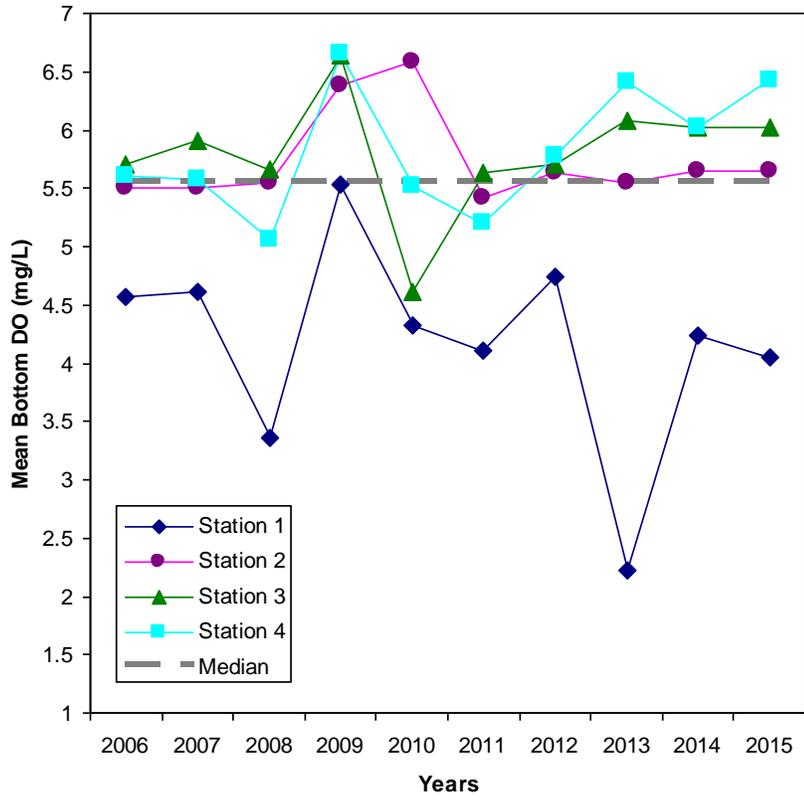


Figure 3-19. Mean bottom dissolved oxygen (mg/L) in Tred Avon River, by station, 2006 - 2015. Dotted line indicates the median of the time-series of annual means.



## **JOB 2: Support multi-agency efforts to assess finfish habitat and ecosystem-based fisheries management.**

Jim Uphoff, Margaret McGinty, Alexis Park, Carrie Hoover, Patrick Donovan, Charles Johnson, and Shaun Miller

### **Introduction**

The objective of Job 2 was to document participation of the Fisheries Habitat and Ecosystem Program (FHEP) in habitat, multispecies, and ecosystem-based management approaches important to recreationally important finfish in Maryland's Chesapeake Bay and Atlantic coast. Activities in this job used information generated by F-63 in communication and fisheries management or were consistent with the goals of F-63. Contributions to various research and management forums by Program staff through data collection and compilation, analysis, and expertise are vital if Maryland is to successfully develop an ecosystem approach to fisheries management.

### **Maryland Fisheries Service – Fisheries Habitat and Ecosystem Program Website**

We continued to populate the website with new reports and information to keep it up to date with project developments. The web site was redesigned in April 2015 to help with navigation. Currently, we are working on compiling reports and presentations to add to the Reports, Presentations, and Publications link on the FHEP website.

### **Publications**

Uphoff, J.H., Jr. and Co-Authors. 2015. Marine and estuarine finfish ecological and habitat investigations. Performance Report fro Federal Aid Grant F-63-R, Segment 4. Maryland Department of Natural Resources, Annapolis, Maryland.

### **Environmental Review Unit Bibliography Database**

FHEP staff continues to compile an Environmental Review Unit database, adding recent literature and additional topics including effectiveness of Best Management Practices.

### **Review of County Comprehensive Growth Plans**

M. McGinty reviewed and commented on several comprehensive growth plans including Harford County, Queens Anne's County, Myersville and St. Michaels. Part of this effort included developing an assessment of fishing license sales by county. This analysis will be revisited in the coming year, to incorporate fishing license sales into spatial prioritization tools. These efforts also included an assessment of local fisheries resources that represent recreational opportunities and the importance to consider fish habitat protection in planning.

### **MD DNR Interagency Effort on Mattawoman Creek-**

FHEP has continued to work with other DNR units and Charles County on its Comprehensive Development Plan (CDP). Charles County has moved towards zoning that will help conserve fisheries. A major portion of Mattawoman Creek's watershed that was slated for future development was downzoned and will not be subject to intense development. Projections of development in Mattawoman Creek's watershed at build-out indicate that measures proposed in CDP will greatly lower growth from the previous plan. The county will be devoting a considerable amount of resources towards

stormwater management in the watershed in order to meet the Total Maximum Daily Load requirements of the Chesapeake Bay Program. As it now stands, these are very favorable changes for recreational fishing in Mattawoman Creek.

### **Cooperative Research**

M. McGinty, A. Park, C. Hoover, P. Donovan, C. Johnson, S. Miller, and B. Redding supported field sampling efforts of various state and federal projects including: the DNR's Coastal Bays Program, Resident Fish Species, Fish Passage Program, the Alosid Project, Resource Assessment Services, Artificial Reefs Initiative, Hatcheries Division Program, Striped Bass Program, Shellfish Monitoring and Assessment Program, and the Fish Health Program.

J. Uphoff, A. Park, C. Hoover, P. Donovan, S. Miller, and B. Redding collaborated with the Fish Health Program at the Oxford Lab assessing Striped bass stomach contents collected from the upper, middle, and lower Chesapeake Bay.

M. McGinty, A. Park, C. Hoover, P. Donovan, C. Johnson, S. Miller, and B. Redding worked with Resource Assessment Services on building housing units for additional conductivity data loggers that were deployed throughout Mattawoman Creek to continuously record conductivity measurements over a period of time.

J. Uphoff, M. McGinty, A. Park, and C. Hoover collaborated with the Hatcheries Division Program to collect additional ichthyoplankton samples in the Choptank River to investigate spawning areas of Alosid species.

M. McGinty assisted in the 10 year review for the State Wildlife Grant, Species of Greatest Conservation Need (SGCN). No tidal species were listed in the previous assessment. M. McGinty reviewed the regional list to determine if the status of these regional species warrants listing them in need of conservation.

A. Park attended the Hatcheries Division Pre-Production meeting. FHEP staff will coordinate with hatcheries and will keep samples from the Choptank River for additional fish larvae/egg identification by USFWS.

A. Park and C. Hoover assisted the Coastal Bays Program with the highly migratory species tagging and data collection at the Mid Atlantic Fishing tournament held in Ocean City.

J. Uphoff, M. McGinty, A. Park, C. Hoover, P. Donovan, and C. Johnson assisted Resource Assessment Services Zebra Mussel monitoring program. Samples were provided from Middle River. The mussels were positively identified as both Zebra Mussels and Dark False Mussels. This discovery was evidence of the spread of Zebra Mussels in Chesapeake Bay.

J. Uphoff and M. McGinty worked with NOAA and TNC on linking Choptank River land use practices to fish habitat in order to set priorities for restoration activities. Choptank River has been chosen by NOAA as a habitat focus area (HFA), one of five nationwide.

J. Uphoff and M. McGinty guided development of an HSI model for the Choptank River, meant to develop a tool to target restoration. They presented information from data evaluation showing that the Choptank River is still supporting healthy habitat for target species.

M. McGinty collaborated with CBNERR staff to discuss how the citizen science data is being used in Fisheries Assessments. Data collected over the past decade, have

allowed Fisheries staff to determine that land use change has caused a decline in spawning habitat occupation. M. McGinty guided development of and reviewed a poster using these data to be presented at the nation NERRs meeting.

M. McGinty participated in a workgroup developed to expand Anne Arundel County's Habitat Protection Areas. This workgroup formed in response to citizen concerns over losses in habitat in urban rivers and the effects of future development on recovery of lost habitat. Staff responded to citizen inquiries regarding regulatory protections of Yellow Perch habitat in the Magothy River. Staff applied study results to inform citizens of the potential to recover habitat and steps they can take to protect the present stock. This information is being examined to determine how it can support expansion of Habitat Protected Areas.

M. McGinty met with consultants engaged in developing an Environmental Sensitivity Inventory for NOAA to be used to determine responses to oil spills. Staff provided data and reviewed maps to refine habitat designations for anadromous spawning areas.

### **Presentations and Outreach**

*Managing Chesapeake Bay's Land Use, Fish Habitat, and Fisheries* presented at the Restore America's Estuaries Conference at National Harbor. J. Uphoff represented a state perspective in a panel discussion on fish habitat at the Restore America's Estuaries conference. Other panelists represented NMFS, the Mid-Atlantic Fisheries Management Council, and the ASMFC. Our presentation was focused on management experience and habitat monitoring and analysis. Others were more focused on conceptual issues.

*Tracking and Understanding Changes: 25 Years of Monitoring Mattawoman Creek (and more)* presented at the 20<sup>th</sup> Annual Maryland Water Monitoring Annual Conference, M. McGinty, presenter.

*Managing Chesapeake Bay's Land Use, Fish Habitat and Fisheries: Developing and Applying Impervious Surface Reference Points* presented as a DNR noon seminar, J. Uphoff, presenter.

*Multispecies Fisheries Management* presented to the Perryville MSSA Chapter, J. Uphoff, presenter.

*Development and Chesapeake Bay's Fish Habitat and Fisheries* presentation for Chesapeake Bay Policy class at Salisbury State University, J. Uphoff, presenter.

*Quantifying development's effect on management: yellow perch example and future management questions* presented at DNR Bay Policy Group, J. Uphoff, presenter.

*Fish Habitat Outcome: What Can Local Governments Do?* presented at the Chesapeake Bay Program Local Government Advisory Committee, Margaret McGinty, presenter.

J. Uphoff attended a forum on Growth and the Future of Chesapeake Bay and was part of a panel discussing fish habitat development.

A. Park presented on summer sampling results and illustrated what summer juvenile fish sampling involved for the Anita C. Leight Estuary Center staff and volunteers. The Bush River is one of FHEP's sampling areas and has been sampled since 2006 by staff and volunteers. The volunteer group samples the Bush River and provides data to FHEP staff.

M. McGinty and C. Hoover assisted with the Explore and Restore Maryland Coastal Streams workshop held at the Somerset Intermediate School. Staff instructed participants about biological indicators of health, in part, through catches in fish traps and via seining, and gave an introduction to fish ecology and coastal fish populations. The potential influences of various land-use types were discussed, and the biodiversity of fish caught was assessed in that context.

### **Program and Staff Development**

A. Park and C. Hoover attended the Bleiker Auto Conference titled “How can we involve more people? As many people as possible?”

A. Park and C. Hoover attended the Vessel Safety and Training Course, conducted by Capt. Mike Simonsen at Oxford. The training allows staff to assist with research projects being conducted on the NOAA Research Vessel R5502.

P. Donovan attended a training titled Visual Storytelling for Environmental Non-profits. The training covered approaches to educating the public about environmental issues using ESRI Story Maps.

### **ASMFC**

J. Uphoff attended meetings of the ASMFC’s Biological and Ecological Reference Point Work Group to discuss ecological reference points for Atlantic menhaden. The ASMFC work group recognized that menhaden management requires additional precautions because of their importance as forage. A biomass dynamic model that includes Striped Bass and Spiny Dogfish predation developed by J. Uphoff is being considered for development of forage reference points. Indicator approaches developed in Job 4 are of interest as well.

M. McGinty attended the spring ASMFC Habitat Committee meeting. Issues discussed included a status review of a habitat bottleneck whitepaper, review of habitat fact sheets, fish passage database, inclusion of habitat considerations in ecosystem-based fisheries management process, impacts and mitigation for coral and seagrass due to a port expansion project in Florida, and a spiny lobster/stone crab trap modification study to reduce movement and habitat impacts.

### **Fisheries Habitat Workgroup**

M. McGinty is currently leading meetings of the Fisheries Habitat Workgroup as MD DNR support staff. The following vision was adopted by the workgroup members: *Protect and restore fisheries habitat, using ecosystem-based management and practical understanding of watershed ecology, to educate and engage people and influence decisions and policies, respecting all voices.* The workgroup members worked on developing goals, elected a workgroup chairman, and reached out additional individuals who were not in attendance to gauge their interest in participating in the workgroup. Currently, the workgroup is meeting monthly.

The workgroup proposed adopting Jones Falls as a focus area. A subset of the workgroup will meet to develop a strategy to promote fish habitat conservation and restoration in the Jones Falls Watershed. The workgroup also identified several habitat subjects for which they needed more information. One attendee identified the need to develop county Fisheries Advisory Groups.

The workgroup has provided members with a number of presentations to help provide insight and guidance into planning, promoting habitat conservation, research, etc. The workgroup members invited Theresa Moore from Valleys Planning Council and Amy Owsley from Eastern Shore Land Conservancy to talk about their work and how they promote conservation in their areas. Beth McGee from Chesapeake Bay Foundation and Elliot Campbell from MD DNR Integrated Policy presented their work and the concept of Ecosystem Services in Maryland and the Chesapeake Bay. Scott Stranko presented information and data on the effects of road salting in Maryland.

### **Bay Agreement**

M. McGinty and J. Uphoff attended the Bay Cabinet meeting to understand what the expectations were for the MD leads for developing strategies to achieve the goals identified in the new Bay Agreement.

J. Uphoff attended the STAC Forage Fish workshop where fishery managers discussed monitoring programs can be integrated to provide insight in Maryland's portion of the Bay and address the new Bay agreement forage fish objective.

J. Uphoff developed seven indicators to address the forage fish outcomes of the new Bay Agreement, "By 2016, develop a strategy for assessing the forage fish base available as food for predatory species in the Chesapeake Bay." Striped bass is the predator of concern in Maryland's portion of the Bay and Atlantic menhaden, bay anchovy, and spot were important forage fish. Condition (proportion without body fat in fall) and relative survival of striped bass described whether striped bass ate enough forage fish; a September-October recreational catch per trip index indicated relative abundance of resident striped bass, and ratios of the three forage species (individually) to striped bass relative abundance and the grams of prey consumed per gram of striped bass indicated intensity of forage-to-predator relationships. These indicators were rated in the most recent year they were available and on their three year trend. This approach was reviewed and accepted by Maryland's GIT representatives. A prototype indicator-based approach for assessing forage fish based on resident striped bass diets in Maryland's portion of Chesapeake Bay was presented to the Bay Program forage fish outcome team. This approach integrates existing monitoring and could be implemented cheaply (perhaps with no additional cost). It seemed to be acceptable, especially since no one else had an approach to talk about and probably 2/3 of the group did not participate in the call. Narrative was forwarded to NCBO for them to work with. Hopefully, parts can be used, and missing aspects identified and filled in quickly.

M. McGinty outlined a strategy for Maryland's effort to support the Chesapeake Bay Agreement, Habitat Goal. This approach involves identifying key sentinel species that represent various habitats and ecoregions in the Bay, mapping them according to their natural limiting factors and applying know stressor information to develop spatial tools. M. McGinty continues to serve as the Maryland lead on the team to support development of regional approaches to identify effective management strategies design to conserve and restore fish habitat. This requires monthly meetings and communications to develop work plans, review public comment, shape research proposals and track progress toward meeting the 2017 milestones outlined in the plan.

M. McGinty participated in several meetings to explore criteria to develop Healthy Watersheds designations in Maryland, to support the Healthy Watersheds Goal.

The priority fish habitat map was referenced in the discussion to include fish habitat needs, but the watershed scale used in the approach was considered too large and intractable. Presently, Healthy Watersheds are designated at a small stream catchment scale. This is considered practical to assure protection through regulations. We address this issue in the Mapping section of this report, noting that stressors influence fish habitat at all scales, and if we want to protect large tracts of anadromous spawning habitat, we will have to conserve large tracts of rural lands to assure watersheds do not exceed the impervious target developed by our program.

### **JOB 3: Developing Priority Fish Habitat Spatial Tools**

Margaret McGinty, Carrie Hoover, Alexis Park, Patrick Donovan, and Jim Uphoff

#### **Introduction**

As the human population in the Chesapeake Bay watershed increases, we need to understand its effects on aquatic systems. Identifying and quantifying stressors associated with development that can be applied as stressor criteria in a visual spatial framework will promote more effective management of habitat and fisheries. In 2015, we continued to monitor conductivity in spawning areas to determine if there was potential to develop conductivity criteria for anadromous spawning habitat mapping based on background levels reported by Morgan et al. (2012). Additionally, we examined the influence of watershed scale on dissolved oxygen limits to fish, to determine if scale influences biological habitat conditions to verify the appropriate scale for mapping watersheds to prioritize management actions. We present results of this effort by topic in the following narrative.

#### **Conductivity Criteria for Anadromous Fish Spawning**

##### **Introduction**

Increased salinity of freshwater due to human activities is one stressor gaining attention from researchers and resources managers. Cañedo-Argüelles et al. (2013) described salinization of rivers as a “global and growing threat” likely to be exacerbated by climate change. Sources of salt include irrigation, sewage effluent, mining and road de-icing practices. Salt input from road de-icing is gaining more attention in Maryland (Stranko et al. 2013). According to the Cary Institute, salt was used as a de-icing agent in the United States as early as 1938 (Kelly et al. 2010). In the early 1940’s national application rates were estimated at 5,000 tons annually (Kelly et al. 2010). Today, these estimates are 10 to 20 million tons per year (Kelly et al. 2010). Kelly et al. (2008) reported that road salt accounts for 91% of salt input in a watershed. Salt enters streams through direct surface runoff and groundwater; salt can persist in groundwater for decades (Kelly et al. 2008; Harte and Trowbridge 2010; Perera et al. 2013). The dominant form of salt used in deicing is NaCl (Kelly et al. 2010), so research has been directed to evaluate the impacts of increased NaCl in freshwater, as well as effects of chloride as it dissociates and become potentially toxic to freshwater organisms. Elevated conductivity, related primarily to chloride from road salt (but including most inorganic acids and bases; APHA 1979), has emerged as an indicator of watershed development (Wenner et al. 2003; Kaushal et al. 2005; Morgan et al. 2007; Morgan et al. 2012). Use of salt as a deicer may lead to both “shock loads” of salt that may be acutely toxic to freshwater biota and elevated baselines (increased average concentrations) of chloride that have been associated with decreased fish and benthic diversity (Kaushal et al. 2005; Wheeler et al. 2005; Morgan et al. 2007; 2012).

Uphoff et al. (2010) observed increased conductivity in freshwater reaches of spawning habitat for River Herring in Mattawoman Creek. They evaluated historical records to establish a background level, with highest concentrations upstream in the vicinity of the urban center of the watershed (see Job 1, Section 1). They established a correlation between increased conductivity and absence of spawning, and identified

increased conductivity as a symptom of urbanization in streams supporting anadromous fish spawning in Maryland (Uphoff et al. 2015).

In 2014, we deployed continuous monitors to continuously measure conductivity in four watersheds known to support Herring spawning: Mattawoman Creek, Piscataway Creek, Deer Creek and Bush River (see Job 1, Section 1). We deployed four additional meters in Mattawoman Creek in June, 2015, to determine variability of conductivity in relation to subwatershed road density. We were interested in defining the persistence of increased conductivity after anadromous fish spawning season to determine if these continuous data could provide insight into how watersheds were changing and guide us in establishing conductivity criteria for anadromous fish spawning streams. Once established, these criteria could be applied in a targeting approach to identify areas for conservation or targeted actions to curb practices that attribute to increased conductivity in spawning areas.

### Methods

*Mainstem Tributary Comparisons* - We deployed four HOBO™ continuous conductivity monitoring meters in four streams at their lowest nontidal Herring spawning sampling station (Figure 1). Two adjacent streams in the Coastal Plain and two in the Piedmont region were sampled. Each pair of streams represented less and more developed watersheds, although degrees of development were not the same for each pair. Each site chosen for continuous conductivity monitoring was sampled in the 1970s by O'Dell et al. (1975, 1980) and again during 2005-2015 (Uphoff et al. 2015). Availability of trained volunteers to handle sampling of Herring eggs and larvae and water quality was an important factor in choosing these streams for spawning surveys conducted during 2005-2015 since project personnel were not available to conduct sampling. These four streams continued to support Herring spawning, but the amount was related to development intensity (Uphoff et al. 2015; see Job 1, Section 1).

In the Coastal Plain, we sampled Mattawoman and Piscataway creeks, two streams in the Washington, DC, growth region. Mattawoman Creek is an urbanizing watershed in northern Charles County that has developed to early suburban density. Structure density in the watershed is estimated at 0.914 per hectare (C/ha) which is equivalent to 10.38% impervious cover (Figure 2). This exceeds the development threshold of 0.83 C/ha (10% impervious) that represents a fish habitat threshold (Uphoff et al. 2011a; Uphoff et al. 2015). Piscataway Creek is located nearest Washington, DC, in Prince Georges County, Maryland. Development in the watershed is presently at 1.49 C/ha (14.12% impervious cover; Figure 3). In the Piedmont Region, we compared Deer Creek to Bynum Run, a tributary of Bush River. These streams are in the growth path of Baltimore. Deer Creek, furthest from Baltimore, is a rural watershed in northern Harford County that empties into the Susquehanna River. Development in the watershed is at 0.24 C/ha (4.48% impervious cover), below the 0.27 C/ha (5% impervious cover) target (Uphoff et al. (2011a; Uphoff et al. 2015; Figure 4). Bynum Run is the largest stream in the Bush River watershed without blockages to impede upstream migration of Herring. Bush River has multiple streams flowing into the watershed as opposed to one major stream. The Bush River watershed is presently developed at 1.51 C/ha, equivalent to 14.21% impervious cover (Figure 5). All meters were deployed at the end of May, 2014, and remained in place through December, 2015, except for the meter at Mattawoman Creek (this site was moved upstream after the site showed evidence of tidal influence in May 2015).

*Mattawoman Creek Continuous Monitoring and Discrete Stations* - We deployed one Onset HOBO™ continuous conductivity monitoring meter in each of four subwatershed tributaries to Mattawoman Creek based on density of roads in the subwatersheds (Figure 6). Additionally, citizen scientists with the Mattawoman Watershed Society committed to sampling conductivity once a month at eight stations along the mainstem of the Creek and five tributaries (Figure 7).

We estimated density of roads and selected two sites in separate subwatersheds that reflected a low density network and two sites in separate subwatershed that reflected a high density network of roads. We obtained a 2012 shapefile of road centerlines and updated it using Google Earth imagery to include housing development that has occurred since the centerline shapefile was created. Road centerlines were clipped using a Mattawoman Creek watershed polygon shapefile. Road centerlines were then buffered on both sides using three different lane widths: 10, 15, and 20 feet. Lanes are typically 10 to 12 feet wide, but since centerlines were not categorized by road type wider lanes were calculated to account for multi-lane roads and on-street parking. Buffer polygons were merged to make a single large polygon. Watershed areas of interest were then used to clip the road buffer polygon shapefile. We calculated road density by subwatershed by dividing total road coverage by watershed area.

The Onset HOBO™ instruments were deployed in fabricated PVC housings and anchored on the bottom of the stream bed using large rocks attached to cables. They were secured to trees on the bank with cable to assure they would not be dislodged. Instruments were programmed to record conductivity and temperature on an hourly basis. Sites were visited every four to six weeks to download data and assure instruments were not dislodged and were still functioning. Meters were cleaned as needed and redeployed. Temperature and conductivity were recorded using handheld YSI meters at the time of meter deployment and when data were downloaded to account for any drift in the meter during deployment. Data were also observed for anomalies related to possible dewatering events. Instantaneous measures were recorded monthly by citizen scientists using handheld YSI Pro2030™ meters.

Continuous monitoring data were plotted and compared among the four tributary stations. Conductivity data from the four study watersheds were plotted against the appropriate Morgan et al. (2012) background levels for Piedmont and Coastal Plain streams in Maryland. Morgan et al. (2012) provided two sets of methods of estimating spring base flow background conductivity for two different sets of Maryland ecoregions, for a total set of four potential background estimates. We chose the option featuring Maryland Biological Stream Survey (MBSS) Coastal Plain and Piedmont regions and the 25<sup>th</sup> percentile background level for conductivity (109 and 150 uS/cm, respectively). We developed a mean conductivity to reference conductivity (MCRC) ratio by dividing the spawning season mean conductivity by the Morgan et al. (2012) background level for each ecoregion. We plotted annual estimates of  $P_{herr}$  at each site against the MCRC ratio to see if this ratio had the potential predict  $P_{herr}$  (regression analysis would follow) or provide a breakpoint between high and low levels of  $P_{herr}$ .

An ANOVA was used to explore differences in conductivity among stations on Mattawoman Creek including the continuous monitoring and discrete sampling stations. Tukey's Studentized Range (HSD) Test was applied to determine which stations were significantly different. Level of significance was set at  $\alpha = 0.05$

We also examined conductivity data recorded concurrently with ichthyoplankton samples to determine if we could derive a conductivity limit associated with proportion of samples with Herring present ( $P_{herr}$ ). Refer to Job 1, Section 1 for details on estimating  $P_{herr}$ . We examined data for the period of record and observed Herring presence was positive between March and May. We only used the site where the conductivity meters were located which represented the lowest site in the watershed. In the Bush River, we used the site on Bynum Run, because it was the largest stream in the watershed that did not have a stream blockage and was most similar to sites on the other streams. We calculated  $P_{herr}$  by year and station and calculated mean conductivity from March through May. We plotted  $P_{herr}$  against mean conductivity to determine if there was a conductivity limit that predicted a change in  $P_{herr}$ .

### **Results and Discussion**

*Mainstem Tributary Comparisons* - Measurement of conductivity from continuous monitors in Deer Creek, Mattawoman Creek, Piscataway Creek, and Bynum Run usually exceeded Morgan et al. (2012) background criteria (Figure 8). These levels were only met 0.09% to 3.6% of the time throughout the year suggesting that these criteria may not be directly applicable to Herring spawning habitat (Table 1). All four streams generally showed the same annual pattern, where highest conductivities were observed in the winter when road salting occurs to treat frozen pavement. They declined in the spring when Herring spawning began (Figure 8). In the two most urbanized watersheds, mean conductivity was greater when estimated in spring (March through May) than annually (Table 2). Spring and annual means were similar in the two less urban watersheds. Ranges of conductivity were narrower in spring samples than annual samples (Table 2), indicating that conductivity was more stable during the spawning season.

Deer Creek's means were similar to Mattawoman Creek's, Piscataway Creek's means were similar to and Bush River's, but the means of the latter two more heavily developed where higher than the means from the two less developed streams (Figure 9). The MCRC ratio increased with development (Figure 10). This is consistent with plots of mean annual spawning season conductivity that was standardized to the same ecoregion reference values for all stream anadromous fish spawning sites against development (Figure 1-11 in Job 1, Section 1).

The plot of MCRC ratio against  $P_{herr}$  suggested a breakpoint at an MCRC ratio of 1.56 for two regions: one where  $P_{herr}$  was usually 0.8 or higher (10 of 12 estimates) and one where it was consistently lower (8 of 10 estimates; Figure 11). This breakpoint was viewed as a threshold for mean conductivity during spawning. When viewed for whole stream estimates of mean conductivity and  $P_{herr}$  (see Figure 1-12B in Job 1, Section 1), values of  $P_{herr}$  (present in Deer and Mattawoman creeks only) in the highest category were only found near or below the MCRC breakpoint of 1.56 identified in this analysis.

The MCRC ratio limit can be used to target watersheds to identify specific issues for management or enforcement actions. To demonstrate this, we acquired conductivity data from the Maryland Biological Stream Survey from 1995 to 2014 (MBSS; Scott Stranko, MD DNR, personal communication). The MBSS applies a probabilistic sampling design to assess streams in Maryland on a five year rotating basis (Mercurio et al. 1999). We pooled the entire MBSS data set, calculated the mean spring conductivity by Maryland's 8-digit subwatershed, and estimated the MCRC ratio. We chose the 8-digit

scale because it is the same scale used to develop priority spawning habitat maps (Uphoff et al. 2013). We applied the proposed MCRC ratio limit of 1.56 and identified subwatersheds exceeding this level. We overlaid these maps with the historical maps documenting Herring spawning areas (O'Dell, 1975, 1980) to show where spawning habitat was and was not meeting the MCRC ratio limit (Figure 12). We can take this map and overlay the priority watersheds to determine which watersheds may need some directed management to address elevated conductivity. Figure 13 shows the high priority watersheds targeted for conservation and restoration with the target conductivity applied. This overlay reveals areas where the proposed MCRC ratio limit is exceeded and offers opportunities to focus management practices such as limiting inputs or minimizing road salting operations. We can also overlay conductivity data to identify “hot spots” where conductivity was elevated compared to other stations. Figure 14 shows an example where conductivity data are plotted in the Choptank River. Figure 15 identifies three areas where conductivity is highest. With this information, we used Google Earth to hone in on areas and determine if there are any discernible features that could be contributing to elevated conductivity. Since the conductivity data are georeferenced, we can go directly to the site and view the area around the site to identify potential sources (Figure 16). At this view, there appeared to be a commercial operation that could be a potential source of higher conductivity effluent. The next step would be to determine if this operation has an effluent permit and determine if the permitted use could potentially be contributing a substance that could change conductivity. This approach demonstrates how a conductivity limit could be used to target management action. A MCRC ratio limit could also be applied in a regulatory framework to classify and protect spawning areas from land use change and operations that degrade habitat.

*Mattawoman Creek Continuous Monitoring and Discrete Stations* - The two sites in Mattawoman Creek subwatersheds dominated by forest (MUT3 and MUT4) had much lower conductivity during 2015 than sites in developed areas with high road densities (Figure 17). This suggests that background levels in a forested landscape are much lower. Uphoff et al. (2010) mined data to establish historical conductivity conditions between 1970 and 1991 and reported conductivity increased toward urban centers, but generally, conductivity measurements less than 100  $\mu\text{S}/\text{cm}$  were common above and below Waldorf (see Figure 1-8 in Job 1, Section 1). O'Dell et al. (1980) included tables in their reports with conductivity data; conductivity in the Bush River was much lower ( $\sim 100 \mu\text{S}/\text{cm}$ ) than presently observed.

We examined pooled monthly conductivity data (April-December, 2015) from Mattawoman Creek collected by citizen scientists to determine if station means were different using ANOVA (Table 3). The ANOVA model confirmed that means were different among stations ( $F=7.39, p<0.0001$ ) and the means test indicated that elevated means existed at stations with higher subwatershed road densities and elevated conductivity persisted downstream even though road densities became lower (see Figure 7). A box and whisker plot and Tukey's groupings indicated that the two stations upstream of watersheds with high road densities had mean conductivities closer to 100  $\mu\text{S}/\text{cm}$ , while high road density stations and downstream low road density stations had means closer to 200  $\mu\text{S}/\text{cm}$  (Figure 18).

We will continue to explore the effects of conductivity on anadromous fish spawning habitat in Maryland. To date, we have documented increases beyond historical

levels and associated increased conductivity with development in the landscape and decreased  $P_{herr}$  (Uphoff et al. 2015). We have also demonstrated an approach to develop a conductivity limit for spawning habitat which can be used to identify potential hot spots and sources of elevated conductivity in a watershed. We plan to expand our effort in 2016 to collect water chemistry samples that will be analyzed to identify the specific ion responsible for increased conductivity. We are exploring the potential to collaborate with a study group to develop bioassay tests for Herring eggs. This would determine if increased conductivity is responsible for direct mortality to eggs, or is just an indicator of habitat change in the watershed that is contributing to observed declines. We will also continue to monitor Mattawoman Creek with continuous monitoring and discrete samples to determine if data can be used to localize sources of elevated conductivity within the watershed.

## **Exploring the Influence of Land Use and Watershed Scale on Dissolved Oxygen Limits**

### **Introduction**

Geographic information systems (GIS) have long been important to natural resource applications (Becco and Brown 2013). Managers depend on mapping resources to better manage and target management to conserve and restore vital habitats. These resources vary by scale, depending on the management focus. Fisheries managers seeking to conserve and restore Brook Trout habitat may work at the scale of a small stream segment or a catchment (Rosi-Marshall et al. 2006), while managers seeking to address habitat challenges for migratory fish like Striped Bass need to consider changes on large regional scales (example Coutant 1990). Both conservation and restoration practices are scale dependant. Over the last two decades attention to scale has increased, with greater understanding of the need to consider larger scale processes and how they influence local habitats and organisms (Levin 1992; Allan et al. 1997). This evolution of understanding is challenged by programs and policies that limit focus of actions to localized scales. In particular, spatial tools created to designate management priorities for migratory anadromous spawning fish like Striped Bass promote management priorities at large watershed scales, while land conservation programs target small parcels of land within these large watersheds. In promoting the anadromous spawning priority tool (Uphoff et al. 2014), we have received requests to scale it down to demonstrate the value of small scale restoration and conservation efforts. We contend that we have scaled it down to the smallest scale practicable that will maintain the integrity of the tool, and we wanted to explore how scale influences habitat response to stressors.

### **Methods**

We chose to focus on dissolved oxygen (DO) responses to land use change. Uphoff et al. (2011a) demonstrated the likelihood of low oxygen in small mesohaline watersheds (7,000 to 60,000 hectares). We examined three large scale watersheds (175,000 to ~2.6 million hectares; Table 4) to determine if we could mimic a similar response of DO to increased development in smaller mesohaline watersheds examined by Uphoff et al. (2011a). We selected three large Chesapeake Bay mesohaline tributaries based on their dominant land use (Figure 19). The James River in Virginia is dominated by forest (71% of the watershed; James River Association 2016), that served as the forested treatment. The Patuxent River in Maryland is approaching the 10% impervious

surface threshold while losing natural land (Patuxent River Commission 1997). The Choptank River, located on the Eastern shore of Maryland, is dominated by agriculture (Maryland Department of Natural Resources, Surf Your Watershed <http://www.dnr.state.md.us/watersheds/surf/>).

We retrieved water quality data from the Chesapeake Bay Program Data hub for each river (<http://www.chesapeakebay.net/data>). We examined the last ten years of data available for each site in the mesohaline area of the estuary. We examined bottom DO conditions in July through September and used box and whisker plots to compare distributions among watersheds. Bottom DO was most likely to reflect nutrient levels and processing in summer (Uphoff et al. 2011a). We used the DO target and limit used by Uphoff et al. (2011a) to evaluate conditions.

### **Results and Discussion**

The predominately forested James River watershed had the best mesohaline bottom DO conditions, followed by the agriculture dominated Choptank River, and worst condition were found in the urbanizing Patuxent River (Table 6). Four sites fell within the mesohaline zone of the lower James River (Figure 20). James River bottom DO was generally above the 5 mg/L target. The Choptank River (Figure 21) had two mesohaline stations. The uppermost station in Figure 21, ET5.1, is not within the mesohaline region, but we included it to visualize the gradient of oxygen conditions from up to downstream. Bottom DO conditions fell below the 5.0 mg/L target, but most measurements were above 4.0 mg/L with a few measurements below the 3.0 mg/L target. (Table 6; Figure 21). Bottom DO in Patuxent River (with four mesohaline stations; Figure 22) improved with downstream distance, but station medians did not meet the target and were generally below the DO limit (Table 6).

These results are consistent with the work we have been conducting over the last decade (Uphoff et al. 2011a; 2011b; 2012; 2013; 2014; 2015). This suggests that impervious thresholds established at the smaller watershed scale should apply at the larger watershed scale. It also emphasizes the importance of continuing to promote conservation of rural landscapes in key fish habitats to assure these habitats continue to thrive and promote fish production.

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Table 1. Percentage of conductivity measurements exceeding the background level of Morgan et al. (2012) for Coastal Plain and Piedmont streams in Maryland.

| Ecoregion     | Background $\mu\text{S}/\text{cm}$ | River      | n     | # Samples > Background | Percent > Background |
|---------------|------------------------------------|------------|-------|------------------------|----------------------|
| Coastal Plain | 108.8                              | Mattawoman | 12644 | 12536                  | 99.1                 |
| Coastal Plain | 108.8                              | Piscataway | 13247 | 13130                  | 99.1                 |
| Piedmont      | 150.3                              | Deer       | 12835 | 12438                  | 96.9                 |
| Piedmont      | 150.3                              | Bush       | 12839 | 12376                  | 96.4                 |

Table 2. Anadromous fish spawning season and annual means and ranges of conductivity ( $\mu\text{S}/\text{cm}$ ) for the four Streams sampled.

| Stream                 | Spawning Season |              | Annual |             |
|------------------------|-----------------|--------------|--------|-------------|
|                        | Mean            | Range        | Mean   | Range       |
| Bynum Run (Bush River) | 411             | 150.4-1213.7 | 326.2  | 32.8-1349.5 |
| Deer Creek             | 205.4           | 130.1-422.0  | 198.7  | 71.6-422.0  |
| Mattawoman Creek       | 194             | 91.8-422.1   | 195.5  | 56.5-762.9  |
| Piscataway Creek       | 376             | 100.3-2401.4 | 274    | 35.1-3001.0 |

Table 3. Analysis of Variance comparing conductivity among mainstem stations along Mattawoman Creek.

| Source          | DF | Sum of Squares | Mean Square | F Value | Pr > F |
|-----------------|----|----------------|-------------|---------|--------|
| Model           | 7  | 107864.0448    | 15409.1493  | 7.39    | <.0001 |
| Error           | 56 | 116717.2863    | 2084.2373   |         |        |
| Corrected Total | 63 | 224581.3311    |             |         |        |

|          |           |          |           |
|----------|-----------|----------|-----------|
| R-Square | Coeff Var | Root MSE | cond Mean |
| 0.480289 | 26.37665  | 45.65345 | 173.0828  |

| Source  | DF | Anova SS    | Mean Square | F Value | Pr > F |
|---------|----|-------------|-------------|---------|--------|
| station | 7  | 107864.0448 | 15409.1493  | 7.39    | <.0001 |

Table 4. Watershed area for three rivers selected to evaluate watershed scale effects on bottom dissolved oxygen dynamics.

| River    | Dominant Land Use | Area (hectares) |
|----------|-------------------|-----------------|
| Choptank | Agriculture       | 454,802         |
| James    | Forest            | 2,589,958       |
| Patuxent | Urban             | 242,682         |

Table 5. Proportion of dissolved oxygen measurements during 2005-2015 that did not meet target and limit criteria in the three mesohaline tributaries.

| River          | Percentage below target (5.0 mg/L) | Percentage below limit (3.0 mg/L) |
|----------------|------------------------------------|-----------------------------------|
| James River    | 12                                 | 0                                 |
| Choptank River | 40                                 | 6                                 |
| Patuxent River | 88                                 | 56                                |

Figure 1. Four stations selected for continuous conductivity monitoring. Deer Creek and Bush River are located in the Piedmont Region. Mattawoman Creek and Piscataway Creek are located in the Coastal Plain Region. Dark dots mark the location of continuous monitors.

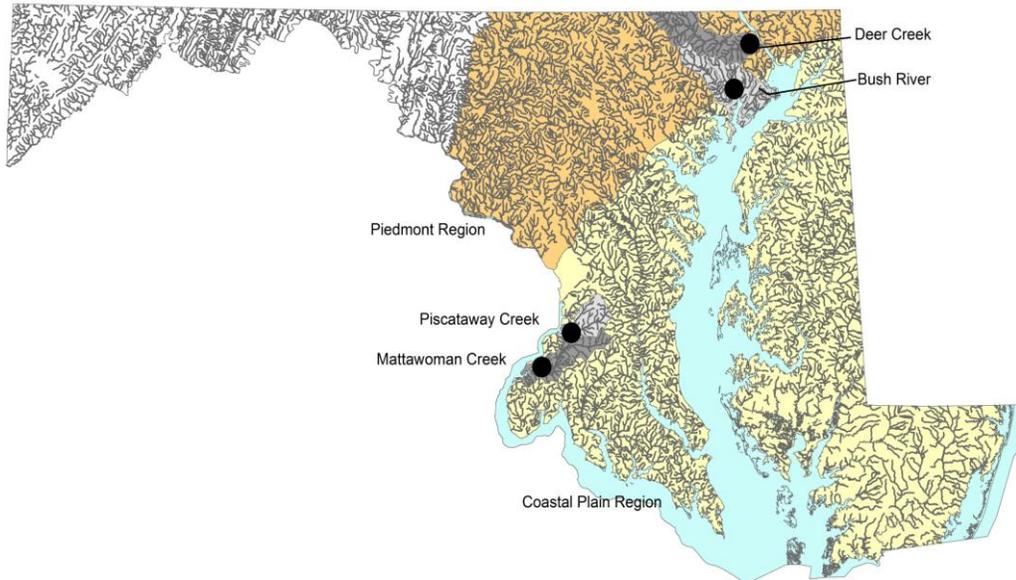


Figure 2. Mattawoman Creek watershed with points representing the centroid of each dwelling the watershed. The light tan color shows the watershed, green areas are individual dots representing an individual dwelling unit. The intensity of green increases with density of dots. Major roadways are indicated by black lines with water (estuary and streams) represented in blue. Note the northeast area of the watershed lies in Prince Georges County, MD. The county has designated this area of the watershed for conservation.

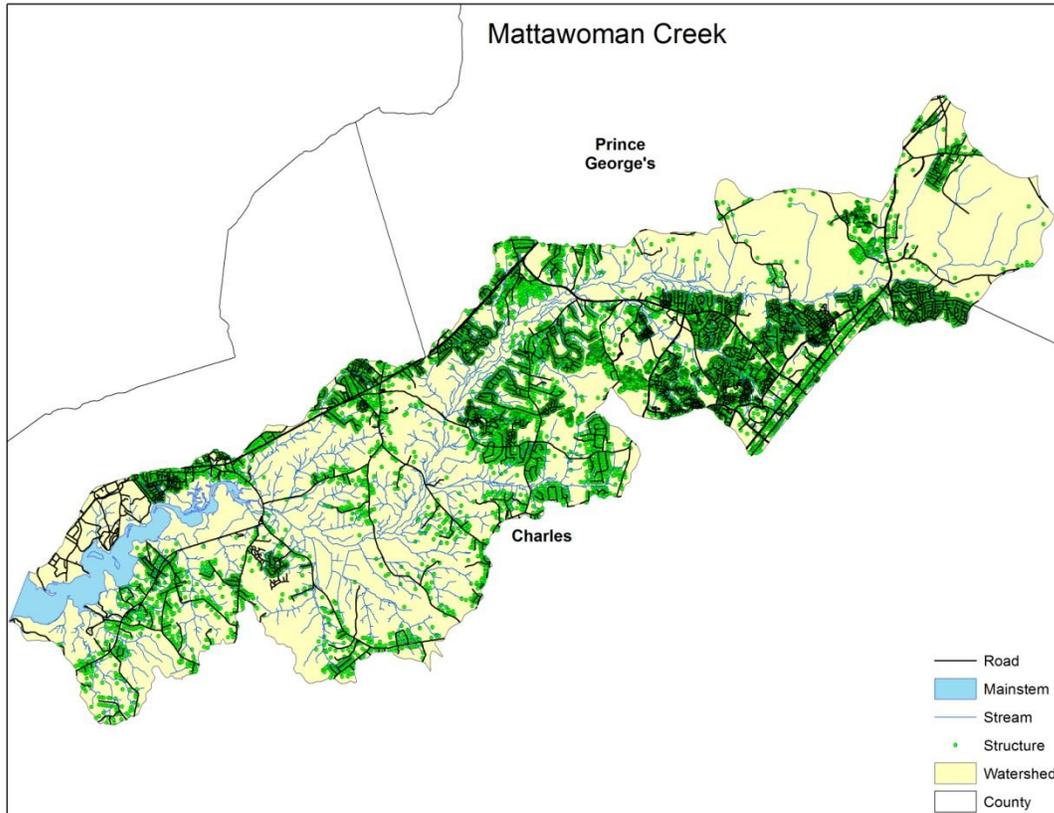


Figure 3. Piscataway Creek watershed with points representing the centroid of each dwelling the watershed. The light tan color shows the watershed, green areas are individual dots representing an individual dwelling unit. The intensity of green increases with density of dots. Major roadways are indicated by black lines with water (estuary and streams) represented in blue.

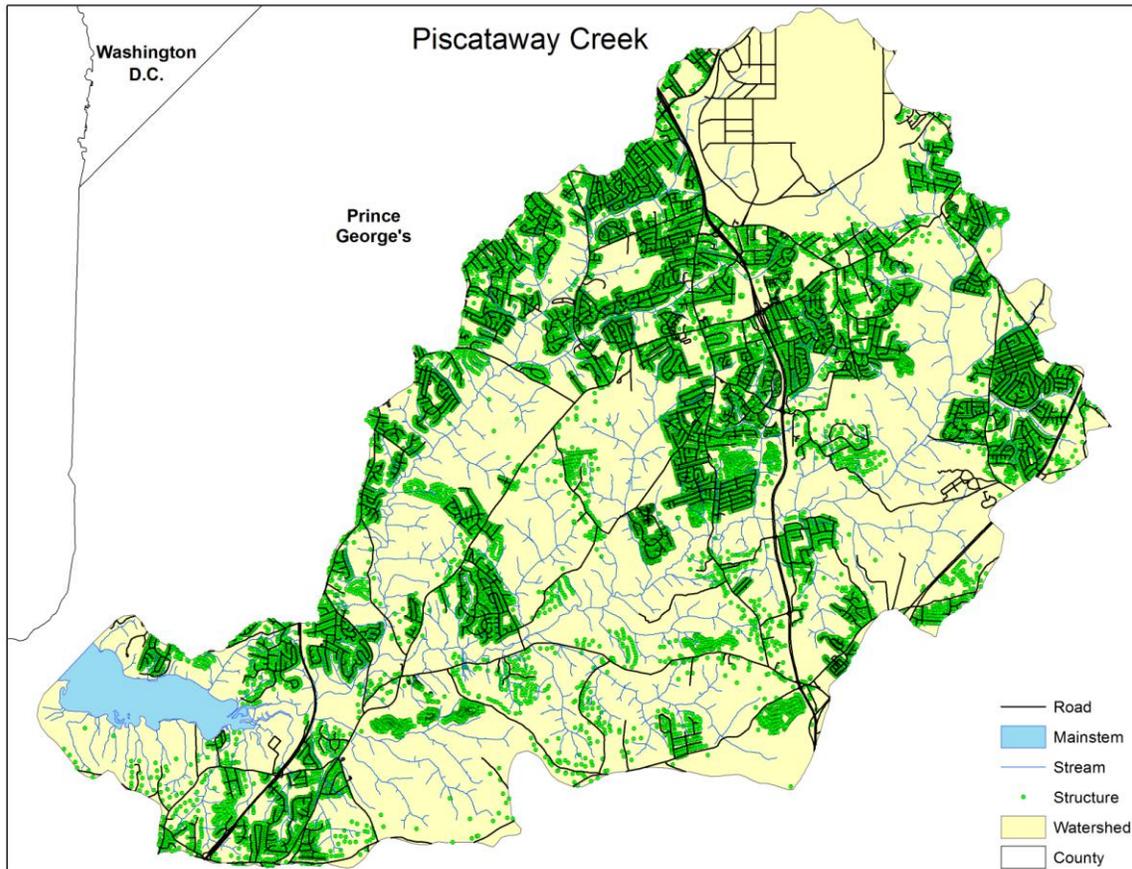


Figure 4. Deer Creek watershed with points representing the centroid of each dwelling the watershed. The light tan color shows the watershed, green areas are individual dots representing an individual dwelling unit. The intensity of green increases with density of dots. Major roadways are indicated by black lines with water (estuary and streams) represented in blue.

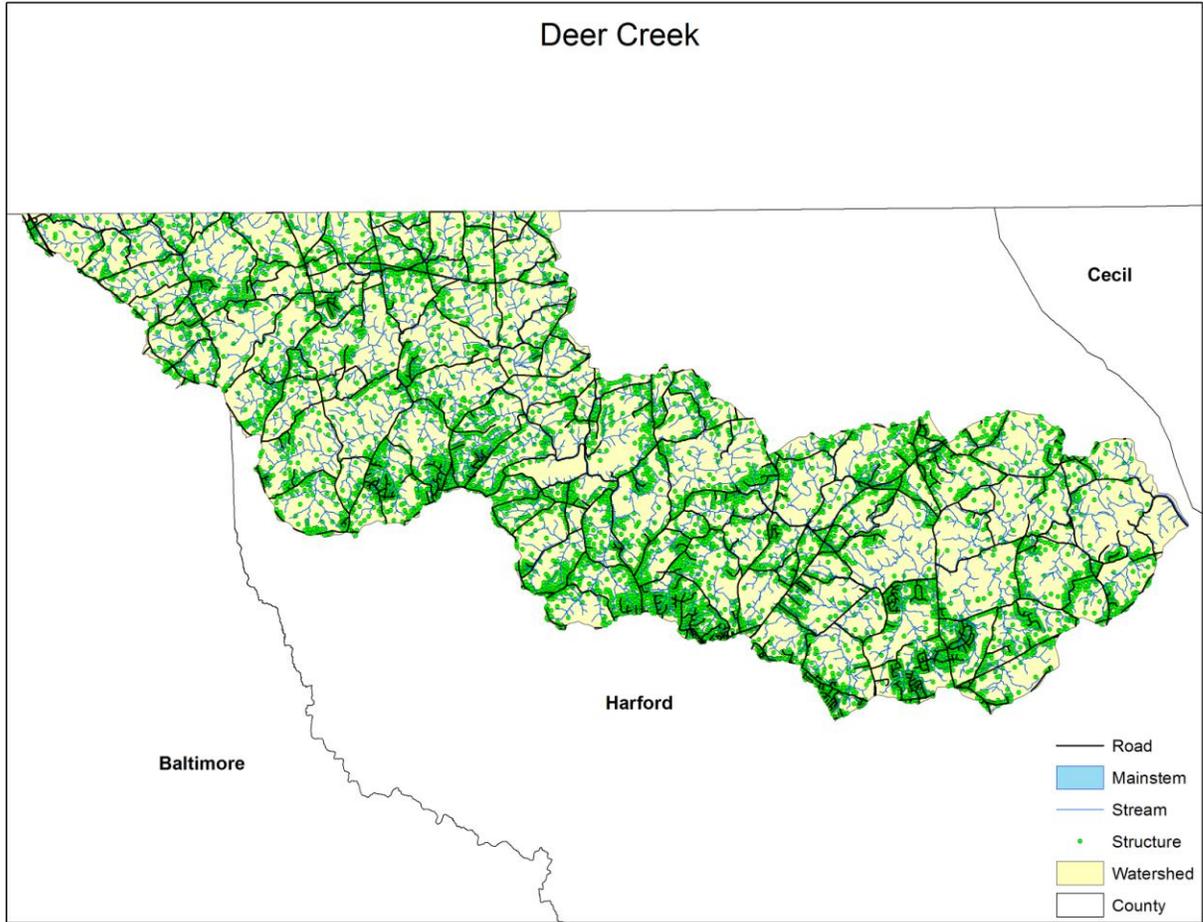


Figure 5. Bush River watershed with points representing the centroid of each dwelling the watershed. The light tan color shows the watershed, green areas are individual dots representing an individual dwelling unit. The intensity of green increases with density of dots. Major roadways are indicated by black lines with water (estuary and streams) represented in blue.

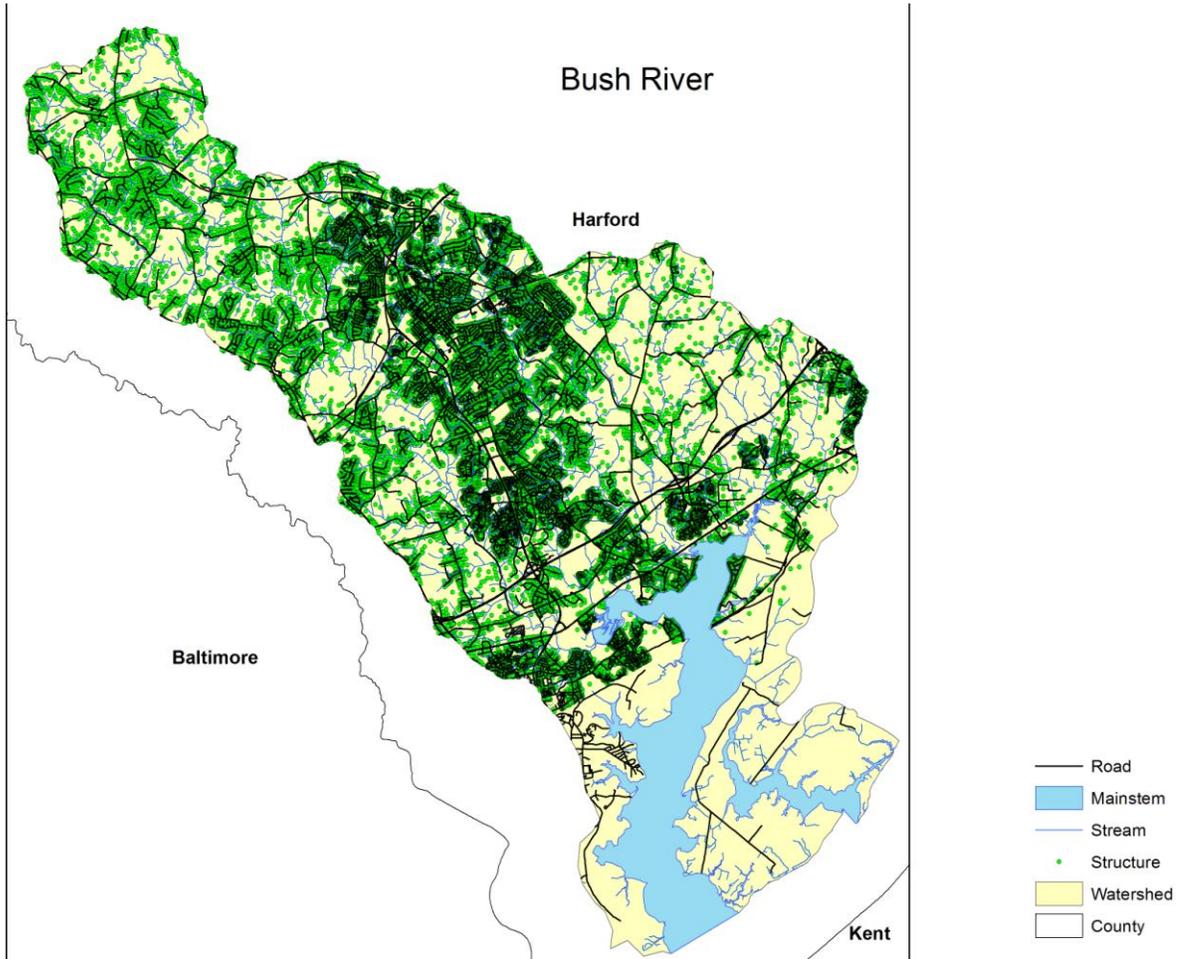


Figure 6. Mattawoman Creek with road density estimated by subwatershed and four stations (black dots) added in 2015 to conduct continuous conductivity monitoring.

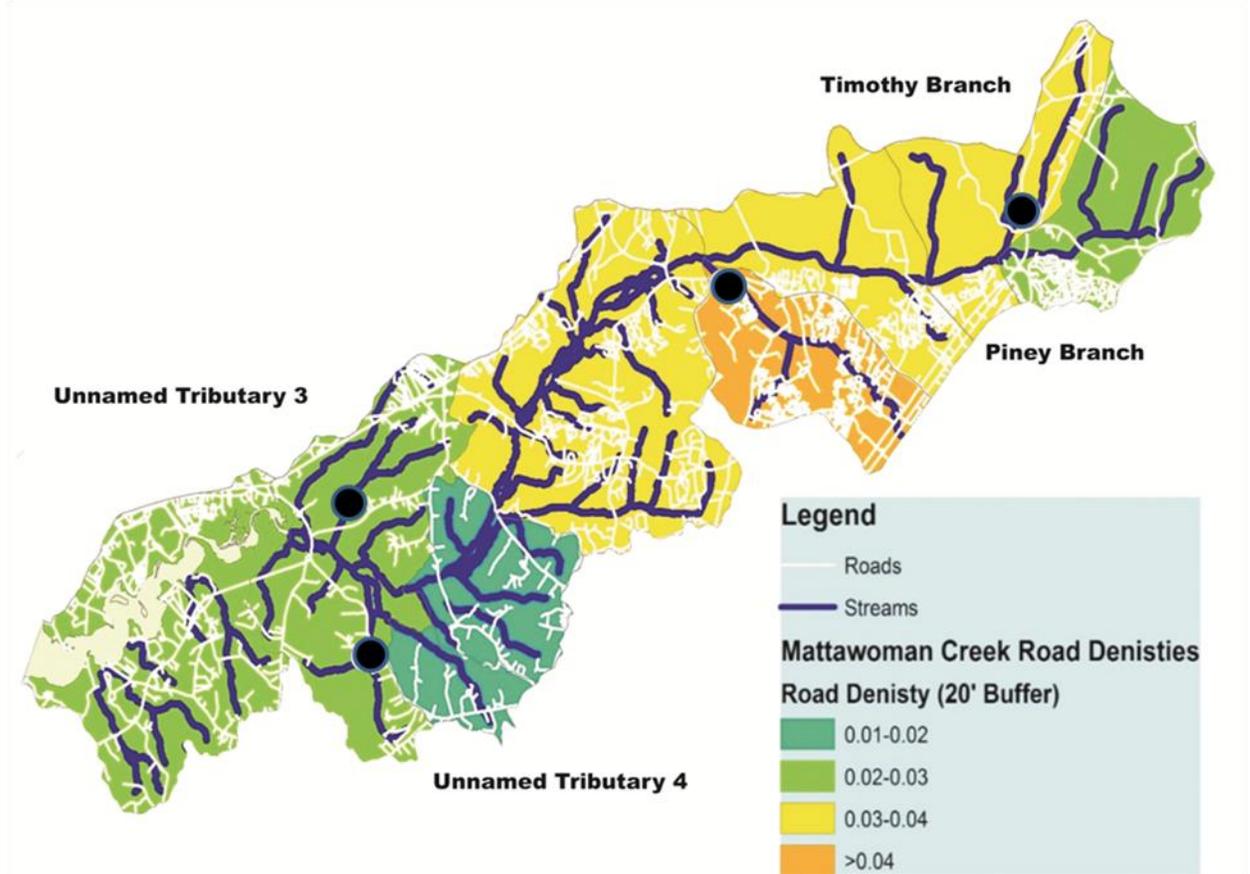


Figure 7. Additional stations (white squares) added by citizen scientists for monthly measurements of conductivity.

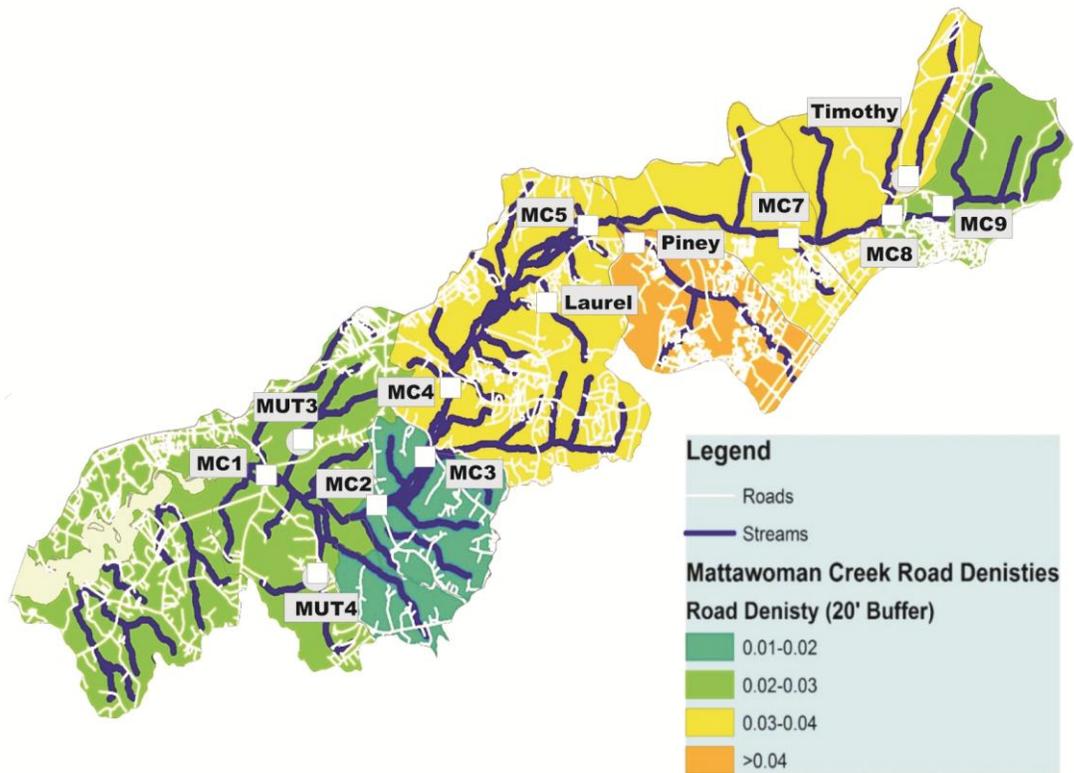


Figure 8. Mainstem continuous monitoring conductivity measurements with the Morgan background level identified by the red line. Note different scales on the y-axes.

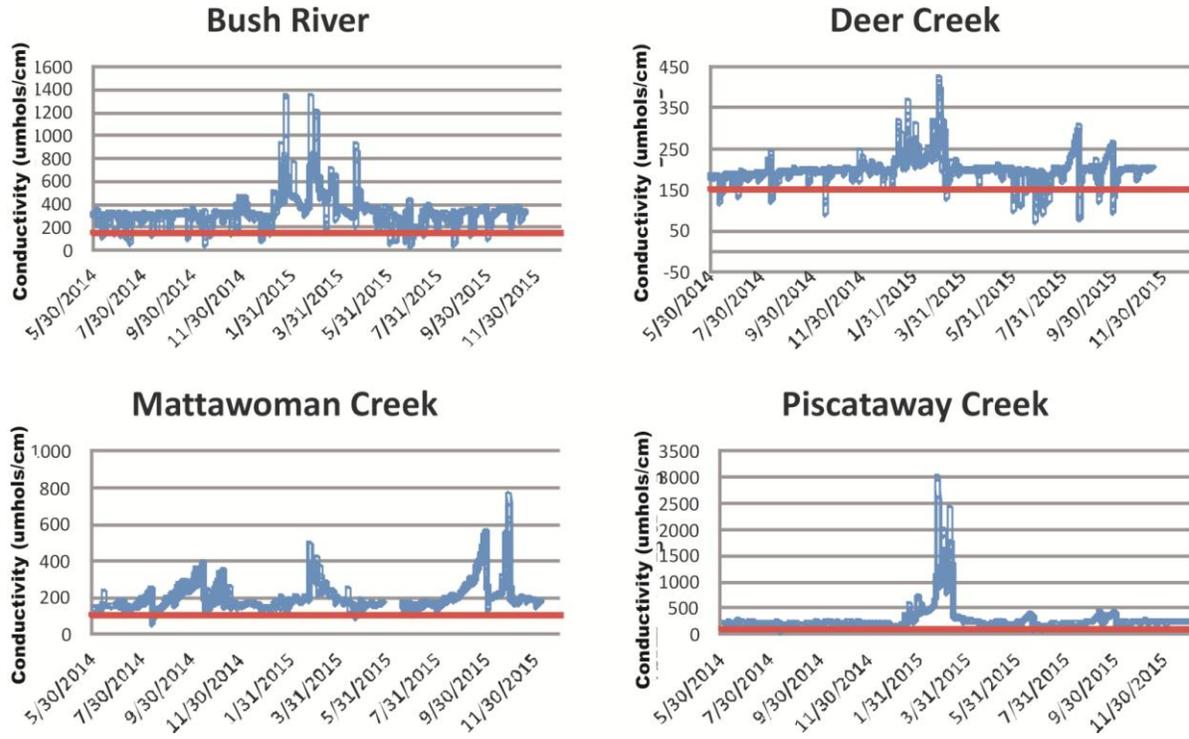


Figure 9. Mean Conductivity by housing density (C/ha) in each watershed.

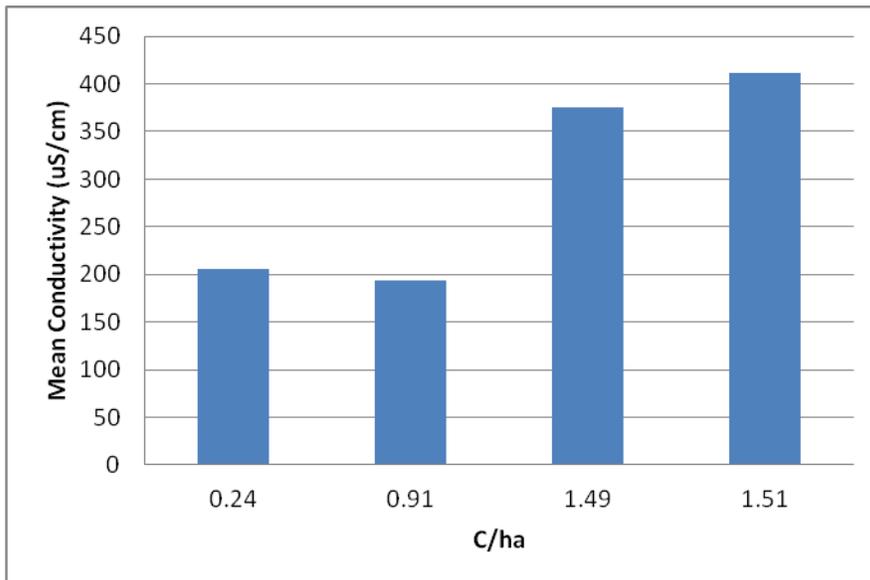


Figure 10. Mean conductivity to reference conductivity ratio (estimated mean/reference level) plotted against housing density (C/ha).

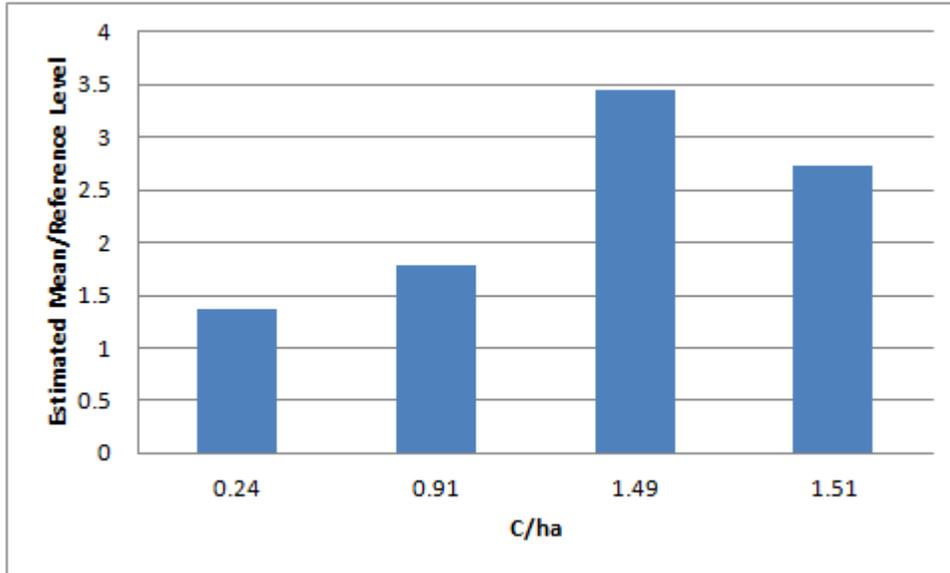


Figure 11. Adjusted  $P_{herr}$  vs adjusted mean conductivity (estimated median / reference level) with the proposed limit set at 1.56.

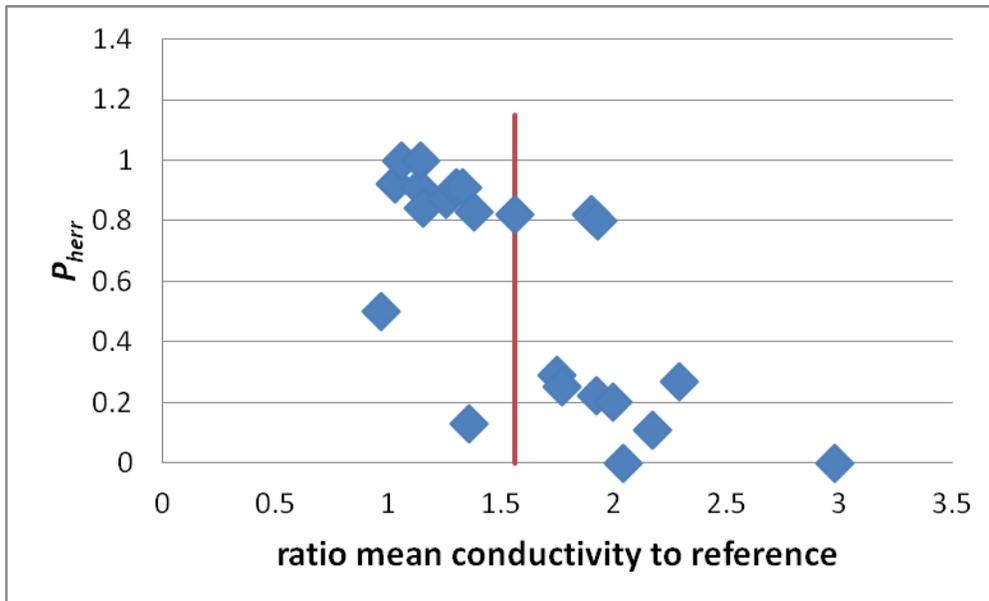


Figure 12. Applying the proposed mean conductivity to reference conductivity ratio limit of 1.56 to spring conductivity measurements during 1995-2014 acquired from the MBSS database. Historical spawning areas are overlaid. Pink shading represents watersheds where the adjusted mean conductivity exceeded the limit, green shaded watersheds reflect the limit is avoided.

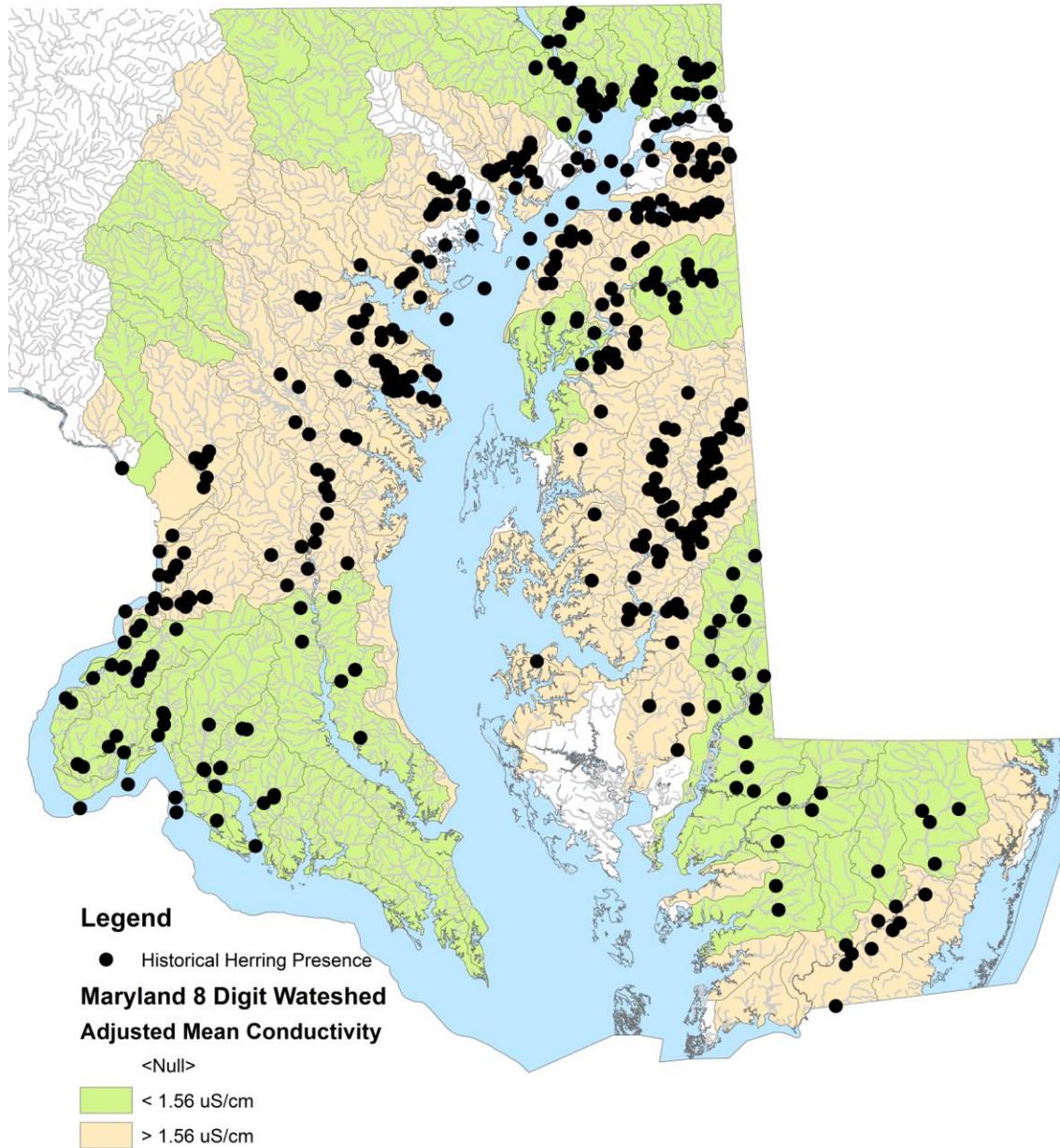


Figure 13. The proposed mean conductivity to reference conductivity ratio (MCRC) for Herring spawning applied to high priority watersheds. Pink shaded areas are high priority watersheds where the MCRC was exceeded.

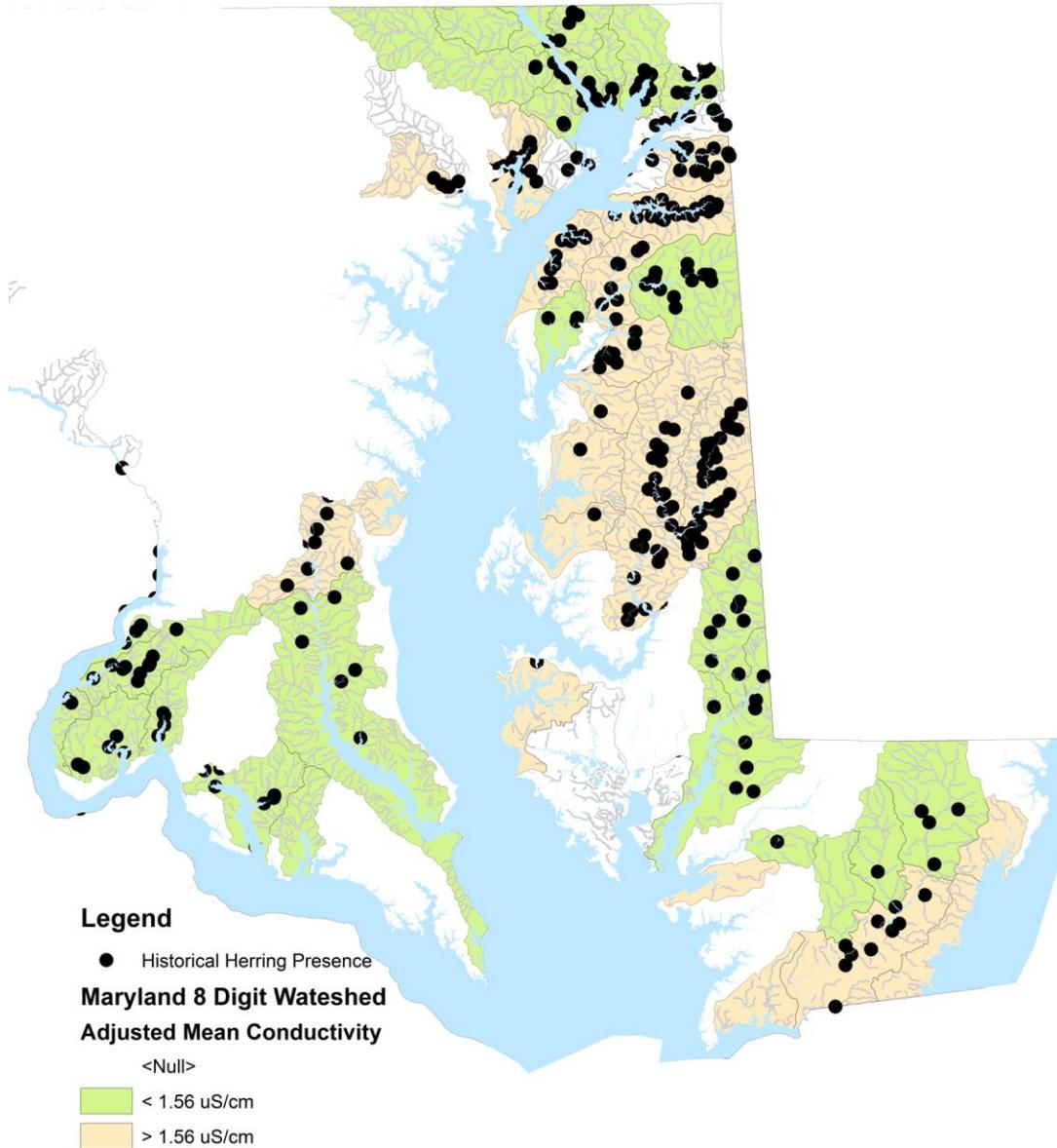


Figure14. Honing in on the Choptank River where the the mean conductivity to reference conductivity ratio exceeded the proposed limit for Herring spawning. MBSS conductivity data from spring are plotted to identify “hot spots” areas where conductivity is higher. Conductivity was partitioned into five categories using the natural breaks function in ArcGIS 10. Larger symbols represent higher concentrations.

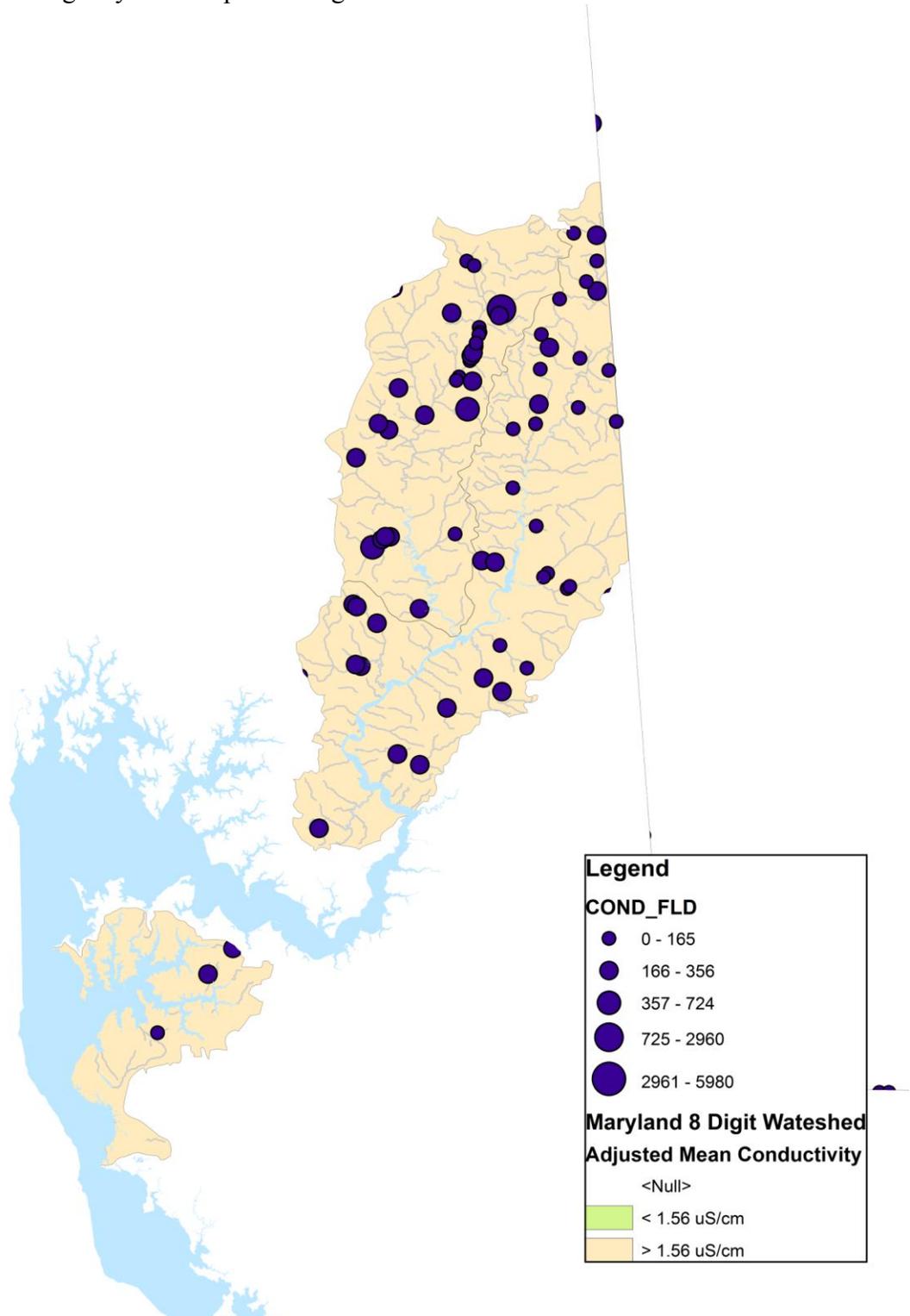


Figure 15. Three “hot spots” on Tuckahoe Creek, a tributary of the Choptank River, identified in the sample watershed where the MCRC was exceeded.

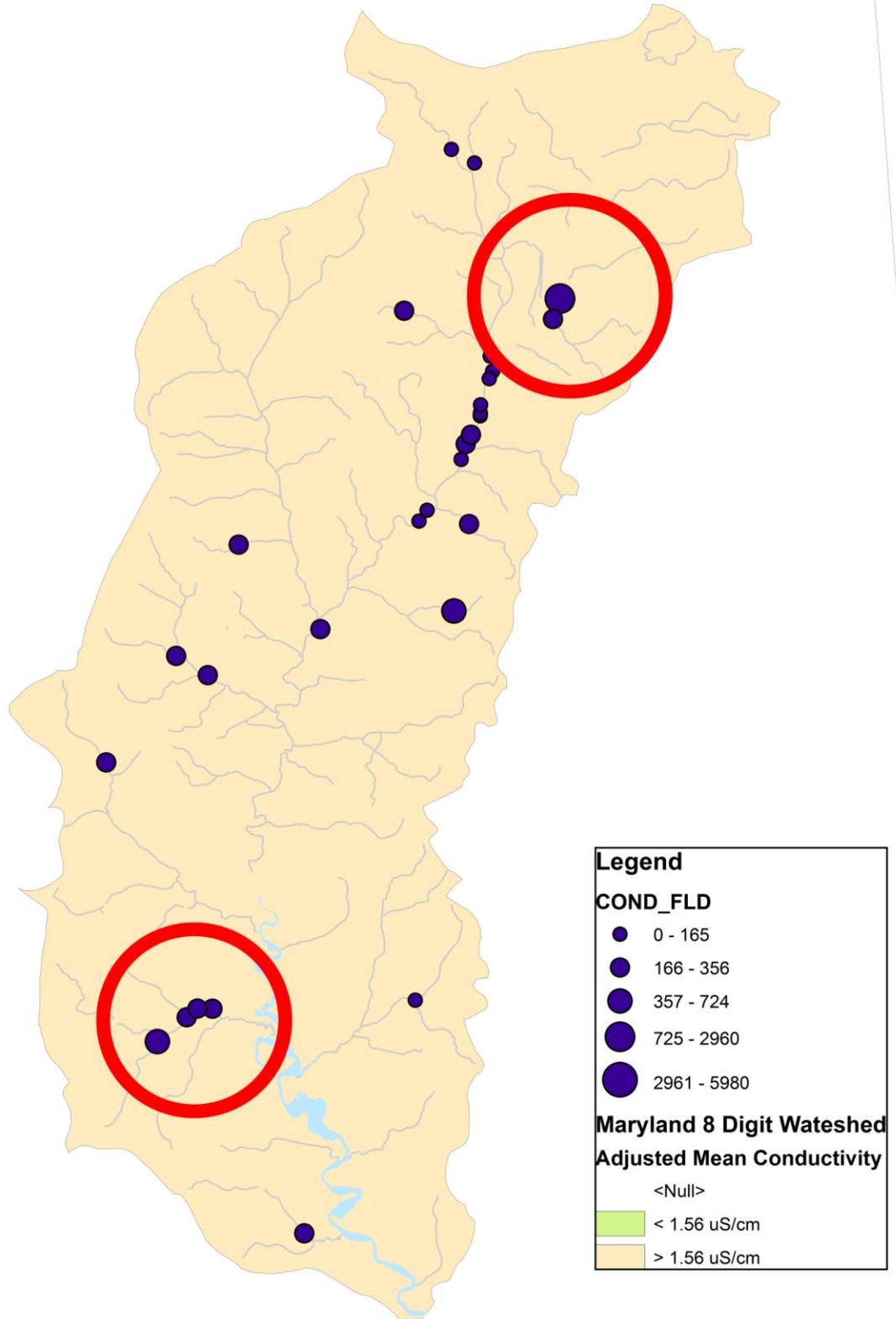


Figure 16. Google earth image showing land use upriver from the site showing elevated conductivity. This is a close up of the area in the lower left hand circle in Figure 15.



Figure 17. Mean daily conductivity at four sites on Mattawoman Creek. Note there were dewatering events between late August and early October at all stations except Timothy Branch. MC1 is Mattawoman Creek Mainstem Station, MUT3, Unnamed Tributary 3, MUT4, Unnamed

Tributary 4. MUT3 and MUT4 are within watersheds with low density of roads. Piney and Timothy Branch are in watersheds with high road densities.

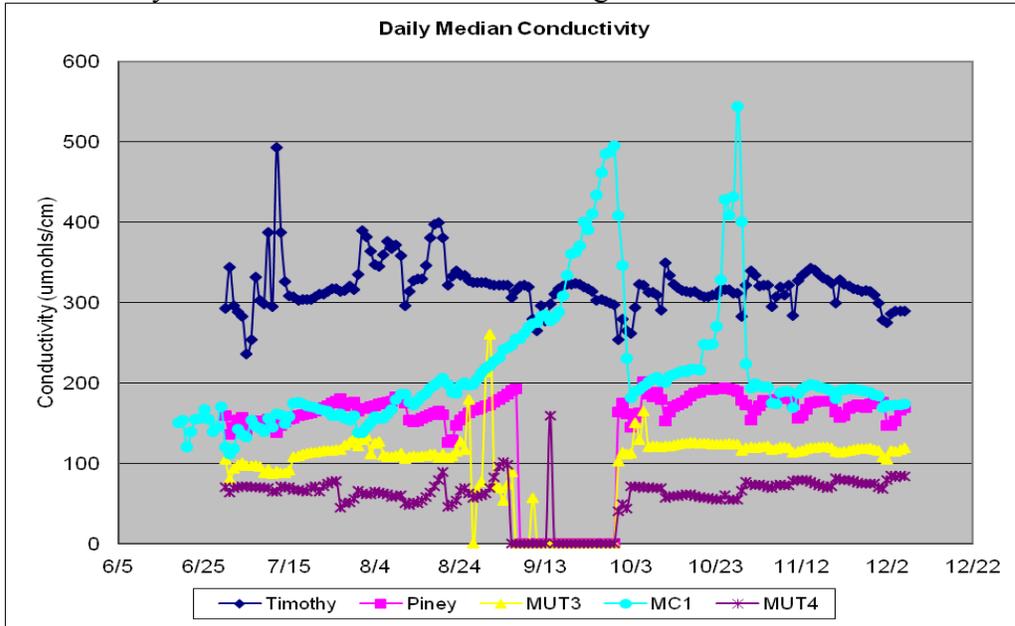


Figure 18. Box and whisker plot showing conductivity measurements of stations sampled by citizen scientists in Mattawoman Creek during 2015. Letters below the station labels represent the Tukey's means test grouping. Stations in the same letter group are not significantly different from one another.

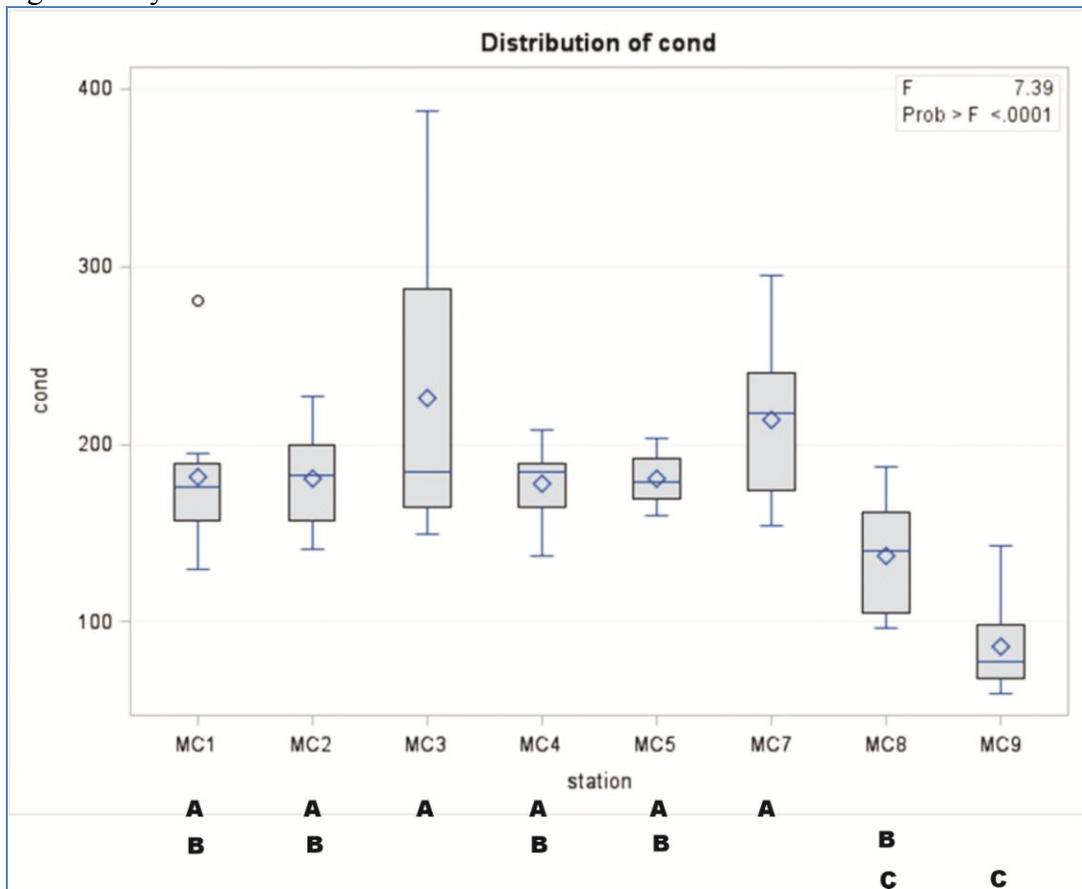


Figure 19. The three rivers chosen for evaluating Dissolved Oxygen targets and thresholds at the larger watershed scale.

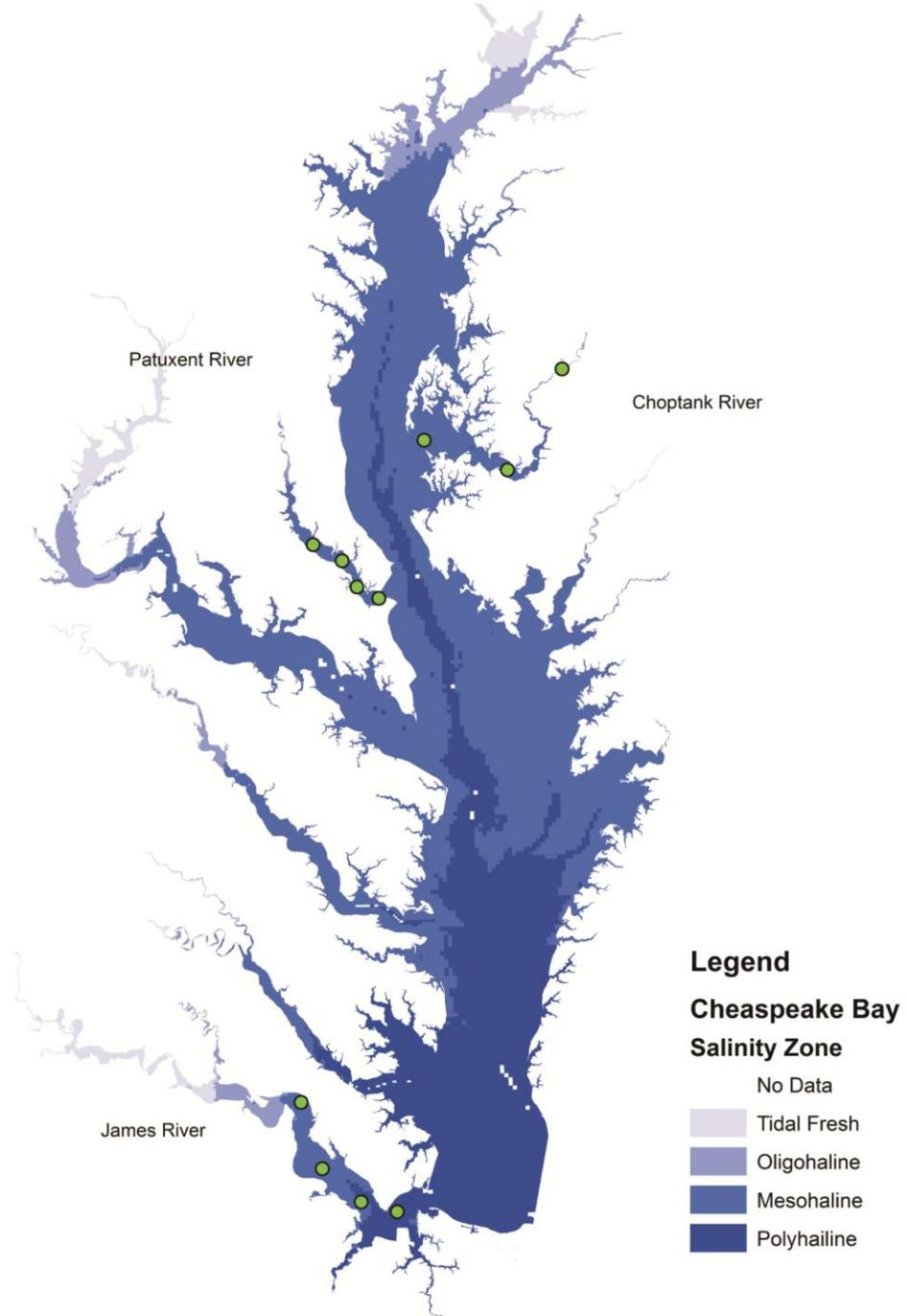


Figure 20. James River sampling locations in the lower mesohaline habitat with a box and whisker plot comparing summer bottom dissolved oxygen among stations, with 3.0 mg/L and 5.0 mg/L DO reference lines (gray lines) identified.

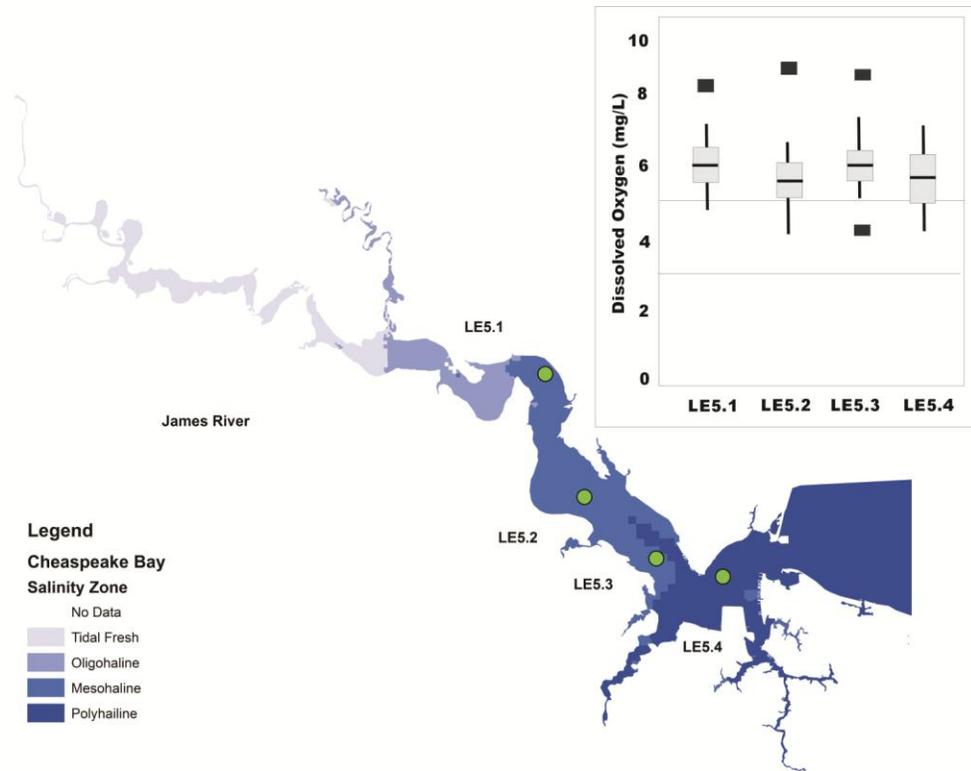


Figure 21. Choptank River sampling locations in the lower mesohaline habitat with a box and whisker plot comparing summer bottom dissolved oxygen among stations, with 3.0 mg/L and 5.0 mg/L DO reference lines (gray lines) identified.

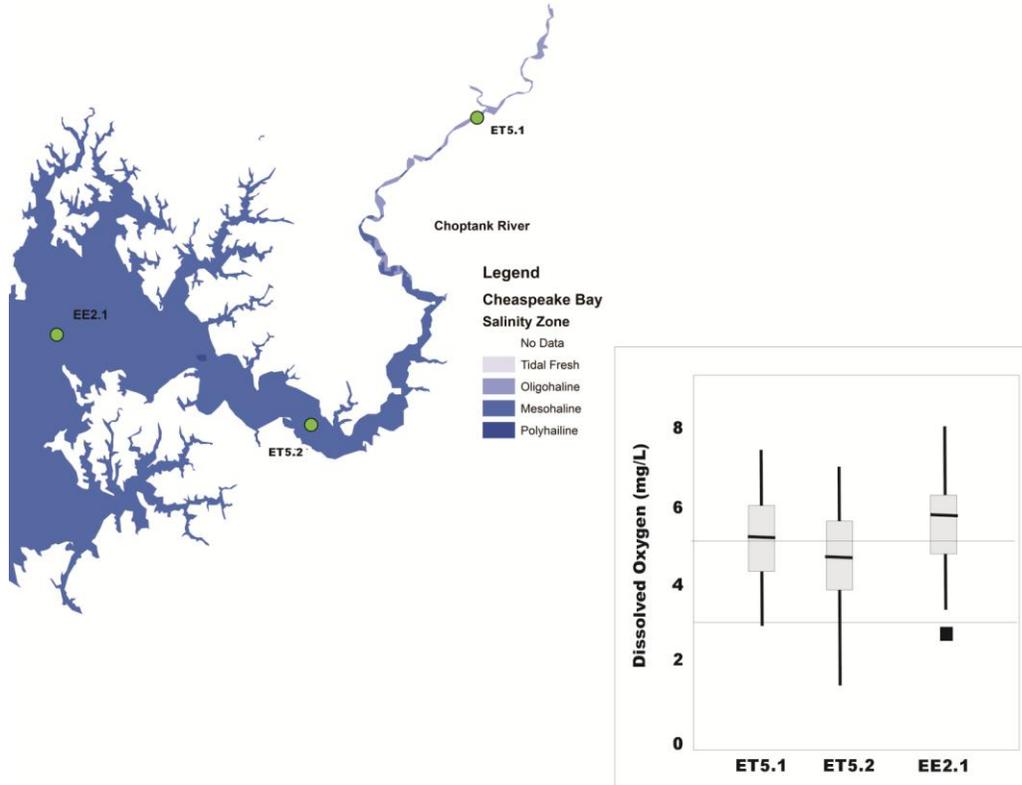
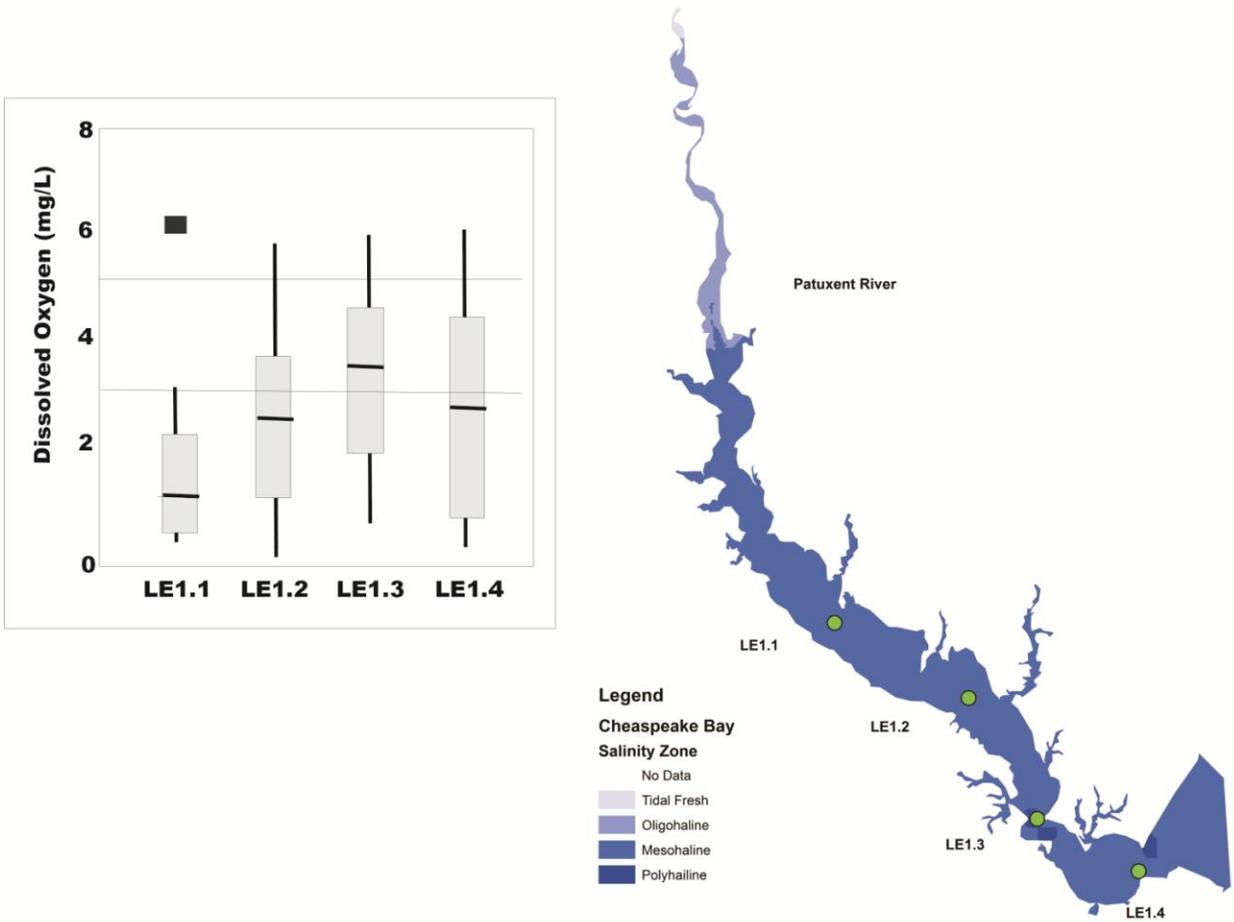


Figure 22. Patuxent River sampling locations in the lower mesohaline habitat with a box and whisker plot comparing summer bottom dissolved oxygen among stations, with 3.0 mg/L and 5.0 mg/L DO reference lines (gray lines) identified.



## **Job 4: Development of ecosystem-based reference points for recreationally important Chesapeake Bay fishes of special concern: Striped Bass nutrition and forage availability benchmarks**

Jim Uphoff, Jim Price (Chesapeake Bay Ecological Foundation), Alexis Park, Carrie Hoover, Shaun Miller, and Brian Redding

### **Abstract**

Monitoring of Striped Bass health (1998-2014), relative abundance (1983-2014), natural mortality (1987-2014), and forage relative abundance in surveys (1959-2014) and fall diets of Striped Bass (2006-2014) provided indicators to assess forage status and Striped Bass well-being in Maryland's portion of Chesapeake Bay. Striped Bass abundance rose rapidly during the mid-1990s and was followed by declines of Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab (major prey) to historical lows. Estimates of proportion without body fat indicated Striped Bass were typically in poor condition and vulnerable to starvation and disease during fall, 1998-2014. Condition was related to Striped Bass relative abundance; the relationship was described by an ascending curve with a plateau of proportion without body fat (~0.70) at high relative abundance. Striped Bass were in good condition during 2008-2010 and 2014 when their relative abundance was at a mid-level, proportion of stomachs without food was low, and some forage indices were higher. Age-0 Atlantic Menhaden dominated biomass of fall diets even though their relative abundance was low. Correlations among survey and diet-based forage indices suggested that fall diets of Striped Bass less than 457 mm were more sensitive to major prey trends than diets of larger Striped Bass. Survival of sublegal and legal sized Striped Bass in upper Bay shifted downwards in the mid-1990s shortly after upper Bay major forage-to-Striped Bass ratios, an indicator of attack success, bottomed out. Compensatory processes that intensified under regulation for highly abundant Striped Bass may undercut anticipated fishery and escapement outcomes from low fishing mortality.

### **Introduction**

Estimates of the Atlantic coast Striped Bass (*Morone saxatilis*) population increased nearly 50-fold between 1982 and 2012, mainly due to growth in the Chesapeake Bay stock (ASMFC 2013; Overton et al. 2015). The Chesapeake Bay stock supports major commercial and recreational fisheries within Chesapeake Bay and along the Atlantic coast of the United States (Richards and Rago 1999; Maryland Sea Grant 2009). After recovery of Atlantic coast Striped Bass was declared in 1995 (Richards and Rago 1999), concern emerged about the impact of high Striped Bass population size on its prey-base (Hartman and Margraf 2003; Hartman 2003; Uphoff 2003; Overton et al. 2015). Striped Bass appear capable of limiting prey populations along the Atlantic coast and in its estuaries (Hartman 2003; Uphoff 2003; Savoy and Crecco 2004; Heimbuch 2008; Davis et al. 2012; Overton et al. 2015). Inadequate prey supply has been a common management problem in lakes (Axon and Whitehurst 1985).

A large contingent of Chesapeake Bay Striped Bass that do not participate in the Atlantic coast migration (mostly males along with some young, immature females;

Setzler et al. 1980; Kohlenstein 1981; Dorazio et al. 1994; Secor and Piccoli 2007) constitute a year-round population of predators that provides the major recreational fishery in Maryland's portion of Chesapeake Bay (hereafter, upper Bay) and an important commercial fishery (Maryland Sea Grant 2009). Reports of Striped Bass in poor condition and with ulcerative lesions increased in Chesapeake Bay shortly after recovery was declared (Overton et al. 2003; Uphoff 2003; Gauthier et al. 2008; Overton et al. 2015). These reports spurred concerns about the effect of low forage fish abundance on Striped Bass health (Uphoff 2003; Overton et al. 2015). Linkage between these phenomena and poor feeding success on Atlantic Menhaden (*Brevortia tyrannus*) and other prey in upper Bay was considered plausible (Uphoff 2003; Overton et al. 2015). Mycobacteriosis, a chronic wasting disease, became an epizootic in Chesapeake Bay (Maryland and Virginia) in the late 1990s and was a likely source of lesions and poor condition (Overton et al. 2003; Jiang et al. 2007; Gauthier et al. 2008; Jacobs et al. 2009b). Challenge studies with Striped Bass linked nutrition with progression and severity of the disease, and survival (Jacobs et al. 2009a). Tagging models indicated that annual instantaneous natural mortality rate (M) of Striped Bass in Chesapeake Bay increased substantially during the mid-1990s while fishing mortality remained low (Jiang et al. 2007; ASMFC 2013).

The Atlantic States Marine Fisheries Commission (ASMFC) is moving to develop forage reference points for managing Atlantic Menhaden along the Atlantic coast and Striped Bass is a predator of concern (SEDAR 2015). Prevalence of mycobacteriosis and natural mortality appear to be less intense outside Chesapeake Bay (Matsche et al. 2010; ASMFC 2013), leaving open the possibility that upper Bay issues would be diluted in a coastal approach. Maryland's fisheries managers and stakeholders want to know whether there is enough forage to support Striped Bass along the Atlantic coast and in upper Bay. Formal assessments of abundance and biomass of Striped Bass and most forage species in upper Bay are lacking due to cost and difficulty in addressing migration. Indicators based on monitoring, such as forage indices, prey-predator ratios, Striped Bass condition indices, and prey abundance in diet samples have been suggested as a basis for assessment (Maryland Sea Grant 2009; SEDAR 2015) and formed the foundation of our approach. Indicators are widely used for environmental reporting, research, and management support (Rice 2003; Jennings 2005).

A nutritional indicator, proportion of Striped Bass without body fat, anchored our approach, providing a measure of condition and potential for starvation that was well-related to feeding of Striped Bass in the laboratory (Jacobs et al. 2013). Lipids are the source of metabolic energy for growth, reproduction, and swimming for fish and relate strongly to foraging success, subsequent fish health, and survival of individual fish and fish populations (Tocher 2003; Jacobs et al. 2013).

While upper Bay Striped Bass feed on a wide range of prey, Atlantic Menhaden, Bay Anchovy (*Anchoa mitchilli*), Spot (*Leiostomus xanthurus*), and Blue Crab (*Callinectes sapidus*) have consistently accounted for most diet biomass (Hartman and Brandt 1995c; Griffin and Margraf 2003; Walter et al. 2003; Overton et al. 2009; Overton et al. 2015). We selected these species as focal prey for forage indices. Indices of focal prey availability were estimated from fishery-independent surveys and fall diets of Striped Bass. Trends in prey survey index-to-Striped Bass index ratios were examined for each focal prey since forage indices alone would not consider interference from other

predators (including their own species) that restricts their feeding success (Ginzburg and Akçakaya 1992; Yodzis 1994; Walters and Martell 2004). Attack success can be indexed from the ratio of prey-to-predator, allowing for the effect of predator interference to be included (Ginzburg and Akçakaya 1992; Yodzis 1994; Ulltang 1996; Uphoff 2003; Walters and Martell 2004).

The ratio of age 3 relative abundance of male Striped Bass in spring gill net surveys (Versak 2015) to their year-class-specific juvenile indices (Durell and Weedon 2015) were used as indicators of change in survival due to natural mortality (SR) prior to recruitment to the fishery (Uphoff et al. 2015). Confining the gill net relative abundance indices to 3 year-old males makes it likely that trends in SR will reflect resident Striped Bass survival before harvest. Age 3 male Striped Bass in the spring gill net survey were nearly always well below legal-size in years when harvest was allowed (Versak 2015), but they could be subject to catch-and-release mortality. We expected SR to vary without trend if natural mortality (M) remained constant. Unfortunately, it became apparent that SR estimates used in Uphoff et al. (2015) were biased because age 3 gill net indices were not reflecting expected trends in abundance of age 3 fish indicated by the stock assessment, juvenile indices, and other indicators. We developed adjusted gill net indices that reflected expected stock changes and used these as the numerator in the SR estimates in this report.

Statistical analyses can provide insight into important processes related to predation (Whipple et al. 2000). We used correlation and regression analyses to examine whether indicators of upper Bay Striped Bass abundance, forage abundance, consumption, condition, and natural mortality estimates were likely to be linked. If compensatory processes were active, we expected lower forage, higher Striped Bass abundance, and poor condition would align with higher natural mortality. If fishing mortality was the primary driver of dynamics (the underlying assumption of single-species assessments; Hare 2014) few, if any, associations or relationships would be apparent among indicators and they would not align with changes in estimated natural mortality.

### **Methods**

Nutritional status (condition) for upper Bay Striped Bass was estimated as the proportion of fish without visible body fat during October-November (P0; Jacobs et al. 2013). Body fat data were collected as part of comprehensive Striped Bass health monitoring in upper Bay initiated after an outbreak of lesions began. Fish were collected by hook-and-line from varying locations during fall, 1998-2014, between Baltimore, Maryland (northern boundary), and the Maryland-Virginia state line (southern boundary; Figure 1). Estimates of P0 were made for all sizes of Striped Bass combined. Standard deviations and confidence intervals (95% CI) of P0 were estimated using the normal distribution approximation of the binomial distribution (Ott 1977).

As Striped Bass experience starvation, lipids are replaced by water, conserving weight loss and hampering the interpretation of weight at length condition indices (Jacobs et al. 2013). Jacobs et al. (2013) presented a target for body moisture (25% or less of fish with starved status) as a surrogate for lipid content estimated from proximate composition of well fed Striped Bass. This target was derived from fall 1990 field collections by Karahadian et al. (1995) - the only field samples available from favorable feeding conditions. A target for visible body fat was not presented in Jacobs et al. (2013)

because the index was not applied in the 1990 collection. However, mean tissue lipid of Striped Bass without visible body fat was reported to be identical to that estimated from percent moisture in the remainder of the data set, meaning that P0 related strongly to the proportion exceeding the moisture criteria. A level of P0 of 0.30 or less was used to judge whether Striped Bass had fed successfully during October-November. Variation of tissue lipids estimated from body fat indices was greater than for moisture and the P0 target accounted for this additional variation plus a buffer for misjudging status. Jacobs et al. (2013) stressed that comparisons of Striped Bass body fat to a nutritional target or threshold in Chesapeake Bay should be based on October-November data since they were developed from samples during that time span.

We used geometric mean catches from fixed station seine and trawl surveys as indicators of relative abundance of most major prey species in upper Bay. A shoreline seine survey targeting age-0 Striped Bass during 1959-2014 provided indices for Atlantic Menhaden, Bay Anchovy, and Spot (Goodyear 1985; Richards and Rago 1999; Durell and Weedon 2015). Additional indices for Spot and Bay Anchovy were estimated from a Blue Crab trawl survey conducted during 1989-2014 (Uphoff 1998; Rickabaugh and Messer 2015; MD DNR 2015a). These surveys sampled major and minor tributaries, sounds adjacent to the mainstem upper Bay, but not the mainstem itself (Figure 1). Sampling occurred during summer through early fall. Density of juvenile Blue Crabs in a stratified random winter dredge survey (1989-2014) that sampled Chesapeake Bay (Maryland and Virginia) was our indicator of Blue Crab relative abundance (Sharov et al. 2003; Jensen et al. 2005; MD DNR 2015b). Spot and Blue Crabs were classified as benthic forage, while Atlantic Menhaden and Bay Anchovy were pelagic (Hartman and Brandt 1995c; Overton et al. 2009). Correlation analyses explored associations of forage indices and P0. Each forage index was divided by its mean for years in common (1989-2014) to place them on the same scale for graphs.

Indicators of feeding success and diet composition during October-November were developed from a citizen-science based Striped Bass diet monitoring program conducted by Chesapeake Bay Ecological Foundation (CBEF) during 2006-2014. During 2014, Striped Bass collected as fish health samples by the Fish and Wildlife Health Program (FWHP) were processed by Fish Habitat and Ecosystem Program personnel for diet information.

Striped Bass diet collections were made in a portion of upper Bay bounded by the William Preston Lane Bay Bridge to the north, the mouth of Patuxent River to the south, and into the lower Choptank River (Figure 1). Striped Bass were collected for diet samples by hook and line fishing. Conditions of the collectors permit issued to CBEF allowed for samples of up to 15 Striped Bass less than 457 mm total length (or TL; hereafter, sublegal sized Striped Bass or fish) and 15 fish 457 mm TL or larger (hereafter, legal sized Striped Bass or fish) per trip during 2006-2014. Most active trips by CBEF occurred in Choptank River, but some occurred in the mainstem Chesapeake Bay. These trips were our source of sublegal sized fish, but legal sized fish were caught as well. Striped Bass kept as samples during active trips were placed in a cooler and either processed immediately or held on ice for processing the next day. Legal sized Striped Bass collections were supplemented by charter boat hook and line catches sampled at a fish cleaning business by CBEF. These fish were predominately from the mainstem Chesapeake Bay. These fish were iced immediately and cleaned at the station upon

return to port. Fish, minus fillets, were held on ice over one to several days by the proprietor of the fish cleaning service and processed at the check station.

Diet collections by FWHP during 2014 were not constrained by collectors permit conditions like CBEF collections. Sampling by FWHP was designed to fill size class categories corresponding to age-classes in an age-length key to assess Striped Bass health. Sublegal and legal sized fish were collected together. Some trips occurred where fish in filled out length classes were discarded (typically sublegal sized fish). Samples were usually obtained by fishing on a charter boat using the techniques considered most effective by the captain (chumming, chunking, or trolling usually). During fall 2014, Striped Bass were obtained on five trips made at five locations in mainstem middle and lower Chesapeake Bay (between the mouth of the Choptank River and mouth of Patuxent River).

Total length of each Striped Bass was recorded and whole fish were weighed on a calibrated scale for CBEF and FWHP samples. Striped Bass length-weight regressions based on that year's October-November samples were used to estimate missing weights from filleted fish in CBEF collections. Diet items of each fish were identified to the lowest taxonomic group. Contents were classified as whole or partially intact. In CBEF collections, total length of intact fish and shrimp, carapace width of crabs, and shell length of intact bivalves were measured. Non-linear allometry equations for converting diet item length to weight (Hartman and Brandt 1995a) were used. In a few cases, equations for a similar species were substituted when an equation was not available. These equations were used to reconstruct diets for Overton et al. (2009) and Griffin and Margraf (2003), and were originally developed and used by Hartman and Brandt (1995a). Soft, easily digested small items such as amphipods or polychaetes that could not be weighed were recorded as present. Empirical relationships developed by Stobberup et al. (2009) were used to estimate relative weight from frequency of occurrence of their general taxonomic category. These soft items were rare in our fall collections, but were more common during other seasons (J. Uphoff, personal observation). Appendix 1 describes processing instructions for FWHP collections and a copy of the form used to enter data from 2014 collections. These 2014 diet data were merged with the rest of the fish health records.

Fall diet collections through 2013 by CBEF were entered and processed as described in Uphoff et al. (2014; 2015). Collections by CBEF for 2014 have not been entered at this time. Data for fall 2014 from FWHP collections had been entered and were used in this report.

Striped Bass diets were analyzed separately for sublegal and legal sized fish. These categories accounted for ontogenic changes in Striped Bass diet, but also reflected unbalanced sample availability to CBEF (sublegal fish could only be collected by fishing for them directly, while legal sized fish were supplemented by cleaning station sampling). The lower limit of fish analyzed in the sublegal category, 286 mm, was the minimum length in common among years during 2006-2013. An upper limit of 864 mm avoided inclusion of large, migratory Striped Bass that reentered upper Bay in late fall.

We confined analysis of food items to those considered recently consumed in an attempt to keep odds of detection as even as possible. Items with "flesh", including whole or partial fish and invertebrates, and intact crab carapaces were considered recently consumed. Hard, indigestible parts such as gizzards, mollusk shells, and backbones were

recorded but excluded from analysis. Partially intact items with flesh were identified to lowest taxonomic group and assigned the mean weight estimated for intact items in the same group. Bait was excluded.

Major items were classified as young-of-year or age 1+ based on published size cut-offs. Size cut-offs (TL) for young-of-year were 174 mm for Atlantic Menhaden (minimum for August-November; SEDAR 2015), 65 mm for Bay Anchovy (VIMS 2015), 200 mm for Spot (VIMS 2015) and 61 mm carapace width for Blue Crab (MD DNR 2015b). This analysis was completed through 2014.

Percentage of food represented by an item (excluding bait) during 2006-2014 was estimated for each Striped Bass size class in numbers and weight based on fish with stomach contents (Pope et al. 2001). Two feeding metrics were calculated for each size class for each year. Relative availability of prey biomass (C) was estimated by dividing the sum of diet item weights by the sum of weight of all Striped Bass sampled (including those with empty stomachs; Pope et al. 2001). Estimates of C were subdivided by contribution of each major prey to overall diet mass (species-specific C). Proportion of Striped Bass with empty stomachs (PE) was also estimated as an indicator of total prey availability (Chipps and Garvey 2007). Standard deviations and confidence intervals (95% CI) of PE were estimated using the normal distribution approximation of the binomial distribution (Ott 1977). Correlation analyses explored associations of PE, C, with P0, and species-specific C with forage indices.

An index of relative abundance of upper Bay resident Striped Bass was not available; therefore, we developed a catch-per-private boat trip index (released and harvested fish) for 1981-2014 from the National Marine Fisheries Service's (NMFS) Marine Recreational Information Program (MRIP; NMFS Fisheries Statistics Division 2015) database. Similar recreational catch per trip indices have been used as abundance indicators in Atlantic coast stock assessments of major pelagic finfish predators: Striped Bass, Bluefish (*Pomatomus saltatrix*), and Weakfish (*Cynoscion regalis*; ASMFC 2009; NEFSC 2012; ASMFC 2013).

This index was estimated as a catch-effort ratio for private and rental boat anglers in Maryland in the MRIP inland fishing area (inshore saltwater and brackish water bodies such as bays, estuaries, sounds, etc, excluding inland freshwater areas; NMFS Fisheries Statistics Division 2015). This Striped Bass recreational fishing index (RI) equaled September-October recreational private and rental boat catch divided by estimates of trips for the private and rental boat sector. Recreational survey estimates are made in two month waves and September-October constituted the fifth wave (NMFS Fisheries Statistics Division 2015). The September-October wave was chosen because portions or the whole wave were continuously open for fishing for legal sized Striped Bass following the 1985-1990 moratorium, making it less impacted by regulatory measures than other waves that opened later. Recreational fishing by boat occurs over the entire portion of the upper Bay and this index would be as close to a global survey of non-migratory upper Bay Striped Bass as could be obtained. Migratory fish were unlikely to have been present during this wave.

The relationship of upper Bay resident Striped Bass relative abundance (RI) to condition (P0) was examined using linear regression. Examination of the plot of P0 and RI suggested that an asymptotic relationship might be possible, so a reciprocal

transformation ( $1 / RI$ ; Sokal and Rohlf 1969) was used to linearize data. Therefore, two models were used: (1)  $P0 = RI$  and (2)  $P0 = 1 / RI$ .

We used forage indices divided by RI (forage index-to-Striped Bass index ratios) as indicators of forage supply relative to Striped Bass demand (relative attack success). Trends in forage-to-Striped Bass ratios were compared to survival derived from tag-based estimates of  $M$  for 457-711 mm, TL, Striped Bass from Chesapeake Bay (ASMFC 2013). Tag-based estimates of natural mortality were determined for two time periods: 1987-1996 and 1997-2011 (ASMFC 2013).

We estimated relative survival for age-3 Striped Bass in upper Bay as relative abundance at age-3 divided by age-0 relative abundance three years prior (juvenile index in  $y-3$ ). Maryland DNR estimates age-specific indices of Striped Bass relative abundance from spawning season gill net surveys on the Potomac River and Head-of-Bay spawning areas (~39% and 47% of Maryland's spawning area; Hollis 1967) and we used their age-3 indices for the numerator. To combine regional estimates, we first standardized each area's time series of gill net catch per unit effort to its time-series mean (1985-2014). These standardized estimates were then averaged for each year (Potomac River was not sampled in 1994) and this average of standardized CPUE was divided by its respective  $JI$  three years prior to estimate  $SR$  (Uphoff et al. 2015).

Gill net indices used in the numerator of  $SR$  in Uphoff et al. (2015) were suggesting either no change in abundance since 1985 or a decrease; we considered both trends implausible when viewed against stock assessment estimates (Figure 2), juvenile indices, and harvest trends. One possibility was for gill net survey catchability to change as an inverse nonlinear function of population size (Peterman and Steer 1981; Crecco and Overholtz 1990; Johnson and Carpenter 1994; VanDeValk et al. 2005; Hubert and Fabrizio 2007). Prey availability, schooling behavior of fish, limited search areas and times for fish and fishermen, non-random behavior of fish and fishermen, and contraction of area occupied by less abundant stocks affect catchability in recreational and commercial fisheries (Peterman and Steer 1981; Crecco and Overholtz 1990; Johnson and Carpenter 1994; VanDeValk et al. 2005; Hubert and Fabrizio 2007). While the stratified random design employed in the spring gill net survey (Versak 2015) should alleviate concerns related to fisher behavior, nonrandom fish behavior that causes changes in catchability with stock size may be of concern (Hubert and Fabrizio 2007).

The expected relationship of survey catch ( $C$ ) per effort ( $f$ ) to abundance ( $N$ ) is

$$(1) C / f = N * q;$$

where  $q$  (catchability) is constant over time (Hubert and Fabrizio 2007). Rearranging this equation as

$$(2) (C / f) / N = q$$

provides a basis for estimating catchability for each survey year. Since we used standardized combined gill net indices for each age, we were estimating relative catchability. Our concern was with trend rather than absolute values of catchability.

Survey gill net catch per effort has been estimated for age-3 males since 1985 (Verak 2015) and abundance at age estimates from the SCAM existed for 1982-2012 (ASMFC 2013). Much of the trend in SCAM abundance estimates should reflect the Chesapeake component of the stock (ASMFC 2013), so equation 2 could be used to estimate relative catchability (relative  $q$ ) trends to examine whether  $q$  was likely to be

changing with stock size. To confirm that relative  $q$  was an inverse function of abundance we fit relative  $q$  to age-3 abundance estimates using

$$(3) q = \alpha \cdot N^B,$$

where  $\alpha$  and  $B$  are parameters describing the nonlinear, inverse relationship of relative  $q$  and abundance at age-3 ( $N$  estimated by the SCAM; Peterman and Steer 1981; Bannerot and Austin 1983; Crecco and Overholtz 1990; Johnson and Carpenter 1994; VanDeValk et al. 2005). The objective function minimized the sum of squared residuals of the observed minus predicted relative  $q$  estimates. Sums of squares of this relationship was compared to those estimated by assuming mean relative  $q$  was equal to the time-series mean (i.e., assuming  $q$  was constant as in equation 1). Evolver, a super solver add-in for Excel (Palisade Corporation 2010) based on a genetic algorithm, was used to estimate parameters after the nonlinear regression program in SAS (Proc NLIN; Freund and Littel 2006) exhibited difficulty in obtaining estimates. Genetic algorithms mimic natural selection by creating an environment where hundreds of possible solutions to a problem can compete with one another, and only the “fittest” survive. They are not vulnerable to local minima and other difficulties encountered with typical optimization algorithms (Freund and Littel 2006; Palisade Corporation 2010). If an inverse relationship of relative  $q$  and  $N$  was present, the slope term  $B$  should be negative and different from zero (Crecco and Savoy 1990). Jackknifing (Efron and Gong 1983) was used to estimate variability (maximum, minimum, median, and 90% percentiles) of  $\alpha$ ,  $B$ , and relative  $q$  for each year, and whether the relationship of relative catchability to abundance was different from constant over the range of stock sizes. The correlation of jackknifed estimates of  $\alpha$  and  $B$  was estimated to examine independence of the two model parameters.

Relative  $q$  could be used directly to adjust gill net CPUE to reflect SCAM abundance as relative  $q \cdot N$ ; however, this would simply duplicate the trend in SCAM estimates for age-3. We wanted annual estimates of SR that were independent of the stock assessment and its schedule (assessments usually lag behind survey information by one or several years).

We expected that the age 3 gill net index in a given year would be a function of the strength of the year-class and catchability. We split the time-series into two categories, one indicating catchability at low abundance (1985-1996; high catchability category = 0) and the other at high abundance (1996-2014; low catchability category = 1; Rose et al. 1986). These categories split the time-series into periods based on when strong year-classes fully recruited into the male spawning population (starting with the 1993 dominant year-class in 1996; Durrell and Weedon 2015). This categorical approach can be used to remove autocorrelation from ecological time-series data (Rose et al. 1986). Serial patterning of residuals in a linear regression of the gill net index against age-3 abundance estimated by SCAM (time-series of Figure 2) indicated autocorrelation was a concern. We used geometric mean juvenile indices from three years prior as the year-class strength variable. We used the multiple regression model:

$$(4) \text{ Gill net index} = \text{catchability category} * \text{juvenile index}.$$

If the categorical model term was significant, an adjusted gill net index time-series was estimated by setting category equal to one for the entire time-series and using the regression equation to estimate a predicted gill net time-series. The unadjusted and adjusted gill net index time-series for age 3 were regressed against estimates of abundance at age from SCAM (ASMFC 2013) to judge effectiveness of the adjustment.

Relative survival (SR) was estimated as the adjusted gill net index for age-3 divided by its respective juvenile index. Trends in SR were compared to two tag-based estimates of survival for legal-size (457-711 mm) Striped Bass from Chesapeake Bay in ASMFC stock assessments (ASMFC 2010; 2013) and to a relative survival trends based on abundance of age-3 Striped Bass estimated by the SCAM (ASMFC 2013). Tag-based estimates of M were determined for two time periods in the ASMFC (2013) stock assessment (1987-1996 and 1997-2011). Annual estimates of M from tagging were available for 1987-2008 (ASMFC 2010). We converted the estimates of M in ASMFC (2010; 2013) to survival (S) using the equation  $S = e^{-M}$  (Ricker 1975). A relative survival estimate based on SCAM (SCAM SR) was estimated the same as SR, but with estimates of age-3 abundance standardized to their time-series mean (reducing leading zeros but producing the same trend) used as the numerator rather than adjusted gill net catch per effort.

After examination of plots of SR and RI, linear regressions and nonlinear power functions were used to examine their relationship. Correlation analysis was used to explore associations with major forage indices. Relative survival was estimated in spring, while RI and forage indices were estimated in summer-fall, so we used SR estimates in the year following the forage index year in these analyses. We removed SR estimates for 1985 from analyses because these fish may have been subject to harvest in 1984 under the 305 mm TL length limit in place. A moratorium was put in place on January 1, 1985, and age-3 Striped Bass in subsequent years would have been protected from harvest through October, 1990 (when it was removed), and by higher size limits (457 mm TL) afterwards.

Level of significance for correlation and regression analyses was  $P \leq 0.05$ . Scatter plots were examined for the need for data transformations. Residuals of regressions were inspected for outliers, trends, and non-normality. If a large outlier was identified, the data from that year was removed and the analysis was rerun. Levels of significance of correlations were not adjusted for multiple comparisons as there is no formal consensus as to when these adjustment procedures should be applied (Nakagawa 2004).

Abbreviations and definitions of indicators and important parameters are summarized in Table 1.

## Results

Striped Bass in upper Bay were usually in poor condition during 1998-2014. The body fat target ( $P_0 \leq 0.30$ ) was met during 2008-2010 and remaining estimates of  $P_0$  were between 0.40 and 0.78 (Fig 3). The 95% confidence intervals of  $P_0$  allowed for separation of years meeting the target condition from remaining estimates (Figure 3).

Major pelagic forage fish indices have been at low levels in upper Bay since the mid-1990s (Figure 4). Atlantic Menhaden seine indices were high during 1971-1994 and much lower during 1959-1970 and 1995-2014. Bay Anchovy seine indices were at low levels following the early to mid-1990s, typically at or below the bottom quartile of indices during 1959-1993. Highest Bay Anchovy trawl indices occurred in 1989-1992 and 2001-2002, while lowest indices occurred during 2006-2011 (Figure 4). There was little agreement between the two sets of Bay Anchovy indices.

Benthic forage indices were low after the 1990s, but years of higher abundance were interspersed during the 2000s (Figure 5). Seine and trawl indices for Spot were similar in trend and indicated high abundance during 1971-1994 and low abundance during 1959-1970 and 1995-2013. Blue Crab densities were highest during 1989-1996, 2009, and 2011 (Figure 5).

Correlation analyses indicated that P0 was not associated with forage indices during 1998-2014 (Table 2), a period of mostly low abundance of major prey. The trawl index for Spot was marginally and negatively correlated ( $r = -0.45$ ,  $P = 0.07$ ) and, counter-intuitively, the Bay Anchovy trawl index was positively correlated ( $r = 0.63$ ,  $P = 0.0076$ ) with P0. Remaining prey indices were not significantly associated with P0 (Table 2).

Samples from 706 sublegal and 1,907 legal sized Striped Bass were analyzed for diet composition during October-November, 2006-2013. Numbers examined during October-November of each year ranged from 47 to 191 sublegal fish and 49 to 327 legal fish.

In combination and by number, Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab (major forage items) accounted for 96.4% of diet items encountered in sublegal Striped Bass collected from upper Bay during fall, 2006-2014. Bay Anchovy accounted for the highest percentage (56.2%); Atlantic Menhaden, 15.4%; Spot 8.7%; Blue Crab, 16.1%; and other items accounted for 3.6%. By weight, sublegal Striped Bass diets in fall were dominated by Atlantic Menhaden (74.2%), followed by Spot (13.9%), Bay Anchovy (5.8%), Blue Crab (2.6%) and other items (3.4%).

Major prey accounted for 89.8% of diet items, by number, encountered in legal sized Striped Bass diets during fall. Atlantic Menhaden accounted for 44.5%; Bay Anchovy, 15.1%; Spot, 9.7%; Blue Crab, 23.3%; and other items, 7.4%. By weight, Atlantic Menhaden predominated in legal fish sampled (77.7% of diet weight); Bay Anchovy accounted for 1.1%; Spot, 6.8%; Blue Crab, 6.4%; and other items, 7.9%.

Nearly all major items eaten by both length classes of Striped Bass in fall were young-of-year. Ninety-nine percent of Atlantic Menhaden eaten ( $N = 957$ ) were below the length cutoff, as were 96.0% of Bay Anchovy ( $N = 1,325$ ), 98.4% of Blue Crab ( $N = 910$ ), and 99.4% of Spot ( $N = 207$ ). Atlantic Menhaden were the heaviest diet item, 33.2 grams on average for both size classes of Striped Bass combined, followed by Spot (12.1 grams), Blue Crab (3.9 grams), and Bay Anchovy (0.8 grams).

Estimates of relative availability of prey biomass ( $C$ , total grams of prey consumed per gram of Striped Bass) for sublegal Striped Bass varied as much as 6.7-times during 2006-2014 (Figure 6). During years of lowest  $C$  (2007 and 2011) varying items contributed to the diet of sublegal fish; during remaining years of higher  $C$ , either Spot (2010) or Atlantic Menhaden (remaining years) dominated diet mass (Figure 6).

Estimates of  $C$  for legal sized Striped Bass varied as much as 3.4-times among years sampled (Figure 7). Atlantic Menhaden dominated diet weight of legal sized fish during October-November (Figure 7).

Estimates of proportion of empty stomachs (PE) of sublegal sized Striped Bass during fall, 2006-2014, ranged between 0.10 and 0.57 (Figure 8). Lowest estimates of PE for sublegal fish (2009-2011 and 2014) could be separated from remaining estimates (except 2008) based on 95% confidence interval overlap. Estimates of PE steadily fell for sublegal sized fish during 2006-2011, increased during 2012-2013, and then fell to the

lowest level in the time-series during 2014. Estimates of PE of legal sized Striped Bass (0.37-0.63) were largely indistinguishable based on 95% confidence intervals up through 2013, with an exception that 2012 was higher than 2007-2011 (Figure 8). The proportion of empty stomachs fell to 0.10 in 2014, well below other years. Variation in relative availability of prey among years, as indicated by PE, was similar for sublegal sized Striped Bass (5.7-times) than legal sized Striped Bass (6.6-times).

Estimates of PE were more strongly correlated with P0 ( $r = 0.62$ ,  $P = 0.07$  for sublegal fish and  $r = 0.48$ ,  $P = 0.18$  for legal fish) during 2006-2014 than estimates of C ( $r = -0.40$ ,  $P = 0.29$  for sublegal fish and  $r = -0.37$ ,  $P = 0.32$  for legal fish). Removing 2008 from the correlation analyses of PE and P0 improved both correlations ( $r = 0.73$ ,  $P = 0.03$  for sublegal fish and  $r = 0.69$ ,  $P = 0.06$  for legal fish). Removal of 2008 (lowest P0 estimate of the time-series) from C and P0 analyses lead to a stronger association for legal fish ( $r = 0.69$ ,  $P = 0.06$ ) than sublegal fish ( $r = 0.53$ ,  $P = 0.18$ ). Examination of the four binary plots suggested shifts in the plots between 2006-2010 and 2011-2014 for both size classes (Figure 9). Similar levels of PE or C resulted in higher levels of P0 during 2011-2014 (Figure 9). These patterns indicated that potential relationships of condition and consumption indices may be more complex than linear associations detected by correlation analyses.

Estimates of individual prey species grams consumed per gram of Striped Bass (relative availability indicated by species-specific C) corresponded to indices of forage abundance for three of four major prey items. Sublegal sized Striped Bass species-specific C estimated during 2006-2014 for Atlantic Menhaden, Spot, and Blue Crab were strongly, positively, and significantly correlated with upper Bay survey forage indices ( $r$  between 0.81 and 0.97; Table 3). Correlations of Bay Anchovy estimates were not significant. Correlations of species-specific C based on legal sized Striped Bass diets were strongly, positively, and significantly correlated with upper Bay survey indices for Spot and Blue Crab ( $r$  between 0.86 and 0.92), but not Atlantic Menhaden or Bay Anchovy (Table 3). Blue Crab and Spot indices analyzed during 2006-2014 offered stronger time-series contrast (highs and lows similar to those exhibited in full survey time-series), while Atlantic Menhaden and Bay Anchovy indices offered limited contrast (indices were low).

In general, relative abundance of Striped Bass (RI) was lowest prior to 1994 (1990-1993 mean RI = 0.5 fish per trip; these were years of low RI with adequate catch precision; Figure 10). Estimates of RI then rose very rapidly to a high level and remained there during 1995-2006 (mean = 2.6). Estimates of RI fell to about half the 1995-2006 mean during 2008-2013 (mean = 1.2) and then rose to 2.5 in 2014 (Figure 10). Proportional standard errors (standard error as a percent of the mean) of the catch estimates were large enough to be of concern ( $> 30\%$ ) during 1981-1989, 2007, and 2014 (NMFS Fisheries Statistics Division 2015). Precision of trip estimates was adequate during 1981, 1983, and after 1985. Estimates were not available for 1982 and 1987 (NMFS Fisheries Statistics Division 2015).

Significant relationships were detected for P0 versus RI ( $r^2 = 0.35$ ,  $P < 0.013$ ) and P0 versus  $1 / \text{RI}$  ( $r^2 = 0.52$ ,  $P < 0.0011$ ; Figure 11); the latter relationship was considered a better description of dynamics due to better fit. Predicted back-transformed P0 rapidly increased from 0.25 to 0.5 as RI increased from 0.8 to 1.5 and then increased slowly towards an asymptote of approximately 0.70 when RI was 3.0 or more. Target P0 would

be reached when predicted RI was less than 0.9 (met during 1983-1993, 2008 and 2010; Figure 10). Upper Bay forage-to-Striped Bass ratios for pelagic prey (Figure 12) and benthic prey (Figure 13) rapidly shifted from high during 1983-1994 to low during 1995-2014.

Estimates of gill net survey relative  $q$  during 1985-2012 were inversely related to abundance of age-3 Striped Bass ( $N$ ) estimated by the SCAM ( $r^2 = 0.48$ ,  $P < 0.0001$ ; Figure 14). The relationship was described by the power function:

$$\text{relative } q = 1565 \cdot N^{-1.46}$$

Ninety percent of jackknifed estimates of  $\alpha$  fell between 543 and 2,890, while 90% of estimates of  $B$  were between -1.39 and -1.49. One estimate (1988, the year of lowest abundance) failed to converge within reasonable bounds for  $\alpha$  and an upper constraint could not be identified. We used a value of  $\alpha$  (3,000) slightly greater than the next highest estimate (2,890) as a constant to estimate  $B$  in this case. Estimates of  $\alpha$  and  $B$  were strongly correlated ( $r = -0.94$ ), indicating sets of pairs of the two parameters that produced similar curves. The power function sums of squares were 52% less than those generated using mean time-series (constant) relative  $q$ .

The multiple regression of gill net index with catchability category (category) and juvenile index (JI) three years prior was significant ( $r^2 = 0.47$ ,  $P = 0.002$ ; Figure 15). The equation describing the relationship was

$$\text{catchability} = 1.07 + (1.09 \cdot \text{JI}) - (0.965 \cdot \text{category});$$

standard errors of the coefficients for the intercept, JI coefficient and category coefficient equaled 0.15, 0.026 and 0.221, respectively. This equation depicted high gill net indices prior to 1996 that were less variable than those observed. Peaks in observed and predicted gill net indices generally matched during 1996-2013 (Figure 15). The observed gill net index was not related to SCAM estimates of age-3 abundance ( $r^2 = 0.026$ ,  $P = 0.41$ ), but the adjusted index was ( $r^2 = 0.76$ ,  $P < 0.0001$ ; Figure 16). The adjusted gill net index strongly mimicked the trend in year-class success indicated by juvenile indices three years prior, but were scaled to standardized gill net index values (Figure 17).

Even though the gill net index appeared to mimic the juvenile indices used in the relative survival denominator, a pattern emerged of higher relative survival of sublegal fish during 1985-1993, falling to lower relative survival by 1996 (Figure 18). This pattern was similar to the high and low survival (based on natural mortality alone) periods estimated from tagging data for legal sized (457-711 mm) Chesapeake Bay male Striped Bass: 77% survival during 1987-1995 and 44% survival during 1996-2012 (Figure 18; ASMFC 2013). Trends in relative survival of age-3 Striped Bass were related to annual tag-based survival estimates for legal-sized fish estimated by ASMFC (2010) during 1987-2008 ( $r^2 = 0.30$ ,  $P = 0.0008$ ; Figure 19). Trends in relative survival during 1985-2012 using standardized SCAM abundance estimates for age-3 in the numerator were related to those based on adjusted gill net indices ( $r^2 = 0.79$ ,  $P < 0.0001$ ; Figure 20). Downward shifts in survival of legal-sized fish and relative survival of sublegal fish coincided with sharp declines in attack success on major prey indicated by bass-to-forage ratios.

Linear and power functions provided very similar fits to relative survival (SR) and relative abundance of resident Striped Bass (RI) during 1986-2014 (power function approximate  $r^2 = 0.44$ ,  $P < 0.0001$ , Proc NLIN, SAS ; linear  $r^2 = 0.43$ ,  $P < 0.0002$ ; Figure

21). The power function described the relationship of relative survival and relative abundance of resident Striped Bass as

$$SR = 0.157 \cdot (RI^{-0.178});$$

standard errors of the multiplier and exponent were 0.0056 and 0.039, respectively. These two parameters were not correlated ( $r = -0.017$ ). The power function predicted that relative survival would decline by 55% over the range of RI. The linear regression equation describing the relationship of relative survival to relative abundance was

$$SR = (-0.024 \cdot RI) + 0.19;$$

standard errors of the slope and intercept were 0.011 and 0.005, respectively. The linear regression predicted that relative survival would decline by 46% over the range of RI during 1986-2014.

Two of three pelagic forage indices and one of three benthic prey indices were significantly correlated with SR during 1986-2014 (Table 4). The Atlantic Menhaden seine index had the highest correlation with SR ( $r = 0.70$ ,  $P < 0.0001$ ), followed by the Bay Anchovy seine index ( $r = 0.58$ ,  $P = 0.001$ ), and the Spot trawl index ( $r = 0.48$ ,  $P < 0.015$ ; but not the seine index for Spot; Table 4). These correlations indicated that major forage in general might be important, but Atlantic Menhaden, Bay Anchovy, and Spot indices were inter-correlated (Table 5), so it was not clear how species-specific the associations with SR were. The Atlantic Menhaden index was significantly correlated with seine indices for Spot and Bay Anchovy and marginally correlated with the Spot trawl index; Spot seine and trawl indices were strongly correlated with one another; and the Bay Anchovy seine index was not associated with either Spot index (Table 5). It is also possible that the significant association of the shorter Spot trawl survey time-series (1989-2014) with SR may have reflected a more detectable signal as Atlantic Menhaden seine indices declined from very high levels during 1986-1989.

### **Discussion**

A rapid rise in Striped Bass abundance in upper Bay during the mid-1990s, followed by a dozen more years at high abundance, coincided with declines in indices of Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab (i.e., major pelagic and benthic prey) to low levels. Striped Bass were often in poor condition during fall 1998-2014 and vulnerable to starvation. Improvements in condition during 2008-2010 and 2014 coincided with lower RI, slight increases in some major forage indices, and higher availability of prey in fall diets. Survival of sublegal and legal sized Striped Bass in upper Bay shifted downwards in the mid-1990s shortly after upper Bay major forage-to-Striped Bass ratios, an indicator of attack success, bottomed out.

Four sets of estimates of trends in Striped Bass survival due to non-fishing causes agreed that survival abruptly declined in the mid-1990s. These sets were not entirely independent. Two tag-based estimates relied on the same data but used different estimation techniques, while two relative survival estimates contained some of the same information (Maryland baywide juvenile index as the denominator in particular). Decreases in tag-based estimates of survival of legal-sized fish could reflect misspecification of parameters such as tag reporting rates that make absolute estimates less reliable (ASMFC 2013); however, relative survival estimates of sublegal Striped Bass based on age-3 adjusted gill net indices or SCAM estimates (based on an assumption of age-specific, temporally constant M) indicated similar trends. The fall in

survival was consistent with a compensatory response to high Striped Bass abundance, low forage, and poor condition. The degree that  $M$  compensates with  $F$  may reduce effectiveness of management measures since total mortality,  $Z$ , is not reduced by harvest restrictions when  $M$  increases as  $F$  decreases (Hilborn and Walters 1992; Hansen et al. 2011; Johnson et al. 2014). Single species stock assessments typically assume that  $M$  is constant and additive with  $F$  to keep calculations tractable (Hilborn and Walters 1992). Animal populations may exhibit additive mortality at low abundance and compensatory mortality at high abundance or compensatory mortality that changes continuously with density (Hansen et al. 2011). Increased  $M$  over time may have serious implications for management since Chesapeake Bay is the main contributor to Atlantic coast fisheries (Richards and Rago 1999; ASMFC 2013). Management of Chesapeake Bay Striped Bass fisheries attempts to balance a trade-off of yield with escapement of females to the coastal migration by controlling fishing mortality, and compensatory mortality would undercut both objectives.

Combined estimates of prevalence and progression of mycobacteriosis indicated that increased  $M$  from the disease alone may be responsible for a 16% reduction in the age 3-8 Chesapeake Bay Striped Bass population (ASMFC 2013). However, mycobacteriosis would not necessarily be the only source of increased  $M$  of Chesapeake Bay Striped Bass. Abundant individuals competing for limited prey may hinder one another's feeding activities, leading to starvation (Yodzis 1994). Under low forage conditions during 1998-2014,  $P_0$  exhibited a strong response to abundance of Striped Bass in upper Bay. At highest levels of Striped Bass abundance, estimates of  $P_0$  were consistently high ( $> 0.50$ ), indicating high vulnerability to starvation. Shifts from high survival during 1987-1996 to low survival afterwards lagged two years behind downward shifts in forage-to-Striped Bass ratios. Dutil and Lambert (2000) found that the response of Atlantic Cod (*Gadus morhua*)  $M$  could be delayed after unfavorable conditions. Similar to Striped Bass, some stocks of Atlantic Cod experienced forage fish declines, followed by declining body condition and increased  $M$ ; starvation caused declines in energy reserves, physiological condition, and enzyme activity (Lilly 1994; Lambert and Dutil 1997; Dutil and Lambert 2000; Shelton and Lilly 2000; Rose and O'Driscoll 2002). Mortality due to starvation is a size-dependent process that represents an alternative (albeit final) response to reduced growth and stunting during food shortages and may be more common than generally perceived (Ney 1990; Persson and Brönmark 2002).

Strong year-classes driven by favorable environmental conditions are a predominant feature of Striped Bass population dynamics (Richards and Rago 1999; Maryland Sea Grant 2009) that may swamp forage supply in upper Bay for periods of time regardless of management regime. Multiple strong year-classes of Striped Bass were produced in upper Bay during 1993-2003 (Durell and Weedon 2015). They were followed by high  $RI$ , low forage indices and ratios, high  $P_0$ , and higher  $M$ . Dominant year-classes had occurred in 1956 and 1958 (Hollis et al. 1967) and an account in Maryland Department of Natural Resources files (circa 1960) on condition of the 1958 dominant year-class of Striped Bass as yearlings described "emaciated Striped Bass" and "Striped Bass in poor condition" that reflected "the enormous population" and an "inadequate food supply". The regulatory regime in place in the late 1950s would have resulted in substantially less demand for forage than management after restoration (Hartman 2003; Uphoff 2003), yet Striped Bass condition was of concern.

Comparisons of PE, C, prey species-specific C, P0, and indices of forage indicated that diets of sublegal sized Striped Bass in fall were more sensitive to major forage species shifts (with the exception of Bay Anchovy) than diets of legal sized fish. Even though their relative abundance was low, Atlantic Menhaden dominated fall diets, by weight, in both size classes of Striped Bass during fall 2006-2014. Consumption of Atlantic Menhaden by sublegal sized Striped Bass reflected a seemingly minor upward trend in relative abundance. Legal sized fish fed largely on Atlantic Menhaden during fall and their success in feeding on them did not reflect the upper Bay Atlantic Menhaden abundance index. Larger Striped Bass may possess physical (increased swimming speed, greater ability to handle larger sized prey, and greater visual acuity) and behavioral skills (more experience in the competitive arena) that allow them to concentrate on Atlantic Menhaden, or they may be compelled to do so to maximize profitability in the trade-off of nutrition obtained and energy expended foraging (Hartman 2000; Ward et al. 2006). Consumption indices for major benthic prey were strongly associated with their relative abundance for both size classes of Striped Bass, suggesting opportunistic feeding. Correlation analyses did not indicate correspondence of Bay Anchovy consumption with relative abundance. Under low abundance indicated by survey indices during 2006-2014, Bay Anchovy constituted 5.8% of fall diet biomass of sublegal sized Striped Bass, contributing more than 25% to diet biomass in three years and less than 6% otherwise; Bay Anchovy comprised 1.2% of legal sized Striped Bass diet biomass across all years. This inconsistency in fall feeding success on Bay Anchovy was present in historical data as well, although interpretation is somewhat clouded by different Striped Bass size classifications among studies. During 1955-1959, Bay Anchovy comprised 38% of Striped Bass (< 600 mm, TL) diet biomass in fall (Griffin and Margraf 2003); the Bay Anchovy seine index for 1959 (earlier years were not available) was high. Bay anchovies comprised less than 5% of fall diets of ages 2+ Striped Bass during 1990-1992 (Hartman and Brandt 1995c), while both Bay Anchovy indices indicated high abundance. Overton et al. (2009) found that during 1998-2001 Bay Anchovy contributed about 30% of fall diet biomass of 300-500 mm, TL, Striped Bass and about 10% of diet biomass of 501-701 mm fish in a region approximating upper Bay. Trawl-based Bay Anchovy indices during 1998-2001 suggested they were more abundant than other major prey during that period, but seine-based Bay Anchovy indices were low.

Our concentration on fall diets did not directly consider some prey items in the “other” category that could be important in other seasons. White Perch (*Morone americana*) and invertebrates other than Blue Crab are important diet items during winter and spring, respectively (Walter et al. 2003; Hartman and Brandt 1995c; Overton 2009). These species did not usually make a large contribution to diet mass during fall, 2006-2014. White Perch from a 2011 dominant year-classes made a large contribution (15.6%) to legal sized Striped Bass diet biomass in 2012.

Age-0 Striped Bass from a strong year-class were encountered diets in both size classes of fish examined during 2011; they were not encountered in other years. Increased cannibalism in Atlantic cod was viewed as a direct result of increased food demand from an increased stock and reduced forage abundance (Ultang 1996), similar to conditions experienced by Striped Bass during 2006-2014.

Associations of PE and C with P0 during fall 2006-2014 indicated that Striped Bass condition may respond quickly to fall feeding success. Fall is a period of active

feeding and growth for resident Striped Bass and forage fish biomass is at its peak (Hartman and Brandt 1995c; Walter and Austin 2003; Overton et al. 2009). Growth patterns of 3 to 6 year-old upper Bay Striped Bass during 1990-1992 exhibited sharp increases in weight during October-November after weight had held steady or declined through summer due to seasonal prey shortages (Hartman and Brandt 1995c; Hartman 2003). Examination of bivariate plots of PE or C with P0 indicated potential for previous feeding history and condition to influence P0 change in fall. We have not completed the diet data base from other seasons to fully address other seasonal feeding patterns.

Overton et al. (2009) provided estimates of percent of Striped Bass stomachs with food during fall 1998-2000 (years combined) from a mid-Bay region that corresponded to our study area that could be converted to PE. Proportion of empty stomachs was 0.54 for fish between 301 and 500 mm TL (approximating our sublegal class) and 0.57 for Striped Bass between 501 and 700 mm (approximating our legal class; Overton et al. 2009). These 1998-2000 estimates were comparable to our highest estimates of PE and reflected high P0 and CPUE, and a nadir in major prey indices (except the Bay Anchovy trawl index) during that period.

Estimates of PE in the 1930s and 1950s suggested considerable variation in feeding success in the past. Hollis (1952) reported PE equaled 0.48 during fall, 1936-1938, for Striped Bass corresponding to our sublegal size class (N = 189). During fall 1956-1958, PE equaled 0.36 (N = 67) for upper Bay Striped Bass corresponding to our sublegal size class; these estimates were made from data used by Griffin and Margraf (2003) to characterize diet during the 1950s (J. Uphoff, unpublished analysis).

All Striped Bass collected during diet monitoring were caught by hook-and-line. Hook-and-line samples are common in marine and estuarine Striped Bass diet studies (Hartman and Brandt 1995b; Walter and Austin 2003; Rudershausen et al. 2005; Nelson et al. 2006; Overton et al. 2008; Overton et al. 2009). Prey availability, schooling behavior of fish, limited search areas and times for fish and fishermen, and non-random behavior of fishermen affect hook-and-line catchability (Peterman and Steer 1981; Johnson and Carpenter 1994; VanDeValk et al. 2005) and could bias diet sampling. Collection gear may affect regurgitation rate, but Chipps and Garvey (2007) did not list hook-and-line sampling as a technique that would result in high rates of regurgitation. Samples analyzed by Hollis (1952) and Griffin and Margraf (2003) were from commercial gill nets, fyke nets, haul seines, pound nets, and by hook-and-line. Most Striped Bass examined in these two studies were collected from pound nets and bias from concentrated prey and feeding within pound nets was possible (Hollis 1952; Overton et al. 2009).

Estimates of RI generally tracked trends in upper Bay Striped Bass juvenile indices (Durell and Weedon 2015) and Atlantic coast stock assessment abundance estimates (ASMFC 2013) that corresponded to ages 2-5 (Uphoff et al. 2015). Exceptions were lower than expected RI during 1989-1993 and higher RI than expected during 2002-2004. These exceptions may reflect changes in angling catchability that were not linearly related to population size (Peterman and Steer 1981; Johnson and Carpenter 1994; VanDeValk et al. 2005) and changes in catch-and-release policy. Catch-and-release fishing was prohibited in upper Bay during 1985-1993 and, while the ban was not easily enforced, many anglers seemed to minimize catch-and-release out of concern for Striped Bass (J. Uphoff, personal observation). A substantial recreational catch-and-release

fishery for Striped Bass arose after the stock recovered and catch-and-release restrictions were removed (Lukacovic and Uphoff 2007).

Diet sampling intensity by CBEF in mid-Bay during October-November, 2006-2013, was comparable to regional effort in Overton et al. (2009). Five to 14 identifiable locations within the larger study area were sampled each year. Overton et al. (2009) sampled eight sites within the same area we sampled. Over the course of four fall seasons, Overton et al. (2009) sampled 702 Striped Bass comparable to our sublegal and legal classes in their mid-Bay region for an average of about 175 fish. The CBEF monitoring program averaged 327 Striped Bass per year (both size classes) during October-November, 2006-2013.

Sampling by CBEF during 2006-2013 and FWHP during 2014 resulted in very different relative sample sizes of sublegal and legal fish. During 2006-2013, CBEF sample sizes of sublegal sized fish averaged 40% of those for legal sized fish. During 2014, 2.6-times more sublegal sized fish were sampled than legal sized ones in FWHP collections.

Due to Mr. Price's health concerns, CBEF collections ended during summer 2015. Collections by CBEF for 2014 will be entered (as will those made for 2015), allowing for comparison with fall consumption estimates based on FWHP sampling. Fish and Wildlife Program sampling in fall will be our platform for future diet and body fat samples.

We had considered estimating P0 for the two size classes used in diet analyses, but found that little information would be lost by combining the estimates based on 2006-2013 data. Differences in values between size classes were minor and trends were closely correlated ( $r = 0.95$ ,  $P < 0.0001$ ).

We did not pursue analyses of forage-to-Striped Bass ratios with P0 and SR since both the numerator and denominator of these ratios were analyzed previously. A general recommendation for data in stock assessments is to only use information once (Cotter et al. 2004).

An upper Bay-only approach for assessing forage and predation was defensible based on migratory behavior of Striped Bass. Contingents that share migration patterns have been identified for Chesapeake Bay Striped Bass based on tagging (Mansueti 1961; Hollis 1967; Setzler et al. 1980; Kohlenstein 1981; Dorazio et al. 1994) and otolith microchemistry (Secor and Piccoli 2007). After spawning in spring, most males and some females (mostly immature) stay within Chesapeake Bay, while the rest migrate to coastal waters (Setzler et al. 1980; Kohlenstein 1981; Dorazio et al. 1994; Secor and Piccoli 2007). Studies of within Chesapeake Bay movements appear confined to upper Bay tagging during 1954-1961 and most of these Striped Bass remained within upper Bay (Mansueti 1961; Hollis 1967; Maryland Sea Grant 2009).

The impact of Striped Bass on its forage base and the impact of forage on Striped Bass are concerns for fisheries management (Hartman 2003; Uphoff 2003; Maryland Sea Grant 2009; SEDAR 2015). Reduced fishing mortality and higher size limits have underpinned management since the mid-1980s, resulting in larger and more abundant Striped Bass and substantial increases in prey demand (Hartman 2003; Uphoff 2003; Maryland Sea Grant 2009; Overton et al. 2015). Few Striped Bass in our legal sized class were present in fall diet studies conducted in the 1930s (Hollis 1952) or 1950s (Griffin 2001; Griffin and Margraf 2003), reflecting effects of smaller minimum length limits and

higher fishing mortality rates on size of surviving fish. An Atlantic Menhaden consumption per Striped Bass recruit analysis indicated that changes to age-at-entry of Striped Bass to the fishery and fishing mortality that approximated past and current regulatory regimes increased demand for Atlantic Menhaden from 11-16% of an unfished population to 31-55% (Uphoff 2003). Overton et al. (2015) estimated that energy consumed per Striped Bass was similar for ages 1-4 during 1955-1959, 1990-1992, and 1998-2001, but age-5 and age-6 fish in the 1950s were able to consume 1.3 and 1.5 times more energy than during 1990-1992 and 1998-2001, respectively. Weight at length of upper Bay Striped Bass in fall samples became more variable following stock recovery in the 1990s, as did length at age of males likely to be residents (ages 4-6) in a spawning season experimental gill net survey (Uphoff 2003; Versak 2015).

With high size limits and low fishing mortality in place since restoration, intraspecific competition for limited forage should be greater for sublegal sized Striped Bass because they compete with one another and legal sized fish. All things being equal, legal sized Striped Bass should forage more efficiently and outcompete sublegal sized fish through greater vision, swimming speed, and experience (Ward et al. 2006). Higher growth rates of sublegal Striped Bass require diversion of more lipids towards growth, while both size classes, primarily consisting of mature males in upper Bay, would divert lipids to reproduction and swimming. However, bioenergetics modeling indicated that excess demand for prey in upper Bay during 1990-1992, when forage-to-Striped Bass ratios were high (nutrition targets were estimated from Striped Bass sampled in fall 1990; Jacobs et al. 2013), was greater for Striped Bass ages 4-6 (approximating the legal-sized class) than younger ages (Hartman and Brandt 1995c). Younger Striped Bass came closer to balancing prey supply and demand, while a dearth of suitably sized larger prey (Atlantic Menhaden and Spot) may have been the primary factor reducing growth of older Striped Bass (Hartman and Brandt 1995b). After the mycobacteriosis epizootic became established in Striped Bass in Chesapeake Bay in the late 1990s, prevalence increased with age, reducing growth and potentially dampening competitive advantages for larger, older fish if nutritional challenges were severe enough (Gauthier et al. 2008; Jacobs et al. 2009b; Latour et al. 2012).

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Table 1. Abbreviations and definitions of indicators and important parameters.

| Abbreviation | Definition   |
|--------------|--|
| C            | Grams of prey consumed per gram of Striped Bass, an indicator of feeding success and prey availability.                  |
| F            | Instantaneous annual fishing mortality rate.   |
| M            | Instantaneous annual natural mortality rate.   |
| PE           | Proportion of Striped Bass with empty stomachs, an indicator of feeding success and prey availability.                   |
| P0           | Proportion of Striped Bass without visible body fat, an indicator of nutritional status (condition).                     |
| RI           | Catch (number harvested and released) of Striped Bass per private and rental boat trip, a measure of relative abundance. |
| SR           | Relative survival of sublegal sized resident Striped Bass to age-3.  |

Table 2. Correlations of survey-based forage indices and proportion of Striped Bass without body fat (P0) during 1998-2014.

| Species and survey | Statistic | P0    |
|--------------------|-----------|-------|
| Atlantic Menhaden  | r         | -0.39 |
| Seine              | P         | 0.12  |
| Bay Anchovy        | r         | -0.02 |
| Seine              | P         | 0.94  |
| Spot               | r         | -0.24 |
| Seine              | P         | 0.35  |
| Blue Crab          | r         | -0.26 |
| Dredge             | P         | 0.32  |
| Bay Anchovy        | r         | 0.63  |
| Trawl              | P         | 0.007 |
| Spot               | r         | -0.45 |
| Trawl              | P         | 0.07  |

Table 3. Correlations of species-specific estimates grams of forage consumed per gram of Striped Bass (C) during October-November and trends in upper Bay forage indices during 2006-2014, by size class. Sublegal sized Striped Bass are < 457 mm, TL, and legal sized fish are  $\geq$  457 mm, TL.

|        |           | Sublegal sized C  |             |         |           |
|--------|-----------|-------------------|-------------|---------|-----------|
| Survey | Statistic | Atlantic Menhaden | Bay Anchovy | Spot    | Blue Crab |
| Seine  | r         | 0.81              | -0.63       | 0.95    |           |
|        | P         | 0.008             | 0.07        | <0.0001 |           |
| Trawl  | r         |                   | 0.003       | 0.97    |           |
|        | P         |                   | 0.99        | <0.0001 |           |
| Dredge | r         |                   |             |         | 0.86      |
|        | P         |                   |             |         | 0.003     |
|        |           | Legal sized C     |             |         |           |
| Seine  | r         | -0.04             | -0.25       | 0.90    |           |
|        | P         | 0.92              | 0.51        | 0.0008  |           |
| Trawl  | r         |                   | -0.26       | 0.92    |           |
|        | P         |                   | 0.50        | 0.0004  |           |
| Dredge | r         |                   |             |         | 0.86      |
|        | P         |                   |             |         | 0.003     |

Table 4. Correlations of relative survival with indices of major forage relative abundance.

| Species, gear     | r    | P      | N  |
|-------------------|------|--------|----|
| Menhaden, seine   | 0.67 | <.0001 | 30 |
| Anchovy, seine    | 0.56 | 0.001  | 30 |
| Anchovy, trawl    | 0.00 | 0.99   | 25 |
| Spot, seine       | 0.26 | 0.17   | 30 |
| Spot, trawl       | 0.48 | 0.02   | 25 |
| Blue Crab, dredge | 0.29 | 0.17   | 25 |

Table 5. Correlations among major forage indices for species where at least one survey was significantly associated with relative survival in Table 4.

| Species          |            | Menhaden | Anchovy | Spot   |
|------------------|------------|----------|---------|--------|
| Gear             | Statistics | Seine    | Seine   | Seine  |
| Anchovy<br>Seine | r          | 0.70     |         |        |
|                  | P          | <.0001   |         |        |
|                  | N          | 30       |         |        |
| Spot<br>Seine    | r          | 0.45     | 0.05    |        |
|                  | P          | 0.01     | 0.80    |        |
|                  | N          | 30       | 30      |        |
| Spot<br>Trawl    | r          | 0.37     | 0.08    | 0.80   |
|                  | P          | 0.07     | 0.69    | <.0001 |
|                  | N          | 26       | 26      | 26     |

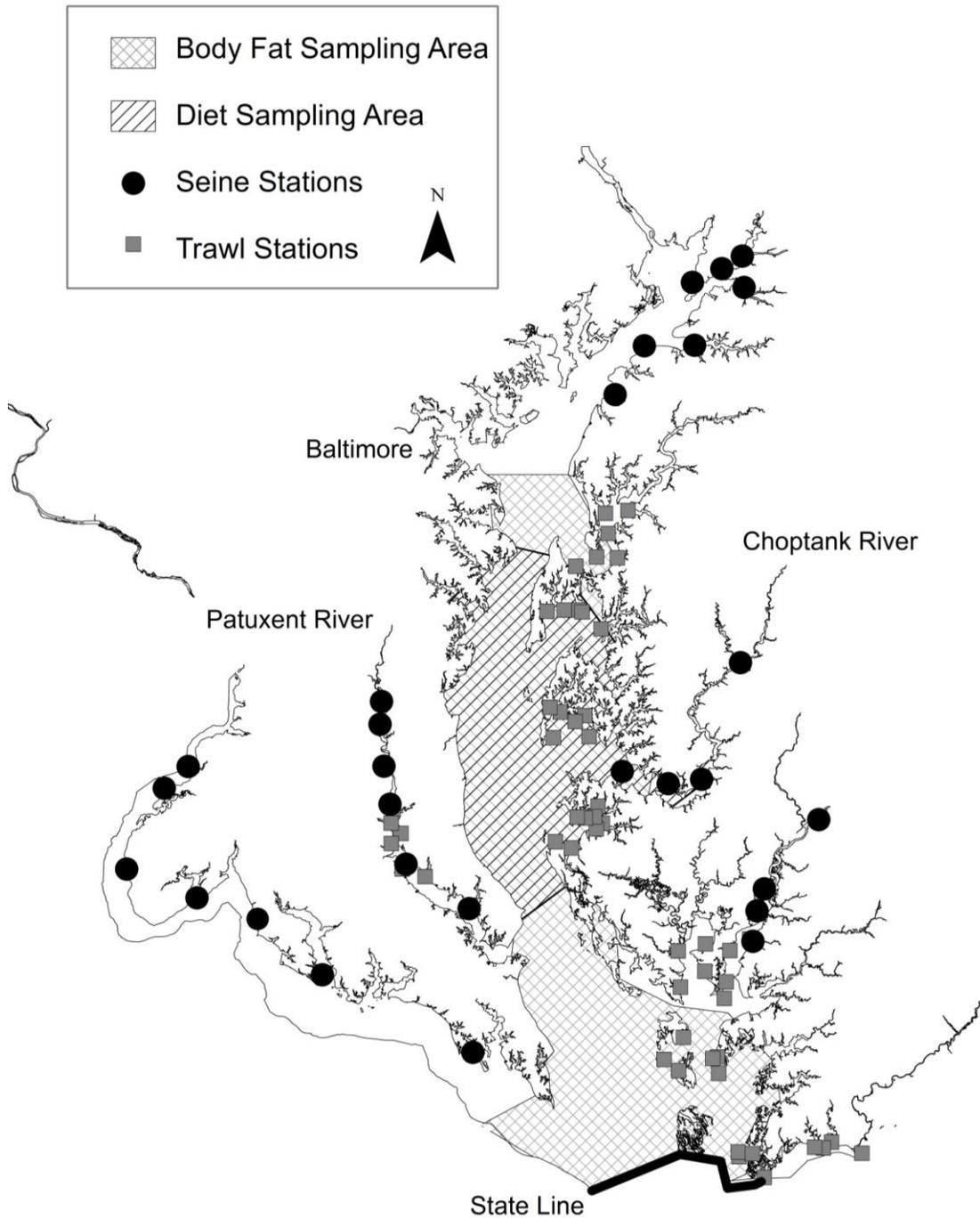


Figure 1. Upper Bay (Maryland's portion of Chesapeake Bay) with locations of forage index sites (black dots = seine site and grey squares = trawl site), and regions sampled for Striped Bass body fat and diet data.

Figure 2. Trends of spawning area gill net survey catch per unit effort (CPUE) of age-3 male Striped Bass and trend in age-3 abundance (N, both sexes) from the Atlantic coast stock assessment during 1985-2012 (ASMFC 2013).

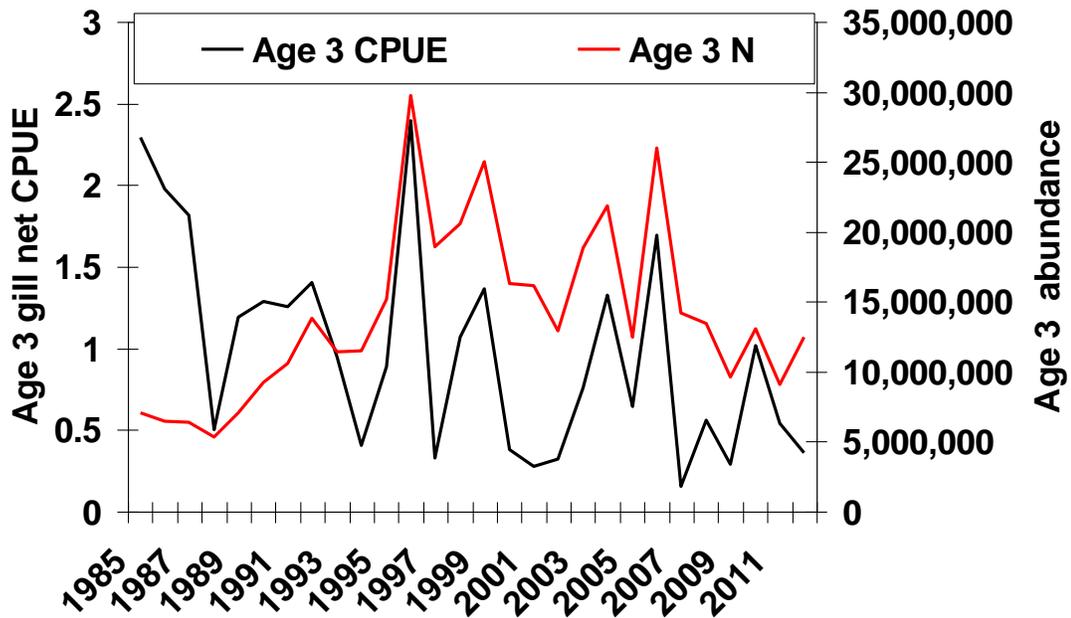


Figure 3. Estimates of the proportion of Striped Bass without body fat and their 95% confidence intervals during October-November, 1998-2014.

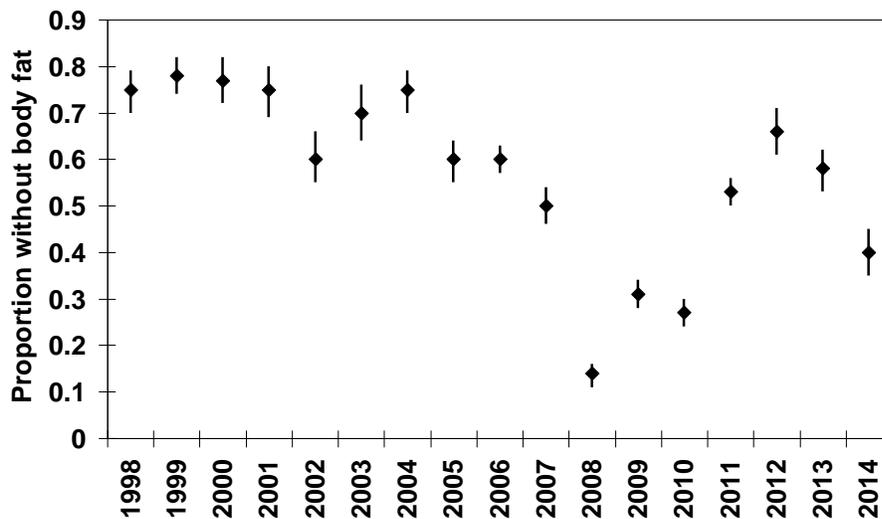


Figure 4. Trends in major pelagic prey of Striped Bass in upper Bay surveys, 1959-2014. Indices were standardized to their 1989-2014 means (time-series in common). Menhaden = Atlantic Menhaden and Anchovy = Bay Anchovy.

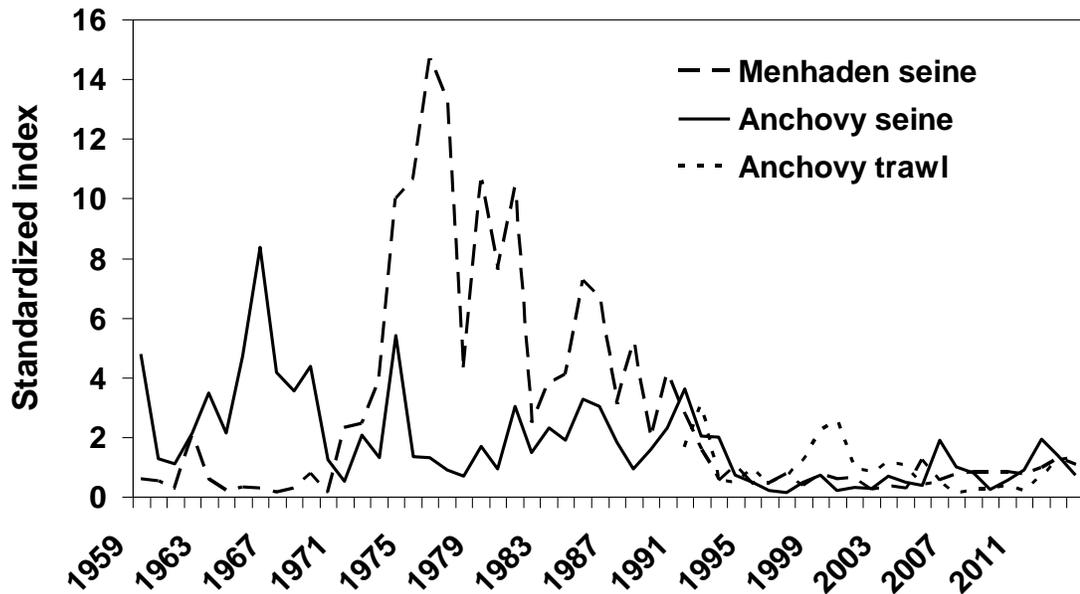


Figure 5. Trends in major benthic prey of Striped Bass in upper Bay surveys. Indices were standardized to their 1989-2014 means (time-series in common).

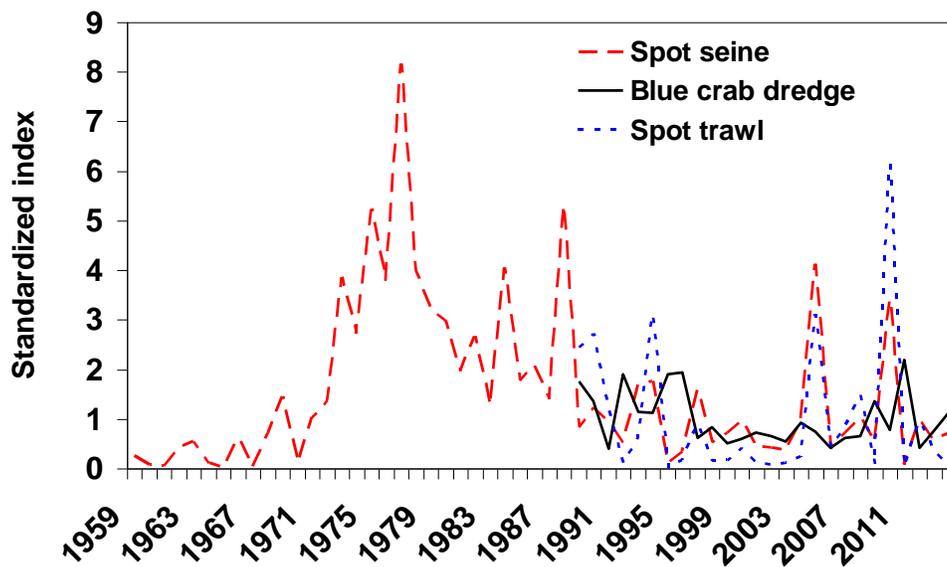


Figure 6. Estimated grams of major forage items consumed per gram of sublegal sized (280-456 mm) Striped Bass during October-November, 2006-2014, and its composition.

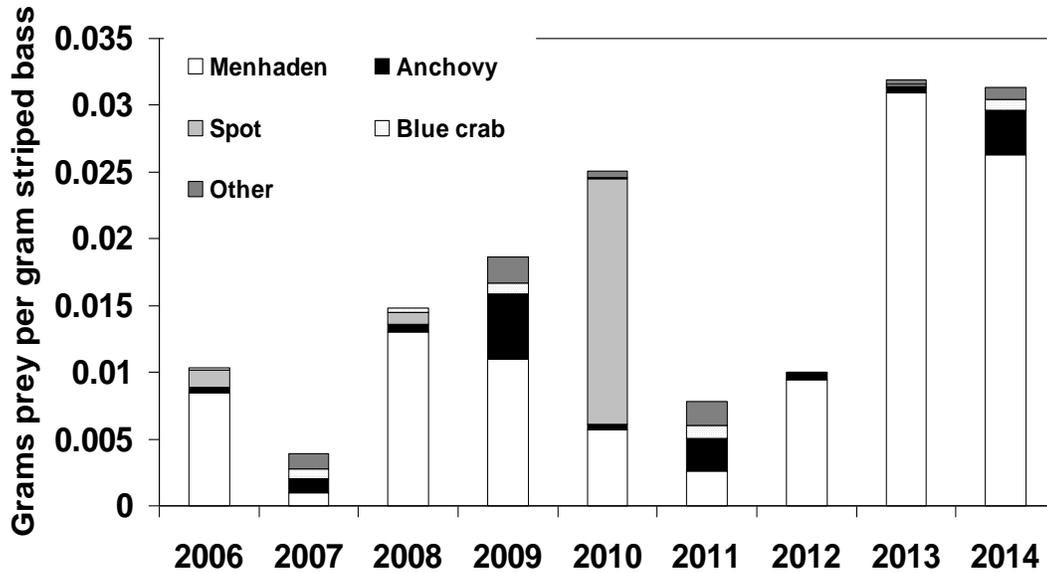


Figure 7. Estimated grams of major forage items consumed per gram of legal sized (457-864 mm) Striped Bass and its composition during October-November, 2006-2014.

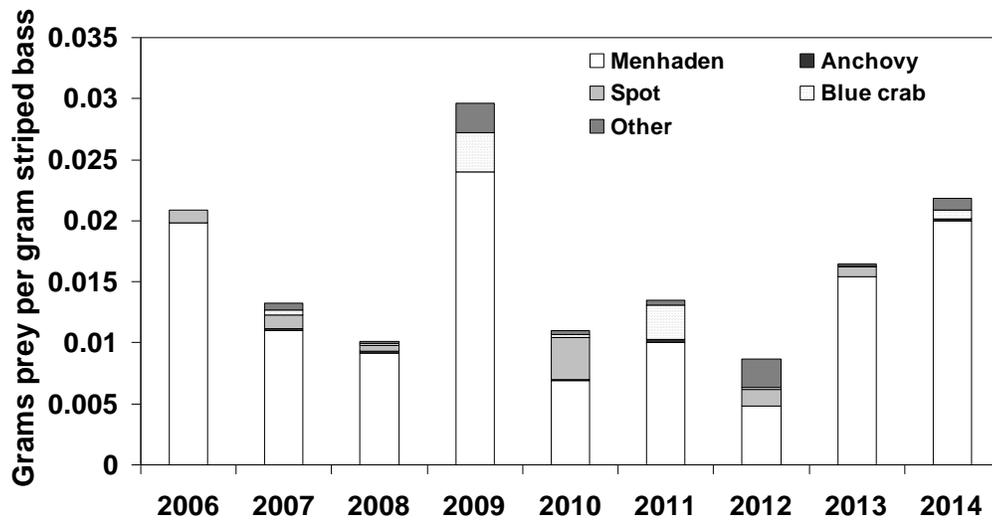


Figure 8. Mean proportion of sublegal sized (286-456 mm) and legal sized (457-864 mm, TL) Striped Bass with empty stomachs during 2006-2013 and its 95% confidence intervals during October-November, 2006-2014.

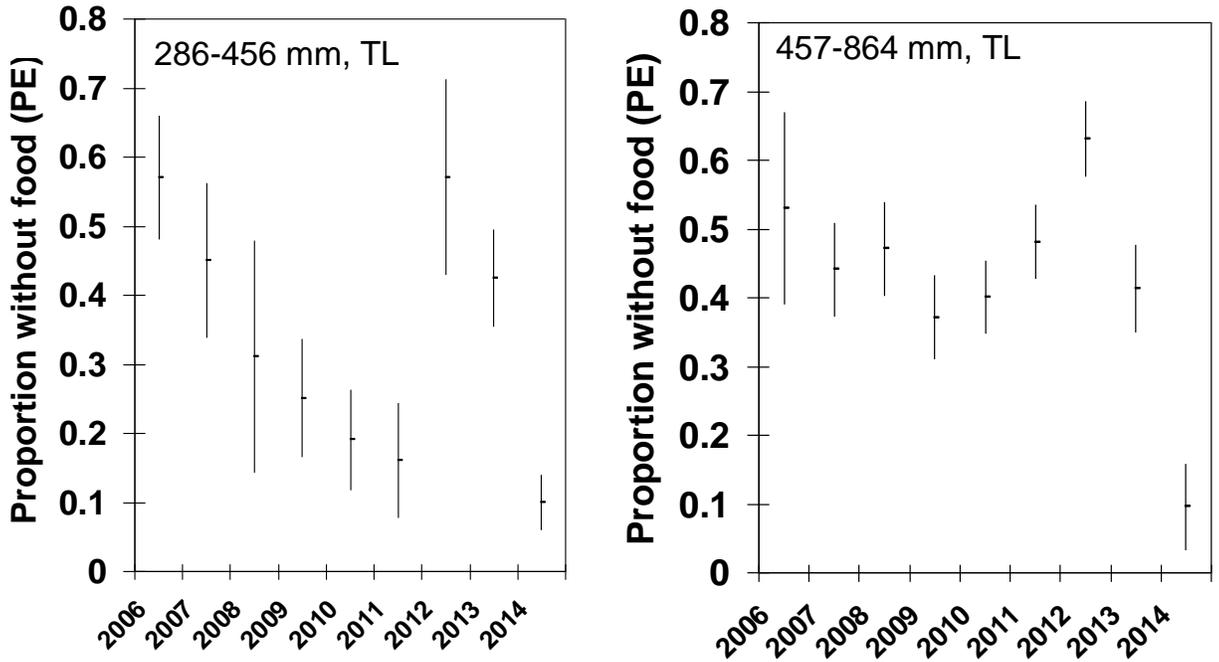


Figure 9. Striped Bass feeding indicators versus proportion without body fat (P0), by size class, during 2006-2014. Red lines indicate 2006-2010 trend and open circle is 2006. Black lines indicate 2011-2014 trend. A is the plot of P0 and proportion of empty stomachs (PE) for sublegal sized fish; B is P0 and PE for legal sized fish; C is P0 and grams consumed per gram of sublegal sized fish; and D is grams consumed per gram of legal sized fish

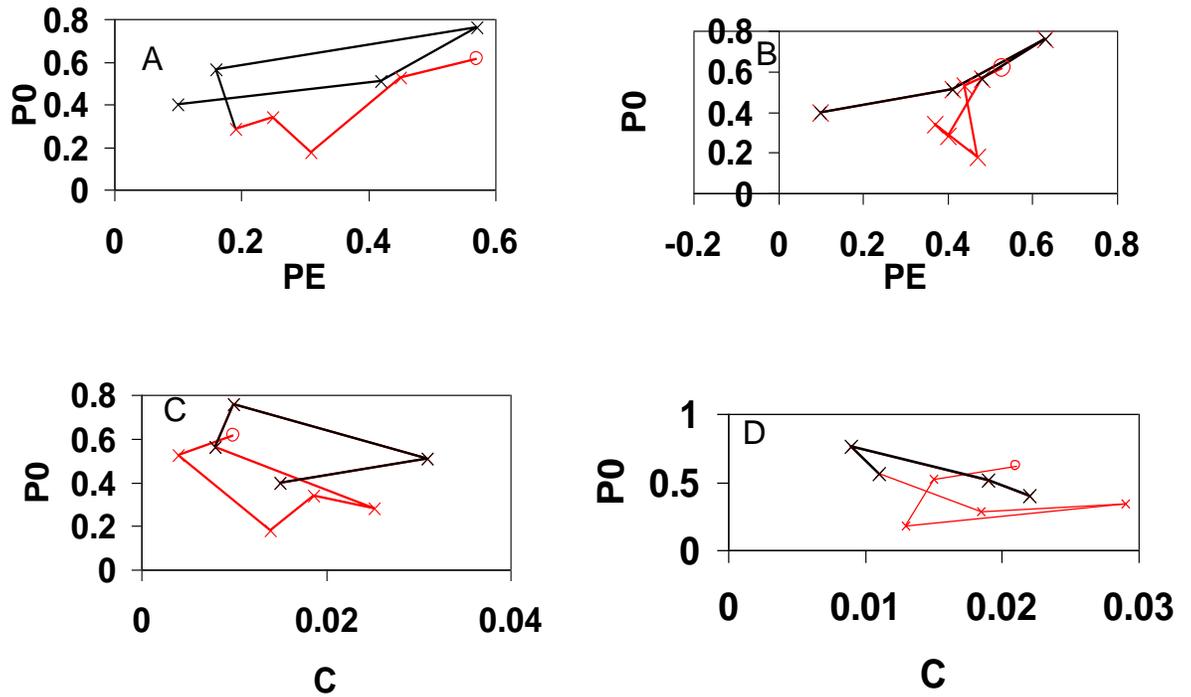


Figure 10. September-October recreational private and rental boat catch-per-trip relative abundance index for Striped Bass (RI) in upper Bay during 1981-2014.

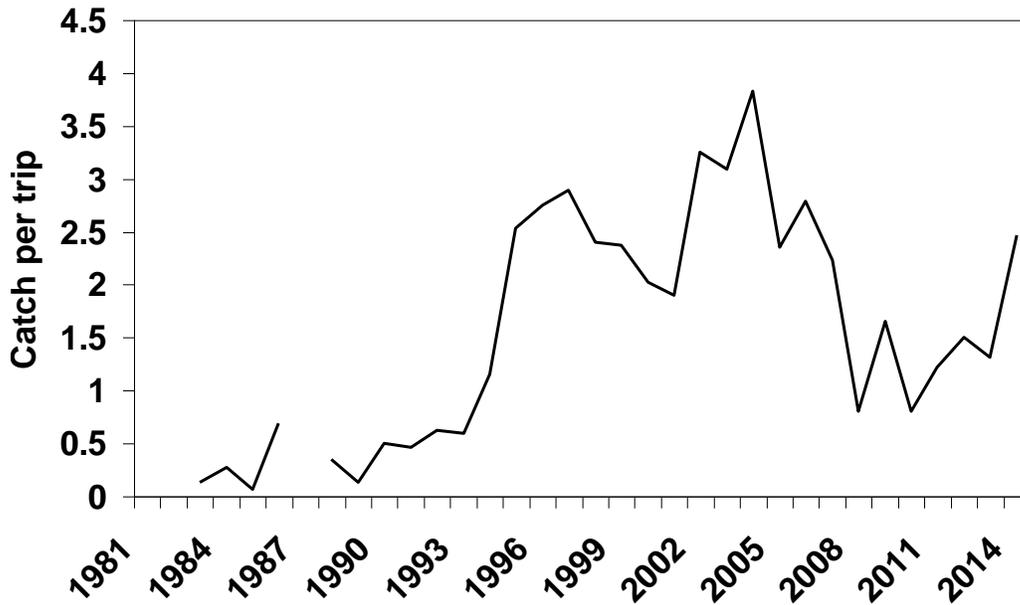


Figure 11. Relationship of proportion of Striped Bass without body fat during October-November and relative abundance of upper Bay striped bass (recreational catch-per-trip index) during 1998-2014. Squares = observations; solid line = prediction; and dotted line indicates the body fat target (below line = good condition).

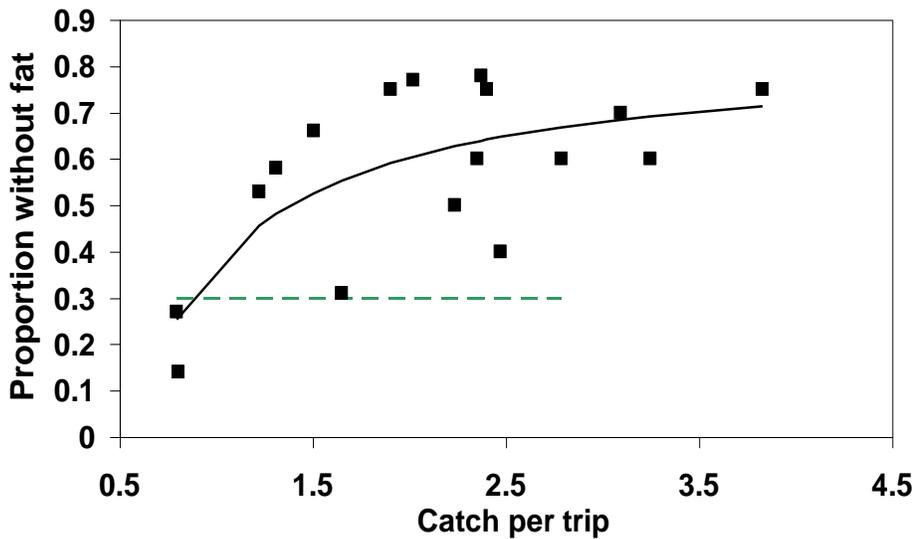


Figure 12. Trends in pelagic forage index-to-Striped Bass index (RI) ratios, 1983-2014. Menhaden = Atlantic Menhaden and Anchovy = Bay Anchovy. Note  $\log_{10}$  scale.

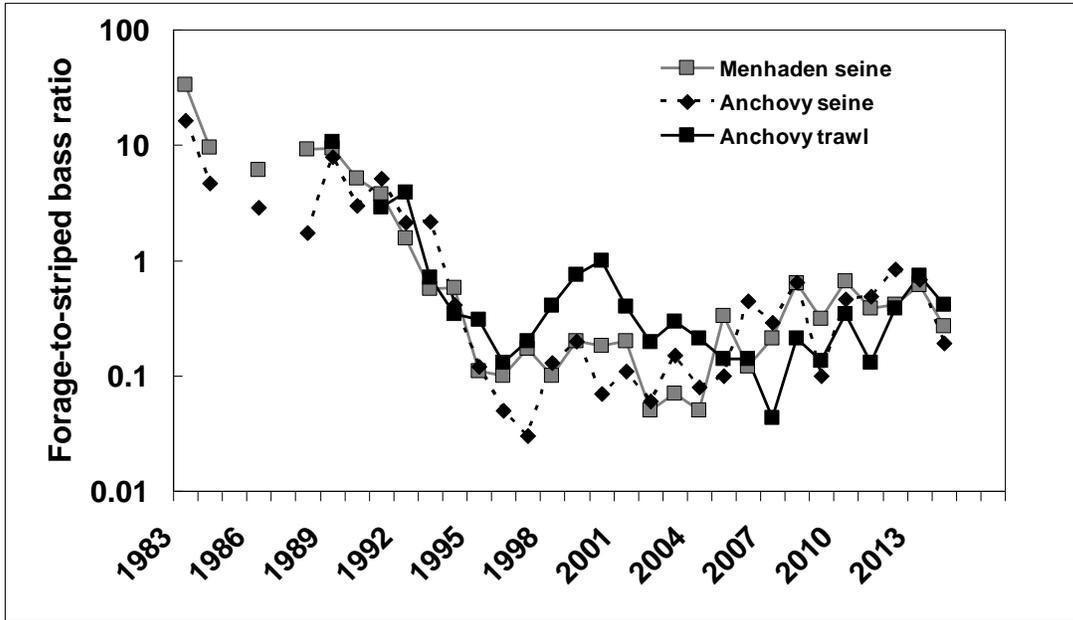


Figure 13. Trends in benthic forage index-to-Striped Bass index (RI) ratios, 1983-2014. Note  $\log_{10}$  scale.

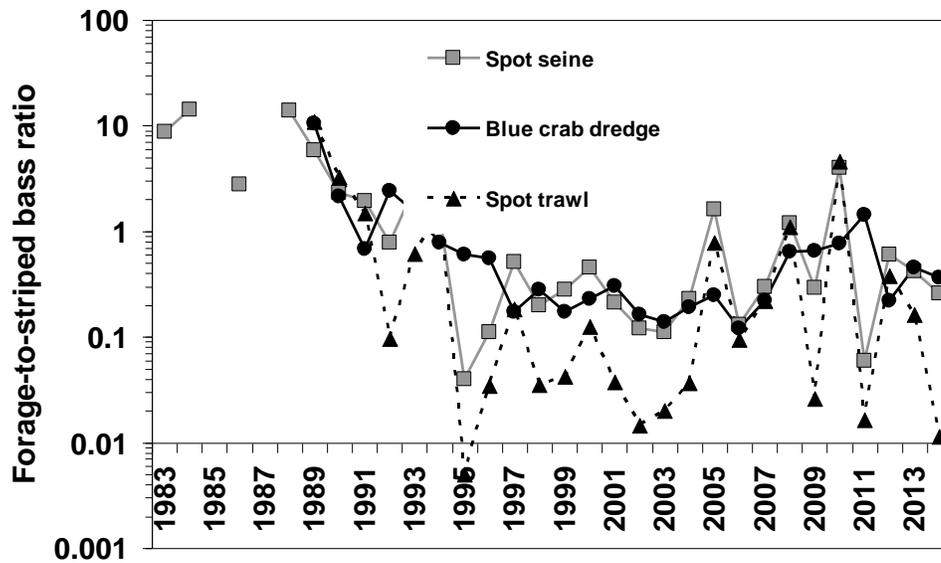


Figure 14. Relationship of relative catchability ( $q$ ) of age-3 male Striped Bass in spawning season and area gill net surveys to age-3 Atlantic coast abundance estimates (ASMFC 2013) during 1985-2012. Squares = observations; solid red line = predicted; and dashed black line = mean relative  $q$ .

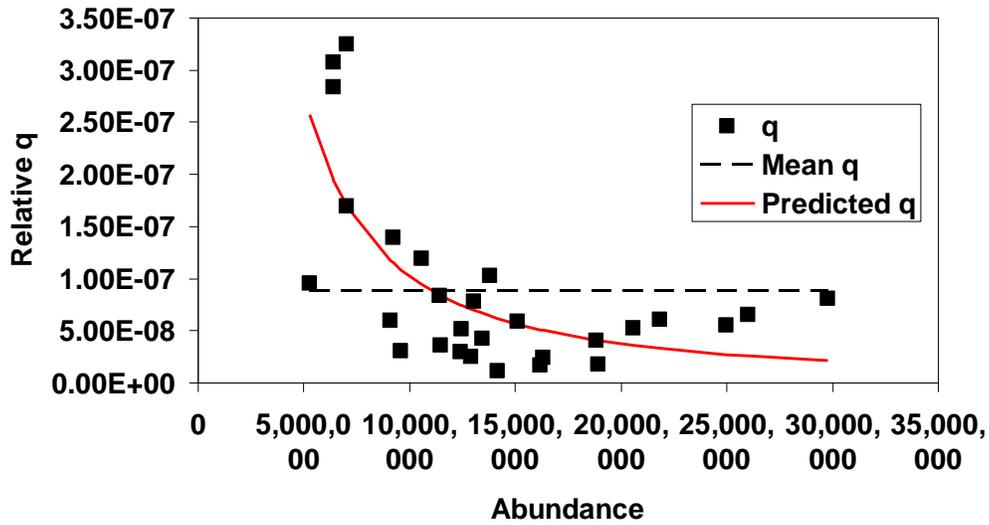


Figure 15. Observed and adjusted gill net catch per unit effort of age-3 male Striped Bass (CPUE; standardized to time-series mean) in spawning area surveys.

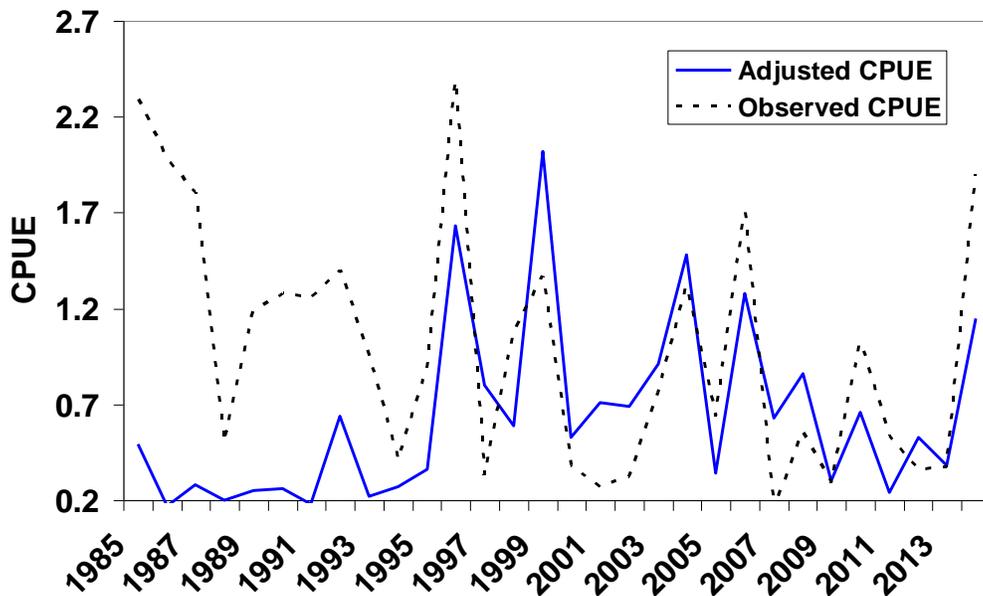


Figure 16. Relationship of adjusted gill net survey catch per effort (CPUE) of age-3 Striped Bass males to their estimated abundance along Atlantic coast (N; ASMFC 2013).

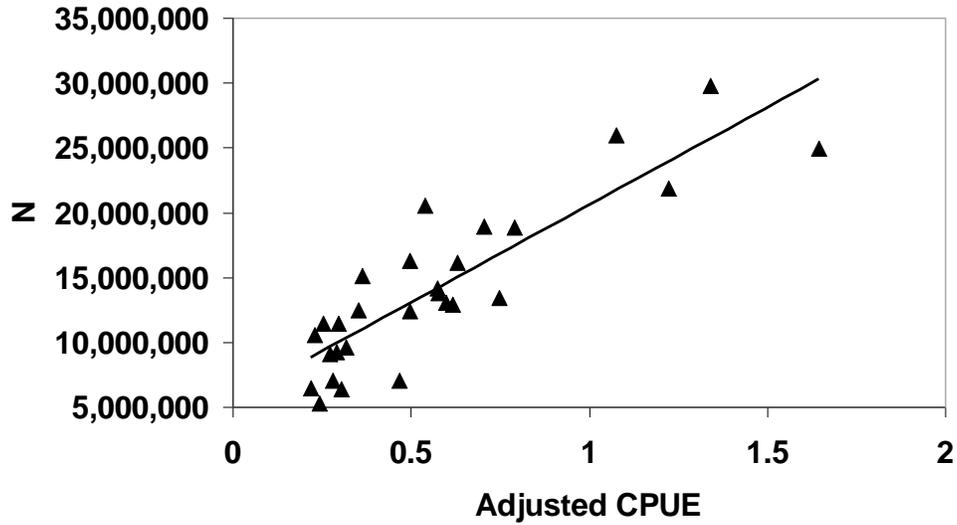


Figure 17. Trends in Adjusted gill net survey catch per effort (CPUE) of age-3 male Striped Bass and the Maryland baywide juvenile index (JI) three years previous .

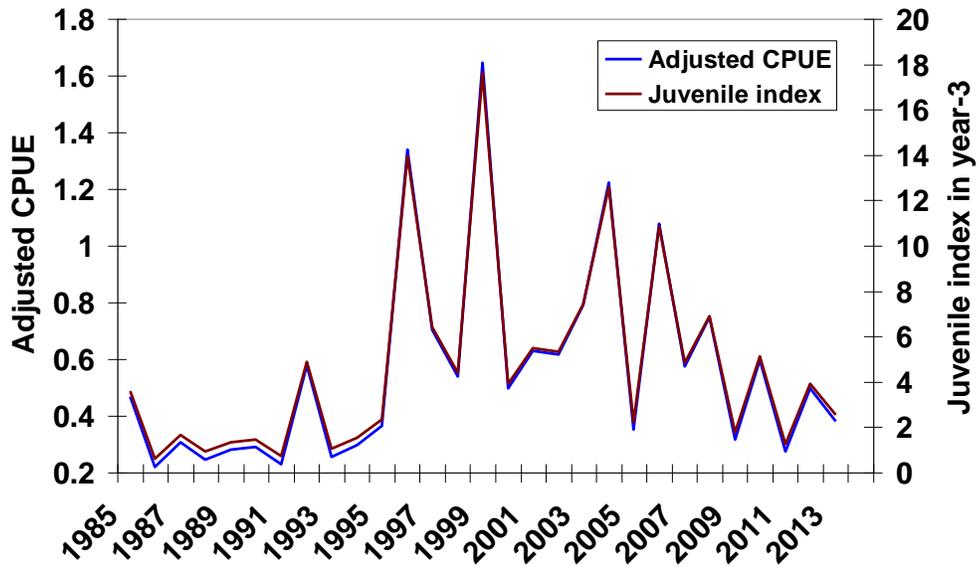


Figure 18. Relative survival of age-3 Striped Bass in upper Bay (adjusted gill net index / juvenile index in year-3; solid line) and trends in tag-based estimates of survival of 457-711 mm, TL, Striped Bass (ASMFC 2013) in Chesapeake Bay (dashed lines). Both estimates indicated trend in survival related to natural mortality. Values are not comparable between estimates.

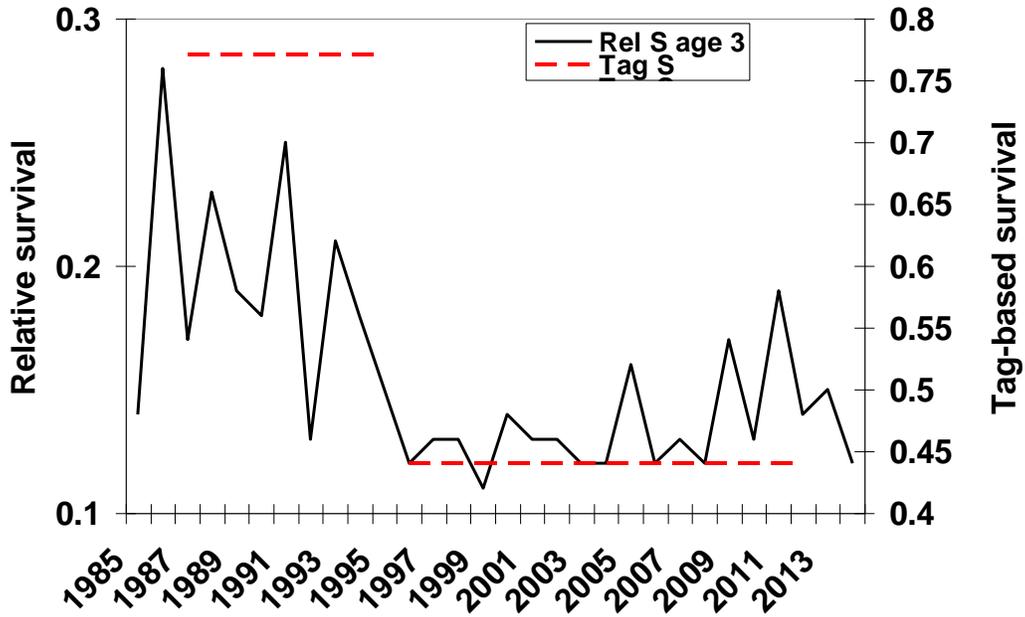


Figure 19. Relative survival of age-3 Striped Bass in upper Bay (adjusted gill net index / juvenile index in year-3; solid line) and annual tag-based estimates of survival of 457-711 mm, TL, Striped Bass during 1987-2008 (ASMFC 2010) in Chesapeake Bay (dashed lines). Both estimates indicated trend in survival related to natural mortality. Values are not comparable between estimates.

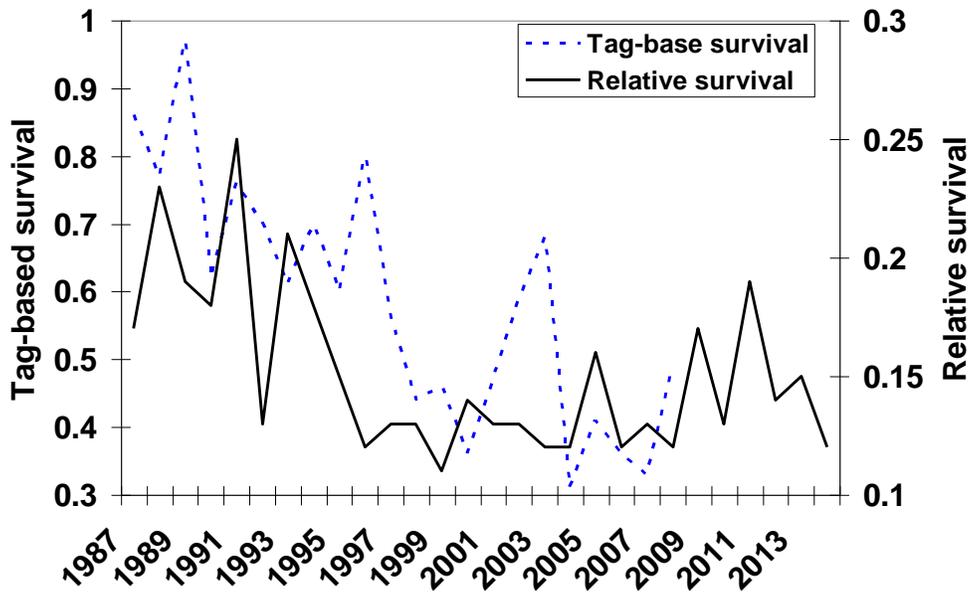


Figure 20. Relative survival for age-3 Striped Bass in upper Bay based on adjusted gill net indices or estimated abundance (N) of age-3 Atlantic coast Striped Bass (standardized to its mean as the numerator of the ratio). The Maryland baywide juvenile index three years prior was the denominator in both cases. Both estimates indicated trend in survival related to natural mortality. Values are not comparable between estimates.

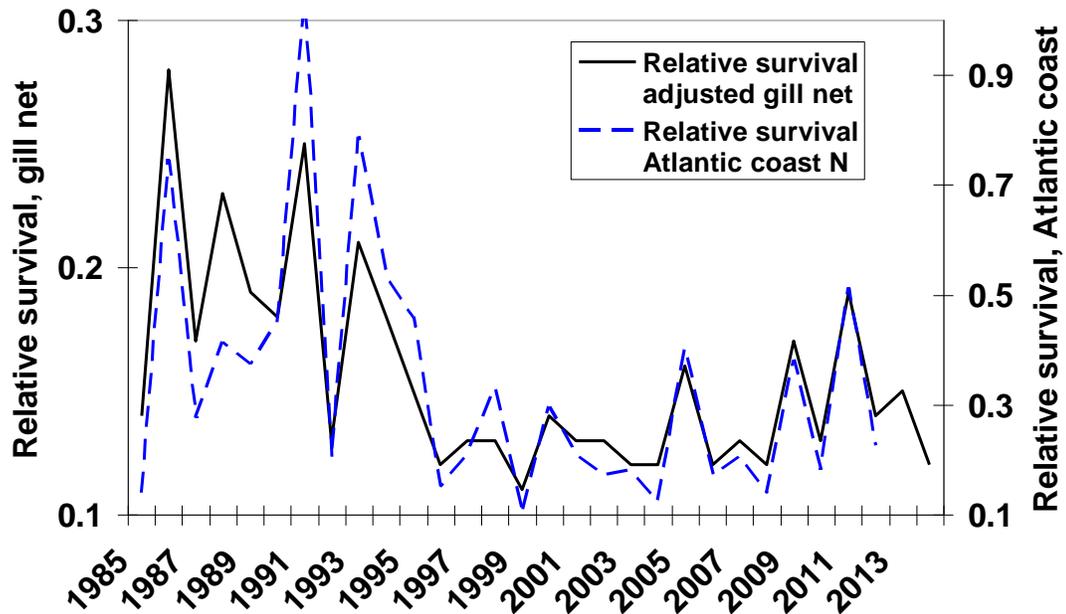
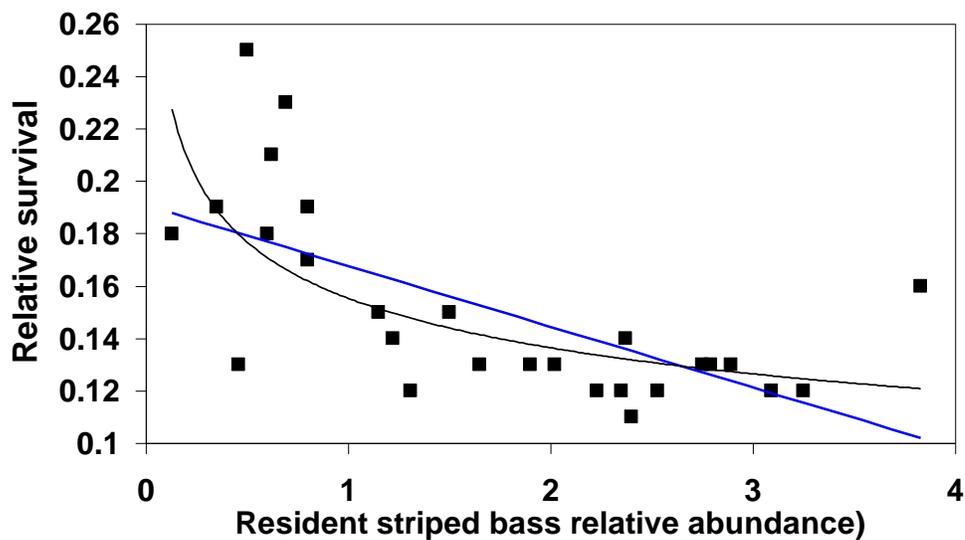


Figure 21. Relationships of relative survival of age-3 Striped Bass (adjusted gill net index / juvenile index in year-3) to relative abundance of resident Striped Bass RI or September-October catch per private boat trip) during 1986-2014.



Appendix 1a. Data sheet instructions for processing FWHP Striped Bass stomach contents.

Bass ID - Record the ID for fish. Do not use dashes. This ID will match these diet data to that individual fish's health data.

Prey Sequence # - This is the arbitrary order that diet items are processed. For anchovies, and Soft and Razor Clams from samples with more than 10 prey items, record the individual samples as 1-10 and the total count and mass of the remaining as 11 (or some other number when you get there if other organisms were eaten in large numbers). The count / mass of the remaining sample is just one number for sequence #.

Prey ID – Labels for commonly encountered prey are provided (Appendix 1b). Use these consistently. Anchovy indicates Bay Anchovy. Striped Anchovy should be identified separately should they ever be encountered. Unknown fish is one that is unidentifiable. If you encounter a new species, identify and record it.

Data = Count. Use that for count data. Enter 0 for none, 1 for any individual fish, and the count for any type of pooled sample where you counted all the organisms (exclude those that were measured individually).

Data = Present. Use for presence-absence. 0 indicates absence if none is the prey ID and 1 indicates the organism was present.

If Prey ID is “None”, enter 0 in both Data = count and Data = present. It's redundant, but avoids having to interpret or insert a 0. All other counts and presence entries only have a blank as an alternative to an entry.

TL is total length in mm – TL is for fish, carapace width (CW) for Blue Crab, shell maximum length (SHL) for Soft and Razor Clams.

Individual mass – record the number determined by scale or volume of the individual organism.

Mass unit – record weight in grams and volume in ml.

Total mass – mass of organisms not measured individually.

Mass unit – record weight in grams and volume in ml.

Appendix 1b. Prey ID diet codes for processing FWHP Striped Bass stomach contents.

| Data type | Item                          | Count or Present | Whole Individual Length | Whole Individual Wt or Vol | Total Wt or Vol | Comment |
|-----------|-------------------------------|------------------|-------------------------|----------------------------|-----------------|---------|
| 1         | crab, blue                    | C                | CW                      | Yes                        | Yes             | No bait |
| 1         | croaker                       | C                | TL                      | Yes                        | Yes             |         |
| 1         | goby                          | C                | TL                      | Yes                        | Yes             |         |
| 1         | herring                       | C                | TL                      | Yes                        | Yes             |         |
| 1         | herring, blueback             | C                | TL                      | Yes                        | Yes             |         |
| 1         | menhaden                      | C                | TL                      | Yes                        | Yes             | No bait |
| 1         | none                          | C                |                         |                            |                 |         |
| 1         | spot                          | C                | TL                      | Yes                        | Yes             | No bait |
| 1         | unknown fish                  | C                |                         |                            | Yes             |         |
| 1         | white perch                   | C                | TL                      | Yes                        | Yes             | No bait |
| 1         | anchovy                       | C                | TL up to 10             | Yes                        | Yes for rest    |         |
| 1         | silverside                    | C                | TL                      | Yes                        |                 |         |
| 2         | clam, razor                   | P or C           | SHL                     | Yes                        | Yes             | No bait |
| 2         | clam, soft                    | P or C           | SHL                     | Yes                        | Yes             |         |
| 2         | crab, mud                     | C                |                         |                            | Yes             |         |
| 2         | shrimp                        | P or C           |                         |                            | Yes             |         |
| 2         | shrimp, grass                 | P or C           |                         |                            | Yes             |         |
| 2         | shrimp, mantis                | P or C           |                         |                            | Yes             |         |
| 2         | shrimp, sand                  | P or C           |                         |                            | Yes             |         |
| 2         | tunicate                      | P or C           |                         |                            | Yes             |         |
| 3         | amphipod                      | P                |                         |                            | Yes             |         |
| 3         | clam                          | P                | SHL                     | Yes                        | Yes             |         |
| 3         | polychaete                    | P                |                         |                            | Yes             |         |
| 3         | soft invert                   | P                |                         |                            | Yes             |         |
| 3         | soft invert w/ shell residue, | P                |                         |                            | Yes             |         |
| 3         | unknown                       | P                |                         |                            | Yes             |         |

|   |                        |                            |              |
|---|------------------------|----------------------------|--------------|
| 4 | Spine                  | P (parts) or<br>C (intact) | BL if intact |
| 5 | parasitic<br>arthropod | P                          |              |

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Appendix 1b (cont).

P or C = type choice. P if parts or can't be counted; C otherwise  
P = presence of uncombinable parts or residue  
C = counts for whole items, combined parts forming an individual,  
or separate parts  
Lengths are in mm  
Lengths for whole or parts combinable into whole fish estimate.  
TL = total length  
BL = backbone length  
SHL = shell max length  
CW = carapace width  
Weight units are grams (g)  
Volume units are ml  
Unknown fish = unidentifiable due to digestion  
Other species can be added

