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MARINE AND ESTUARINE FINFISH ECOLOGICAL AND HABITAT INVESTIGATIONS

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Fishing and Boating Services
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December, 2022

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For today and tomorrow,
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inspires people to enjoy and live in
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and to protect what makes Maryland
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Bay, our diverse landscapes and our
living and natural resources.

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This report was completed during December, 2022. It consists of summaries of activities for Projects 1–4 under this grant cycle. All pages are numbered sequentially; there are no separate page numbering systems for each Project. Project 1 activities are reported in separate numbered sections. For example, Project 1, section 1 would cover development reference points (Project 1) for stream spawning habitat of anadromous fish (Section 1). Tables in Project 1 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://dnr.maryland.gov/fisheries/Pages/FHEP/pubs.aspx . Table 1 provides the page number for each Project and section.

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

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MD - Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern

Executive Summary

Purpose - Project 1 investigates two general alternative hypotheses relating recreationally important species to development and-or agriculture. The first hypothesis is that there is a level of a particular land-use that does not significantly alter habitat suitability and the second is that there is a threshold level of land-use that significantly reduces habitat suitability (production from this habitat diminishes). The null hypothesis would be an absence of differences. In general, we expect habitat deterioration to manifest itself as reduced survival of sensitive live stages (usually eggs or larvae) or limitations on use of habitat for spawning or growth (eggs-adults). In either case, we would expect that stress from habitat would be reflected by dynamics of critical life stages (abundance, survival, growth, condition, etc.).

Spatial Analyses - We used property tax map-based counts of structures (C) in a watershed, standardized to hectares (C/ha), as our indicator of watershed development. Estimates of C/ha that were equivalent to watershed development reference points for Chesapeake Bay fisheries of 5% IS (impervious surface; 5% = 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 0.37, 0.86, and 1.35 C/ha, respectively. Maryland Department of Planning estimates of percent of watershed in agriculture, forest, and wetlands in 2010 were used for other land use categories.

Recent improvements to spatial resolution of land cover necessitated revisiting the relationship between C/ha and %IS. Recent land cover estimates became available at 1 m x 1 m resolution for the entire Chesapeake Bay watershed; resolution of land use data currently used to estimate %IS from C/ha have 30m x 30 m resolution. A non-linear power function provided a very good fit to the high-resolution data and will be used in the future to predict %IS from C/ha. Estimates of C/ha that were equivalent to 5% IS, 10% IS, and 15% IS) were estimated as 0.31, 0.84, and 1.51 C/ha, respectively. The relationship that produced these estimates was developed too late for this report.

Section 1, Stream Ichthyoplankton - Anadromous fish spawning in Patuxent River was assessed during 2021 and these data were added to the time-series that began in 2005 (9 watersheds; N = 36). Watershed land use for these watersheds ranged from rural to suburban. Proportion of plankton net samples with Herring eggs and-or larvae (P_{herr}) provided a reasonably precise indicator of habitat occupation. Regression analyses that included spawning stock categories (0 for low during 2005-2011 and 1 for high during 2012-2019), indicated significant and logical relationships among P_{herr} and C/ha (R^2 = 0.73) or P_{herr} and conductivity (R^2 = 0.67) consistent with the hypothesis that development was detrimental to stream spawning. Predicted P_{herr} declined by 50% over the range of observed C/ha (0.07-1.52); and increased by 60% between the two spawning stock categories. Predicted P_{herr} declined by 50% over the range of observed conductivity standardized to its baseline (1.14-2.19) and increased by 66% between the two spawning stock categories. The high spawning stock category in the analysis of 2005-2021 corresponded with closure of Maryland's River Herring fisheries in 2011, closure of most other in-river fisheries along the Atlantic Coast by 2012, and caps on River Herring bycatch in coastal Atlantic Herring and Atlantic Mackerel fisheries.

Estimates of P_{herr} were consistently high in watersheds dominated by agriculture. Importance of forest cover could not be assessed with confidence since forest cover estimates may have included residential tree cover. Conductivity was positively related with C/ha in our analysis and with urbanization in other studies. General development targets and limits for C/ha or IS worked reasonably well in characterizing habitat conditions for stream spawning of Herring.

Preliminary data from Patuxent River suggested a localized impact of development on Herring spawning. Stream drift net stations, located above the tide line at Route 214, that were within or just below the developed Laurel-Bowie area had much lower P_{herr} (0.17; N = 36) than stations below this region in the tidal, more rural portion of the watershed that was sampled by conical nets towed by boat (P_{herr} = 0.89; N = 47). The Choptank River, sampled in 2017, had a similar sampling design (drift nets upstream and boat samples downstream; Uphoff et al. 2018), but was subject to low development throughout the watershed. Estimated P_{herr} in Choptank River during 2017 was 0.74 (N = 43) upstream where drift nets were employed and 0.88 (N = 58) where boat samples were taken.

Section 2: Section 2, Yellow Perch Larval Presence-Absence Sampling – Sassafras and Choptank Rivers were sampled with conical plankton nets towed from boats during 2021. These data were added to the time-series that extends back to 1965 (18 watersheds, N = 104). Data were either mined from historical surveys (intermittent availability) or were conducted specifically for Yellow Perch larval presence absence. 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 postlarval stage.

Estimates of L_p were negatively related to development and positively associated with forest and agriculture. Development was an important influence on Yellow Perch egg and larval dynamics and negative changes generally conformed to development targets and thresholds. Widespread low L_p occurred sporadically in Chesapeake Bay subestuaries with rural watersheds and appeared to be linked to high winter temperatures.

Years of high spring discharge favor anadromous fish recruitment in Chesapeake Bay and may represent episodes of hydrologic transport of accumulated organic matter from riparian marshes and forests of watersheds that fuel zooplankton production and feeding success. Amount of organic matter present in L_p samples was negatively influenced by development in Chesapeake Bay subestuaries. Wetlands appeared to be an important source of organic matter in the subestuaries we studied.

Section 2.1: Striped Bass spawning and larval habitat status – This section deviates from the Project description described in *Purpose*. An overfishing declaration and successive poor year-classes of Striped Bass in Maryland spawning areas during 2019-2021 sparked concern about degradation of Striped Bass spawning and larval nursery habitat in Chesapeake. We have assembled historical data and oriented some of our spring monitoring to respond to these concerns.

We updated the proportion of tows with Striped Bass eggs (*Ep;* an indicator of spawning stock), the Maryland baywide juvenile index (JI; an indicator of recruitment), and a relative larval survival index (RLS, baywide JI / *Ep*). These time-series start in the mid- to late 1950s. Trends in pH, conductivity, and alkalinity in the Choptank River were compared between the 1980s and 2014-2021. We added analyses to detect changes in historic (1950s-present) spawning area temperature and flow patterns that may be influencing recent year-class success of

Striped Bass in the four major spawning areas (Head-of-Bay and Potomac, Choptank, and Nanticoke rivers).

Egg production in 2021, based on baywide *Ep* (0.67), was not in the top tier of estimates (roughly 0.80 or greater), but there was a very high chance it was above levels when it was depleted enough to affect year-class success during 1982-1988. Estimated RLS in 2021 was just above a poor survival criterion; most of the poor RLS estimates were concentrated in 1980-1991. Measurements of pH in Choptank River between 1986-1991 and 2013-2021 indicated improvement (higher, more stable averages and less variability of individual measurements) that would have lowered toxicity of metals implicated in poor recruitment in some Striped Bass spawning areas during the 1980s. Average alkalinity was at least 3-times higher in 2021 compared to 1986-1991. It seems unlikely that poor survival of larvae during 2019-2021 could be attributed to a return of toxic water quality conditions implicated in poor recruitment during the 1980s.

Means or medians of days between 12ºC and 20ºC water temperature milestones indicative of the beginning and ending of spawning, respectively, during 2000-2021 were 10 days to 12 days shorter (respectively) than during 1954-1992 in Choptank and Nanticoke rivers. Changes were not uniform among temperature milestones. Early milestones (first egg collected and 12ºC) appeared to be the least affected and later milestones (16ºC and 20ºC) were progressively earlier. The portion of the spawning period when most eggs were collected in historic collections with counts (days from 12ºC to 16ºC) has shortened and potentially lethal high temperatures (indicated by days to 20ºC) were being reached earlier. In addition to these general changes, 3 years during 2000-2021 (of 9 available) had very short spans between 12ºC and 16ºC (2 days) and 2021 had the earliest date that 12ºC was reached in the entire time-series. Our temperature milestones generally captured most Striped Bass egg and larval production based on counts in historic datasets.

Below average flow conditions were less conducive to formation of strong year-classes (JIs in the top quartile) and poor year-classes (JIs in the bottom quartile) were more likely. Above average flows resulted in a higher chance that strong year-classes would be formed and a modest reduction in occurrence of poor year-classes. When all spawning areas were combined during the recent period of high productivity were combined into a single analysis (1993-2020; N = 112 area and year combinations), there were 4 strong year-classes when flows were below average and 24 strong year-classes when flow was at or above average. There were 17 poor year-classes when flow was below average and 13 when it was at or above average. When 1993-2020 was split in half (14 years each), below average flows were less common during the first half (1993-2006) than the second in the Potomac (7 in the first half and 10 in the second), Choptank (4 and 7), and Nanticoke (5 and 8) rivers. There was no change in the Head-of-Bay (5 years of below average flow in each half). Frequency of below average flow conditions has increased since 2006 in 3 of the 4 spawning areas (no change in Susquehanna River), increasing odds that a lesser year-class will be formed and decreasing the odds that strong baywide year-class will form.

Managing for low exploitation rates and high spawning stock would be expected to provide extended age structure that allows for diverse spawning behaviors over a protracted time period that are expected to stabilize recruitment in the face of warming winter and spring temperatures. However, the time span between temperature milestones contracted in the Choptank River in the last two decades, concentrating egg production in a shorter period. When spawning is concentrated in a shorter time period, egg mortality events kill a larger proportion of a year's spawn. It is unclear whether increased egg production within this compressed spawning window can offset temperature related egg mortality numerically, but potential for more eggs to result in more larvae cannot be ruled out. However, if mistiming of zooplankton blooms with first-feeding larvae is important, its density-dependent nature may limit successful management. It becomes possible in years of higher survival of eggs that larval survival and subsequent recruitment will be capped at a low level due to inadequate larval foraging no matter how many eggs are pumped into the spawning area if zooplankton production is misaligned with first-feeding larvae.

Section 3 - Estuarine Fish Community Sampling - Subestuaries sampled during summer 2021 were limited by a COVID-19 hiring freeze. We sampled Tred Avon River (C/ha = 0.78); a developing, mesohaline tributary of the Choptank River located in Talbot County, continuing a time-series started in 2006. We also sampled rural, tidal-fresh Sassafras River (C/ha = 0.11), located at the Head-of-Bay, for a second year. These data were added to the time-series that started in 2003 (23 watersheds, N = 150).

Mean bottom DO remained below the 3.0 mg/L threshold at Tred Avon River's station 01 (nearest development in Easton) during July-September, 2021; mean bottom DO for stations 02 and 03 remained under the time-series median, while station 04 (furthest downstream) increased above the time-series median. A decline in bottom DO at station 02 has been observed over the last five years and may represent downstream progression of declining water quality caused by increased development. Frequency of below target (3.0 mg/L) and threshold bottom DO measurements have increased in Tred Avon River since 2006, primarily reflecting deterioration at station 01. Less developed watersheds adjacent to Tred Avon River have not exhibited deterioration. Five species were in the top 90% of finfish caught in the Tred Avon River from 2006 to 2021, Bay Anchovy (56.8%), Spot (17.0%), White Perch (adults and juveniles; 7.7%), Hogchoker (7.3%), and Striped Bass (adults and juveniles; 3.5%). An additional 32 species comprised the last 7.6%. Changes in modified PSDs for White Perch in Tred Avon River for trawl samples primarily reflected recruitment of year-classes into the stock or quality categories; only about 40% of modified PSDs were above 10% in Tred Avon River.

The Sassafras River watershed is predominately agricultural. In 2021, bottom DO readings for the Sassafras River did not fall below the threshold but did fall below the 5 mg/L target (8%). Sassafras River bottom trawl catches for 2020–2021 were composed of White Perch (adults and juveniles; 72.4%), Spot (12.4%), Blue Catfish (4.7%), and other species (9 species; 10.5%). Eight species comprised the top 90% of species of finfish in seine samples: White Perch (juveniles and adults; 43.5%), Spottail Shiner (11.0%), Gizzard Shad (10.7%), Blueback Herring (6.8%), Pumpkinseed (5.2%), Inland Silverside (4.6%), Striped Bass (juveniles and adults; 4.2%), and Atlantic Menhaden (4.0%); an additional 22 other species (10.1%) were collected. Fish abundance did not appear impacted by DO since it was above threshold level throughout shallow and bottom water habitat. The Head-of-Bay subestuaries we have sampled were primarily habitat for smaller White Perch and modified PSDs were very low.

Correlation analyses of bottom DO with temperature and C/ha in subestuaries sampled since 2003 indicated that bottom DO responded differently depending on salinity classification. Mean bottom DO in summer surveys declined with development in mesohaline subestuaries, reaching average levels below 3.0 mg/L when development was beyond its threshold; occupation of bottom channel habitat diminishes at or below target DO. Mean bottom DO did not decline in oligohaline or tidal-fresh subestuaries. The extent of bottom channel habitat that can be occupied does not diminish with development in tidal-fresh and oligohaline subestuaries due to low DO; however, more localized, or episodic habitat issues seem to be important. Sampling of DO in dense submerged aquatic vegetation (SAV) beds in tidal-fresh Mattawoman Creek in 2011 indicated low DO within the beds. Ammonia toxicity that was associated with high SAV coverage was suspected as a cause of boom-and-bust dynamics of trawl GMs in Mattawoman Creek during the 2000s. Harmful algal blooms (HABS) appear to be a negative habitat feature of low salinity subestuaries in the Head-of-Bay region; HABS, sometimes with fish kills, have occurred in Gunpowder (2004 and 2017), Middle River (2015), and Sassafras River (2018 and 2020), that have greatly contrasting dominant land uses (urban in Gunpowder and Middle, agricultural in Sassafras). During 2020, Sassafras River was subject to HABs throughout summer sampling, but fish kills were not detected; HABs were not observed there during 2021.

Median bottom DO in mesohaline subestuaries increased as agricultural coverage went from 3 to 39%; these watersheds were located on the western shore. Agricultural coverage was 43-72% for watersheds of eastern shore subestuaries and the DO trend appeared to be stable or slightly declining. A dome-shaped quadratic model of median bottom DO and agricultural coverage that did not account for regional differences fit the data well. Below threshold median bottom DO was predicted when agricultural coverage fell below 18%. Median bottom DO was predicted to peak at about 50% agricultural coverage and modest declines in bottom DO would occur through 72% of their watershed covered in agriculture. Predicted median bottom DO at the highest level of agriculture observed would equal 4.3 mg/L, between the DO target and threshold. Agricultural coverage and C/ha were strongly and inversely correlated, so the positive trend of DO at low agricultural coverage was likely to reflect development’s negative impact.

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Common Background for Project 1, Sections 1-3

"*It is the whole drainage basin, not just the body of water, that must be considered as the minimum ecosystem unit when it comes to man's interests.*" (Odum 1971).

Fishing has been the focus of assessments of human-induced perturbations of fish populations (Boreman 2000) and biological reference points (BRPs) have been developed to guide how many fish can be safely harvested from a stock (Sissenwine and Shepherd 1987). Managers also take action to avoid negative impacts from habitat loss and pollution that might drive a fish population to extinction (Boreman 2000) and typically control fishing to compensate for these other factors. A habitat-based corollary to the BRP approach would be to determine to what extent habitat can be degraded before adverse conditions cause habitat suitability to decline significantly or cease.

Forests and wetlands in the Chesapeake Bay watershed have been converted to agriculture and residential areas to accommodate increased human populations since colonial times (Brush 2009). These watershed alterations have affected major ecological processes and have been most visibly manifested in Chesapeake Bay eutrophication, hypoxia, and anoxia (Hagy et al. 2004; Kemp et al. 2005; Fisher et al. 2006; Brush 2009). Human population growth since the 1950s added a suburban landscape layer to the Chesapeake Bay watershed (Brush 2009) that has been identified as a threat (Chesapeake Bay Program or CBP 1999). Land in agriculture has been relatively stable, but fertilizer and pesticide use became much more intensive (use had increased) in order to support population growth (Fisher et al. 2006; Brush 2009). Management of farming practices has become more intense in recent decades in response to eutrophication (Kemp et al. 2005; Fisher et al. 2006; Brush 2009). Through previous research under F-63, we have identified many negative consequences of watershed development on Bay habitat of sportfish and have used this information in attempts to influence planning and zoning (Interagency Mattawoman Ecosystem Management Task Force 2012) and fisheries management (Uphoff et al. 2011). We have less understanding of the consequences of agriculture on sportfish habitat and have redirected some effort towards understanding impacts of this land use on sportfish habitat.

Project 1 investigates two general alternative hypotheses relating recreationally important species to development and agriculture. The first hypothesis is that there is a level of a particular land-use that does not significantly alter habitat suitability and the second is that there is a threshold level of land-use that significantly reduces habitat suitability (production from this habitat diminishes). The null hypothesis would be an absence of differences. In general, we expect habitat deterioration to manifest itself as reduced survival of sensitive live stages (usually eggs or larvae) or limitations on use of habitat for spawning or growth (eggs-adults). In either case, we would expect that stress from habitat would be reflected by dynamics of critical life stages (abundance, survival, growth, condition, etc.).

Development associated with increased population growth 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) that have ecological, economic, and societal consequences (Szaro et al. 1999). Ecological stress from development of the Bay watershed conflicts with demand for fish production and recreational fishing opportunities from its estuary (Uphoff et al. 2011; Uphoff et al 2015). 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; Branco et al. 2016).

Impervious surface is used as an indicator of development because of compelling scientific evidence of its effect in freshwater systems (Wheeler et al. 2005; NRC 2009) and because it is a critical input variable in many water quality and quantity models (Arnold and Gibbons 1996; Cappiella and Brown 2001). Impervious surface itself increases runoff volume and intensity in streams, leading to increased physical instability, erosion, sedimentation, thermal pollution, contaminant loads, and nutrients (Beach 2002; Wheeler et al. 2005; NRC 2009: Hughes et al. 2014 a and b). Urbanization may introduce additional industrial wastes, contaminants, stormwater runoff and road salt (Brown 2000; NRC 2009; Benejam et al. 2010; McBryan et al. 2013; Branco et al. 2016; Kaushal et al. 2018; Baker et al. 2019) that act as ecological stressors and are indexed by impervious surface. The NRC (2009) estimated that urban stormwater is the primary source of impairment in 13% of assessed rivers, 18% of lakes, and 32% of estuaries in the U.S., while urban land cover only accounts for 3% of the U.S. land mass.

Measurable adverse changes in physical and chemical characteristics and living resources of estuarine systems have occurred at IS of 10-30% (Mallin et al. 2000; Holland et al. 2004; Uphoff et al. 2011; Seitz et al. 2018; Uphoff et al. 2022). Habitat reference points based on IS have been developed (ISRPs) for Chesapeake Bay estuarine watersheds (Uphoff et al. 2011). They provide a quantitative basis for managing fisheries in increasingly urbanizing Chesapeake Bay watersheds and enhance communication of limits of fisheries resources to withstand development-related habitat changes to fishers, land-use planners, watershed-based advocacy groups, developers, and elected officials (Uphoff et al. 2011; Interagency Mattawoman Ecosystem Management Task Force 2012). These guidelines have held for Herring stream spawning, Yellow Perch larval habitat (they are incorporated into the current Maryland's tidal Yellow Perch management plan; MD DNR 2017), and summer habitat in tidal-fresh subestuaries (Uphoff et al. 2015). Conserving watersheds at or below 5% IS would be a viable fisheries management strategy. Increasingly stringent fishery regulation might compensate for habitat stress as IS increases from 5 to 10%. Above a 10% IS threshold, habitat stress mounts and successful management by harvest adjustments alone becomes unlikely (Uphoff et al. 2011; Interagency Mattawoman Ecosystem Management Task Force 2012; Uphoff et al. 2022). A preliminary estimate of IS in Maryland's portion of the Chesapeake Bay watershed in 2018 equaled 9.3%. We expect adverse habitat conditions for important forage and gamefish to worsen with future growth. Managing this growth with an eye towards conserving fish habitat is important to the future of sportfishing in Maryland.

We now consider tax map derived development indices as the best source for standardized, readily updated, and accessible watershed development indicators in Maryland and have development targets and thresholds based on it that are the same as ISRPs (Topolski 2015; Uphoff et al. 2020; see **General Spatial and Analytical Methods used in Project 1, Sections 1-3**). Counts of structures per hectare (C/ha) had strong relationships with IS (Uphoff et al. 2020). Tax map data can be used as the basis for estimating target and threshold levels of development in Maryland and these estimates can be converted to IS. 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 0.37, 0.86, and 1.35 C/ha, respectively. Tax map data provide a development time-series that goes back to 1950, making retrospective analyses possible (Uphoff et al. 2020). Development in Maryland's portion of the Chesapeake Bay watershed, approximately 0.17 C/ha in 1950, reached 0.75 C/ha in 2018. The target level of C/ha is 0.37 and the threshold is 0.86 (Uphoff et al. 2022).

The area of major spawning tributaries used by Striped Bass, White Perch, Yellow Perch, Alewife, Blueback Herring, Hickory Shad, and American Shad are typically on the receiving end of large amounts of agricultural drainage because of their location at the junction of large fluvial systems and brackish estuaries. Trends in juvenile indices of these species are similar, indicating similar influences on year-class success (Uphoff 2008).

Agricultural pesticides and fertilizers were thought to be potential sources of toxic metals implicated in some episodic mortality of Striped Bass larvae in Bay spawning tributaries in the early 1980s (Uphoff 1989; 1992; Richards and Rago 1999; Uphoff 2008; Uphoff et al. 2020). A correlation analysis of Choptank River watershed agricultural best management practices (BMPs) and estimates of postlarval survival during 1980-1990 indicated that as many as four BMPs were positively associated with survival (Uphoff 2008). Two measures that accounted for the greatest acreage, conservation tillage and cover crops, were strongly associated with increased postlarval survival (r = 0.88 and r = 0.80, respectively). These correlations cannot explain whether toxicity was lowered by BMPs, but it is possible that reduced contaminant runoff was a positive byproduct of agricultural BMPs aimed at reducing nutrients (Uphoff 2008).

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). Hypoxia is also associated with suburban landscapes in mesohaline Chesapeake Bay subestuaries (Uphoff et al. 2011; Uphoff et al 2022). Hypoxia's greatest impact on gamefish habitat occurs during summer when its extent is greatest, but hypoxic conditions are present at lesser levels during spring and fall (Hagy et al. 2004; Costantini et al. 2008). Episodic hypoxia may elevate catch rates in various types of fishing gears by concentrating fish at the edges of normoxic waters, masking associations of landings and hypoxia (Kraus et al. 2015).

Habitat loss due to hypoxia in coastal waters is often associated with fish avoiding DO that reduces growth and requires greater energy expenditures, as well as lethal conditions (Breitburg 2002; Eby and Crowder 2002; Bell and Eggleston 2005). There is evidence of cascading effects of low DO on demersal fish production in marine coastal systems through loss of invertebrate populations on the seafloor (Breitburg et al. 2002; Baird et al. 2004). A long-term decline in an important Chesapeake Bay pelagic forage fish, Bay Anchovy, may be linked to declining abundance of the common calanoid copepod *Acartia tonsa* in Maryland's portion of Chesapeake Bay that, in turn, may be linked to rising long-term water temperatures and eutrophication that drive hypoxia (Kimmel et al. 2012; Roman et al. 2019; Slater et al. 2020). Crowding in nearshore habitat, if accompanied by decreased growth due to competition, could lead to later losses through size-based processes such as predation and starvation (Breitburg 2002; Eby and Crowder 2002; Bell and Eggleston 2005). Exposure to low DO appears to impede immune suppression in fish and Blue Crabs, leading to outbreaks of lesions, infections, and disease (Haeseker et al. 1996; Engel and Thayer 1998; Breitburg 2002; Evans et al. 2003). Exposure of adult Carp to hypoxia depressed reproductive processes such as gametogenesis, gonad maturation, gonad size, gamete quality, egg fertilization and hatching, and larval survival through endocrine disruption even though they were allowed to spawn under normoxic conditions (Wu et al. 2003). Endocrine disruption due to hypoxia that could reduce population spawning potential has been detected in laboratory and field studies of Atlantic Croaker in the Gulf of Mexico (Thomas and Rahman 2011) and Chesapeake Bay (Tuckey and Fabrizio 2016).

A hypoxia based hypothesis, originally formed to explain die-offs of large adult Striped Bass in southeastern reservoirs, links increased natural mortality and deteriorating condition in Chesapeake Bay through a temperature-oxygen squeeze (mismatch of water column regions of desirable temperature and dissolved oxygen in stratified Chesapeake Bay during summer; Coutant 1985; Price et al. 1985; Coutant 1990; Coutant 2013). Constantini et al. (2008), Kraus et al. (2015), and Itakura et al. (2021) examined the impact of hypoxia on 2 year-old and older Striped Bass in Chesapeake Bay through bioenergetics modeling and acoustic tagging and concluded that a temperature-oxygen squeeze by itself was not limiting for Striped Bass. However, Groner et al (2018) suggested that Striped Bass are living at their maximum thermal tolerance and that this is driving increased mycobacteriosis and associated mortality. Adequate levels of Striped Bass prey can offset negative effects of warm temperatures and suboptimal dissolved oxygen in reservoirs (Thompson et al. 2010; Coutant 2013).

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MD – Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern
General Spatial and Analytical Methods used in Project 1, Sections 1-3

Spatial Methods - 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 was estimated for 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 MD DOP 2019). 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 MD DOP's GIS database. Files were managed and geoprocessed in ArcGIS 10.3.1 from Environmental Systems Research Institute (ESRI 2015). 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, although there is typically a two-year lag in processing by MD DOP. 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.

During 2003-2010, we used impervious and watershed area estimates made by Towson University from Landsat, 30-meter pixel resolution satellite imagery (eastern shore of Chesapeake Bay in 1999 and western shore in 2001) as our measure of development for each watershed (Barnes et al. 2002). They became outdated and C/ha provided a readily updated substitute. Uphoff et al. (2012) developed an nonlinear power function to convert annual estimates of C/ha during 1999-2000 for watersheds sampled during 2003-2009 (Table 1) to estimates of percent impervious surface (IS) calculated by Towson University from 1999-2000 satellite imagery. This equation was used to convert each year's C/ha estimates to IS.

Recalculation of this conversion equation was necessary in 2018 due to a new time-series provided by MD DOP, as well as inconsistencies found in the data for some watersheds up to 2002 (M. Topolski, MD DNR, personal communication). Historic data were recalculated using 2002 MdProperty View data (previously 1999 data had been used) which corrected data deficiencies in the 2000 and 2001 data, as well as errors in the 1999 data (Table 1; M. Topolski, MD DNR, personal communication). The same watersheds and years used to estimate the original nonlinear relationship (Uphoff et al. 2012) were used in the update to maintain continuity.

A linear regression described the updated relationship well:

$$IS = (10.129 \cdot C/ha) + 1.286; (r^2 = 0.905; P < 0.0001; \text{Figure 1}).$$

Revised 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 0.37, 0.86, and 1.35 C/ha, respectively. The previous C/ha estimates, based on a nonlinear power function, corresponding to 5%, 10%, and 15% IS were 0.27, 0.83, and 1.59, respectively (Uphoff et al. 2018).

Percent of watershed in agriculture, forest, and wetlands were estimated from MD DOP spatial data. The MD DOP forest cover estimates have a minimum mapping unit of 10 acres that mixes forest cover in residential areas (trees over lawns) with true forest cover, clouding interpretation of forest influence (R. Feldt, MD DNR Forest Service, personal communication). An urban category was available as well, but was not featured in many subsequent analyses since we have adopted C/ha as our preferred index of development. Urban land consisted of high and low density residential, commercial, and institutional acreages and was not a direct measure of IS.

Land use and land cover (LULC) shapefiles were available for 1973, 1994, 1997, 2002, and 2010 for each Maryland jurisdiction and as an aggregated statewide file. Metadata for the LULC categories is available for download from MD DOP. The statewide LULC shapefiles were clipped using boundary shapefiles for each watershed of interest. Once clipped, polygon geometry was recalculated. Polygons designated as water were omitted when calculating watershed area; that is only land was considered when calculating the ratio of LULC for each category. For each LULC category, polygons were queried and its land area in hectares was calculated. The land use total was divided by the watershed total to the nearest tenth of a hectare and multiplied by 100%.

Updating Impervious Surface Coverage with High Resolution Land Cover - Recent improvements to spatial resolution of land cover (from 30 x 30 m that we currently used) to 1 x 1 m provided by the Chesapeake Conservancy's Conservation Innovation Center affords the opportunity to recalibrate the relationship between tax data and %IS. Recalibration of this relationship is particularly relevant as these high-resolution land cover data become the authoritative source of current and future on-the-ground conditions. For example, Chesapeake Conservancy's Conservation Innovation Center has contracted with the Chesapeake Bay Program to produce additional high resolution land cover datasets for the years 2017/2018 and 2021/2022 (Walker et al. 2022). Appendix 1 describes analyses that Marek Topolski conducted for F-63 that determined the best approach for converting C/ha to IS based on these new data. The best method described in Appendix 1 will replace what is used now in future reports.

Statistical Analyses – A combination of correlation analysis, plotting of data, and curve-fitting was commonly used to explore trends among land use types (land that was developed or in agriculture, forest, or wetland) and among fish habitat responses. Typical fish habitat responses were the proportion of stream samples with Herring eggs and-or larvae (P_{herr}; Section 1); proportion of subestuary samples with Yellow Perch larvae (L_p; Section 2); or subestuary bottom dissolved oxygen, fish presence-absence or relative abundance, and fish diversity in summer (Section 3).

Correlations among watershed estimates of C/ha and percent of watershed estimated in urban, agriculture, forest, and wetland based on MD DOP spatial data were used to describe associations among land cover types. These analyses explored (1) whether C/ha estimates were correlated with another indicator of development, percent urban and (2) general associations among major landscape features in our study watersheds. Scatter plots were inspected to examine whether nonlinear associations were possible. Land use was assigned from MD DOP estimates for 1973, 1994, 1997, 2002, or 2010 that fell closest to a sampling year. We were particularly interested in knowing whether these land uses might be closely correlated enough (r greater than 0.80; Ricker 1975) that only one should be considered in analyses of land use and L_p and P_{herr}. We further examined relationships using descriptive models as a standard of comparison (Pielou 1981). Once the initial associations and scatter plots were examined, linear or nonlinear regression analyses (power, logistic, or Weibull functions) were used to determine the general shape of trends among land use types. This same strategy was pursued for analyses of land use and L_p or P_{herr}. Level of significance was reported, but potential management and biological significance took precedence over significance at $P < 0.05$ (Anderson et al. 2000; Smith 2020). We classified correlations as strong, based on $r \geq 0.80$; weak correlations were indicated by $r < 0.50$; and moderate correlations fell in between. Relationships indicated by regressions were considered strong at $r^2 \geq 0.64$; weak relationships were indicated by $r^2 \leq 0.25$; and moderate relationships fell in between. Confidence intervals (95% CIs were standard output) of the model parameters for each indicator species were estimated to examine whether parameters were different from 0 (Freund and Littell 2006). If parameter estimates were often not different from 0, rejection of the model was considered. Residuals of regressions were inspected for trends, non-normality, and need for additional terms. A general description of equations used follows, while more specific applications will be described in later sections.

Linear regressions described continuous change in variable Y as X changed:

$$Y = (m \cdot X) + b;$$

where m is the slope and b is the Y-intercept (Freund and Littel 2006). Multiple regression models accommodated an additional variable (Z):

$$Y = (m \cdot X) + (n \cdot Z) + b;$$

where n is the slope for variable Z and other parameters are as described previously (Freund and Littel 2006). We did not consider multiple regression models with more than two variables. Potential dome-shaped relationships were examined with quadratic models (Freund and Littell 2006):

$$Y = (m \cdot X) + (n \cdot X^2) + b.$$

The linear regression function in Excel or Proc REG in SAS (Freund and Littel 2006) was used for single variable linear regressions. Multiple linear and quadratic regressions were analyzed with Proc REG in SAS (Freund and Littell 2006).

Examination of scatter plots suggested that some relationships could be nonlinear, with the Y-axis variable increasing at a decreasing rate with the X-axis variable and we fit power, logistic growth, or Weibull functions to these data using Proc NLIN in SAS (Gauss-Newton algorithm). The power function described a relationship with a perceptible, but declining increase in Y with X by the equation:

$$Y = a \bullet (X)^b;$$

where a is a scaling coefficient and b is a shape parameter. The symmetric logistic growth function described growth to an asymptote through the equation:

$$Y = b / ((1 + ((b – c) / c) \bullet (\exp (-a \bullet X)));$$

where a is the growth rate of Y with X, b is maximum Y, and c is Y at X = 0 (Prager et al. 1989). The Weibull function is a sigmoid curve that provides a depiction of asymmetric ecological relationships (Pielou 1981). A Weibull curve described the increase in Y as an asymmetric, ascending, asymptotic function of X:

$$Y = K \bullet \{1 - \exp [-(X / S)^b]\};$$

where K was the asymptotic value of Y as X approached infinity; S was a scale factor equal to the value of Y where Y = 0.63 • K; and b was a shape factor (Pielou 1981; Prager et al. 1989).

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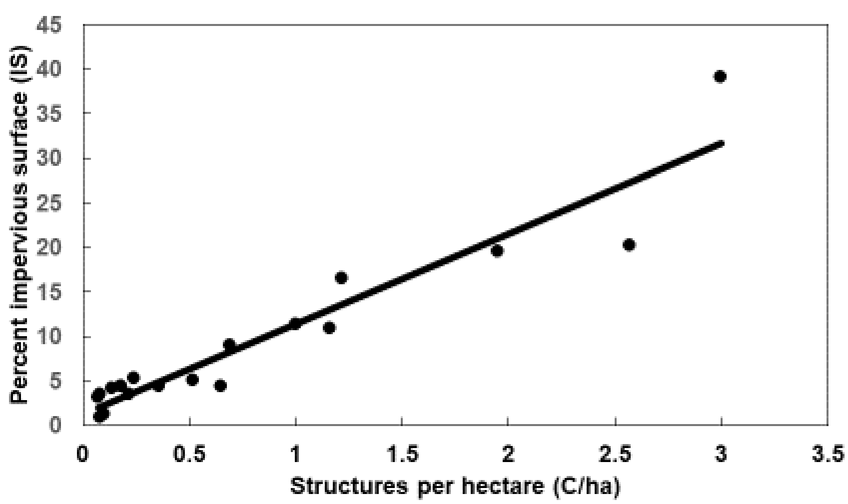
Table 1. Structures per hectare (C/ha) and percent impervious surface estimates (IS) used to estimate the relationship for predicting IS from C/ha. Old C/ha were estimates used previous to this report and New C/ha were revised estimates used to estimate the current relationship.

Watershed	Old C/ha	New C/ha	IS
Nanjemoy Creek	0.08	0.08	0.9
Bohemia River	0.10	0.10	1.2
Langford Creek	0.07	0.07	3.1
Wye River	0.08	0.08	3.4
Miles River	0.23	0.22	3.4
Corsica River	0.14	0.14	4.1
Wicomico River west	0.29	0.18	4.3
Northeast River	0.36	0.36	4.4
Gunpowder River	0.03	0.65	4.4
St Clements Bay	0.19	0.18	4.4
West River Rhode River	0.55	0.52	5.0
Breton Bay	0.25	0.24	5.3
Mattawoman Creek	0.71	0.69	9.0
South River	1.23	1.16	10.9
Bush River	0.98	1.00	11.3
Piscataway Creek	1.34	1.22	16.5
Severn River	2.14	1.95	19.5
Magothy River	3.01	2.57	20.2
Middle River	7.39	3.00	39.1

Figure 1. Relationship of structures per hectare (C/ha) and percent impervious surface (IS).



Project 1, Appendix 1
Estimating Impervious Surface Coverage from High Resolution Land Cover Data and Property Tax Information
Marek Topolski

Introduction

Property tax map-based counts of structures in a watershed, standardized to hectares (C/ha), is used as our main indicator of development (Uphoff et al. 2012, 2022; Topolski 2015; see General Spatial and Analytical Methods used in Project 1, Sections 1-3). Tax maps are graphic representations of individual property boundaries and existing structures that help State tax assessors locate properties (Maryland Department of Planning or MD DOP 2019). Estuarine watershed percent impervious surface (%IS) predicted from (C/ha) was first presented in Uphoff et al. (2010) as a nonlinear power curve and estimation was recalibrated in 2018 as a linear model following revisions of tax map data used to estimate C/ha (Uphoff et al. 2019). The reference impervious surface dataset used for the tax index was developed by Towson University for Maryland 12-digit subwatersheds from Landsat Thematic Mapper (TM) imagery from the year 2000 (see Project 1 General Spatial and Analytical Methods used in Project 1, Sections 1-3). Spatial resolution of the Landsat TM imagery was 30 m x 30 m and appropriate for regional planning (Vogelman et al. 1998). Therefore, regional scale land cover data were used to make subwatershed (i.e., subregional) %IS estimates that were then compared to spatially explicit local tax data.

The requirements of the Environmental Protection Agency's Chesapeake Bay total maximum daily load (TMDL; Executive Order 13508 2009) will require precision planning not possible using the coarse resolution of Landsat TM data. Chesapeake Conservancy's Conservation Innovation Center was contracted by the Chesapeake Bay Program to develop high-resolution, 1 m x 1 m, land cover data for the Chesapeake Bay watershed (2020). The land cover raster created was a composite of LiDAR, imagery, and land cover data having varied spatial resolutions for the years 2013/2014. The composite raster improved spatial representation and delineation of existing impervious surface boundaries compared to datasets, such as the National Landcover Dataset (NLCD), derived solely from Landsat TM imagery (Figure 1) thereby increasing accuracy of watershed %IS estimates. Direct comparison of watershed %IS estimates cannot be made between the Towson University and Chesapeake Conservancy datasets since they are based on different years; however, within year comparisons of %IS predicted from the tax index and %IS developed from each land cover dataset can be compared.

Percent impervious surface is a convenient measure of watershed development that encapsulates a variety of stressors (Chithra et al. 2015) and is an indicator of fish habitat condition (Booth et al. 2002; Stranko et al. 2008; Uphoff et al. 2011). Effects on sensitive fish species such as brook trout are evident when impervious surface is as low as 1 % and their absence occurs at 2-4 %IS (Boward et al. 1999; Stranko et al. 2008). Fish reproductive success has been linked to habitat condition, where increasing %IS corresponds to diminishing egg viability and larval survival (Uphoff et al. 2009). Uphoff et al. (2011) proposed a 5.5 %IS target as a fishery management strategy that could be used in concert with harvest management and reintroduction actions. Furthermore, they cautioned that at 10 %IS comprehensive watershed management measures or acceptance of diminished fishery productivity will be necessary. Maryland Department of Natural Resources (MD DNR) developed management guidelines for aquatic condition across a range of watershed development (C/ha) and equivalent %IS levels for consideration during the comprehensive [land] planning process (The Interagency Mattawoman Ecosystem Management Task Force 2012) and inclusion in fishery management plan development (MD DNR 2017). To further simplify resource management, reference points that delineate target (5 %IS), threshold (10 %IS), and impaired (15 %IS) watershed condition and corresponding C/ha have been recommended in this report (see Project 1 Section 1-3).

Recent improvements to spatial resolution of land cover provides the opportunity to recalibrate the relationship between tax data and %IS. Recalibration of this relationship is particularly relevant as these high-resolution land cover data become the authoritative source of current and future on-the-ground conditions. For example, Chesapeake Conservancy's Conservation Innovation Center has contracted with the Chesapeake Bay Program to produce additional high resolution land cover datasets for the years 2017/2018 and 2021/2022 (Walker et al. 2022). Several questions should be addressed during this revision to the tax index. First, does the current suite of estuarine watersheds [used previously to calibrate %IS with C/ha] have a sufficient range of rural to urban development to predict %IS? Second, what model best explains the relationship between C/ha and %IS estimates from the Chesapeake Conservancy 2013/2014 data? Third, do the existing tax index and 2013/2014 Chesapeake Conservancy index produce comparable estimates of %IS? From these analyses, the goal is a tax index model to estimate %IS (based on Chesapeake Conservancy estimates) from C/ha for both current watershed development scenarios and for retrospective analyses (time series dating back to 1950) of %IS change within watersheds.

Methods

A suite of 33 estuarine watersheds were selected for the analyses (Figure 2). Twenty-nine watersheds have been periodically surveyed by MD DNR fish habitat programs; 19 of these had been used to calculate tax-based indices of %IS. Fourteen additional watersheds were included to ensure a broad range of land development intensity and spatial distribution. Of these 14, four non-surveyed watersheds were included: two because of their high level of development (Baltimore Harbor and Bird River), one freshwater tributary of the Susquehanna River (Broad Creek), and one in higher salinity water of Tangier Sound near the Maryland/Virginia border (Big Annemessex River). Watershed boundaries were delineated from the Maryland 12-digit subwatershed polygon shapefile, available from the MD iMAP data portal (https://data.imap.maryland.gov/), by their Maryland 8-digit code (MDE8NAME). The spatial extent of ten watersheds (Baltimore Harbor, Blackwater River, Broad Creek: Lower Choptank, Bush River, Elk River, Gunpowder River, Harris Creek, Tred Avon River, Wicomico River: Eastern Shore, and Wicomico River: Western Shore) varied from a single 8-digit delineation and so the watershed boundaries were adjusted by the inclusion or exclusion of Maryland 12-digit subwatershed codes (DNR12DIG; Table 1; Figure 2). For example, Baltimore Harbor was upstream of Colgate Creek and excluded headwaters except Gwynns Falls and Jones Falls, Aberdeen Proving Grounds (APG) at the mouth of Bush River was excluded from the watershed along with Swan Creek, Elk River was limited to the watershed upstream of the C&D Canal, and Gunpowder River excluded the Middle River watershed. Maryland 8-digit watersheds are comparable to U.S. Geological Survey (USGS) Hydrologic Unit Code (HUC) 8 watersheds and Maryland 12-digit subwatersheds are comparable to USGS HUC 12 subwatersheds (M. Topolski, MD DNR, personal observation).

Two raster-based datasets of land cover were used for this study; one dataset was prepared by Towson University and the other by the Chesapeake Conservancy's Conservation

Innovation Center. Percent impervious surface estimates derived by Towson University were for the year 2000 from 30 m x 30 m Landsat TM imagery and used for the initial and revised tax indices previously discussed. Pixels were classified as deciduous forest, evergreen forest, herbaceous agriculture, herbaceous urban, impervious high, impervious low, bare ground, and open water then summarized by classification for each 12-digit subwatershed. Pixels classified as impervious high were estimated to be 90 %IS and pixels classified as impervious low were estimated to be 50 %IS. Watershed %IS was calculated as the number of impervious acres divided by the number of land acres multiplied by 100. These estimates of land cover and %IS per 12-digit subwatershed were then provided to MD DNR in table form. The 12-digit subwatershed data was imported into RStudio software package and compiled as the 12-digit land area weighted average %IS for each of the estuarine watersheds previously described (Table 1).

Chesapeake Conservancy 2013/2014 land cover estimates were available for download as a 1 m x 1 m resolution raster for the entire Chesapeake By watershed or for each state's portion of the Chesapeake Bay watershed (https://www.chesapeakeconservancy.org/conservation-innovation-center/high-resolution-data/land-cover-data-project/). The Chesapeake Conservancy 2013 raster was derived from multiple datasets including Light Detection and Ranging (LiDAR) elevation data, USGS National Agriculture Imagery Program (NAIP) imagery, orthoimagery (where available), county planimetrics, statewide and federal road datasets, and National Wetlands Inventory (NWI) polygons (Chesapeake Conservancy 2020). The raster included several pixel classifications of impervious surfaces (structures, impervious surfaces, impervious roads, tree canopy over structures, tree canopy over impervious surfaces, and tree canopy over impervious roads) that allowed for calculation of %IS. The Maryland statewide land cover raster was imported into ArcGIS Pro software, reprojected to the NAD 1983 StatePlane Maryland FIPS 1900 (meters) coordinate system, and pixels were reclassified as impervious, pervious, or water. Total impervious, pervious, and water surfaces per 12-digit subwatershed were calculated using the Zonal Statistics as Table tool where each 12-digit subwatershed was a distinct zone. Since pixels were 1 m x 1 m, total count was equivalent to the total area in m^2 for each pixel classification per zone. Tables were imported into the RStudio software package, joined by 12-digit subwatershed, %IS (excluding water) per 12-digit subwatershed was calculated, then compiled as the 12-digit land area weighted average %IS for each estuarine watershed previously described (Table 1).

Tax data are made available by jurisdiction as point shapefiles from the Maryland Department of Planning as part of the MdProperty View datasets (https://planning.maryland.gov/Pages/OurProducts/DownloadFiles.aspx). Tax datasets for the years 2000 and 2013 were selected to correspond with the Towson University 2000 and Chesapeake Conservancy 2013/2014 datasets, respectively. ArcGIS Pro was used to query each of the 24 jurisdictions' tax data for primary structures built between the years 1700-2000 and 1700-2013. For each of these jurisdiction datasets, structures were spatially joined to their corresponding 12-digit subwatershed. These 24 shapefiles were compiled into a single statewide point shapefile and the sum of number of structures per 12-digit subwatershed was exported as a table. Total number of structures per 12-digit subwatershed data was then imported into the RStudio software package, summed by watershed (previously described), divided by watershed land hectares to convert to C/ha, then joined with watershed %IS estimates (previously described).

Structures per hectare for the 2000 and 2013 tax data were based on different estimates of land area for each 12-digit subwatershed. For 2000 tax data, shapefiles (vector) were used to subtract the area classified as water, delineated using a shoreline shapefile for the year 1990 (published in 2003) and a lakes/reservoirs (≥0.4 ha or 1 acre) shapefile (published in 2005), from the total 12-digit subwatershed polygon area. These vector-based estimates of land area were used in conjunction with 2000 tax data to calculate C/ha. The shoreline and lakes/reservoirs shapefiles were downloaded from the MD iMAP data portal (https://data.imap.maryland.gov/). For 2013 tax data, land area was calculated by subtracting the water area per 12-digit subwatershed derived using the Chesapeake Conservancy 2013/2014 raster data from the corresponding 12-digit subwatershed's total polygon area. Percent impervious surface estimates for each raster dataset also excluded pixels classified as water.

Both nonlinear (derived in 2010) and linear (2018 revision) models have been used to estimate %IS based on the relationship between Towson University %IS estimates and tax data (C/ha). The 2018 model revision, published in 2019 (TU2019), in response to revised 2000 C/ha data. Since Chesapeake Conservancy land cover data of significantly higher resolution was available, model form (linear versus nonlinear) was revisited. The initial tax index from 2010 was based on a power model ($\%IS = a * (C/ha)^b$) where the amount of change in %IS decreased as C/ha increased, "a" was a scaling coefficient, and "b" was a shape parameter: $\%IS = 10.98 * (C/ha)^{0.63}$ (Uphoff et al. 2012). The revised tax index (TU2019) used a linear model to describe the change in %IS as C/ha increased: $\%IS = 1.286 + (C/ha) * 10.129$ (Uphoff et al. 2019). Linear and power models were developed using the %IS estimates from the Chesapeake Conservancy 2013/2014 data and the corresponding 2013 tax data for the original 19 watersheds and the expanded set of 33 watersheds. Models were evaluated and compared based on standardized residual plots, minimization of residual standard error (RSE), coefficient of determination (R^2), overlap of 90% confidence intervals, and corrected Akaike Information Criterion (AICc). Nonrandom patterning in the plot of residuals versus fitted values is an indicator of incorrect model specification(Forthofer et al. 2007; Meloun and Militký 2011; Altman and Krzywinski 2016). Calculation of R^2 is not appropriate for nonlinear models (Kvålseth 1983; Spiess and Neumeyer 2010), and so linear regressions of power model predicted %IS and observed %IS were used to approximate R^2 for the two power models. Lack of confidence interval overlap is indicative of smaller t-test p values and significant differences, at the specified α criterion, in model estimates (Cumming and Finch 2005; Cornell Statistical Consulting Unit 2020). Corrected Akaike information criteria (AICc), which adjusts for small sample size, is a measure of a model's likelihood of being the best choice considering the data and parameters considered (Burnham and Anderson 2001). Models with a lower AICc score are a better choice; the difference between a model's AICc score and the minimum AICc (Δ_i where i is an individual model) score provides a "strength of evidence" ranking of the models. Values of $\Delta_i < 2$ have substantial support, while $\Delta_i > 10$ have essentially no support (Burnham and Anderson 2001). Evidence ratios, which indicate the comparative probability of one model being better than the other (Motulsky and Christopoulos 2003), were also calculated.

Accuracy of the TU2019 linear model was evaluated through comparison with the Chesapeake Conservancy 2013/2014 data and models. Percent impervious surface for each watershed was estimated for the year 2013 using the TU2019 model and 2013 tax data (C/ha). Analysis of covariance (ANCOVA) was conducted to compare the preferred linear model with the TU2019 model for differences in slope and y-intercept. Significant difference in regression slopes is an indication that the two models describe different rates of change in %IS as C/ha increases; whereas a significant difference in y-intercept indicates that the magnitude of %IS is different for a given value of C/ha. Confidence intervals for the preferred linear and nonlinear Chesapeake Conservancy 2013/2014 models were compared with the TU2019 model's confidence interval to estimate C/ha values where %IS estimates become significantly different. Data were developed using ArcGIS Pro and all analyses were done using RStudio.

Results

For the year 2000, land area hectare estimates based on the vector shapefiles were always greater than from the 30-meter resolution raster (Table 1). Land area decreased in all watersheds from the 2000 vector estimate to the 2013/2014 1-meter raster estimate; land area was consistently underestimated by the 2000 30-meter raster in all but three watersheds (Blackwater River, Fishing Bay, and Transquaking River). Watershed land area derived from shapefiles ranged from 2,753 to 113,637 ha (mean=22,937 ha and median=16,342 ha); for the original 19 watersheds the range in land area did not differ although the mean (22,364 ha) and median (14,773 ha) land area were lower. Estimated watershed land area from 30-meter resolution raster data was 2,221 to 109,665 ha (mean=21,261 and median=15,905); while the range did not differ for the original 19 watersheds the mean (21,329 ha) was greater and median (13,769 ha) was lower (Table 1). Watershed land area based on 1-meter resolution raster data ranged from 2,716 to 113,435 ha (mean=22,030 ha and median=16,221 ha); among the original 19 watersheds the land area range did not differ but the mean (22,257 ha) was greater and the median (14,675 ha) was lower (Table 1).

Structures per hectare in 2000 ranged from a low of 0.027 C/ha in Transquaking River to a high of 5.298 C/ha in Baltimore Harbor (mean = 0.758 C/ha and median = 0.282 C/ha among all watersheds); the original 19 watersheds ranged from 0.065 C/ha in Langford Creek to 3.002 C/ha in Middle River (mean=0.775 C/ha and median=0.358 C/ha among the original watersheds; Table 1). All watersheds increased in C/ha from 2000 to 2013. Transquaking River had the lowest C/ha at 0.030 and Baltimore Harbor remained the most developed with C/ha of 6.180 (mean=0.892 C/ha and median=0.378 C/ha among all watersheds; Table 1) in 2013. For the original 19 watersheds in 2013, the lowest C/ha was 0.075 in Langford Creek and 3.381 C/ha in Middle River (mean=0.905 C/ha and median=0.477 C/ha among the original watersheds; Table 1).

Watershed %IS in 2000, derived from Towson University 12-digit subwatershed estimates, ranged from 0.94% in Nanjemoy Creek to 46.39% in Baltimore Harbor (mean=8.65% and median=4.95%); for the original 19 watersheds %IS estimates varied from 0.94% in Nanjemoy Creek to 39.11% in Middle River (mean=9.01% and median=4.95%; Table 1). High resolution %IS estimates for 2013 were lower than the 2000 estimates in 12 of the 33 watersheds. Blackwater River and Fishing Bay had a low of 1.21 %IS while Baltimore Harbor had a high of 38.68 %IS (mean=8.90% and median=4.90%); among the original 19 watersheds Nanjemoy Creek had a low of 1.93 %IS and Middle River had a high of 30.39 %IS (mean=9.29% and median=6.58%; Table 1).

Percent impervious surface was estimated for each watershed in 2013 using the four models developed. Estimates were generally comparable to observed %IS calcualted from the Chesapeake Conservancy 2013/2014 data (Table 2). On average, the absolute difference between modeled %IS and observed %IS was 0.82 -1.16 percentage points. Among the models, maximum absolute difference between a watershed's modeled and observed %IS varied from 4.06-11.27 percentage points (Table 2).

Linear models were significant for the original ($P<0.0001$, $R^2=0.978$) and expanded ($P<0.0001$, $R^2=0.956$) sets of watersheds (Table 3). Both the original and expanded watershed models' y-intercepts were significantly greater than zero ($P<0.0001$; Table 3) and over-estimated %IS when C/ha was less than about 0.250 (Figure 3). Standardized residuals for both linear models increased from 0 %IS to about 5 %IS after which no trend was evident although the expanded watershed standardized residuals remained above zero (Figure 3). The linear model for original watersheds had a lower RSE (1.195) than the expanded watersheds model (RSE=1.901; Table 3).

Both the original and expanded power models along with the scaling coefficient and shape parameters were significant at $P<0.0001$ (Table 3). Approximate R^2 was comparable between to the models: 0.972 for the original watersheds power model and 0.982 for the expanded watersheds power model (Table 3). Power model residuals were initally greater than zero, decreased from positive to negative value from about 4 %IS to about 6 %IS (original) but were randomly distributed from about 3 %IS to about 10 %IS (expanded), and then remained negative until about 20 %IS when the standardized residuals became randomly distributed around zero (Figure 3). Residual standard error was 1.343 for the original watersheds power model and decreased to 1.210 for the expanded watersheds power model.

Ninety percent confidence intervals for the original watershed linear and power models overlapped except when C/ha was less than about 0.10 at which point the linear model estimated higher %IS per C/ha (Figure 4). When the expanded set of watersheds was considered, the linear and power models' 90% confidence intervals deviated below about 0.15 C/ha, between about 0.65-2.60 C/ha, and above about 6.15 C/ha (Figure 4); specifically the linear model estimated higher %IS at low and high values (less than about 0.15 C/ha and greater than about 6.15 C/ha) and the %IS estimate was lower between about 0.65-2.60 C/ha (Figure 4).

For the original set of watersheds, the linear model had the lower AICc score (63.72) compared to the power model (AICc=70.21, $\Delta_i=6.49$, Table 4). However, since Δ_i was <10 and >2 the power model was a reasonable alternative. The greater number of watersheds in the expanded set that had low (<0.5) and high (>2) C/ha had considerable influence on determination of model liklihood. The power model had an AICc=111.21 which was considerably lower than the linear model with AICc=139.41 ($\Delta_i=28.20$, Table 4). While all models were significant, AICc scores indicated that the preferred linear model was based on the original watersheds and that the preferred nonlinear power model was based on the expanded set of watersheds. Confidence interval comparison between the two preferred models (original watersheds linear model and expanded watersheds power model) show significant deviation when C/ha was less than about 0.10 and about 0.65-1.55 C/ha; the linear model's confidence intervals were projected for the expanded watershed data range and they deviated at about 3.55 C/ha (Figure 4).

The two preferred models were then compared with the TU2019 model currently used for %IS estimation from C/ha. The TU2019 model's slope was significantly different from the preferred linear model's slope (ANCOVA P=0.0055) but the y-intercepts did not differ (ANCOVA P=0.3081, Table 5). Ninety percent confidence intervals diverged and no longer overlaped when C/ha was greater than about 1.20 (Figure 5). Considerable overlap of 90% confidence intervals occured between the TU2019 model and the preferred nonlinear power model (Figure 6), and they did not diverge until C/ha exceeds about 1.60. Percent impervious surface estimates generated by the FHEP have been based on the TU2019 model; tax data C/ha values of 0.070, 0.367, 0.860, and 1.354 correspond to 2, 5, 10, and 15 %IS, respectively. Relative to these %IS levels relevent to resource management (2, 5, 10, and 15), the C/ha equivalents are -0.041, 0.348, 0.997, and 1.646 from the preferred linear model (original watersheds) and 0.084, 0.312, 0.844, and 1.509 from the preferred power model (expanded watersheds), respectively (Table 6). Negative C/ha values such as those estimated by the preferred linear model when %IS ≤2 were not realistic since C/ha cannot be less than zero; therefore the nonlinear power model derived from the 2013/2014 Chesapeake Conservancy data for the expanded set of watersheds should be used for %IS estimation from C/ha.

Discussion

Residual analysis of models developed from the high-resolution Chesapeake Conservancy 2013/2014 data indicated that the C/ha to %IS relationship was nonlinear when considered across a broad range of land development, i.e. the expanded set of watersheds. A power function was a good fit to the nonlinear relationship and was attractive due to its simplicity. Expanding the set of watersheds doubled the number of very low density residential and rural watersheds (< 0.405 C/ha = < 1 C/acre; MD DOP 2018), including an additional three rural watersheds (≤ 0.081 C/ha = ≤ 0.2 C/acre), and added the high density residential Baltimore Harbor (≥ 4.047 C/ha = ≥ 10 C/acre). The increased number of watersheds resulted in the power model being the better choice given the data and the recommended model for estimating %IS from tax data. In circumstances where C/ha was at or below levels of development that would impact sensitive aquatic species (0.084 C/ha or 2 %IS) and exceeded the ecological threshold of 10 %IS (0.844 C/ha), the power models tend to slightly underpredict and overpredict %IS, respectively. These under and over predictions are outside of the target (5 %IS) and threshold (10 %IS) benchmarks previously identified for resource management.

The inability to detect the nonlinear C/ha to %IS relationship with the Towson University 2000 data may reflect the lack of spatial resolution in the %IS estimates. When C/ha was less than equal about one, the TU2019 model was a good fit with the estimated Chesapeake Conservancy 2013/2014 %IS and was a close approximation to the expanded watershed power model. Existing TU2019 C/ha reference points for target (5 %IS), threshold (10 %IS), and impairment (15 %IS) levels of development, compared to the preferred nonlinear model, were 0.054 C/ha higher, 0.016 C/ha higher, and 0.156 C/ha lower, respectively. These differences translated to a 0.55 %IS underestimate at target C/ha, a 0.16 %IS underestimate at threshold C/ha, and a 1.58 %IS overestimate at impairment C/ha by the TU2019 model. While differences were expected between model reference points and %IS estimates, particularly since they were from distinctly different models based on significantly different spatial resolutions of data, the target and threshold reference points and %IS estimates were strikingly similar.

Although not explicitly examined in this analysis, linking total %IS present to C/ha inherently incorporates vehicular infrastructure such as roads and parking lots into the model. The nonlinear nature of the relationship suggests that vehicular infrastructure per structure may differ between rural, suburban, and urban development. Vehicular infrastructure is highly variable in footprint (number of lanes and lane width among residential streets, rural routes, expressways, and interstate highways), density (parking space orientation and multi-level parking structures), and proximity to structures. Furthermore, tax coordinates are the parcel centroid rather than within a structure's footprint. These spatial variabilities are particularly evident at small spatial extents. Caution should be exercised if the tax model is to be used for %IS estimation at the small spatial extents of National Hydrography Dataset (NHD) catchments. Excluding catchments that did not intersect land, NHD catchments in Maryland numbered 14,597 (compared to 1,104 12-digit subwatersheds) and were comparatively small having a mean of 195 ha and median of 106 ha, although NHD catchment size was as large as 4,883 ha (personal observation). Eleven percent of these catchments were intersected by roads without having a tax record, meaning %IS would be underestimated by the tax model for these NHD catchments.

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Table 1. Watersheds included in the analysis and their estimated land area (hectares), index of development (C/ha), and estimate of actual percent impervious surface (%IS; TU = Towson University and CC = Chesapeake Conservancy). Different estimates of land hectares had to be used based on the data source: vector for C/ha 2000, 30 m for %IS TU 2000, and 1 m for both C/ha 2013 and %IS CC 2013. ‡Watersheds modified from their MDE8NAME delineation. *Watersheds included to increase the sample from the original 19 to a total of 33.

Watershed	Vector hectares	30 m hectares	1 m hectares	C/ha 2000	C/ha 2013	%IS TU 2000	%IS CC 2013
Baltimore Harbor ‡*	35,855	29,743	35,813	5.298	6.180	46.39	38.68
Big Annemessex River*	9,019	8,626	8,671	0.086	0.099	2.43	2.14
Bird River*	6,725	5,785	6,683	2.189	2.643	26.00	24.57
Blackwater River‡*	39,031	37,596	36,339	0.031	0.041	1.23	1.21
Bohemia River	10,757	10,682	10,748	0.100	0.114	1.22	3.61
Breton Bay	14,285	13,714	14,154	0.240	0.378	5.29	5.81
Broad Creek*	10,544	10,417	10,530	0.177	0.205	1.14	4.28
Broad Creek: Lower Choptank‡*	4,728	4,539	4,657	0.256	0.300	4.41	4.63
Bush River‡	36,009	33,621	35,834	1.203	1.257	11.28	13.73
Corsica River	9,676	9,359	9,632	0.141	0.259	4.12	3.63
Deer Creek*	37,724	37,269	37,598	0.208	0.242	1.24	4.31
Elk River ‡*	21,040	20,411	20,937	0.500	0.601	5.31	8.59
Fishing Bay*	39,031	37,596	36,339	0.031	0.041	1.23	1.21
Gunpowder River ‡	113,637	109,665	113,435	0.648	0.732	5.52	8.15
Harris Creek‡*	3,694	3,560	3,620	0.331	0.396	5.98	4.79
Langford Creek	9,641	9,360	9,512	0.065	0.075	3.12	2.21
Magothy River	9,205	7,919	9,141	2.572	2.804	20.02	20.71
Mattawoman Creek	24,430	23,046	24,377	0.692	0.917	8.96	9.30
Middle River	2,753	2,221	2,716	3.002	3.381	39.11	30.39
Miles River	11,062	10,807	10,977	0.224	0.256	3.44	4.54
Nanjemoy Creek	18,891	18,436	18,707	0.080	0.093	0.94	1.93
Northeast River	16,342	15,961	16,221	0.358	0.477	4.34	7.87
Piscataway Creek	17,634	15,905	17,603	1.221	1.494	16.48	13.30
Sassafras River*	19,580	19,170	19,512	0.092	0.110	2.40	2.98
Severn River	17,937	16,112	17,857	1.950	2.305	19.44	20.01
South River	14,773	13,769	14,675	1.162	1.373	10.93	13.18
St. Clements Bay	12,054	11,555	11,959	0.182	0.216	4.42	3.93
Transquaking River*	28,008	27,363	27,222	0.027	0.030	0.95	1.61
Tred Avon River‡*	9,538	9,094	9,445	0.570	0.763	7.74	9.31
West River & Rhode River	6,604	6,322	6,540	0.516	0.601	4.95	6.58
Wicomico River: Eastern Shore‡*	47,325	45,206	46,734	0.506	0.604	7.85	8.78
Wicomico River: Western Shore‡	58,807	56,807	58,452	0.282	0.361	4.29	4.90
Wye River	20,410	19,987	20,335	0.084	0.097	3.34	2.73

Table 2. Percent impervious surface for each watershed estimated from the Chesapeake Conservancy 2013/2014 land cover data (CCLC 2013) and estimated %IS predicted by the four models for the year 2013. *Watersheds not included in the linear original and power original models.

		Modeled % impervious surface			
Watershed	%IS CC2013	Linear original	Linear expand	Power original	Power expand
Baltimore Harbor *	38.68	49.95	44.36	42.78	40.10
Big Annemessex River*	2.14	3.08	3.58	1.90	2.24
Bird River*	24.57	22.69	20.64	22.55	22.17
Blackwater River*	1.21	2.63	3.19	0.98	1.21
Bohemia River	3.61	3.20	3.68	2.11	2.48
Breton Bay	5.81	5.23	5.45	5.21	5.71
Broad Creek*	4.28	3.90	4.29	3.29	3.73
Broad Creek: Lower Choptank*	4.63	4.63	4.93	4.38	4.86
Bush River	13.73	12.00	11.34	12.88	13.20
Corsica River	3.63	4.31	4.65	3.92	4.39
Deer Creek*	4.31	4.18	4.53	3.72	4.18
Elk River *	8.59	6.95	6.94	7.39	7.89
Fishing Bay*	1.21	2.63	3.19	0.98	1.21
Gunpowder River	8.15	7.96	7.82	8.57	9.05
Harris Creek*	4.79	5.37	5.57	5.39	5.90
Langford Creek	2.21	2.90	3.42	1.54	1.85
Magothy River	20.71	23.93	21.72	23.58	23.10
Mattawoman Creek	9.30	9.38	9.06	10.16	10.59
Middle River	30.39	28.38	25.59	27.15	26.33
Miles River	4.54	4.29	4.63	3.88	4.35
Nanjemoy Creek	1.93	3.03	3.54	1.81	2.14
Northeast River	7.87	5.99	6.11	6.21	6.72
Piscataway Creek	13.30	13.83	12.93	14.67	14.89
Sassafras River*	2.98	3.17	3.65	2.06	2.42
Severn River	20.01	20.09	18.38	20.35	20.16
South River	13.18	12.90	12.12	13.77	14.04
St. Clements Bay	3.93	3.98	4.36	3.41	3.86
Transquaking River*	1.61	2.55	3.11	0.77	0.97
Tred Avon River*	9.31	8.20	8.03	8.85	9.32
West River & Rhode River	6.58	6.95	6.94	7.38	7.89
Wicomico River: Eastern Shore*	8.78	6.97	6.96	7.42	7.92
Wicomico River: Western Shore	4.90	5.10	5.34	5.04	5.53
Wye River	2.73	3.07	3.57	1.87	2.22

Table 3. Regression model parameters from 2013 C/ha and %IS derived from 2013 Chesapeake Conservancy data. Approximated R^2 for power models was from regression of predicted and observed estimates.

CCLC2013 model	Coefficient	Estimate	SE	t	P
Linear original	y-intercept	2.317	0.378	6.127	<0.0001
	slope	7.707	0.288	26.756	<0.0001
	F=716	DF=1,17	P=<0.0001	R^2=0.977	RSE=1.195
Linear expanded	y-intercept	2.913	0.404	7.203	<0.0001
	slope	6.707	0.26	25.766	<0.0001
	F=664	DF=1,31	P=<0.0001	R^2=0.955	RSE=1.901
Power original	a	10.842	0.406	26.72	<0.0001
	b	0.754	0.04	18.69	<0.0001
	DF=17	RSE=1.343		$R^2_{Approx.}$=0.972	
Power expanded	a	11.255	0.284	39.59	<0.0001
	b	0.698	0.02	35.37	<0.0001
	DF=31	RSE=1.210		$R^2_{Approx.}$= 0.982	

Table 4. Comparison of AICc statistics for CCLC2013 linear and nonlinear models for both the original and expanded sets of watersheds. Δ_i and the evidence ratio are for comparison of model liklihood.

Dataset	Model	K	AICc	Δi	Evidence Ratio
Original	CCLC2013 linear	3	63.72	0.00	1.00
	CCLC2013 power	3	70.21	6.49	25.66
Expanded	CCLC2013 power	3	111.21	0.00	1.00
	CCLC2013 linear	3	139.41	28.20	1.33×10^6

Table 5. Analysis of covariance (ANCOVA) to test for differences in slope (slope:model) and y-intercept (model) between the TU2019 model and the preferred CCLC2013 linear model.

Coefficient	Estimate	Std Error	t-value	P
y-intercept	2.3173	0.7155	3.239	0.0027
slope	7.7074	0.5449	14.144	<0.0001
model	-1.0312	0.9965	-1.035	0.3081
slope:model	2.4214	0.8160	2.967	0.0055

Table 6. Development (C/ha) associated with benchmark %IS derived using the TU2019 model and CCLC2013 models. The TU2019 C/ha values are included for comparison of benchmarks derived from coarse (Towson University) and fine (Chesapeake Conservancy) resolution impervious cover estimates.

	%IS & corresponding C/ha			
Model	2% IS	5% IS	10% IS	15% IS
TU2019 index original	0.070	0.367	0.860	1.354
CCLC2013 linear original	-0.041	0.348	0.997	1.646
CCLC2013 linear expanded	-0.136	0.311	1.057	1.802
CCLC2013 Power original	0.106	0.358	0.898	1.538
CCLC2013 power expanded	0.084	0.312	0.844	1.509

Figure 1. Side-by-side comparison of 30 m x 30 m (Landsat TM) and 1 m x 1 m (Chesapeake Conservancy) resolution raster data that depict the same area of developed land in Prince George's County, Maryland. The Landsat TM data was collected in 2011 and classified by the Multi-Resolution Land Characteristics consortium according to land cover for the National Land Cover Database. Additional land classifications are not shown for clarity. Chesapeake Conservancy's land cover data was collected in 2013/2014 and classification explicitly identified land cover as either impervious or pervious categories.

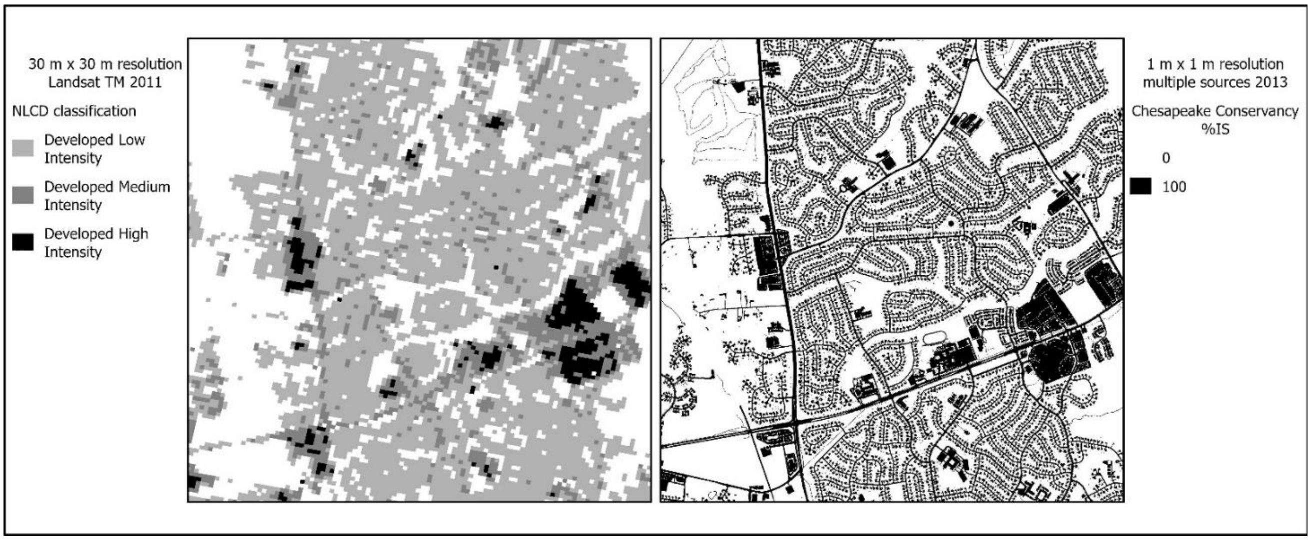


Figure 2. Map of Maryland's Chesapeake Bay showing the location of watersheds used for the original tax index (gray) and additional watersheds used in the expanded analysis (green).

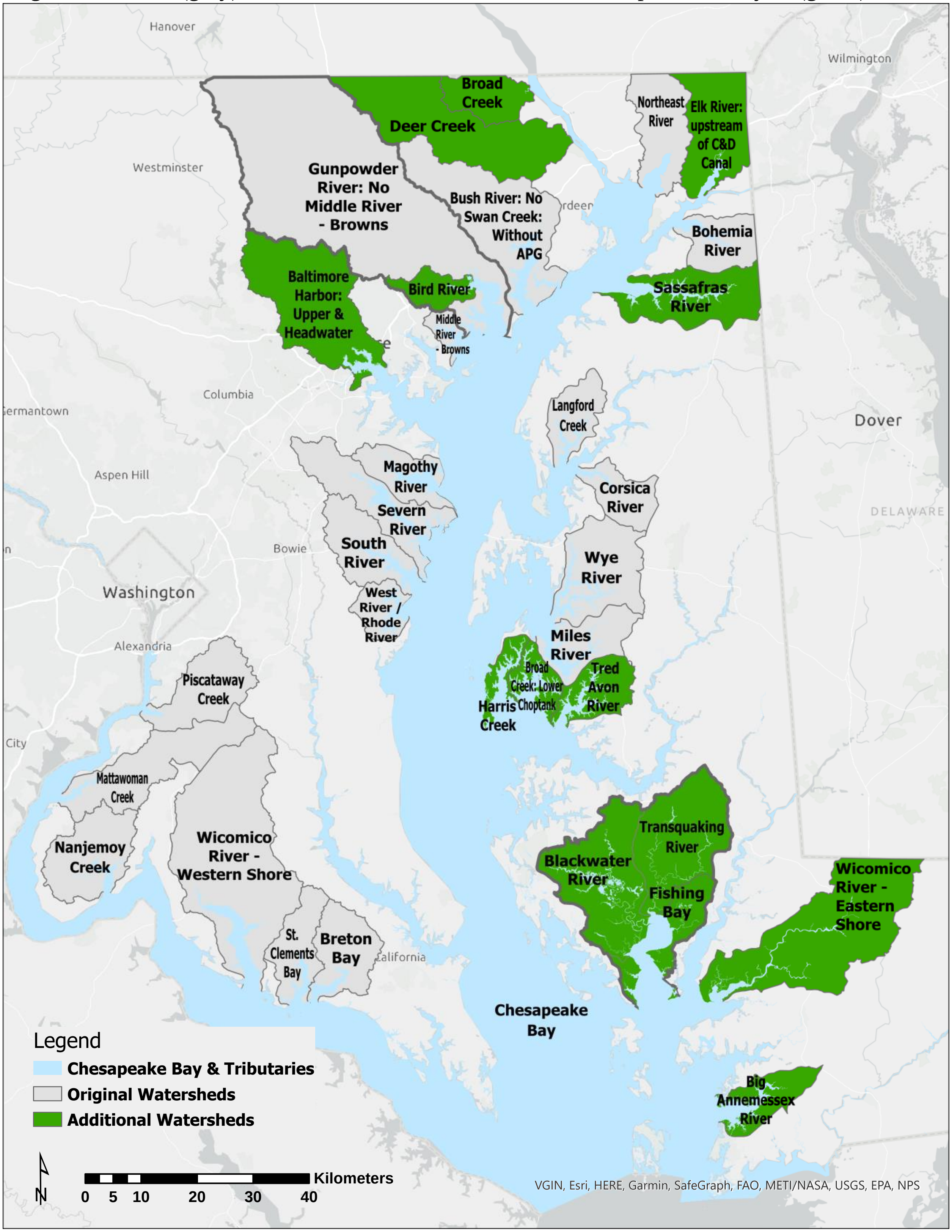


Figure 3. Linear (A) and power (B) models for the original set of 19 watersheds and the expanded set of 33 watersheds derived from Chesapeake Conservancy 2013/2014 land cover and MdProperty View tax data. Standardized residual plots for each model are directly below the model plots. Data, line-of-best-fit, and residuals for the original watersheds are in blue.

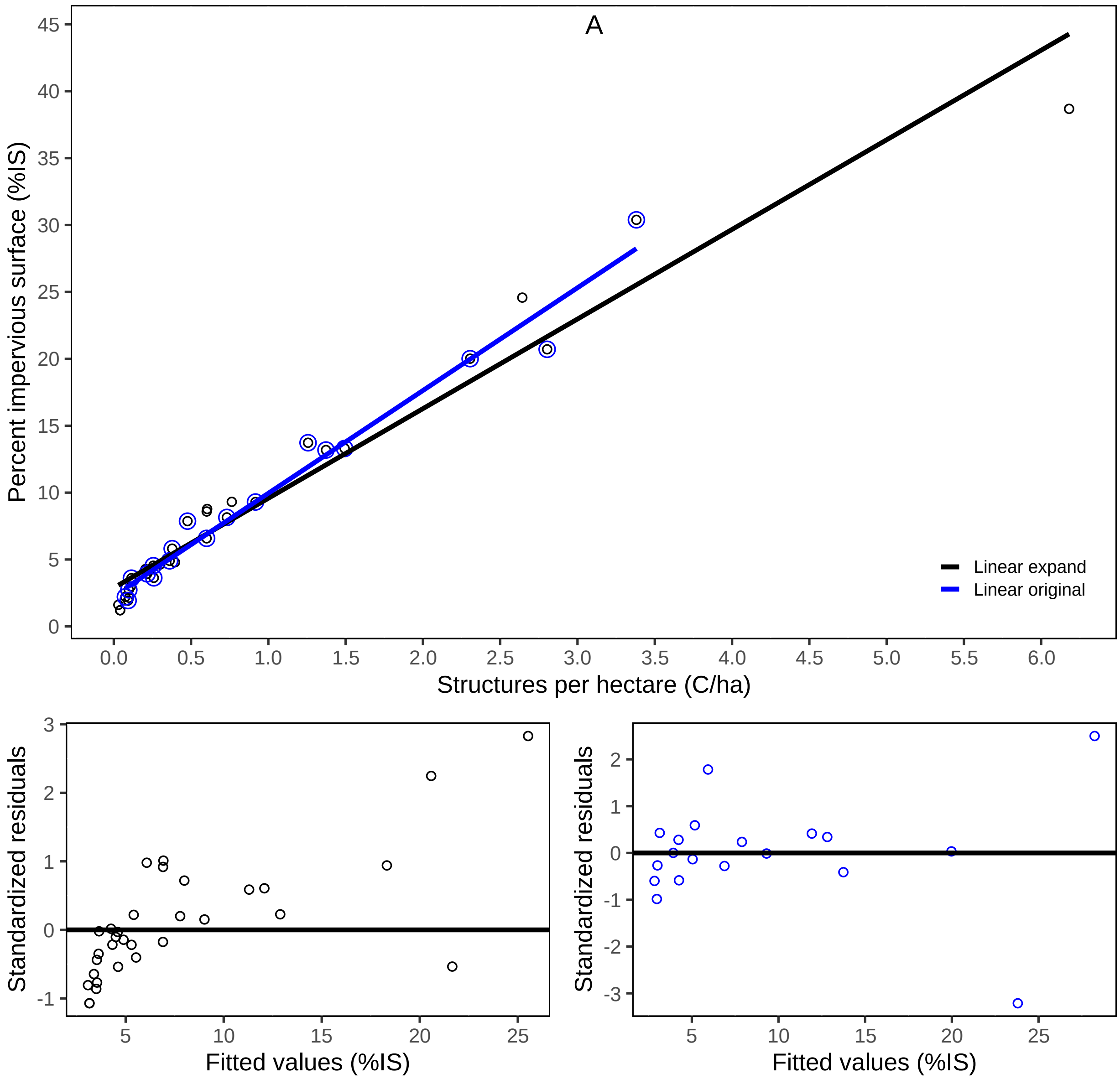


Figure 3 cont.

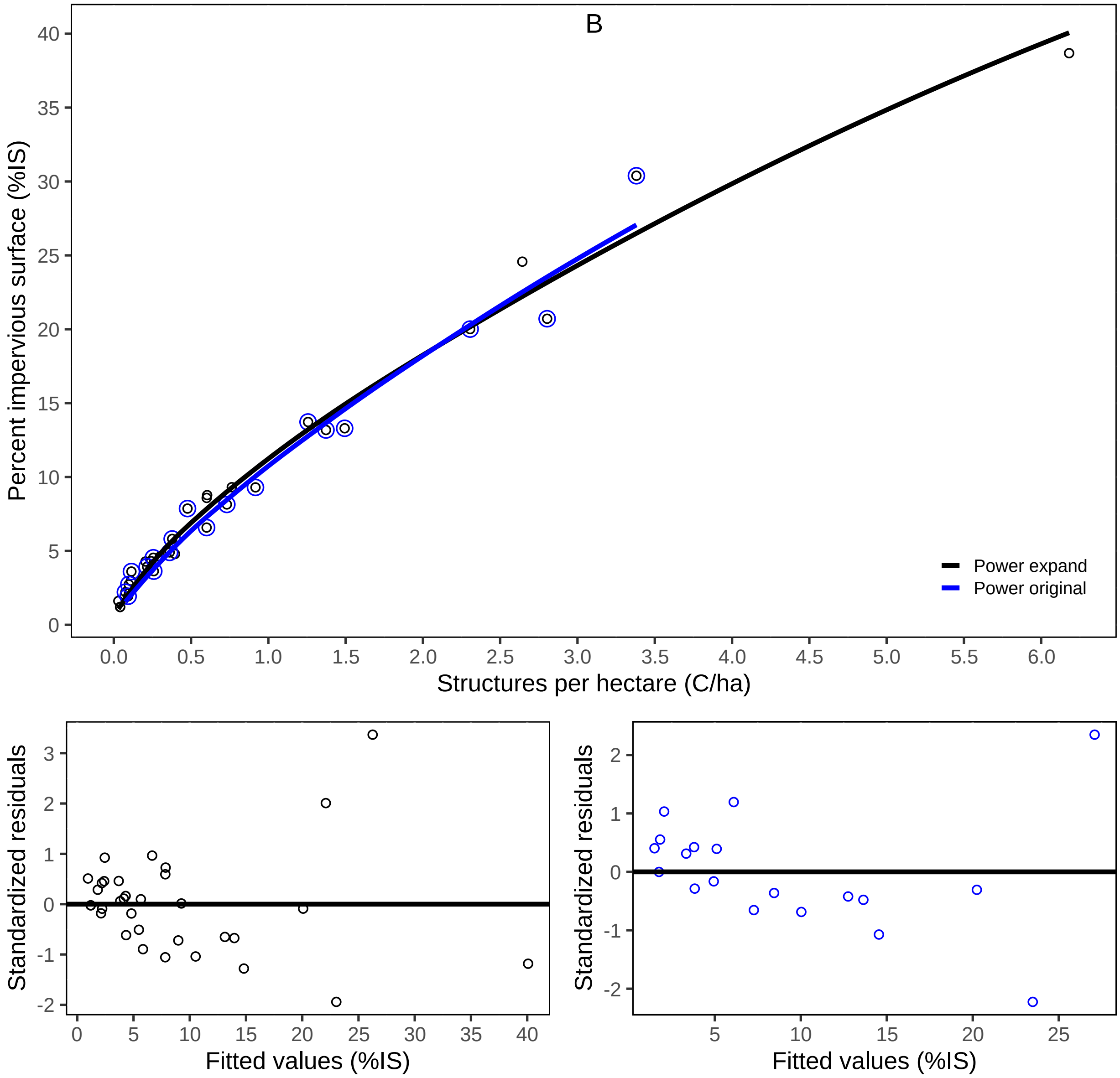


Figure 4. Comparison of linear and power models using CCLC2013 %IS estimates for the original watersheds (A) and the expanded watersheds (B). Plot C is a comparison of the CCLC2013 preferred linear and power models. Linear models are blue and power models are black. Ninety percent confidence intervals are shown for each model with corresponding colors: light blue for linear and gray for power.

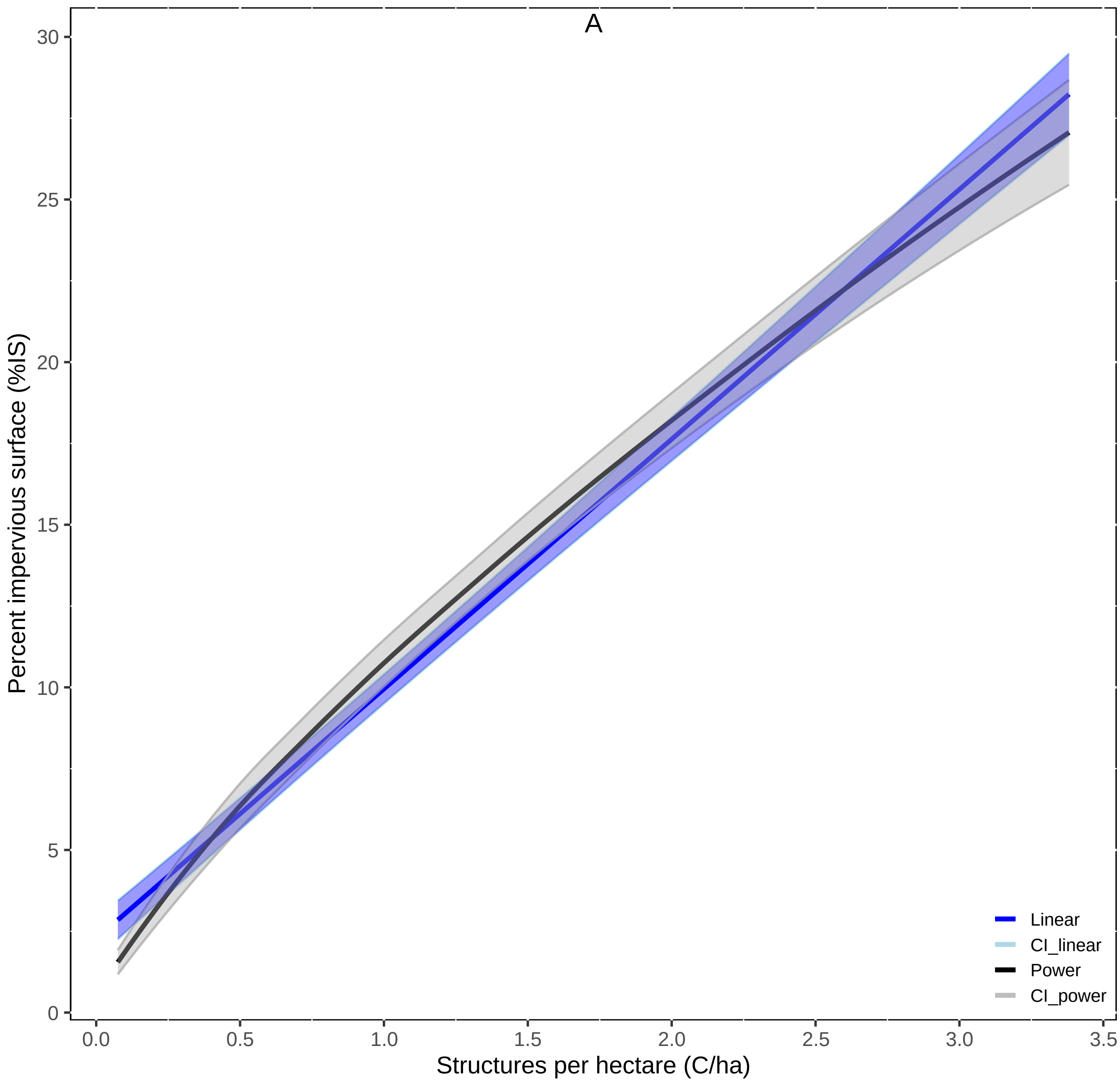


Figure 4 cont.

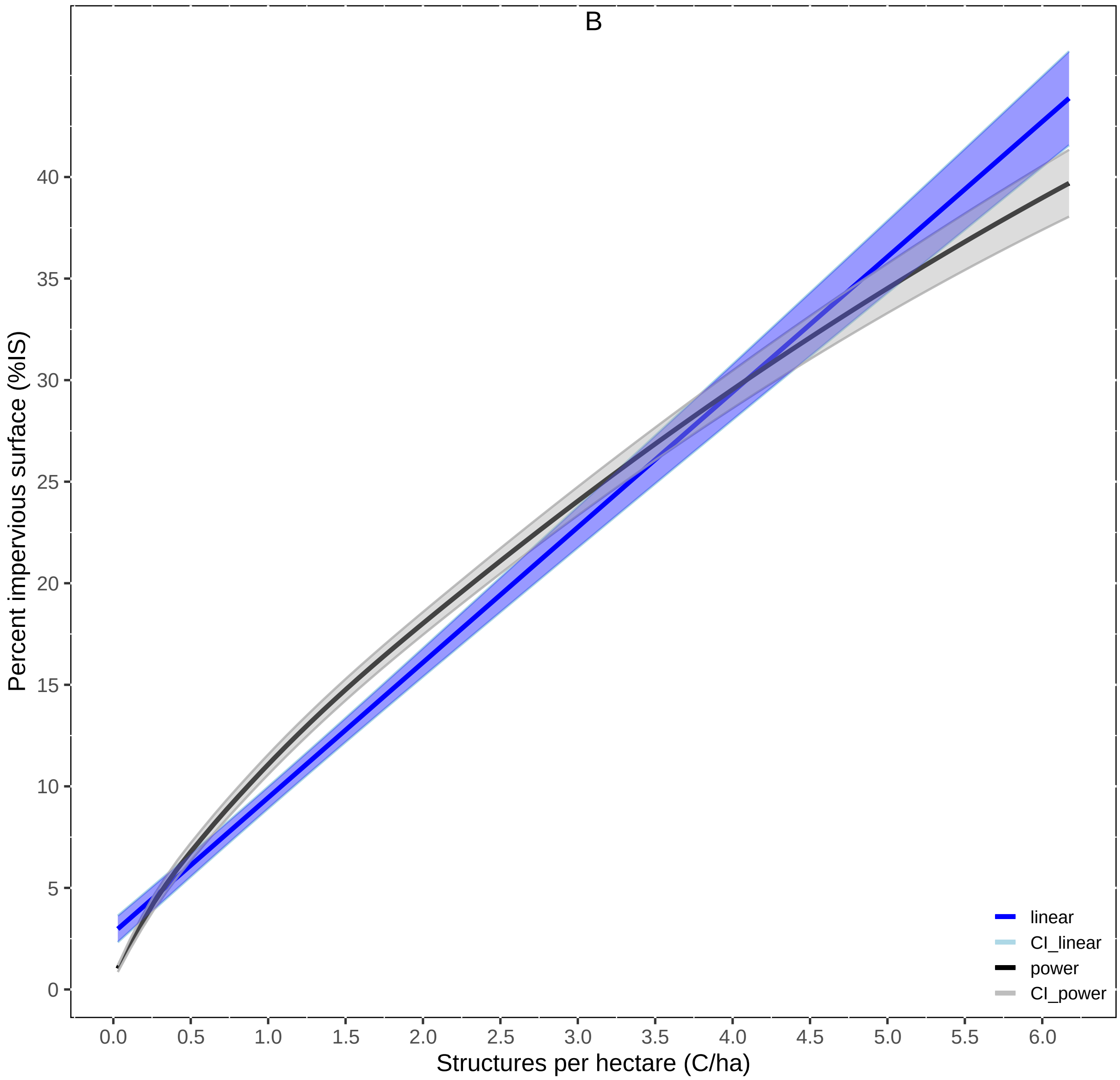


Figure 4 cont.

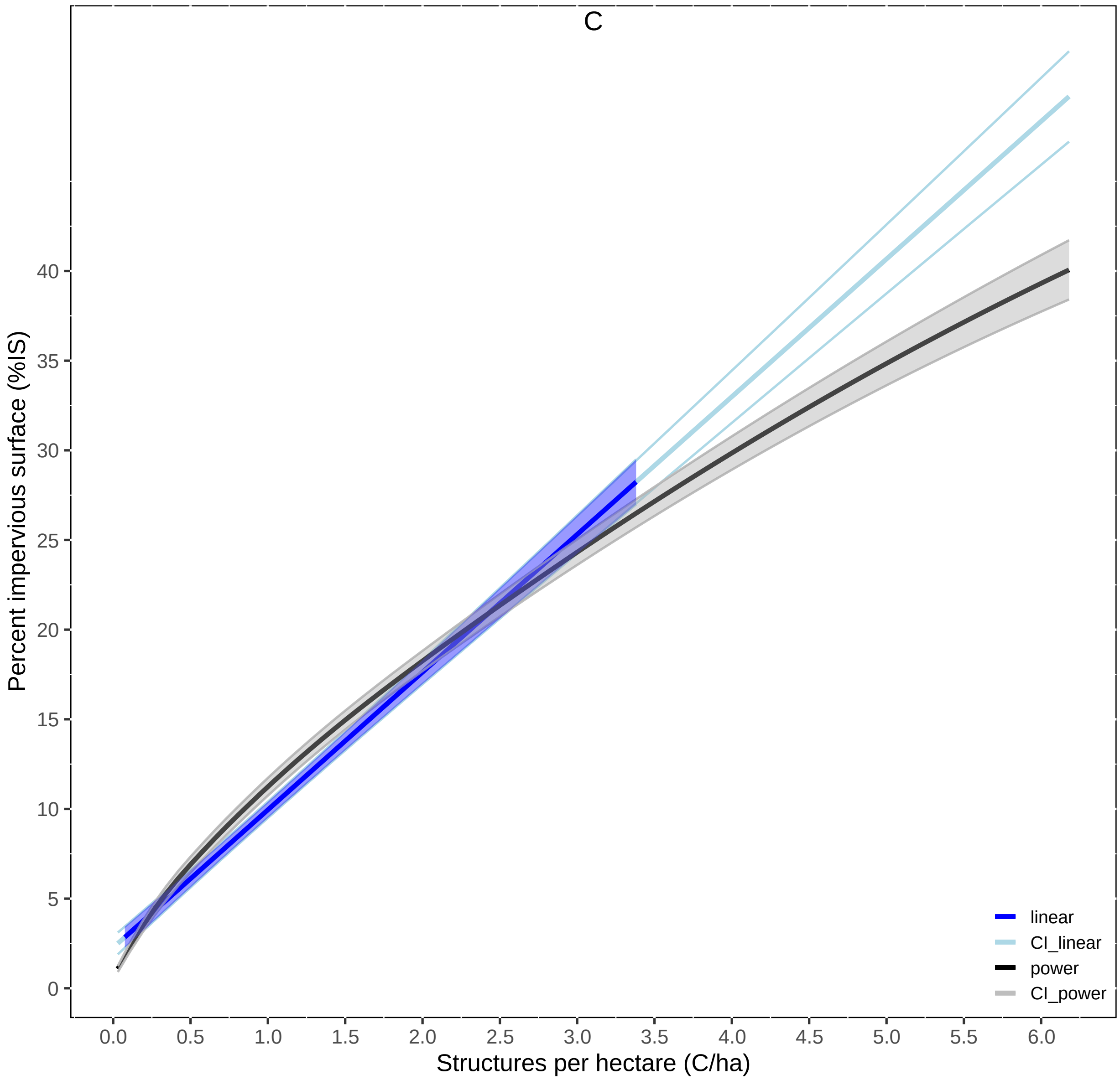


Figure 5. Comparison of tax index models based on the high-resolution Chesapeake Conservancy 2013/2014 data and the TU2019 model applied to 2013 tax data (C/ha). The TU2019 model was compared with (A) the CCLC2013 linear model derived from the original set of watersheds and (B) the CCLC2013 power model derived from the expanded set of watersheds. The CCLC2013 models are in blue and the TU2019 model in in black. Ninety percent confidence intervals for the models are shown with corresponding colors: light blue for CCLC2013 and gray for TU2019.

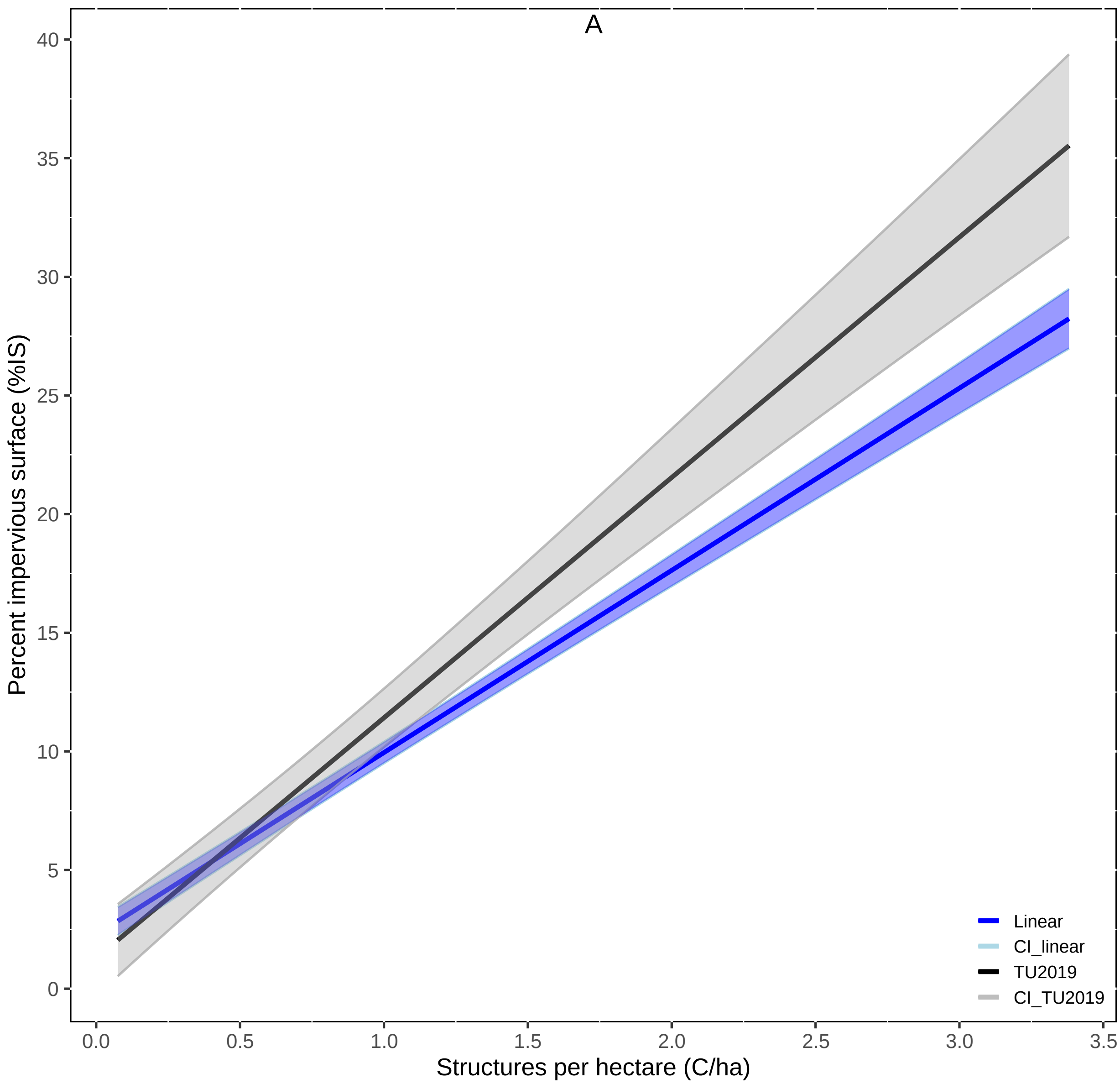
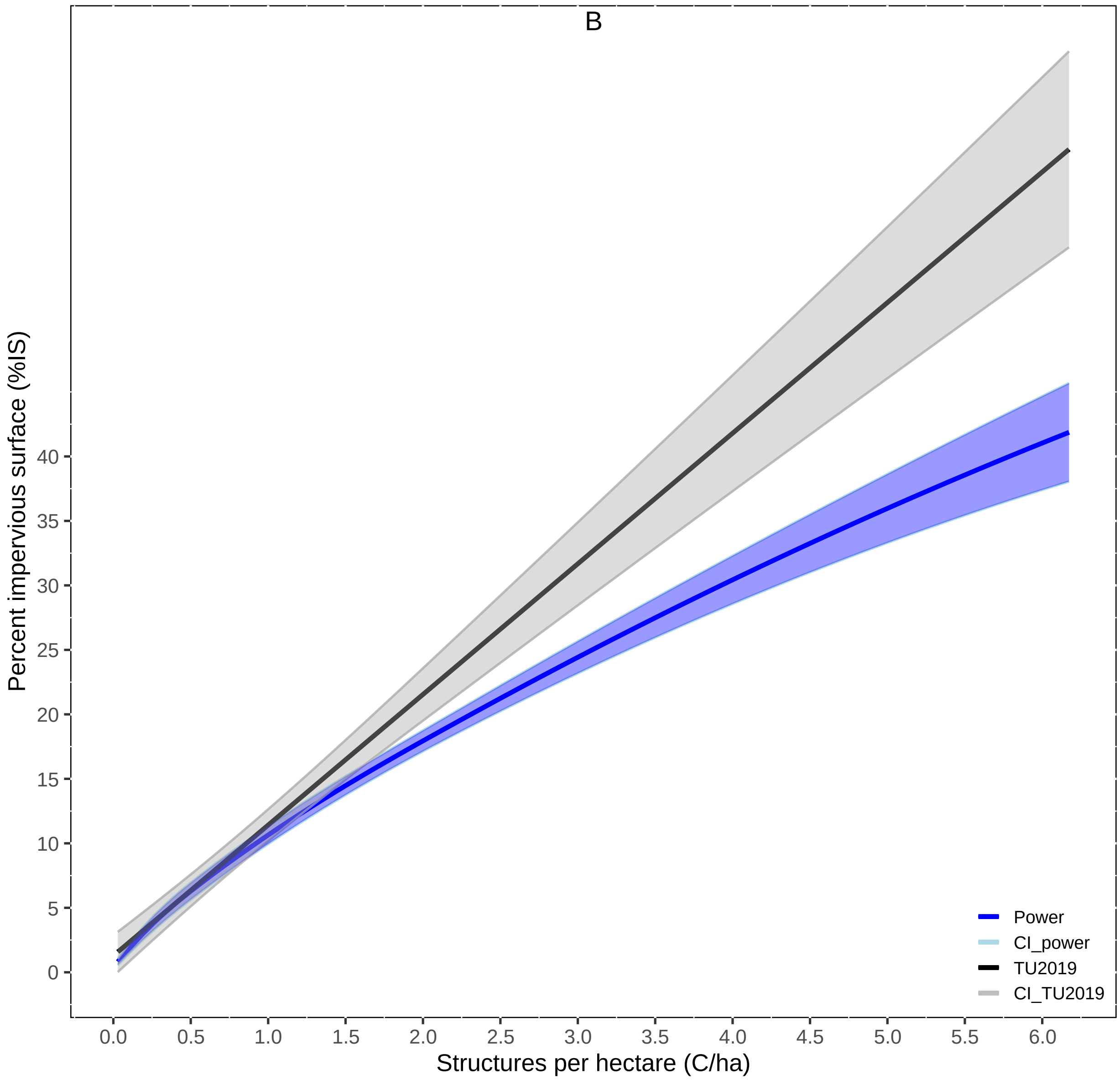


Figure 5 cont.



MD – Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern
Section 1: Stream Ichthyoplankton Sampling

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

Introduction

Urbanization associated with increased population growth became a factor in the decline of diadromous fishes in the late 20th century (Limburg and Waldman 2009). Increases in impervious surface have altered hydrology and increased diadromous fish habitat loss (Limburg and Waldman 2009). Anadromous fish egg densities (Alewife and White Perch) in the Hudson River exhibited a strong negative threshold response to urbanization (Limburg and Schmidt 1990). We were interested in understanding how reference points for development (impervious surface reference points or ISRPs, or C/ha reference points) developed for Chesapeake Bay subestuaries (Uphoff et al. 2011) were related to anadromous fish spawning in streams in Maryland's portion of Chesapeake Bay.

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; O'Dell and Mowrer 1984; Mowrer and McGinty 2002; Uphoff et al. 2020). 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 sampling methods of O'Dell et al. (1975; 1980) and O'Dell and Mowrer (1984) were used to sample Mattawoman Creek (2008-2018), Piscataway Creek (2008-2009 and 2012-2014), Bush River (2005-2008 and 2014), Deer Creek (2012-2015), Tuckahoe Creek (2016-2017), Choptank River (2016-2017), Patapsco River (2013-2017), Chester River (2019), and Patuxent River (2021 preliminary data; Figure 1-1).

Mattawoman and Piscataway Creeks are adjacent Coastal Plain watersheds along an urban gradient emanating from Washington, DC (Table 1-1; 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 within a conservation district located entirely in the Piedmont north of Baltimore, near the Pennsylvania border (Clearwater et al. 2000). Bush River and Deer Creek drainages are adjacent to each other. The Choptank River drainage, which includes Tuckahoe Creek, is a major eastern shore tributary of the Chesapeake Bay within the Coastal Plain and has a watershed dominated by agriculture. The Patapsco River watershed is located within Coastal Plain and Piedmont provinces, with rolling hills over much of its area that are characteristic of the eastern division of the Piedmont province, while to the southeast the watershed lies in the Coastal Plain bordering the western side of the Chesapeake Bay (O'Dell et al. 1975). Fluvial Patapsco River meets the Chesapeake Bay and forms the port of Baltimore. The Chester River, located on the eastern shore, is a fluvial-tidal system located in the Coastal Plain. Agriculture is predominant in its watershed (O'Dell et al. 1975; Table 1-1; Figure 1-1). The Patuxent River, located within the Piedmont and Coastal Plain provinces, is a major tributary of the Chesapeake Bay and is the largest river that is located entirely within the state of Maryland. The upper portion of the drainage (MD Route 214 north, including the Little Patuxent River drainage) is located between Washington, D.C. and Baltimore, while the middle portion of the drainage (MD Route 214 south to Hall Creek) extends through Anne Arundel, Prince George's, and Calvert counties (O'Dell and Mowrer 1984; Figure 1-1). The Patuxent River is urbanized, with extensive development that has affected water quality and physical characteristics of the system (O'Dell and Mowrer 1984; Uphoff et al. 2018; Table 1-1; Figure 1-1).

We developed two indicators of anadromous fish spawning in a watershed based on presence-absence of eggs and-or larvae: occurrence at a site (a spatial indicator) and proportion of samples with eggs and-or 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) and O'Dell and Mowrer (1984). This spatial indicator was compared to the extent of development in the watershed (counts of structures per hectare or C/ha; Topolski 2015) between the 1970s and the present. An indicator of habitat occupation in space and time from collections that started 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's eggs and-or larvae was compared to level of development (C/ha) and conductivity, an indicator of water quality strongly associated with development (Wang and Yin 1997; Paul and Meyer 2001; Wenner et al. 2003; Morgan et al. 2007; Carlisle et al. 2010; Morgan et al. 2012; Moore et al. 2017; Bird et al. 2018).

In addition, we attempted to address the possibility that proportion of samples with anadromous Herring may have been impacted by spawning stock abundance increases due to more restrictive coast-wide regulatory measures implemented over the past decade. Closures of most in-river fisheries along the Atlantic Coast were in place by 2012 (including Maryland in 2011; ASMFC 2019) and caps on River Herring bycatch in Atlantic Herring and Atlantic Mackerel fisheries that started in 2014 (MAFMC 2019) could have boosted Herring spawning stock. Increases in presence of Herring eggs and-or larvae due to regulatory measures (or other large-scale factors such as decreased predation or increased at-sea survival due to improved feeding and-or environmental conditions) should potentially have been evident across three watersheds studied before and after regulatory measures were put in place. Increases in spawning stock abundance over time would have the potential to bias estimated relationships of C/ha and conductivity with indicators of anadromous Herring stream spawning intensity.

Methods

Stream sites sampled for anadromous fish eggs and larvae during 2005-2019 and 2021 were typically at road crossings that O'Dell et al. (1975; 1980) and O'Dell and Mowrer (1984) determined were anadromous fish spawning sites during the 1970s and 1980s. O'Dell et al. (1975; 1980) and O'Dell and Mowrer (1984) summarized spawning activity as the presence of an anadromous fish species (White Perch, Yellow Perch, or Herring) group's egg, larva, or adult at a site sampled with stream drift ichthyoplankton nets and wire traps.

All collections during 2005-2019 and 2021, with the exception of Deer Creek during 2012-2015, Choptank River and Tuckahoe Creek during 2016-2017, Patapsco River during 2013-2017, Chester River during 2019, and Patuxent River during 2021 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 1-2). Tributary sites

MUT4 and MUTX were selected based on volunteer interest and added in 2010 and 2014, respectively; MUTX was discontinued in 2015 due to restricted access and limited indication of spawning. All mainstem sites were sampled in 2016-2018, while the only tributary site sampled was MUT3; beaver dams blocked spawning access to MUT4 and MUT5. 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; Uphoff et al. 2015). Deer Creek sites SU01-SU04 were sampled in 2012 and sampling continued in 2013-2015 with the addition of site SU05 (Figure 1-5). Choptank River (CH100-CH111; Figure 1-6) and Tuckahoe Creek (TUC101-TUC110; Figure 1-7) sites were sampled in 2016-2017. Patapsco River samples (four sites; Figure 1-8) were collected during 2013-2017 by U.S. Fish and Wildlife Service and were added to this data set. Chester River (CH19001-CH19016; Figure 1-9) was sampled during 2019 to provide up-to-date information for the Queen Anne County comprehensive growth plan. Patuxent River (12 sites; Figure 1-10) was sampled during 2021 to provide information for the Anne Arundel County comprehensive growth plan. Table 1-2 summarizes sites, dates, and sample sizes in Mattawoman, Piscataway, Deer, and Tuckahoe creeks, and Bush, Choptank, Patapsco, Chester, and Patuxent rivers during 2005-2019 and 2021.

Ichthyoplankton samples were collected in all systems and years 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. Collections in Choptank River and Tuckahoe Creek during 2016-2017, and in Patuxent River in 2021, were made using stream drift nets at wadable sites or using a conical plankton net towed from a boat (see Section 2 for a description of ichthyoplankton sampling by boat) at sites too deep to wade (Uphoff et al. 2017; 2018). These techniques mimic collections made by O’Dell et al. (1980) within the Choptank River drainage, specifically Tuckahoe Creek, and by O’Dell and Mowrer (1984) within the Patuxent River drainage. For both types of collections, 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 both in the jar and on top of the lid before it was sealed. Samples were fixed immediately with 10% buffered formalin after collection by DNR staff, or were placed in a cooler with ice for transport and preserved after a volunteer team was finished sampling for the day. Water temperature (°C), conductivity (μS/cm), and dissolved oxygen (DO, mg/L) were recorded at each site using either a hand-held YSI Model 85 meter or YSI Pro2030 meter. Meters were calibrated for DO each day prior to use. All data were recorded on standard field data sheets and double-verified at the site during volunteer collections. Approximately 2-ml of rose bengal dye was added to each sample in order to stain the organisms pink 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. With the exception of 2018, all eggs and-or larvae were removed and 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 systematically sorted 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. Results for Patuxent River in 2021 are preliminary as, due to time and staffing limitations, picking is not currently complete. 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 the presence or absence of each of the above was recorded. 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 previous surveys.

Collections and sample processing were adjusted in 2018 due to anticipated time and staffing limitations. Mattawoman Creek volunteers received training on field identification of Herring eggs and larvae prior to the start of the season, and if they were able to determine presence in the field the sample was not retained. Samples that they could not determine conclusively contained Herring, or ones in which no eggs or larvae were observed in the field, were preserved for laboratory examination. In the lab, samples were sorted only for presence of Herring eggs and-or larvae. Once a Herring egg or larvae was encountered, processing of the sample was considered complete, regardless of how much of it had been gone through.

Methods used to estimate development (C/ha) and land use indicators (percent of watershed in agriculture, forest, wetlands, and urban land use) are explained in **General Spatial and Analytical Methods used in Project 1, Sections 1-3.** Development targets and limits and general statistical methods (analytical strategy and equations) are described in this section as well. Specific spatial and analytical methods for this section of the report are described below.

Watershed area draining into the Herring spawning areas (hereafter, watershed), land use, and C/ha in those Herring spawning areas were estimated. Mattawoman Creek's watershed was 24,430 ha and estimated C/ha increased from 0.87 to 0.97 during 2008-2018; Piscataway Creek's watershed was 17,634 ha and estimated C/ha increased from 1.41 to 1.50 during 2008-2014; Bush River's watershed was 36,009 ha and estimated C/ha increased from 1.37 to 1.52 during 2005-2014; and Deer Creek, a spawning stream with low development, had a watershed of 37,724 ha and estimated C/ha was 0.24 during 2012-2015 (Table 1-1). The upper portion of the Choptank River (watershed area = 38,285 ha and developmental level = 0.18 C/ha) and a tributary of the Choptank River, Tuckahoe Creek (watershed area = 39,364 ha and developmental level = 0.07 C/ha), were added in 2016-2017; and the Chester River drainage (watershed area = 77,751 and developmental level = 0.13 C/ha) was sampled in 2019. These three systems are all spawning streams with high agricultural influence and low watershed development. The Patuxent River (watershed area = 100,181 ha and developmental level = 1.34 C/ha) was sampled in 2021 (Table 1-1; Figure 1-1). Deer Creek, Choptank River and Tuckahoe Creek, Chester River, and Patuxent River (all upper river, and two middle river, stream drift net sites) were sampled by DNR biologists from the Fishery Management Planning and Fish Passage Program. Four middle Patuxent River collections were made by boat by DNR biologists from the Fish Health and Hatcheries, Anadromous Species Division, all at no charge to this grant. Patapsco River's watershed equaled 93,730 ha and estimated C/ha was 1.11 in 2013 and 1.15 in 2017. Collections in the Patapsco River were made by U.S. Fish and Wildlife Service and were provided at no charge to this grant.

Conductivity was measured for each date and stream site (mainstem and tributaries) during 2008-2018 from Mattawoman Creek, but only mainstem measurements were summarized for each year. Mainstem sites would be influenced by development in Waldorf, the major urban influence on the watershed, while the monitored tributaries would not (Figure 1-2). Unnamed tributaries were excluded from calculation of summary statistics to capture conditions in the largest portion of habitat. 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 (although they can provide within watershed low versus high development comparisons for spawning streams in years when all were sampled). Conductivity was measured with each sample in Deer Creek in 2012-2015, in the Choptank River and Tuckahoe Creek in 2016-2017, in the Patapsco River in 2013-2017, in the Chester River drainage in 2019, and in the Patuxent River in 2021.

Presence of eggs and-or larvae of White Perch, Yellow Perch, and Herring at each station was compared to past surveys to determine which sites still supported spawning. The only exception was Mattawoman Creek in 2018 when only presence of Herring eggs and-or larvae was determined. We used the criterion of detection of eggs and-or larvae at a site (O'Dell et al. 1975; 1980; O'Dell and Mowrer 1984) as evidence of spawning. Raw data from early 1970s and 1980s collections were not available to formulate other metrics.

Sites where Herring spawning was detected (site occupation) during the current study and historical studies were compared to changes in C/ha. Historical site occupation was available for Mattawoman Creek mainstem stations sampled in 1971 by O'Dell et al. (1975) and Hall et al. (1992) during 1989-1991. Hall et al. (1992) collected ichthyoplankton with 0.5 m diameter plankton nets (3:1 length to opening ratio and 363μ mesh set for 2-5 minutes, depending on flow) suspended in the stream channel between two posts instead of stream drift nets. Historical site occupation was available for Piscataway Creek in 1971 (O'Dell et al. 1975), Deer Creek in 1972 (O'Dell et al. 1975), Bush and Patapsco Rivers in 1973 (O'Dell et al. 1975), Tuckahoe Creek in 1976-77 (O'Dell et al. 1980), Chester River in 1975-1977 (O'Dell et al. 1980), and Patuxent River in 1980-1983 (O'Dell and Mowrer 1984).

The proportion of samples where Herring eggs and-or larvae were present (P_{herr}; described below) was estimated for Mattawoman Creek mainstem stations (MC1-MC4) during 1991 and 2008-2018, Piscataway Creek (2008-2009 and 2012-2014), Bush River (2005-2008 and 2014), Deer Creek (2012-2015), Choptank River (2016-2017), Tuckahoe Creek (2016-2017), Patapsco River (2013-2017), Chester River drainage (2019), and Patuxent River (2021). Counts of Herring eggs and larvae were available for Mattawoman in 1991 (C/ha = 0.48) in a tabular summary in Hall et al. (1992) at the sample level and these data were converted to presence-absence. Herring was the only species group with adequate sample sizes for annual proportion of sample 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 corresponded to O'Dell et al. (1975) sites 1, 2, and 3 respectively (Figure 1-5). Two additional sites, SU02 and SU03 were sampled and analyzed in this system as well. The mainstem of the Choptank River had not been sampled previously, so 12 stations (going from downstream to upstream, CH100-CH111; Figure 1-6) were added in that system for analysis.

Tuckahoe Creek stations TUC101, TUC102, TUC103, TUC108, TUC109, and TUC110 (going from downstream to upstream) correspond to O'Dell et al. (1980) sites 4, 5, 6, 8, 11, and 12 respectively (Figure 1-7). Four additional sites were sampled in this system and analyzed as well. Sampling in the Patapsco River was within an area similar to that of O'Dell et al. (1975), but sites were different (Figure 1-8). All sites sampled within the Chester River drainage correspond to sites sampled by O'Dell et al. (1980; Figure 1-9). Eight of the twelve sites sampled within the Patuxent River correspond to sites sampled by O'Dell and Mowrer (1984; Figure 1-10).

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:

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

The 90% confidence intervals were constructed as:

$$^{(3)}\ P_{herr} \pm (1.645 \bullet \mathrm{SD}).$$

Two regression approaches were used to examine possible linear relationships between C/ha or standardized conductivity and P_{herr}: simple linear regression and multiple regression using two dependent variables, a categorical variable to indicate two levels of spawning stock (low and high), and C/ha or standardized conductivity. Simple linear 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}, and estimates of watershed percentage that was agriculture or forest with P_{herr}. Data were from Mattawoman, Piscataway, Deer and Tuckahoe creeks, and Bush, Choptank, Patapsco, Chester and Patuxent rivers (estimate of P_{herr} for Patuxent River is preliminary). Thirty-seven sets of estimates of C/ha, percent agriculture, percent forest, and P_{herr} were available (1991 estimates for Mattawoman Creek could be included), while 36 estimates were available for standardized conductivity (Mattawoman Creek conductivity 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}, that either linear or curvilinear relationships might be applicable to C/ha with standardized conductivity and standardized conductivity with P_{herr}, and that quadratic relationships best described the relationships of percentage of a watershed that was either agriculture or forest and P_{herr} (see Uphoff et al. 2018 for additional information and results). Nonlinear power functions were used to fit curvilinear models. Simple 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 (both had two parameters) model was considered the best description if a moderate or strong relationship was suggested, 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 for physiographic province by dividing by an estimate of the background expected from a stream absent anthropogenic influence (Morgan et al. 2012). Piedmont and Coastal Plain streams in Maryland have different background levels of conductivity, and 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 25th 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 μS/cm) when development was relatively low (Hall et al. 1992). Background conductivity used to standardize median conductivities was 109 μS/cm in Coastal Plain streams and 150 μS/cm in Piedmont streams (Morgan et al. 2012). For Bush, Patapsco, and Patuxent rivers, watersheds that run through both physiographic provinces, conductivities were standardized using the 150 μS/cm of Piedmont streams since sampling locations were solely within that region.

Multiple regression of C/ha or standardized conductivity and spawning stock class against P_{herr} assumed slopes were equal for two stock size categories, but intercepts were different (Neter and Wasserman 1974; Rose et al. 1986; Freund and Littell 2006). This common slope would describe the relationship of C/ha or standardized conductivity to P_{herr}, while the intercept would indicate the effect of high or low spawning stock size. This analysis was conducted for the 2005-2021 time-series and excluded 1991. These analyses were initially done in Excel and run again in SAS (Proc Reg) to confirm the estimates. Spawning stock size was modeled as an indicator variable in the multiple regression with 0 indicating lower spawning stock prior to the full implementation of river closures and bycatch reductions (2005-2011) and 1 indicating higher spawning stock following these measures (2012-2021). Categorizing spawning stock was necessary because P_{herr} would be the indicator of spawning stock size for each watershed and the dependent variable in the analysis if used as a continuous variable. None of the watersheds studied had independent indicators of spawning stock size. Rose et al. (1986) presented the use of categorized variables and linear regression as an alternative to Box-Jenkins models and time-series regression. In addition to standard regression output, we also used the type II sums of squared partial correlation coefficients to examine the amounts of variation in P_{herr} explained by each independent term in the multiple regression models after holding the other constant (Ott 1977; Sokal and Rohlf 1981; Afifi and Clark 1984).

Results

Development level of Piscataway, Mattawoman, and Deer creeks, Bush, Chester, and Patuxent rivers, and the Choptank River drainage (which includes Tuckahoe Creek) watersheds started at approximately 0.05 C/ha in 1950, while Patapsco River was approximately 0.20 C/ha at this time (Figure 1-11). Surveys conducted by O'Dell et al. (1975, 1980) in the 1970s, and O'Dell and Mowrer (1984) in the 1980s, sampled largely rural watersheds (C/ha < 0.28) except for Piscataway Creek (C/ha = 0.47), Patapsco River (C/ha = 0.44), and Patuxent River (C/ha = 0.33). By 1991, C/ha in Mattawoman Creek was similar to that of Piscataway in 1970. By the mid-2000s, Bush and Patuxent Rivers and Piscataway Creek were at higher suburban levels of development (~1.36 C/ha) than Mattawoman Creek (~0.81 C/ha) and Patapsco River (~1.02 C/ha). Deer Creek (zoned for agriculture and preservation) and the Choptank and Chester River drainages (predominantly agricultural) remained rural through 2019 (0.24, 0.18, and 0.13 C/ha, respectively; Figure 1-11).

Conductivities were usually elevated beyond background levels in all streams studied during 2008-2021 and median conductivities ranged from 1.14- to 2.42-times expected background levels (Table 1-3). In general, Deer Creek and Choptank River appeared to have consistently low conductivity, Patapsco River and Piscataway Creek had consistently high conductivity, and Patuxent River had the highest conductivities seen to date. Mattawoman Creek exhibited the highest inter-annual variation (1.14- to 1.94-times background). Bush River (1.39- to 1.69-times), Tuckahoe Creek (~1.46-times), and Chester River (1.66-times) were similarly elevated, even though Tuckahoe Creek and Chester River were much more rural (Table 1-3).

Herring spawning was detected at all mainstem stations sampled in Mattawoman Creek (MC1-MC4) during 1971 and 1991 (Table 1-4). Herring spawning in fluvial Mattawoman Creek was detected at two mainstem sites during 2008-2009 and all four mainstem stations during 2010-2018. Herring spawning was not detected at tributary site MUT3 during 2008-2010, but was consistently present during 2011-2016. Herring spawning was not detected in 2017 at MUT3, but was in 2018. Spawning was intermittently detected at MUT4 and MUT5 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 2013-2017, at MC2 during 2013-2014 and 2016-2017, and at MC3 during 1971 and 2016. Yellow Perch stream spawning in Mattawoman Creek has only been detected at MC1 in all surveys conducted since 1971, with the exceptions of 2009 and 2012 when spawning was not detected (Table 1-4). Presence of White Perch and Yellow Perch spawning in Mattawoman Creek was not determined in 2018 due to time and staffing limitations.

Herring spawning was detected at all mainstem sites in Piscataway Creek in 2012-2014 (Table 1-5). Stream spawning of anadromous fish had nearly ceased in Piscataway Creek between 1971 and 2008-2009. 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. There were far less occurrences of White and Yellow Perch eggs and larvae 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 of these sites 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; evidence of spawning was absent in 2013 and 2015, while spawning was detected at two and three sites in 2012 and 2014, respectively (Table 1-7).

While the Choptank River itself had not been sampled prior to 2016 (Table 1-8), O'Dell et al. (1980) reported Herring, White Perch, and Yellow Perch spawned in its drainage (Tuckahoe Creek) during 1976-1977 (Table 1-9). Twelve sites were sampled during 1976-1977 after installation of a fish ladder at the dam for the lake at Tuckahoe State Park. Sampling sites were established above and below the dam to determine the effectiveness of the fish ladder in passing anadromous and estuarine species (O'Dell et al. 1980). During 1976-1977, White Perch, Yellow Perch, and Herring were collected downstream of the dam/fishway, while White Perch were documented on the upstream side. O'Dell et al. (1980) noted that this species might have been trapped behind the dam when it was built and that its presence did not necessarily indicate successful migration through the fish ladder since no other species were documented on the upstream side. Sites in common between current sampling (2016-2017) and the O'Dell et al. (1980) study included the furthest downstream stations TUC101-TUC103 and the furthest upstream stations TUC108-TUC110 (Table 1-9). Herring spawning was detected at all sites sampled in 2017 with the exception of TUC109. A new fish ladder was installed in 1993 to replace the one referenced in O'Dell et al. (1980) and has been shown to pass Herring (J. Thompson, MD DNR, personal communication). White Perch spawning was detected in all but the two most upstream sites, both of which were located above the dam. In 2017, Yellow Perch spawning was detected at all sites below the dam, with the exception of TUC105, but not above the dam (Table 1-9).

Herring, White Perch, and Yellow Perch spawning during 2013-2017 occurred within the same reach of Patapsco River as that sampled by O'Dell et al. (1975; Figure 1-8, Table 1-10). Herring spawning was detected at all sites sampled in the Patapsco River in 2013-2017, with the exception of MBSS 593 in 2016. White Perch and Yellow Perch spawning was more variable, with spawning presence being detected in as few as one site, and as many as all sites, throughout the sampling period (Table 1-10).

Sites sampled in 2019 in the Chester River drainage match a subset of those sampled from 1975-1977 by O'Dell et al. (1980). Herring spawning was detected at a larger number of sites in 2019 than during 1975-1977, while White Perch spawning was detected at roughly the same number of sites, although locations differed, and Yellow Perch spawning detection decreased (Figure 1-9; Table 1-11).

Herring spawning was detected in the upper portion of Patuxent River at two of the six sites that matched O'Dell and Mowrer (1984) collections in 1980 (AFC10-4 and AFC3-165), and at one site (AFC3-163) where Herring spawning previously was not detected (Figure 1-10; Table 1-12). White Perch spawning was only detected in the upper portion of Patuxent River at half the number of sites as collections in 1980, and no Yellow Perch spawning was detected in this area in 2021, even though presence was noted at two sites by O'Dell and Mowrer (1984). Yellow Perch larvae were frequently encountered, however, during sampling of the Patuxent River Striped Bass spawning area during 2015-2016 (Uphoff et al. 2017). Results from samples collected in the middle portion of Patuxent River in 2021 are preliminary, with 59 out of 64 samples currently processed. Only two stations (AFC3-161 and site 1) in the middle potion of Patuxent River matched sampling locations of O'Dell and Mowrer (1984; Figure 1-10, Table 1-12). Herring spawning was detected in 2021 at these two locations, as well as the other four sites in the middle portion of Patuxent River. It should be noted, however, that 2021 ichthyoplankton collections were made at the same time DNR biologists were monitoring for, and making collections of, Hickory Shad adults in middle Patuxent River (MD DNR 2022). White Perch spawning was detected at five out of six middle Patuxent River sites in 2021, two of which matched presence detected in 1982 by O'Dell and Mowrer (1984). Yellow Perch spawning has not been detected in any of the samples processed so far for 2021, but was found at the two stations sampled by O'Dell and Mowrer (1984; Table 1-12).

The 90% confidence intervals of P_{herr} (Figure 1-12) 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 3 (1991, 2011, and 2013-2018). 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). Patapsco River was characterized by spawning at level 2 (2013 and 2017) and 3 (2014-2016). Deer Creek (2012-2015), Tuckahoe Creek (2016-2017), Choptank River (2016-2017), and Chester River (2019) are the least developed watersheds and were characterized by the highest level of Herring spawning (level 3) in all years sampled. In 2021 Herring spawning in the Patuxent River was characterized at level 3 (Figure 1-12). These categories may have been influenced by harvest regulations that we believe are having a positive effect on P_{herr}, through increased spawning stock (see below), and did not exclusively reflect level of development.

Estimates of P_{herr} increased in Bush River, and Mattawoman and Piscataway creeks during 2005-2018 (Figure 1-13). Counterintuitively, increases coincided with increased development for watersheds sampled before and after 2011; P_{herr} in Mattawoman Creek (C/ha increasing from 0.87 to 0.93) approached levels exhibited in streams in rural watersheds (P_{herr} as high as 0.78), while P_{herr} in developed Bush River and Piscataway Creek watershed streams (C/ha increasing from 1.37 to 1.52 and 1.41 to 1.50, respectively) increased to a lesser extent (to P_{herr} as high as 0.47; Figure 1-13). Remaining systems were sampled after 2011. Estimates of P_{herr} in Choptank and Chester rivers, and Deer and Tuckahoe creeks were high and steady through 2019 (0.62 to 0.87), while estimates for Patapsco River were lower and more variable. The estimate of P_{herr} in Patuxent River was high (Figure 1-13), however ichthyoplankton collections there were concurrent with monitoring and collections of spawning adult Hickory Shad (MD DNR 2022).

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 1-13). The relationship of C/ha with standardized median conductivity was linear, moderate, and positive ($r^2 = 0.35$, $P = 0.0001$, $N = 36$; Table 1-13; Figure 1-14). Estimates of P_{herr} were linearly, moderately, and negatively related to C/ha ($r^2 = 0.49$, $P < 0.0001$, $N = 37$; Figure 1-15). Negative linear and curvilinear (power function) regressions similarly described weak relationships of P_{herr} and standardized median conductivity ($r^2 = 0.16$, $P = 0.0171$; or approximate $r^2 = 0.15$, $P < 0.0001$, respectively), with linear regression explaining only slightly more variability ($N = 36$; Figure 1-15). Low estimates of P_{herr} (≤ 0.4) were much more frequent beyond the C/ha threshold (0.86 C/ha) or when standardized conductivity was 1.8-times or more than the baseline level (Figure 1-15). Estimates of P_{herr} were consistently above 0.6 in the four watersheds dominated by agriculture (Deer Creek, Tuckahoe Creek, Choptank River, and Chester River; Figure 1-15). The only watershed in this analysis dominated by forest cover was Mattawoman Creek and only one estimate (1991 at 62.6% forest cover and C/ha = 0.48) represented development below the C/ha threshold. The 1991 estimate of P_{herr} was above 0.6 and was consistent with watersheds dominated by agriculture. Remaining estimates for Mattawoman Creek were represented by 53.9% forest cover with C/ha increasing from 0.87 in 2008 to 0.97 in 2018. Samples were not collected in Mattawoman Creek in 2019, but it is the system with the longest data set. Additional analyses have been performed on these data in previous years; see Uphoff et al. (2019) for more information.

Plots of residuals against year for linear regressions of C/ha or standardized conductivity and P_{herr} indicated an increasing trend (Figure 1-16); residuals were all negative prior to 2011 and nearly all positive afterwards for either model. Predictions based on these models were likely to be biased.

The C/ha and spawning stock time category multiple regression explained 73% of variation in P_{herr} ($P < 0.0001$; Table 1-14). The intercept (mean = 0.51, SE = 0.08) and both coefficients (C/ha slope = -0.27, SE = 0.05; spawning stock coefficient = 0.31, SE = 0.06) were estimated with reasonable precision (CV < 30%). Predicted P_{herr} declined by 50% over the range of observed C/ha (0.07-1.52; Figure 1-17). Predicted P_{herr} increased by 60% between the two spawning stock categories (Table 1-14). Only the high spawning stock category contained estimates from the three land use types.

The standardized conductivity and spawning stock time category multiple regression explained 67% of variation in P_{herr} ($P < 0.0001$; Table 1-15). The intercept (mean = 0.64, SE = 0.12) and both coefficients (standardized conductivity slope = -0.29, SE = 0.07; spawning stock coefficient = 0.42, SE = 0.06) were estimated with reasonable precision (CV < 33%). Predicted P_{herr} declined by 50% over the range of observed standardized conductivity (1.14-2.42; Figure 1-17). Predicted P_{herr} increased by 66% between the two spawning stock categories (Table 1-15). Only the high spawning stock category contained estimates from all three land use types (Figure 1-17). Standardized median conductivities in excess of 1.75 were exclusively from watersheds categorized as urban. Higher standardized median conductivity (up to about 1.60) in agricultural and forested watersheds did not appear to be associated with distinctly lower P_{herr}; declines appeared concurrent with higher conductivity associated with urban development (Figure 1-17).

An increasing trend in residuals, evident in the simple linear regressions of P_{herr} against C/ha or standardized conductivity, was eliminated (or nearly so) for the multiple regressions that added a spawning stock size time category (Figure 1-18). Linear regressions of residuals from the multiple regressions and year in Figure 1-18 indicated a slight increasing trend over time was possible for standardized conductivity ($r^2 = 0.13$, $P = 0.03$) but unlikely for C/ha ($r^2 = 0.04$, $P = 0.24$). Cook's distance statistics identified 2011 as an outlier in both multiple regressions; the 2011 estimate of P_{herr} was more consistent with the high spawning stock (2012-2018) period than the low. This may have indicated some benefit by regulatory actions prior to the in-river fisheries deadline (2012; ASMFC 2019), including Atlantic coast bycatch reduction. Improved survival to maturity in response to declines in undescribed non-fishing related sources of at-sea losses (predation and feeding) could have contributed to increased spawning stock or supplied an alternative hypothesis to harvest reductions for the increase.

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 that accounted for shifting spawner abundance between 2005-2011 and 2012-2021, indicated significant and logical relationships among P_{herr} and C/ha consistent with the hypothesis that urbanization was detrimental to stream spawning. Predicted P_{herr} declined by 50% over the range of observed C/ha (0.07-1.52). 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.

Our preliminary data from Patuxent River suggested a localized impact of development on Herring spawning. Drift net stations, located above the tide line at Route 214 that were within or just below the developed Laurel-Bowie area had much lower P_{herr} (0.17; N = 36) than stations below this region in the tidal, more rural portion of the watershed that was sampled by boat (P_{herr} = 0.89; N = 47). The Choptank River, sampled in 2017, had a similar sampling design (drift nets upstream and boat samples downstream; Uphoff et al. 2018), but was subject to low development throughout the watershed. Estimated P_{herr} in Choptank River during 2017 was 0.74 (N = 43) upstream where drift nets were employed and 0.88 (N = 58) where boat samples were taken. We will explore this further in the next annual report with finalized data.

Higher standardized conductivity (up to about 1.6-times higher) in agricultural and forested watersheds did not appear to be associated with distinctly lower P_{herr}. Declines in P_{herr} appeared with higher conductivity in developing watersheds, suggesting that other urban stressors accompanied increasing conductivity. 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; Moore et al. 2017; Bird et al. 2018; Kaushal et al. 2018; Baker et al. 2019). Salt pollution and human-accelerated weathering have shifted the chemical composition of major ions in fresh water and increased salinization and alkalinization (freshwater salinization syndrome) across North America (Kaushal et al 2018). Coupled changes in conductivity, major ions, and pH began in the early and middle twentieth century and have influenced the water quality of most of the streams in the eastern United States. Densities of urban and agricultural land within a watershed can be strong predictors of base cations and pH in streams and rivers. In developed areas with colder climates, road salt is an important source of salinization. Agriculture can contribute significant bicarbonate and base cations from liming, potash, and fertilizer applications. Different mixtures of salt ions (such as sodium, bicarbonate, magnesium, sulfate, etc.) produce differential toxicity to aquatic life (Kaushal et al. 2018).

Uphoff et al. (2017) reported that there were strong, negative correlations between agricultural watershed percentages with C/ha; that forest cover and agriculture were strongly and negatively correlated; and that forest cover was poorly correlated with C/ha (Uphoff et al. 2017). The MD DOP forest cover estimate mixes forest cover in residential areas (trees over lawns) with true forest cover, clouding interpretation of forest influence. Uphoff et al. (2017) determined that subsequent analyses with P_{herr} beyond comparisons with C/ha were likely to be confounded by the close negative correlations, so we did not pursue statistical analyses with land uses other than C/ha. Our preference for using C/ha in analyses was two-fold: we have already done considerable work using C/ha, and C/ha provides a continuous rather than episodic time-series. However, we did note when these other land uses were predominant for particular P_{herr} outcomes. Estimates of P_{herr} were consistently high in watersheds dominated by agriculture, while importance of forest cover could not be assessed with confidence since it was possible that forest cover estimates included residential tree cover in Mattawoman Creek's watershed (our only forested watershed).

An unavoidable assumption of regression analyses of P_{herr}, C/ha, and standardized conductivity was that watersheds at different levels of development were a substitute for time-series. Extended time-series of watershed-specific P_{herr} 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 province effects in analyses with that variable. Differential changes in physical stream habitat and flow with urbanization, due to differences in geographic provinces, could also have influenced 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. Squared type II partial correlation coefficients for regressions of C/ha with P_{herr} were higher (0.40; Table 1-14) than for standardized conductivity (0.33; Table 1-15), possibly reflecting the wider coverage of stressors by C/ha.

Liess et al. (2016) developed a stress addition model for meta-analysis of toxicants that combined additional stressors of aquatic vertebrates and invertebrates and found that the presence of multiple environmental stressors could amplify the effects of toxicants 100-fold. This general concept may offer an explanation for the difference in fit of P_{herr} with C/ha and median standardized conductivity, with conductivity accounting for water quality and C/ha accounting for multiple stressors. This concept may also warn against expectations of overcoming Herring spawning stream habitat deterioration due to development through stringent management of directed fisheries and bycatch. An underlying negative relationship of P_{herr} with C/ha was present, but only described how the spatial and temporal distribution of earliest life stages of Herring may be impacted. Increasingly frequent poor juvenile indices of Blueback Herring and Alewife in the urbanizing Patuxent River after the late 1990s did not indicate that increased spawning stock has overcome deterioration of habitat (Uphoff et al. 2018).

Based on a simple plot and linear regression of C/ha and P_{herr}, it appeared that spawning both declined and became more variable as development increased. However, increasing variability likely was an artifact of increasing spawning stock size with time. Once a time category term, that we assumed accounted for changing spawner abundance, was added to the P_{herr} and C/ha regression, the variability about the predicted slopes was reduced considerably. Maryland closed its Herring fisheries in 2011, and most other in-river fisheries along the Atlantic Coast were closed by 2012 (AFMFC 2019). Caps on Herring bycatch in Atlantic Herring and Atlantic Mackerel fisheries were also implemented in 2014 (MAFMC 2019), and estimates of P_{herr} increased concurrently with these reductions.

The 2017 ASMFC River Herring stock assessment update indicated that 16 stocks experienced increasing abundance, two experienced decreasing abundance, eight experienced stable abundance, and 10 did not experience a discernable trend in abundance over the final 10 years of the times series (2006-2015; ASMFC 2019). Long-term monitoring of adult Blueback Herring and Alewife during spawning runs in the Nanticoke River, however, has not indicated an increase in recent years (Bourdon and Jarzynski 2020), but Herring may have increased in the Head-of-Bay region (Bourdon 2022).

Urbanization and physiographic province both affect discharge and sediment supply of streams (Paul and Meyer 2001; Cleaves 2003). These, in turn, could affect location, substrate composition, and extent and success of spawning. Processes such as flooding, riverbank erosion, and landslides vary by geographic province (Cleaves 2003) and influence physical characteristics of anadromous fish spawning streams. Coastal Plain streams have slow flows and sand or gravel bottoms (Boward et al. 1999). Unconsolidated layers of sand, silt, and clay underlie the Coastal Plain, with broad plains of low relief and wetlands characterizing the natural terrain (Cleaves 2003). Most Piedmont streams are of moderate slope with rock or bedrock bottoms (Boward et al. 1999), and the region is underlain by metamorphic rocks and characterized by narrow valleys and steep slopes, with regions of higher land between streams in the same drainage. 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.

Alewife spawn in sluggish flows, while Blueback Herring spawn in sluggish to swift flows (Pardue 1983). American Shad selected spawning habitat based on macrohabitat features (Harris and Hightower 2011) and spawned in moderate to swift flows (Hightower and Sparks 2003). Spawning substrates for Herring include gravel, sand, and detritus (Pardue 1983), and these can be impacted by development. Strong impacts of urbanization on lithophilic spawners include loss of suitable substrate, increased embeddedness, lack of bed stability, and siltation of interstitial spaces (Kemp 2014). Broadcasting species, such as Herring, could be severely affected since they do not clean substrate during spawning or provide protection to eggs and larvae in nests (Kemp 2014). Urbanization affects the quality and quantity of organic matter, another source of spawning substrate (detritus) in streams (Pardue 1983; Paul and Meyer 2001) that feed into subestuaries. While organic matter may be positively impacted by nutrients, it can also be negatively impacted by fine sediment from agriculture (Piggot et al. 2015).

Elevated conductivity, related primarily to chloride from road salt (although it includes 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; Kaushal et al. 2018; Baker et al. 2019). Use of salt as a deicer may lead to both "shock loads" of salt that may be acutely toxic to freshwater biota, as well as 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 of spawning adults and disrupt 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 Blueback 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. Sodium chloride is the dominant form of salt pollution with freshwater salinization syndrome, but increases in different mixtures of salt ions such as bicarbonate, magnesium, sulfate, etc., are part of the syndrome (Kaushal et al. 2018).

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 assumed that spawning sites detected in the 1970s and 1980s indicated the extent of habitat. O'Dell et al. (1975; 1980) and O'Dell and Mowrer (1984) summarized spawning activity as the presence of any species group's egg, larva, or adult (latter from wire fish trap sampling) for all samples 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 and early 1980s 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. 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 with Herring (P_{herr}) incorporated spatial and temporal presence-absence and provided an economical, precise, alternative to the O'Dell et al. (1975; 1980) and O'Dell and Mowrer (1984) estimates 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; Ward et al. 2017), 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 would 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.

Volunteer-based sampling of stream spawning during 2005-2019 used only stream drift nets, while O'Dell et al. (1975; 1980), O'Dell and Mowrer (1984), 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; Uphoff 1997).

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 these 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 and their watershed size, Department of Planning (DOP) land use designation and estimates of land use types, and level of development (C/ha) during years sampled. DOP Year = the year DOP estimated land use that best matches sample year. Bush (w/o APG) refers to the portion of the Bush River watershed not including Aberdeen Proving Grounds.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	Watershed Size (ha)	Primary Land Use
Bush (w/o APG)	2005	2002	1.37	25.4	35	36,009	Urban
Bush (w/o APG)	2006	2002	1.41	25.4	35		
Bush (w/o APG)	2007	2010	1.43	18	29.9		
Bush (w/o APG)	2008	2010	1.45	18	29.9		
Bush (w/o APG)	2014	2010	1.52	18	29.9		
Chester	2019	2010	0.13	65.9	24.8	77,751	Agriculture
Choptank	2016	2010	0.18	55	27.8	38,285	Agriculture
Choptank	2017	2010	0.18	55	27.8		
Deer	2012	2010	0.24	44.6	28.4	37,724	Agriculture
Deer	2013	2010	0.24	44.6	28.4		
Deer	2014	2010	0.24	44.6	28.4		
Deer	2015	2010	0.24	44.6	28.4		
Mattawoman	1991	1994	0.48	13.8	62.6	24,430	Forest
Mattawoman	2008	2010	0.87	9.3	53.9		
Mattawoman	2009	2010	0.88	9.3	53.9		
Mattawoman	2010	2010	0.90	9.3	53.9		
Mattawoman	2011	2010	0.91	9.3	53.9		
Mattawoman	2012	2010	0.90	9.3	53.9		
Mattawoman	2013	2010	0.91	9.3	53.9		
Mattawoman	2014	2010	0.93	9.3	53.9		
Mattawoman	2015	2010	0.94	9.3	53.9		
Mattawoman	2016	2010	0.95	9.3	53.9		
Mattawoman	2017	2010	0.96	9.3	53.9		
Mattawoman	2018	2010	0.97	9.3	53.9		

Table 1-1 cont.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	Watershed Size (ha)	Primary Land Use
Patapsco	2013	2010	1.11	24.4	30.4	93,730	Urban
Patapsco	2014	2010	1.12	24.4	30.4		
Patapsco	2015	2010	1.13	24.4	30.4		
Patapsco	2016	2010	1.14	24.4	30.4		
Patapsco	2017	2010	1.15	24.4	30.4		
Patuxent	2021	2010	1.34	20.5	35.1	100,181	Urban
Piscataway	2008	2010	1.41	10	40.4	17,634	Urban
Piscataway	2009	2010	1.43	10	40.4		
Piscataway	2012	2010	1.47	10	40.4		
Piscataway	2013	2010	1.49	10	40.4		
Piscataway	2014	2010	1.50	10	40.4		
Tuckahoe	2016	2010	0.07	66.6	25.4	39,364	Agriculture
Tuckahoe	2017	2010	0.07	66.6	25.4		

Table 1-2. Summary of subestuary watersheds sampled, years sampled, number of sites sampled, first and last dates of sampling, and stream ichthyoplankton sample sizes (N). Please note: (*) beside 2021 Patuxent indicates preliminary data.

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
Chester	2019	14	18-Mar	7-May	8	93
Choptank	2016	12	17-Mar	18-May	10	101
Choptank	2017	11	9-Mar	24-May	14	109
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
Mattawoman	2016	5	13-Mar	22-May	11	55
Mattawoman	2017	5	5-Mar	28-May	13	65
Mattawoman	2018	5	11-Mar	19-May	11	55
Patapsco	2013	4	19-Mar	30-May	22	40
Patapsco	2014	4	4-Apr	29-May	19	28
Patapsco	2015	4	25-Mar	28-May	18	32
Patapsco	2016	4	7-Mar	2-Jun	26	40
Patapsco	2017	4	9-Mar	6-Jun	21	40
Patuxent *	2021	12	18-Mar	9-Jun	18	95
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
Tuckahoe	2016	10	16-Mar	16-May	12	97
Tuckahoe	2017	10	8-Mar	23-May	11	102

Table 1-3. Summary statistics of conductivity (µS/cm) for mainstem stations in Deer, Mattawoman, Piscataway, and Tuckahoe creeks, and Bush, Chester, Choptank, Patapsco, and Patuxent rivers during 2005-2021. Unnamed tributaries were excluded from analysis. Tinkers Creek was included with mainstem stations in Piscataway Creek. Please note: (*) beside 2021 Patuxent indicates preliminary data.

	Bush					Chester	Choptank		Deer			
Conductivity	2005	2006	2007	2008	2014	2019	2016	2017	2012	2013	2014	2015
Mean	269.5	206.3	262.5	236.5	276.7	175.8	130.7	129.7	174.9	175.6	170.3	191.8
Standard Error	25.4	5	16	6.1	15.0	4.0	1.4	1.0	1.02	1.5	1.4	0.9
Median	229.5	208.1	218.7	233.9	253.4	181.5	133.2	129.8	176.8	177.7	171.7	193.5
Kurtosis	38.2	2.3	22.5	6.5	3.2	-0.40	2.41	-0.05	17.22	13.88	9.21	7.43
Skewness	5.8	-0.7	3.8	0.1	1.6	-0.37	-1.07	-0.07	-3.78	-2.25	-2.42	-1.97
Range	1860.8	320.8	1082.5	425.2	605.6	164	89	49	39.3	122	66	51
Minimum	79.2	0.0	104.5	9.8	107.0	85	74	107	140.2	93	116	156
Maximum	1940.0	320.8	1187.0	435.0	712.6	249	163	156	179.5	215	183	207
Count	81.0	106.0	79.0	77.0	60.0	93	101	109	44	87	60	75

	Mattawoman										
Conductivity	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018
Mean	120.1	244.5	153.7	147.5	128.9	126.1	179.4	181.8	180.3	151.2	160.7
Standard Error	3.8	19.2	38	2.8	1.9	2.4	9.1	6.5	4.1	3.7	4.4
Median	124.6	211	152.3	147.3	130.9	126.5	165.8	172.5	188.8	150.2	165.5
Kurtosis	2.1	1.41	1.3	8.29	-0.26	5.01	0.33	1.49	-0.80	-0.55	2.99
Skewness	-1.41	1.37	0.03	1.72	-0.67	-1.70	1.00	1.33	-0.68	-0.36	-1.70
Range	102	495	111	117	49	96	261	185	93	102	120
Minimum	47	115	99	109	102	63	88	130	121	91	79
Maximum	148	610	210	225	151	158	350	315	214	193	198
Count	39	40	43	44	44	48	48	44	44	52	44

Table 1-3 cont.

	Patapsco					Patuxent	Piscataway					Tuckahoe	
Conductivity	2013	2014	2015	2016	2017	2021	2008	2009	2012	2013	2014	2016	2017
Mean	406.2	282.5	346.8	310.4	340.3	354.3	218.4	305.4	211.4	245	249.4	152.2	155.9
Standard Error	48.7	8.0	18.2	30.6	15.1	9.0	7.4	19.4	5.9	6.9	11.1	2.4	1.7
Median	304.9	279.5	324	262.7	310	363.1	210.4	260.6	195.1	238.4	230	159.6	160.5
Kurtosis	12.13	-0.24	5.04	17.97	2.22	0.90	-0.38	1.85	0.11	-0.29	2.56	-0.29	-0.18
Skewness	3.33	0.42	1.97	3.99	1.36	-0.97	0.75	1.32	0.92	0.73	1.50	-0.68	-0.61
Range	1554	166	487	1055	432	391	138	641	163	173	274	103	82
Minimum	245	219	216	188	175	103	163	97	145	181	174	85	103
Maximum	1799	385	703	1243	607	494	301	737	308	354	449	188	185
Count	40	28	32	40	40	91	29	50	44	44	36	97	102

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 2008-2018. 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.

	Year														
Station	1971	1989	1990	1991	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018
	Herring														
MC1	1	1	1	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	1	1	1
MC3	1			1	1	1	1	1	1	1	1	1	1	1	1
MC4	1			1	0	0	1	1	1	1	1	1	1	1	1
MUT3	1				0	0	0	1	1	1	1	1	1	0	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	1	1	
MC2	0	0	1	0	0	0	0	0	0	1	1	0	1	1	
MC3	1			0	0	0	0	0	0	0	0	0	1	0	
	Yellow Perch														
MC1	1	1	1	1	1	0	1	1	0	1	1	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.

	Year				
Station	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. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Choptank River during 2016-2017. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-6.

	Year					
Station	2016			2017		
	Herring	White Perch	Yellow Perch	Herring	White Perch	Yellow Perch
CH100	1	1	1	1	1	1
CH101	1	1	1	1	1	1
CH102	1	1	1	1	1	1
CH103	1	1	1	1	1	1
CH104	1	1	1	1	1	1
CH105	1	1	1	1	1	1
CH106	1	1	1	1	1	1
CH107	1	1	0	1	1	0
CH108	1	1	0	1	1	0
CH109	1	1	1	1	1	0
CH110	1	0	0	1	0	0
CH111	0	0	0			

Table 1-9. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Tuckahoe Creek during 1976-1977 and 2016-2017. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-7.

	Year		
Station	1976-77	2016	2017
	Herring		
TUC101	1	1	1
TUC102	1	1	1
TUC103	1	1	1
TUC104		1	1
TUC105		1	1
TUC106		1	1
TUC107		1	1
TUC108	0	1	1
TUC109	0	1	0
TUC110	0	0	1
	White Perch		
TUC101	1	1	1
TUC102	1	1	1
TUC103	1	1	1
TUC104		1	1
TUC105		1	1
TUC106		1	1
TUC107		1	1
TUC108	1	1	1
TUC109	0	0	0
TUC110	0	0	0
	Yellow Perch		
TUC101	1	1	1
TUC102	1	1	1
TUC103	1	1	1
TUC104		1	1
TUC105		1	0
TUC106		1	1
TUC107		1	1
TUC108	0	0	0
TUC109	0	0	0
TUC110	0	0	0

Table 1-10. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Patapsco River during 1973 and 2013-2017. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-8.

O'Dell Sampling (1973)	
Station	Herring
Inland 1	0
Inland 2	1
Inland 3	1
Inland 4	1
Inland 5	0
	White Perch
Inland 1	1
Inland 2	1
Inland 3	0
Inland 4	1
Inland 5	0
	Yellow Perch
Inland 1	1
Inland 2	0
Inland 3	0
Inland 4	0
Inland 5	1

	Year				
Station	2013	2014	2015	2016	2017
			Herring		
USFWS Down River	1	1	1	1	1
USFWS Upriver	1	1	1	1	1
MBSS 591	1	1	1	1	1
MBSS 593	1	1	1	0	1
			White Perch		
USFWS Down River	0	1	1	1	1
USFWS Upriver	1	1	1	1	1
MBSS 591	0	1	0	1	1
MBSS 593	0	0	0	0	0
			Yellow Perch		
USFWS Down River	1	1	1	1	1
USFWS Upriver	1	0	1	1	0
MBSS 591	0	0	0	1	0
MBSS 593	0	0	0	1	0

Table 1-11. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Chester River during 1975-1977 and 2019. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-9.

	Year	
Station	1975-77	2019
	Herring	
CH19001	0	1
CH19002	0	1
CH19003	1	1
CH19004	0	0
CH19005	1	1
CH19006	1	1
CH19007	0	1
CH19008	0	1
CH19009	1	1
CH19010	1	1
CH19011	1	1
CH19012	1	1
CH19014	1	1
CH19015	1	1
	White Perch	
CH19001	0	1
CH19002	0	1
CH19003	1	1
CH19004	1	0
CH19005	1	1
CH19006	1	1
CH19007	0	0
CH19008	0	0
CH19009	1	1
CH19010	1	1
CH19011	1	1
CH19012	1	0
CH19014	0	1
CH19015	1	1
	Yellow Perch	
CH19001	1	1
CH19002	1	0
CH19003	1	1
CH19004	0	0
CH19005	1	0
CH19006	1	0
CH19007	0	0
CH19008	0	0
CH19009	0	0
CH19010	0	0
CH19011	1	0
CH19012	0	0

CH19014	0	0
CH19015	1	0

Table 1-12. Site-specific presence-absence of Herring (Blueback Herring, Hickory Shad, and Alewife), White Perch, and Yellow Perch stream spawning in Patuxent River during 1980-1982 and 2021. 0 = site sampled, but spawning not detected; 1 = site sampled, spawning detected; and blank indicates no sample. Station locations are identified on Figure 1-10.

	Patuxent	
Station	1980-1982	2021
	Herring	
AFC3-163	0	1
AFC3-164	0	0
AFC3-165	1	1
AFC3-114	1	0
AFC10-4	1	1
AFC10-8	1	0
AFC3-188		1
AFC3-161	1	1
1	1	1
2		1
3		1
4		1
	White Perch	
AFC3-163	1	1
AFC3-164	1	1
AFC3-165	1	1
AFC3-114	1	0
AFC10-4	1	0
AFC10-8	1	0
AFC3-188		0
AFC3-161	1	1
1	1	1
2		1
3		1
4		1
	Yellow Perch	
AFC3-163	1	0
AFC3-164	1	0
AFC3-165	0	0
AFC3-114	0	0
AFC10-4	0	0
AFC10-8	0	0
AFC3-188		0
AFC3-161	1	0
1	1	0
2		0
3		0
4		0

Table 1-13. 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	P	
Regression	1	1.68691	1.68691	18.47	0.0001	
Residual	34	3.10543	0.09134			
Total	35	4.79234				
$r^2 = 0.3520$						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	1.19567	0.10938	10.93	<.0001	0.97338	1.41796
C / ha	0.45274	0.10535	4.30	0.0001	0.23865	0.66683

Linear Model	Proportion of samples with herring eggs or larvae (P_{herr}) = Structure density (C/ha)					
ANOVA	df	SS	MS	F	P	
Regression	1	1.2426	1.2426	34.05	<.0001	
Residual	35	1.27735	0.0365			
Total	36	2.51996				
$r^2 = 0.4931$						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.83881	0.06763	12.40	<.0001	0.70152	0.97611
C / ha	-0.38417	0.06584	-5.84	<.0001	-0.51782	-0.25051

Linear Model	Proportion of samples with herring eggs or larvae (P_{herr}) = Standardized conductivity					
ANOVA	df	SS	MS	F	P	
Regression	1	0.38707	0.38707	6.29	0.0171	
Residual	34	2.0915	0.06151			
Total	35	2.47857				
$r^2 = 0.1562$						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.94215	0.18736	5.03	<.0001	0.56139	1.3229
Standardized conductivity	-0.2842	0.1133	-2.51	0.0171	-0.51444	-0.05395

Table 1-14. Summary statistics of the multiple regression model for development level (C/ha) and spawning stock time category versus proportion of samples with Herring eggs and-or larvae (P_{herr}).

ANOVA				Multiple Regression		
Source	df	SS	MS	F	P	
Regression	2	1.8092	0.9046	44.6	<.0001	
Residual	33	0.66937	0.02028			
Total	35	2.47857				
$r^2 = 0.7299$						
	Estimate	SE	t Stat	P-value	Squared Partial Corr Type I	Squared Partial Corr Type II
Intercept	0.51158	0.0786	6.51	<.0001	.	.
C / ha	-0.27453	0.05341	-5.14	<.0001	0.48518	0.44463
Time category	0.3118	0.05701	5.47	<.0001	0.47542	0.47542

Table 1-15. Summary statistics of the multiple regression model for standardized conductivity (annual median/province background) and spawning stock time category versus proportion of samples with Herring eggs and-or larvae (P_{herr}).

ANOVA				Multiple Regression		
Source	df	SS	MS	F	P	
Regression	2	1.6702	0.8351	34.09	<.0001	
Residual	33	0.80837	0.0245			
Total	35	2.47857				
$r^2 = 0.6739$						
	Estimate	SE	t Stat	P-value	Squared Partial Corr Type I	Squared Partial Corr Type II
Intercept	0.64351	0.12522	5.14	<.0001	.	.
Standardized conductivity	-0.28779	0.0715	-4.03	0.0003	0.15617	0.32930
Time category	0.42151	0.05824	7.24	<.0001	0.61350	0.61350

Figure 1-1. Watersheds sampled for stream spawning anadromous fish eggs and larvae during 2005-2021. Coastal Plain and Piedmont Regions are indicated.

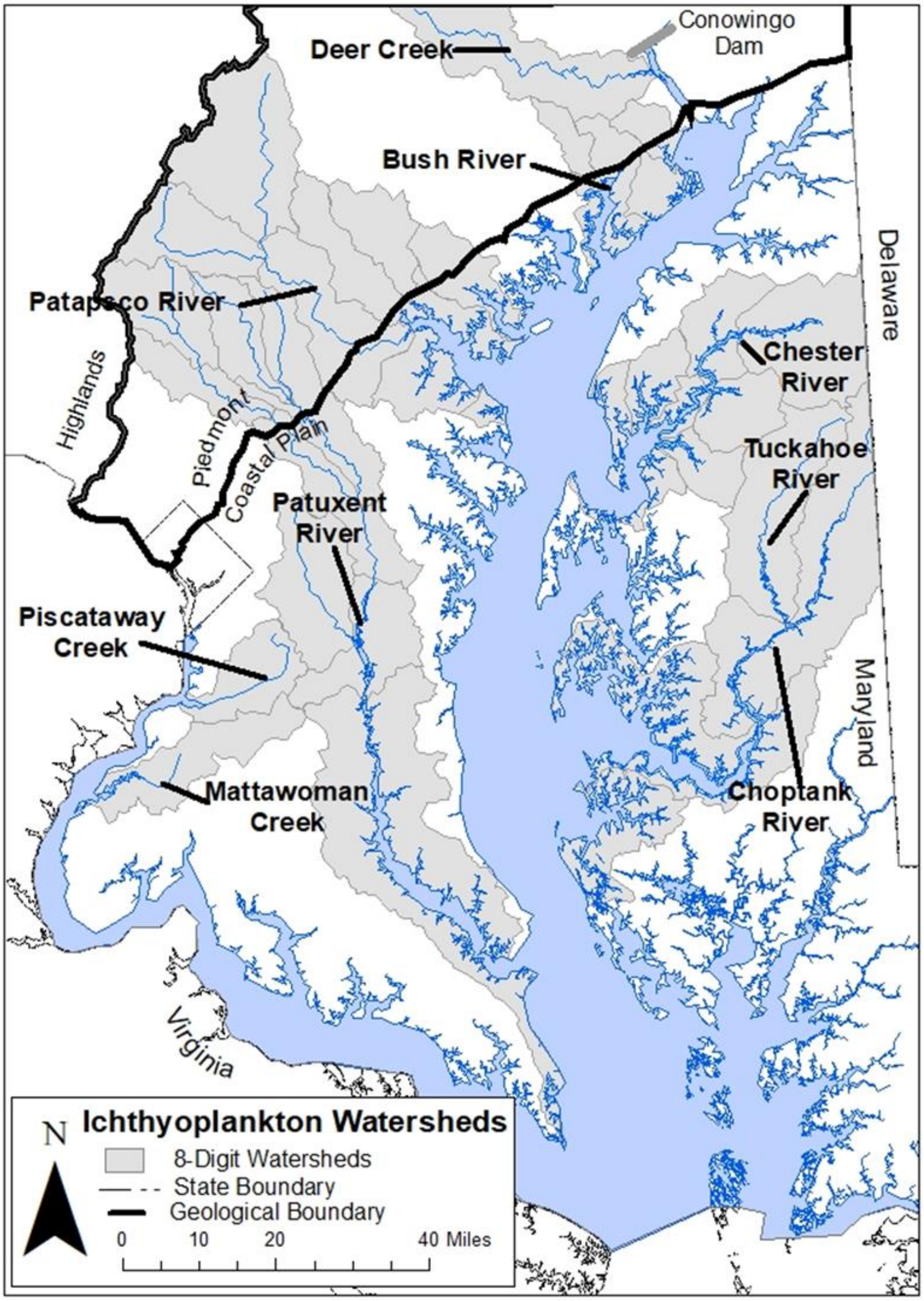


Figure 1-2. Mattawoman Creek's 1971 (O'Dell et al. 1975) and 2008-2018 sampling stations. Bar approximates lower limit of development associated with the town of Waldorf.

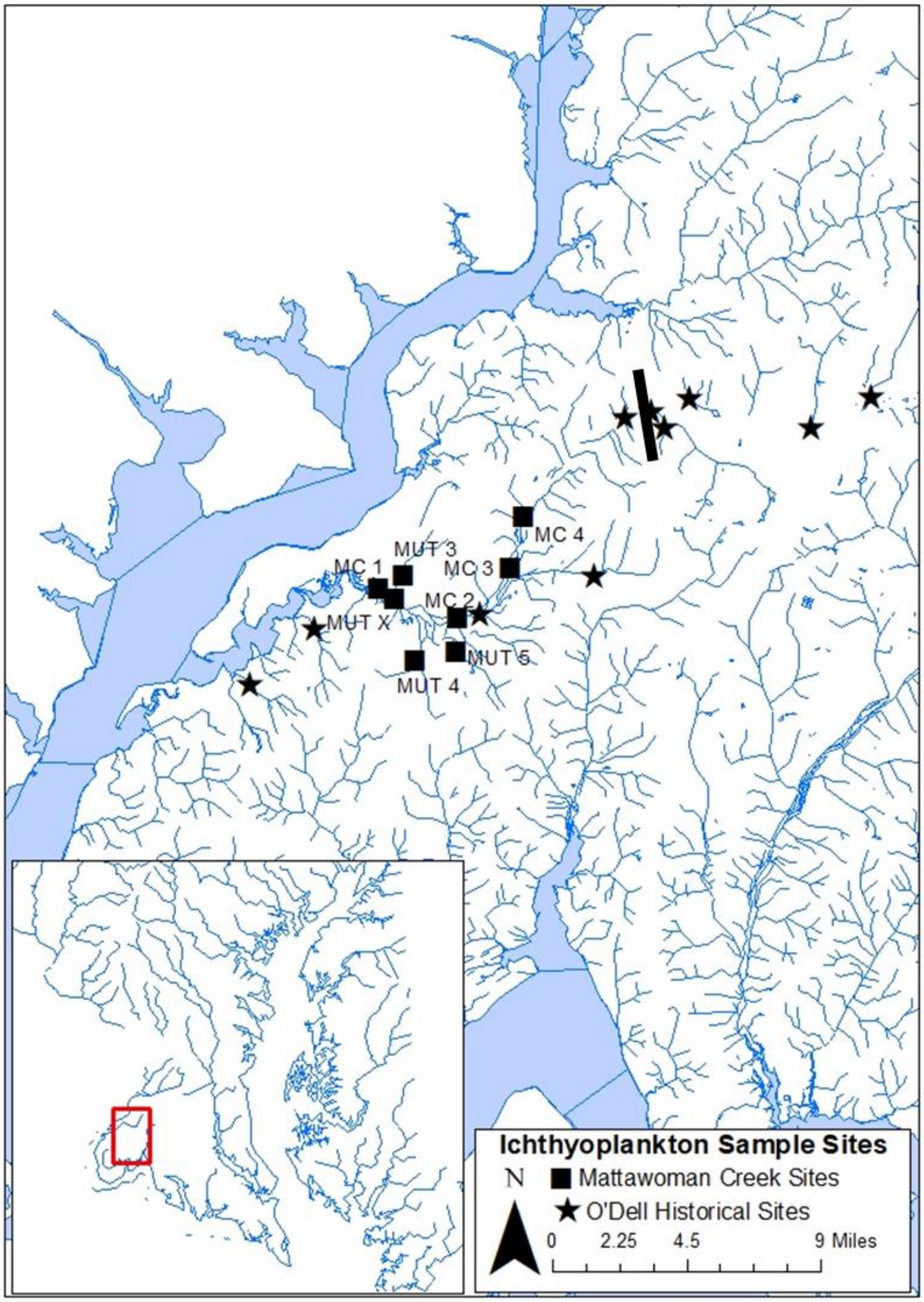


Figure 1-3. Piscataway Creek's 1971 (O'Dell et al. 1975), 2008-2009, and 2012-2014 sampling stations.

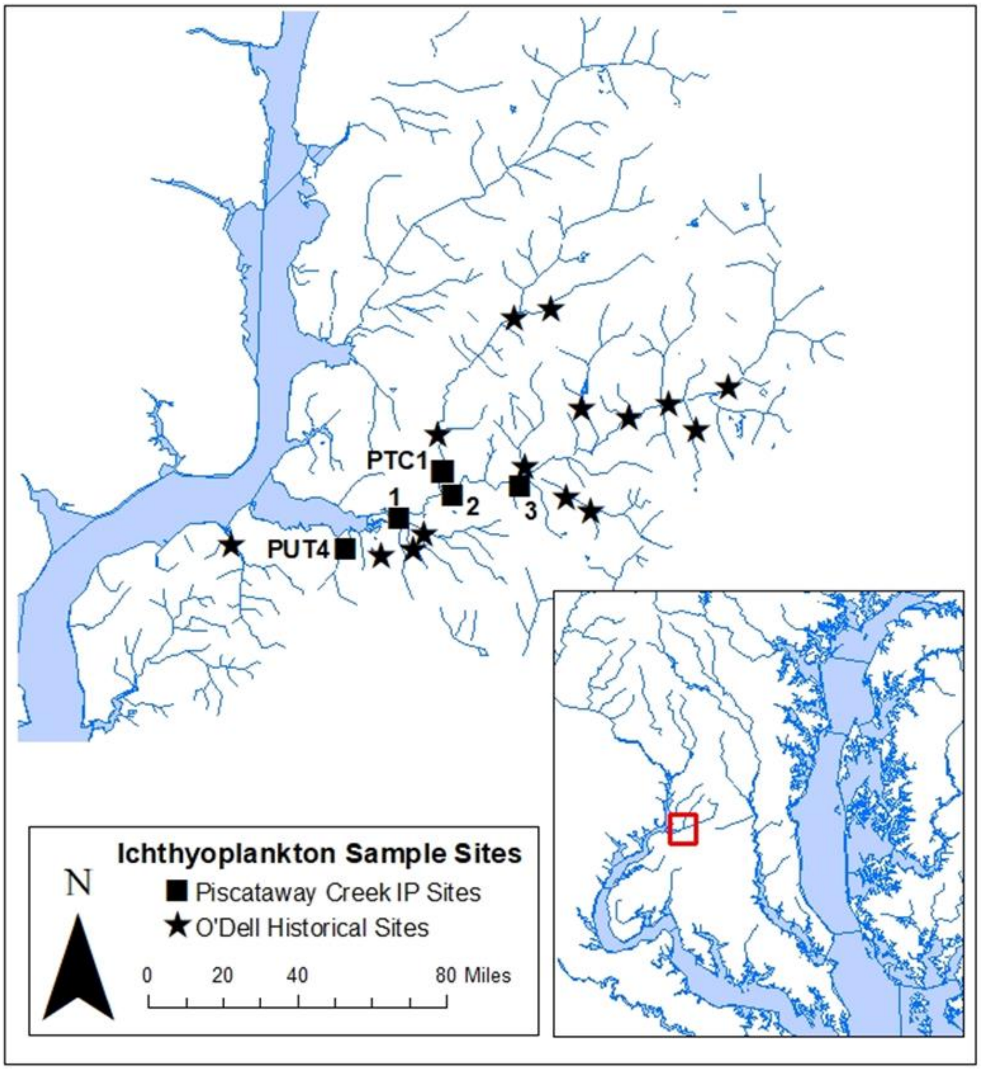


Figure 1-4. Bush River's 1973 (O'Dell et al. 1975), 2005-2008, and 2014 sampling stations. Stations in Aberdeen Proving Grounds (APG) have been separated from other Bush River stations. Line delineates APG streams that were excluded.

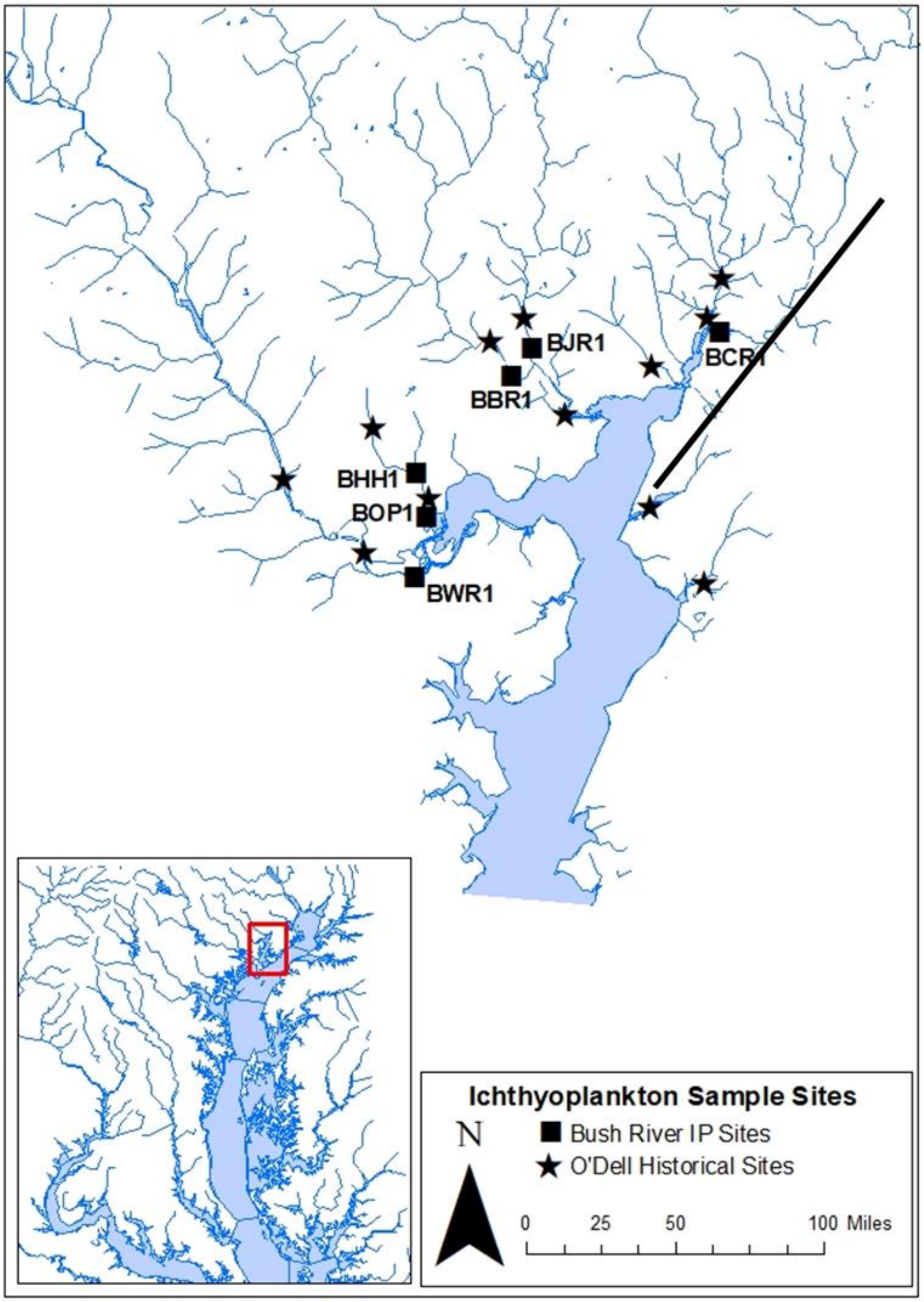
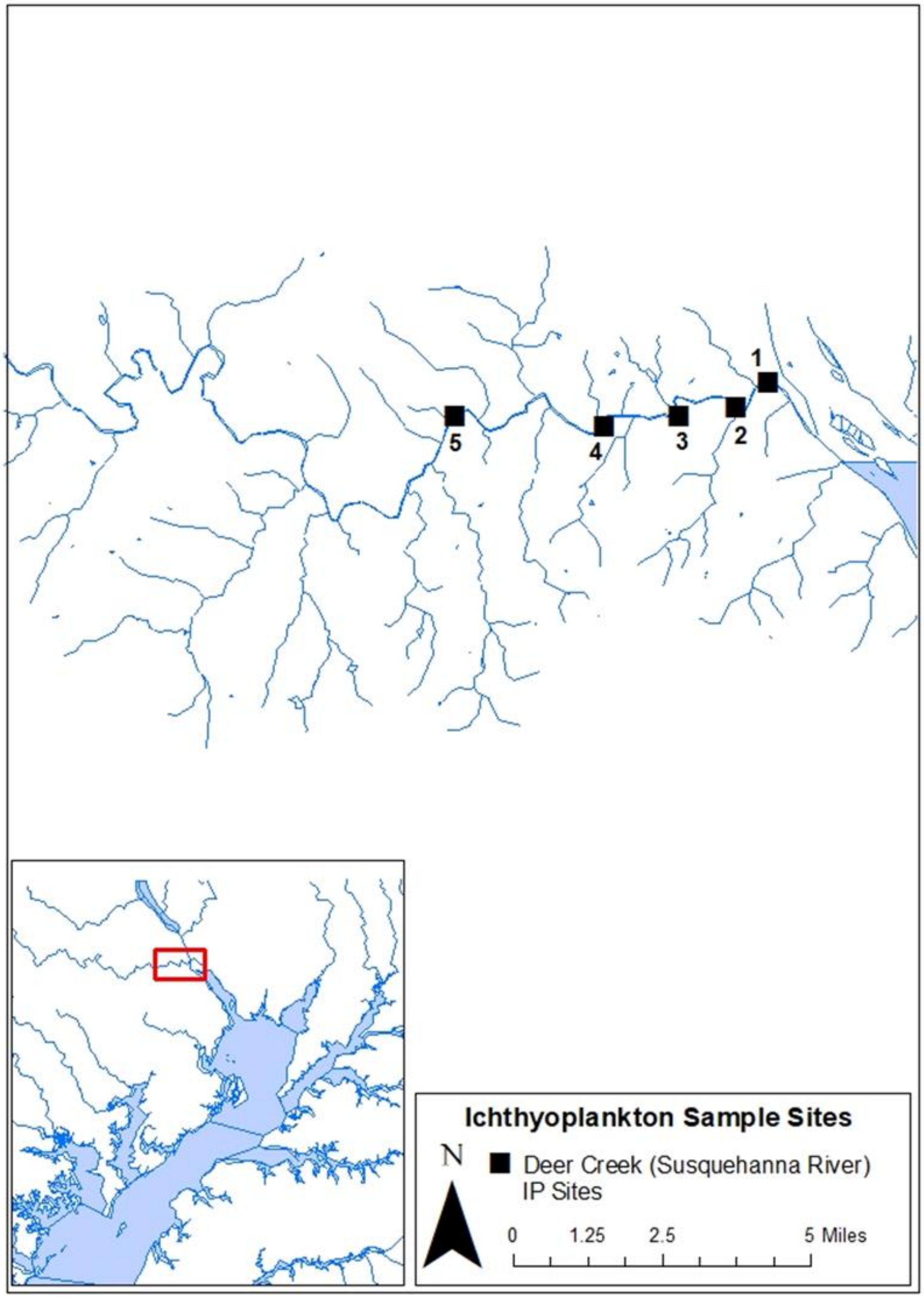


Figure 1-5. Deer Creek's 1972 (O'Dell et al. 1975) and 2012-2015 sampling stations.



Figures 1-6 and 1-7. Choptank River and Tuckahoe Creek's 2016-2017 sampling stations. Stars indicate sites only sampled by O'Dell et al. (1980). D = drift net and T = towed 0.5 m diameter net. stations TUC101, TUC102, TUC103, TUC108, TUC109, and TUC110 correspond to O'Dell et al. (1980) sites

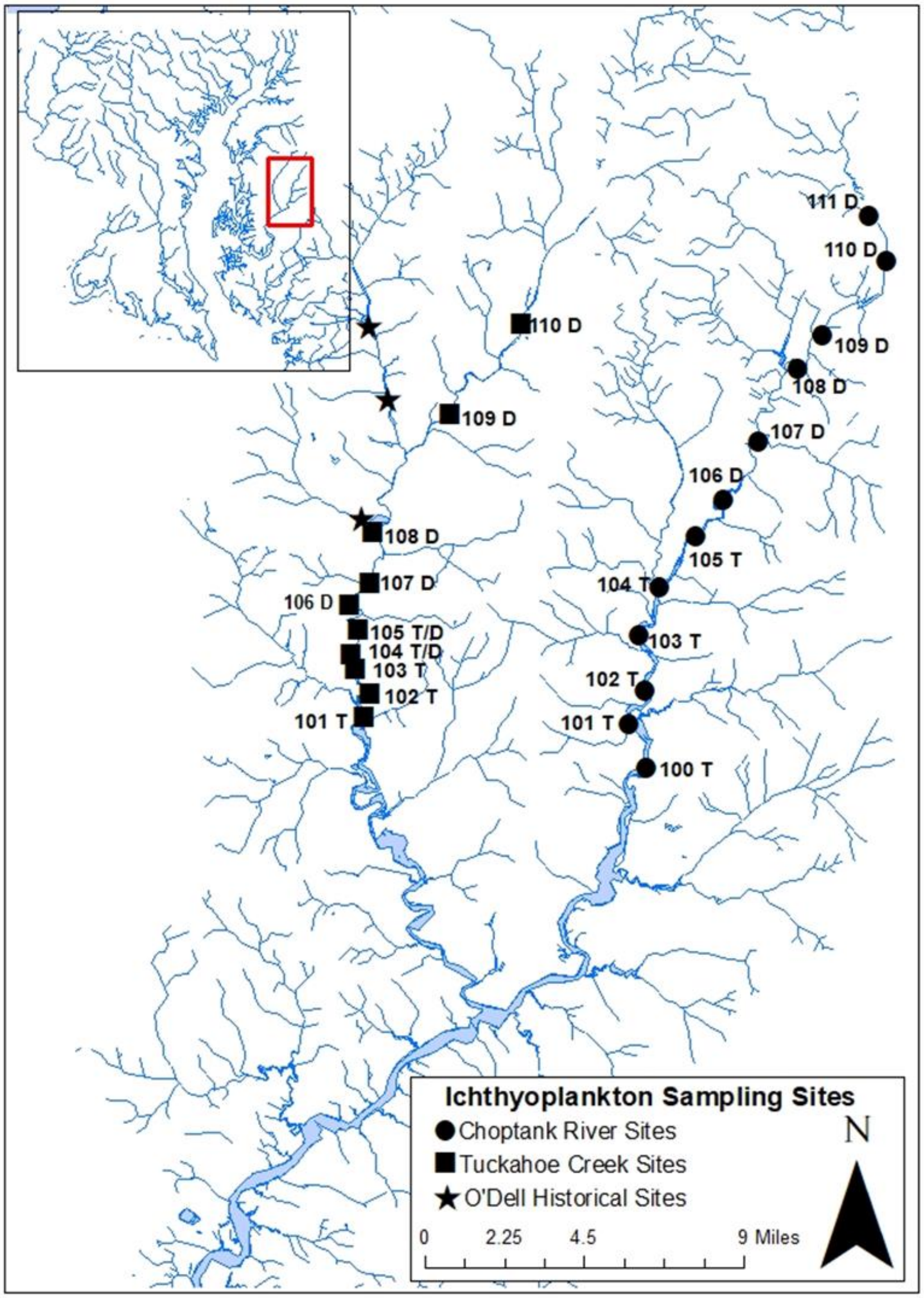


Figure 1-8. Patapsco River's 1973 (O'Dell et al. 1975) and 2013-2017 sampling stations.

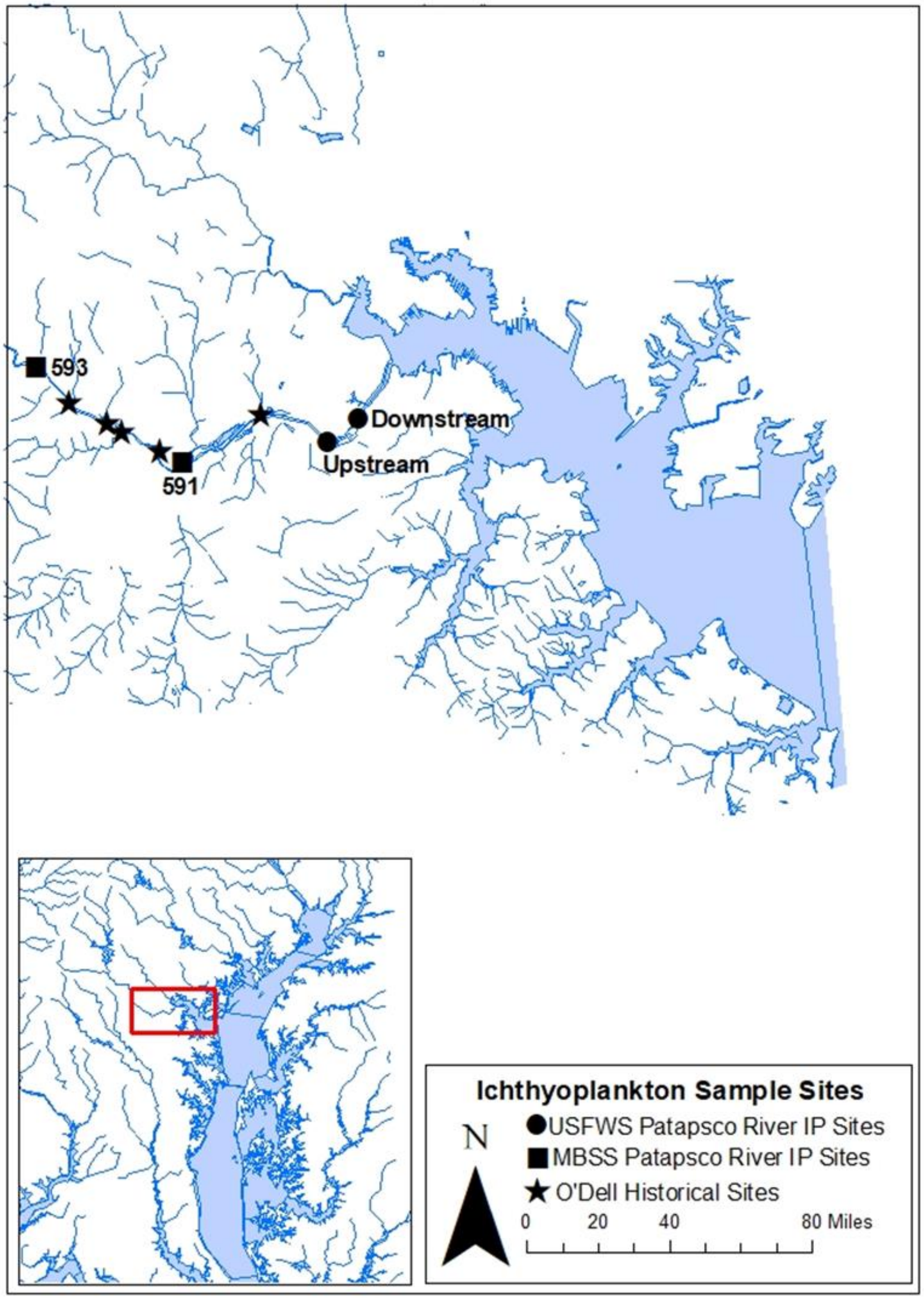


Figure 1-9. Chester River's 1975-1977 (O'Dell et al. 1980) and 2019 sampling stations.

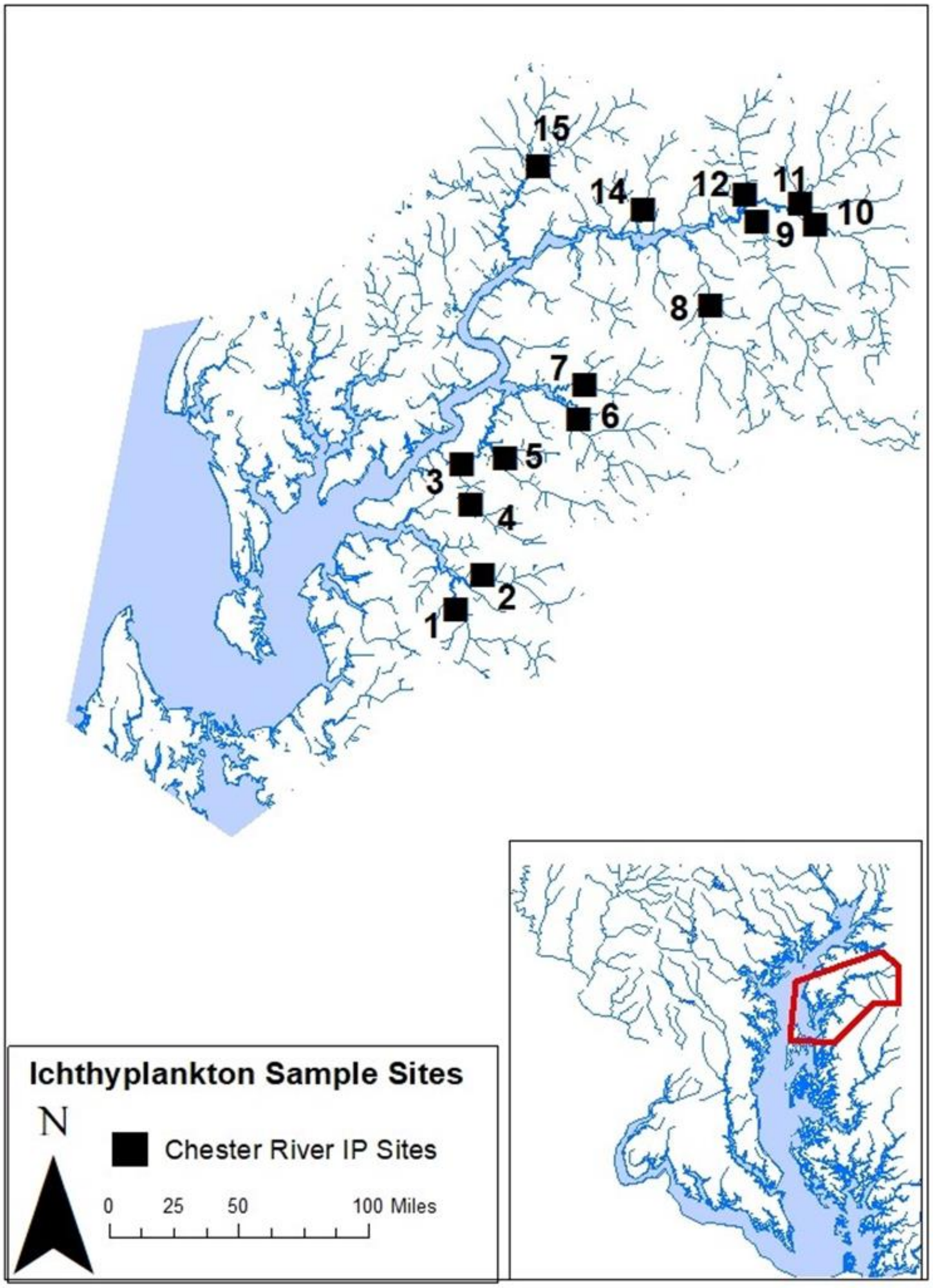


Figure 1-10. Patuxent River's 1980-1982 (O'Dell and Mowrer 1984) and 2021 sampling stations.

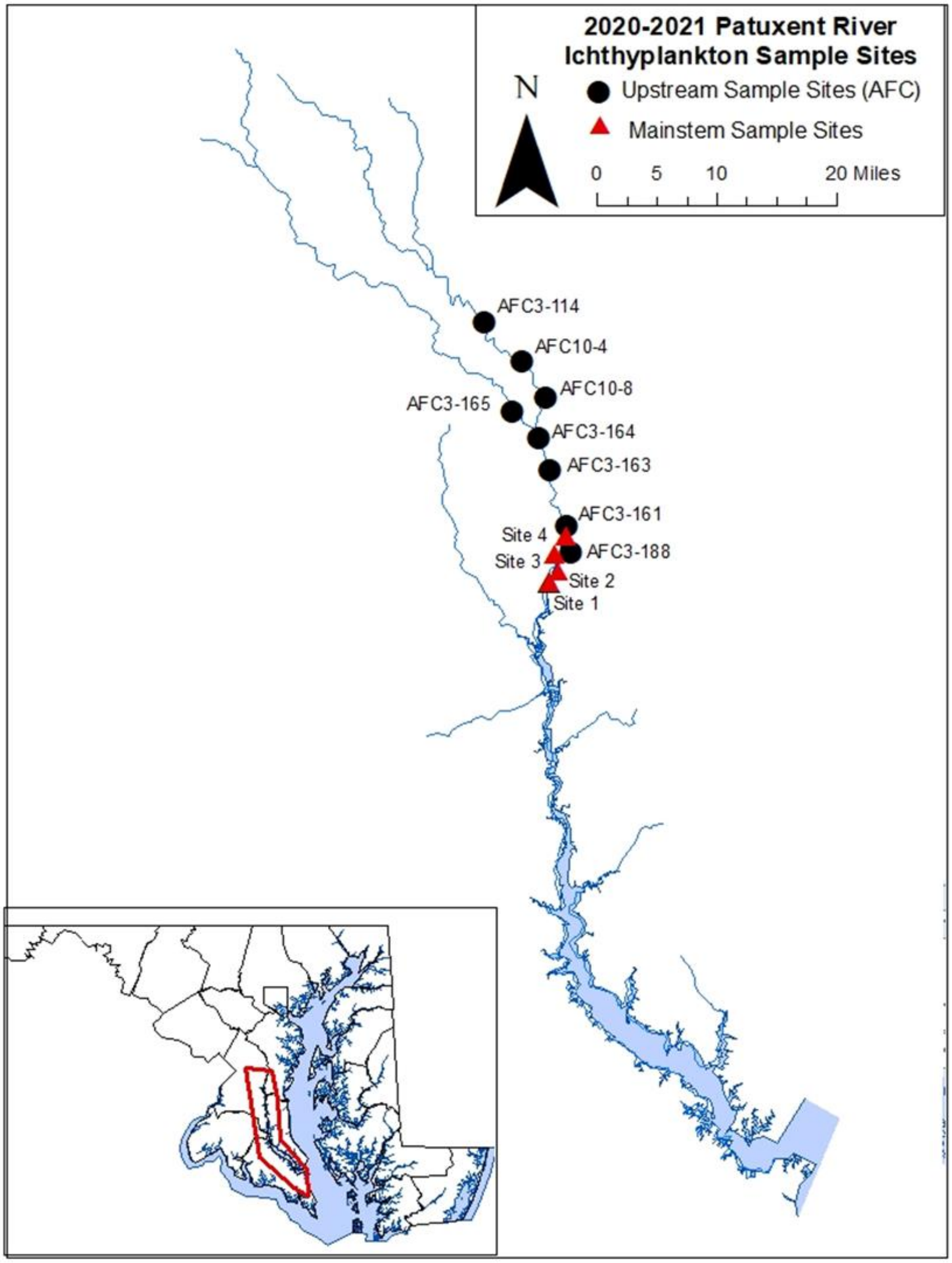


Figure 1-11. Trends in counts of structures per hectare (C/ha) during 1950-2021 in Deer, Mattawoman, and Piscataway creeks, Bush, Patapsco, and Patuxent rivers, and Chester and Choptank River drainages. Estimates of C/ha were only available to 2016 or 2020, depending on Department of Planning data updates. Large symbols indicate years when stream ichthyoplankton was sampled.

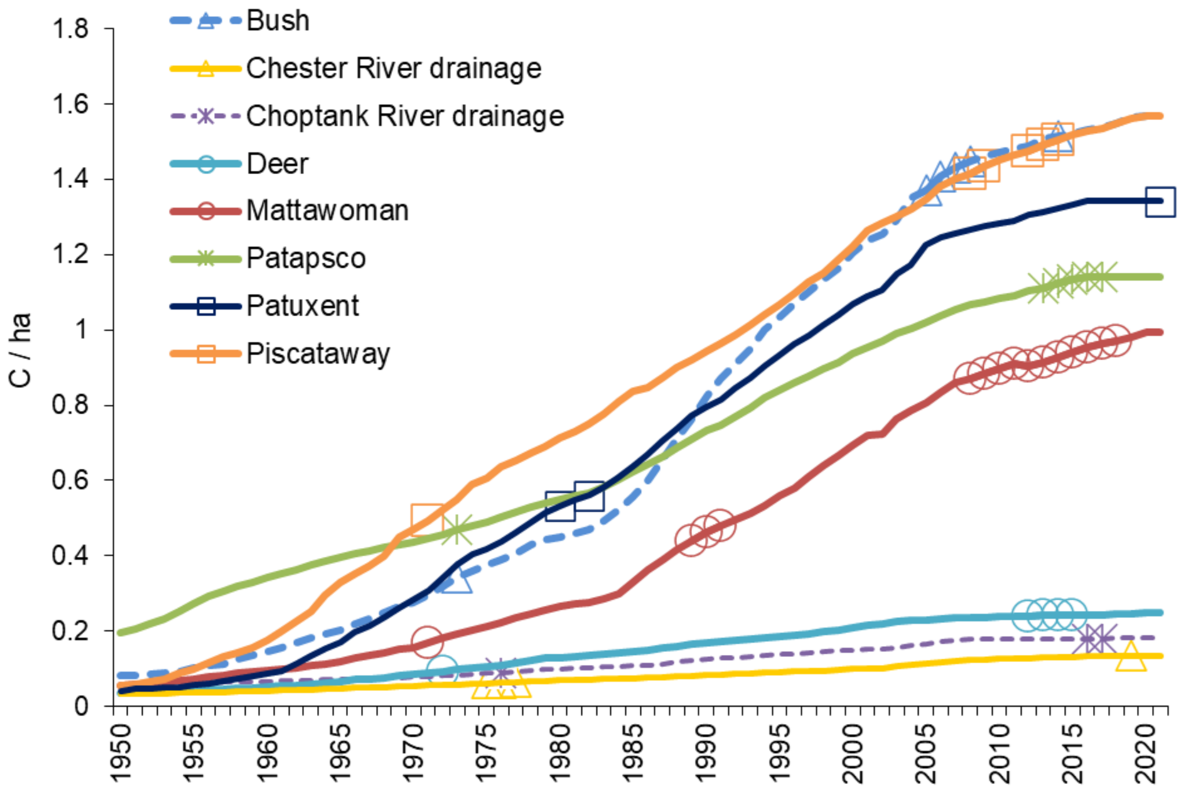


Figure 1-12. Proportion of samples (P_{herr}) with Herring and their 90% confidence intervals for stream ichthyoplankton surveys in Mattawoman, Piscataway, Deer, and Tuckahoe creeks, and Bush, Choptank, Patapsco, Chester, and Patuxent rivers.

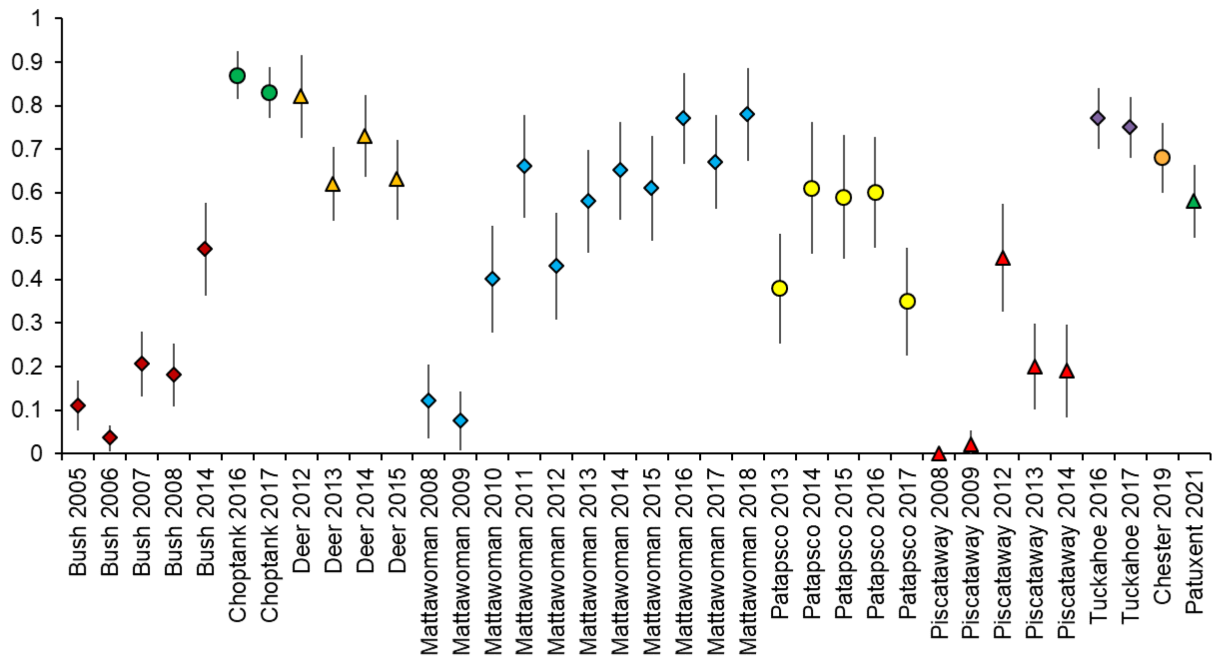


Figure 1-13. Trends in P_{herr} (proportion of stream samples with Herring eggs and-or larvae) by watershed. Watersheds sampled in both early (2005-2011) and late (2012-2021) spawning periods are indicated by large triangles.

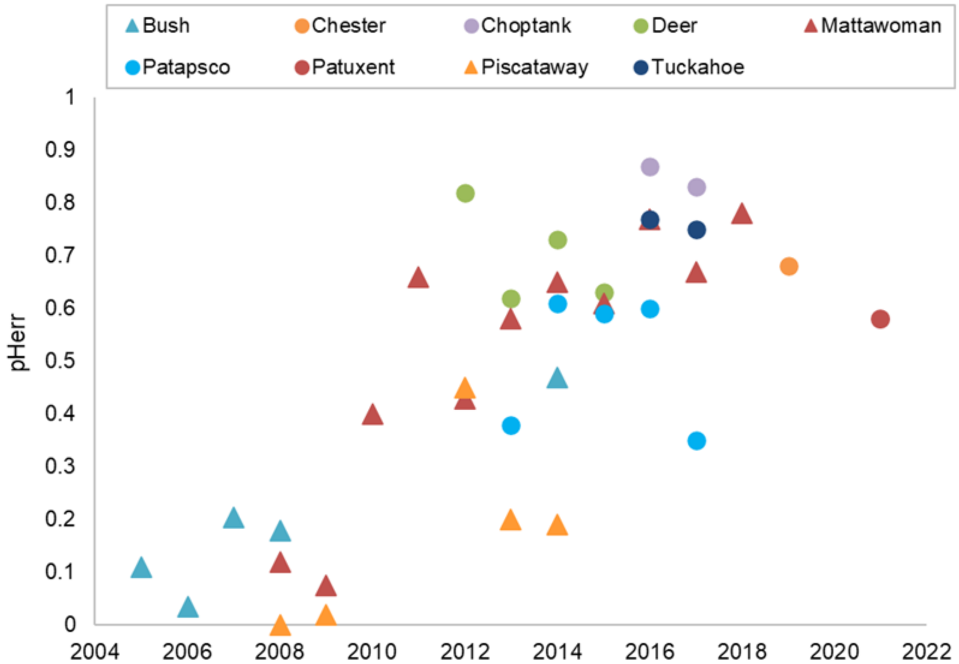


Figure 1-14. Standardized median conductivity during spring spawning surveys and level of development (C/ha) with dominant Department of Planning land use designations. Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).

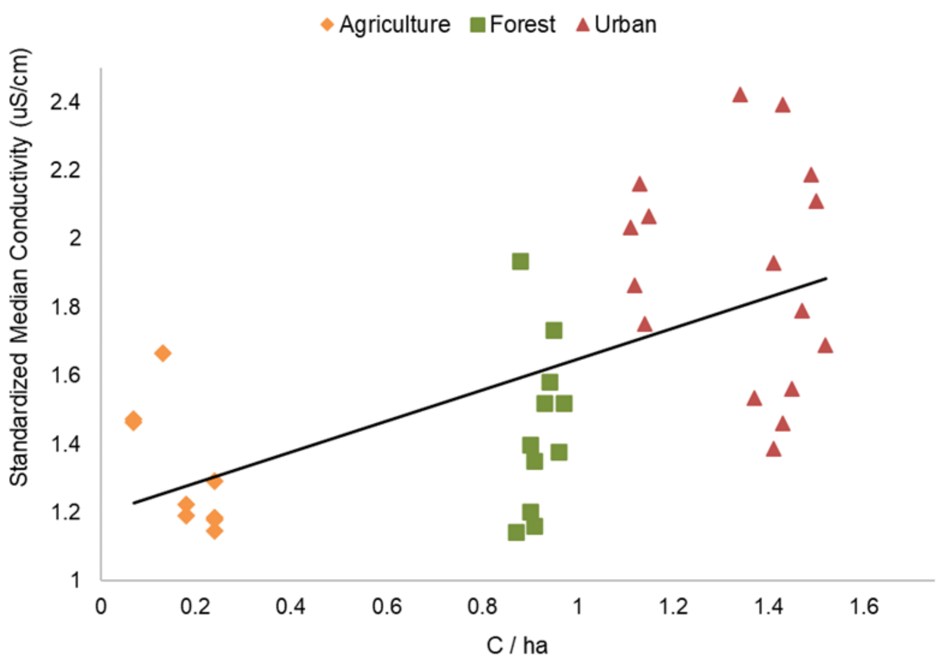


Figure 1-15. (A) Proportion of stream samples with Herring eggs and-or larvae (P_{herr}) and level of development (C/ha) with dominant Department of Planning land use designations. (B) P_{herr} and standardized median spawning survey conductivity (uS/cm). Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).

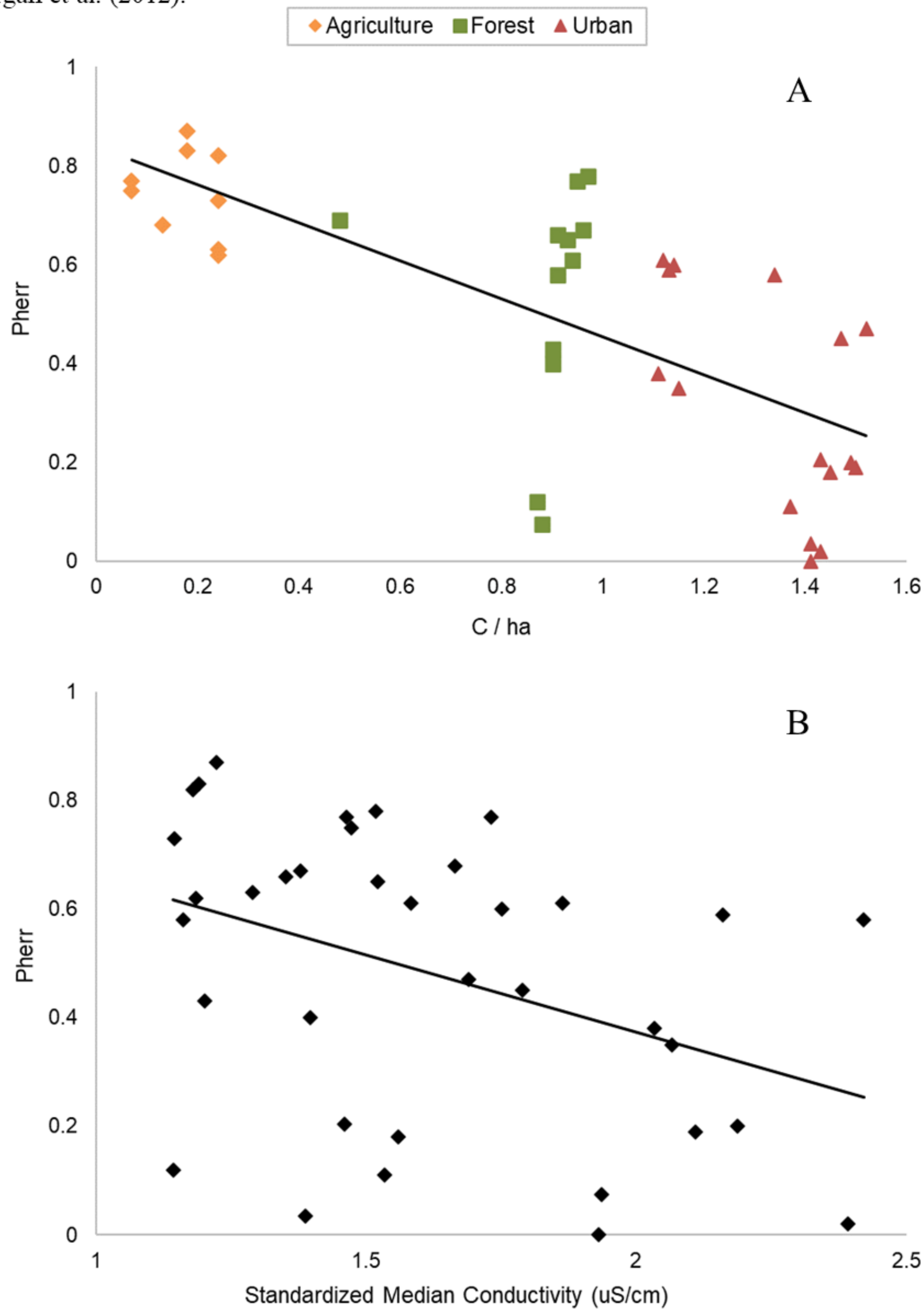


Figure 1-16. Serial patterns of residuals versus year for regressions of P_{herr} (proportion of stream samples with Herring eggs and-or larvae) and (A) level of development (C/ha) or (B) standardized median spawning survey conductivity (uS/cm). Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on Morgan et al. (2012).

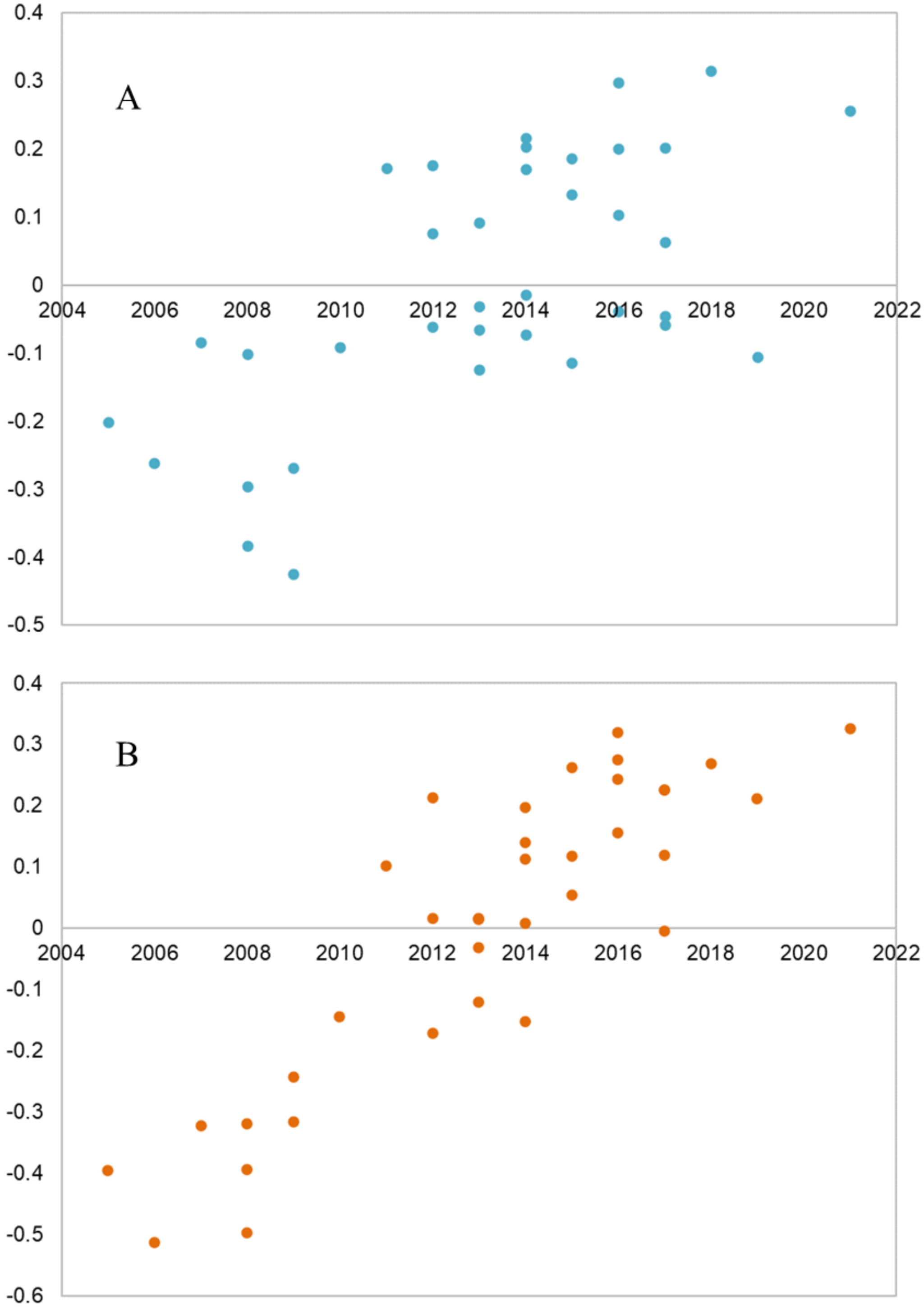
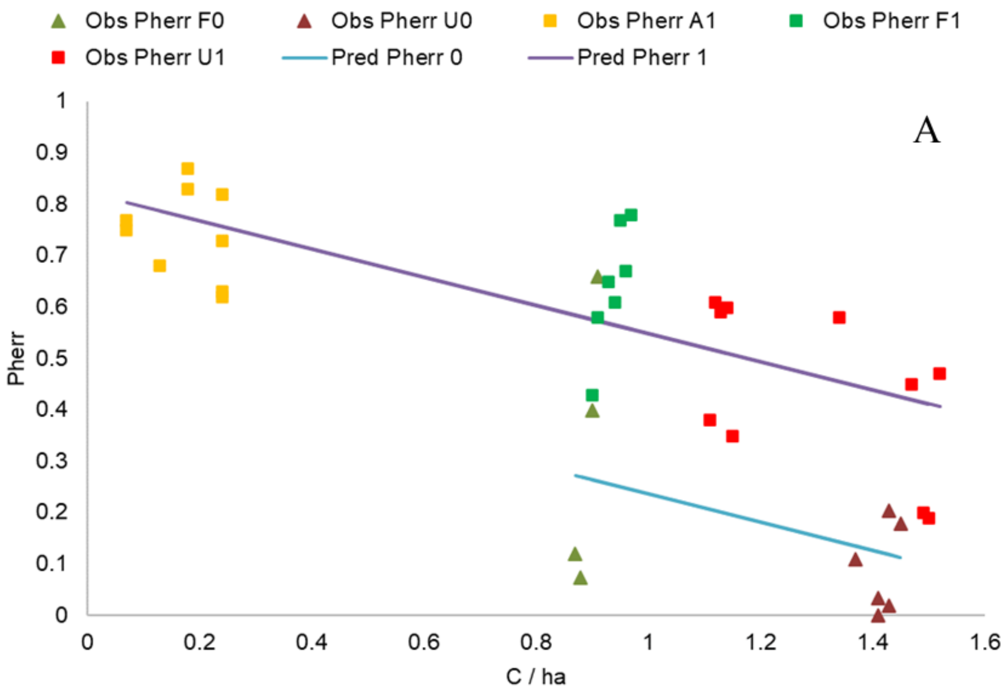


Figure 1-17. Plots of regressions of P_{herr} (proportion of stream samples with Herring eggs and-or larvae) against (A) level of development (C/ha) or (B) standardized median spawning survey conductivity (uS/cm) with spawning stock time categories (0 = 2005-2011; 1 = 2012-2021) included. Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).



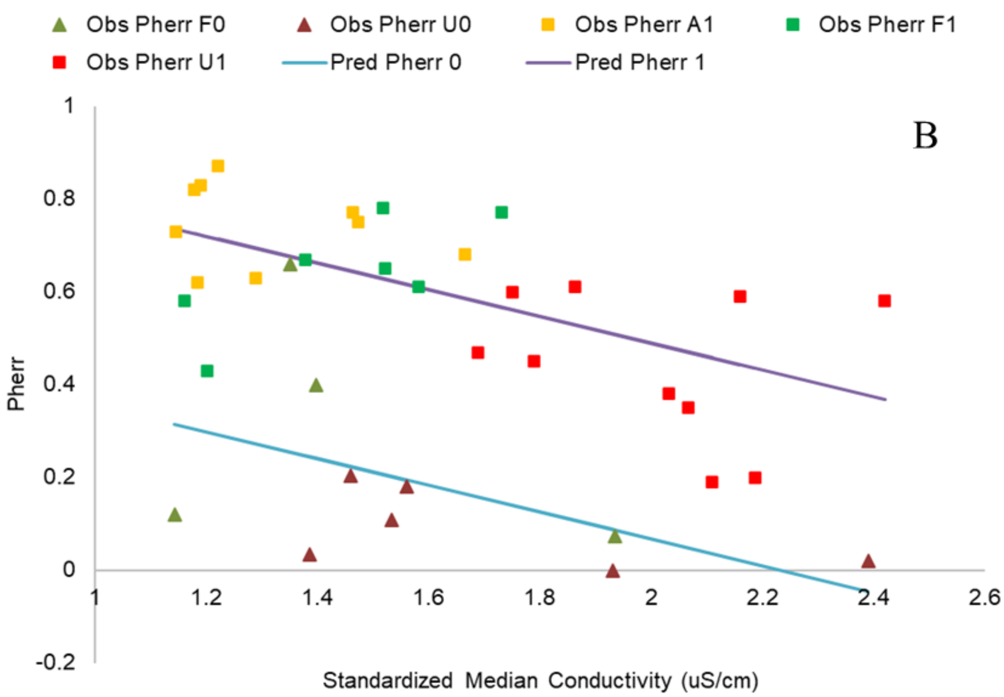
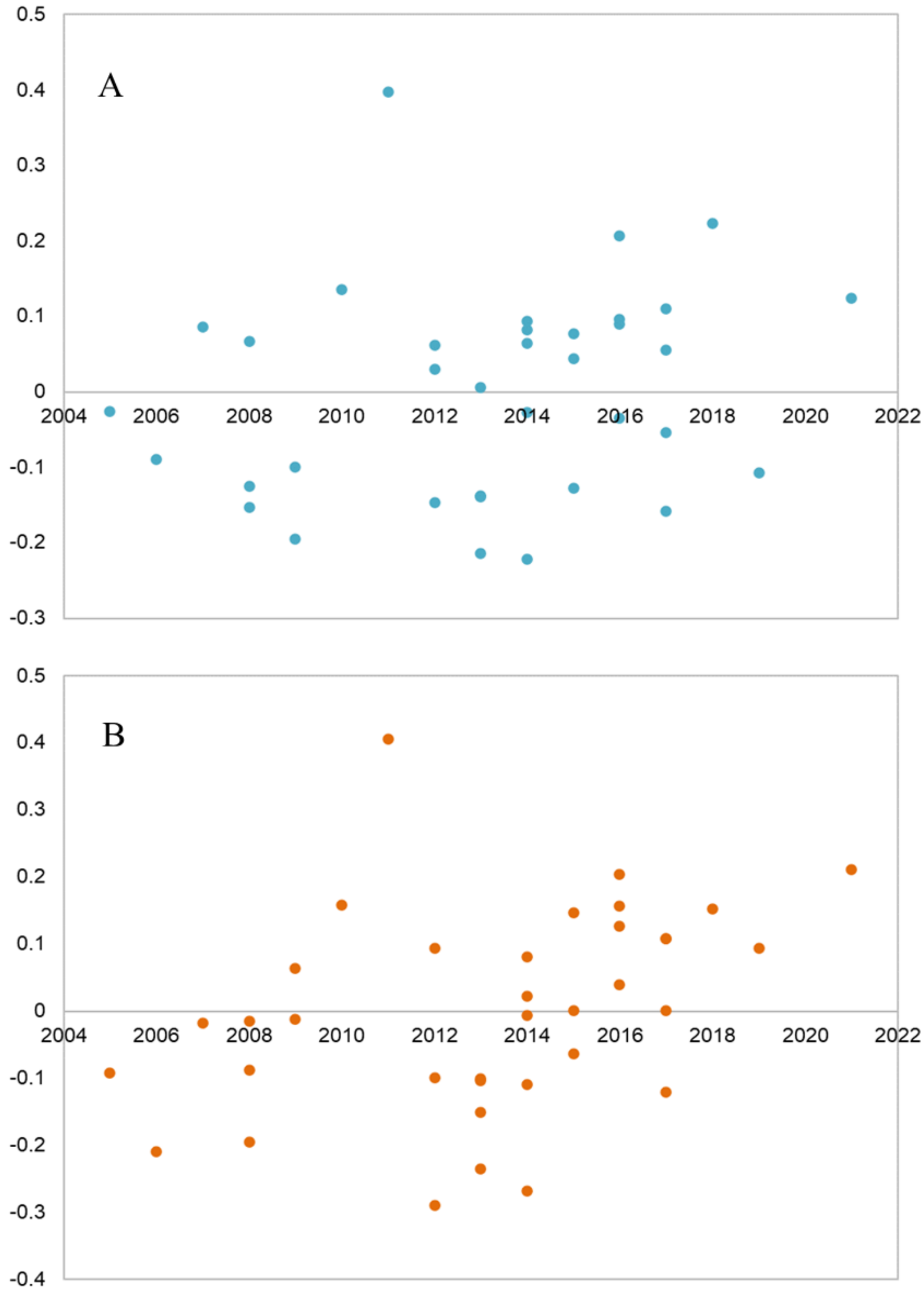


Figure 1-18. Residuals versus year for multiple regressions of spawning stock size time category and (A) level of development (C/ha) or (B) standardized median spawning survey conductivity (uS/cm) against proportion of stream samples with Herring eggs and-or larvae (P_{herr}). Median conductivity was standardized to background estimates for Coastal Plain and Piedmont regions based on estimates in Morgan et al. (2012).



MD – Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern
Section 2: Estuarine Yellow Perch Larval Presence-Absence Sampling

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

Introduction

Annual L_p, or the proportion of tows containing 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 postlarval stage. Presence-absence sampling for Yellow Perch larvae was conducted in the upper tidal reaches of the Choptank and Sassafras Rivers in 2021 (Figure 2-1). Sampling started the last week of March, and the first week of April, respectively, and continued through the first week of May in the Choptank, and the last week of April in the Sassafras.

Choptank and Sassafras Rivers are located in the Coastal Plain on the eastern side of Chesapeake Bay (Figure 2-1). The Sassafras River is a small tributary of Chesapeake Bay with a 19,580 Ha watershed. Agricultural is the primary land use (64% of the watershed) and development is low (C/ha = 0.11; Table 2-1). The subestuary is classified as oligohaline (salinity < 0.05‰) based on two Chesapeake Bay Program (CBP) monitoring stations; one within Sassafras River (ET 3.1) and another just below the mouth in the mainstem Bay (CB 2.2; MD DNR 2022a). Sassafras River represented the only low development, low salinity watershed with agriculture as its dominant land use with Yellow Perch spawning.

Choptank River is a large tributary of Chesapeake Bay with a watershed of 110,016 Ha. Land use is similar to Sassafras River (Table 2-1). Salinity classification runs from mesohaline at the mouth (CBP site EE2.1), to oligohaline at Ganey's Wharf, in the Yellow Perch larval nursery and Striped Bass spawning area (CBP site ET5.1; MD DNR 2022a). Nursery conditions for Yellow Perch larvae and Striped Bass eggs and larvae (see Section 2.1) could be surveyed concurrently in Choptank River and that influenced it being chosen for monitoring. An overfishing declaration and successive years of poor recruitment of Striped Bass have generated concern in the fisheries management and angling community. There has been unease expressed about degradation of Striped Bass spawning and larval nursery habitat in Chesapeake Bay. We have assembled historical data (Uphoff et al. 2020; 2022) and reoriented some of our spring monitoring in 2022 to respond to Striped Bass habitat concerns while maintaining Yellow Perch larval monitoring.

In 2021 we used regression analyses to examine relationships among land use types (development, agriculture, forest, and wetlands), L_p, organic matter availability, and watershed size. We also examined a hypothesis that watershed land use impacted related organic matter (OM) dynamics.

Methods

Choptank and Sassafras Rivers were sampled by program personnel in 2021. The Nanticoke River has been voluntarily sampled by the Maryland Fishing and Boating Services Shad and Herring program, during its normal operations in years past, without charge to this grant. Due to time and staffing limitations, however, collections were only done in the Nanticoke on three days during 2021, and these data were not used for analyses.

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). Each sample was collected in a glass jar which was then emptied into a dark pan to check for Yellow Perch 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 pan contained enough OM to obscure seeing larvae, it was 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. Temperature, dissolved oxygen (DO), conductivity, pH, and salinity were measured at each site on each sample date, and, in 2021, alkalinity was added to the suite of water quality parameters collected in Choptank River (see Section 2.1 of this report for additional information and results).

Ten sites were sampled twice weekly in all systems (Figure 2-1) unless weather or salinity did not allow. Boundaries of areas sampled were determined from Yellow Perch larval presence in estuarine surveys conducted during the 1970s and 1980s (O'Dell 1987) when this information was available. In larger subestuaries with designated Striped Bass areas (Choptank, Nanticoke, Patuxent, Wicomico, and Chester rivers), boundaries were the same as the legal Striped Bass spawning areas. Historical estimates of L_p were initially developed from surveys conducted for Striped Bass eggs and larvae in the Choptank and Nanticoke rivers (Uphoff 1993) and continuity with past surveys was maintained by sampling these Striped Bass spawning areas.

In general, sampling to determine L_p began during the last week of March or first week of April and ended after larvae were absent (or nearly so) for two consecutive sampling rounds, usually mid-to-late April, depending on larval presence and catchability. 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 for L_p conducted 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; see also Section 2.1), 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, or early in the second week, of April 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 with a 1:3 mouth to length ratio 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, 1:8 mouth to length ratio) mounted in the cod-end were used in the Choptank River during 1980-

1990 (Uphoff 1997; Uphoff et al. 2005; Uphoff et al. 2022). Survey designs for the Choptank and Nanticoke Rivers were described in Uphoff (1997) and Uphoff et al. (2020).

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

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

where $N_{present}$ equaled the number of samples with Yellow Perch larvae present during the L_p period and N_{total} equaled the total number of samples during the L_p period. Sites used to estimate L_p did not include downstream or upstream sites beyond the range where larvae were found. 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:

$$^{(3)}\ L_p \pm 1.96 \cdot SD; \text{ (Ott 1977).}$$

For this report we also started to explore using temperature limits to estimate L_p, specifically to determine an endpoint to use for analyses, as sampling continues for Striped Bass spawning estimates beyond Yellow Perch spawning season (see Section 2.1). We used past surveys to look at the cumulative frequency of presence by temperature, and we conducted both an all-years combined analysis, and one that split plankton trawl years (see Section 2.1) from conical plankton net years to see if the upper range of temperature was different. Prior to 1994 many systems had actual count data available, and these were converted to presence-absence for inclusion with the rest of the data set. Water temperature increments were in units of 1ºC (for example, the increment at 16ºC consisted of presence for all samples from 16.0 to 16.99ºC). Sample sizes within increments varied, but the trend was expected to be reasonably representative. Cumulative frequency of presence for each temperature increment was expressed as a percent of the total. Increments with most rapid growth in cumulative percent were considered to represent important temperatures for Yellow Perch larvae. Several systems, which had five years of data or more available, were analyzed separately (Bush, Choptank, Nanticoke, and Severn rivers, and Mattawoman and Nanjemoy creeks), and then an analysis containing all years and systems available was conducted as well (see Section 2 in Uphoff et al. 2022 for a description of time-series available).

Methods used to estimate development (C/ha) and land use indicators (percent of watershed in agriculture, forest, wetlands, and urban land use) are explained in **General Spatial and Analytical Methods used in Job 1, Sections 1-3.** Development targets and limits, and general statistical methods (analytical strategy and equations) are described there as well. Specific spatial and analytical methods for Section 2 are described below.

Estimates of C/ha and MD DOP land cover (agriculture, forest, and wetland) percentages were used as measures of watershed land use for analyses (Table 2-1). Whole watershed estimates were used with the following exceptions: Nanticoke, Choptank, Chester, Wicomico (eastern shore region of Maryland or ES), and Patuxent River watersheds were truncated at the lower boundaries of their Striped Bass spawning areas, and estimates for Choptank and Nanticoke River watersheds stopped at the Delaware border (latter due to lack of comparable land use data). Estimates of C/ha were available from 1950 through 2020 for Yellow Perch analyses (M. Topolski, MD DNR, personal communication).

Uphoff et al. (2012) developed L_p thresholds for brackish (salinity > 2.0‰ in the subestuary outside of the larval nursery) and tidal-fresh systems (salinity always ≤ 2.0‰). Choptank River was classified as brackish, while Sassafras River was classified as tidal-fresh.

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 (0.40) 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 within the same time span (13th to 17th 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 in decline, as opposed to occasional depressions (Uphoff et al. 2012).

Linear regression was used to evaluate time trends in L_p in two large subestuaries with extended time-series: Choptank River (1986-2021; N = 26) and Nanticoke River (1965-2019; N = 20). Neither time-series was continuous; Choptank River estimates were available for 1980-1990, 1998-2004, and 2013-2021, while the Nanticoke River estimates were available for 1965, 1967, 1968, 1970, 1971, 2004-2009, and 2011-2019.

Two regression approaches were used to examine possible linear relationships 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 subestuaries and 1 indicating brackish subestuary 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 impervious surface (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 that may cause endocrine disruption, leading to poor egg and larval viability (Wu et al. 2003; 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. These same analyses were repeated using percent agriculture and percent forest land cover estimates in place of C/ha in regressions with L_p. Regression analyses were also used to examine relationships between C/ha, watershed size and salinity, and their effects on L_p.

We used Akaike Information Criteria adjusted for small sample size, AICc, to evaluate the models that describe hypotheses that related changes in L_p to either C/ha for each salinity category (separate slopes), or to C/ha and salinity category (common slopes, separate intercepts; Burnham and Anderson 2001; Freund and Littel 2006):

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

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 Δ_i, (AIC_{ci} – minimum AIC_c), where i is an individual model, for the tidal-fresh or brackish regression compared to the multiple regression. The Δ_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 (land use type that predominated in the watershed) when interpreting plots of salinity classification (brackish or tidal-fresh), C/ha, and L_p. Dominant land use (agriculture, forest, or urban) was determined from Maryland Department of Planning estimates for 1973, 1994, 1997, 2002, or 2010 that fell closest to a sampling year (MD DOP 2020). Urban land consisted of high and low density residential, commercial, and institutional acreages (MD DNR 1999).

We used OM0 (proportion of samples without organic material, i.e., proportion with rank = 0) as our indicator of detritus availability. OM0 estimates were available for 2011-2021. The distribution of OM ranks assigned to samples were highly skewed towards zero, and few ranks greater than one were reported. We were specifically interested in the relationship of the amount of organic matter to development, and regressed OM0 against C/ha. 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 power and logistic growth functions to these data.

We were interested in links among OM0, percent wetlands in a watershed, and C/ha. Examination of the plot of percent wetlands and C/ha suggested that the relationship was nonlinear, with percentage of wetlands decreasing at a decreasing rate with C/ha, and appeared to be a mirror image of the plot of OM0 and C/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 linear functions to these data sets, respectively.

Results

Sampling in 2021 began on Choptank River on March 25 and lasted until May 6, while sampling on Sassafras River began on April 5 and concluded on April 27. Samples through April 23 and April 19 were used to estimate L_p in Choptank and Sassafras Rivers, respectively. Samples were collected at three stations in the Nanticoke on April 8, at eight stations on April 13, and at 10 stations on April 20. Collections in the Nanticoke were not used for estimating L_p in 2021 due to low sample size.

The estimate of mean L_p was just below the tidal-fresh threshold (0.65) in the Sassafras River (L_p = 0.60; 95% CI's did overlap the threshold), and just above the brackish threshold (0.40) in the Choptank River (L_p = 0.41), during 2021 (Figure 2-2). Comparisons of L_p during 2021 with historical estimates for brackish subestuaries is plotted in Figure 2-3 and for tidal-fresh estimates in Figure 2-4. The range of C/ha values available for analysis with L_p was 0.05-2.84 for brackish subestuaries and 0.11-3.33 for tidal-fresh (Table 2-1).

Strong relationships of L_p with year were not evident in the Choptank River or Nanticoke River. Estimates of L_p in Choptank River during 1986-2021 exhibited little indication of decline (r^2 = 0.06; P = 0.21), while a decline of L_p of about 0.005 per year was detected during 1965-2019 (predicted L_p declined from 0.63 to 0.38) in the Nanticoke River (r^2 = 0.17; P = 0.07; Figure 2-3). Both of these subestuaries are rural, land use is dominated by agriculture, and they have been closed to commercial fishing since 1989 (Piavis 2005).

Separate linear regressions of C/ha and L_p by salinity category indicated that C/ha was modestly and negatively related to L_p and L_p was, on average, higher in tidal-fresh subestuaries than in brackish subestuaries ($P \leq 0.0009$; Table 2-2; Figure 2-5). Estimates of C/ha accounted for 25% of variation of L_p in brackish subestuaries and 30% in tidal-fresh subestuaries. Based on 95% CI overlap, intercepts were different between tidal-fresh (mean = 0.91, SE = 0.08) and brackish (mean = 0.59, SE = 0.03) subestuaries. Mean slope for C/ha estimated for tidal-fresh subestuaries (mean = -0.26, SE = 0.07) were steeper, but 95% CI's overlapped CI's estimated for the slope of brackish subestuaries (mean = -0.16, SE = 0.03; Table 2-2). 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 moderate fit of L_p with C/ha ($r^2 = 0.30$; Table 2-2) as separate regressions for each salinity type. Intercepts of tidal-fresh and brackish subestuaries equaled 0.91 and 0.59, respectively; the common slope was -0.18. Predicted L_p over the observed ranges of C/ha available for each salinity type would decline from 0.58 to 0.13 in brackish subestuaries and from 0.88 to 0.04 in tidal-fresh subestuaries (Figure 2-5).

Estimates of L_p were weakly related to agriculture ($r^2 = 0.15$, P = 0.001) and forest ($r^2 = 0.02$, P = 0.2635) in brackish tributaries (Table 2-2; Figure 2-5). Regressions of L_p and agriculture and forest in tidal-fresh subestuaries were very similar to that found in brackish ones, but sample sizes were lower so their level of significance was above 0.05 (Table 2-2). Results of linear regressions of L_p with percent agriculture or forest did not explain enough variation to be of interest for management. Regression analysis did not suggest a relationship of wetlands with L_p in subestuaries of either salinity type so additional analyses were not conducted.

Akaike's Information Criteria values equaled 9.3 for the regression of C/ha and L_p for brackish subestuaries, 9.8 for tidal-fresh estuaries, and 11.4 for the multiple regression that included salinity category (Table 2-3). Calculations of Δi for brackish or tidal-fresh versus multiple regressions were approximately 2.04 and 1.58 (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 (Table 2-3).

Additional regressions examining the effects of watershed size and salinity on the relationship between C/ha and L_p indicated that considering either separately improved the regression fit similarly (overall, $r^2 = 0.15$, P < 0.0001; size, $R^2 = 0.24$, P < 0.0001; and salinity, $R^2 = 0.30$, P < 0.0001), but combining them into a single model did not improve the fit and size was no longer significant (combined $R^2 = 0.31$; salinity, P = 0.0037 and size, P = 0.2236). Considering size separately, all tidal-fresh systems are within the small-system size category, so fit did not change from previous analyses ($r^2 = 0.31$, P = 0.0009; Tables 2-2 and 2-4, respectively). The relationship between C/ha and L_p in small, brackish systems was better explained, however ($r^2 = 0.56$, P <.0001; Table 2-4). A relationship between C/ha and L_p was not detected for large systems (Table 2-4).

Based on cumulative frequency of presence for the individual systems analyzed, and for the larger data set (full time series), it was determined that the cumulative catch distribution showed the greatest increase between 12ºC (full time series cumulative P = 0.290) and 17ºC (full time series cumulative P = 0.877) or 18ºC (full time series cumulative P = 0.930; Figure 2-6). Estimates of L_p using these temperature cutoffs were tabulated along with our original estimates (Table 2-5). We have started the process of determining which would be best to use. This work should be finalized in the next annual report.

In 2021, temperatures were similar in rural, agricultural Choptank and Sassafras Rivers, but DO and pH values were significantly higher in the Sassafras (Table 2-6; Figure 2-7). Harmful algal blooms have been documented several times in the Sassafras River (see Section 3 of Uphoff et al. 2022; MD DNR 2022b) and while a phytoplankton bloom was not noted during sampling, this could contribute to the high DO and pH values observed. While these differences are not likely to be fatal to Yellow Perch larvae, they do point to differences in dynamics and conditions among tributaries and years.

Although we have analyzed these data by distinguishing tidal-fresh and brackish subestuaries, inspection of Table 2-1 indicated an alternative interpretation based on primary land use estimated by MD DOP. Predominant land use at lower levels of development may influence intercept estimates. Rural watersheds with below threshold development (at or below C/ha target) in tidal-fresh subestuaries were dominated by forest, with only a single low development, low salinity watershed with agricultural as its dominant land use available (Figure 2-8). Dominant land cover estimated by MD DOP for watersheds of tidal-fresh subestuaries was split between forest (C/ha = 0.46-0.95; 18 observations) and urban (C/ha ≥ 1.17; 14 observations). Nearly all rural land in brackish subestuary watersheds was in agriculture (C/ha ≤ 0.22; 50 observations), while forest land cover was represented by six observations from Nanjemoy Creek (C/ha = 0.09) and two from Wicomico River (eastern shore; C/ha = 0.68). The range of L_p was similar in brackish subestuaries with forest and agricultural cover, but the distribution shifted towards higher L_p in the limited sample from Nanjemoy Creek. Urban land cover predominated in 13 observations of brackish subestuaries (C/ha ≥ 1.24; Table 2-1; Figure 2-8). 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.

Estimates of C/ha and OM0 were significantly related. A non-linear power function provided a moderate fit to the data (approximate $r^2 = 0.41$, P <.0001; N = 44), depicting OM0 increasing towards 1.0 at a decreasing rate as C/ha approached 1.50 (Figure 2-9). The relationship was described by the equation:

$$^{(5)}\ OM0 = 0.79 \cdot ((C/ha)^{0.25}).$$

Approximate standard errors were 0.05 for parameters a and b. A logistic growth function fit these data similarly, but one term was not significantly different from zero, so the model was rejected.

Percent wetlands (determined from the most recent MD DOP estimates in 2010) and development, and OM0 and wetland percentage were negatively related. An inverse power function provided a moderate fit of C/ha and percent wetland (approximate $r^2 = 0.41$, P <.0001, N = 44; Figure 2-10), while the relationship of OM0 and wetland percentage was linear ($r^2 = 0.52$, P <.0001, N = 44; Figure 2-10). These relationships 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.

Discussion

General patterns of land use and L_p emerged from analyses: L_p was negatively related to development, positively associated with forest and agriculture, and not associated with wetlands. Wetlands appeared to be an important source of organic matter for subestuaries.

Rural features (agriculture, forest, and wetlands) were negatively correlated with development in the watersheds monitored for L_p (Uphoff et al. 2017). A broad range of L_p (near 0 to 1.0) was present up to 1.3 C/ha. Beyond 1.3 C/ha, estimates of L_p values were ≤ 0.65. A full range of L_p values occurred in subestuaries with agricultural watersheds (C/ha was < 0.22). A forest cover classification in a watershed was associated with higher L_p (median L_p = 0.78) than agriculture (median L_p = 0.55) or development (median L_p = 0.35), but these differences may have also reflected dynamics unique to brackish or tidal-fresh subestuaries since all but one agricultural watershed had brackish subestuaries, and nearly all forested watersheds had tidal-fresh subestuaries.

At least five factors can be identified that potentially contribute to variations in L_p: salinity, summer hypoxia, maternal influence, winter temperature, and watershed development. Some of these factors may not be independent and there is considerable potential for interactions among them.

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 Yellow Perch larvae collected in Choptank River (based on counts) during 1980-1985 were from 1‰ or less. Approximately 85% of Yellow Perch larvae collected by Dovel (1971) from Magothy and Patuxent rivers, and Head-of-Bay, during 1963-1967 were collected at salinity 1‰ or less.

Severn River offers the most extensive evidence of salinity changes in a subestuary. These changes were concurrent with development from 0.35 to 2.30 C/ha. During 2001-2003 salinity within Severn River's estuarine Yellow Perch larval nursery ranged between 0.5 and 13‰ (C/ha was ~ 2.0); 93% of measurements were above the salinity requirement for eggs and larvae of 2‰ (Uphoff et al. 2005). Muncy (1962) and O'Dell's (1987) descriptions of upper Severn River salinity suggested that the nursery was less brackish in the 1950s through the 1970s than at present (C/ha was 0.35 in 1950 and rose to 1.01 by 1976), although a single cruise by Sanderson (1950) measured a rise in salinity with downstream distance similar to what Uphoff et al. (2005) observed. Most Yellow Perch spawning in Severn River during 1958 occurred in waters of 2.5‰ or less (Muncy 1962). Mortality of Yellow Perch eggs and prolarvae in experiments generally increased with salinity and was complete by 12‰ (Sanderson 1950; Victoria et al. 1992). Uphoff et al. (2005) estimated that nearly 50% of the historic area of estuarine nursery for Yellow Perch was subject to salinities high enough to cause high mortality. Salinity in the estuarine nursery of Severn River varied without an annual pattern even though conditions went from extremely dry to extremely wet (Uphoff et al. 2005).

As development increases, rainfall flows faster across the ground and more of it reaches fluvial streams rather than recharging groundwater (Cappiella and Brown 2001; Beach 2002). In natural settings, very little rainfall is converted to runoff and about half is infiltrated into underlying soils and the water table (Cappiella and Brown 2001). These pulses of runoff in developed watersheds alter stream flow patterns and could be at the root of the suggested change in salinity at the head of the Severn River estuary where the larval nursery is located (Uphoff et al. 2005).

In our studies, suburban mesohaline subestuaries commonly exhibit summer hypoxia in bottom channel waters, but it is less common in agricultural watersheds (see Section 3). Stratification due to salinity is an important factor in development of hypoxia in mesohaline subestuaries, while hypoxia is rarely encountered in tidal-fresh and oligohaline subestuaries (see Section 3). Depressed egg and larval viability in fish due to endocrine disruption may follow inadequate DO the previous summer (Wu et al. 2003; Uphoff et al. 2005; Thomas and Rahman 2011; Tuckey and Fabrizio 2016). Ovaries of Yellow Perch are repopulated with new germ cells during late spring and summer after resorptive processes are complete (Dabrowski et al. 1996, Ciereszko et al. 1997) and hypoxic conditions are well developed by the time we begin our summer habitat assessments in early July (see Section 3).

Hypoxia in coastal waters reduces fish growth and condition due to increased energy expenditures to avoid low DO and compete for reduced food resources (Zimmerman and Nance 2001; Breitburg 2002; Stanley and Wilson 2004). Reproduction of mature female fish is higher when food is abundant and condition is good (Marshall et al. 1999; Lambert and Dutil 2000; Rose and O'Driscoll 2002; Tocher 2003), but stress may decrease egg quality (Bogevik et al. 2012). A female Yellow Perch's energetic investment provides nutrition for development and survival of its larvae until first feeding (Heyer et al. 2001) and differences in Yellow Perch larval length, yolk volume, and weight were attributed to maternal effects in Lake Michigan (Heyer et al. 2001).

Widespread low L_p occurs sporadically in Chesapeake Bay subestuaries with rural watersheds and appears to be linked to high winter temperatures (Uphoff et al. 2013). During 1965-2012, estimates of L_p less than 0.5 did not occur when average March air temperatures were 4.7°C or less (N = 3), while average March air temperatures of 9.8°C or more were usually associated with L_p estimates of 0.5 or less (7 of 8 estimates). Estimates of L_p between this temperature range exhibited high variation (0.2 – 1.0, N = 27; Uphoff et al. 2013). In Yellow Perch, a period of low temperature is required for reproductive success (Heidinger and Kayes 1986; Ciereszko et al. 1997). Recruitment of Yellow Perch continuously failed in Lake Erie during 1973-2010 following short, warm winters (Farmer et al. 2015). Subsequent lab and field studies indicated reduced egg size, energy and lipid content, and hatching success followed short winters even though there was no reduction in fecundity. Whether this reduced reproductive success was due to metabolic or maternal endocrine pathways could not be determined (Farmer et al. 2015).

Yellow Perch and Striped Bass larvae are found in the same regions of large tidal rivers in Chesapeake Bay (Uphoff 1991; 2020). Copepods, typically of the genus *Eurytemora*, were important prey of Striped Bass and Yellow Perch larvae (MD Sea Grant 2009; Uphoff et al. 2017). Winter water temperature has also been found to have an influence on peak abundances of an important zooplankton prey (*Eurytemora carolleeae*) of larval Striped Bass, which could affect recruitment in the spring (Millette et al. 2020). Millette et al. (2020) found that low temperature delayed development timing and increased the size of peak spring abundance of copepod nauplii in Chesapeake Bay Striped Bass larval nurseries. Results suggest that cold winters, in conjunction with freshwater discharge, explained up to 78% of annual recruitment variability in Striped Bass due to larvae occurring at the same time as high concentrations of their prey (Millette et al. 2020). Given the high correlation of Striped Bass and Yellow Perch juvenile indices in Maryland's portion of Chesapeake Bay and high concurrence of their larvae in their nursery (Uphoff et al. 2020), we would expect these same factors would impact Yellow Perch recruitment.

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) offered an explanation for 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 > 1.32; 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). 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 through 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). A hatchery program attempted to raise Severn River Yellow Perch larvae and juveniles for mark-recapture experiments, but egg viability declined drastically by the early 2000s and Choptank River brood fish had to be substituted (Uphoff et al. 2005). Estimates of L_p from Severn River were persistently low during the 2000s. 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 Severn River and similarly impacted western shore subestuaries (Magothy and South rivers) in the mid-to-late 1990s.

These reconstituted fisheries were likely supported by juvenile Yellow Perch that migrated from the upper Bay nursery rather than internal production (Uphoff et al. 2005). A sudden upward shift in both Yellow Perch juvenile indices and mesozooplankton relative abundance occurred in the early 1990s in the Head-of-Bay region which coincided with a downward shift in annual chlorophyll a averages at two Head-of-Bay monitoring stations (Uphoff et al. 2013). This shift in Head-of-Bay productivity was followed by reports of increased angling success in western shore subestuaries below the Head-of-Bay: Rock and Curtis creeks and Severn, South, and Magothy rivers (Piavis and Uphoff 1999). Declines in L_p in the Magothy, Severn, and South rivers indicated a loss of productivity. All eleven estimates of L_p have been below the threshold in the three western shore subestuaries with well-developed watersheds during 2001-2016, while estimates from Head-of-Bay subestuaries have typically been above the threshold (4 of 7 Bush River estimates, 2 of 3 Elk River estimates, and 5 of 5 Northeast River estimates). 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. The term "regime shift" has been used to suggest these types of changes in productivity are causally connected and linked to other changes in an ecosystem (Steele 1996; Vert-pre et al. 2013).

Amount of organic matter present was negatively influenced by development. Estimates of C/ha and OM0 were moderately related, and a non-linear power function depicted OM0 increasing towards 1.0 at a decreasing rate with C/ha. 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). Development associated with increased human population growth in the Chesapeake Bay watershed converts natural sources of organic matter (forests and welands) to agricultural, residential and industrial uses that alter and lessen the supply of watershed organic matter.

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 year-class success of Head-of-Bay Yellow Perch (Uphoff et al. 2013).

Higher DO and pH values in rural Sassafras River likely reflected higher primary production by phytoplankton. The possibility exists that this could lead to lower zooplankton production and lower juvenile abundance, although these mechanisms are not clearly understood. RNA/DNA analyses during 2015 and 2016 did not indicate reduced larval condition in urbanized Patuxent River when compared with rural Choptank River; however, presence of OM and subsequent feeding success of first-feeding Yellow Perch was negatively influenced by development in multiple subestuaries (Uphoff et al. 2017). 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 River and Patapsco River 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 as C/ha 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 (https://www.epa.gov/chesapeake-bay-tmdl/chesapeake-bay-tmdl-fact-sheet). 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 and has been associated with increased, decreased, and undetectable changes in OM that may reflect diversity of farming practices (Stanley et al. 2012). In our study, agricultural watersheds (all eastern shore) had most of the lower OM0 scores (indicating more OM), while OM0 levels were higher and distributed similarly among watersheds that were predominately in development (all western shore) or forest (eastern and western shore). The MD DOP forest cover estimates have a minimum mapping unit of 10 acres that mixes forest cover in residential areas (trees over lawns) with true forest cover, clouding interpretation of forest influence (R. Feldt, MD DNR Forest Service, personal communication).

Annual L_p (proportion of tows with Yellow Perch larvae during a standard period of time, and where larvae would be expected, provided an economically collected measure of the product of egg production and egg through early postlarval survival. Declines in survival for older Yellow Perch life stages would not be detected using L_p alone. We used L_p as an index to detect "normal" and "abnormal" egg and early larvae dynamics. We considered L_p estimates from subestuaries that were persistently lower than those measured in other subestuaries indicative of abnormally low survival. Remaining levels were considered normal. Assuming catchability does not change greatly from year to year, egg production and egg through early postlarval survival would need to be high to produce strong L_p, but only one factor needed to be low to result in lower L_p.

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). Counts coupled with gear efficient at collecting larger, older larvae would be needed to estimate mortality rates. Tighter budgets necessitate development of low-cost indicators of larval survival and relative abundance in order to pursue an ecosystem approach to 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 in previous years (Uphoff et al. 2017) required laboratory analysis. These latter two analyses represented separate studies rather than a requirement for estimating L_p (Uphoff et al. 2017).

We have relied on correlation and regression analyses to judge the effects of watershed development on Yellow Perch larval dynamics (see Uphoff et al. 2017). Interpretation of the influence of salinity class or major 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.

Hilborn (2016) reviewed the use of correlation in fisheries and ecosystem management and this advice should apply to regression analyses that we used since the underlying math is very similar. Ideally, manipulative experiments and formal adaptive management should be employed. In large-scale aquatic ecosystems these opportunities are limited and are not a possibility for us. Correlations may not be causal, but they represent 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).

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 DO, 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. Estimates of proportions of ichthyoplankton net tows with Yellow Perch larvae (L_p) during 1965-2021 and data used for regressions with counts of structures per hectare (C/ha), percent agriculture, percent forest, and percent wetland. Salinity class 0 = tidal-fresh (≤ 2.0‰) and 1 = brackish (> 2.0‰). Land use percentages and overall primary land use were determined from Maryland Department of Planning estimates for 1973, 1994, 1997, 2002, or 2010 that were closest to a sampling year.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	% Wetland	% Urban	Primary Land Use	Salinity	Lp
Bush (w/ APG)	2006	2002	1.17	21	36.3	5.5	37	Urban	0	0.79
Bush (w/ APG)	2007	2010	1.19	14.9	32.1	5.5	46.4	Urban	0	0.92
Bush (w/ APG)	2008	2010	1.20	14.9	32.1	5.5	46.4	Urban	0	0.55
Bush (w/ APG)	2009	2010	1.21	14.9	32.1	5.5	46.4	Urban	0	0.86
Bush (w/ APG)	2011	2010	1.23	14.9	32.1	5.5	46.4	Urban	0	0.96
Bush (w/ APG)	2012	2010	1.24	14.9	32.1	5.5	46.4	Urban	0	0.28
Bush (w/ APG)	2013	2010	1.25	14.9	32.1	5.5	46.4	Urban	0	0.15
Chester	2019	2010	0.13	66.6	24.5	0.8	7.8	Agriculture	1	0.73
Choptank	1980	1973	0.07	65.2	30.6	2	2.1	Agriculture	1	0.59
Choptank	1981	1973	0.07	65.2	30.6	2	2.1	Agriculture	1	0.82
Choptank	1982	1973	0.07	65.2	30.6	2	2.1	Agriculture	1	0.80
Choptank	1983	1973	0.07	65.2	30.6	2	2.1	Agriculture	1	0.33
Choptank	1984	1994	0.07	64	29.2	2.3	4.4	Agriculture	1	0.64
Choptank	1985	1994	0.07	64	29.2	2.3	4.4	Agriculture	1	0.85
Choptank	1986	1994	0.07	64	29.2	2.3	4.4	Agriculture	1	0.80
Choptank	1987	1994	0.08	64	29.2	2.3	4.4	Agriculture	1	0.76
Choptank	1988	1994	0.08	64	29.2	2.3	4.4	Agriculture	1	0.63
Choptank	1989	1994	0.08	64	29.2	2.3	4.4	Agriculture	1	0.64
Choptank	1990	1994	0.08	64	29.2	2.3	4.4	Agriculture	1	0.66
Choptank	1998	1997	0.10	63.6	27.7	2.2	6.4	Agriculture	1	0.61
Choptank	1999	1997	0.11	63.6	27.7	2.2	6.4	Agriculture	1	0.75
Choptank	2000	2002	0.11	63.9	27.1	2.1	6.9	Agriculture	1	0.27
Choptank	2001	2002	0.11	63.9	27.1	2.1	6.9	Agriculture	1	0.26
Choptank	2002	2002	0.11	63.9	27.1	2.1	6.9	Agriculture	1	0.38
Choptank	2003	2002	0.11	63.9	27.1	2.1	6.9	Agriculture	1	0.52
Choptank	2004	2002	0.12	63.9	27.1	2.1	6.9	Agriculture	1	0.41
Choptank	2013	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.47
Choptank	2014	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.68
Choptank	2015	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.82

Table 2-1 cont.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	% Wetland	% Urban	Primary Land Use	Salinity	Lp
Choptank	2016	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.90
Choptank	2017	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.40
Choptank	2018	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.44
Choptank	2019	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.69
Choptank	2021	2010	0.13	60.9	25.6	2.1	11.2	Agriculture	1	0.41
Corsica	2006	2002	0.21	64.3	27.4	0.4	7.9	Agriculture	1	0.47
Corsica	2007	2010	0.22	60.4	25.5	0.1	13.2	Agriculture	1	0.83
Elk	2010	2010	0.59	28	38.7	1.1	31.2	Forest	0	0.75
Elk	2011	2010	0.59	28	38.7	1.1	31.2	Forest	0	0.79
Elk	2012	2010	0.60	28	38.7	1.1	31.2	Forest	0	0.55
Langford	2007	2010	0.07	70.2	20.4	1.5	8	Agriculture	1	0.83
Magothy	2009	2010	2.74	1.2	21	0	76.8	Urban	1	0.10
Magothy	2016	2010	2.84	1.2	21	0	76.8	Urban	1	0.10
Mattawoman	1990	1994	0.46	13.8	62.6	0.9	22.5	Forest	0	0.81
Mattawoman	2008	2010	0.87	9.3	53.9	2.8	34.2	Forest	0	0.66
Mattawoman	2009	2010	0.88	9.3	53.9	2.8	34.2	Forest	0	0.92
Mattawoman	2010	2010	0.90	9.3	53.9	2.8	34.2	Forest	0	0.82
Mattawoman	2011	2010	0.91	9.3	53.9	2.8	34.2	Forest	0	0.98
Mattawoman	2012	2010	0.90	9.3	53.9	2.8	34.2	Forest	0	0.20
Mattawoman	2013	2010	0.91	9.3	53.9	2.8	34.2	Forest	0	0.47
Mattawoman	2014	2010	0.93	9.3	53.9	2.8	34.2	Forest	0	0.78
Mattawoman	2015	2010	0.94	9.3	53.9	2.8	34.2	Forest	0	1.00
Mattawoman	2016	2010	0.95	9.3	53.9	2.8	34.2	Forest	0	0.82
Middle	2012	2010	3.33	3.4	23.3	2.1	71	Urban	0	0.00
Nanjemoy	2009	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.83
Nanjemoy	2010	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.96
Nanjemoy	2011	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.98
Nanjemoy	2012	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.03
Nanjemoy	2013	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.46
Nanjemoy	2014	2010	0.09	12.4	68.7	4.1	14.7	Forest	1	0.82

Table 2-1 cont.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	% Wetland	% Urban	Primary Land Use	Salinity	Lp
Nanticoke	1965	1973	0.05	46.6	43.4	8.1	1.9	Agriculture	1	0.50
Nanticoke	1967	1973	0.05	46.6	43.4	8.1	1.9	Agriculture	1	0.43
Nanticoke	1968	1973	0.06	46.6	43.4	8.1	1.9	Agriculture	1	1.00
Nanticoke	1970	1973	0.06	46.6	43.4	8.1	1.9	Agriculture	1	0.81
Nanticoke	1971	1973	0.06	46.6	43.4	8.1	1.9	Agriculture	1	0.33
Nanticoke	2004	2002	0.11	46.3	40.7	7.4	5.5	Agriculture	1	0.49
Nanticoke	2005	2002	0.11	46.3	40.7	7.4	5.5	Agriculture	1	0.67
Nanticoke	2006	2002	0.11	46.3	40.7	7.4	5.5	Agriculture	1	0.35
Nanticoke	2007	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.55
Nanticoke	2008	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.19
Nanticoke	2009	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.41
Nanticoke	2011	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.55
Nanticoke	2012	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.04
Nanticoke	2013	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.43
Nanticoke	2014	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.35
Nanticoke	2015	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.64
Nanticoke	2016	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.67
Nanticoke	2017	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.22
Nanticoke	2018	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.28
Nanticoke	2019	2010	0.11	45	39.4	7.4	8.1	Agriculture	1	0.41
Northeast	2010	2010	0.46	31.1	38.6	0.1	28.9	Forest	0	0.68
Northeast	2011	2010	0.46	31.1	38.6	0.1	28.9	Forest	0	1.00
Northeast	2012	2010	0.47	31.1	38.6	0.1	28.9	Forest	0	0.77
Northeast	2013	2010	0.47	31.1	38.6	0.1	28.9	Forest	0	0.72
Northeast	2014	2010	0.48	31.1	38.6	0.1	28.9	Forest	0	0.77
Patuxent	2015	2010	1.24	20.5	35.1	1	41.7	Urban	1	0.72
Patuxent	2016	2010	1.25	20.5	35.1	1	41.7	Urban	1	0.82

Table 2-1 cont.

River	Sample Year	DOP Year	C / ha	% Ag	% Forest	% Wetland	% Urban	Primary Land Use	Salinity	Lp
Piscataway	2008	2010	1.41	10	40.4	0.2	47	Urban	0	0.47
Piscataway	2009	2010	1.43	10	40.4	0.2	47	Urban	0	0.39
Piscataway	2010	2010	1.45	10	40.4	0.2	47	Urban	0	0.54
Piscataway	2011	2010	1.46	10	40.4	0.2	47	Urban	0	0.65
Piscataway	2012	2010	1.47	10	40.4	0.2	47	Urban	0	0.16
Piscataway	2013	2010	1.49	10	40.4	0.2	47	Urban	0	0.50
Sassafras	2021	2010	0.11	64.1	25.9	1.3	8.3	Agriculture	0	0.60
Severn	2002	2002	2.02	8.6	35.2	0.2	55.8	Urban	1	0.16
Severn	2004	2002	2.09	8.6	35.2	0.2	55.8	Urban	1	0.35
Severn	2005	2002	2.15	8.6	35.2	0.2	55.8	Urban	1	0.40
Severn	2006	2002	2.18	8.6	35.2	0.2	55.8	Urban	1	0.27
Severn	2007	2010	2.21	5	28	0.2	65.1	Urban	1	0.30
Severn	2008	2010	2.24	5	28	0.2	65.1	Urban	1	0.08
Severn	2009	2010	2.25	5	28	0.2	65.1	Urban	1	0.15
Severn	2010	2010	2.26	5	28	0.2	65.1	Urban	1	0.03
South	2008	2010	1.32	10.2	39.2	0.5	48.8	Urban	1	0.14
Wicomico (ES)	2017	2010	0.68	30.1	36.8	2.3	29.9	Forest	1	0.53
Wicomico (ES)	2018	2010	0.68	30.1	36.8	2.3	29.9	Forest	1	0.38

Table 2-2. Summary of results of regressions of proportions of tows with Yellow Perch larvae (L_p) and (A) counts of structures per hectare (C/ha), (B) percent agriculture, and (C) percent forest. Separate regressions by salinity (tidal-fresh ≤ 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			(A) Brackish			
Source	df	SS	MS	F	P	
Model	1	1.15545	1.15545	22.88	<.0001	
Error	69	3.48415	0.05049			
Total	70	4.6396				
r^2	0.2490					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.58924	0.03121	18.88	<.0001	0.52698	0.6515
C / ha	-0.16257	0.03399	-4.78	<.0001	-0.23037	-0.09477

ANOVA			(A) Tidal-Fresh			
Source	df	SS	MS	F	P	
Model	1	0.68442	0.68442	13.37	0.0009	
Error	31	1.58737	0.05121			
Total	32	2.27179				
r^2	0.3013					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.91029	0.08231	11.06	<.0001	0.74242	1.07817
C / ha	-0.26099	0.07139	-3.66	0.0009	-0.40659	-0.11539

ANOVA			(A) Multiple Regression			
Source	df	SS	MS	F	P	
Model	2	2.16749	1.08375	21.25	<.0001	
Error	101	5.15066	0.051			
Total	103	7.31815				
r^2	0.2962					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.82927	0.05018	16.53	<.0001	0.72974	0.92881
C / ha	-0.18096	0.0308	-5.88	<.0001	-0.24206	-0.11987
Salinity	-0.23126	0.05035	-4.59	<.0001	-0.33114	-0.13137

Table 2-2 cont.

ANOVA			(B) Brackish			
Source	df	SS	MS	F	P	
Model	1	0.67986	0.67986	11.85	0.001	
Error	69	3.95973	0.05739			
Total	70	4.6396				
r^2	0.1465					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.32154	0.06213	5.18	<.0001	0.19759	0.44549
% Ag	0.00441	0.00128	3.44	0.001	0.00186	0.00697

ANOVA			(B) Tidal-Fresh			
Source	df	SS	MS	F	P	
Model	1	0.08915	0.08915	1.27	0.2691	
Error	31	2.18263	0.07041			
Total	32	2.27179				
r^2	0.0392					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.56965	0.08212	6.94	<.0001	0.40217	0.73714
% Ag	0.00439	0.0039	1.13	0.2691	-0.00356	0.01233

ANOVA			(B) Multiple Regression			
Source	df	SS	MS	F	P	
Model	2	1.17578	0.58789	9.67	0.0001	
Error	101	6.14237	0.06082			
Total	103	7.31815				
r^2	0.1607					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.56922	0.04806	11.84	<.0001	0.47388	0.66456
% Ag	0.00441	0.00124	3.56	0.0006	0.00195	0.00687
Salinity	-0.24754	0.06093	-4.06	<.0001	-0.36841	-0.12668

Table 2-2 cont.

ANOVA			(C) Brackish			
Source	df	SS	MS	F	P	
Model	1	0.08391	0.08391	1.27	0.2635	
Error	69	4.55568	0.06602			
Total	70	4.6396				
r^2	0.0181					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.40878	0.09624	4.25	<.0001	0.21679	0.60078
% Forest	0.0029	0.00257	1.13	0.2635	-0.00223	0.00802

ANOVA			(C) Tidal-Fresh			
Source	df	SS	MS	F	P	
Model	1	0.22195	0.22195	3.36	0.0766	
Error	31	2.04984	0.06612			
Total	32	2.27179				
r^2	0.0977					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.28896	0.19999	1.44	0.1585	-0.11892	0.69684
% Forest	0.00856	0.00467	1.83	0.0766	-0.000969	0.01808

ANOVA			(C) Multiple Regression			
Source	df	SS	MS	F	P	
Model	2	0.63813	0.31906	4.82	0.01	
Error	101	6.68003	0.06614			
Total	103	7.31815				
r^2	0.0872					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.4702	0.10414	4.52	<.0001	0.26362	0.67679
% Forest	0.00421	0.00225	1.87	0.0643	-0.000256	0.00868
Salinity	-0.10819	0.05596	-1.93	0.056	-0.21921	0.00282

Table 2-3. Summary of Akaike's Information Criteria from regressions of proportions of tows with Yellow Perch larvae (L_p) and counts of structures per hectare (C/ha) for each salinity category, and a multiple regression using salinity as a class variable.

Model	MSE	n	K	neg2loge(MSE)	2K	2K(K+1)	(n-K-1)	AICc	Delta brackish	Delta fresh
Categorical	0.051	104	4	2.975929646	8	40	99	11.4	2.04	1.58
Fresh	0.05121	33	3	2.971820454	6	24	29	9.8		
Brackish	0.05049	71	3	2.985979982	6	24	67	9.3		

Table 2-4. Summary of results of regressions of proportions of tows with Yellow Perch larvae (L_p) and (A) small system counts of structures per hectare (C/ha), or (B) large system counts of structures per hectare (C/ha). Separate regressions by salinity (tidal-fresh ≤ 2.0 ‰ and brackish > 2.0 ‰) are presented for small systems only as all large systems are brackish.

ANOVA			(A) Small Brackish			
Source	df	SS	MS	F	P	
Model	1	1.22963	1.22963	26.19	<.0001	
Error	21	0.98609	0.04696			
Total	22	2.21572				
r^2	0.555					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.68482	0.06774	10.11	<.0001	0.54393	0.8257
C / ha	-0.22159	0.0433	-5.12	<.0001	-0.31164	-0.13154

Table 2-4 cont.

ANOVA			(A) Small Tidal-Fresh			
Source	df	SS	MS	F	P	
Model	1	0.68442	0.68442	13.37	0.0009	
Error	31	1.58737	0.05121			
Total	32	2.27179				
r^2	0.3013					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.91029	0.08231	11.06	<.0001	0.74242	1.07817
C / ha	-0.26099	0.07139	-3.66	0.0009	-0.40659	-0.11539

ANOVA			(B) Large Brackish			
Source	df	SS	MS	F	P	
Model	1	0.04312	0.04312	1.24	0.275	
Error	27	0.93777	0.03473			
Total	28	0.98088				
r^2	0.044					
	Coefficients	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.58982	0.04075	14.47	<.0001	0.50621	0.67343
C / ha	0.13265	0.11906	1.11	0.275	-0.11164	0.37694

Table 2-5. Estimates of proportions of ichthyoplankton net tows with Yellow Perch larvae (L_p) and their standard deviations (SD) using original methodology, a 17°C temperature cutoff, and an 18°C temperature cutoff in systems having five years or more of data. N/C indicates there was "no change" from the previous column's estimate.

River	Sample Year	Original Lp and SD	17°C cutoff Lp and SD	18°C cutoff Lp and SD
Bush (w/ APG)	2006	0.79 ± 0.05	0.56 ± 0.05	N/C
Bush (w/ APG)	2007	0.92 ± 0.04	0.93 ± 0.03	N/C
Bush (w/ APG)	2008	0.55 ± 0.05	0.76 ± 0.06	N/C
Bush (w/ APG)	2011	0.96 ± 0.02	N/C	N/C
Bush (w/ APG)	2012	0.28 ± 0.06	0.42 ± 0.08	0.34 ± 0.07
Bush (w/ APG)	2013	0.15 ± 0.06	0.07 ± 0.03	N/C
Choptank	2013	0.47 ± 0.06	0.46 ± 0.07	0.48 ± 0.06
Choptank	2014	0.68 ± 0.06	0.46 ± 0.05	0.42 ± 0.05
Choptank	2015	0.82 ± 0.05	0.84 ± 0.06	0.81 ± 0.06
Choptank	2016	0.90 ± 0.04	0.56 ± 0.06	0.59 ± 0.05
Choptank	2017	0.40 ± 0.05	0.42 ± 0.05	N/C
Choptank	2018	0.44 ± 0.05	N/C	N/C
Choptank	2019	0.69 ± 0.06	0.68 ± 0.06	N/C
Choptank	2021	0.41 ± 0.06	0.50 ± 0.06	N/C
Mattawoman	2008	0.66 ± 0.06	0.58 ± 0.06	N/C
Mattawoman	2009	0.92 ± 0.04	0.90 ± 0.03	N/C
Mattawoman	2010	0.82 ± 0.05	0.70 ± 0.06	N/C
Mattawoman	2011	0.98 ± 0.01	N/C	0.92 ± 0.03
Mattawoman	2012	0.20 ± 0.05	0.24 ± 0.06	N/C
Mattawoman	2013	0.47 ± 0.06	0.38 ± 0.08	0.53 ± 0.06
Mattawoman	2014	0.78 ± 0.05	0.65 ± 0.06	N/C
Mattawoman	2015	1.00	N/C	N/C
Mattawoman	2016	0.82 ± 0.06	1.00	0.90 ± 0.06

Table 2-5 cont.

River	Sample Year	Original Lp and SD	17°C cutoff Lp and SD	18°C cutoff Lp and SD
Nanjemoy	2009	0.83 ± 0.05	0.74 ± 0.05	N/C
Nanjemoy	2010	0.96 ± 0.03	0.78 ± 0.05	N/C
Nanjemoy	2011	0.98 ± 0.01	N/C	0.92 ± 0.03
Nanjemoy	2012	0.03 ± 0.02	0.04 ± 0.03	N/C
Nanjemoy	2013	0.46 ± 0.07	0.26 ± 0.07	0.35 ± 0.07
Nanjemoy	2014	0.82 ± 0.04	0.86 ± 0.04	0.78 ± 0.05
Nanticoke	2007	0.55 ± 0.07	0.69 ± 0.06	N/C
Nanticoke	2008	0.19 ± 0.05	0.12 ± 0.03	0.11 ± 0.03
Nanticoke	2009	0.41 ± 0.07	0.27 ± 0.05	0.25 ± 0.05
Nanticoke	2011	0.55 ± 0.06	0.44 ± 0.07	0.52 ± 0.06
Nanticoke	2012	0.04 ± 0.02	0.04 ± 0.03	0.04 ± 0.02
Nanticoke	2013	0.43 ± 0.06	0.39 ± 0.08	0.41 ± 0.07
Nanticoke	2014	0.35 ± 0.07	0.29 ± 0.06	N/C
Nanticoke	2015	0.64 ± 0.06	0.55 ± 0.06	N/C
Nanticoke	2016	0.67 ± 0.10	0.42 ± 0.08	0.38 ± 0.07
Nanticoke	2017	0.22 ± 0.07	0.04 ± 0.04	0.22 ± 0.07
Nanticoke	2018	0.28 ± 0.08	0.21 ± 0.06	N/C
Nanticoke	2019	0.41 ± 0.06	0.40 ± 0.08	N/C
Severn	2006	0.27 ± 0.05	0.21 ± 0.04	N/C
Severn	2007	0.30 ± 0.05	N/C	N/C
Severn	2008	0.08 ± 0.04	0.06 ± 0.03	N/C
Severn	2009	0.15 ± 0.05	0.09 ± 0.03	N/C
Severn	2010	0.03 ± 0.03	N/C	N/C

Table 2-6. Summary of water quality parameter statistics for Choptank and Sassafras Rivers sampled 2021. Mean pH was calculated from H^+ concentrations, then converted to pH.

System/Year		*Temperature (°C)*	*DO (mg/L)*	*Conductivity (µS/cm)*	*pH*
	Mean	15.43	7.47	462.60	6.98
	Standard Error	0.15	0.13	85.46	
	Median	15.33	7.42	152	6.95
	Mode	15.14	7.42	128	6.94
Choptank 21	Kurtosis	-0.93	-0.56	6.10	0.02
	Skewness	-0.31	0.00	2.60	1.37
	Minimum	12.62	5.34	96	6.83
	Maximum	17.23	10.15	3326	7.43
	Count	75	75	75	75
	Mean	14.99	10.75	722.18	8.05
	Standard Error	0.20	0.19	22.69	
	Median	15.29	11.19	756	8.36
	Mode	.	12.05	791	8.02
Sassafras 21	Kurtosis	-0.20	-1.13	-0.38	-0.92
	Skewness	-0.84	-0.39	-0.40	-0.41
	Minimum	11.66	8.32	367	7.50
	Maximum	16.91	13.11	993	8.89
	Count	50	50	50	50

Figure 2-1. Subestuaries sampled for Yellow Perch larval presence-absence studies, 2006-2021. Watersheds of subestuaries sampled in 2021 are highlighted in green and have bolded text labels; watershed delineation is for Maryland only.

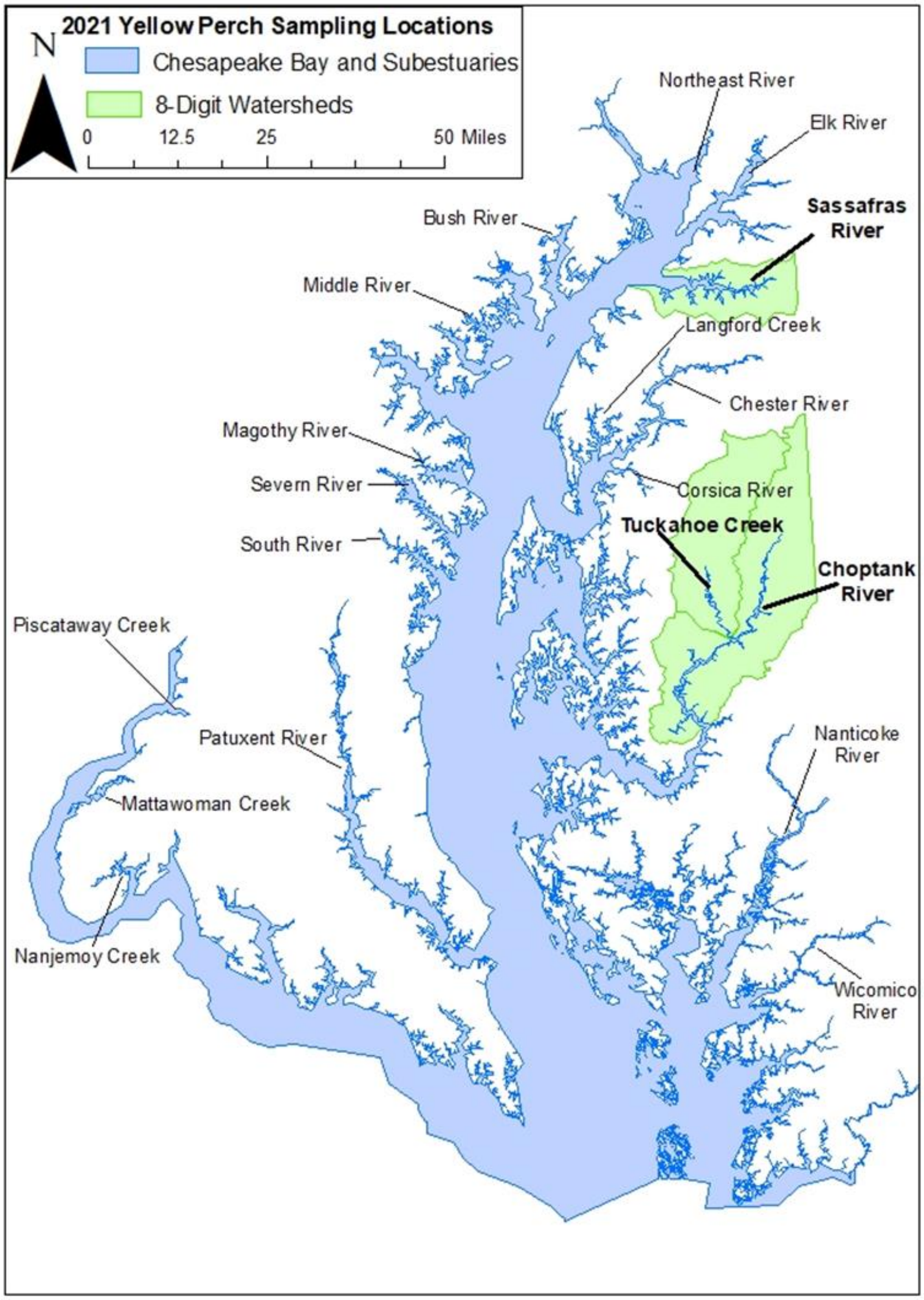


Figure 2-2. Proportion of tows with larval Yellow Perch (*L*p) and their 95% confidence intervals in systems studied during 2021. Mean *L*p of brackish Choptank River, and tidal-fresh Sassafras River, is indicated by green triangle and blue circle, respectively. Brackish subestuary *L*p threshold is indicated by a green dotted line, and tidal-fresh is blue.

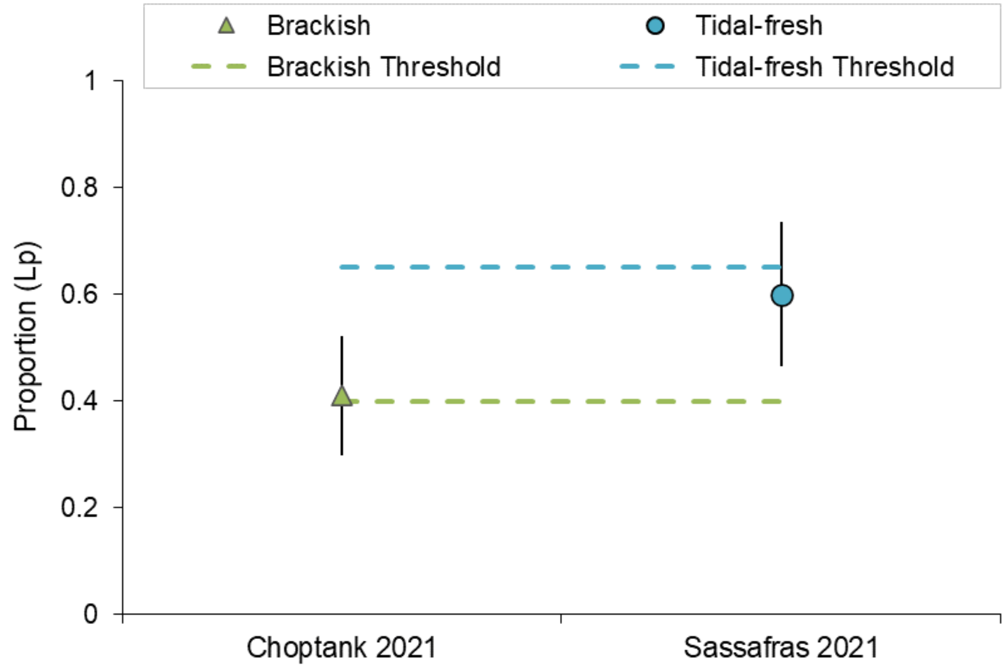


Figure 2-3. Proportion of tows with Yellow Perch larvae (*Lp*) for brackish subestuaries, during 1965-2021. Dotted line provides threshold for persistent poor *Lp* exhibited in developed brackish subestuaries. Dominant land use is indicated by symbol color (gold = agriculture, green = forest, and red = urban).

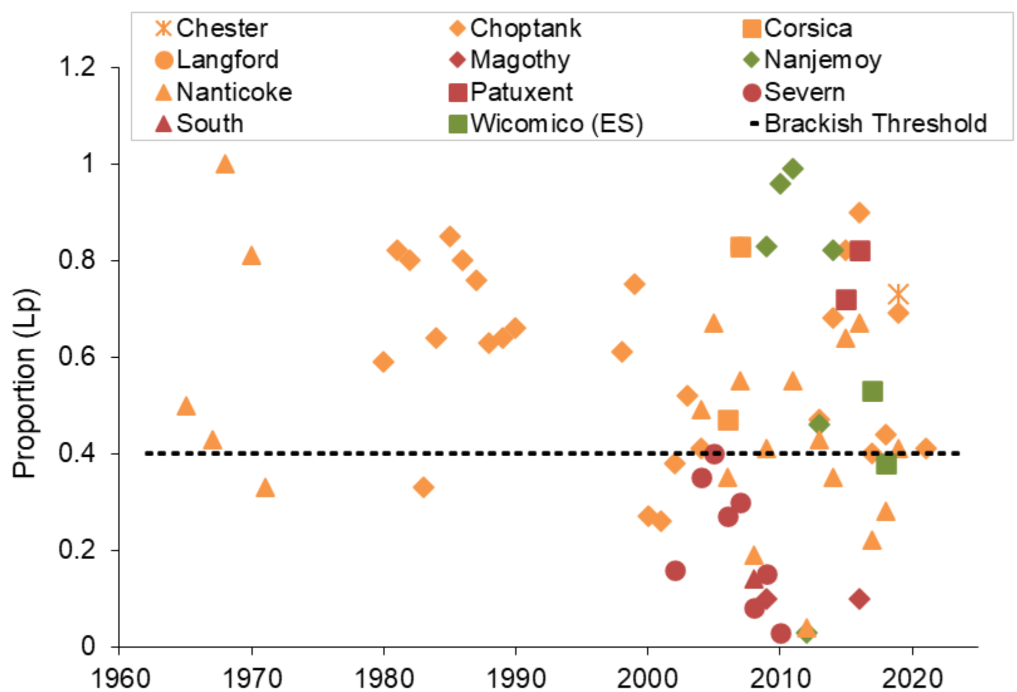


Figure 2-4. Proportion of tows with Yellow Perch larvae (*Lp*) for tidal-fresh subestuaries, during 1990-2021. Dotted line provides reference for consistent poor *Lp* exhibited in a more developed tidal-fresh subestuary (Piscataway Creek). Dominant Department of Planning land use is indicated by symbol color (gold = agriculture, green = forest, and red = urban).

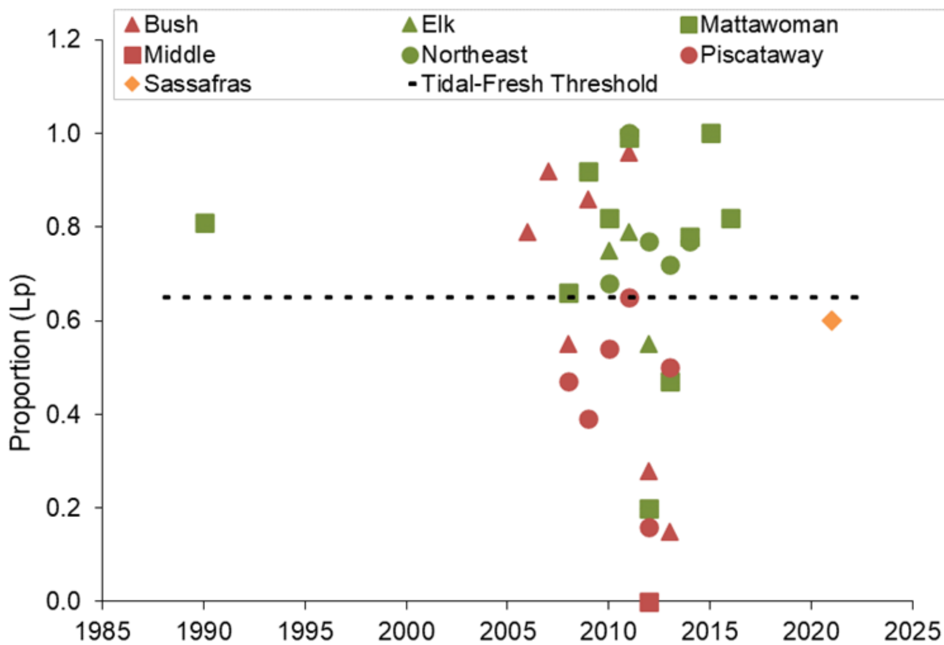


Figure 2-5. Relationship of proportion of plankton tows with Yellow Perch larvae (*Lp*) and (A) development (structures per hectare or C/ha), (B) percent agriculture, and (C) percent forest, indicated by multiple regression of fresh and brackish subestuaries combined (prediction = MR) and separate linear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

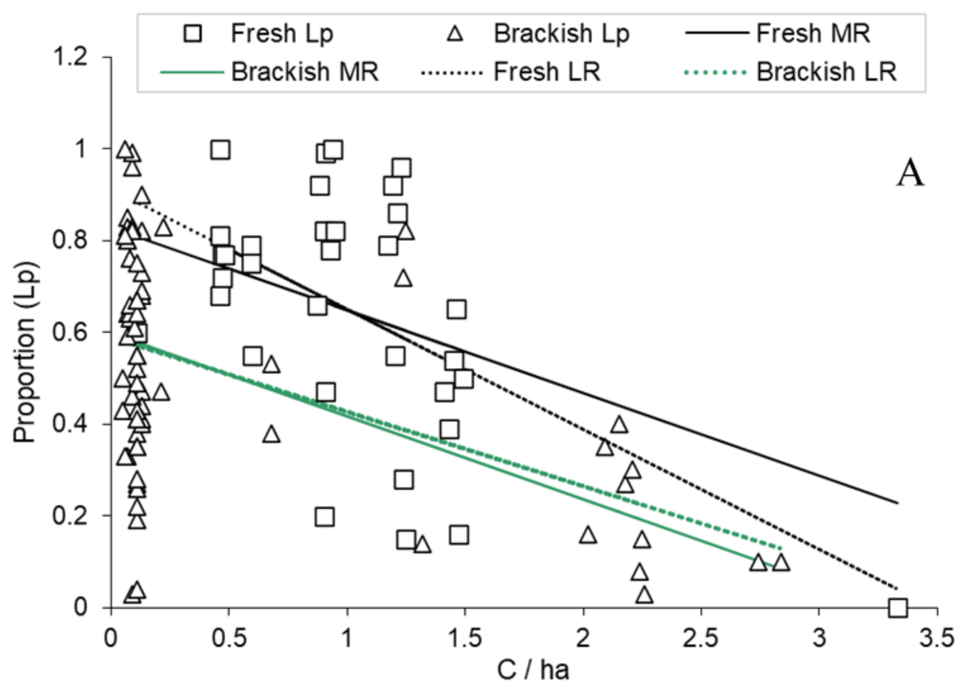
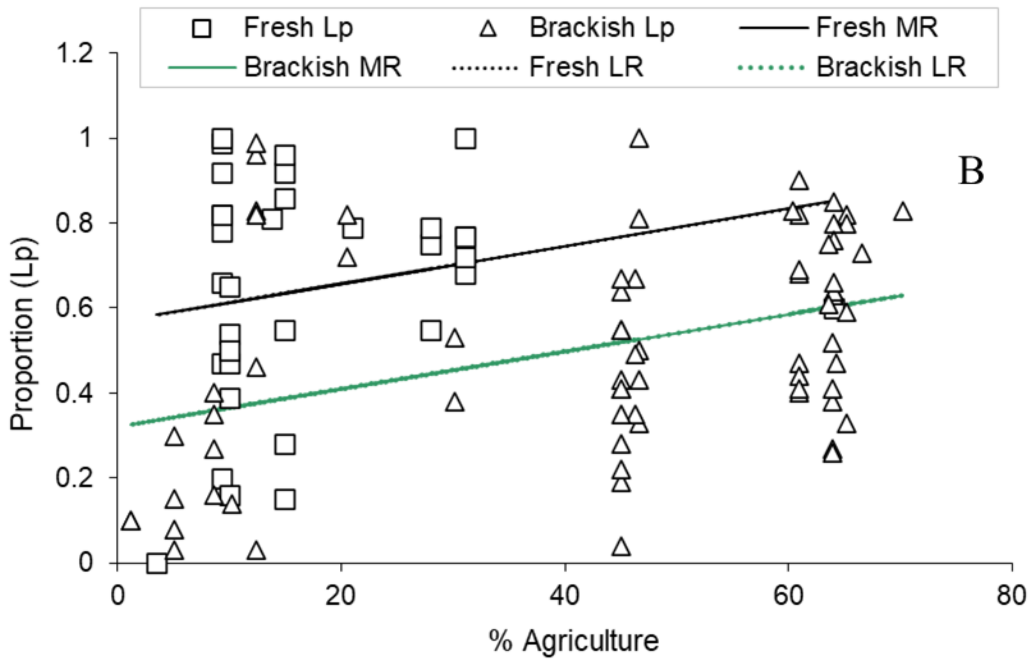


Figure 2-5 cont.



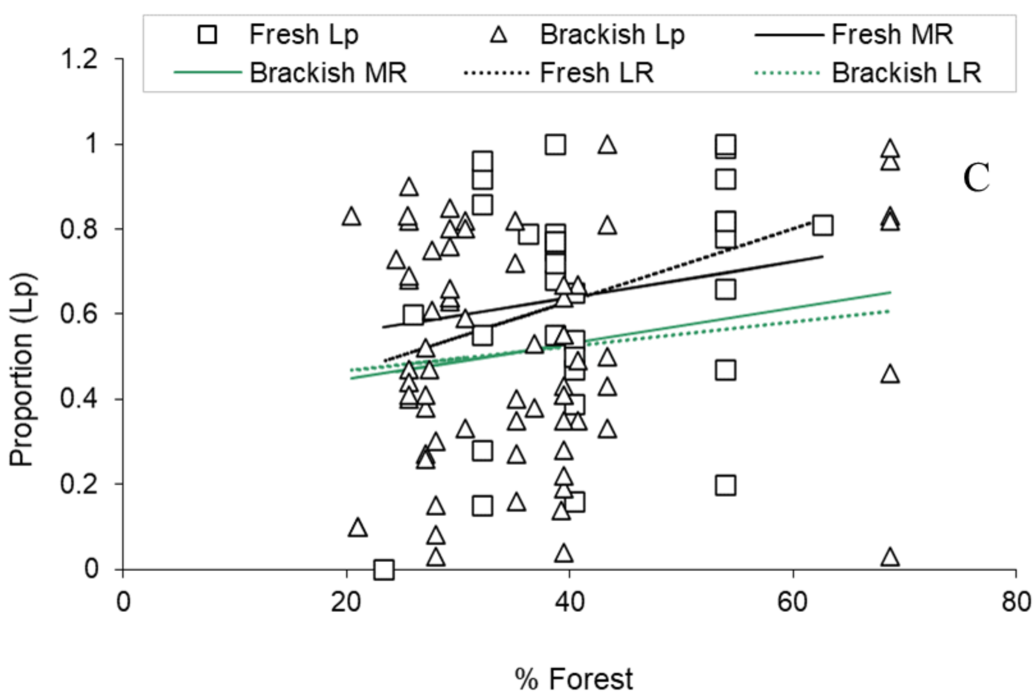


Figure 2-6. Cumulative proportion of presence of Yellow Perch larvae, collected in ichthyoplankton surveys, by 1°C temperature increments. Prior to 1994 many surveys had actual count data available, while after 1994 data was only recorded as presence-absence. All counts were converted to presence-absence for inclusion in this data set (see Section 2 in Uphoff et al. 2022 for a complete description of time-series available).

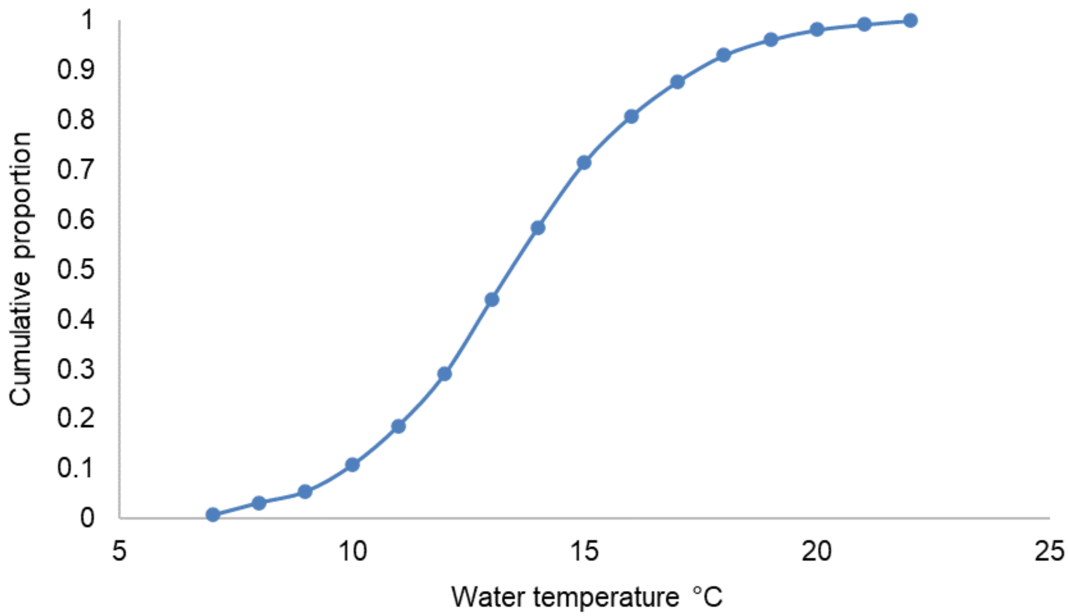


Figure 2-7. Individual values of water temperature, DO, conductivity, and pH, by date sampled, in Choptank and Sassafras Rivers during 2021.

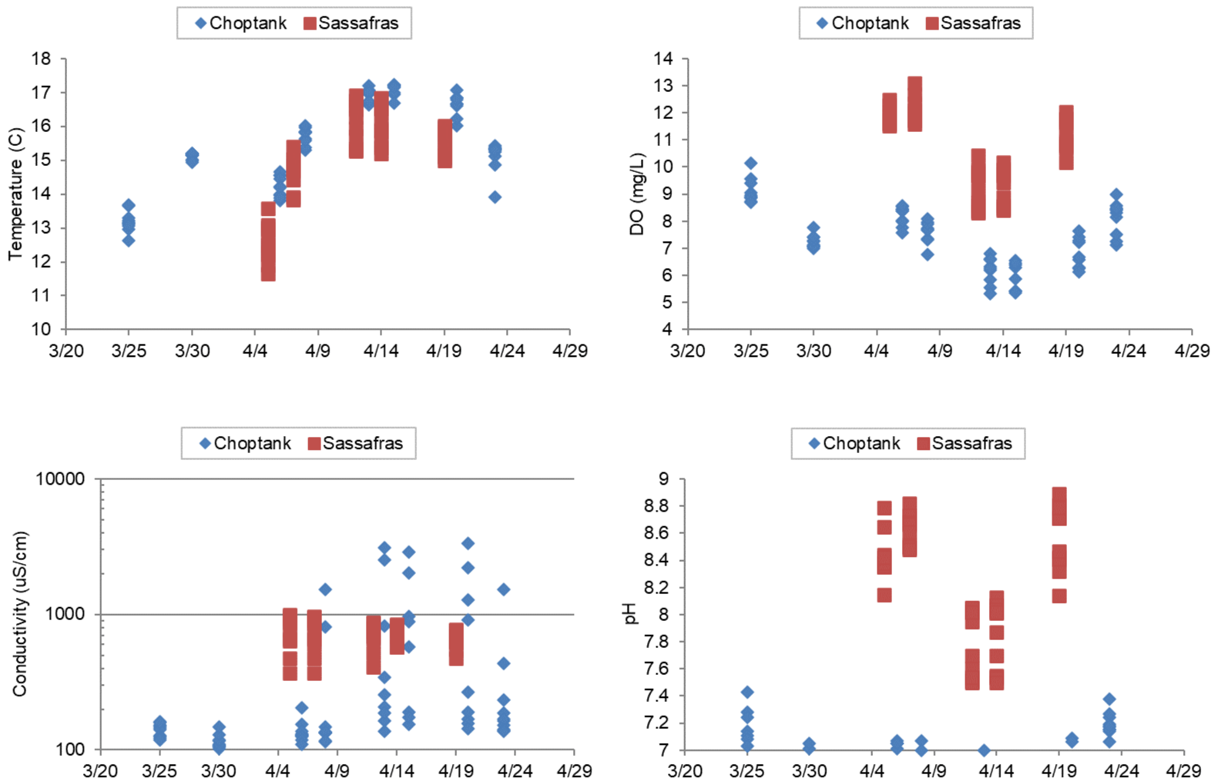
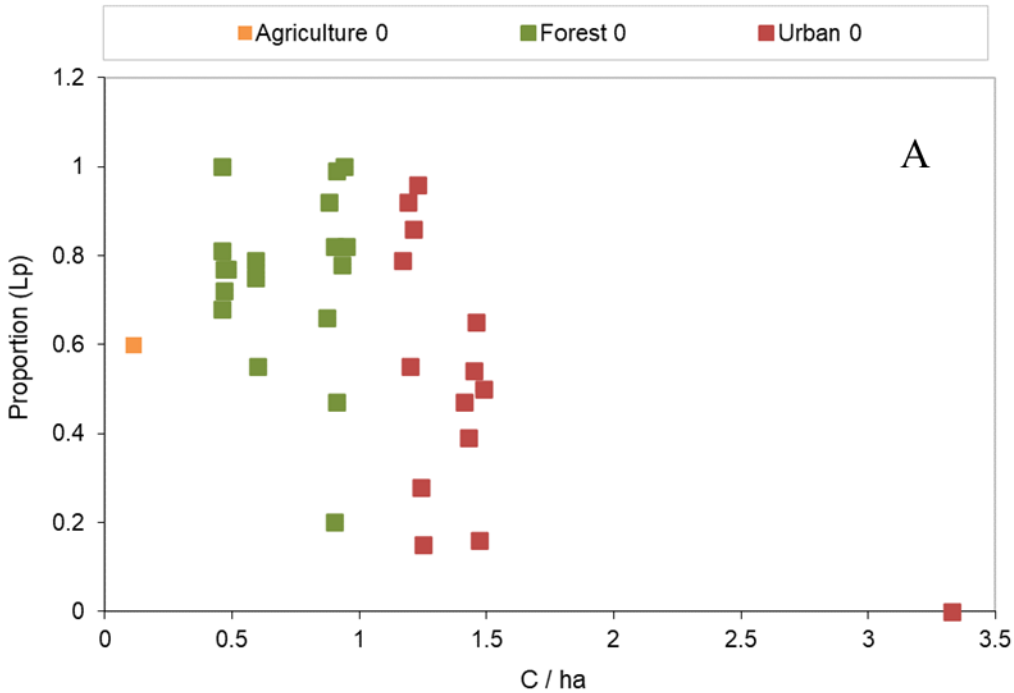


Figure 2-8. Proportion of plankton tows with Yellow Perch larvae (*Lp*) plotted against development (C/ha) with Department of Planning land use designations and salinity class indicated by symbols. Squares and a "0" behind land use indicate tidal-fresh subestuaries (A), while diamonds and a "1" indicate brackish subestuaries (B).



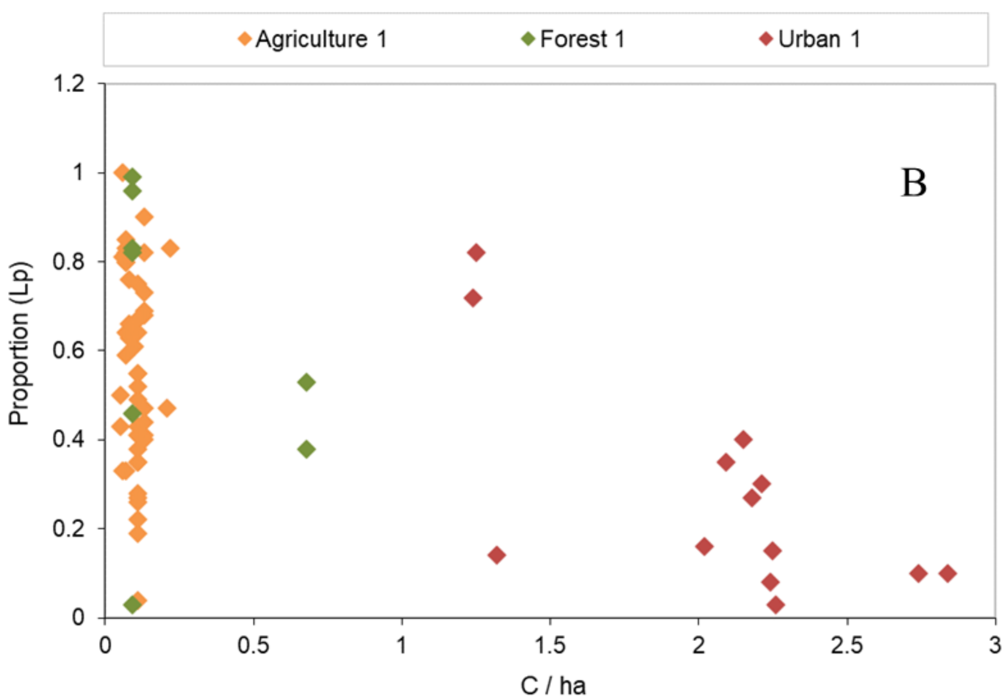


Figure 2-9. Relationship of proportion of plankton tows without detritus (OM0) and development (structures per hectare or C/ha). Dominant Department of Planning land use is indicated by symbol color (gold = agriculture, green = forest, and red = urban).

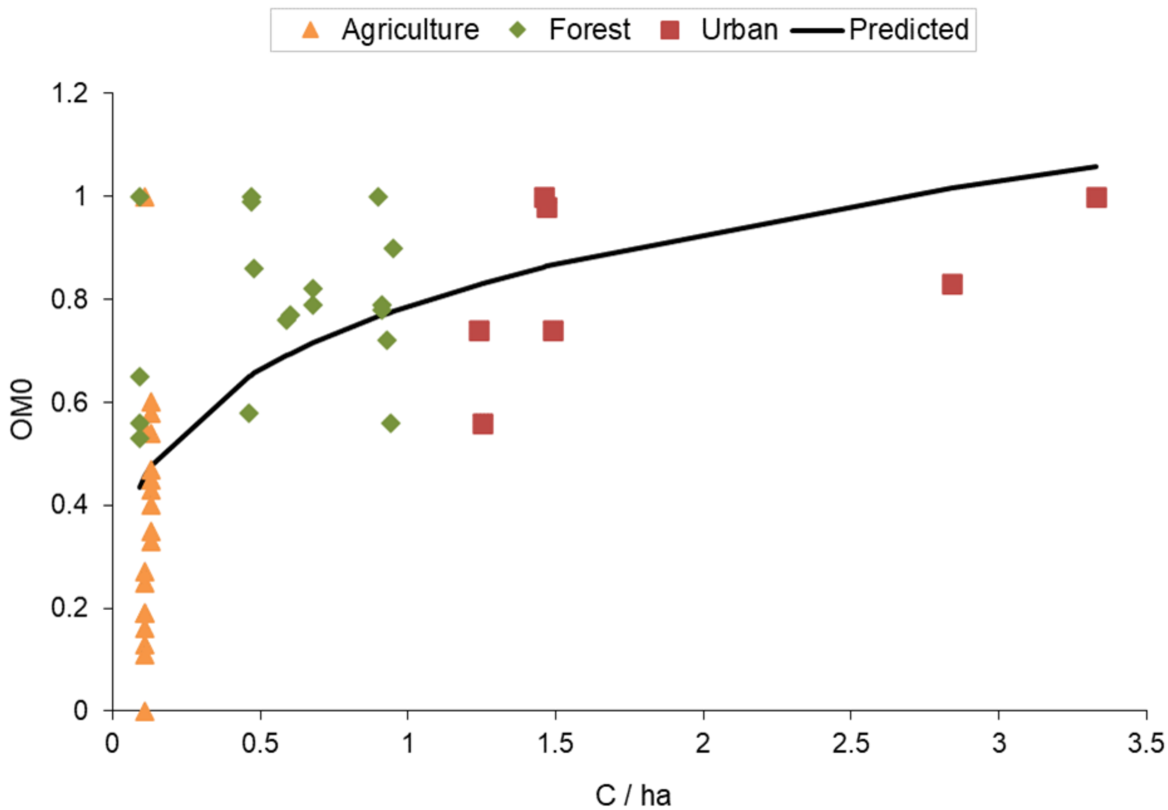
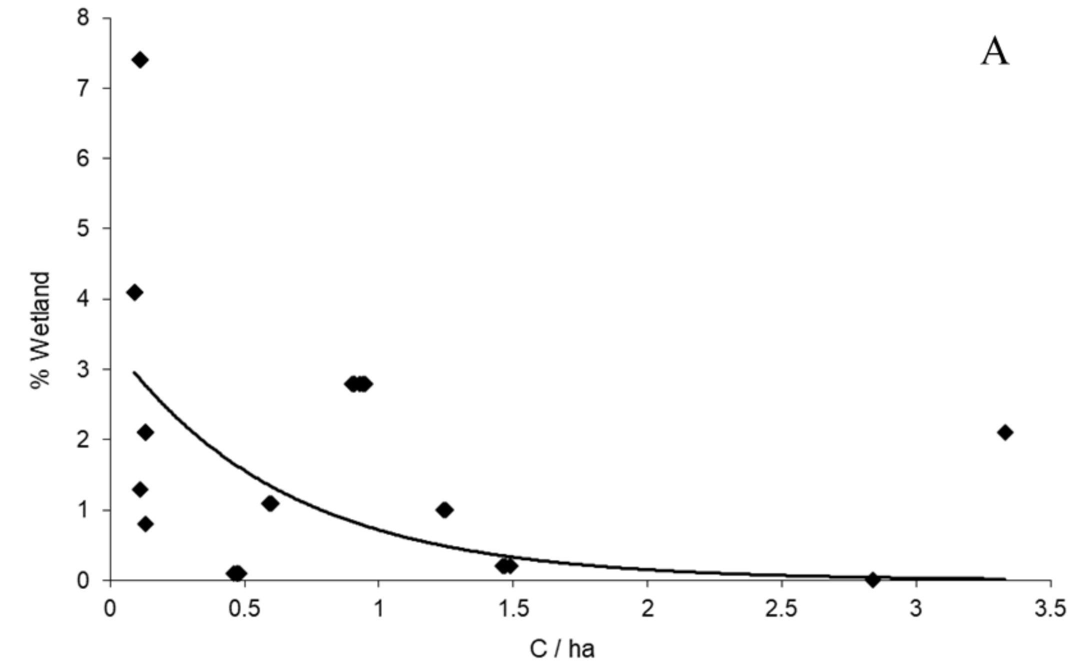
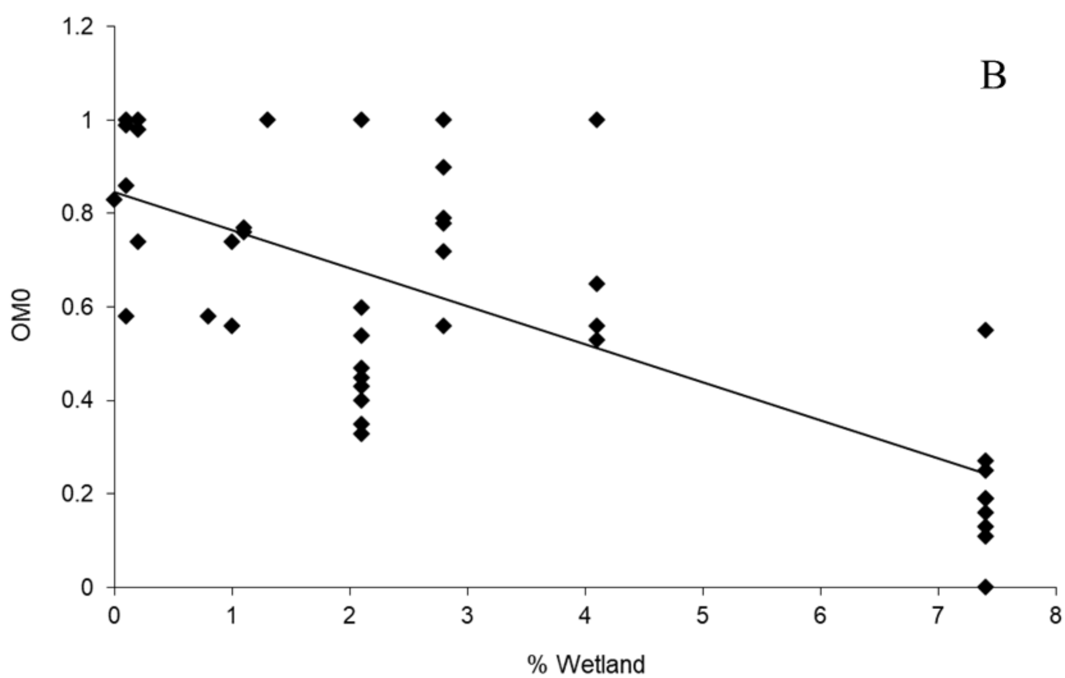


Figure 2-10. (A) Relationship of percent wetlands per watershed and level of development (C/ha). (B) Proportion of samples without organic material (OM0) and percent wetlands per watershed.





MD – Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern
Section 2.1: Investigation of Striped Bass spawning and larval habitat status in Maryland

Jim Uphoff, Alexis Park, Carrie Hoover, Margaret McGinty, and Marek Topolski

Executive Summary

In 2021, we updated the proportion of tows with Striped Bass eggs (*Ep*) and the Maryland baywide juvenile index (JI), and then estimated larval survival (RLS, baywide JI / *Ep*). Trends in pH, conductivity, and alkalinity in the Choptank River were compared between the 1980s and 2014-2021. We added analyses to detect changes in historic (1950s-present) spawning area temperature and flow patterns that influence year-class success of Striped Bass in the four major spawning areas (Head-of-Bay and Potomac, Choptank, and Nanticoke rivers).

Egg production in 2021, based on Choptank River *Ep* (0.67), was not in the top tier of estimates (roughly 0.80 or greater), but there was a high chance it was above levels during 1982-1988 when it was depleted enough to affect year-class success. Estimated RLS in 2021 was just above the poor survival criterion; most of the poor RLS estimates were concentrated in 1980-1991. Measurements of pH in Choptank River between 1986-1991 and 2013-2021 indicated improvement (higher, more stable averages and less variability of individual measurements) that would have lowered toxicity of metals implicated in poor recruitment in some Striped Bass spawning areas during the 1980s. Average alkalinity was at least 3-times higher in 2021 compared to 1986-1991. It seems unlikely that poor survival of larvae during 2019-2021 could be attributed to a return of toxic water quality conditions implicated in poor recruitment during the 1980s.

Means or medians of days between 12ºC and 20ºC water temperature milestones indicative of the beginning and ending of spawning, respectively, during 2000-2021 were 10 days to 12 days shorter (respectively) than during 1954-1992 in Choptank and Nanticoke rivers. Changes were not uniform among temperature milestones. Early milestones (first egg collected and 12ºC) appeared to be the least affected and later milestones (16ºC and 20ºC) were progressively earlier. The portion of the spawning period when most eggs were collected in historic collections with counts (days from 12ºC to 16ºC) has shortened and lethally high temperatures (indicated by days to 20ºC) were being reached earlier. In addition to these general changes, 3 years during 2000-2021 (of 9 available) had very short spans between 12ºC and 16ºC (2 days) and 2021 had the earliest date that 12ºC was reached in the entire time-series. Our temperature milestones generally captured most Striped Bass egg and larval production based on counts in historic datasets.

Below average flow conditions during 1957-2020 were less conducive to formation of strong year-classes and poor year-classes were more likely. Above average flows resulted in a higher chance that strong year-classes would be formed and a modest reduction in occurrence of poor year-classes. When all spawning areas were combined during the recent period of high productivity,1993-2020 (N = 112 area and year combinations), there were 4 strong year-classes when flows were below average and 24 strong year-classes when flow was at or above average. There were 17 poor year-classes when flow was below average and 13 when it was at or above average. When the 1993-2020 was split in half (14 years each), below average flows were less common during the first half (1993-2006) than the second in the Potomac (7 in the first half and

10 in the second), Choptank (4 and 7), and Nanticoke (5 and 8) rivers. There was no change in the Head-of-Bay (5 years of below average flow in each half).

Managing for low exploitation rates and high spawning stock would be expected to provide extended age structure that allows for diverse spawning behaviors over a protracted time period that are expected to buffer recruitment in the face of warming winter and spring temperatures. However, the time span between temperature milestones contracted in the Choptank River in the last two decades, concentrating egg production in a shorter period. When spawning is concentrated in a shorter time period, egg mortality events kill a larger proportion of a year's spawn. It is unclear whether increased egg production within this compressed spawning window can offset temperature related egg mortality numerically, but potential for more eggs to result in more larvae cannot be ruled out. However, if mistiming of zooplankton blooms with first-feeding larvae is important, it may limit successful management. In years of higher survival of eggs, larval survival and subsequent recruitment may be capped at a low level due to inadequate larval foraging no matter how many eggs are pumped into the spawning area if zooplankton production is misaligned with first-feeding larvae.

Introduction

An overfishing declaration and successive poor year-classes of Striped Bass in Maryland spawning areas during 2019-2021 have generated concern in the fisheries management and angling community. Although much of this concern has focused on the abundance of spawning stock, there has been unease expressed about degradation of Striped Bass spawning and larval nursery habitat in Chesapeake Bay (J. Uphoff, personal observation). We have assembled historical data and oriented some of our spring monitoring to respond to these concerns. This report updates efforts begun in the last two annual reports (Uphoff et al. 2020; 2022) to assess spawning and larval habitat and adds analyses of some environmental factors (river flow and water temperature) that are considered drivers of Striped Bass year-class success (Maryland Sea Grant 2009).

Year-class success of Chesapeake Bay Striped Bass is largely determined within the first three weeks of life in early spring and is a product of egg abundance and highly variable survival through the postlarval stage (Uphoff 1989; 1993; Houde 1996; Maryland Sea Grant 2009; Shideler and Houde 2014; Martino and Houde 2010; Secor et al. 2017). Spawning and larval nursery habitat (both are basically the same) is concentrated in limited fresh to low salinity tidal reaches of 16 Chesapeake Bay tributaries within the Coastal Plain; the estuarine turbidity maximum is particularly important (Hollis et al. 1967; Grant and Olney 1991; Schaaf et al. 1993; North and Houde 2001; 2003; Secor 2007; Uphoff 2008; Maryland Sea Grant 2009; Martino and Houde 2010).

Water temperature and flow conditions are important influences on year-class success of Striped Bass. Temperature may directly impact recruitment through mortality of eggs and larvae due to lethally low or high temperatures and indirectly via its influence on the timing of zooplankton blooms for first-feeding larvae (match-mismatch hypothesis), while flow may be associated with zooplankton dynamics, nursery volume, location of the nursery, advection from the nursery, and water quality and toxicity of contaminants (Hollis et al. 1967; Uphoff 1989; 1992; Secor and Houde 1995; North and Houde 2001; 2003; Maryland Sea Grant 2009; Martino and Houde 2010; Shideler and Houde 2014; Secor et al. 2017; Millette et al. 2020). Positive and negative relationships and associations of Chesapeake Bay tributary flow to Striped Bass early life stage survival and year-class success have been detected (Kernehan et al. 1981; Uphoff 1989; 1992; Rutherford et al. 1997; Martino and Houde 2010; Millette et al. 2020).

Winter-spring climate variability was considered a prime environmental driver of Striped Bass recruitment (Wood and Austin 2009) and multiple studies have cited cooler and wetter winters and springs as favorable (Maryland Sea Grant 2009; Martino and Houde 2010; Millette et al. 2020). During the past 70 years the Chesapeake Bay has experienced nearly a 2˚C rise in mean surface water temperature and long-term warming could alter timing of spawning and survival of eggs and early larvae (Maryland Sea Grant 2009; Peer and Miller 2014). Hinson et al. (2021) determined that warming in Chesapeake Bay was occurring at a more rapid rate during May-October than November-April. The seasonal split during April-May coincides with Striped Bass spawning and larval development in the Chesapeake Bay region. Modeling of the effect of likely temperature increase scenarios on Striped Bass spawning in the Hudson River from 2010 to the 2090s indicated spawning will occur earlier and be of shorter duration (Nack et al. 2019).

The Atlantic States Marine Fisheries Commission (ASMFC) has determined that Atlantic coast Striped Bass are overfished based on its most current stock assessment covering 1982-2017 (ASMFC 2019). The spawning stock biomass (SSB) estimates contain Delaware River and Hudson River stocks, but are dominated by the Chesapeake Bay stock (NEFSC 2019). High spawning stock biomass (SSB) reference points currently in use are not a product of stock-recruitment analysis, but appear to reflect an expectation that higher spawning stock will positively influence recruitment. Management of Striped Bass along the Atlantic Coast strives to achieve high SSB levels through targets and limits that reflect SSB when it was considered recovered (1995) after the period of depletion (Richards and Rago 1999; ASMFC 2003; NEFSC 2019). Based on SSB estimates from a statistical catch at age model, Striped Bass have been overfished since 2013 based on its most current stock assessment and target SSB has never been achieved (ASMFC 2019; NEFSC 2019). An egg production index independent of this model, based on egg presence-absence in Chesapeake Bay ichthyoplankton surveys, did not indicate that stock levels are low enough to impact recruitment (Uphoff et al. 2020).

Maryland has measured year-class success (recruitment) of Striped Bass in four major Chesapeake Bay spawning and nursery areas (Head-of-Bay, Potomac River, Nanticoke River, and Choptank River) with a shore zone seine survey of young-of-year juveniles since 1954 (Hollis et al. 1967; Durell and Weedon 2021) and the juvenile index (JI) has proven to be a reliable indicator of recruitment to Atlantic coast fisheries (Schaefer 1972; Goodyear 1985; Richards and Rago 1999; Maryland Sea Grant 2009). Strong year-classes failed to appear during 1971-1992, but a pattern of strong year-classes appearing every few years returned to Maryland's portion of Chesapeake Bay in 1993 (Maryland Sea Grant 2009; Durell and Weedon 2021). During 1993-2018, year-class success has been a mix of poor to strong year-classes that were characteristic of a previous period of high productivity that spanned 1958-1970 (Uphoff et al. 2020). Year-class success has been low during 2019-2021 (Durell and Weedon 2021), but has narrowly avoided an ASMFC (2003; 2010) criterion defining poor year-class success.

Uphoff (1993; 1997) used historical ichthyoplankton survey data to develop a Striped Bass egg presence-absence index (*Ep* or proportion of samples with eggs) of spawning stock status during 1955-1995 in Maryland spawning areas. An *Ep* time-series has been maintained through 2021, although it became a low priority in the 2000s as catch-at-age modeling became the primary stock assessment method (Uphoff et al. 2020). An index of relative larval survival, the ratio of the juvenile index to *Ep* (RLS = JI / *Ep*), was used for retrospective examination of the relative importance of egg and larval habitat on Striped Bass year-class success (Uphoff et al.

2020). Patterns in this ratio provided an indication of changes in egg and larval habitat conditions without specification of the myriad factors (water quality variables, food availability, water temperature, etc.) that determined habitat suitability (Uphoff et al. 2020).

Toxic water quality conditions encountered by striped bass larvae were implicated in episodic mortalities in some spawning areas in the 1980s and 1990s (Uphoff 1989; 1992; Hall et al. 1993; Richards and Rago 1999). Since 2014, we have collected basic water quality data (temperature, conductivity, dissolved oxygen or DO, and pH) on the spawning grounds of several Striped Bass spawning areas as we investigated the impact of urbanization (Uphoff et al. 2020). During 2021, we added alkalinity to the suite of water quality variables sampled on the Choptank River spawning grounds. Low survival of Striped Bass postlarvae during 1980-1988 in the Choptank River estimated from ichthyoplankton surveys was associated with low pH, alkalinity, and conductivity that could have influenced toxicity of metals (Uphoff 1989; 1992). Water quality in Choptank River ichthyoplankton surveys (Uphoff 1992) was consistent with descriptions for in situ and on-site toxicity tests conducted in Choptank and Nanticoke rivers during 1984-1990 (Hall et al. 1993). Acidic conditions and toxic metals (Al, Cu, Zn, Cd, Cr, Pb, and As) were associated with high mortality of Striped Bass larvae in bioassays conducted during 1984-1990 in Choptank and Nanticoke rivers (Hall et al. 1993; Richards and Rago 1999).

C. Hoover mined historical reports and Maryland DNR data sheets to create a spreadsheet with georeferenced data on distribution of anadromous fish eggs and larvae (Striped Bass, White Perch, Yellow Perch, and Alosids) and water quality in Maryland's Striped Bass spawning areas (Uphoff et al. 2022). Most of this information was focused on Striped Bass. Water quality parameters available varied, but were generally confined to temperature (˚C), salinity (‰), and tide stage until the early 1980s. During the 1980s and after, dissolved oxygen (DO; mg/L), pH, and conductivity (µS/cm) were monitored more routinely (Uphoff et al. 2022).

Uphoff et al. (2020) examined long-term (1950s to present), concurrently collected water temperature and egg distribution data from some, but not all spawning areas contained in the data set compiled for Uphoff et al. (2022). This examination suggested that water temperature (21˚C) indicative of the end of spawning and-or poor survival of hatched larvae was occurring earlier in recent years. Temperatures approaching and exceeding 21˚C fall on a rapidly ascending limb of instantaneous daily mortality rates of larvae that would negate benefit from late spawning (Secor and Houde 1995). There appeared to be a general upward shift in Choptank River spawning area average water temperature between 1986-1991 and 2014-2019 during a standard period (April 1 – May 8) used for comparisons. The 21˚C cutoff was sometimes breached later in the 1950s and 1978-1979 than during the 1990s or 2015-2019 in Patuxent River and Chester River, but not in Wicomico River (Uphoff et al. 2020). In this report, we looked at temperature patterns for the two spawning areas with the most extensive time-series, the Choptank and Nanticoke rivers.

We examined four spawning milestones that we felt were reasonably straightforward to interpret: date that the first egg was collected, and the dates when 12ºC, 16 ºC, and 20ºC were consistently met. We used the cumulative distribution of eggs or larvae collected by temperature or salinity increment to evaluate the cut offs used to estimate *Ep* and evaluate temperature milestones. Spawning in Chesapeake Bay rivers generally occurs between 12ºC and 23 ºC (Peer and Miller 2014), but temperatures above 21ºC are generally not suitable (Uphoff 1993). Secor and Houde (1995) found temperature oscillations had an important influence on egg production when they fluctuated in the range l0°C-20°C. Episodic mortalities of eggs and newly hatched larvae occurred when temperatures fell below 12 ºC (Uphoff 1989; Rutherford and Houde 1995; Peer and Miller 2014). Olney et al. (1991) reported that for most years, peak egg production in the Pamunkey and Rappahannock rivers occurred with rising temperatures between 15°C and 18°C. Cohort-specific mortality rates of Striped Bass larvae are strongly temperature dependent, with both early (<14 ºC) and late (>21 ºC) cohorts experiencing higher mortality (Secor and Houde 1995; Peer and Miller 2014). We selected 20ºC as an upper temperature boundary since egg presence-absence surveys sometimes cut off sampling just prior to when 21ºC was anticipated to occur; 16ºC represented the midpoint of the range and was a temperature where larval cohort survival was expected to be high (Secor and Houde 1995).

Uphoff et al. (2020) explored the long-term (1957-2019) influence of Choptank River March-April flow on $\log_e$-transformed JIs and a weak relationship was found. Patterning of residuals indicated the relationship was not stable over time with sets of years having stronger or weaker responses to flow. A particularly positive shift in the relationship of flow and the Choptank River JI was reflected by frequent strong year-classes during 1993-2007. The most recent period that started in 2008 coincided with lower flows in April and, while strong year-class have occurred (2011 and 2015), they appeared less frequently than in 1993-2007. We expanded this analysis to include all four spawning areas with JIs and explored relationships for both long-term (1957-2020) and the most recent period of high productivity (1993-2020) identified by Uphoff et al. (2020). Ichthyoplankton studies and modeling of Striped Bass egg and larval dynamics in Chesapeake Bay spawning areas during this period have linked recruitment success to higher river discharge (Secor and Houde 1995; North and Houde. 2001; 2003; North et al. 2005; Martino and Houde 2010; Secor et al. 2017; Millette et al. 2020). The long-term data set would be subject to extra variability due to shifts in productivity and low spawning stock; these impacts would be minimized during 1993-2020.

In summary, we updated the following metrics developed in Uphoff et al. (2020) through 2021 in this report: *Ep*, JI, RLS, temperature, DO, pH, salinity, and conductivity. Covid restrictions prevented other sampling during 2020 and only the JI was available. Alkalinity, measured in the Choptank River during 2021, was added. We added analyses to detect changes in spawning area temperature and flow patterns that may be influencing recent year-class success of Striped Bass in the four major spawning areas. Details of previous work that was updated in this report can be found in Job1, Section 2.1 of Uphoff et al. (2020). A description of the compiled Striped Bass spawning area data can be found in Project 1, Section 2 of Uphoff et al. (2022).

Methods

Study area **-** Maryland's portion of Chesapeake Bay contains 11 Striped Bass spawning areas (4 more are in Virginia; Olney et al. 1991), comprising an estimated 57,448 ha (Figure 2.2.1; Hollis et al. 1967). The entire Chesapeake Bay has a surface area of 1,160,000 ha (Malmquist 2009). On an egg production basis, Maryland's spawning areas were estimated to produce approximately 69% of the Chesapeake Bay total (Uphoff 2008).

The four largest Maryland spawning areas are sampled for the MD JI: Head-of-Bay (drowned river valley of the Susquehanna River, 27,225 ha), Potomac River (22,162 ha), Nanticoke River (3,034 ha), and Choptank River (1,734 ha); remaining spawning areas in Maryland are 23-1,011 ha (Hollis et al. 1967). These four largest spawning areas comprise 94% of Maryland's total surface area (Hollis et al. 1967). Two Maryland spawning areas, Patuxent and Potomac rivers, are located on the west side of Chesapeake Bay, the Head-of-Bay is in the center and is furthest north, and remaining spawning areas are on the east side (Figure 2.2.1).

Proportion of ichthyoplankton tows with Striped Bass eggs (Ep) 2021 update – Surveys included in the time-series were considered to have covered most to all of the spawning season and spawning area through multiple sampling events. We confined analysis to spawning areas sampled for the Maryland juvenile index to view status and trends (Choptank River, Head-of-Bay, Potomac River, and Nanticoke River; Hollis et al. 1967; Durell and Weedon 2021). Elk River was considered a proxy for the Head-of-Bay when the latter spawning area was not entirely sampled. Previously summarized Striped Bass ichthyoplankton surveys (1955-2019; Uphoff 1997; Uphoff et al. 2020) were used as a starting basis for the *Ep* time-series. Stratified random sampling designs for Choptank, and Nanticoke rivers used to sample for *Ep* were described in Uphoff (1997) and these designs were used during 2021.

Surveys prior to 1994 varied in tow durations, tow configuration, and net and mesh sizes (Uphoff 1993; 1997; Uphoff et al. 2020). Surveys to estimate *Ep* during 1994-2021 were standardized to techniques of the longest running early time-series (Nanticoke River, 1955-1981; Uphoff 1997). These surveys used 2-minute tows made against the current at the surface with a 0.5-m diameter plankton net made of 0.5 mm Nitex mesh and a 3:1 length-to-mouth diameter ratio. If eggs were readily seen in a sample during or after processing, the sample was discarded, and presence of eggs was recorded. If a sample was fully rinsed and the sampler was confident that eggs were absent, it was discarded and absence of eggs was recorded. In these cases, the net was rinsed thoroughly without a jar before taking the next sample. If a sample had been completely processed and the sampler was unsure if eggs were present or not, the sample was preserved in 5-10% buffered formalin, rose bengal stain was added to aid detection, and it was sorted in the laboratory.

Sample trips during 1994-2021 were usually made twice per week, spaced 2-4 days apart. Sampling was conducted until a 21˚C water temperature cutoff criterion was met (Uphoff 1993; 1997; Uphoff et al. 2020) or was very likely to be met before the next scheduled sampling visit based on water temperature and forecast air temperatures. In a few years, persistent cool temperatures during late spring did not allow water temperatures to rise above 21˚C for a long period even though egg catches had tapered off and a judgement was made to discontinue sampling. Sites with greater than 2.0‰ salinity usually were randomly replaced within the same sample strata (if possible) by lower salinity sites during sampling to minimize including non-spawning habitat (Uphoff 1997). More than 99% of Striped Bass eggs collected (and counted) in Choptank River during 1980-1985 were collected at 2.0‰ salinity or less (Uphoff 1989). Historic field collections were not subject to these criteria and they were applied during analysis.

We restricted analysis to collection dates between the first sample containing an egg and when water temperature reached 21°C. Sites with salinity greater than 2.0‰ and stations past outer boundaries where eggs were not collected during an entire season were excluded to minimize zeros representing non-spawning habitat (Uphoff 1993; 1997). Stations where eggs were not collected located between stations where eggs were present were included in analyses.

The proportion of tows with one egg or more and its 90% confidence interval were estimated using the normal distribution to approximate the binomial probability distribution (Ott 1977). This approximation can be used when the sample size is greater than or equal to 5 divided by the smaller of the proportion of positive or zero tows (Ott 1977). Surveys that did not meet this sample size requirement were not included. The proportion of tows with eggs was estimated for each spawning area and year, and for an annual baywide estimate (described below) as:

$$(1)\ Ep = N_{present} / N_{total};$$

where $N_{present}$ equaled the number of qualifying samples with Striped Bass eggs present and N_{total} equaled the total number of qualifying samples. The SD of *Ep* was estimated as:

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

Ninety percent confidence intervals were constructed as:

$$(3)\ Ep \pm (1.645 \cdot SD);\ \text{(Ott 1977).}$$

In cases where cool temperatures persisted and sampling ended before 21˚C, we calculated overall mean *Ep* for all dates sampled, recalculated each mean (*j*) with each sample date (*i*) excluded, Ep_{ji}, and then examined the distribution of Ep_{ji} to judge influence of a single date. A late sample date that represented an outlier was expected to noticeably depress Ep_{ji} lower than combinations of sample dates preceding it and the date prior was used as the terminal date. If late dates did not represent an outlier, estimates of Ep_{ji} were expected to be distributed evenly above and below *Ep* and these dates would be included.

Uphoff (1997) concluded that *Ep* in one or more spawning areas could represent baywide spawning stock status since consistent differences in tow times, net diameters, and spawning areas were not detected (Uphoff 1997). We pooled available annual data from these spawning areas to estimate baywide *Ep* using equation 1, its SD using equation 2, and its 90% CI using equation 3. Five Elk River surveys were redundant with Head-of-Bay surveys and were not used to estimate baywide *Ep*.

Juvenile index 2021 update - We used annual geometric mean catches of Striped Bass juveniles per standard seine haul at permanent stations in Head-of-Bay, and Potomac, Choptank, and Nanticoke rivers (combined) as the juvenile index (JI; Durell and Weedon 2021). Baywide (Maryland's portion of Chesapeake Bay) and spawning area specific JI's were available online from the MD DNR *Juvenile Striped Bass Survey* website https://dnr.maryland.gov/fisheries/pages/striped-bass/juvenile-index.aspx ; we converted the 95% CI's provided to 90% CI's.

The JI was derived annually from sampling at 22 fixed stations within Maryland's portion of the Chesapeake Bay (Durell and Weedon 2021). There were seven stations each in the Potomac River and Head-of-Bay and four each in the Nanticoke and Choptank Rivers. Two seine hauls, a minimum of thirty minutes apart, were taken at each site on each sample round. Sampling occurred during July prior to 1962 (44 samples per year), during July and August during 1962-1965 (88 samples), and during July, August, and September after 1965 (132 samples; Durell and Weedon 2021).

Relative Larval Survival (RLS) 2021 update - I used the JI and baywide *Ep* to estimate annual relative larval survival (RLS) during 1957-2021 as:

$$(4)\ RLS = JI / \text{baywide } Ep\ \text{(Uphoff et al 2020).}$$

Estimates of the JI concurrent with Ep were available for 1957-2021 (Durell and Weedon 2021). The baywide *Ep* time-series started in 1955 and continued through 2021; estimates were not available for 1958-1960 and 2020.

Confidence intervals (90%) were developed for RLS ratios using an Excel add-in, @Risk, to simulate distributions reported for numerators and denominators using Latin Hypercube sampling to recreate input distributions (Palisade Corporation 2016). Each annual RLS estimate was simulated 5,000-times. Annual means and SDs of *Ep* were used for the denominator in simulations. Juvenile indices, based on geometric means, were back-transformed into the mean of $\log_e$-transformed catches (+1) and its SE was used. Geometric means were recreated for the numerator for each simulation (Uphoff et al. 2020).

The Striped Bass management plan specifies a criterion for recruitment failure as three consecutive years of juvenile indices lower than 75% of all other values in the dataset during 1957-2009 (lowest quartile; ASMFC 2003; 2010). We used the same series of years to develop criteria for low and high RLS. We adopted the lowest quartile of RLS during 1957-2009 as a criterion for poor egg-larval survival. Conversely, we chose the upper quartile as an indicator of high egg-larval survival; a strong year-class criterion is not suggested in the management plan. The probability of falling below the poor larval survival criterion was estimated by using the RLS mean and SD to estimate its cumulative probability distribution and the proportion below the criterion was an estimate of risk. The probability of meeting or exceeding the high larval survival criterion was estimated by using the RLS mean and SD to estimate its cumulative probability distribution and the proportion above the criterion was an estimate of this probability.

We expressed deviations between the relative status indicated by the JI and RLS by standardizing each variable by its time-series mean. This deviation was expressed for each year as:

$$(5)\ (SJI_t - SRLS_t) / SJI_t;$$

where SJI_t is the standardized juvenile index in year *t* and $SRLS_t$ is standardized RLS in year *t*.

Water quality 2021 update – Choptank River and Nanticoke River were sampled during 2021; FHEP sampled the Choptank River, while the Anadromous Fish Project sampled the Nanticoke River in addition to their regular survey. Measurements of water temperature (ºC), pH, dissolved oxygen (mg/L), conductivity (µS/cm), and salinity (‰) were made at the surface during each site visit with a YSI model 556 water quality multimeter during 2014-2021. In 2021, FHEP supplied the Anadromous Fish Project with a YSI 556 that could measure pH. These meters were calibrated frequently. The Choptank River is turbulent and did not show signs of stratification during 1983-1991 surveys (J. Uphoff, personal observation), so surface measurements should have been comparable to those at multiple depths. This observation would apply to the Nanticoke River as well.

During 2021, an additional water quality parameter, total alkalinity (mg/L $CaCO_3$), was measured in Choptank River using a YSI 9500 Photometer. The Photometer was calibrated for use with YSI photometer color standards, and the transmittance test (program 000), just prior to the beginning of the season. The color standards came with a sheet which provided certified transmittance values, and as long as the photometer produced a similar result (within a specific +/- margin of error), it was working properly. Water samples were collected just below the surface (0.5 m) in Nalgene bottles that were triple rinsed on location before the final collection was made. Bottles were kept in a small cooler while sampling was being conducted, and total alkalinity was measured within 24 hours after collection. Bottles were shaken prior to removing a 10 ml sample, which was then added to a round glass test tube for processing. All collections were free of debris and particulates, so “blanks” were made using the same water from each site just prior to the reagent being added. After reading the blank, one total alkalinity tablet (Alkaphot) was crushed and mixed into a sample until all particles had dissolved. Samples were allowed to stand for exactly one minute before remixing and were then read immediately using the Phot 2 program on the Photometer. The YSI Photometer 9500 has a minimum detection limit of 10 mg/L, a working range of 0-500 mg/L, and a tolerance of ±7 mg/L at 200 mg/L for the total alkalinity test.

Water quality analyses were split into two categories. The first examined changes in pH, total alkalinity, and conductivity. These variables were associated with toxic conditions encountered by larvae in the 1980s in the Nanticoke and Choptank rivers during the 1980s (Hall et al. 1993). The second, described in its own section (below), looked at long-term changes in water temperature on the spawning grounds of these two rivers.

Water quality surveys were conducted in Choptank River spawning area during 1983-1985, but they focused on fewer fixed stations that did not span the spawning area (Uphoff 1989; 1992). After 1985, sampling spanned the entire spawning area. Four fixed stations were sampled in Choptank River during 1986 and the stratified random design described in Uphoff (1997) was employed afterwards (Uphoff 1992). Choptank River data for 1980-1991 existed in a data base in a format that had not been supported for years; documentation for the data base was scanty but water quality data was extracted from it. Historical summaries were developed for Nanticoke River in 1992 and 1993 from water quality measurements reported in Houde et al. (1996).

Summary water quality statistics included mean, median, minimum, maximum, and the interval encompassing 90% of measurements over a standard time period usually relevant to eggs, prolarvae, and postlarvae (measurements available during April 1-May 8; Uphoff 1989; 1992; Houde et al. 1996) and relative area (salinity $\leq$ 2.0 ‰). Means and medians would provide some indication of chronic conditions, while maximums and minimums would capture acute conditions. The 90% data interval would provide an indication of how extreme minimums and maximums were. Estimates of pH were converted to H+ concentration to estimate the mean and then converted to mean pH.

Salinity – To evaluate the 2.0‰ salinity cut off used to estimate *Ep,* we used historic Choptank River or Nanticoke River surveys with counts of eggs to determine cumulative distributions with salinity for each river. Salinity increments were in units of 0.5‰ (for example, the increment at 0‰ consisted of sums for all samples from 0 to 0.499‰). Two types of numeric data were present, counts and densities; densities were not expressly delineated in the data set, but the presence of decimals was considered an indicator. (A count or density indicator has since been added to the dataset). We confined analyses of egg distribution with salinity to data based on counts.

Water temperature – We explored how egg and larval abundance responded to water temperature by using past surveys with counts of eggs and larvae (prolarvae and postlarvae combined) to determine cumulative distributions of their respective counts with water temperature. Water temperature increments were in units of 1ºC (for example, the increment at 16ºC consisted of sums for all samples from 16.0 to 16.99ºC). Sample sizes within increments varied, but the trend was expected to be reasonably representative. Cumulative sums for each increment were expressed as a percent of the total. Increments with most rapid growth in cumulative percent were considered to represent important temperatures for eggs or larvae. Choptank River and Nanticoke River data were analyzed separately.

Larval analyses were confined to surveys employing gears capable of capturing prolarvae, and postlarvae. Choptank River data from 1980-1989 were used and methods were described in Uphoff (1989; 1992). Nanticoke River data were from 1992 and 1993 and methods were described in Houde et al. (1996) and Secor et al. (2017). These distributions were compared to the temperature milestones (described below).

We examined four spawning milestones in the Choptank River and Nanticoke River time-series that were reasonably straightforward to interpret: date that the first egg was collected, and the dates when 12ºC, 16 ºC, and 20ºC were consistently met. All dates were expressed as days from April 1 (day = 0). To be considered consistent, temperatures could not be single, isolated measurements; a date with multiple readings at milestone would be selected. Intervals between sampling visits had to be no more than weekly for a survey to be included. In some cases, sampling from a single site was all that was available (a few years in the Choptank River), but most surveys had multiple sites spanning most or all of the spawning area. Measurements from the upper reaches of the spawning grounds were sometimes rejected since these areas warm quickly before detectable spawning activity. Dates indicating when the first egg was detected or 12ºC or 20ºC were consistently met had to be preceded by one day without eggs detected or lower temperatures, respectively. These criteria were not met in all years, so time-series varied among milestones.

Surveys from the Nanticoke River during 1954-1981, 1985, 1989, 1992-1994, 2004-2019, and 2021 were used (Uphoff et al. 2022). The Choptank river time-series consisted of 1954, 1957-1962, 1980-1989, 1994, 1997-2004, 2013-2019, and 2021 (Uphoff et al. 2022). J. Uphoff carefully examined spreadsheets containing either Nanticoke River or Choptank River time-series by eye and determined the first eligible date for each criterion. These dates were plotted against year to determine trends. Choptank River and Nanticoke River data were combined for these summaries and plots. These two spawning areas are adjacent to each other in the Coastal Plain. These plots did not seem to exhibit a continuous change over the time-series and a threshold around 2000 was suggested. We estimated the median date for a milestone for each year through 2021 and then examined the frequency that dates exceeded or fell below the median prior to 2001 and after 2000. We used a one-way t-test for equal variances to assess the hypothesis that there was no difference in the mean number of days at a particular temperature milestone before 2000 and after (Dowdy and Weardon 1991). The alternative hypothesis was that days to a particular milestone were less after 2000 than before 2000. An F-test was used to test for equality of variances in cases where the equal variance assumption seemed questionable (Dowdy and Weardon 1991).

Flow – Two approaches were used to explore how spring flow may have influenced Striped Bass recruitment. One approach used correlation analysis to examine the strength of linear associations of spring flow with the area-specific Striped Bass JI's (Head-of-Bay, Potomac River, Choptank River, and Nanticoke River). The second was probability based, examining the frequency of strong or poor year-classes in relation to average long-term flow levels. This latter approach did not depend on linear dynamics.

We considered March-May monthly flow to be most likely to influence recruitment since these months immediately precede and encompass spawning and larval development periods. We examined two time-series: one that covered the entire JI time-series available in Durell and Weedon (2021; 1957-2020; hereafter, the long-term flow data) and a second that was identified in Uphoff et al. (2020) as a recent period of high productivity, 1993-2020). Long-term correlations had a greater possibility of the JI – flow association reflecting additional factors affecting productivity (low stock size and toxic water quality were possible influences during portions of 1971-1992; Uphoff et al. 2020). Monthly average flow for each year (in cubic feet per second or CFS) were obtained from the US Geological Survey gauging stations at Marietta, PA (Susquehanna River), for the Head-of Bay; Little Falls, MD, for the Potomac River; Greensboro, MD, for the Choptank River; and Bridgeville, DE, for the Nanticoke River from the National Water Information System: Web Interface (https://waterdata.usgs.gov/). Correlations of the long-term area specific JIs with March, April, or May average flow were used as a basis for determining which months were included in subsequent analyses. We were looking for two-month combinations that immediately preceded and included the bulk of spawning activity for subsequent analyses (correlation and probability) with JIs. We used correlation analysis to examine associations in spawning season trends in flow among the four spawning areas and the associations among area specific JIs.

The probability-based approach looked at each spawning area separately during 1993-2020. Each set of spawning season flow records were standardized to their 1957-2020 means to provide similar scales. Area-specific JIs were plotted against standardized flows and classified into poor, strong, or in-between year-classes. The frequency of poor, strong, or in-between year-classes occurring below or at and above average flow (standardized flow = 1.0) was determined for each spawning area. The Striped Bass management plan specifies a criterion for recruitment failure as three consecutive years of juvenile indices lower than 75% of all other values in the dataset during 1957-2009 (lowest quartile; ASMFC 2003; 2010). We adopted the lowest quartile of the current high productivity period, 1993-2020, as our poor year-class criterion and the upper quartile as an indicator of a strong year-class. Strong year-classes are particularly important to the fishery and we determined how often upper quartile year-classes were present simultaneously in two, three, for four spawning areas during a year for the full time-series and the current high productivity period.

Statistical considerations - Correlations were classified as strong, based on $r \geq + 0.80$ (Ricker 1975). Weak correlations were indicated by $r < + 0.50$; and moderate correlations fell in between. We considered strong and moderate correlations or relationships to be of interest. Levels of significance were reported, but potential for management and biological significance took precedence over $P \leq 0.05$ (Anderson et al. 2000; Smith 2020).

Results

Proportion of ichthyoplankton tows with Striped Bass eggs (Ep) 2021 update – Sample size was sufficient for estimating Ep in the Choptank River (N = 90) during 2021; too few samples were available from the Nanticoke River (N = 20) for an estimate. The estimate of Ep in Choptank River during 2021 was 0.67 (SD = 0.05; Figure 2.1.2) and this estimate served as the baywide Ep estimate (Figure 2.1.3) as well. As baywide Ep, the 2021 estimate was within the bounds exhibited since Ep recovered in 1989 (Ep bounds = 0.57-0.90), although its 90% CI did not overlap some of the higher estimates (Figure 2.1.3). It was clearly separated from the 90% CI's of lower baywide Ep estimates during 1982-1988; estimates during this period were reflected by JI's lower than expected given their estimates of relative survival (Uphoff et al. 2020).

Juvenile index 2021 update – The Baywide JI was 1.65 in 2021 (Figure 2.1.4; Durell and Weedon 2021).

Relative larval survival 2021 update – We adopted the lowest quartile of RLS (<2.07) during 1957-2009 as a criterion for poor egg-larval survival and the upper quartile (>6.73) as an indicator of high egg-larval survival. Estimated RLS was 2.46 in 2021. The simulated mean was 2.50 and the SD was 0.36. The probability of falling below the poor RLS criterion in 2021 was 0.12 and the probability that survival was above the high RLS criterion was 0.

Recovery to a higher frequency of strong RLS began in 1993 (Figure 2.1.5). During 1993-2001, the first half of the high productivity period, there were 18 estimates (2020 did not have an estimate). Five years were above the strong RLS criterion and none fell below the poor RLS criterion. After 2001, second half of the high productivity period, three single years of poor RLS returned, but six years of strong RLS also occurred. This distribution of year-classes was similar to the period of high productivity during 1961-1970 (2 poor years of RLS and 3 years of strong RLS; 10 years total) and was dissimilar to the extended period of low productivity during

1971-1992 when there were 9 years of poor RLS and 2 years of high RLS (22 years total; Figure 2.1.5).

With the exception of 1982-1988, deviations between standardized RLS and standardized JIs during 1957-2021 fell between -0.21 and 0.23 (hereafter, the normal range; Figure 2.1.6). During 1982-1988, larger negative deviations occurred, -0.38 to -1.12; these large negative deviations were interpreted as an indication of the effect of low *Ep*. The deviation for 2021, -0.12, was within the normal range (Figure 2.1.6).

Water quality 2021 update - During 2021, median pH during the April 1-May 8 standard time period in Choptank River was 7.07 and measurements ranged between 6.83 and 8.10 (Table 2.2.1; Figure 2.1.7). This continued the pattern of above neutral, stable pH measurements since 2014 in Choptank River. Medians during 2014-2019 ranged from 7.19-7.42, minimums ranged between 6.56 and 7.05, and maximums were between 7.50 and 8.07. Measurements of pH during 1986-1991 were generally acidic and exhibited higher annual and interannual variation. Median pH during 1986-1991 ranged from 6.18 to 7.15, minimums ranged from 5.75-6.50, and maximum pH measurements were between 6.46 and 9.15 (Table 2.1.1; Figure 2.1.7).

Standard period Choptank River total alkalinity measurements during 2021 (photometer) were much higher than measurements during 1986-1991 (titration; Table 2.1.1; Figure 2.1.7). During 2021, median total alkalinity was 70 mg/L and ranged between 30 and 110 mg/L. During 1986-1991, median total alkalinity varied from 19 to 23 mg/L, and minimums ranged from 7 to 20 mg/L. Maximum total alkalinity was lower during 1986-1989 (22-32 mg/L) and rose during 1990-1991 (37 and 45 mg/L, respectively); the 5th and 95th percentile of annual measurements during 1986-1991 confirmed the trend of stable low measurements (5th percentile) throughout the period and an increase in higher measurement (95th percentile) in the latter two years (Table 2.1.1; Figure 2.1.7).

We could not discern potential patterns in the conductivity summary statistics from Choptank River during the standard period that would suggest differences between 1986-1991 and 2014-2021 (Table 2.1.1). Standard period median, minimum, and maximum conductivity measurements during 2021 were 186, 115, and 3695 $\mu S/cm^2$. The range for median, minimum, and maximum measurements during 1986-2019 were 161-560 $\mu S/cm^2$, 93-135 $\mu S/cm^2$, and 3,660-4,881 $\mu S/cm^2$, respectively (Table 2.1.1).

Salinity – Based on egg counts, 99.5% of eggs in Choptank River (113,313 eggs during 1954-1991) and 94.1% of eggs in Nanticoke River (79,023 eggs during 1954-1985) were collected at salinity less than 2‰. The 2‰ cut-off used for *Ep* sampling and analysis was very likely to capture most egg deposition on the spawning grounds.

Water temperature – Based on cumulative counts of eggs in Choptank River during 1954-1991 (N = 113,503 eggs), the cumulative catch distribution increased rapidly between 12ºC (cumulative P = 0.057) and 16ºC (cumulative P = 0.932; Figure 2.1.8). A total of 2,322 samples were available for Choptank River and 690 contained eggs. In the Nanticoke River during 1954-1985, the cumulative catch distribution (N = 105,336 eggs) increased rapidly between 11ºC (cumulative P = 0.005) and 16ºC (cumulative P = 0.837; Figure 2.1.8). A total of 1,436 samples were available for Nanticoke River and 821 samples collected eggs.

Cumulative distributions of larvae in the Choptank River and Nanticoke River were shifted to the right of egg distributions and did not ascend as steeply. Cumulative counts of larvae in Choptank River during 1980-1989 (N = 42,562 larvae; Uphoff 1989; 1992) exhibited their greatest increase between 15ºC (cumulative P = 0.092) and 17ºC (cumulative P = 0.805), then exhibited a lesser, but steady increase through 20ºC (cumulative P = 0.968; Figure 2.1.9). A total of 2,054 samples were taken in Choptank River and 612 contained larvae. Based on cumulative sum of density estimates of larvae in Nanticoke River during 1992-1993 (Houde et al. 1996; Secor et al. 2017), larvae exhibited their greatest increase between 14ºC (cumulative P = 0.089) and 16ºC (cumulative P = 0.580), then exhibited a lesser, but steady increase through 20ºC (cumulative P = 0.908; Figure 2.1.9). A total of 192 samples were taken in Nanticoke River and 151 contained larvae.

Visual inspection of the plot of days from April 1 that the first egg was collected in the Choptank and Nanticoke rivers (combined) indicated that date was later during 1954-1986 (span of years that first egg capture date could be estimated) than 2000-2021 (Figure 2.1.10). On average, the number of days from April 1 that the first egg was collected in available surveys was later during 1954-1986 (mean = 7.64, SE = 1.04, N = 33) than 2000-2021 (mean = 4.64, SE = 1.25, N = 14); a one-tailed t-test indicated this 3-day decrease was significant (P = 0.05). The median number of days from April 1 was 7 during 1954-1986 and 6 days during 2000-2021 surveys. Minimums of days since April 1 were similar for the time periods; day = -4 for surveys within 1954-1986 and day = -3 during 2000-2021. There was a substantial decrease in maximums of days since April 1: day = 23 during 1954-1986 versus day = 11 for 2000-2021. There were 6 surveys with days since April 1 that the first egg was collected during 1954-1986 that were greater than day 11; day 11 was the maximum for 2000-2021. There was a 23-day range in days since April 1 during 1954-1999 and a 15-day range during 2000-2021.

The plot of days from April 1 that 12ºC was reached in the Choptank and Nanticoke rivers (combined) indicated earlier and later dates were more likely during 1954-1992 (span of years that first date at 12ºC could be estimated) than 2001-2019 (Figure 2.1.11). During 1954-1992, There were 6 surveys with days since April 1 at or before day = 2 and 6 surveys at or after day 16. There were 3 surveys that met the former and 2 that met the latter during 2001-2021 (Figure 2.1.11). A t-test (P = 0.32) did not indicate that the average date that 12ºC was reached in available surveys was different during 1954-1992 (mean = 10.2, SE = 1.23; N =28) and 2001-2021 (mean = 8.25, SE = 1.60; N =16). The median number of days from April 1 was 11 during 1954-1992 surveys and 8 during 2000-2021 surveys. During 1954-1992, minimums of days since April 1 were day = -3 for surveys within 1954-1986 and day = -7 during 2000-2021. Maximums of days since April 1, day = 20 were the same for the two periods. There was a 23-day range in days since April 1 during 1954-1999 and a 27-day span during 2000-2021.

The plot of days from April 1 indicated that 16ºC was reached in the Choptank and Nanticoke rivers (combined) later during 1954-1999 (span of years that surveys could address this criterion) than 2000-2021 (Figure 2.1.12). A t-test (P < 0.0007) indicated that the average number of days since April 1 that 16ºC was reached in available surveys was greater during 1954-1999 (mean = 22.9, SE = 1.15; N =46) than 2000-2021 (mean = 16.6, SE = 1.41; N = 26). Median number of days from April 1 was 22 during 1954-1999 surveys and 16.5 during 2000-2021 surveys. During 1954-1999, minimums of days since April 1 when 16ºC was reached were day = 8 for surveys within 1954-1986 and day = 4 during 2000-2021. Maximums of days since April 1 were day = 42 for 1954-1999 and 31 for 2000-2021. There was a 34-day range in days since April 1 that 16ºC was reached during 1954-1999 and a 27-day span during 2000-2021 (Figure 2.1.12).

The plot of days from April 1 that 20ºC was reached in the Choptank and Nanticoke rivers (combined) indicated it was later during 1954-1998 (span of years that surveys could address this criterion) than 2000-2021 (Figure 2.1.13). Only Choptank River estimates of days since April 1 were available for 2000-2021; Nanticoke River sampling ended early as personnel left for other monitoring projects. A t-test ($P < 0.0032$) indicated that the average number of days since April 1 that 20ºC was reached in available surveys was greater during 1954-1998 (mean = 42.2, SE = 1.64; N =42) than 2000-2021 (mean = 32.8, SE = 2.48; N = 16). Median number of days from April 1 was 42 during 1954-1998 surveys and 32 during 2000-2021 surveys. During 1954-1998, minimums of days since April 1 when 20ºC was reached were day = 22 for surveys within 1954-1986 and day = 16 during 2000-2021. Maximums of days since April 1 were day = 64 for 1954-1998 and 47 for 2000-2021. There was a 44-day range in days since April 1 that 20ºC was reached during 1954-1998 and a 31-day span during 2000-2021.

In the Nanticoke River during 1954-1992, there were 12 surveys where the days between 12ºC and 16ºC were shorter than between 16ºC and 20ºC and 7 years that were the opposite (Figure 2.1.14). Span of days between 12ºC and 20ºC in Nanticoke River averaged 34.3 days, the median was 30 days, 19 days was the minimum, and 59 days was the maximum (Figure 2.1.14).

In the Choptank River during 1954-2021, there were 10 years where days between 12ºC and 16ºC were shorter than between 16ºC and 20ºC, 2 years with no difference, and 4 years where days between 12ºC and 16ºC were longer than between 16ºC and 20ºC (Figure 2.1.15). There was a noticeable decrease in the span of days between 12ºC and 20ºC between 1954-1987 (mean = 28.5 days, median = 31.5, minimum = 14, and maximum = 40) and 2001-2021 (mean = 22.4 days, median = 20, minimum = 7, and maximum = 37). Of particular note were 3 years since 2000 (2013, 2017, and 2019) where the span of days between 12ºC and 16ºC were only 2 days (Figure 2.1.15); this short period warming of the earlier portion of spawning season was not evident in either the Nanticoke or Choptank data available prior to 2000. In the Nanticoke River prior to 2000, 7 of 19 years had single digit spans between 12ºC and 16ºC and the minimum was 3 days. In Choptank River during 1955-1987, there were 2 of 6 days with single digit spans with a minimum of 5 days. In the Choptank River since 2000, there have been 5 of 8 days with single digit spans (Figure 2.1.15). Overall, the span of days between 12ºC and 16ºC in single digits has gone from less than half the span of days prior to 2000 to more than half after 2000.

When water temperature data from Choptank and Nanticoke rivers are compiled into a single record, the average span between the 12ºC and 20ºC decreased from 33.7 days during 1954-1992 to 24.4 days during 2000-2021. Median spans declined from 32 days to 20 days, respectively. In addition to these general changes, 3 years during 2000-2021 (of 9 available) had the shortest documented spans between 12ºC and 16ºC (2 days) and 2021 had the earliest date that 12ºC was reached in the entire time-series. Average number of days between 12ºC and 16ºC declined from 14.3 during 1954-1992 (median = 14) to 8.1 days during 2000-2021 (median = 5).

Flow – Annual monthly average flows (cubic feet per second or CFS) for March-May and area-specific JIs during 1957-2020 were summarized for the Head-of-Bay (Table 2.1.2), Potomac River (Table 2.1.3), Choptank River (Table 2.1.4), and Nanticoke River (Table 2.1.5). Generally, correlations of March, April, or May flow with area specific JIs were significant at $P < 0.05$, but not strong. (Table 2.1.6). Strongest correlations were observed for April in Potomac ($r = 0.39$), Choptank ($r = 0.25$), and Nanticoke ($r = 0.32$) rivers, followed by March ($r = 0.35$, 0.25, and 0.24, respectively; $P < 0.06$). Correlations of May flows with JIs for these three spawning areas were weak ($r = 0.05$ to 0.12, $P > 0.36$). May flows had the strongest correlation with Head-of-Bay JIs ($r = 0.43$, $P = 0.003$), followed by April ($r = 0.21$, $P = 0.09$), and March ($r = -0.11$, $P = 0.38$; Table 2.1.6). The mean of March and April flows were chosen for correlations with Potomac River, Choptank River, and Nanticoke River JIs in subsequent analyses (Table 2.1.7).

The mean of April and May flows were chosen for Head-of-Bay (Table 2.1.7). These two-month combinations are referred to as spawning period flows.

During 1957-2020, spawning period flows and JIs were better correlated in Head-of-Bay and Potomac River ($r = 0.42$, $P = 0.0005$ and $r = 0.44$, $P = 0.0003$, respectively) than Choptank and Nanticoke rivers ($r = 0.30$, $P = 0.016$ and $r = 0.29$, $P = 0.018$, respectively). Correlations of spawning period flow and JIs during the recent high productivity period (1993-2020) were moderate in the Head-of-Bay and Potomac River ($r = 0.54$, $P = 0.0033$ for both), poorer in Choptank River ($r = 0.44$, $P = 0.021$), and weak in Nanticoke River ($r = 0.19$, $P = 0.33$).

Strong correlations ($r \geq 0.84$) of annual spawning season flows during 1957-2020 were found between Potomac River and Head-of-Bay or Choptank River and Nanticoke River (Table 2.1.8). Correlations between Potomac River and Choptank River or Nanticoke River were moderate ($r = 0.72$ and 0.76, respectively). Correlations were moderate for Head-of-Bay flows and Choptank River or Nanticoke River ($r = 0.56$ and 0.60, respectively). All correlations of flow among rivers were significant at $P < 0.0001$ (Table 2.1.8).

Correlations among spawning area JIs fit the general patterns exhibited by flow, but were not as strong. In the long-term (1957-2020) analysis, moderate correlations of JIs were found between the Choptank and Nanticoke rivers ($r = 0.66$, $P < 0.0001$) and weak for remaining combinations ($r = 0.32$-0.48, $P \leq 0.008$; Table 2.1.9). Results were similar for the recent period of high productivity (1993-2020) with the exception of a moderate correlation arising between Head-of-Bay and Potomac River JIs ($r = 0.54$, $P = 0.0023$; Table 2.1.9).

During 1993-2020, standardized spawning season flow into the Head-of-Bay spawning area ranged between 0.48- and 2.51-times average flow. Head-of-Bay JIs in the upper quartile (strong year-class; $JI \geq 8.8$) did not occur unless April-May flow was above average for the long-term time-series (Figure 2.1.16). Strong year-classes appeared from 1.08-times the long-term average through the highest flow (2.5-times average). Poor Head-of-Bay year-classes (bottom quartile of JIs; $JI \leq 2.3$) occurred when spawning season flows were 0.48- to 1.51-times average flow. Juvenile indices between these criteria occurred at flows 0.64- to 2.48-times the average; all but one of these year-classes were found between 0.64- and 1.49-times the long-term average.

During 1993-2020, standardized spawning season flow into the Potomac River spawning area ranged between 0.35- and 2.73-times average flow. Two Potomac River JIs in the upper quartile ($JI \geq 5.8$) occurred at below average flows (0.68- and 0.95-times the long-term average), but the strongest JIs (JIs > 2-times the upper quartile JI) were found at 1.19- to 2.73-times the long-term average (Figure 2.1.17). Poor year-classes ($JI \leq 2.2$) occurred at flows 0.57- to 2.16-times the average; all but one was between 0.57- and 1.36-times the average. Potomac River JIs between these criteria occurred at flows 0.35- to 1.72-times the average (Figure 2.1.17).

During 1993-2020, standardized spawning season flow in the Choptank River ranged between 0.30- and 2.40-times average flow. Strong year-classes in the Choptank River ($JI \geq 20.6$) were present when flows were 1.01- to 1.72-times the long-term average (Figure 2.1.18). There was one JI that fell very slightly below the upper quartile boundary that occurred at 0.83-times the average. Poor year-classes ($JI \leq 2.4$) were present at flows 0.30- to 1.63-times the long-term average. Year-classes between the poor and strong year-class criteria were found at 0.39- to 2.40-times the average flow (Figure 2.1.18).

During 1993-2020, standardized spawning season flow in Nanticoke River ranged between 0.46- and 2.34-times average flow. Strong year-classes in the Nanticoke River ($JI \geq 5.8$) were present when flows were 0.83- to 1.69-times the long-term average (Figure 2.1.19). Strongest JIs (JIs > 2-times the upper quartile JI) were found at 1.15- to 1.69-times the long-term average. Poor year-classes (JI ≤ 1.8) were present at flows 0.46- to 2.34-times the long-term average. Year-classes between the poor and strong year-class criteria were found at 0.49- to 1.75-times the average flow (Figure 2.1.19).

Discussion

2021 updates - Egg production in 2021, based on baywide *Ep* (0.67), was not in the top tier of estimates during 1993-2021 (roughly 0.80 or greater), but there was a high chance it was above levels during 1982-1988 when it was depleted enough to affect year-class success. Four top quartile baywide JIs were present during 1993-2021 when *Ep* was within the top tier (1993, 1996, 2003, and 2011), while 5 were present when *Ep* was at a similar level to 2021 (2000, 2001, 2005, 2015, and 2018). Estimated RLS in 2021 was just above the poor survival criterion; most of the poor RLS estimates were concentrated in 1980-1991. Estimates of RLS near or below the poor survival criterion were absent during 1993-2001, but returned afterward and have occurred intermittently. Three years of relatively low baywide JIs have occurred since 2019 and presumably 3 years of low RLS have occurred as well; *Ep* (denominator for RLS) was not estimated in 2020 due to Covid restrictions on sampling, but *Ep* is assumed to be in the same mid-range as 2019 and 2021 (0.70 and 0.67, respectively). Three consecutive years of relatively low JIs and RLS is worrisome and will impact the fishery in the future. However, it is not unprecedented; three years of relatively low RLS preceded the strong 1970 year-class. It remains to be seen whether a continued period of low productivity is in the offing.

Measurements of pH in Choptank River between 1986-1991 and 2013-2021 indicated improvement (higher, more stable averages and less variability of individual measurements) that would have lowered toxicity of metals implicated in poor recruitment in some Striped Bass spawning areas during the 1980s (Uphoff 1989; 1992; Hall et al. 1993; Richards and Rago 1999; Uphoff et al. 2020). Average alkalinity was at least 3-times higher in 2021 compared to 1986-1991. Low survival of Striped Bass larvae during the 1980s in the Choptank River estimated from ichthyoplankton surveys and in situ bioassays were associated with low pH, alkalinity, and conductivity that could have influenced toxicity of metals (Uphoff 1989; 1992; Hall et al. 1993; Richards and Rago 1999). Increases in pH, alkalinity, and RLS coincided with actions that reduced acidity and deposition of toxic metals in acid rain, increased implementation of conservation agriculture that reduced use of inorganic fertilizers and pesticides (a potential source of metals) and decreased erosion (sediment is a vector for contaminants); alkalinity of freshwater increased in the U.S. as well (Uphoff et al. 2020). While measurements of metals are unavailable, it seems unlikely that poor survival of larvae during 2019-2021 could be attributed to a return of toxic water quality conditions implicated in poor recruitment during the 1980s.

Water temperature and flow - In this report we added analyses to detect changes in spawning area temperature and flow patterns that may be influencing recent year-class success of Striped Bass in the four major spawning areas. We detected changes in spawning season water temperature and flow that were concurrent with decreased RLS and year-class success since the mid-2000s. Water temperature and flow conditions are important influences on year-class success of Striped Bass (Hollis et al. 1967; Uphoff 1989; 1992; Secor and Houde 1995; Rutherford et al. 1997; North and Houde 2001; 2003; Maryland Sea Grant 2009; Martino and Houde 2010; Shideler and Houde 2014; Secor et al. 2017; Millette et al. 2020). Temperature impacts recruitment through direct mortality of eggs and larvae due to lethally low or high temperatures (density-independent mortality) and indirectly via its influence on the timing of zooplankton blooms for first-feeding larvae (match-mismatch hypothesis; density-dependent mortality), while flow may be associated with zooplankton dynamics, nursery volume, advection from the nursery, and water quality and toxicity of contaminants (Hollis et al. 1967; Uphoff 1989; 1992; Secor and Houde 1995; North and Houde 2001; 2003; Maryland Sea Grant 2009; Martino and Houde 2010; Shideler and Houde 2014; Secor et al. 2017; Millette et al. 2020).

Water temperature and spawning changes were similar to expectations described by MD Sea Grant (2009) and Nack et al. (2019). Mismatches between the occurrence of larvae and environmental conditions favorable for their survival were considered likely under projected warming scenarios (MD Sea Grant 2009). Higher temperatures during spring would likely have negative effects on larval survival due to a more rapid spring to summer transition that reduces when temperatures are most favorable for larval survival (MD Sea Grant 2009).

Our temperature milestones generally captured most Striped Bass egg and larval production based on counts in historic datasets (1950s to 1990s). Cumulative catch distributions of Striped Bass eggs increased rapidly between 12ºC and 16 ºC in the Choptank and Nanticoke rivers, indicating most eggs were collected when these temperatures prevailed. Eggs do not have an escape response (Bulak 1993) and changes would reflect hatching or death. The larval cumulative catch distribution gained most rapidly between 14ºC and 17ºC, followed by a lesser, but steady, increase to 20ºC. Changes in larval distribution would have been related to growth and its effect on increasing mobility of larvae and changes in catchability with size, as well as mortality.

Means or medians of days between 12ºC and 20ºC milestones during 2000-2021 were 10 days to 12 days shorter (respectively) than during 1954-1992 in Choptank and Nanticoke rivers. Changes were not uniform among temperature milestones. Early milestones appeared to be the least affected. While analysis of the average first date that eggs were collected indicated that date had shifted about 3 days earlier between time periods, but earlier attainment of 12ºC in 2000-2021 (about 2-3 days) was not fully supported. As the milestones progressed in magnitude, average dates of occurrence progressed between 1954-1992 and 2000-2021 (7 days earlier at 16ºC and 10 days for 20ºC). The portion of the spawning period when most eggs were collected (days from 12ºC to 16ºC) has shortened and lethally high temperatures (indicated by days to 20ºC) were being reached earlier. In addition to these general changes, 3 years during 2000-2021 (of 9 available) had very short spans between 12ºC and 16ºC (2 days) and 2021 had the earliest date that 12ºC was reached in the entire time-series. During 1954-1992, the transition from 12ºC to 16ºC took a week or less with 5 of 19 Nanticoke River surveys and 2 of 7 Choptank River surveys; after 2000, 4 of 9 Choptank River surveys exhibited a transition of a week or less. The transition from 16ºC to 20ºC took a week or less in 2 of 19 Nanticoke River surveys during 1954-1992 and in 1 of 7 Choptank River surveys; after 2000, 3 of 9 Choptank River surveys made this transition in a week or less.

Survival of striped bass larvae is highest at 18°C (Secor and Houde, 1995; MD Sea Grant 2009). In the past, average springtime temperatures in Chesapeake Bay typically fell near 18°C for approximately 2 to 3 weeks during April and May before consistently remaining above 20°C at the onset of summer (MD Sea Grant 2009). Warming in Chesapeake Bay now occurs at a more rapid rate and duration of suitable temperatures for larval development is shorter by 10 days on average after 2000.

Water temperature analyses presented here and in Uphoff et al. (2020) have not covered the two largest Striped Bass spawning areas, Head-of-Bay and Potomac River. Peer and Miller (2014) analyzed catches from Maryland's spring gill net monitoring of adult Striped Bass on these two spawning grounds during 1985-2010 and found that females moved onto Head-of-Bay and Potomac River spawning grounds approximately 3 d earlier for every 1ºC increase in spring water temperature. Further analysis of spring gill net data (1985-2020) indicated that timing of a 14°C milestone was about 3-5 days earlier and that the date that cumulative catch of females reached 100% was 8-9 days earlier, but date that 25% of catch was reached had not changed (A Guiliano, MD DNR, personal communication).

Below average flow conditions were less conducive to formation of strong year-classes and poor year-classes were more likely. Above average flows resulted in a higher chance that strong year-classes would be formed and a modest reduction in occurrence of poor year-classes. When all spawning areas were combined during 1993-2020 (N = 112 area and year combinations), there were 4 strong year-classes when flows were below average and 24 strong year-classes when flow was at or above average. There were 17 poor year-classes when flow was below average and 13 when it was at or above average. When the 1993-2020 high productivity period was split in half (14 years each), below average flows were less common during the first half (1993-2006) than the second in the Potomac (7 in the first half and 10 in the second), Choptank (4 and 7), and Nanticoke (5 and 8) rivers. There was no change in the Head-of-Bay (5 years of below average flow in each half).

Frequency of below average flow conditions has increased since 2006 in 3 of the 4 spawning areas (no change in Susquehanna River), increasing odds that a lesser year-class will be formed and decreasing the odds that strong baywide year-class will form. General timing of spawning season flows associated with JIs were similar (March-April) for Potomac River, Choptank River, and Nanticoke River, and later (April- May) for Susquehanna River. The watersheds of the three rivers with higher frequency of low flows fall roughly along similar latitudes, while the Susquehanna River drains to the north. Average winter water temperatures were lower in Head-of-Bay than in Choptank River (Millette et al. 2020), indicating these latitude differences could reflect local climate. Flow and year-class patterns detected here also suggested differences between the large fluvial rivers draining three geographic provinces and smaller spawning rivers located on the Coastal Plain. The Susquehanna and Potomac rivers flow through the Coastal Plain, Piedmont, and Appalachian geographic provinces while Choptank and Nanticoke rivers are adjacent Coastal Plain rivers on the eastern shore of Chesapeake Bay. Strongest correlations among spawning period flows were indicated for rivers draining similar provinces.

Years of high spring discharge favor anadromous fish recruitment in Chesapeake Bay and may represent episodes of hydrologic transport of accumulated terrestrial carbon (organic matter or OM) from watersheds that fuel zooplankton production and feeding success (McClain et al. 2003; Hoffman et al. 2007; Martino and Houde 2010; Shideler and Houde 2014). Under natural conditions, riparian marshes and forests would provide OM subsidies in high discharge years, while phytoplankton would be the primary source of OM in years of lesser flow (Hoffman et al. 2007). Differences in watershed characteristics of land draining into the Striped Bass spawning areas may influence their sources of OM. Choptank and Nanticoke rivers are largely agricultural watersheds (40-49% of watershed non-water area) with modest forest cover (18-25%) and extensive non-tidal and tidal wetlands (18-19%); wetlands would be an important source of OM (Table 2.1.10). Potomac and Susquehanna rivers have proportionally less agriculture (21-23%), more forest cover (57-60%) and less wetlands (1-2%; Table 2.1.10); OM would more likely be derived from upland forest sources.

Correlations of spawning period flows and JIs were positive and weak to moderate for all four spawning areas during 1993-2020. Linear modeling techniques alone would not necessarily have provided a full understanding of flow and year-class success dynamics among spawning areas. Juvenile indices increased throughout the ranges of flows exhibited in Susquehanna and Potomac rivers, while highest flows were not associated with strong year-classes in the Choptank and Nanticoke rivers. Much of the work that linked high flows, zooplankton, and Striped Bass year-class success were conducted in Potomac River or Head-of-Bay (Rutherford et al. 1997; North and Houde 2001; 2003; Martino and Houde 2010; Shideler and Houde 2014), and those relationships may not apply as well to Choptank and Nanticoke rivers.

Alignment of strong year-classes among all four spawning areas was rare and alignment among three areas was most common. During 1957-2021, upper quartile year-classes aligned between two spawning areas six times, among three areas ten times, and among four areas twice. Strong year-classes appeared singly on two occasions. During the current period of high productivity (1993-2021, a period of 29 years), strong year-classes aligned between two spawning areas four times, among three areas eight times, and among four areas once; a single spawning area produced an upper quartile year-class on one occasion. During 1957-1970 (14 years), there was one instance of alignment of two areas, two instances of three areas, and one instance of four areas aligning; there was not an instance of a single area producing an upper quartile year-class. There was only one year during 1971-1992 that there was alignment of strong year-classes among spawning areas (2 areas in 1989) and one year (1992) had a single strong year-class.

Head-of-Bay, Potomac River and Choptank River were the most common three spawning area combination of upper quartile year-classes (4 of the 10 years with 3 area combinations) during 1957-2021; followed by Head-of-Bay, Potomac River, and Choptank River (3 years), Head-of-Bay, Choptank and Nanticoke River (2 years); and a single instance for Head-of-Bay, Potomac River, and Nanticoke River. Two-area combinations (one year for each) were present for Head-of-Bay and Potomac River, Potomac River and Choptank River, and Choptank River and Nanticoke River. Choptank River contributed most frequently to the combinations (11), followed by Potomac River (9), Nanticoke River (8), and Head-of-Bay (7). Head-of-Bay and Potomac River each had a year when they were the only spawning area to have a year-class in its upper quartile.

Caveats – Surface tows were used in most surveys, and we considered them representative of egg distribution throughout the water column. Kernehan et al. (1981) found that abundance of eggs in Head-of-Bay increased from surface to bottom but Dovel (1971) did not. Using ANOVA, Uphoff (1997) did not find significant differences in $\log_e$-transformed midwater, bottom, and inshore egg catches in the Choptank River or $\log_e$-transformed surface, mid-depth, and bottom egg densities in Head-of-Bay spawning areas (data set of Kernehan et al. 1981). Other studies of vertical distribution of striped bass eggs have yielded a variety of results (Kernehan et al. 1981).

Water temperature milestones were conceptually straightforward, but a bit ambiguous in practice at times. Sites in the upper reaches of the spawning areas appear to warm quicker than downstream, but early spawning was typically downstream (J. Uphoff, MD DNR, personal observation). Use of upper sites where early spawning was not likely could have negatively biased dates when 12ºC was relevant to spawning dynamics. There were also instances that impacted all three temperature milestones individually when they were reached at multiple stations considered relevant, followed by a sustained decrease and an interval before they were reached again. The initial occurrence at multiple stations was used for the temperature. Sampling interval could have an impact as well. None of the surveys were conducted daily and most were conducted several days a week with a maximum interval of a week for inclusion in analysis. Spawning season temperatures can be volatile and longer intervals are more likely to miss important events than shorter ones.

This analysis was constrained to the Choptank and Nanticoke River spawning areas (both watersheds located in the Coastal Plain) because of their long time-series and more current sampling. These areas were sampled more frequently because their size made them tractable for small boats used by DNR surveys that made up the vast majority of available data. None of these surveys were specifically designed to monitor for long-term temperature changes and they represent "targets of opportunity" for addressing the effect of climate warming on Striped Bass spawning. Head-of-Bay and Potomac River have not had ichthyoplankton surveys that qualified for *Ep* analysis since 1996 (Uphoff 1997; Uphoff et al. 2020). The absence of information on the 20ºC milestone from Nanticoke River beyond 1993 was not anticipated and the dynamics of all three milestones since 2000 were based the Choptank River alone. Nanticoke and Choptank rivers were combined to understand pre-2000 dynamics under an assumption that spawning season temperatures were not likely to be different.

There were considerable differences in total alkalinity measurements in Choptank River during 1986-1991 and 2021. Alkalinity during 1986-1991 was measured by titration and with a photometer during 2021. Measurements during 2021 were well above the minimum tolerance of the photometer and were within the working range and it seems reasonable to conclude the differences were real and unrelated to different methods.

Use of juvenile index quartiles to designate poor and strong year-classes was convenient and use of the lower quantile as a poor year-class marker based on the use of the Baywide MD JI by ASMFC (ASMFC 2003; 2010). Time periods used for quartiles should reflect similar underlying dynamics (spawning stock and environmental forcing), although that may be difficult to determine with confidence, particularly during periods of transition. For Striped Bass in Maryland's portion of the Chesapeake Bay, RLS and *Ep* can be used to identify periods of productivity (Uphoff et al. 2020). However, quartiles may not align with the needs of the fishery. The fishery has been generally described as driven by strong year-classes (Florence 1980; Rago and Goodyear 1987; Rago 1992; Richards and Rago 1999; Secor 2000; Uphoff et al. 2020), but some of the lesser year-classes within the upper quartile may not entirely meet the expectations of the fishery.

Magnitude of an upper quartile index may not translate directly into fish available to the fishery due to changing natural mortality. Martino and Houde (2012) detected density-dependent mortality of age 0 Striped Bass in Chesapeake Bay, supporting a hypothesis that density dependence in the juvenile stage can contribute significantly to regulation of year-class strength. Tagging models indicated that annual instantaneous natural mortality rates (M) of legal sized Striped Bass in Chesapeake Bay increased substantially during the mid-1990s while annual instantaneous fishing mortality rates (F) remained low (Kahn and Crecco 2006; Jiang et al. 2007; ASMFC 2013; NEFSC 2019). The rise in M in the mid-to-late 1990s was consistent with a compensatory response to high Striped Bass abundance, low forage, and poor condition (Uphoff et al. 2022).

Upper quartiles of area specific JIs of the four spawning areas were not always similar in magnitude. During 1993-2021, boundaries of quantiles for the Potomac and Nanticoke rivers were the same, 5.8-16.0; the Head-of-Bay was somewhat higher, 8.8-18.5; and the upper quartile of the Choptank River JI was much higher, generally ranging between 20.6-33.0 with one JI at 86.7. Whether the difference in the upper quartile is a real difference in abundance or a matter of relative scale is question that requires egg production and egg-larval survival estimates from Choptank River and one other system during a year that strong year-classes were produced in both (this is not easy to plan for). Boundaries for poor year-classes were fairly similar (2.2-2.8). To some degree, the Baywide JI is statistically weighted by the number of sites and dates in each surveyed nursery (24 each in Choptank and Nanticoke rivers, and 42 each in Potomac River and Head-of-Bay since 1966; Durell and Weedon 2021). Weighting by spawning area size would severely discount the Nanticoke and Choptank rivers, and would make strong density-dependent assumptions about egg abundance and survival of eggs and larvae. During the 1970s, there was a weighting scheme based on spawning area and season landings that was calculated, but weighted JIs were not very different from the basic calculation and it was not used in management (J. Uphoff, MD DNR, personal observation).

Management implications - The primary management strategy for Atlantic coast Striped Bass is to keep SSB at a very high level. Managing for low exploitation rates and high SSB would be expected to provide extended age structure that allows for diverse spawning behaviors (older Striped Bass tend to spawn earlier than young ones; Hollis 1967; Peer and Miller 2014) that are expected to stabilize recruitment in the face of warming winter and spring temperatures (Secor 2007; MD Sea Grant 2009). Although not explicitly stated, this strategy appears associated with an expectation of protracted spawning seasons. However, the time span between temperature milestones contracted in the Choptank River in the last two decades, concentrating egg production in a shorter period. When spawning is concentrated in a shorter time period, egg mortality events kill a larger proportion of a year's spawn (Richards and Rago 1999). It is unclear whether increased egg production within this compressed spawning window can offset temperature related egg mortality numerically, but potential for more eggs to result in more larvae cannot be ruled out. However, if mistiming of zooplankton blooms with first-feeding larvae is important, its density-dependent nature may limit successful management. It becomes possible in years of higher survival of eggs that larval survival and subsequent recruitment will be capped at a low level due to inadequate larval foraging no matter how many eggs are pumped into the spawning area if zooplankton production is misaligned with first-feeding larvae. Addressing hypotheses about roles of density-independent and density-dependent egg and larval mortality due water temperature, flow, and zooplankton interactions will require intense ichthyoplankton surveys capable of sampling eggs through postlarvae.

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Table 2.1.1. Summary of pH, conductivity (µS/cm^2), and total alkalinity (mg/L) during a standard period (April 1 – May 8), 1986-1991 and 2014-2021. Surveys had similar geographic scales.

pH							
Year	Mean	Median	95th%	5th%	Minimum	Maximum	N
1986	7.04	7.15	7.76	6.71	5.75	9.15	628
1987	6.76	6.78	7.07	6.54	6.30	7.45	249
1988	6.93	7.02	8.01	6.53	6.45	8.40	122
1989	6.17	6.18	6.39	6.00	5.78	6.46	139
1990	6.97	7.03	7.19	6.78	6.50	7.34	150
1991	6.74	7.02	7.51	6.13	5.86	8.20	222
2014	7.09	7.19	7.80	6.80	6.70	8.00	96
2015	7.39	7.42	7.83	7.11	7.05	8.07	96
2016	7.22	7.27	7.68	6.92	6.68	7.85	88
2017	7.23	7.27	7.55	7.01	6.87	7.76	100
2018	7.12	7.15	7.68	6.83	6.71	7.86	90
2019	7.18	7.25	7.55	6.92	6.56	8.10	100
2021	7.05	7.07	7.38	6.86	6.83	7.50	100
Conductivity							
Year	Mean	Median	95th%	5th%	Minimum	Maximum	N
1986	858	560	2480	126	94	3950	628
1987	893	372	3175	144	132	4410	250
1988	910	363	3686	186	177	4390	122
1989	426	194	1824	132	93	3750	148
1990	650	161	3053	136	129	3660	144
1991	603	217	3092	147	126	4090	212
2014	669	177	3101	118	111	4881	96
2015	673	208	2956	137	126	3934	96
2016	963	416	3538	150	93	4389	88
2017	991	535	3054	149	135	3664	100
2018	619	207	2652	135	122	3770	90
2019	464	166	2185	128	124	3496	100
2021	636	186	2703	133	115	3695	100

Table 2.1.1 (continued).

	Total alkalinity (mg/L)						
	Mean	Median	95th%	5th%	Minimum	Maximum	Count
1986	22	23	26	15	13	26	155
1987	24	24	31	19	17	32	99
1988	21	22	23	20	20	23	21
1989	20	22	22	13	13	22	42
1990	20	19	30	12	11	37	146
1991	20	20	28	10	7	45	173
2021	67	70	116	43	30	110	80

Table 2.1.2. Head-of Bay (Susquehanna River) March-May flow (CFS) and the Head-of-Bay JI (Durell and Weedon 2021), 1957-2020.

Year	March	April	May	JI
1957	56,350	105,400	30,970	1.92
1958	23,260	78,050	137,900	22.07
1959	44,020	57,570	77,810	0.95
1960	51,280	28,120	126,200	3.28
1961	61,910	97,260	110,300	7.46
1962	26,290	84,540	112,300	3.68
1963	15,800	104,900	52,340	3.01
1964	29,150	134,900	72,470	15.41
1965	44,750	42,600	58,500	0.76
1966	55,780	78,740	35,210	15.89
1967	30,390	83,780	64,640	3.92
1968	41,010	57,620	34,870	6.13
1969	29,140	29,190	61,100	12.21
1970	68,710	52,700	139,500	13.71
1971	74,340	105,600	63,960	10.45
1972	30,470	114,500	92,710	4.95
1973	60,980	65,630	80,560	11.71
1974	47,590	62,110	93,470	6.75
1975	83,140	81,440	48,710	2.34
1976	98,840	64,490	42,980	2.70
1977	27,160	126,700	85,790	4.99
1978	36,430	108,900	90,250	6.51
1979	43,190	138,900	63,180	4.56
1980	11,940	68,580	103,100	1.43
1981	102,400	34,610	36,150	0.17
1982	49,730	84,640	75,500	2.98
1983	42,990	51,720	123,300	0.61
1984	109,300	52,340	124,300	2.24
1985	35,050	53,720	50,040	0.19
1986	62,990	98,720	57,790	0.90
1987	20,600	55,290	89,050	0.16
1988	51,540	46,810	34,360	2.25
1989	24,670	39,650	61,470	8.54
1990	85,400	36,950	52,180	2.20

Table 2.1.2 (continued).

Year	March	April	May	JI
1991	50,410	71,450	51,640	1.99
1992	21,500	61,920	65,820	0.87
1993	16,580	76,870	235,100	15.00
1994	53,390	142,700	147,300	12.88
1995	23,190	46,510	33,700	2.85
1996	66,560	74,270	74,660	15.00
1997	48,210	74,590	41,290	6.15
1998	77,180	99,620	87,840	4.32
1999	42,910	56,750	59,620	1.91
2000	42,880	84,900	91,050	8.84
2001	34,110	51,410	87,830	7.15
2002	39,680	41,920	45,180	1.35
2003	27,710	110,200	73,810	11.89
2004	25,630	84,800	75,840	4.17
2005	48,200	63,450	109,000	8.48
2006	58,110	28,120	31,860	0.95
2007	16,210	99,720	72,490	8.21
2008	85,390	125,900	58,970	2.33
2009	39,020	51,040	45,650	2.85
2010	31,340	82,920	45,250	2.90
2011	35,270	162,700	148,000	5.79
2012	38,800	45,390	23,370	0.44
2013	54,830	44,610	52,570	3.29
2014	25,530	48,810	88,630	8.02
2015	11,670	48,080	92,500	7.20
2016	74,210	43,250	33,210	1.14
2017	51,350	55,270	94,800	18.52
2018	87,430	61,180	74,350	14.48
2019	76,590	67,680	75,540	2.33
2020	54,810	62,850	61,300	1.95

Table 2.1.3. Potomac River March-May flow (CFS) and the Potomac River JI (Durell and Weedon 2021), 1957-2020.

Year	March	April	May	JI
1957	15,790	20,750	8,046	1.78
1958	28,960	29,460	25,980	3.93
1959	8,949	12,890	10,830	0.61
1960	16,230	32,940	21,820	2.44
1961	30,210	34,270	16,470	12.82
1962	45,900	23,750	9,891	6.70
1963	45,060	8,432	4,982	0.54
1964	30,660	18,270	15,630	9.40
1965	28,430	16,820	7,914	1.10
1966	12,460	11,790	13,990	5.08
1967	35,920	9,116	17,270	1.02
1968	24,130	8,289	12,680	0.39
1969	8,385	7,058	3,921	0.12
1970	16,100	34,590	9,215	10.98
1971	21,410	10,760	15,340	3.48
1972	27,460	24,800	25,010	0.96
1973	17,300	35,140	18,940	1.10
1974	11,200	20,990	10,050	0.69
1975	30,130	15,170	19,750	3.56
1976	11,070	12,340	4,887	1.46
1977	23,810	23,000	4,720	0.78
1978	42,840	16,140	30,760	3.33
1979	37,880	18,380	16,880	1.15
1980	23,330	30,960	25,300	1.04
1981	7,545	13,100	10,000	0.68
1982	29,050	13,370	7,995	3.50
1983	25,270	48,260	24,560	0.62
1984	34,430	47,860	19,930	1.42
1985	13,030	11,480	9,686	1.45
1986	25,240	11,880	6,841	3.09
1987	16,180	45,580	15,300	3.01
1988	8,315	11,680	38,930	0.22
1989	19,810	10,570	40,410	1.15
1990	7,403	12,260	13,000	0.38
1991	25,840	16,090	6,145	0.84
1992	19,010	18,350	12,890	6.00

Table 2.1.3 (continued).

Year	March	April	May	JI
1993	62,740	57,850	12,530	15.96
1994	67,370	27,780	18,020	2.01
1995	10,700	5,810	11,410	4.48
1996	30,310	22,080	28,270	13.60
1997	31,430	10,900	6,743	3.67
1998	47,840	28,100	26,000	4.42
1999	16,450	13,520	5,143	5.84
2000	17,630	16,100	6,979	3.52
2001	17,200	20,540	7,456	5.01
2002	6,225	12,180	14,780	3.95
2003	46,450	28,760	33,550	12.81
2004	20,480	31,840	15,960	2.36
2005	24,940	24,150	10,060	7.92
2006	5,291	10,250	7,475	2.42
2007	32,510	23,240	6,882	2.20
2008	20,640	20,500	33,500	1.40
2009	4,495	17,360	27,060	3.75
2010	45,970	13,580	9,934	2.17
2011	34,260	43,940	33,450	7.18
2012	18,440	7,171	9,514	0.95
2013	19,530	14,360	19,050	3.13
2014	18,510	18,490	34,770	1.07
2015	23,820	18,150	8,526	6.07
2016	13,350	6,728	19,430	2.36
2017	9,010	15,120	26,020	3.82
2018	12,080	23,220	33,090	2.97
2019	30,850	22,170	26,420	1.27
2020	9,138	16,420	20,820	1.05

Table 2.1.4. Choptank River March-May flow (CFS) and the Choptank River JI (Durell and Weedon 2021), 1957-2020.

Year	March	April	May	JI
1957	268.7	111	42.3	1.16
1958	401.3	308.8	298.2	11.01
1959	163.7	168.8	49.3	0.09
1960	200.4	165.2	72.8	4.31
1961	354.1	277.4	114.5	5.40
1962	337.2	312.6	56.2	3.14
1963	372.2	67.4	41.8	2.01
1964	280.6	257	111.3	4.92
1965	180.8	123.4	43.7	2.18
1966	43.7	47.2	57.8	5.52
1967	248.2	89.5	121.3	2.80
1968	326.8	92.4	113.1	3.85
1969	196.4	131	81	2.55
1970	135.4	367.7	93	25.75
1971	203.1	153.6	86.9	2.51
1972	218.5	243.7	173.6	5.36
1973	189.5	281.3	132.3	0.43
1974	194.2	228.7	138.8	3.55
1975	402.8	226.3	234.8	2.71
1976	148.8	94.4	40	0.89
1977	109.9	86.8	30.3	0.81
1978	557.1	144.7	202.9	2.65
1979	370.9	182.9	134.5	1.12
1980	265.1	266.8	178.9	0.60
1981	82.9	150	163.9	0.84
1982	207.1	193.5	95.9	5.68
1983	420.6	649.4	228.6	0.64
1984	468.5	429.3	200.4	2.13
1985	73.9	65	55.5	1.78
1986	197.9	102	46.7	0.32
1987	255.8	139.1	107.5	3.06
1988	110.8	121	155.3	0.40
1989	390.2	304	457.3	28.10
1990	137.6	223.7	288.9	1.34

Table 2.1.4 (continued).

Year	March	April	May	JI
1991	182.6	192.1	88.7	4.42
1992	197.8	109.8	115	2.07
1993	512.2	312.4	107.8	27.87
1994	826.3	331.3	121	7.71
1995	221.8	84	107.2	9.96
1996	235.9	397.1	279.9	33.29
1997	262.5	238.5	222.5	3.95
1998	535.7	155.5	123	21.10
1999	241.9	161.2	52.7	20.01
2000	406.7	314.2	105.5	12.53
2001	370.5	226.7	123.7	86.71
2002	61.6	86.3	135.2	0.38
2003	521.9	310.7	235.8	20.56
2004	128.5	389.7	110.9	9.52
2005	239.8	426.7	187.5	16.81
2006	92.5	97.1	52	2.81
2007	257.8	462.7	82.3	7.87
2008	213.8	124.6	299.7	0.34
2009	82.9	212.8	184.4	6.61
2010	566.4	217.5	82.9	2.23
2011	275	215.9	91.5	26.14
2012	208.2	90.9	60.2	0.08
2013	227.8	194.9	155.1	3.53
2014	323.3	256	235.5	6.28
2015	439.6	213.7	86	21.69
2016	185.3	107.9	260.7	0.64
2017	208.6	192.1	243.8	3.40
2018	322.3	204.8	363.7	8.85
2019	382.7	183.6	195.1	1.97
2020	163.9	236.7	171.3	0.11

Table 2.1.5. Nanticoke River March-May flow (CFS) and the Nanticoke River JI (Durell and Weedon 2021), 1957-2020.

Year	March	April	May	JI
1957	201.7	103.6	53	0.9
1958	373.2	300	183.7	17.9
1959	80.5	105.5	48.3	0.8
1960	137.6	167.6	68.3	2
1961	272.6	195	103.7	1.3
1962	241.5	173.2	68.8	3.1
1963	237.2	96.1	57.7	1.7
1964	211	208.8	96.5	6.1
1965	106.5	109.8	60.1	5.5
1966	63	70.2	110.9	1.6
1967	104	74	103.5	2.2
1968	174.9	101	79.9	3.9
1969	102.1	113.9	76.9	3
1970	122.5	188.7	119.3	6.3
1971	167.8	140.4	103.3	1.1
1972	157.8	147.3	170.9	5.2
1973	126.9	147.8	97.3	0.6
1974	110.3	113.6	96.5	2.1
1975	222.4	165.6	154.8	2.6
1976	107	81.3	54.1	1
1977	61.5	68.1	58	0.7
1978	255.8	147.1	185	2.3
1979	279.6	141.1	92	0.7
1980	154.7	193.7	149	0.8
1981	82.1	98.9	107.1	1.2
1982	125.4	128	91.6	3.1
1983	179.4	268.9	144	0.6
1984	241.7	248	189.6	0.8
1985	62.7	47.8	51.6	0.9
1986	101.9	70.9	47.7	1.2
1987	159.9	128.5	109.7	1.4
1988	83.6	103.7	101.1	0.3
1989	188.7	238.5	211.1	1.9
1990	101.9	139.4	218.6	0.6

Table 2.1.5 (continued).

Year	March	April	May	JI
1991	106.7	114.2	77.1	0.5
1992	122.7	94.9	90.1	1.7
1993	261	207.4	120.6	4.6
1994	421.5	285.1	125.4	9.1
1995	109.2	74.1	78.4	3.8
1996	147.6	180.4	173	19.1
1997	177.1	176.8	129.7	1.7
1998	316.5	181.2	153.9	2.7
1999	148.8	123.3	67.3	5.5
2000	178.8	184.9	113.2	10.9
2001	188.1	176.1	77.2	20.3
2002	51.7	89.2	96.9	4.9
2003	268.5	212	168.8	3.3
2004	95.9	171.5	97.7	9.7
2005	168.6	184.5	156.2	1.1
2006	96.5	80.4	46.2	1.6
2007	151.5	216.5	102.9	5.4
2008	92.1	68.6	120.2	0.7
2009	60.7	134.7	162.1	4.2
2010	388.7	177.7	82.9	3
2011	116	122.1	69.3	13
2012	82.1	55.9	45.3	0.4
2013	155.4	118.7	97.5	4.1
2014	170.2	171.5	125	5.1
2015	205.4	135.2	76.9	25.7
2016	129.2	97.3	132.3	0.7
2017	101.2	89.1	82.8	2.2
2018	110.9	106	227.7	5.8
2019	182.6	127.5	160.7	2.7
2020	69.3	84.9	92.5	1.4

Table 2.1.6. Correlation of spawning area specific juvenile indices (JI) with average monthly flow (cubic feet per second) for March, April, and May during 1957-2020. Statistic r is the correlation coefficient and P is the level of significance. N = 64 for each comparison.

Spawning Area	Statistic	March	April	May
Head-of-Bay	r	-0.11	0.21	0.43
	P	0.3838	0.0919	0.0003
Potomac	r	0.35	0.39	0.05
	P	0.005	0.0013	0.6878
Choptank	r	0.25	0.25	0.12
	P	0.0478	0.0446	0.3604
Nanticoke	r	0.24	0.32	0.07
	P	0.0571	0.0096	0.5732

Table 2.1.7. Average annual flow during two-month periods used in correlation analyses with spawning area JIs, 1957-2020. Average = 1957-2020 mean flow used to standardize spawning area flows.

Spawning area:	Head-of-Bay	Potomac	Choptank	Nanticoke
Flow months:	April-May	March-April	March-April	March-April
Year	Average Flow (CFS)			
1957	67,575	18,229	191	153
1958	108,466	29,206	356	337
1959	67,856	10,887	166	93
1960	77,964	24,448	183	152
1961	103,887	32,207	316	234
1962	98,648	35,007	325	208
1963	78,189	27,046	222	168
1964	103,173	24,567	269	210
1965	50,680	22,720	153	108
1966	56,618	12,130	45	67
1967	74,053	22,738	170	89
1968	46,059	16,339	212	139
1969	45,407	7,732	164	108
1970	96,811	25,193	250	155
1971	84,439	16,172	179	154
1972	103,426	26,152	231	153
1973	73,217	26,074	235	137
1974	78,047	16,015	211	112
1975	64,807	22,773	316	194
1976	53,559	11,695	122	94
1977	105,910	23,412	99	65
1978	99,422	29,709	354	202
1979	100,419	28,290	278	211
1980	86,123	27,082	266	174
1981	35,393	10,277	116	90
1982	79,995	21,339	200	127
1983	88,097	36,577	533	223
1984	88,910	41,035	449	245
1985	51,850	12,268	70	55
1986	77,920	18,670	151	87
1987	72,447	30,639	198	144
1988	40,483	9,970	116	93
1989	50,739	15,266	348	213
1990	44,690	9,792	180	120

Table 2.1.7 (continued).

Spawning area:	Head-of-Bay	Potomac	Choptank	Nanticoke
Flow months:	April-May	March-April	March-April	March-April
Year	Average Flow (CFS)			
1991	61,383	21,045	187	110
1992	63,902	18,685	155	109
1993	157,282	60,335	414	235
1994	145,038	47,900	583	354
1995	40,000	8,295	154	92
1996	74,468	26,262	315	164
1997	57,667	21,333	251	177
1998	93,633	38,132	349	250
1999	58,209	15,009	202	136
2000	88,025	16,878	361	182
2001	69,919	18,843	300	182
2002	43,577	9,154	74	70
2003	91,707	37,750	418	241
2004	80,247	26,067	257	133
2005	86,598	24,551	332	176
2006	30,021	7,730	95	89
2007	85,882	27,951	359	183
2008	91,886	20,571	170	81
2009	48,301	10,822	147	97
2010	63,776	30,040	395	285
2011	155,230	39,021	246	119
2012	34,200	12,898	151	69
2013	48,655	16,987	212	137
2014	69,046	18,500	290	171
2015	70,654	21,031	329	171
2016	38,148	10,093	147	114
2017	75,359	12,015	200	95
2018	67,873	17,559	265	108
2019	71,674	26,581	285	156
2020	62,062	12,719	200	77
1957-2020 Average	62,616	22,128	242	143

Table 2.1.8. Correlations of spawning period flow among spawning areas during 1957-2020. All correlations were significant at $P < 0.0001$. N = 64 for each comparison.

	Nanticoke	Choptank	Potomac
Choptank	0.88		
Potomac	0.72	0.76	
Head-of-Bay	0.56	0.60	0.84

Table 2.1.9. Correlations among area specific juvenile indices for the entire time-series (1957-2021, N = 65) and the recent period of high productivity (1993-2021, N = 29).

		Head-of-Bay	Potomac R.	Choptank R.
Area	Statistic		1957-2021	
Potomac R.	r	0.48		
	P	<.0001		
Choptank R.	r	0.32	0.48	
	P	0.0084	<.0001	
Nanticoke R.	r	0.42	0.36	0.66
	P	0.0006	0.0033	<.0001
			1993-2021	
Potomac R.	r	0.54		
	P	0.0023		
Choptank R.	r	0.3	0.49	
	P	0.1192	0.0068	
Nanticoke R.	r	0.3	0.32	0.67
	P	0.1082	0.0857	<.0001

Table 2.1.10. Major land use estimates for Head-of-Bay, Potomac River, Choptank River, and Nanticoke River based on Chesapeake Conservancy high resolution estimates for 2013-2014 (Chesapeake Conservancy 2022). Agriculture = cropland and pasture/hay categories; Wetlands = tidal, floodplain, and other; Forest is their forest category; and Developed = 7 remaining categories. Estimates were made by M. Topolski, MD DNR.

	Spawning area			
Land use	Head-of-Bay	Potomac R.	Choptank R.	Nanticoke R.
Developed	14.7	20.8	15.3	16.2
Wetland	1.7	1.2	17.9	18.9
Forest	60.2	57.0	17.8	25.3
Agriculture	23.4	21.0	49.1	39.6

Figure 2.2.1. Location of Striped Bass spawning and larval nursery habitat in MD's portion of Chesapeake Bay based on average salinity less than 2 ppt (see Job 3, Uphoff et al. 2017). These areas encompass spawning areas described in (Hollis 1967), but do not exactly duplicate them.

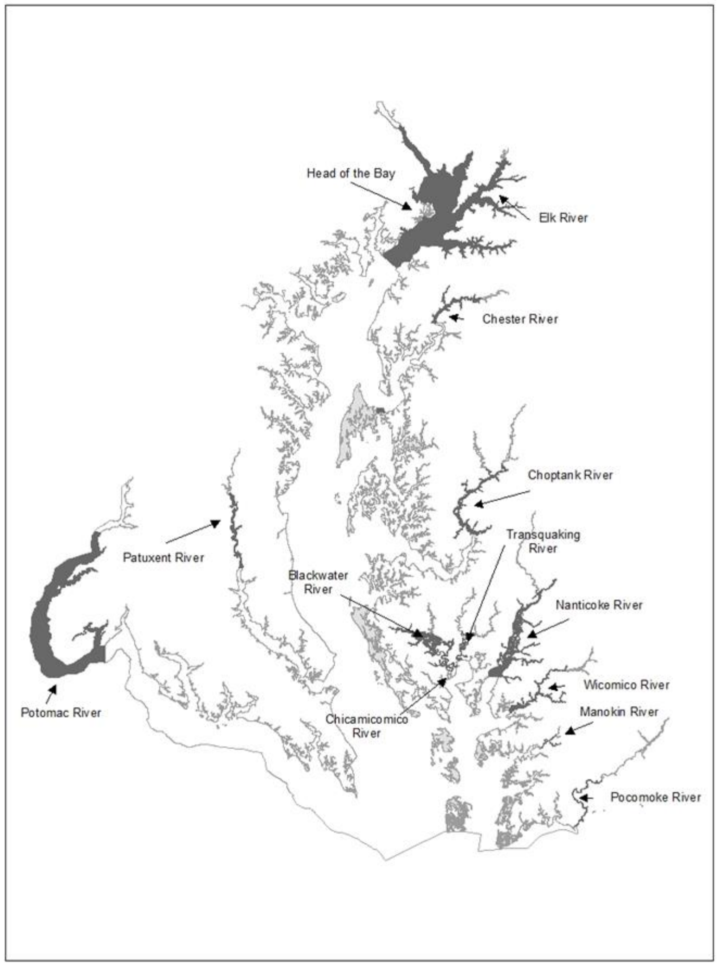


Figure 2.1.2. Spawning area specific proportion of tows with Striped Bass eggs (*Ep*) estimated from surveys in juvenile index rivers conducted during 1955-2021. Elk River represents a portion of the Head-of-Bay.

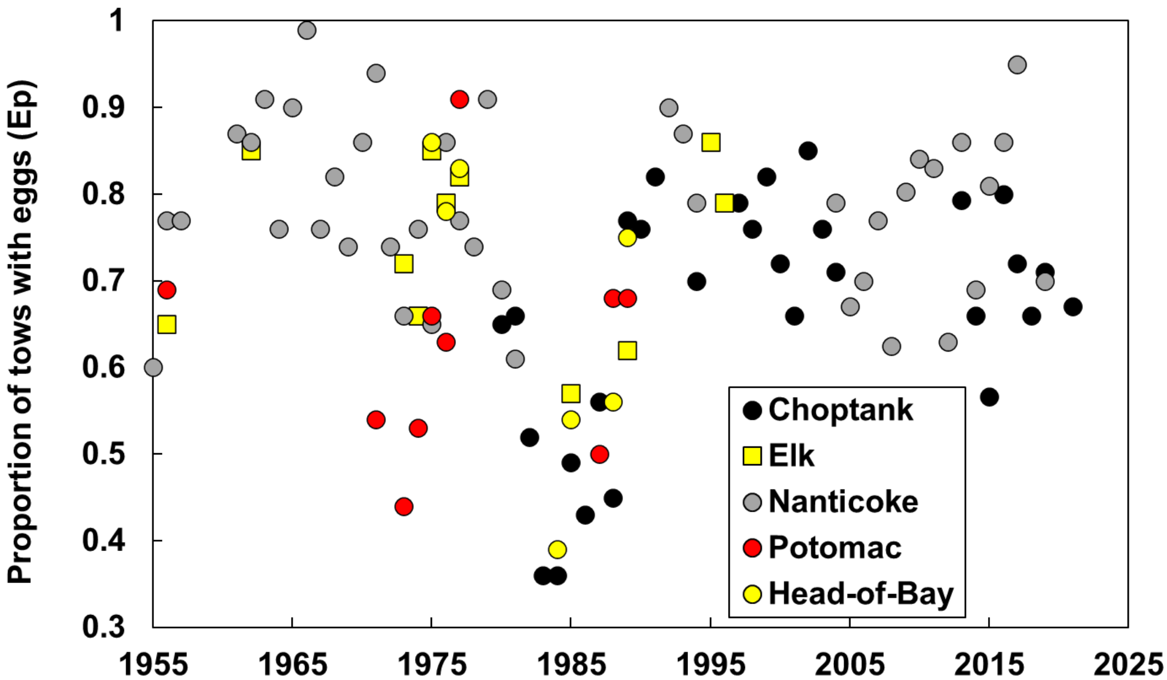


Figure 2.1.3. Baywide (Maryland's spawning areas) proportion of tows with Striped Bass eggs (*Ep;* diamond) and its 90% CI (line) estimated from surveys in juvenile index rivers conducted during 1955-2021. Baywide estimate pools available data from spawning surveys conducted in four areas surveyed for the juvenile index: Head-of-Bay, Potomac River, Nanticoke River, and Choptank River.

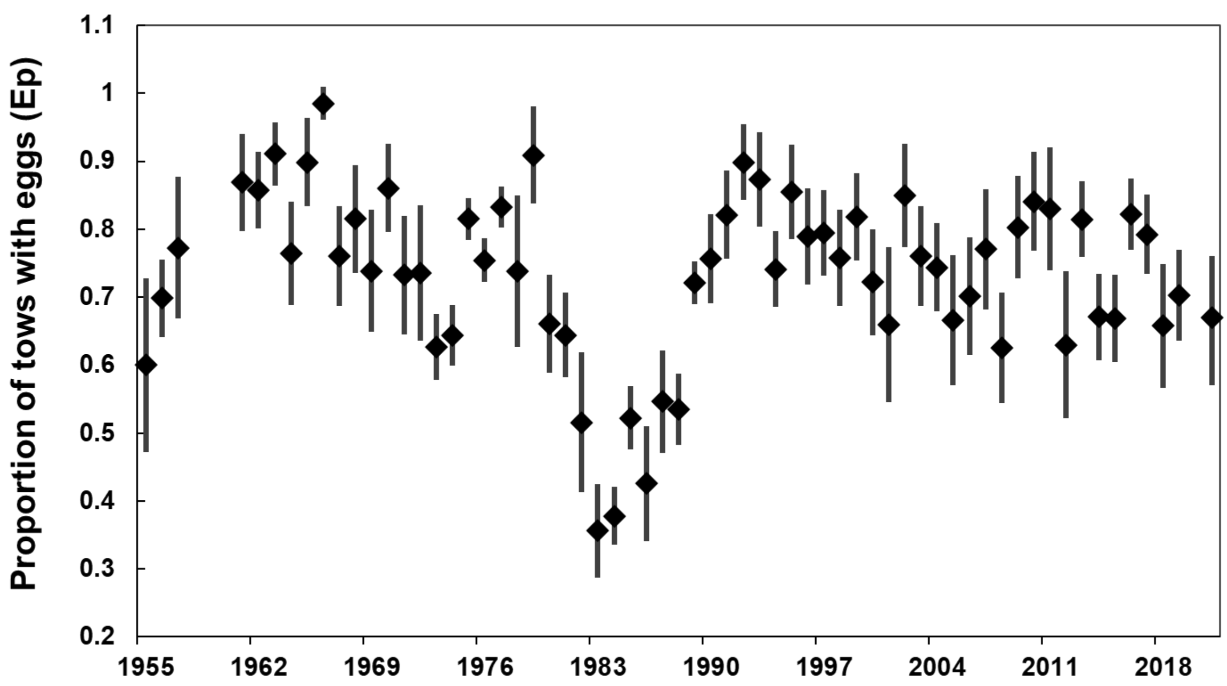


Figure 2.1.4. Baywide Striped Bass juvenile indices (geometric mean catch per standard seine haul; diamonds) and their 90% confidence interval (line) estimated for Maryland's major spawning areas during 1957-2021 (Durrell and Weedon 2021).

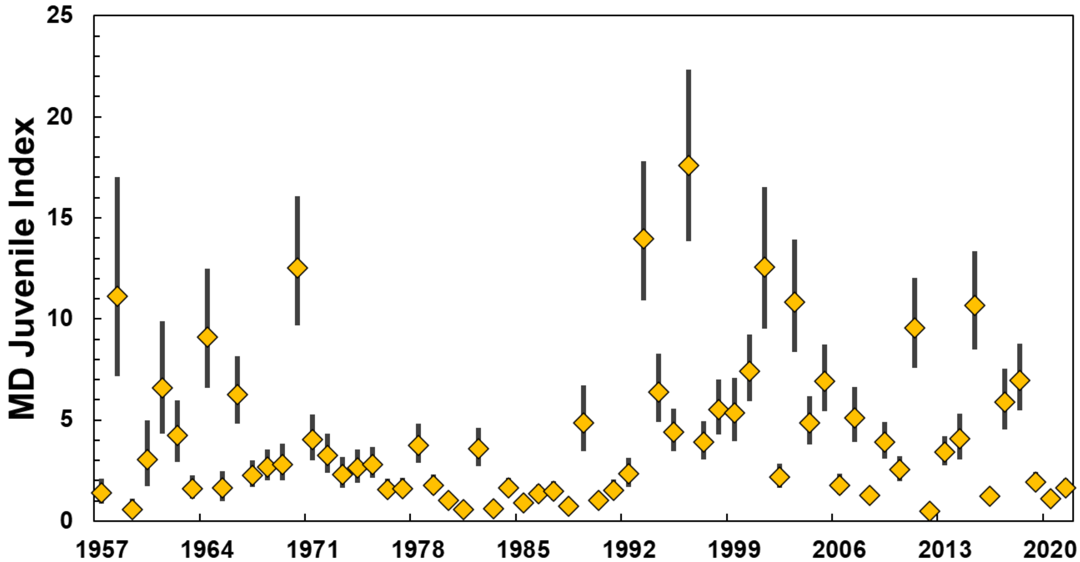


Figure 2.1.5. Relative larval survival (baywide JI / baywide Ep) mean and 90% CIs, 1957-2021. Quartiles (green and grey dashed lines) based on 1957-2009 (ASMFC JI base years); upper quartile = green dashed line and lowest quartile = grey dashed line.

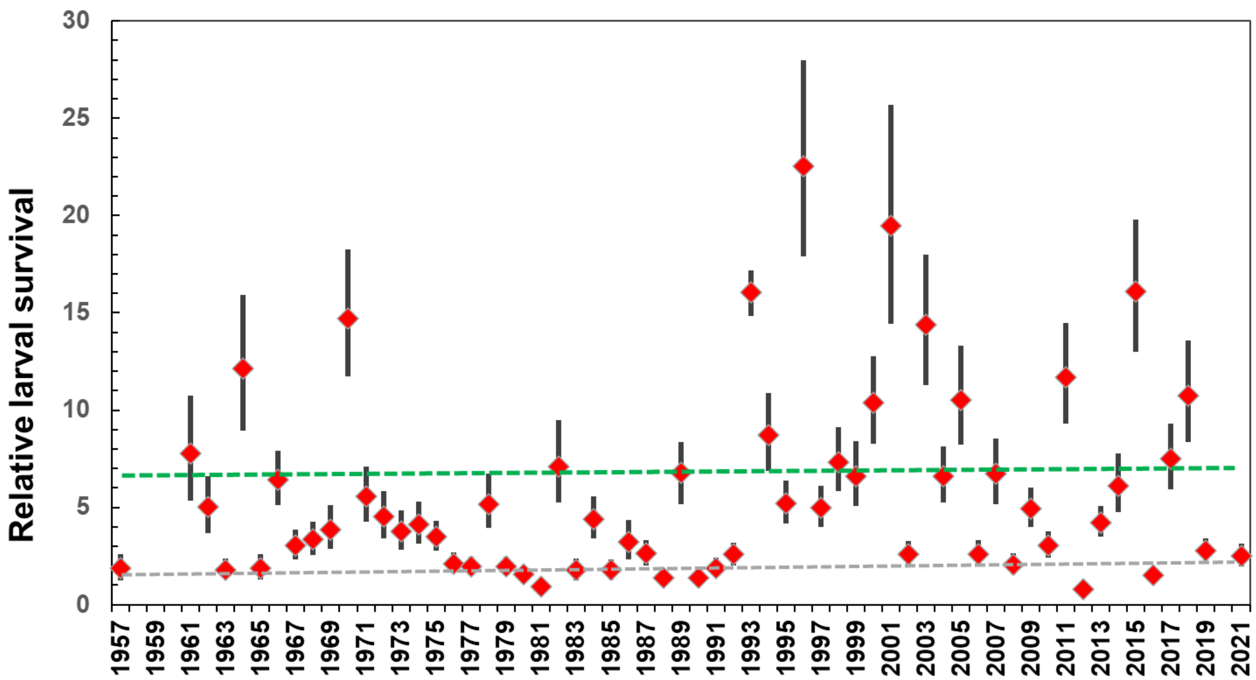


Figure 2.1.6. Difference of standardized (std) JI and standardized relative larval survival (RLS) as proportion of standardized JI during 1957-2021. Large negative deviations indicate overfishing in 1982-1988. Indices standardized to mean of common years (same scale).

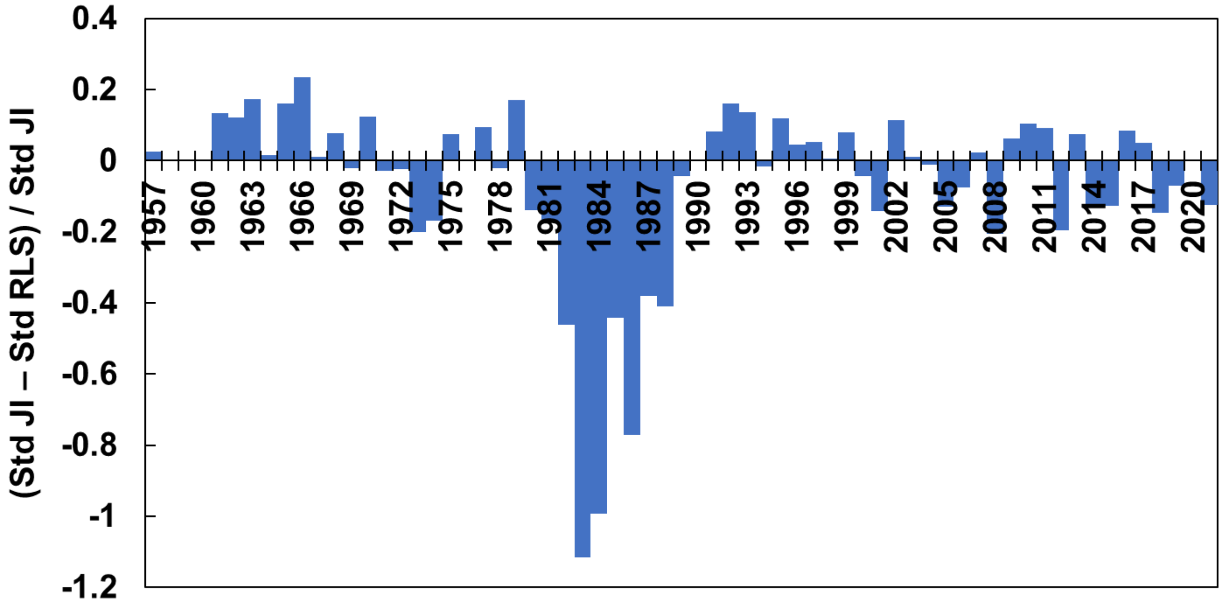
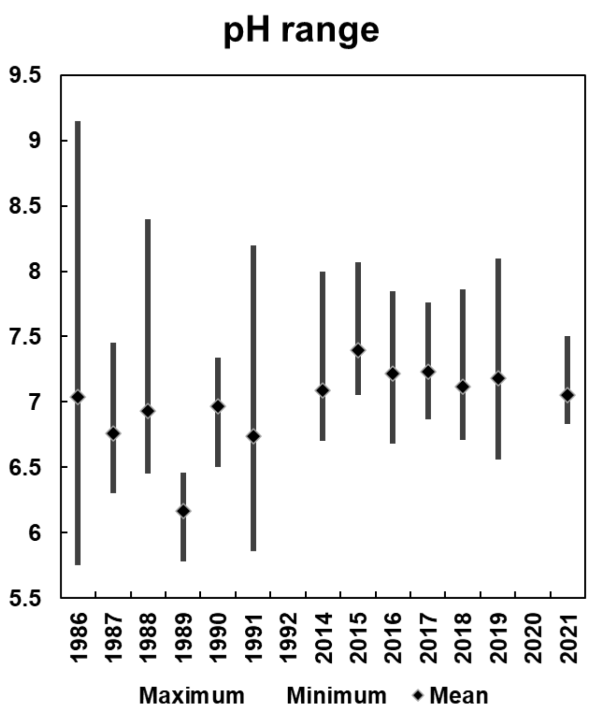


Figure 2.1.7. Choptank pH and alkalinity mean and range during April 1 – May 7, 1986-1991 and 2014-2021.



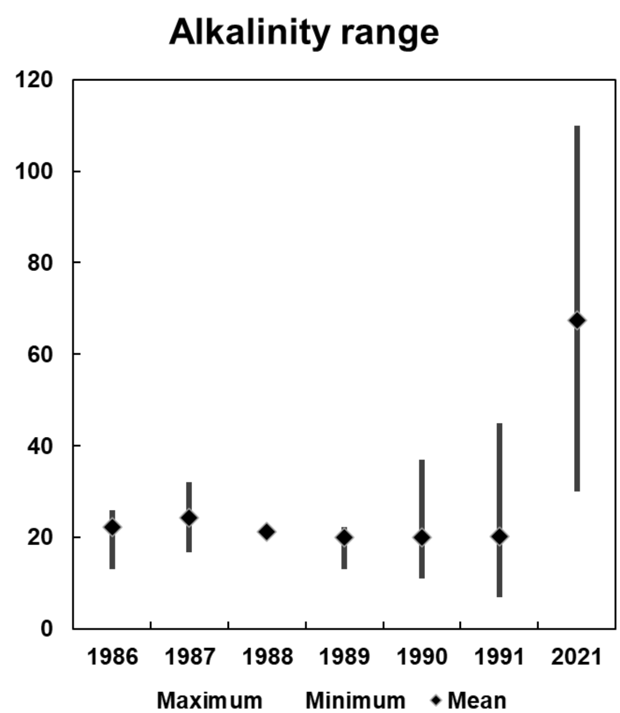


Figure 2.1.8. Cumulative proportion of eggs collected in ichthyoplankton surveys, by 1ºC temperature increment, in the Choptank and Nanticoke Rivers. Surveys with counts only. Surveys were conducted between 1954 and 1993. Number of eggs were 113,503 for the Choptank River and 105,336 for the Nanticoke River.

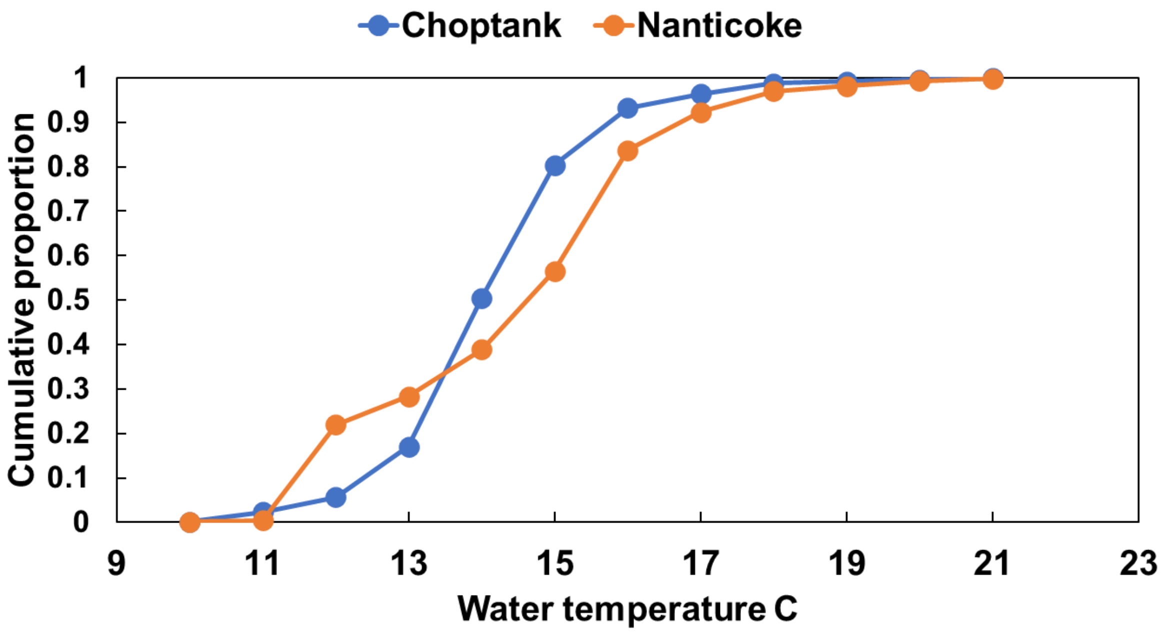


Figure 2.1.9. Cumulative proportion of larvae (prolarvae and postlarvae) collected in ichthyoplankton surveys, by 1ºC temperature increment, in the Choptank and Nanticoke Rivers. Surveys with counts only. Surveys were conducted during 1980-1989 in Choptank River and 1992-1993 in Nanticoke River. Gears in these surveys were considered capable of sampling larval life stages effectively. Number of larvae were 42,562 for the Choptank River. Nanticoke River data were reported as density and this was used as a substitute for number for estimating cumulative proportion.

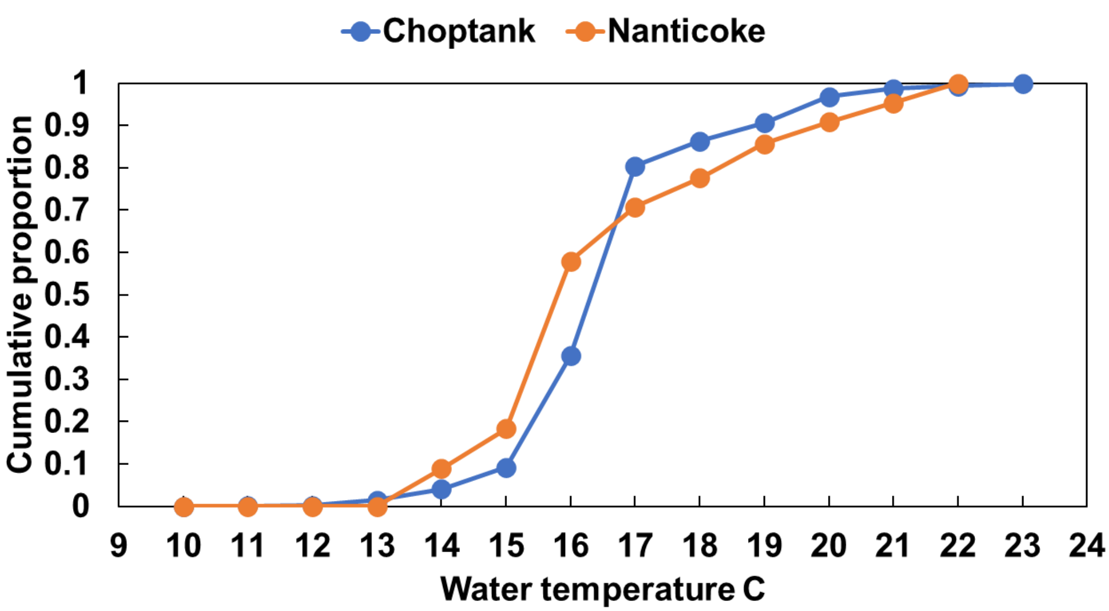


Figure 2.1.10. Days from April 1 (day = 0) that the first egg was collected in Choptank River and Nanticoke River Striped Bass ichthyoplankton surveys during 1954-2021. Median = median day for both rivers combined (day 7).

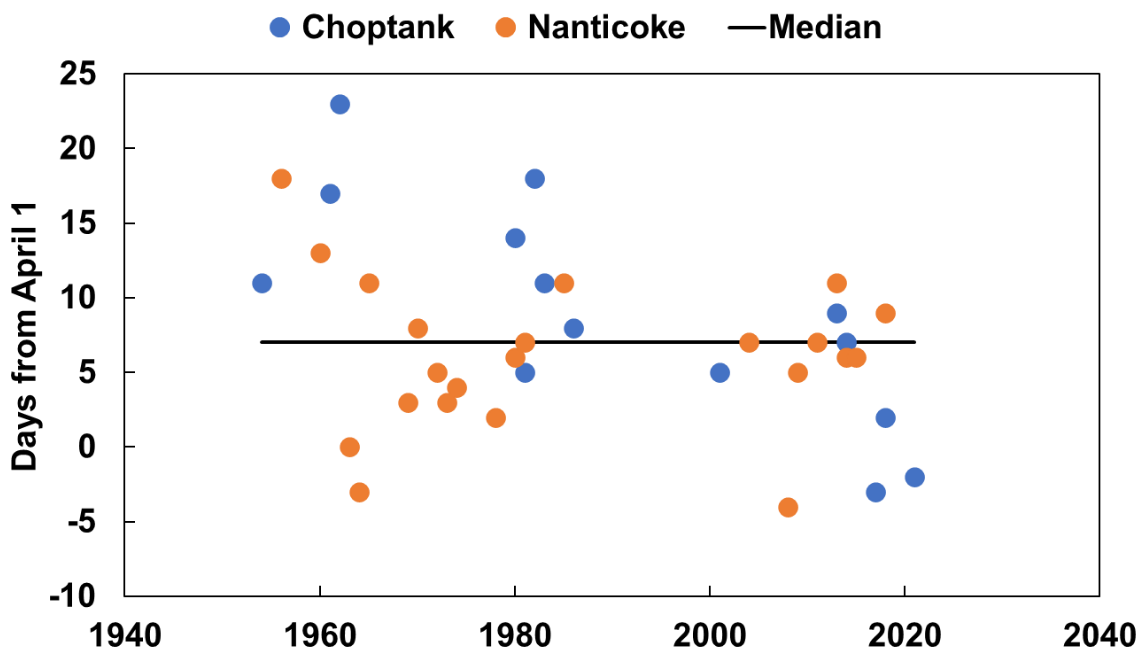


Figure 2.1.11. Days from April 1 (day = 0) that 12ºC was reached in Choptank River and Nanticoke River Striped Bass ichthyoplankton surveys during 1954-2021. Median = median day for both rivers combined (day 11).

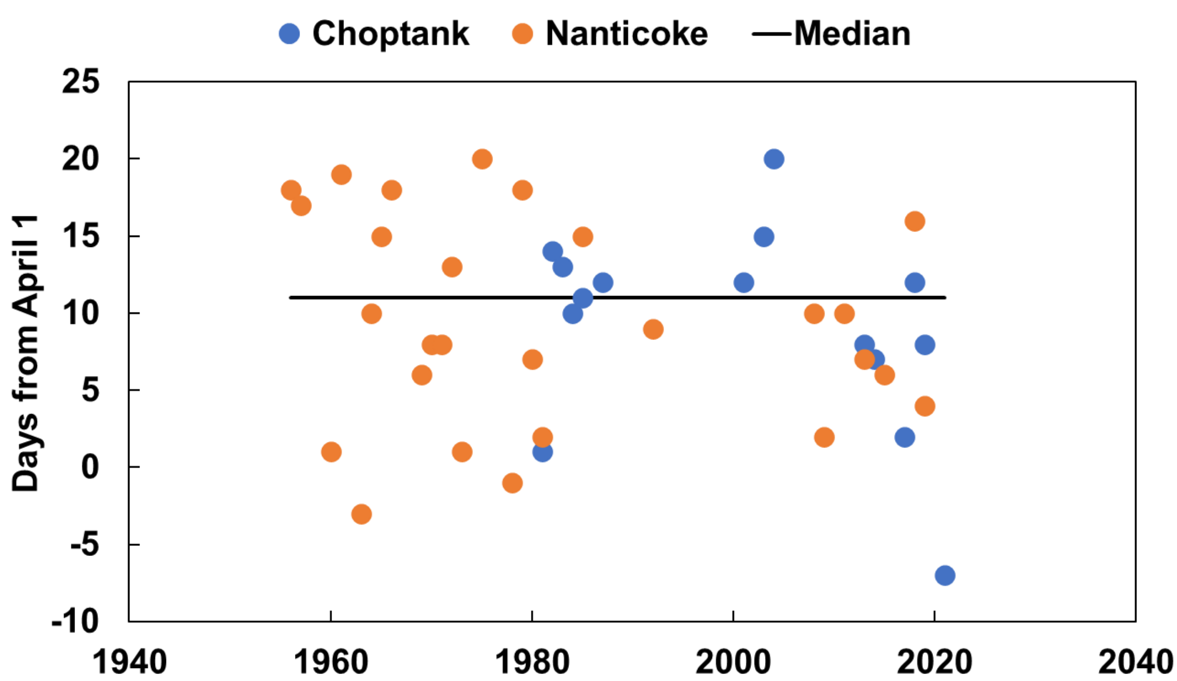


Figure 2.1.12. Days from April 1 (day = 0) that 16ºC was reached in Choptank River and Nanticoke River Striped Bass ichthyoplankton surveys during 1954-2021. Median = median day for both rivers combined (day 19).

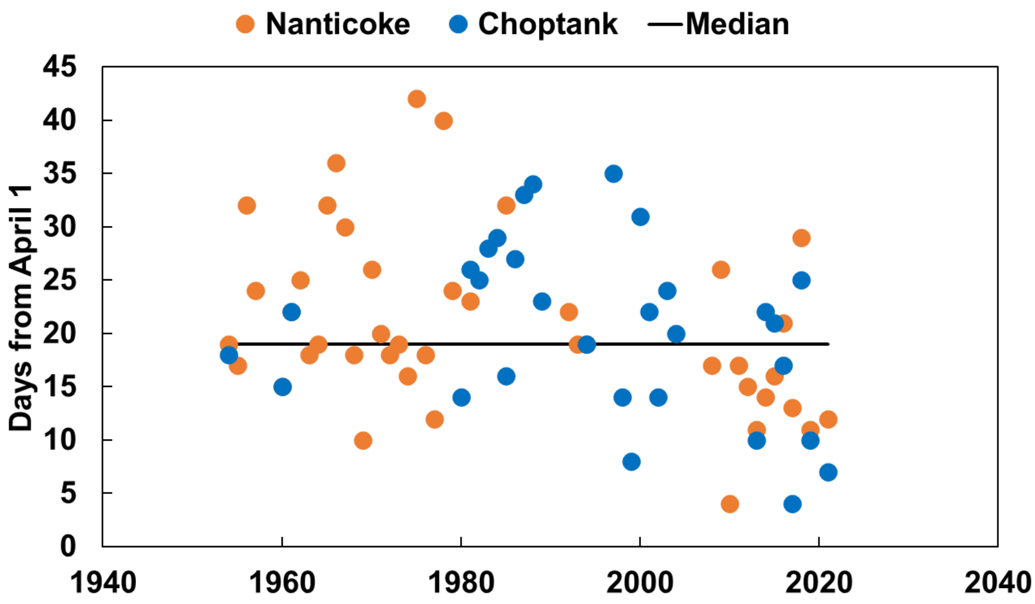


Figure 2.1.13. Days from April 1 (day = 0) that 20ºC was reached in Choptank River and Nanticoke River Striped Bass ichthyoplankton surveys during 1954-2021. Median = median day for both rivers combined (day 41).

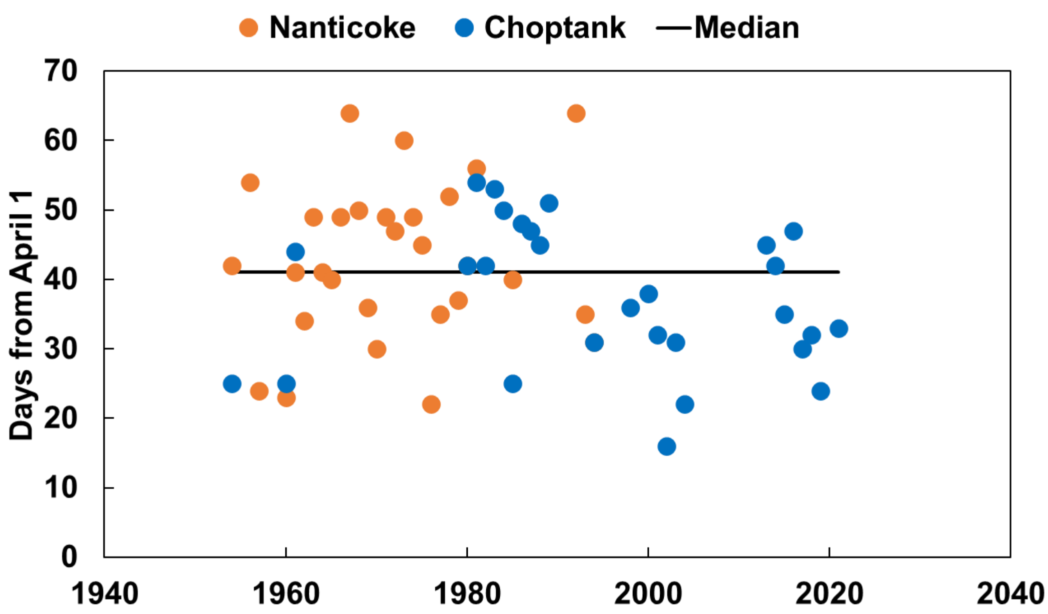


Figure 2.1.14. Surveys with all days from April 1 (day = 0) that 12ºC, 16ºC, and 20ºC were reached in the Nanticoke River present during 1954-1992.

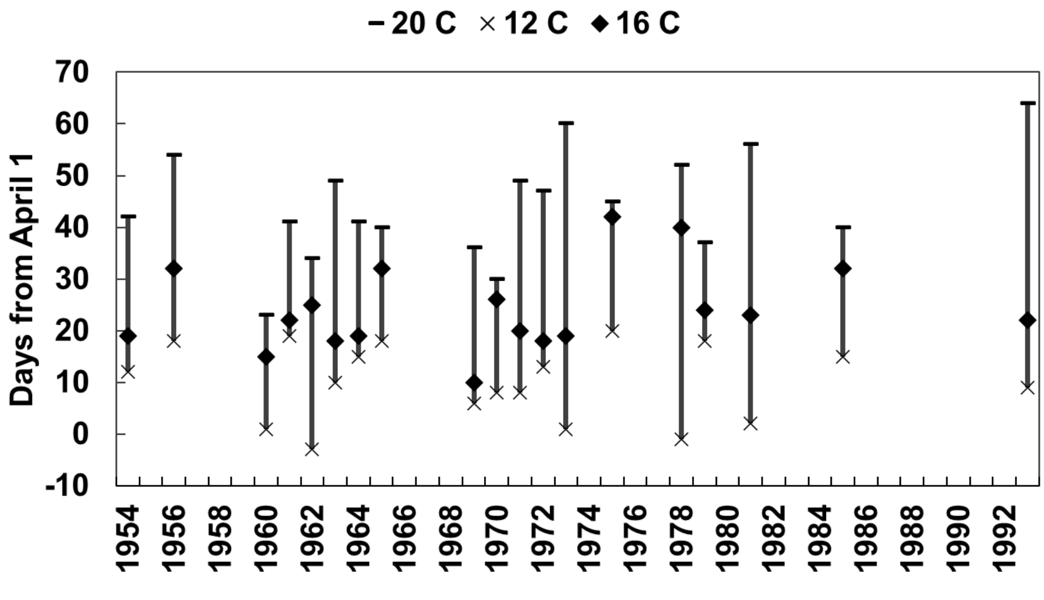


Figure 2.1.15. Days from April 1 (day = 0) that 12ºC, 16ºC, and 20ºC were reached in the Choptank River during 1954-2021.

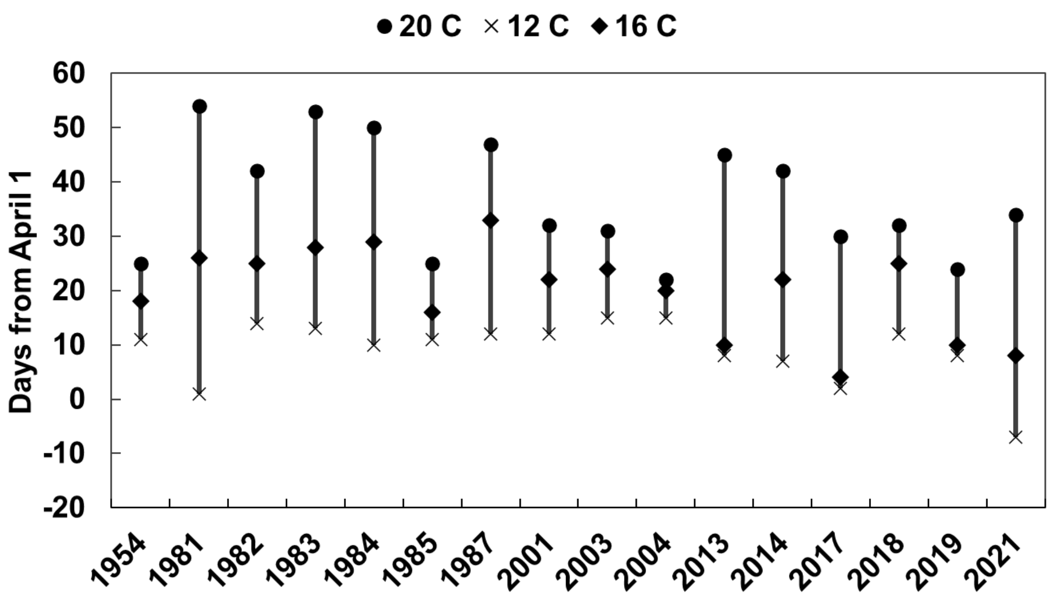


Figure 2.1.16. Head-of-Bay 1993-2020 April-May mean standardized flow vs the Head-of-Bay geometric mean juvenile index (JI), with JI percentiles. Annual mean flows are standardized to the 1957-2020 mean.

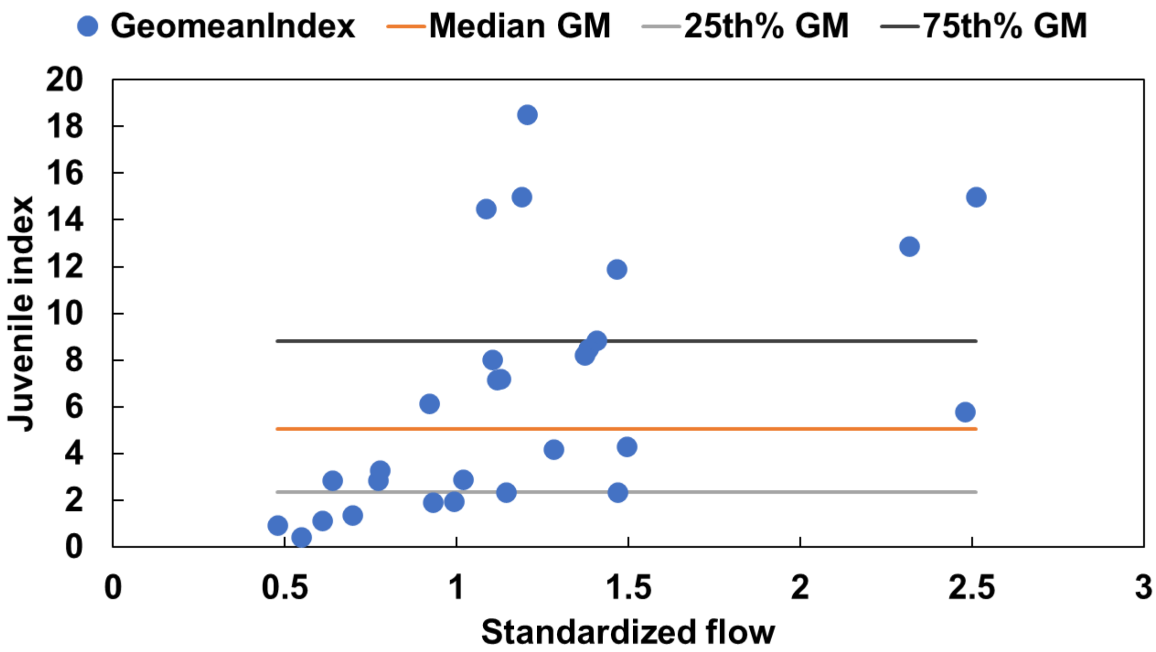


Figure 2.1.17. Potomac River 1993-2020 March-April mean standardized flow vs the Head-of-Bay geometric mean juvenile index (JI), with JI percentiles. Annual March-April mean flows are standardized to the 1957-2020 mean.

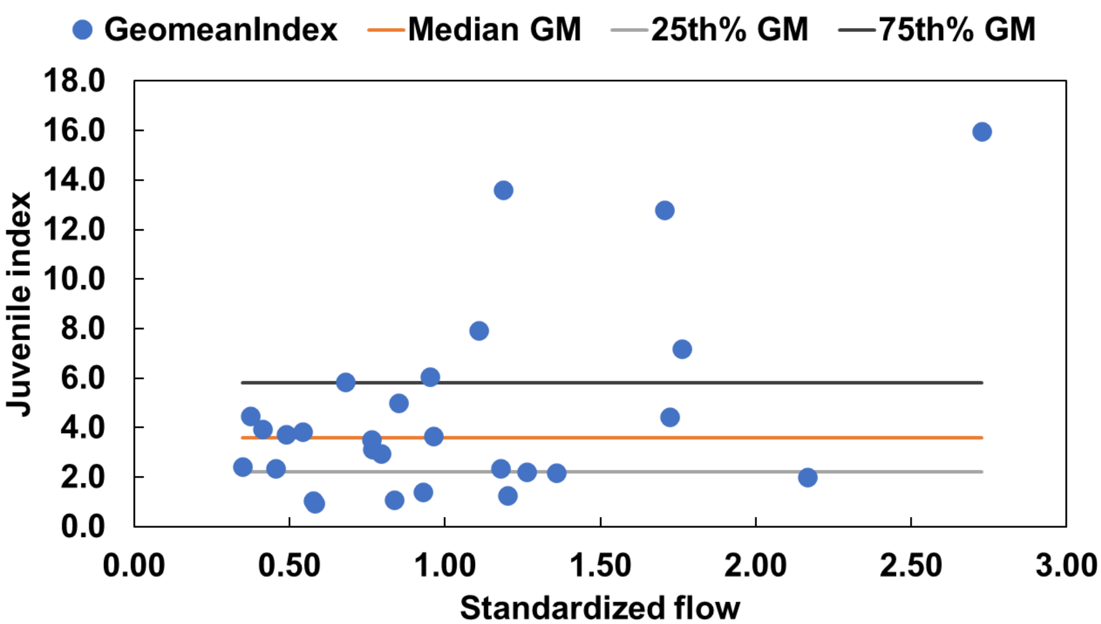


Figure 2.1.18. Choptank River 1993-2020 March-April mean standardized flow vs the Head-of-Bay geometric mean juvenile index (JI), with JI percentiles. Annual March-April mean flows are standardized to the 1957-2020 mean.

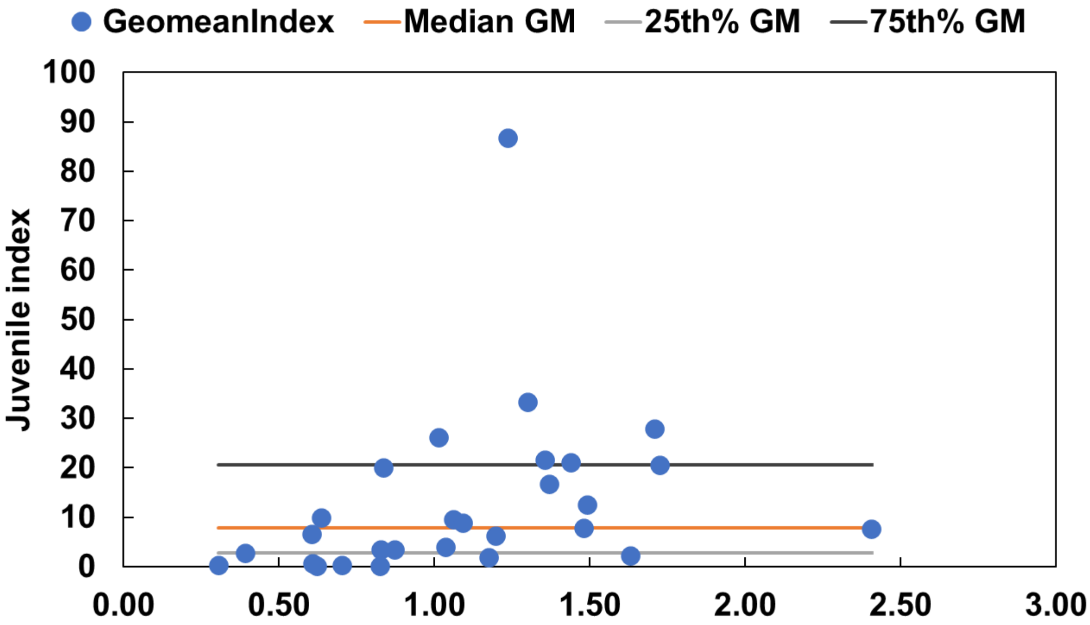
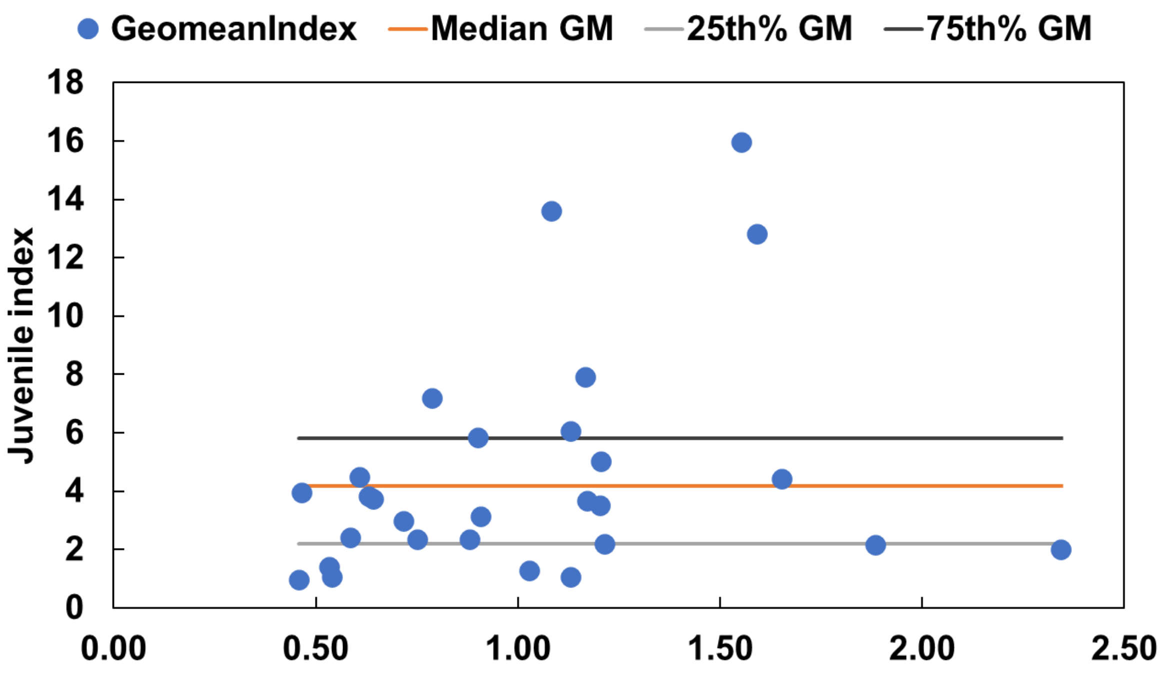


Figure 2.1.19. Nanticoke River 1993-2020 March-April mean standardized flow vs the Head-of-Bay geometric mean juvenile index (JI), with JI percentiles. Annual March-April flows are standardized to the 1957-2020 mean.



MD - Marine and estuarine finfish ecological and habitat investigations
Project 1: Development of habitat-based reference points for recreationally important Chesapeake Bay fishes of special concern
Project 1, Section 3 - Estuarine Fish Community Sampling

Alexis Park, Carrie Hoover, Margaret McGinty, Jim Uphoff

Changes to Project 1 Activities due to Coronavirus

The choice of subestuaries sampled during summer 2021 was limited by a COVID-19 hiring freeze which limited personnel. The subestuaries chosen had resource issues of interest and continued long-term data sets.

Introduction

Human population growth since the 1950s added a suburban landscape layer to the Chesapeake Bay (or Bay) watershed (Brush 2009) that has been identified as a threat (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; Meals et al. 2010; Sharpley et al. 2013; Zhang et al. 2016). These are the basic trade-off in land use facing Maryland as its population grows (Maryland Department of Planning; MD DOP 2020a) and they have ecological, economic, and societal consequences (Szaro et al. 1999).

Water quality and aquatic habitat are altered by agricultural activity and urbanization. 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 of the Bay (Hagy et al. 2004; Kemp et al. 2005; Fisher et al. 2006; Brush 2009; Zhang et al. 2016). Land in agriculture has been relatively stable but farming itself has become much more intensive (fertilizer and pesticide use has increased) to support crop production and 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; McBryan et al. 2013; Branco et al. 2016) that act as ecological stressors and alter fish production. Extended exposure to biological and environmental stressors affects fish condition and survival (Rice 2002; Barton et al. 2002; Benejam et al. 2008; Benejam et al. 2010; Branco et al. 2016). 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. Todd et al. (2019) reviewed impacts of three interacting drivers of marine urbanization (resource exploitation, pollution, and proliferation of manmade marine structures) and described negative impacts that were symptomatic of urban marine ecosystems. Taylor and Suthers (2021) outlined how urban estuarine fisheries management was defined by unique ecological attributes of urbanized estuaries, the socio-economic objectives of anglers, and bottlenecks to productivity of exploited species.

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. 2020). Uphoff et al. (2011) estimated target and limit impervious surface reference points (ISRPs) for productive juvenile and adult fish habitat in brackish (mesohaline; 5.0 – 18.0 ‰; Oertli, 1964) 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 of mesohaline subestuaries at a target of 5.5 % IS (expressed as IS equivalent to that estimated by the methodology used by Towson University for 1999-2000) 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 5.0 mg/L (target DO). Mean bottom DO seldom exceed 3.0 mg/L above 10 % IS (suburban threshold; Uphoff et al. 2011). Although bottom DO concentrations were negatively influenced by development (indicated by IS) in mesohaline subestuaries, Uphoff et al. (2020) have found adequate concentrations of DO in bottom channel habitat of tidal-fresh (0 – 0.5 ‰) and oligohaline (0.5 – 5.0 ‰) 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 absent, allowing for more mixing.

In 2021, we continued to evaluate summer nursery and adult habitat for recreationally important finfish in tidal-fresh, oligohaline, and mesohaline subestuaries of the Chesapeake Bay. In this section, we analyzed the associations of land use (i.e., agriculture, forest, urban, and wetlands) and C/ha (structures per hectare) on the annual median bottom DO among subestuaries sampled during 2003 – 2021. We evaluated the influence of watershed development on target species presence-absence and abundance, total abundance of finfish, and finfish species richness. We continued to examine Tred Avon River; a tributary of the Choptank River located in Talbot County (Table 3-1; Figure 3-1). We sampled the Sassafras River, located at the Head-of-Bay, for a second year in 2021 (Table 3-1; Figure 3-1). We examined associations among relative abundance of all finfish from Choptank River and the Head-of-Bay with Tred Avon River and Sassafras River to evaluate potential contributions of the two large outside regions to the abundance in tributaries and subestuaries in our study. We added a more detailed evaluation of species composition, abundance, and richness to our analysis to better understand the possible changes occurring throughout the subestuaries of the Chesapeake Bay.

Methods

Each subestuary sampled was classified into a salinity category based on the Venice System for Classification of Marine Waters (Oertli 1964). Tidal-fresh ranged from 0 – 0.5 ‰; oligohaline, 0.5 – 5.0 ‰; and mesohaline, 5.0 – 18.0 ‰ (Oertli 1964). Salinity influences distribution and abundance of fish (Allen 1982; Cyrus and Blaber, 1992; Hopkins and Cech 2003) and DO (Kemp et al. 2005). We calculated an arithmetic mean of all bottom salinity and measurements for all years available through 2020 to determine salinity class of each subestuary. We sampled one mesohaline Tred Avon River subestuary located in Talbot County during 2021. We have sampled Tred Avon River since 2006. In 2021 we sampled the Sassafras River, a tidal-fresh subestuary located between Kent and Cecil Counties, for a second summer. In 2021, we were able to conduct both bottom trawling and beach seining; we were not able to conduct beach seines because of a contact advisory due to high *Microcytis* levels in 2020.

Sampling of the Tred Avon River (Figure 3-1) began in 2006, one year ahead of a substantial development project. We have continued monitoring Tred Avon River in anticipation of DO and fish community changes as its watershed continues to develop (Figure 3-1). Talbot County and the town of Easton (located at the upper Tred Avon River) have active programs to mitigate runoff and provides an opportunity to evaluate how well up-to-date stormwater management practices maintain subestuary fish habitat. Starting in 2012, we assessed adjacent subestuaries that were less developed (Figure 3-1): Broad Creek (through 2017 and in 2020) and Harris Creek (through 2016; Uphoff et al. 2015; 2016; 2017; Figure 3-1).

We used property tax map-based counts of structures in a watershed (C), standardized to hectares (C/ha), as our indicator of development (Uphoff et al. 2012; Topolski 2015). Estimates of C/ha and Maryland Department of Planning land use and water percentages were used for analyses of data from mesohaline subestuaries sampled during 2003 – 2021 (Table 3-2). Maryland DOP only has structure estimates available through 2020; 2020 estimates were used to represent 2021 in analyses. Methods used to estimate development (C/ha) and land use indicators (percent of agriculture, forest, wetlands, urban land use, and water in the watershed) are explained in **General Spatial and Analytical Methods used in Project 1, Sections 1-3.**

Development targets and limits, and general statistical methods (analytical strategy and equations) are described in **General Spatial and Analytical Methods used in Project 1, Sections 1 – 3** as well. Specific spatial and analytical methods for this section of the report are described below.

2021 Sampling - Ideally, four evenly spaced haul seine and bottom trawl sample sites were in the upper two-thirds of each subestuary. Lower portions of a subestuary were not sampled to minimize the impact of mainstem water and maximize subestuary watershed influence. We used GPS to record latitude and longitude at the beginning and end of each trawl site, while latitude and longitude at seining sites were taken at the seine starting point on the beach. We focused on using previously sampled historical sites in 2021 at each of the previously sampled subestuaries unless they were no longer accessible. Seine sites in the Sassafras River were sampled for the first time in 2021; during 2020, seine sites were established but not sampled due to health risks from a harmful algal bloom (HAB) that occurred within the river throughout the sampling season. Seine data was acquired from a juvenile index (JI) monitoring station at Sassafras River Natural Resource Management Area (NRMA) for 2020 to examine the inshore fish community (E. Durell, MD DNR, personal communication); in 2021, we evaluated seine data between JI and FHEP seine sites. Sites were sampled once every two weeks during July – September, totaling six visits per system during 2021. The number of total samples collected from each system varied based on the number of sites available, SAV interference, weather/tidal influences, and equipment issues. All sites on one river were sampled on the same day, usually during morning through mid-afternoon. Sites were numbered from upstream (station 01) to downstream (station 04). The crew determined whether to start upstream or downstream based on tidal direction; this helped randomized potential effects of location and time of day on catches and dissolved oxygen, as well as assisted the crew with seine site availability. 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 due to availability, then the crew leader deviated from the sample route to accommodate this need. Bottom trawl sites were generally in the channel, adjacent to haul seine sites. At some sites, beach seines could not be made because of permanent obstructions, dense SAV beds, or lack of beaches. Bottom trawl and beach seine sampling was conducted one right after the other at a site to minimize time of day or tidal influences between samples.

Water Quality Sampling - Water quality parameters were recorded at all stations for every individual sampling event in 2021. Temperature (ºC), DO (mg/L), conductivity (µS/cm), salinity (parts per thousand; ppt = ‰), and pH were recorded at the surface, middle, and bottom of the water column at the trawl sites depending on depth, and at the surface of each 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 mg/L and a threshold of 3.0 mg/L (Batiuk et al. 2009; Uphoff et al. 2011). The target criterion was originally derived from laboratory experiments but was also associated with asymptotically high presence of target species in trawl samples from bottom channel habitat in mesohaline subestuaries (Uphoff et al. 2011). Target DO was 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. Presence of target species in bottom channel trawls declined sharply when bottom DO fell below the 3.0 mg/L threshold in mesohaline subestuaries (Uphoff et al. 2011). We estimated the percentages of DO samples in each subestuary that did not meet the target or threshold for all DO samples (surface, middle, and bottom DO) and for bottom DO alone. Percentages not meeting target or threshold conditions were termed “violations”, but the term did not have a regulatory meaning. The percentages of

DO measurements that met or fell below the 5 mg/L target (V_{target}) or fell at or below the 3 mg/L threshold ($V_{threshold}$) were estimated as:

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

and

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

where N_{target} was the number of measurements meeting or falling below 5 mg/L, $N_{threshold}$ was the number of measurements falling at or below 3 mg/L, and N_{total} was total sample size. Separate Pearson correlation analyses were conducted for surface or bottom temperature or C/ha with surface or bottom DO for all subestuaries sampled since 2003. This analysis explored multiple hypotheses related to DO conditions. Structures per hectare estimates were considered proxies for nutrient loading and processing due to development in the subestuaries in this analysis (Uphoff et. al 2011). Water temperature would influence system respiration and stratification (Kemp et al. 2005; Murphy et al. 2011; Harding et al. 2016). Conducting correlation analyses by salinity classification provided a means of isolating the increasing influence of salinity on stratification from the influence of 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; Harding et al. 2016). 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 survey 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.

We obtained land use estimates for our watersheds from the Maryland Department of Planning (MD DOP) for 2002 and 2010 (MD DOP 2020b). The MD DOP provides agriculture, forest, urban, and wetlands estimates periodically rather than annually, but C/ha is estimated annually. Median summer bottom DO estimates made before 2010 were compared with 2002 MD DOP land use estimates and those made for 2010 – 2021 were matched with 2010 MD DOP estimates (the most current available). Four categories of land use (percent in agriculture, forest, urban, and wetlands) were estimated based on the land portion of the watershed (water area was excluded from these categories). A fifth category, percent in water, was estimated based on the water plus land area of the watershed. Newer land use estimates have not been released by MD DOP.

We analyzed the associations of land use (i.e., agriculture, forest, urban, and wetlands) and C/ha (structures per hectare) with annual median bottom DO among mesohaline systems sampled during 2003 – 2021 using correlation analysis. We further examined the influence of percent of land in agriculture on median bottom DO using linear, multiple linear, and quadratic regression models. We focused this analysis on mesohaline subestuaries because bottom DO does not exhibit a negative response to development in the other salinity categories.

Water Quality in Mesohaline Tributaries of the Choptank River - In 2021, we sampled four stations in Tred Avon River (Figure 3-2). We contrasted Tred Avon River to previously sampled tributaries of the Choptank River, Broad Creek (sampled during 2012 – 2017, 2020) and Harris Creek (2012 – 2016; Figure 3-3). Trajectories of C/ha since 1950 were plotted for the three Choptank tributaries, Broad Creek, Harris Creek, and Tred Avon River. Bottom DO measurements during 2006 – 2021 were plotted against C/ha and percent of target and threshold DO violations were estimated using all measurements combined (surface, middle, and bottom) and for bottom DO only. Annual mean bottom DO (depth most sensitive to violations) in Tred Avon River at each station for 2006 – 2021 was estimated and plotted by year. We examined correlations of Secchi depths, SAV coverage, DO, pH, and salinity within the three Choptank tributaries.

An ANOVA was used to examine differences in mean bottom DO among stations in Broad Creek, Harris Creek, and Tred Avon River. Tukey Studentized Range and Tukey Honestly

Significant Difference (HSD) tests examined whether stations within each tributary and subestuary were significantly different from one another. An overall median DO was calculated for all time-series data available for each system and used to detect how annual station DO compared with the time-series median. Correlation analysis of annual median DO measurements was used among the three Choptank tributaries.

Water Quality in Head-of-Bay Subestuaries - In 2021, we sampled the Sassafras River for a second summer and seined for the first time (Figure 3-2). Sassafras represented the only Maryland low salinity subestuary with a watershed dominated by agriculture. Sassafras River was within the Head-of-Bay region and Bohemia River (2006), Bush River (2006 – 2010), Gunpowder River (2009 – 2016), Middle River (2009 – 2017), and Northeast River (2007 – 2017) were previously sampled in this region (Figure 3-4). Trajectories of C/ha since 1950 were plotted for the Head-of-Bay subestuaries. Bottom DO measurements during 2006 – 2021 were plotted against C/ha and percent of target and threshold DO violations were estimated using all measurements combined (surface, middle, and bottom) and for bottom DO only. Annual mean bottom DO for each station within the Head-of-Bay subestuaries for 2006 – 2021 was estimated and plotted by year. We examined annual medians of Secchi depths, DO, pH, and salinity within the Head-of-Bay subestuaries, additional analyses involving Secchi depths, DO, pH, and salinity were limited due to the quantity of data. A subset of Bush River sampling years, 2006 – 2010, were summarized for this report; years sampled by citizen volunteers, 2011 – 2020, were excluded because not all stations were sampled consistently each year, therefore, data was incomplete.

An ANOVA was used to evaluate station differences in mean bottom DO; Tukey Studentized Range and Tukey Honestly Significant Difference (HSD) tests examined which station(s) within each subestuary were significantly different from others. An overall median bottom DO was calculated for all time-series data available for each system and compared with annual mean station DO.

Finfish Community Sampling - Surveys focused on twelve target species of finfish that fell within four broad life history groups: anadromous (American Shad, Alewife, Blueback Herring, Striped Bass), estuarine residents (semi-anadromous White Perch and Yellow Perch, and estuarine Bay Anchovy), marine migrants (Atlantic Menhaden and Spot), and tidal-fresh forage (Spottail Shiner, Silvery Minnow, and Gizzard Shad). Except for White Perch, adult sportfish of the target species were rare, 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 the Bay (directly or as forage); they are well represented in 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; Deegan et al. 1997). Gear specifications and techniques were selected to be compatible with past and present MD DNR Fishing and Boating Services' surveys (Carmichael et al. 1992; Bonzek et al. 2007; Durell and Weedon 2021).

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 cod-end, 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 offshore in the same direction as the tide in the same general area as the seine site. 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 then emptied into a tub for processing.

A 30.5 m × 1.2 m bag-less beach seine, constructed of untreated knotted 6.4 mm stretch mesh nylon, 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 from 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 tub for processing. The distance the net was stretched from shore, maximum depth of the seine haul, primary and secondary bottom types (i.e., gravel, sand, mud, and shell), and percent of seine area containing submerged aquatic vegetation were recorded. All fish captured were identified to species and counted. Striped Bass and Yellow Perch were separated into two age categories, juveniles (JUV; young of year = YOY) and adults (ages 1+). White Perch were separated into three age categories based on size and life stage, juveniles, small adults (ages 1+ fish measuring < 200 mm), and harvestable size adults (fish measuring > 200 mm). Harvestable size adult White Perch were measured, and the measurements were recorded for a modified proportional stock density analysis (PSD; Willis et al. 1993).

Three basic metrics of finfish community composition were estimated for tributaries and subestuaries sampled: geometric mean (GM) catch of all species, total number of species (species richness), and species comprising 90 % of the catch. The GM of seine and trawl catches were 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). In addition, we noted which target species were within the group that comprised 90% of fish collected, grouping the remaining 10% of species into the “other species” category. We summarized these metrics by salinity type since some important ecological attributes (DO and high or low SAV densities) appeared to reflect salinity class.

We plotted species richness in seine and trawl collections against C/ha by salinity class for all years in our database. A greater range of years (1989 – 2021) was available for beach seine samples than the 4.9 m bottom trawl (2003 – 2021) due to a change from the 3.1 m trawl used during 1989–2002 (Carmichael et al. 1992). Gear comparisons between the 3.1 m and 4.9 m trawls can be reviewed in Uphoff et al. (2016). We set a minimum number of samples (15 for seine and trawl) for a subestuary in a year to include estimates of species richness based on species accumulation versus sample size analyses in Uphoff et al. (2014). This eliminated years where sampling in a subestuary ended early due to site losses (typically from SAV growth) or high tides. We separated all subestuaries sampled by salinity class, then ranked their 2003 – 2021 bottom trawl GMs by year for all species combined to find where the 2021 subestuaries sampled ranked when compared to other subestuaries in their respective salinity classes.

A modified Proportional Stock Density (PSD; Anderson 1980; Anderson and Neumann 1996; Neumann and Allen 2007) was calculated using trawl catch data for White Perch in the tributaries and subestuaries of Talbot County and the subestuaries of Head-of-Bay for each year available to estimate an annual proportion of the adult population of interest to anglers. Low PSD percentages indicate higher densities of small fish (Anderson 1980; Neumann and Allen 2007). Proportional stock density is calculated using length-frequency data and provides population dynamics information (Anderson and Neumann 1996; Neumann and Allen 2007). Normally, a PSD is calculated as:

$$PSD = ((N \geq L_{Quality}) / (N \geq L_{Stock})) \times 100;$$

where N is the number of White Perch caught in each subestuary that were quality length or stock length or greater. Quality length ($L_{Quality}$) refers to the number of White Perch at the minimum length most anglers like to catch (≥ 200 mm TL; Piavis and Webb 2021). Stock length (L_{Stock}) refers to the number of White Perch at the minimum length of fish that provides a recreational value (≥ 125 mm TL; Piavis and Webb 2021). We substituted for stock length with the total number of small adults plus harvestable length White Perch to estimate a modified PSD since we did not measure small adults. White Perch greater than or equal to 130 mm TL is 20 – 26% of the world record length TL (Gablehouse et al 1984) is considered stock length category minimum; 125 mm TL is used as the length cut-off for White Perch in Chesapeake Bay recruitment and length-frequency assessments (Piavis and Webb 2021). Modified stock length category included small adults under 200 mm TL and could have fish as small as 90 mm TL. White Perch greater than or equal to 200 mm TL were measured to the nearest millimeter. White Perch greater than or equal to 200 mm TL corresponded to the quality length category minimum (36 – 41% of the world record TL) proposed by Gablehouse et al. (1984); 200 mm TL is used as the quality length category minimum length cut-off for White Perch in Chesapeake Bay (Piavis and Webb 2021). These data provided an opportunity to evaluate the influence of development on the availability of fish for anglers to harvest.

Fish Community Sampling in Mesohaline Tributaries and Subestuaries in Talbot County - In 2021, we examined correlations of 4.9 m bottom trawl geometric mean catches of all finfish or adult White Perch within the three Choptank tributaries. We estimated GMs of trawl and seine catches, modified PSD of White Perch, and species composition. We used a percent similarity index to evaluate variation in finfish species composition among the three Choptank tributaries trawl stations by year (Kwak and Peterson 2007). Finfish species abundances at a trawl station were standardized to percentages by dividing the abundance of each finfish species in a trawl station by the total number of fish collected at that trawl station, by year. The similarity among stations, P_{jklm} was calculated as:

$$\sum \text{minimum} \, (p_{ji}, \, p_{ki}, \, p_{li} \; p_{mi});$$

where p_{ji}, p_{ki}, p_{li}, and p_{mi} refers to the finfish species abundance of one particular finfish species *i* in trawl stations *j*, *k*, *l*, and *m*, by year, and the minimum indicates that the smallest of the four relative abundances was used in the summation (Kwak and Peterson 2007). The percent similarity index varies from 0% (no species in common) to 100 % (all species in common) and is considered a robust measure (Kwak and Peterson 2007).

In addition to our standard fish metrics, we also compared adult White Perch trawl GMs from Broad Creek, Harris Creek, and Tred Avon River using correlation analysis. White Perch adults were consistently abundant and represented the only adult gamefish that routinely appeared in samples.

Fish Community Sampling in Head-of-Bay Subestuaries - In 2021, we sampled the Sassafras River for a second summer to collect information on fish habitat status and conducted haul seines for the first time. Sassafras represented the only low salinity subestuary with a watershed dominated by agriculture. Sassafras River metrics were compared with previously sampled Head-of-Bay subestuaries: Bohemia River (2006), Bush River (2006 – 2010), Gunpowder River (2009 – 2016), Middle River (2009 – 2017), and Northeast River (2007 – 2017).

Annual GMs of total fish relative abundance and their 95 % CIs were estimated for 4.9 m trawl and beach seine. The top 90 % of finfish species occurring in annual trawl and seine catches were estimated for each subestuary time-series. Due to increased HABs in the upper Sassafras River, staff compiled seine data collected by the Juvenile Striped Bass Survey (Juvenile Index or JI) at the Sassafras River Natural Resource Management Area (NRMA monitoring station) in the Head-of-Bay for catch composition in 2020. The JI monitoring station was sampled monthly (July, August, and September), using replicate seine hauls, a minimum of thirty minutes apart, were taken at each site in each month. The NRMA station was located 1.61 km (1.0 miles) downriver of trawl site 04, where HABs were not as prolific. The NRMA seine GM was calculated only using the first seine haul (comparative to FHEP sampling methods) and had only three samples.

Results and Discussion

2021 Water Quality Summary – Table 3-3 provides summary statistics for surface and bottom water quality for each tributary and subestuary sampled in 2021. Both Sassafras and Tred Avon Rivers had bottom DO readings less than the target level (5.0 mg/L) during 2021: Sassafras River, 8%; and Tred Avon River, 46% (Table 3-4). Five percent of all DO measurements (surface, middle, and bottom) from Sassafras River were below the target; and

23% were below in Tred Avon River. In 2021, the Sassafras River did not have any bottom DO estimates below the 3 mg/L threshold. Seventeen percent of Tred Avon River bottom DO measurements were below the threshold (Table 3-4).

Salinity in the Tred Avon River was within mesohaline bounds in 2021 (Table 3-4). Sassafras River was classified as a tidal-fresh subestuary in 2021.

Dissolved Oxygen Dynamics – Analyses of DO with temperature and C/ha in subestuaries sampled since 2003 (Table 3-5) indicated that DO respond to temperature and C/ha differently depending on salinity classification (Table 3-6). Mean bottom DO in summer surveys declined below the threshold level in mesohaline subestuaries but did not in oligohaline or tidal-fresh (Figure 3-5). There were a few years in summer surveys where mean bottom DO fell below the target in oligohaline subestuaries but remained above 4.0 mg/L; these below target conditions would not affect occupation of this habitat (Uphoff et al. 2011). Mean surface DO in summer surveys did not fall below the threshold in tidal-fresh and oligohaline subestuaries, but two mesohaline subestuaries (Chester River, 2011 – 2012; Corsica River, 2012) fell below the target conditions (Figure 3-6).

A moderate negative association of surface dissolved oxygen (DO) and a strong negative association of bottom DO with corresponding mean water temperatures at depth were detected for oligohaline subestuaries by correlation analyses (Table 3-6), suggesting respiration was a factor in oligohaline subestuaries. Oligohaline subestuaries were shallower than most subestuaries of the other salinity categories, making them more likely to be warmer throughout. Associations of temperature and DO were weak in mesohaline and tidal-fresh subestuaries. A strong negative association between bottom DO and C/ha was found in mesohaline subestuaries; mesohaline subestuaries were where strongest stratification was expected. Oligohaline and tidal-fresh subestuaries were less likely to stratify strongly 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). Remaining correlations were weak. Given that multiple comparisons were made, correlations that had a significant *P* might be considered spurious if one rigorously adheres to significance testing (Nakagawa 2004; Anderson et al. 2000; Smith 2020). Sample sizes of mesohaline subestuaries (N = 88) were over twice as high as oligohaline (N = 33) or tidal-fresh subestuaries (N = 49), so ability to detect significant associations in mesohaline subestuaries was greater (Table 3-6).

Depletion of bottom DO to below target levels in mesohaline subestuaries with suburban-urban watersheds resulted in lost habitat. Uphoff et al. (2011) determined that the odds of adult and juvenile White Perch, juvenile Striped Bass, Spot, and Blue Crabs being present in shore zone seine samples from mesohaline subestuaries were not influenced by development, but odds of these target species being present in bottom channel trawl samples were negatively influenced by development through its negative influence on DO.

The extent of bottom channel habitat that can be occupied does not appear to diminish due to low DO with increasing watershed development in tidal-fresh and oligohaline subestuaries. However, more localized, or episodic habitat issues seem to be important. 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; 2016). Unfortunately, it was not feasible for us to routinely monitor fish within the beds and the impact on target finfish could not be estimated. Ammonia toxicity that was potentially associated with high SAV coverage was suspected as a cause of boom-and-bust dynamics of trawl GMs in Mattawoman Creek during the 2000s (Uphoff et al. 2016). During 2015, the oligohaline Middle River subestuary experienced an extensive fish kill attributed to HABs (MDE 2016). During 2020, Sassafras River was subject to HABs throughout summer sampling, but fish kills were not detected; no HABs were observed during 2021.

Land Use Categories, C/ha, and Mesohaline Subestuary Bottom Dissolved Oxygen - Correlation of agriculture with C/ha was negative and considered moderate, bordering on strong ($r = -0.76$; $P < 0.0001$); the correlation of urban land cover with C/ha was positive and considered strong (r = 0.89; $P < 0.0001$; Table 3-7). Correlation between forest cover with agriculture cover was negative and considered moderate (r = -0.57; $P < 0.0001$); urban cover with agriculture was negative and considered strong (r = -0.81; $P < 0.0001$). Wetland cover and C/ha were negative and considered weak (r = -0.26; $P = 0.02$). Remaining pairings of categories were not well correlated (Table 3-7).

After inspection of scatter plots, agricultural cover was further divided into regional categories (east and west of Chesapeake Bay) reflecting lower percentages of forest cover on the eastern shore, for analyses with DO in mesohaline subestuaries (Figure 3-7). Two western shore sub-regions reflected agricultural coverage: subestuaries located on the western shore of Chesapeake Bay (Magothy, Rhode, Severn, and South Rivers) fluctuated between 2.6% to 34.1% agricultural coverage, while lower Potomac River watersheds (Breton Bay, St. Clements, and Wicomico Rivers) ranged from 31.6% to 38.6% agricultural coverage. Eastern shore watersheds were divided into two divisions: Choptank River - Miles River drainages (Broad and Harris Creeks, Miles River, and Tred Avon River) ranged from 42.6% to 53.7% agricultural coverage and mid-eastern shore watersheds (Chester, Corsica, Miles, Wye Rivers, and Langford Creek) ranged from 53.7% to 71.6% agricultural coverage.

Inspection of the scatter plot of percent of watershed in agriculture versus median bottom DO in mesohaline subestuaries indicated an ascending limb of median DO when agricultural coverage went from 2.6% to 40.9% comprised entirely of western shore subestuaries (Figure 3-7). Median DO measurements beyond this level of agricultural coverage (42.6% – 71.6% agriculture) were from eastern shore subestuaries and the DO trend appeared to be stable or declining. Development was predominant at low levels of agriculture (< 20% agricultural coverage). Agricultural coverage and C/ha were inversely correlated, so the positive trend of DO with agriculture when agricultural coverage was low was likely to reflect development's negative impact.

We split agricultural coverage and median bottom DO data into western and eastern regions and used a linear regression for each region to describe regional changes in annual median subestuary bottom DO with percent agriculture. The relationship was positive and considered strong for the western shore (slope = 0.13; SE = 0.02; $r^2 = 0.73$; $P < 0.0001$; N = 21; Table 3-8) and negative and weak for the eastern shore (slope = -0.03; SE = 0.01; $r^2 = 0.15$; $P = 0.0024$; N = 60; Table 3-8). Predictions of median DO for mesohaline western shore subestuaries rose from 0.46 mg/L at 2.6% agricultural coverage to 5.24 mg/L at 38.6%. Predictions of median DO for mesohaline eastern shore subestuaries started at 5.40 mg/L at 42.6% agricultural coverage, increased to 5.51 mg/L at 50.1%, and then decreased to 4.39 mg/L at 71.6%. A quadratic regression of median bottom DO versus agricultural coverage described the relationship of median bottom DO with agricultural coverage well ($R^2 = 0.57$, $P < 0.001$; Table 3-9; Figure 3-7). Median bottom DO residuals were inspected and then plotted against agricultural coverage; residuals did not indicate substantial bias. However, residuals suggested that the predications at the highest coverage (≥ 65%) may have negatively biased. In addition, mesohaline subestuaries experiencing heavy rainfall from 2018 to 2020 did not create noticeable changes in the relationship.

Water Quality Summary in Mesohaline Tributaries and Subestuaries in Talbot County – Percentages of land in agriculture (43% – 49%), forest (20% – 27%), and urban (23% – 34%) categories were similar among the three Choptank tributaries (MD DOP 2020b; Table 3-10; Figure 3-1); however, wetlands varied among the three systems, comprising <1% of Broad Creek's watershed, 6% of Harris Creek's, and 1% of Tred Avon River watershed. Water comprised a larger fraction of the area in Broad and Harris Creeks (57% and 62%) than Tred Avon River (24%; i.e., water to watershed ratios were higher in the former; Table 3-10; MD DOP 2020b).

Tax map estimates of C/ha indicated that the Tred Avon River watershed was subjected to more development than Broad Creek and Harris Creek watersheds (Figure 3-8) and more than indicated by the Maryland Department of Planning urban category (Table 3-10). Time-series for all watersheds started at a rural level of development (C/ha ranged from 0.05 to 0.2) in 1950. Harris Creek watershed passed the rural development target (C/ha = 0.38) in 2009, while Broad Creek (C/ha = 0.30) is still under the rural development target. Faster growth occurred in Tred Avon River's watershed and the rural development target was passed in 1982, reaching 0.78 C/ha in 2020 (Figure 3-8). Development accelerated noticeably in the Tred Avon River watershed during 1996-2011 and then slowed. Tred Avon River's watershed has been approaching the suburban threshold (C/ha = 0.86).

During 2021, 46% of Tred Avon River bottom DO samples were below the target and 17% were below the threshold (Table 3-11; Figure 3-9). During 2006–2021, 9% of bottom DO measurements from Tred Avon River were below the DO threshold and 39% were below the DO target. Less than 1% of Broad Creek bottom DO measurements during 2012–2017 and 2020 were below the threshold and 15% were below the target. Harris Creek did not have any bottom DO measurements that fell below the threshold, and 3% were below the target during 2012–2016 (Table 3-11; Figure 3-9).

There was more variation in annual summer median DO in Tred Avon River (4.5 mg/L – 6.3 mg/L; Figure 3-10) than in Broad Creek (5.6 mg/L – 6.6 mg/L) and Harris Creek (5.7 mg/L – 6.4 mg/L; Figure 3-10). Correlations of median bottom DO between Tred Avon River and Broad Creek, or Harris Creek were modest to low, while Broad and Harris Creek's correlations were very low (Table 3-12). Years available for correlation analysis were low (5–7 years) and this pattern does not seem meaningful.

An ANOVA of Tred Avon River stations and bottom DO during 2006–2021 indicated significant differences among stations (F = 64.88; DF = 3; $P < 0.0001$; N = 382). Tukey Studentized Range and Tukey Honestly Significant Difference (HSD) tests indicated that bottom DO at station 01 (station at Easton, Maryland) was significantly lower than downstream stations 02, 03, and 04 (Figure 3-3). This decline in bottom DO with upstream distance was consistent with other mesohaline tributaries with high impervious surface (Uphoff et al. 2011). The mean and SE for bottom DO at all stations in Tred Avon River for all years were 5.14 mg/L and 0.08, respectively. Mean and SE for bottom DO at station 01 were 3.64 mg/L and 0.17; station 02 was 5.55 mg/L and 0.11; station 03 was 5.68 mg/L and 0.10; and station 04 was 5.69 mg/L and 0.10. Mean bottom DO at station 01 was the lowest of the time-series in 2020 (2.13 mg/L), closely followed by 2013 (2.23 mg/L; Figure 3-11). Deterioration of DO at the uppermost station (station 01; Figure 3-11) since 2012 indicated that stormwater from increased watershed development around Easton was the source of poor water quality rather than runoff from the whole watershed or water intruding from downstream. Station 2 mean bottom DO has declined since 2018 and is at the lowest of the time-series in 2021 (4.8 mg/L; Figure 3-11).

An ANOVA of Broad and Harris Creeks station bottom DO measurements did not indicate significant differences among stations in either of the subestuaries during sampling years. Annual station means in subestuaries varied without trend around the time-series median for all sites (Figure 3-11). The mean and SE for bottom DO at all stations for all years were 5.99 mg/L and 0.08 in Broad Creek and Harris Creek, 6.21 mg/L and 0.07, respectively.

Tred Avon River, the subestuary with the most developed watershed exhibited low DO at Easton and had a low percentage of water hectares per area of water and land (24%, respectively). Broad and Harris Creeks had higher percentages, 57% and 62%, respectively. A low percentage may limit intrusion of "good" mainstem water into a subestuary and increase the importance of internal nutrient loading and processing.

Median Secchi depths fluctuated slightly from year to year, while yearly ranges of Secchi depths revealed larger fluctuations within each system (Figure 3-12). Upper ranges were generally higher in Harris and Broad Creeks than in Tred Avon River. Tred Avon River median Secchi depths ranged from 0.4 m to 0.75 m during 2006 – 2021; from 0.5 m to 0.9 m in Broad Creek during 2012 – 2017, 2020; and from 0.5 m to 1.1 m in Harris Creek during 2012 – 2016 (Figure 3-12). The three Choptank River tributaries Secchi depths were strongly correlated with each other (Table 3-13).

Tred Avon River, Broad Creek, and Harris Creek SAV coverage were combined in the VIMS (2021) mouth of the Choptank River region estimates. Coverage of SAV increased substantially from 1% in 2012 to 11.8% in 2017 and declined to 6.1% in 2020 (Figure 3-13); since mapping started, the least SAV coverage was recorded in 1991 at 0.3%. The percentage of SAV coverage has remained above the time-series median of 4.9% since 2014 and displayed a similar trend present in the 1990s (Figure 3-13). The 2018 survey was only partially mapped. An SAV estimate for 2021 was not available at the time of this report.

Median pH in Tred Avon River from 2006 to 2021 ranged from 7.5 (2007) to 8.1 (2019; Figure 3-14). Broad Creek median pH during 2012 – 2017 and 2020 ranged from 7.8 (2014) to 8.1 (2015). Harris Creek median pH during 2012-2016 ranged from 7.7 (2013, 2014) to 7.9 (2012; Figure 3-14). Median pH estimates in Broad Creek and Harris Creek were strongly correlated, but remaining combinations were not (Table 3-14).

All salinity measurements remained in the mesohaline classification for the Choptank River tributaries; salinity ranged varied ever so slightly in Tred Avon River in 2021 (Figure 3-15). Overall, salinity range in 2021 for Tred Avon River appeared normal compared to previous sampled years. Highest salinities for Tred Avon River and the other Choptank subestuaries were observed in 2016, 12.8‰ in Tred Avon River and 13.6 ‰ in both Broad and Harris Creeks (Figure 3-15). Lowest salinity measurements differed by year in each subestuary, 2011 in Tred Avon River (7.5‰), 2013 in Broad Creek (10.2‰), and 2014 in Harris Creek (10.0‰; Figure 3-15). Median salinities of all three Choptank tributaries were positively and strongly correlated among each other; these strong correlations among these tributaries reflected their proximity to one another and minimal input of freshwater from their small drainages (Table 3-15). Individual observations of bottom salinity and bottom DO at trawl stations in Tred Avon River indicated there was a moderate positive association between the two parameters which supported watershed runoff as an influence on DO ($r = 0.365$; $P < 0.0001$). Broad and Harris Creeks exhibited a positive, weak association at each trawl station between bottom salinity and bottom DO suggesting minimal influence of the former on the latter (Broad Creek: $r = 0.118$; $P = 0.136$; Harris Creek: $r = 0.141$; $P = 0.131$).

In 2021, there was little indication that low DO was more widespread than usual, nor did the other water quality measurements offer an obvious connection to changes in finfish abundance. Tred Avon River’s station 01 mean bottom DO, which has declined since 2014, slightly increased in 2021 but remained below the threshold; mean bottom DO for stations 02 and 03 remained under the time-series median, while station 04 increased above the time-series median in 2021. A decline in bottom DO at station 02 has been observed over the last five years. Bottom DO declines at station 02 may represent the downstream progression of declining water quality caused by the increased development, like the decline observed at station 01.

Water Quality Summary in Head-of-Bay Subestuaries – Sassafras River was added to our sampling during 2020 as the only non-mesohaline subestuary with an agriculturally dominated watershed. We obtained a second year of water quality data from the Sassafras River in 2021. Sassafras River was contrasted with other non-mesohaline subestuaries sampled in the Head-of-Bay region. Estimated percentages of watershed in agriculture (3% – 68%), forest (23% – 39%), urban (8% – 71%), and wetlands (0.1% – 3%) varied throughout the Head-of-Bay subestuaries (MD DOP 2020b; Table 3-16; Figure 3-1). Water comprised a larger fraction of the Middle River drainage (28%) than in the Sassafras River (15%), Bohemia River (11%), Bush and Northeast Rivers (both 9%); Gunpowder River (5%; MD DOP 2020b) had the lowest fraction of water coverage (Table 3-16). Bohemia River was another subestuary with an agricultural watershed, but watershed effects on its fish community were difficult to detect because of the marine migrants that came from nearby Chesapeake & Delaware Canal (J. Uphoff, personal communication).

Estimates of C/ha indicated that the Middle River (2020: C/ha = 3.42) has been subject to greatest development in the Head-of-Bay (Figure 3-16), more than indicated by the Maryland Department of Planning urban category (Table 3-2). Bohemia (C/ha = 0.12) and Sassafras Rivers

(C/ha = 0.11) were below the rural development target (IS 5% = 0.38) in 2020. Time-series for Bohemia, Bush, Gunpowder, Northeast, and Sassafras Rivers started at a rural level of development (C/ha ranged from 0.03 to 0.09) in 1950. Middle River's level of development was already above the suburban level target (IS 10% = 0.86) in 1950 (C/ha = 0.97). Northeast River's C/ha progressed slowly, exceeding a rural level in 2003 but remaining below the suburban level target in 2020 (C/ha = 0.50). Gunpowder River progressed a little more quickly than Northeast River, exceeding a rural level in 1979 but remained below the suburban level target in 2020 (C/ha = 0.76). Bush River developed above the C/ha target in 1976, exceeded the threshold in 1991, and reached 1.37 (well-developed suburb) in 2005, continuing a steady incline in development in 2020 (C/ha = 1.57; Figure 3-16).

In 2021, bottom DO readings for the Sassafras River did not fall below the threshold (3.0 mg/L) but did fall below the target (5.0 mg/L) level (8%); 5% of all DO readings fell below the target level (Table 3-17). In addition, the Chesapeake Bay program Sassafras River monitoring station (CBP ET3.1), located near the MD route 213 bridge, did not record DO readings below threshold or target levels in 2021 (Figure 3-17). Since 1989, CBP ET3.1 has only recorded DO measurements below target level in 1991 (32%), 1992 (17%), and 1995 (25%; Figure 3-17); no threshold violations have been recorded. Other Head-of-Bay subestuaries sampled in previous years by FHEP all had target level breaches: 20% of Bohemia River readings; 2% of Bush River; 3% of Gunpowder River; 20% of Middle River; and 10% of Northeast River (Table 3-17). Only two subestuaries at the Head-of-Bay had bottom DO measurements that breached the threshold, Middle River (1%) and Northeast River (2%; Figure 3-18). The Chesapeake Bay Program Head-of-Bay monitoring station (CBP CB1.1), located at the mouth of the Susquehanna River, did not record DO readings below threshold or target levels since 1989 (Figure 3-17).

Sassafras River bottom DO measurements in 2021 ranged from 4.56 mg/L to 8.48 mg/L and median bottom DO was 6.45 mg/L (Figure 3-19). In 2021, Sassafras River bottom DO means were 5.60 mg/L at station 01, 5.88 mg/L at station 02, 6.62 mg/L at station 03, and 7.10 mg/L at station 04 (Figure 3-20). Bottom DO at stations 01 – 03 declined from 2020 to 2021 and were at or below the time-series median; station 04 was the only station to have an increase, although minor, in bottom DO (Figure 3-20). The CBP ET3.1 monitoring station bottom DO in 2021 ranged from 5.2 mg/L to 6.9 mg/L with a median bottom DO of 5.9 mg/L (Figure 3-17). The other Head-of-Bay subestuaries (Bohemia, Bush, Gunpowder, Middle, and Northeast Rivers) station annual mean bottom DO readings fluctuated above and below their time-series median (Figure 3-20). Middle River was an exception; stations 01 and 02 sometimes diverged from stations 03 and 04. Bottom DO could not be collected at Gunpowder River stations 02 and 03 during 2016 because stations were too shallow (depth was below 1.82 m).

The overall mean and SE for bottom DO in Bohemia River for 2006 was 6.41 mg/L and 0.31, respectively; 7.21 mg/L and 0.24 for Bush River (2006 – 2010); 6.75 mg/L and 0.10 for Gunpowder River (2009 – 2016); 6.10 mg/L and 0.10 for Middle (2009 – 2017); 6.93 mg/L and 0.11 for Northeast River (2007 – 2017); and 6.68 mg/L and 0.17 for Sassafras River in 2020 – 2021. The CBP CB1.1 monitoring station summer (July – September; n = 160) overall mean and SE was 7.16 mg/ L and 0.06 from 1989 to 2021, respectively. Bottom DO measurements for CBP CB1.1 in 2021 were 5.7 mg/L in July; 7.3 mg/L and 7.6 mg/L in August; and 8.9 mg/L in September. During 2021, median summer bottom DO at CBP CB1.1 was 7.45 mg/L (Figure 3-17). Correlation analyses of annual survey median bottom DO among Head-of-Bay subestuaries suggested weak associations among Gunpowder River, Bush River, Northeast River, and Middle River, when they were sampled in adjacent years, and very weak associations that were either positive or negative among the remaining subestuaries, respectively. Bohemia River was limited to only a single year of data and Sassafras River was not sampled in adjacent years, therefore, neither system could not be used in this analysis.

Differences in mean bottom DO among stations in the each of the Head-of-Bay subestuaries for years sampled were not detected with ANOVA in Bohemia, Bush, Gunpowder, Northeast, and Sassafras Rivers (Table 3-18). An ANOVA of Middle River stations and bottom

DO during 2009 – 2017 indicated significant differences among stations (F = 9.11; DF = 3; $P < 0.0001$; N = 201; Table 3-18). Tukey Studentized Range and Tukey Honestly Significant Difference (HSD) tests indicated that bottom DO at station 01 (located at the head of Dark Head Creek in front of Wilson Point Park landing) was significantly lower than downstream stations 03 and 04 (Figure 3-20). Station 02 (located at Clark Point where Middle River branch and Dark Head Creek converge) was significantly lower than downstream station 03 (Figure 3-20). Lower DO at the uppermost stations (station 01 and 02) indicated a negative influence of watershed development.

Median Secchi depths had variable annual ranges (Figure 3-21). Bohemia River median Secchi depths ranged from 0.2 m to 0.8 m during 2006; from 0.2 m to 0.6 m in Bush River during 2006–2010; from 0.2 m to 0.9 m in Gunpowder River during 2009–2016; from 0.0 m to 1.5 m in Middle River during 2009–2017; from 0.2 m to 1.0 m in Northeast River during 2007–2017; and from 0.3 m to 0.5 m in Sassafras River during 2020–2021 (Figure 3-21). Gunpowder and Middle Rivers showed the greatest variation in Secchi depths during 2014–2016. In 2015, MD DNR biologists discovered and confirmed zebra mussel presence in the Middle River.

All pH measurements in the tidal-fresh and oligohaline subestuaries investigated between 2006 and 2021 ranged from 6.3 (Middle, 2014) to 9.45 (Northeast, 2017), respectively. Bohemia River median pH for 2006 was 7.33 (Figure 3-22). Bush River median pH from 2006 to 2010 ranged from 6.95 (2007) to 7.90 (2009). Gunpowder River median pH from 2009 to 2016 ranged from 7.34 (2016) to 7.90 (2010). Middle River median pH from 2009 to 2017 ranged from 7.14 (2016) to 8.10 (2011). Northeast River median pH from 2007 to 2017 ranged from 7.80 (2008) to 8.70 (2010). Sassafras River median pH from 2020 to 2021 ranged from 7.68 (2021) to 8.26 (2020; Figure 3-22). The yearly ranges of pH within Bush, Gunpowder, Middle, Sassafras, and Northeast Rivers varied slightly to considerably; Bohemia River only had one year of pH measurements collected (Figure 3-22).

Salinity classifications in Head-of-Bay subestuaries were typically stable as either tidal-fresh or oligohaline; however, some fluctuated between tidal-fresh and oligohaline. Oligohaline subestuaries consisted of Bohemia River, which was sampled in 2006; Bush River, 2006–2010; Gunpowder River, 2009–2016; and Middle River, 2009–2017. The tidal-fresh subestuaries sampled were Northeast River from 2007 to 2017, and Sassafras River from 2020 to 2021. Salinity range varied the least in Northeast River and was greatest in Middle River for all years sampled (Figure 3-23). Highest salinity for the Head-of-Bay subestuaries differed by year in each subestuary, 3.5‰ in Bohemia River (2006), 3.6‰ in Bush River (2010), 6.74‰ in Gunpowder River (2016), 8.53‰ in Middle River (2016), 3.3‰ in Northeast River (2008), and 3.1‰ in Sassafras River (2020; Figure 3-23). Lowest salinity measurements differed by year in each subestuary, 2006 in Bohemia River (0.1‰), 2006 in Broad Creek (0.1‰), 2015 in Gunpowder (0.1‰), 2020 in Bush River (0.4‰), 2011 in Middle (0.5‰, 2009), 2017 in Northeast River (0.06‰), and 2021 in Sassafras River (0.21‰; Figure 3-23).

In the fall of 2015, a fish kill occurred in Middle River. The Maryland Department of the Environment reported that the fish kill was caused by high amounts of toxic algae, *Karlodinium veneficum*, whose toxin causes gill damage to fish when in high concentrations (MDE 2016; 2017). A toxic algae event occurred in the Sassafras in 2020, but a fish kill did not occur. A previous microcystin toxin event in the Sassafras River involving *Oscillatoria lemnosa* was noted in 2018; no fish kill was recorded (SR 2020).

2021 Finfish Community Summary - Geometric mean catch per seine haul ranged from 78 to 122 among the two subestuaries sampled during 2021 (Tred Avon River and Sassafras River, respectively; Table 3-19). Between 23 and 30 species were encountered in seine samples (Table 3-19). Sassafras River seine catch was substantially larger in 2021 due to only three seine samples (first seine hauls only) examined in 2020; in 2021, 19 seine samples were conducted in Sassafras River and 24 in Tred Avon River (Table 3-19).

A plot of species richness in seine samples against C/ha during 1989–2021 did not suggest a strong relationship in tidal-fresh, oligohaline, or mesohaline subestuaries (Figure 3-24).

Tidal-fresh subestuary watersheds were represented by a limited range of C/ha (0.43–0.69). Oligohaline subestuary watersheds were represented by the widest range of C/ha (0.08–3.33) of the three salinity classes. Mesohaline subestuary watersheds were represented by a larger number of surveys (N = 78; C/ha range = 0.07–2.68) than tidal-fresh and oligohaline subestuaries (N = 23 and 36, respectively; Figure 3-24).

A total of 6,300 fish representing 39 species were captured by beach seines in 2021 (Table 3-19). Ten species comprised 90% of the total fish caught in 2021, including (from greatest to least) Atlantic Menhaden, White Perch (adults), Atlantic Silverside, Mummichog, Bay Anchovy, Striped Killifish, White Perch (juveniles), Gizzard Shad, Spottail Shiner, Blueback Herring, and Striped Bass (juveniles). Atlantic Menhaden, Bay Anchovy, Blueback Herring, Gizzard Shad, Spottail Shiner, Striped Bass, and White Perch represented target species. Seven target species were present among species comprising 90% of the seine catch throughout all subestuaries: Atlantic Menhaden, Bay Anchovy, and White Perch were observed in both subestuaries; Gizzard Shad, Spottail Shiner, and Striped Bass in one subestuary (Table 3-19).

Geometric mean catches per trawl were between 11 and 18 during 2021 (Table 3-20). Both subestuaries had 24 samples (four stations) in 2021. Tred Avon River had the greatest GM (18), and Sassafras River had the lowest (11; Table 3-20). A plot of trawl GMs against C/ha (all subestuaries during 2003–2021) declined with development in mesohaline subestuaries and a possible negative threshold response at C/ha between 0.8 and 1.2 (Figure 3-25). Trawl GM catches did not exhibit an obvious decline with C/ha in tidal-fresh and oligohaline subestuaries (Figure 3-25).

Number of species captured by trawl in subestuaries sampled during 2021 ranged from 12 to 14 (Table 3-20). A plot of species richness in trawl samples against C/ha (all subestuaries during 2003–2021) did not indicate a relationship of development and number of species for tidal-fresh (species richness ranging from 14 to 25) or oligohaline subestuaries (species richness ranging from 12 to 26; Figure 3-26). Species richness (ranging from 3 to 23) declined in mesohaline subestuaries as C/ha advanced beyond the threshold (C/ha = 0.86 = 10% IS; Figure 3-26).

A total of 1,082 fish and 20 fish species were captured by bottom trawl during 2021 (Table 3-20). Five species comprised 90% of the total catch for 2021 (from greatest to least): Spot, White Perch (adult), White Perch (juvenile), Hogchoker, Bay Anchovy, and Blue Catfish; three of the five species were target species; Blue Catfish (only found in the Sassafras River) and Hogchoker were the exception. Target species comprising 90% of the catch in both subestuaries sampled during 2021 were White Perch (adult) and Spot (Table 3-20).

Subestuaries in 2021 had low GMs and species richness for their salinity classes and C/ha. Tred Avon River has a localized upper-subestuary DO issue that has worsened since 2014. GMs appeared to bottom out following a decline of Bay Anchovies. Sassafras River had a HAB in 2020 that caused water quality issues throughout most of the upper subestuary. In 2021, juvenile Blue Catfish were observed in the Sassafras River at stations 02–04, with station 04 having the highest quantity; the number of Blue Catfish observed in 2021 was triple the number of Channel Catfish when compared to 2020, when Channel Catfish numbers were substantially greater than Blue Catfish.

Finfish Community Summary in Mesohaline Tributaries in Talbot County – Mesohaline Tred Avon River, sampled in 2021, had the lowest bottom trawl GM ranking compared to Tred Avon River's previous sampling years and ranked, 78th out of 88 mesohaline subestuaries (Table 3-21). Annual GMs of catches of all species of finfish in 4.9 m bottom trawls in Broad Creek, Harris Creek, and Tred Avon River for all sampling years and their 95% CIs were plotted (Figure 3-27).

Correlations of trawl GMs among the three Choptank tributaries did suggest coherence in annual relative abundance of all finfish species combined (Table 3-22). Strong positive correlations of GMs were present between Broad Creek and Harris Creek (r = 0.95, P = 0.01, N = 5); Broad Creek and Tred Avon River (r = 0.96, P = 0.001, N = 7); a positive, moderate correlation was present between Tred Avon River and Harris Creek (r = 0.84, *P* = 0.07, N = 5; Table 3-22). Correlations of trawl GMs among the three Choptank tributaries using only target species suggest a slightly stronger positive correlations between Broad Creek and Harris Creek (r = 0.96, *P* = 0.01, N = 5), Broad Creek and Tred Avon River (r = 0.86, *P* = 0.014, N = 7), and Tred Avon River and Harris Creek (r = 0.79, *P* = 0.11, N = 5; Table 3-23). Correlations of beach seine GMs with bottom trawl GMs for Choptank tributaries were not significantly correlated in annual relative abundance of all finfish species, Broad Creek (r = -0.369, *P* = 0.415, N = 7); Harris Creek, (r = -0.493, *P* = 0.400, N = 5); and Tred Avon River, (r = 0.004, *P* = 0.987, N = 16), respectively. Correlation between beach seine GM with bottom trawl GM for Choptank tributaries among the three Choptank tributaries using only target species were weakly correlated in Broad Creek (r = 0.006, *P* = 0.989, N = 7), Harris Creek (r = 0.489, *P* = 0.404, N = 7), and Tred Avon River (r = 0.443, *P* = 0.086, N = 16).

Bay Anchovy was the most abundant species found throughout the Choptank River tributaries, making up greater than 50% of species present in systems when they were sampled during 2006 – 2021 (Figure 3-28). Two species in Broad Creek comprised the top 90% of finfish caught from 2012 to 2017 and in 2020, Bay Anchovy (87.8%) and Weakfish (2.8%); 27 other species made up the remaining 10% of species observed. Harris Creek had three species in the top 90% during 2012 – 2016: Bay Anchovy (85.8%), White Perch (adults and juveniles; 2.6%), and Weakfish (2.1%), with 25 other species making up the remaining 10% of species observed. Five species were in the top 90% of finfish caught in the Tred Avon River from 2006 to 2021, Bay Anchovy (56.8%), Spot (17.0%), White Perch (adults and juveniles; 7.7%), Hogchoker (7.3%), and Striped Bass (adults and juveniles; 3.5%; Figure 3-28); all except Hogchoker were target species. An additional 32 species comprised the last 7.6% (Figure 3-28). In these comparisons of samples with years combined, the number of other species appeared to be a negative function of how many years were sampled.

Species comprising the top 90% collected in Tred Avon River trawl samples were similar during 2019–2021 (4 species); 2011 and 2018 had the highest species richness (6 species; Figure 3-29). The usually common Bay Anchovy, last observed in the top 90% in 2019, reappeared and was among the top 90% during 2021. Summer Flounder appeared in the top 90% of species in 2021 (2.6%); they were not observed in the 90% of species since sampling started in 2006. Spot presence has increased in the top 90% of species since 2018, 12% (2018), 15% (2019), 37% (2020), and 45% (2021). In Broad Creek, five species were present in the top 90% compared to one to three species in 2012–2017. Tred Avon River and Broad Creek had an increase in the species comprising the top 90% starting in 2018, but this appeared to reflect reduced prevalence of Bay Anchovy (Figure 3-29). Bay anchovies transfer energy from zooplankton to higher levels of the food web and are a major prey for smaller piscivorous fishes in Chesapeake Bay, (Hartman and Brandt 1995; Christensen et al. 2009; Overton et al. 2015) and depletion could have ramifications for production of Striped Bass, Weakfish, and Bluefish in these subestuaries.

Percent similarity in trawl sample finfish species composition among stations 01–04 in the Tred Avon River was at its lowest in 2019 (7%) but increased in 2020 (to 16%) and again in 2021 (to 41%); percent similarity was above 50% during 2007–2017 (Figure 3-30). During 2006 and 2018–2021, the similarity index was below 50%, reflecting possible impacts of heavy rainfall during 2018–2019 and subsequent low salinity on fish community composition (Figure 3-30). Percent similarity in Broad Creek, sampled in 2020, fell but did not show the same drastic drop that appeared in the Tred Avon River; Broad Creek remained above 50%. Harris Creek was above 40% but was not sampled during the period of large change for Tred Avon River (Figure 3-30). Previous analyses in 2018 (Uphoff et al. 2018), suggested wet years with lower salinity had species composition dissimilar to dry years with higher salinity. Prevalent species in bottom trawl samples shifted during 2003–2021 (Figure 3-31). White Perch, Spot, and Bay Anchovy were predominant during 2003–2010; the latter two species predominated in 2012; Bay Anchovy predominated during 2013–2017; White Perch during 2018–2019; Bay Anchovy and Spot during

2020; and Hogchoker and Spot during 2021. Low salinity in 2011 was not accompanied by loss of Bay Anchovy in all mesohaline tributaries as it was during 2018–2021 (Figure 3-31).

Tred Avon River adult White Perch trawl GMs in 2009–2011, 2014–2016, and 2021 were at or fell below the median time-series GM (6; Figure 3-32). The greatest White Perch GM in Tred Avon River was in 2012 (16) and the lowest was in 2010 (2). Tred Avon River White Perch trawl GMs have declined since 2019. During 2016, adult White Perch GMs in Broad and Harris Creeks and Tred Avon River were similar (5; Figure 3-32). In 2020, White Perch GMs in Broad Creek (9) and Tred Avon River (12; Figure 3-32) were greater than the time-series median; Broad Creek recorded its highest White Perch GM in 2020. Correlations of White Perch GMs among Choptank tributaries were weakly positive (Table 3-24).

Modified PSDs for White Perch in Choptank tributaries (Broad Creek, Harris Creek, and Tred Avon River) for 4.9 m trawl samples varied among subestuaries and years but were generally lower in Tred Avon River (Table 3-25; Figure 3-33). In 2020, modified PSD in Broad Creek (14.7%) was half of the previous modified PSD calculated in 2017 (31.3%). Tred Avon River demonstrated a similar decline in modified PSDs since 2018; modified PSD ranged from 4.1% (2012) to 25.8% (2018), and an increase after 2016 reflected the size progression of the strong 2011 year-class (juvenile index = 35.2, respectively; Durell and Weedon 2021) into harvestable size. The decline after 2018 may indicate recruitment of two top quartile year-classes (2014 and 2015 juvenile indices = 14.4 and 14.8, respectively) into the stock category. The 2011 year-class followed a stretch of lesser year-classes during the 2000s (Durell and Weedon 2021). The less developed Choptank River tributary, Harris Creek, had higher modified PSDs for trawl samples than Tred Avon River during corresponding sampling years (2012 – 2016). Most modified PSDs were greater than 10% in Broad and Harris Creeks; only about 40% of modified PSDs were calculated above 10% in Tred Avon River (Table 3-25).

Seine GMs (relative abundance of all species combined) for Choptank River tributaries, Broad and Harris Creeks indicated similar status for years in common; lower seine GMs were present throughout sampling years in Tred Avon River, except 2015 (Figure 3-34). Seine GMs in the three Choptank River tributaries were highest during 2015. Seine GMs for all finfish were lowest in Broad Creek in 2012 (106), Harris Creek in 2012 (130), and Tred Avon River in 2008 (77). Tred Avon River seine GMs had a sharp, one year peak in 2015 and have generally been in a similar range over the rest of the time-series (Figure 3-34).

Seven species comprised the top 90% of finfish in beach seines when all years were combined in Tred Avon River (Figure 3-35). Tred Avon River's (2006 – 2021) top species were Atlantic Silverside (36.1%), Atlantic Menhaden (19.2%), White Perch (14.8%), Striped Killifish (7.7%), Mummichog (7.7%), Bay Anchovy (3.7%), and Banded Killifish (2.7%); an additional 48 other species (8.2%) were collected in Tred Avon River. Only three species in the top 90% were target species, Atlantic Menhaden, Bay Anchovy, and White Perch. Broad Creek (2012 – 2017, 2020) also had 7 species in the top 90% of finfish collected, Atlantic Silverside (34.8%), Atlantic Menhaden (20.8%), Striped Killifish (10.9%), Mummichog (8.9%), Banded Killifish (8.7%), White Perch (4.7%), and Sheepshead Minnow (2.9%); an additional 32 other species (8.2%) were collected in Broad Creek. Harris Creek, not sampled in 2020, had only 6 species in the top 90% of finfish from 2012 to 2016, with an additional 32 other species collected (Figure 3-35).

In 2021, finfish trawl catches in Tred Avon River bottom channel fell to their lowest levels for all years sampled, while inshore seine catches were lower than previous years but not the lowest. Tred Avon River trawl catches were slightly lower than both 2018 and 2020, seven of the sixteen years sampled have had trawl GMs lower than the time-series median (96) with four of those years being the last four sampled, 2018 – 2021. Typically, low finfish catches in the bottom channel within mesohaline systems are associated with increased development and low DO measurements. A change in the species present and richness in bottom trawl catches in 2021 was notable for Tred Avon River (Figure 3-29); all mesohaline systems saw a noteworthy shift in species composition in bottom trawl catches from 2018 to 2021 as well (Figure 3-31). Bay

Anchovy returned to Tred Avon River in 2021 but are less abundant. Spot increased noticeably in 2021 and has continued to increase in abundance over the last four years. The changes in species composition could reflect changes in salinity increased development and DO violations, and conditions outside of Tred Avon River where other processes important to year-class strength occur.

Finfish Community Summary in Head-of-Bay Subestuaries – Geometric means of catches of all species sampled in Head-of-Bay subestuaries since 2003 varied considerably among years. Sassafras River had a GM of 23 in 2020 and ranked 33rd and in 2021 had a GM of 11 and ranked 34th out of 36 fresh-tidal subestuaries sampled with bottom trawls since 2003 (Table 3-21). Bohemia River, an oligohaline system, previously sample in 2006, ranked 31st out of 33 with a GM of 115. Bush River, an oligohaline system, sampled during 2006–2010, achieved its highest ranking in 2010 with a GM of 473 (3rd out of 33). Gunpowder River, an oligohaline system, previously sampled during 2009 – 2016, achieved its highest ranking in 2010 with a GM of 401 (5th out of 33). Middle River, an oligohaline system, previously sampled from 2009 to 2017, ranked 2nd out of 33 with a GM of 520 in 2011. Northeast River, a tidal-fresh system, sampled during 2007–2017, had its greatest GM (392) ranking 2nd out of 36 in 2010. Annual GMs of catches of all species of finfish in 4.9 m bottom trawls in Head-of-Bay subestuaries for all sampling years and their 95% CIs were plotted (Figure 3-36). Bush River GMs ranged from 153 (2006) to 474 (2010); Gunpowder River, ranged from 147 (2013) to 402 (2010); Middle River, ranged from 75 (2017) to 521 (2011); and Northeast River, ranged from 97 (2016) to 306 (2010). Sassafras River had the lowest trawl GMs among all the Head-of-Bay subestuaries (Figure 3-36). To some extent, these rankings depend on whether the varying time-series contain good year-classes of non-marine target species. The short time-series for Sassafras River did not contain strong year-classes of our target species. Longer time-series (5-11 years) had higher rankings.

Modified PSD data revealed White Perch primarily use Head-of-Bay subestuaries as nursery habitat. Modified PSDs fluctuated between 0% and 1.4% (Table 3-26). The earliest quality size White Perch appeared in the Head-of-Bay subestuaries was in 2011, prior years indicate that only stock size White Perch were caught while sampling. After 2011, quality size White Perch are regularly present in the Head-of-Bay subestuaries sampled although modified PSDs were extremely low.

Sassafras River bottom trawl catches for all sampling years, 2020–2021, were composed of White Perch (adults and juveniles; 72.4%), Spot (12.4%), Blue Catfish (4.7%), and other species (9 species; 10.5%; Figure 3-37). Blue Catfish replaced Channel Catfish that were in the top 90% of species caught in 2020 (Figure 3-38). White Perch was the dominant species for all years combined in all the Head-of-Bay subestuaries trawl samples and ranged from 56% to 83% of all finfish (Figure 3-37). Finfish species composition for 4.9m bottom trawl catch in Head-of-Bay subestuaries, by sampling year, indicated White Perch (juveniles and adults) were the predominant species throughout all years and subestuaries (Figure 3-38). Bay Anchovies were prevalent throughout the Head-of-Bay subestuaries, but they were minimal or absent in some years in some subestuaries. Bush River, Gunpowder River, and Middle River had greater species richness over sampling years (Figure 3-38).

Seine GM (first haul only to match our sampling) at the JI – NRMA station in the Sassafras River was 88 in 2020 and 59 in 2021 (Table 3-27). In 2021, the seine GM for FHEP stations was 78, slightly higher than the JI – NRMA seine GM and was substantially greater than the FHEP trawl GM (11); both FHEP trawl and JI – NRMA seine GMs indicated a decline between 2020 and 2021 (Table 3-27). Thirty species were observed in the FHEP seine hauls (19 samples) at the four stations in 2021 (Figure 3-39); twice as many species were observed in shallow water habitat compared to bottom water habitat (12; Tables 3-19 and 3-20). Eight species comprised the top 90% of species of finfish in Sassafras River FHEP seine samples (Figure 3-43). The top 90% of species were White Perch (juveniles and adults; 43.5%), Spottail Shiner (11.0%), Gizzard Shad (10.7%), Blueback Herring (6.8%), Pumpkinseed (5.2%), Inland Silverside (4.6%), Striped Bass (juveniles and adults; 4.2%), and Atlantic Menhaden (4.0%); an additional 22 other species (10.1%) were collected. Six of the eight species in the top 90% in the FHEP stations were target species; nontarget species were Pumpkinseed and Inland Silverside (Figure 3-39). Fish abundance did not appear impacted by DO since it was above threshold level throughout shallow and bottom water habitat, only 5% of all DO fell below target level (Table 3-4).

Nine species comprised the top 90% of finfish in Sassafras River JI – NRMA beach seines in 2021 (Figure 3-40). The top 90% of species were White Perch (juveniles and adults; 25.5%), Striped Bass (juveniles and adults; 13.3%), Bay Anchovy (12.2%), Spottail Shiner (11.2%), Atlantic Silverside (9.2%), Inland Silverside (7.1%), Gizzard Shad (7.1%), Pumpkinseed (3.5%), and American Shad (2.5%); an additional 7 other species (8.2%) were collected. Six of the nine species in the top 90% in JI – NRMA station were our target species; nontarget species were Atlantic Silverside, Inland Silverside, and Pumpkinseed (Figure 3-40).

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Table 3-1. Percent impervious cover (IS), structures per hectare (C/ha), watershed area (land hectares), area of tidal water (water hectares), and salinity class for the subestuaries sampled in 2021.

2021 Sampled Subestuaries						
Area	**Subestuary**	**IS**	**C/ha**	**Land Hectares**	**Water Hectares**	**Salinity Class**
Upper-Bay	Sassafras River	2.4	0.11	19,595	3,461	Tidal-Fresh
Mid-Bay	Tred Avon River	9.2	0.78	9,561	3,086	Mesohaline

Table 3-2. Estimates of structures per hectare (C/ha) and land use percentages from Maryland Department of Planning (2002 and 2010) for subestuaries sampled 2003 – 2021.

River	Year	C/ha	Agriculture	Wetland	Forest	Urban
Breton Bay	2003	0.27	23.8	0.8	56.1	18.7
Breton Bay	2004	0.28	23.8	0.8	56.1	18.7
Breton Bay	2005	0.30	23.8	0.8	56.1	18.7
Broad Creek	2012	0.29	42.6	0.4	25.4	31.5
Broad Creek	2013	0.30	42.6	0.4	25.4	31.5
Broad Creek	2014	0.30	42.6	0.4	25.4	31.5
Broad Creek	2015	0.30	42.6	0.4	25.4	31.5
Broad Creek	2016	0.30	42.6	0.4	25.4	31.5
Broad Creek	2017	0.30	42.6	0.4	25.4	31.5
Broad Creek	2020	0.30	42.6	0.4	25.4	31.5
Bush River	2006	1.17	25.4	3.2	35.0	36.2
Bush River	2007	1.19	25.4	3.2	35.0	36.2
Bush River	2008	1.20	25.4	3.2	35.0	36.2
Bush River	2009	1.21	25.4	3.2	35.0	36.2
Bush River	2010	1.22	18.0	3.2	29.9	47.8
Chester River	2007	0.14	66.5	2.0	25.8	5.8
Chester River	2008	0.14	66.5	2.0	25.8	5.8
Chester River	2009	0.15	66.5	2.0	25.8	5.8
Chester River	2010	0.15	64.2	2.0	24.7	8.9
Chester River	2011	0.15	64.2	2.0	24.7	8.9
Chester River	2012	0.15	64.2	2.0	24.7	8.9
Chester River	2018	0.15	64.2	2.0	24.7	8.9
Chester River	2019	0.16	64.2	2.0	24.7	8.9
Corsica River	2003	0.17	64.3	0.4	27.4	7.9
Corsica River	2004	0.18	64.3	0.4	27.4	7.9
Corsica River	2005	0.19	64.3	0.4	27.4	7.9
Corsica River	2006	0.21	64.3	0.4	27.4	7.9
Corsica River	2007	0.22	64.3	0.4	27.4	7.9
Corsica River	2008	0.24	64.3	0.4	27.4	7.9
Corsica River	2010	0.24	60.4	0.1	25.5	13.2
Corsica River	2011	0.25	60.4	0.1	25.5	13.2
Corsica River	2012	0.25	60.4	0.1	25.5	13.2
Corsica River	2018	0.27	60.4	0.1	25.5	13.2
Corsica River	2019	0.28	60.4	0.1	25.5	13.2
Gunpowder River	2009	0.72	30.6	1.0	32.1	35.6
Gunpowder River	2010	0.72	30.6	1.0	32.1	35.6
Gunpowder River	2011	0.73	30.6	1.0	32.1	35.6
Gunpowder River	2012	0.73	30.6	1.0	32.1	35.6
Gunpowder River	2013	0.73	30.6	1.0	32.1	35.6
Gunpowder River	2014	0.73	30.6	1.0	32.1	35.6
Gunpowder River	2015	0.74	30.6	1.0	32.1	35.6
Gunpowder River	2016	0.74	30.6	1.0	32.1	35.6
Harris Creek	2012	0.39	44.9	5.6	19.7	29.8
Harris Creek	2013	0.39	44.9	5.6	19.7	29.8
Harris Creek	2014	0.39	44.9	5.6	19.7	29.8
Harris Creek	2015	0.39	44.9	5.6	19.7	29.8
Harris Creek	2016	0.39	44.9	5.6	19.7	29.8

Table 3-2 (Cont).

Langford Creek	2006	0.07	71.6	1.5	23.0	3.9
Langford Creek	2007	0.07	71.6	1.5	23.0	3.9
Langford Creek	2008	0.07	71.6	1.5	23.0	3.9
Langford Creek	2018	0.07	70.2	1.5	20.4	8.0
Langford Creek	2019	0.07	70.2	1.5	20.4	8.0
Magothy River	2003	2.68	2.6	0.0	27.8	69.5
Mattawoman Creek	2003	0.76	11.9	1.2	59.4	27.4
Mattawoman Creek	2004	0.79	11.9	1.2	59.4	27.4
Mattawoman Creek	2005	0.81	11.9	1.2	59.4	27.4
Mattawoman Creek	2006	0.83	11.9	1.2	59.4	27.4
Mattawoman Creek	2007	0.86	11.9	1.2	59.4	27.4
Mattawoman Creek	2008	0.87	11.9	1.2	59.4	27.4
Mattawoman Creek	2009	0.88	11.9	1.2	59.4	27.4
Mattawoman Creek	2010	0.90	9.3	2.8	53.9	34.2
Mattawoman Creek	2011	0.91	9.3	2.8	53.9	34.2
Mattawoman Creek	2012	0.90	9.3	2.8	53.9	34.2
Mattawoman Creek	2013	0.91	9.3	2.8	53.9	34.2
Mattawoman Creek	2014	0.93	9.3	2.8	53.9	34.2
Mattawoman Creek	2015	0.94	9.3	2.8	53.9	34.2
Mattawoman Creek	2016	0.95	9.3	2.8	53.9	34.2
Middle River	2009	3.30	4.5	2.2	27.9	63.9
Middle River	2010	3.32	3.4	2.1	23.3	71.0
Middle River	2011	3.33	3.4	2.1	23.3	71.0
Middle River	2012	3.33	3.4	2.1	23.3	71.0
Middle River	2013	3.34	3.4	2.1	23.3	71.0
Middle River	2014	3.35	3.4	2.1	23.3	71.0
Middle River	2015	3.36	3.4	2.1	23.3	71.0
Middle River	2016	3.38	3.4	2.1	23.3	71.0
Middle River	2017	3.38	3.4	2.1	23.3	71.0
Miles River	2003	0.24	53.7	0.9	27.2	18.1
Miles River	2004	0.24	53.7	0.9	27.2	18.1
Miles River	2005	0.24	53.7	0.9	27.2	18.1
Miles River	2020	0.26	49.0	0.8	26.7	23.1
Nanjemoy Creek	2003	0.08	15.1	4.1	73.1	7.6
Nanjemoy Creek	2008	0.09	15.1	4.1	73.1	7.6
Nanjemoy Creek	2009	0.09	15.1	4.1	73.1	7.6
Nanjemoy Creek	2010	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2011	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2012	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2013	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2014	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2015	0.09	12.4	4.1	68.7	14.7
Nanjemoy Creek	2016	0.09	12.4	4.1	68.7	14.7
Northeast River	2007	0.44	36.7	0.1	42.7	20.1
Northeast River	2008	0.44	36.7	0.1	42.7	20.1
Northeast River	2009	0.45	36.7	0.1	42.7	20.1
Northeast River	2010	0.46	31.1	0.1	38.6	28.9
Northeast River	2011	0.46	31.1	0.1	38.6	28.9

Table 3-2 (Cont.)

Northeast River	2012	0.47	31.1	0.1	38.6	28.9
Northeast River	2013	0.47	31.1	0.1	38.6	28.9
Northeast River	2014	0.48	31.1	0.1	38.6	28.9
Northeast River	2015	0.48	31.1	0.1	38.6	28.9
Northeast River	2016	0.49	31.1	0.1	38.6	28.9
Northeast River	2017	0.49	31.1	0.1	38.6	28.9
Piscataway Creek	2003	1.30	12.8	0.3	45.8	40.6
Piscataway Creek	2006	1.38	12.8	0.3	45.8	40.6
Piscataway Creek	2007	1.40	12.8	0.3	45.8	40.6
Piscataway Creek	2009	1.43	12.8	0.3	45.8	40.6
Piscataway Creek	2010	1.45	10.0	0.2	40.4	47.0
Piscataway Creek	2011	1.46	10.0	0.2	40.4	47.0
Piscataway Creek	2012	1.47	10.0	0.2	40.4	47.0
Piscataway Creek	2013	1.49	10.0	0.2	40.4	47.0
Piscataway Creek	2014	1.50	10.0	0.2	40.4	47.0
Rhode/West Rivers	2003	0.55	34.1	0.8	45.3	19.8
Rhode/West Rivers	2004	0.56	34.1	0.8	45.3	19.8
Rhode/West Rivers	2005	0.56	34.1	0.8	45.3	19.8
Sassafras River	2020	0.11	64.1	1.3	25.9	8.3
Sassafras River	2021	0.11	64.1	1.3	25.9	8.3
Severn River	2003	2.06	8.6	0.2	35.2	55.8
Severn River	2004	2.09	8.6	0.2	35.2	55.8
Severn River	2005	2.15	8.6	0.2	35.2	55.8
Severn River	2017	2.38	5.0	0.2	28.0	65.1
South River	2003	1.24	15.2	0.4	45.6	38.8
South River	2004	1.25	15.2	0.4	45.6	38.8
South River	2005	1.27	15.2	0.4	45.6	38.8
St. Clements River	2003	0.19	38.6	0.9	48.6	11.8
St. Clements River	2004	0.20	38.6	0.9	48.6	11.8
St. Clements River	2005	0.20	38.6	0.9	48.6	11.8
Tred Avon River	2006	0.69	50.1	1.0	21.6	27.2
Tred Avon River	2007	0.71	50.1	1.0	21.6	27.2
Tred Avon River	2008	0.73	50.1	1.0	21.6	27.2
Tred Avon River	2009	0.74	50.1	1.0	21.6	27.2
Tred Avon River	2010	0.75	43.2	0.8	21.6	33.6
Tred Avon River	2011	0.75	43.2	0.8	21.6	33.6
Tred Avon River	2012	0.75	43.2	0.8	21.6	33.6
Tred Avon River	2013	0.76	43.2	0.8	21.6	33.6
Tred Avon River	2014	0.76	43.2	0.8	21.6	33.6
Tred Avon River	2015	0.76	43.2	0.8	21.6	33.6
Tred Avon River	2016	0.77	43.2	0.8	21.6	33.6
Tred Avon River	2017	0.77	43.2	0.8	21.6	33.6
Tred Avon River	2018	0.77	43.2	0.8	21.6	33.6
Tred Avon River	2019	0.78	43.2	0.8	21.6	33.6
Tred Avon River	2020	0.78	43.2	0.8	21.6	33.6
Tred Avon River	2021	0.78	43.2	0.8	21.6	33.6
Wicomico River	2003	0.19	34.7	4.6	48.5	12.0
Wicomico River	2010	0.21	31.6	4.6	44.9	18.7
Wicomico River	2011	0.21	31.6	4.6	44.9	18.7
Wicomico River	2012	0.21	31.6	4.6	44.9	18.7
Wicomico River	2017	0.22	31.6	4.6	44.9	18.7
Wye River	2007	0.10	67.7	0.7	23.5	8.1
Wye River	2008	0.10	67.7	0.7	23.5	8.1
Wye River	2018	0.10	64.9	0.6	23.0	10.9
Wye River	2019	0.10	64.9	0.6	23.0	10.9

Table 3-3. Summary of water quality parameter statistics collected during both seine and trawl samples for subestuaries in 2021. Measurements for pH were calculated from H+ concentrations and converted back to pH.

		Surface Measurements					Bottom Measurements					
System	Statistics	Temp ©	DO (mg / L)	Cond (umhols)	Salinity	pH	Temp ©	DO (mg / L)	Cond (umhols)	Salinity	pH	Secchi
Sassafras River	Mean	27.65	7.83	789.51	0.38	7.80	27.27	6.30	787.29	0.38	7.59	0.45
	Standard Error	0.22	0.16	44.01	0.02	8.69	0.28	0.21	63.89	0.03	8.51	0.02
	Median	27.85	7.75	661.00	0.32	7.95	27.59	6.45	665.50	0.33	7.68	0.50
	Mode	29.52	7.62	1101.00	0.30	7.97	.	.	797.00	0.21	7.68	0.50
	Kurtosis	-1.09	1.28	0.96	1.16	-0.38	-1.36	-0.36	1.29	1.42	-0.28	-0.99
	Skewness	-0.05	-0.23	1.17	0.85	-0.21	-0.19	0.12	1.33	1.36	0.29	-0.22
	Minimum	25.32	4.99	444.00	0.03	8.82	25.38	4.56	440.00	0.21	8.33	0.25
	Maximum	30.78	10.50	1626.00	0.82	7.24	29.53	8.48	1627.00	0.82	7.26	0.60
	Count	43	43	43	43	43	24	24	24	24	24	24
Tred Avon River	Mean	28.72	6.61	16789.63	9.60	7.83	28.04	4.64	17284.67	10.14	7.63	0.46
	Standard Error	0.33	0.15	149.57	0.22	9.09	0.34	0.26	137.79	0.09	8.60	0.02
	Median	29.20	6.56	17005.00	9.90	7.87	28.39	5.11	17204.00	10.08	7.72	0.50
	Mode	26.36	7.45	.	9.90	7.96	29.18	.	.	9.74	7.74	0.50
	Kurtosis	8.82	0.16	0.15	30.68	-0.64	-1.22	0.87	0.04	0.09	-0.58	-0.87
	Skewness	1.88	-0.43	-0.97	-5.07	-0.24	-0.46	-1.22	0.07	0.20	-0.28	-0.07
	Minimum	25.22	4.18	14222.00	0.35	8.18	25.24	1.19	15786.00	9.17	7.96	0.30
	Maximum	39.44	8.69	18321.00	10.82	7.43	30.50	6.13	18641.00	11.03	7.20	0.60
	Count	48	48	48	48	48	24	24	24	24	24	24

Table 3-4. Percentages of all dissolved oxygen (DO) measurements and all bottom DO measurements that did not meet target (5.0 mg/L) or threshold (3.0 mg/L) conditions for each subestuary sampled in 2021. C/ha = structures per hectare. N = number of samples.

				All DO		Bottom DO	
Subestuary	**Salinity Class**	**C/ha**	**N**	**% < 5.0 mg/L**	**N**	**% < 5.0 mg/L**	**% < 3.0 mg/L**
Sassafras River	Tidal-Fresh	0.11	91	5%	24	8%	0%
Tred Avon River	Mesohaline	0.78	96	23%	24	46%	17%

Table 3-5. Subestuaries sampled during 2003 – 2021, by salinity class, with C/ha (watershed structures per hectare), mean annual surface and bottom temperatures, and mean annual surface and bottom dissolved oxygen (mg/L).

			Temperature (ºC)		Dissolved Oxygen (mg / L)	
River	Year	C / ha	Surface	Bottom	Surface	Bottom
Mesohaline						
Blackwater River	2006	0.04	28.14	27.98	5.27	4.12
Breton Bay	2003	0.27	26.40	25.69	8.10	3.75
	2004	0.28	27.01	25.95	7.36	3.73
	2005	0.30	28.62	27.51	6.98	3.99
Broad Creek	2012	0.29	27.50	26.60	8.30	5.97
	2013	0.30	27.30	26.49	7.26	5.76
	2014	0.30	27.62	26.64	7.65	5.78
	2015	0.30	28.05	27.05	7.93	6.63
	2016	0.30	29.16	28.33	7.30	6.16
	2017	0.30	27.01	26.29	7.50	6.11
	2020	0.30	27.94	27.57	7.55	5.57
Chester River	1995	0.11	26.70	26.32	7.58	6.28
	1996	0.11	23.86	23.63	7.62	6.71
	1997	0.11	27.02	26.32	7.66	6.37
	1998	0.11	28.00	27.49	7.40	6.26
	2007	0.14	25.59	24.18	5.38	4.53
	2008	0.14	25.09	25.35	5.24	4.20
	2009	0.15	25.79	25.77	5.74	5.21
	2010	0.15	26.12	24.97	5.84	5.71
	2011	0.15	25.31	25.41	4.90	4.28
	2012	0.15	27.12	27.12	4.67	4.39
	2018	0.15	27.54	26.90	6.83	6.00
	2019	0.16	27.45	27.05	6.75	5.77
Corsica River	2003	0.17	25.90	26.13	6.50	4.67
	2004	0.18	27.18	26.88	5.57	4.57
	2005	0.19	28.54	28.14	6.48	3.08
	2006	0.21	27.39	26.84	7.55	4.05
	2007	0.22	25.94	25.82	6.24	4.22
	2008	0.24	26.20	25.22	7.32	4.21
	2010	0.24	34.36	26.62	5.69	5.01
	2011	0.25	27.00	27.01	5.30	3.28
	2012	0.25	27.79	27.47	4.71	3.40
	2018	0.27	27.23	26.71	7.02	5.12
	2019	0.28	27.24	27.04	6.82	4.39
Fishing Bay	2006	0.03	26.23	25.28	7.24	6.79
Harris Creek	2012	0.39	26.55	26.42	7.44	6.35
	2013	0.39	26.39	26.05	7.02	6.01
	2014	0.39	27.61	26.68	6.84	4.84
	2015	0.39	26.62	26.62	7.19	6.56
	2016	0.39	27.82	27.75	6.65	6.02
Langford Creek	2006	0.07	27.05	26.52	6.95	5.68
	2007	0.07	26.23	25.48	6.69	5.68
	2008	0.07	27.47	26.65	6.85	5.05
	2018	0.07	27.08	31.78	6.40	5.10
	2019	0.07	27.77	27.51	6.69	5.07
Magothy River	2003	2.68	25.70	25.31	7.30	2.04
Miles River	2003	0.24	25.50	25.60	6.50	4.09
	2004	0.24	25.75	25.64	6.08	5.47
	2005	0.24	28.03	27.44	5.96	3.31
	2020	0.26	27.88	26.90	6.50	3.42

Table 3-5 (Cont.)

Rhode River	2003	0.47	25.00	24.69	7.10	4.80
	2004	0.47	27.00	26.95	6.58	5.39
	2005	0.48	27.78	27.16	6.50	4.03
Severn River	2003	2.06	26.30	24.75	7.60	1.57
	2004	2.09	27.42	26.18	7.05	2.64
	2005	2.15	28.01	26.23	7.07	0.96
	2017	2.38	26.93	26.07	6.86	1.78
South River	2003	1.24	25.40	24.56	7.60	2.61
	2004	1.25	25.79	25.48	6.46	3.77
	2005	1.27	27.57	26.67	6.02	2.49
St. Clements River	2003	0.19	26.00	25.29	8.20	3.48
	2004	0.20	26.08	25.78	6.84	4.61
	2005	0.20	27.12	26.36	6.85	4.42
Transquaking River	2006	0.03	26.68	22.75	5.75	5.85
Tred Avon River	2006	0.69	27.12	26.72	6.18	5.34
	2007	0.71	26.85	26.59	6.49	5.39
	2008	0.73	26.28	25.61	6.90	4.83
	2009	0.74	26.15	26.03	7.37	6.31
	2010	0.75	27.47	26.93	7.08	5.26
	2011	0.75	28.48	28.18	6.82	5.11
	2012	0.75	27.27	27.16	7.02	5.47
	2013	0.76	26.79	26.39	7.15	5.00
	2014	0.76	26.66	26.51	6.12	5.90
	2015	0.76	28.00	27.60	6.92	5.54
	2016	0.77	28.89	28.44	7.27	5.15
	2017	0.77	26.49	26.13	7.01	5.04
	2018	0.77	27.79	27.34	7.34	4.81
	2019	0.78	28.62	28.22	6.79	4.49
	2020	0.78	28.29	28.11	6.91	4.35
	2021	0.78	28.72	28.04	6.61	4.64
West River	2003	0.64	24.90	24.31	7.40	4.84
	2004	0.65	26.83	26.59	7.37	5.58
	2005	0.66	27.96	27.15	6.72	3.99
Wicomico River	2003	0.19	25.40	23.83	7.00	5.85
	2010	0.21	25.43	25.30	6.06	5.21
	2011	0.21	27.08	26.89	5.57	4.30
	2012	0.21	27.57	27.38	6.59	5.44
	2017	0.22	26.70	25.73	7.55	4.62
Wye River	2007	0.10	26.75	26.45	7.08	5.70
	2008	0.10	26.98	26.22	5.70	5.11
	2018	0.10	28.36	27.78	8.07	4.67
	2019	0.10	27.68	27.67	6.33	4.68
			Oligohaline			
Bohemia River	2006	0.11	26.79	26.02	7.01	6.41
Bush River	2006	1.17	25.48	24.28	7.96	7.47
	2007	1.19	27.02	26.42	7.68	6.54
	2008	1.20	26.59	24.20	9.00	5.43
	2009	1.21	25.88	24.34	9.41	8.54
	2010	1.22	27.72	23.80	7.79	7.04

Table 3-5 (Cont.)

Gunpowder River	2009	0.72	25.71	26.05	7.39	6.79
	2010	0.72	25.17	25.91	7.89	7.13
	2011	0.73	25.09	25.56	8.28	7.14
	2012	0.73	26.48	25.93	8.19	6.71
	2013	0.73	25.85	27.46	8.05	6.10
	2014	0.73	26.65	26.15	7.28	5.76
	2015	0.74	27.51	27.65	8.02	6.63
	2016	0.74	27.70	26.46	7.43	6.18
Middle River	2009	3.30	26.50	25.78	7.27	6.07
	2010	3.32	24.65	24.20	8.44	7.11
	2011	3.33	27.13	26.42	8.35	7.33
	2012	3.33	28.05	26.60	8.82	5.21
	2013	3.34	27.12	26.46	7.58	5.79
	2014	3.35	26.56	26.01	7.55	6.04
	2015	3.36	28.47	27.20	8.20	6.23
	2016	3.38	28.87	27.82	7.56	5.69
	2017	3.38	25.54	25.17	7.80	5.36
Nanjemoy Creek	2003	0.08	25.90	28.80	7.30	4.96
	2008	0.09	27.53	26.58	7.85	6.65
	2009	0.09	26.31	24.64	7.05	7.49
	2010	0.09	26.50	24.80	7.66	7.02
	2011	0.09	29.34	28.55	6.13	5.30
	2012	0.09	26.18	25.92	6.73	5.98
	2013	0.09	26.88	26.30	6.76	5.86
	2014	0.09	26.78	26.36	7.66	6.25
	2015	0.09	27.40	27.10	7.16	6.32
	2016	0.09	28.49	28.21	6.86	5.16
			Tidal Fresh			
Mattawoman Creek	2003	0.76	26.00	25.75	9.00	8.81
	2004	0.79	27.33	27.14	8.34	7.95
	2005	0.81	28.77	28.09	7.74	7.27
	2006	0.83	27.05	26.44	7.10	6.50
	2007	0.86	26.89	26.85	6.70	6.48
	2008	0.87	26.40	24.52	7.97	6.33
	2009	0.88	26.20	26.64	7.92	7.86
	2010	0.90	26.21	26.10	6.95	6.62
	2011	0.91	27.08	27.46	6.33	6.51
	2012	0.90	26.70	26.82	7.40	7.00
	2013	0.91	26.35	25.94	9.22	8.40
	2014	0.93	26.73	26.24	7.48	6.17
	2015	0.94	27.91	26.84	8.66	7.74
	2016	0.95	28.47	28.03	6.96	6.54
Northeast River	2007	0.44	26.83	26.43	9.73	7.75
	2008	0.44	25.35	24.98	8.43	7.70
	2009	0.45	26.33	25.55	9.35	7.36
	2010	0.46	25.90	26.21	7.76	6.78
	2011	0.46	25.97	25.71	6.87	5.79

Table 3-5 (Cont.)

	2012	0.47	27.78	27.59	7.88	6.03
	2013	0.47	26.61	26.11	9.33	7.06
	2014	0.48	26.94	26.52	7.72	6.81
	2015	0.48	26.66	26.23	7.84	6.17
	2016	0.49	27.95	26.86	8.81	7.10
	2017	0.49	26.38	25.68	9.38	7.80
Piscataway Creek	2003	1.30	25.60	24.63	10.20	8.33
	2006	1.38	28.16	24.97	8.70	6.85
	2007	1.40	27.47	26.00	8.57	7.60
	2009	1.43	26.72	27.07	8.56	6.62
	2010	1.45	27.07	25.08	9.36	7.63
	2011	1.46	28.25	30.07	9.05	9.47
	2012	1.47	27.92	25.51	9.53	9.34
	2013	1.49	27.19	26.22	9.87	7.65
	2014	1.50	26.98	26.28	8.66	7.33
Sassafras River	2020	0.11	28.14	27.27	9.83	7.06
	2021	0.11	27.65	27.27	7.83	6.30

Table 3-6. Pearson correlations (r) of mean survey surface and bottom dissolved oxygen (DO; mg/L) with water temperatures at depth (surface and bottom) and with watershed development (C/ha = structures per hectare) from subestuaries sampled during 2003 – 2021, by salinity class. Level of significance = *P*. N = sample size.

DO Depth	Statistics	Temperature	C / ha
		Mesohaline	
Surface	r	-0.005	0.234
	P	0.963	0.028
	N	88	88
Bottom	r	0.073	-0.587
	P	0.498	<.0001
	N	88	88
		Oligohaline	
Surface	r	-0.309	0.404
	P	0.080	0.020
	N	33	33
Bottom	r	-0.602	-0.083
	P	0.0002	0.644
	N	33	33
		Tidal Fresh	
Surface	r	0.041	-0.162
	P	0.779	0.265
	N	49	49
Bottom	r	0.060	-0.098
	P	0.684	0.501
	N	49	49

Table 3-7. Pearson correlations (r) of C/ha for mesohaline subestuaries sampled during 2003 – 2021 with Maryland Department of Planning (DOP) land use categories. Pearson correlations (r) between land use categories estimated by MD DOP for 2002 and 2010. *P* = level of significance. N = sample size. Duplicate entries of C/ha for mesohaline subestuaries from 2003 to 2021 were not included in analysis.

			Land Use Categories			
	Statistics	**C/ha**	**Agriculture**	**Forest**	**Wetland**	**Urban**
C/ha	r					
	P	1				
	N					
Agriculture	r	-0.76				
	P	<.0001	1			
	N	81				
Forest	r	0.07	-0.57			
	P	0.52	<.0001	1		
	N	81	81			
Wetland	r	-0.26	0.02	0.01		
	P	0.02	0.89	0.93	1	
	N	81	81	81		
Urban	r	0.89	-0.81	-0.12	-0.01	
	P	<.0001	<.0001	0.27	0.93	1
	N	81	81	81	81	

Table 3-8. Statistics and parameter estimates for regional (western and eastern shores) linear regressions of median bottom dissolved oxygen (DO; mg/L) versus percent agricultural coverage.

Linear Model	**Western Shore: Median Bottom DO = Agriculture (%)**					
ANOVA	df	SS	MS	F	Significance F	
Regression	1	52.61	52.61	53.84	<.0001	
Residual	20	19.54	0.98			
Total	21	72.15				
r^2 = 0.7292						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	0.50	0.48	1.04	0.31	-0.50	1.50
Agriculture (%)	0.13	0.02	7.34	<.0001	0.09	0.17

Linear Model	**Eastern Shore: Median Bottom DO = Agriculture (%)**					
ANOVA	df	SS	MS	F	Significance F	
Regression	1	6.87	6.87	10.10	0.0024	
Residual	59	40.14	0.68			
Total	60	47.01				
r^2 = 0.1462						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	6.87	0.56	12.29	<.0001	5.75	7.99
Agriculture (%)	-0.03	0.01	-3.18	0.0024	-0.05	-0.01

Table 3-9. Statistics and parameter estimates for a quadratic regression of median bottom dissolved oxygen (DO; mg/L) versus percent agricultural coverage (western and eastern shore combined).

Linear Model	**Median Bottom DO = Agriculture (%) Coverage**					
ANOVA	df	SS	MS	F	Significance F	
Regression	2	88.02	44.01	53.25	<.0001	
Residual	80	66.11	0.83			
Total	82	154.13				
r^2 = 0.5711						
	Estimate	SE	t Stat	P-value	Lower 95%	Upper 95%
Intercept	-0.05	0.47	-0.1	0.92	-0.99	0.90
Agriculture (%)	0.22	0.02	9.43	<.0001	0.18	0.27
Agriculture (%)^2	-0.002	0.0003	-8.13	<.0001	-0.003	-0.002

Table 3-10. Percent of watershed in major land use categories estimated by Maryland Department of Planning (DOP) for each of the Choptank River subestuaries. Land use estimates are determined from MD DOP 2010 data. The first four land use categories contain only land area (hectares) of the watershed; water area (hectares) is removed from each of these categories. Water is the percent of water hectares per area of water and land.

	Choptank River Subestuaries		
Land Use Category	**Broad Creek**	**Harris Creek**	**Tred Avon River**
Agriculture	43	45	43
Forest	25	20	22
Urban	31	30	34
Wetlands	<1	6	1
Water	57	62	24

Table 3-11. Percentages of all dissolved oxygen (DO; mg/L) measurements (surface, middle, and bottom) and all bottom DO measurements that did not meet target (5.0 mg/L) or threshold (3.0 mg/L) conditions during July – September for years sampled. N = sample size.

				All DO		Bottom DO	
Subestuary	**Year**	**C / ha**	**N**	**% < 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%
	2016	0.30	76	4%	22	9%	0%
	2017	0.30	72	3%	22	9%	0%
	2020	0.30	79	6%	24	21%	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%
	2016	0.39	79	3%	23	9%	0%
Tred Avon River	2006	0.69	91	19%	24	38%	0%
	2007	0.71	93	11%	23	26%	4%
	2008	0.73	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	0.76	103	15%	26	31%	15%
	2014	0.76	96	11%	24	21%	0%
	2015	0.76	96	8%	24	21%	13%
	2016	0.77	96	13%	24	38%	13%
	2017	0.77	89	17%	24	42%	13%
	2018	0.77	110	17%	28	50%	14%
	2019	0.78	96	30%	24	71%	17%
	2020	0.78	96	27%	24	63%	17%
	2021	0.78	96	23%	24	46%	17%

Table 3-12. Pearson correlations (r) of annual median bottom dissolved oxygen (DO; mg/L) for Broad Creek, Harris Creek, and Tred Avon River with year and among subestuaries. *P* = level of significance. N = number of annual median DO measurements for each subestuary sampled.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	-0.07		
	P	0.91	1	
	N	5		
Tred Avon River	r	0.57	0.73	
	P	0.18	0.16	1
	N	7	5	

Table 3-13. Pearson correlations (r) of annual survey median Secchi depths for Broad Creek, Harris Creek, and Tred Avon River among subestuaries. *P* = level of significance. N = number of annual survey median Secchi depths.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.972		
	P	0.006	1	
	N	5		
Tred Avon River	r	0.876	0.928	
	P	0.010	0.023	1
	N	7	5	

Table 3-14. Pearson correlations (r) of annual median pH for Broad Creek, Harris Creek, and Tred Avon River among subestuaries. *P* = level of significance. N = number of annual survey median pH estimates.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.937		
	P	0.019	1	
	N	5		
Tred Avon River	r	0.490	0.174	
	P	0.264	0.779	1
	N	7	5	

Table 3-15. Pearson correlations (r) of annual survey median salinity (ppt; ‰) for Broad Creek, Harris Creek, and Tred Avon River among subestuaries. *P* = level of significance. N = number of annual survey median salinity estimates.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.990		
	P	0.001	1	
	N	5		
Tred Avon River	r	0.995	0.979	
	P	<.0001	0.004	1
	N	7	5	

Table 3-16. Percent of watershed in major land use categories estimated by Maryland Department of Planning (DOP 2010) for each of Head-of-Bay subestuary. The first four land use categories contain only land area (hectares) of the watershed; water area (hectares) is removed from each of these categories. Water is the percent of water hectares per area of water and land.

	Head-of-Bay Subestuaries					
Land Use Category	**Bohemia River**	**Bush River**	**Gunpowder River**	**Middle River**	**Northeast River**	**Sassafras River**
Agriculture	68.0	18.0	30.6	3.4	31.1	64.1
Forest	22.8	29.9	32.1	23.3	38.6	25.9
Urban	7.7	47.8	35.6	71.0	28.9	8.3
Wetlands	1.5	3.2	1.0	2.1	0.1	1.3
Water	10.7	9.2	5.3	27.9	9.2	15.0

Table 3-17. Percent of all dissolved oxygen (DO; mg/L) measurements (surface, middle, and bottom) and all bottom DO measurements that did not meet target (5.0 mg/L) or threshold (3.0 mg/L) conditions during July – September, by year sampled, for Head-of-Bay subestuaries. N = number of DO measurements.

				All DO		Bottom DO	
Subestuary	Year	C / ha	N	% < 5.0 mg/L	N	% < 5.0 mg/L	% < 3.0 mg/L
Bohemia River	2006	0.11	72	17%	20	20%	0%
Bush River	2006	1.17	49	0%	11	0%	0%
Bush River	2007	1.19	48	2%	11	0%	0%
Bush River	2008	1.20	45	2%	3	33%	0%
Bush River	2009	1.21	50	2%	8	13%	0%
Bush River	2010	1.22	41	7%	5	0%	0%
Gunpowder River	2009	0.72	65	2%	18	0%	0%
Gunpowder River	2010	0.72	58	0%	13	0%	0%
Gunpowder River	2011	0.73	41	0%	9	0%	0%
Gunpowder River	2012	0.73	60	3%	15	7%	0%
Gunpowder River	2013	0.73	51	4%	12	17%	0%
Gunpowder River	2014	0.73	49	0%	15	0%	0%
Gunpowder River	2015	0.74	54	0%	13	0%	0%
Gunpowder River	2016	0.74	50	2%	8	0%	0%
Middle River	2009	3.30	70	7%	22	18%	5%
Middle River	2010	3.32	78	4%	22	14%	0%
Middle River	2011	3.33	60	2%	18	0%	0%
Middle River	2012	3.33	77	13%	24	42%	0%
Middle River	2013	3.34	71	7%	23	17%	0%
Middle River	2014	3.35	68	1%	23	4%	0%
Middle River	2015	3.36	67	7%	24	17%	0%
Middle River	2016	3.38	59	8%	22	23%	0%
Middle River	2017	3.38	72	11%	24	33%	8%
Northeast River	2007	0.44	86	3%	23	9%	0%
Northeast River	2008	0.44	74	7%	19	11%	0%
Northeast River	2009	0.45	78	1%	23	4%	0%
Northeast River	2010	0.46	71	1%	17	0%	0%
Northeast River	2011	0.46	88	13%	24	33%	13%
Northeast River	2012	0.47	82	7%	24	21%	0%
Northeast River	2013	0.47	85	2%	24	8%	0%
Northeast River	2014	0.48	80	1%	24	4%	0%
Northeast River	2015	0.48	85	5%	24	13%	4%
Northeast River	2016	0.49	84	0%	24	0%	0%
Northeast River	2017	0.49	93	1%	24	4%	0%
Sassafras River	2020	0.11	75	0%	24	0%	0%
Sassafras River	2021	0.11	91	5%	24	8%	0%

Table 3-18. Statistics and parameter estimates for Head-of-Bay subestuaries linear regressions of bottom dissolved oxygen (mg/L) versus stations.

Linear Model			Bohemia River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	0.14	0.14	0.07	0.80	
Residual	18	36.58	2.03			
Total	19	36.72				
r^2 = 0.0038						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	6.62	0.84	7.83	<.0001	4.84	8.39
Station	-0.08	0.29	-0.26	0.80	-0.68	0.53

Linear Model			Bush River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	0.02	0.02	0.01	0.93	
Residual	155	438.16	2.83			
Total	156	438.19				
r^2 = 0.0						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	6.36	0.36	17.64	<.0001	5.65	7.08
Station	0.01	0.17	0.09	0.93	-0.32	0.35

Linear Model			Gunpowder River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	1.06	1.06	0.98	0.33	
Residual	101	110.22	1.09			
Total	102	111.29				
r^2 = 0.0096						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	6.55	0.23	28.00	<.0001	6.08	7.01
Station	0.08	0.08	0.99	0.33	-0.08	0.24

Linear Model			Middle River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	34.73	34.73	16.98	<.0001	
Residual	200	409.15	2.05			
Total	201	443.88				
r^2 = 0.0782						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	5.19	0.24	21.32	<.0001	4.71	5.67
Station	0.37	0.09	4.12	<.0001	0.19	0.55

Linear Model			Northeast River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	10.86	10.86	3.53	0.06	
Residual	248	762.72	3.08			
Total	249	773.58				
r^2 = 0.014						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	6.46	0.27	23.81	<.0001	5.93	6.99
Station	0.19	0.10	1.88	0.06	-0.01	0.38

Linear Model			Sassafras River			
ANOVA	df	SS	MS	F	Significance F	
Regression	1	2.66	2.66	2.00	0.16	
Residual	46	61.00	1.33			
Total	47	63.66				
r^2 = 0.0417						
	Estimate	SE	t Stat	P value	Lower 95%	Upper 95%
Intercept	6.15	0.41	15.11	<.0001	5.33	6.97
Station	0.21	0.15	1.42	0.16	-0.09	0.51

Table 3-19. Beach seine catch summary, 2021. C/ha = structures per hectare. GM CPUE = geometric mean catches per seine sample. Sassafras River data acquired from Juvenile Index monitoring station Sassafras River Natural Resource Management Area (NRMA). Italics designate target species. Young of the year or juveniles = Juv.

River	Stations Sampled	Number of Samples	Number of Species	Comprising 90% of Catch	C / ha	Total Catch	GM CPUE
Sassafras River	4	19	30	*White Perch (Adults)* *White Perch (Juv)* *Spottail Shiner* *Gizzard Shad* *Blueback Herring* Pumpkinseed Inland Silverside *Atlantic Menhaden* *Striped Bass (Juv)* *Bay Anchovy*	0.11	1,766	78
Tred Avon River	4	24	23	*Atlantic Menhaden* Atlantic Silverside Mummichog *Bay Anchovy* Striped Killifish *White Perch (Adults)*	0.78	4,534	122
Grand Total	8	43	39	*Atlantic Menhaden* *White Perch (Adults)* Atlantic Silverside Mummichog *Bay Anchovy* Striped Killifish *White Perch (Juv)* *Gizzard Shad* *Spottail Shiner* *Blueback Herring* *Striped Bass (Juv)*		6,300	

Table 3-20. Bottom trawl catch summary, 2021. C/ha = structures per hectare. GM CPUE = geometric mean catches per trawl sample. Italics designate target species. Young-of-the-year or juveniles = Juv.

River	Stations Sampled	Number of Samples	Number of Species	Comprising 90% of Catch	C / ha	Total Catch	GM CPUE
Sassafras River	4	24	12	*White Perch (Juv)*	0.11	540	11
				White Perch (Adult)			
				Blue Catfish			
				Spot			
Tred Avon River	4	24	14	*Spot*	0.78	542	18
				Hogchoker			
				Bay Anchovy			
				White Perch (Adult)			
				Summer Flounder			
Grand Total	8	48	20	*Spot*		1,082	
				White Perch (Adult)			
				White Perch (Juv)			
				Hogchoker			
				Bay Anchovy			
				Blue Catfish			

Table 3-21. Subestuaries sampled during 2003 – 2021, grouped by salinity class and ranked by annual 4.9 m trawl catch geometric mean (GM) of all species combined.

River	Year	GM	Rank
Mesohaline			
Broad Creek	2014	401	1
Corsica River	2003	378	2
Miles River	2003	313	3
Broad Creek	2012	294	4
West River	2003	272	5
Langford Creek	2007	264	6
Tred Avon River	2010	264	7
Rhode River	2003	262	8
Chester River	2011	259	9
Langford Creek	2006	258	10
Corsica River	2004	251	11
Corsica River	2011	238	12
Corsica River	2009	215	13
Tred Avon River	2014	192	14
Harris Creek	2014	174	15
Corsica River	2006	174	16
Chester River	2010	172	17
Wye River	2007	170	18
Rhode River	2005	163	19
Corsica River	2012	162	20
Langford Creek	2008	161	21
Corsica River	2010	161	22
Tred Avon River	2008	155	23
Tred Avon River	2012	155	24
Harris Creek	2012	155	25
Chester River	2007	152	26
Broad Creek	2017	148	27
Broad Creek	2016	147	28
Broad Creek	2013	142	29
Tred Avon River	2007	137	30
Corsica River	2007	131	31
Fishing Bay River	2006	131	32
Transquaking River	2006	131	33
Chester River	2012	130	34
West River	2005	125	35
Tred Avon River	2016	121	36
Chester River	2008	120	37
Wicomico River	2010	120	38
Wye River	2008	114	39
Wicomico River	2012	110	40
Corsica River	2005	109	41
Tred Avon River	2009	104	42
Broad Creek	2015	103	43
Tred Avon River	2017	98	44
Tred Avon River	2011	92	45

Table 3-21 (Cont.)

Harris Creek	2013	89	46
Corsica River	2008	86	47
Miles River	2004	82	48
Wicomico River	2017	81	49
Tred Avon River	2015	80	50
Chester River	2019	78	51
Tred Avon River	2013	77	52
Chester River	2009	76	53
Tred Avon River	2006	76	54
Miles River	2005	72	55
Wicomico River	2011	65	56
Wicomico River	2003	59	57
South River	2003	55	58
St. Clements River	2005	54	59
Harris Creek	2016	51	60
Langford Creek	2019	42	61
Harris Creek	2015	40	62
Rhode River	2004	38	63
Tred Avon River	2019	37	64
South River	2005	35	65
Blackwater River	2006	35	66
Breton Bay	2005	34	67
West River	2004	34	68
Broad Creek	2020	34	69
Magothy River	2003	33	70
Corsica River	2019	32	71
St. Clements River	2003	31	72
Tred Avon River	2020	29	73
Langford Creek	2018	27	74
South River	2004	21	75
Tred Avon River	2018	20	76
Wye River	2019	19	77
Tred Avon River	2021	18	78
Breton Bay	2003	18	79
St. Clements River	2004	17	80
Breton Bay	2004	16	81
Severn River	2017	16	82
Corsica River	2018	16	83
Wye River	2018	12	84
Miles River	2020	9	85
Severn River	2004	5	86
Severn River	2003	5	87
Severn River	2005	3	88
Oligohaline			
Nanjemoy Creek	2013	576	1
Middle River	2011	520	2
Bush River	2010	473	3
Nanjemoy Creek	2015	416	4
Gunpowder River	2010	401	5
Nanjemoy Creek	2014	396	6

Table 3-21 (Cont.)

Gunpowder River	2011	394	7
Nanjemoy Creek	2011	385	8
Bush River	2007	324	9
Bush River	2009	319	10
Middle River	2010	315	11
Nanjemoy Creek	2010	309	12
Nanjemoy Creek	2016	297	13
Middle River	2009	292	14
Gunpowder River	2009	289	15
Middle River	2015	286	16
Nanjemoy Creek	2009	284	17
Middle River	2016	261	18
Middle River	2014	251	19
Nanjemoy Creek	2012	224	20
Gunpowder River	2012	224	21
Gunpowder River	2014	219	22
Gunpowder River	2015	218	23
Bush River	2008	210	24
Nanjemoy Creek	2008	209	25
Gunpowder River	2016	206	26
Middle River	2013	181	27
Bush River	2006	152	28
Middle River	2012	148	29
Gunpowder River	2013	147	30
Bohemia River	2006	115	31
Nanjemoy Creek	2003	93	32
Middle River	2017	74	33
Tidal-Fresh			
Mattawoman Creek	2014	580	1
Northeast River	2010	392	2
Piscataway Creek	2011	320	3
Northeast River	2014	291	4
Northeast River	2011	290	5
Piscataway Creek	2010	290	6
Mattawoman Creek	2013	283	7
Mattawoman Creek	2004	252	8
Piscataway Creek	2014	221	9
Mattawoman Creek	2015	217	10
Mattawoman Creek	2011	208	11
Northeast River	2009	198	12
Northeast River	2012	191	13
Mattawoman Creek	2005	187	14
Northeast River	2013	186	15
Piscataway Creek	2013	184	16
Northeast River	2008	152	17
Northeast River	2015	150	18
Northeast River	2007	149	19
Mattawoman Creek	2016	149	20

Table 3-21 (Cont.)

Mattawoman Creek	2003	144	21
Piscataway Creek	2012	119	22
Northeast River	2017	105	23
Piscataway Creek	2009	105	24
Northeast River	2016	96	25
Mattawoman Creek	2010	84	26
Mattawoman Creek	2006	75	27
Mattawoman Creek	2012	72	28
Mattawoman Creek	2007	56	29
Piscataway Creek	2003	42	30
Piscataway Creek	2006	28	31
Mattawoman Creek	2008	27	32
Sassafras River	2020	23	33
Sassafras River	2021	11	34
Piscataway Creek	2007	8	35
Mattawoman Creek	2009	6	36

Table 3-22. Pearson correlations (r) of annual 4.9m trawl finfish catch geometric mean (GM; all species combined) for Broad Creek, Harris Creek, and Tred Avon River, with year and among subestuaries. *P* = level of significance. N = number of annual GMs for each subestuary.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.95		
	P	0.01	1	
	N	5		
Tred Avon River	r	0.96	0.84	
	P	0.001	0.07	1
	N	7	5	

Table 3-23. Pearson correlations (r) of annual 4.9m trawl finfish catch geometric mean (GM; only target species) for Broad Creek, Harris Creek, and Tred Avon River, with year and among subestuaries. *P* = level of significance. N = number of annual GMs for each subestuary.

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.96		
	P	0.01	1	
	N	5		
Tred Avon River	r	0.86	0.79	
	P	0.014	0.11	1
	N	7	5	

Table 3-24. Pearson correlations (r) of annual 4.9m trawl catch of adult White Perch geometric mean (GM) for Choptank subestuaries, Broad Creek, Harris Creek, and Tred Avon River, with year and among each subestuary. Level of significance of Pearson correlation = P. Sample size (N) for the number of adult White Perch GM measurements for each subestuary sampled. Bold numbers indicate a significant association ($\alpha = 0.05$).

	Statistics	Broad Creek	Harris Creek	Tred Avon River
Broad Creek	r			
	P	1		
	N			
Harris Creek	r	0.020		
	P	0.975	1	
	N	5		
Tred Avon River	r	0.626	0.447	
	P	0.132	0.450	1
	N	7	5	

Table 3-25. Modified proportional stock density (PSD) of White Perch in Choptank River subestuaries are the proportion of 4.9m trawl and beach seine samples with quality length or greater White Perch. N_{TOTAL} is the total number of White Perch (all juveniles and adults) captured in trawl and seine catches. Number of L_{STOCK} is the number of all adult White Perch (adults age +1). Number of $L_{QUALITY}$ is the number of harvestable adults (≥ 200 mm).

Subestuary	Years	N_{TOTAL}	N L_{STOCK}	N $L_{QUALITY}$	Modified PSD
Broad Creek	2012	496	496	38	7.7%
	2013	170	149	22	14.8%
	2014	184	51	18	35.3%
	2015	750	173	14	8.1%
	2016	138	123	17	13.8%
	2017	16	16	5	31.3%
	2020	281	265	39	14.7%
Harris Creek	2012	285	285	52	18.2%
	2013	292	285	28	9.8%
	2014	115	72	20	27.8%
	2015	556	109	34	31.2%
	2016	169	169	58	34.3%
Tred Avon River	2006	1881	1719	117	6.8%
	2007	1350	1064	62	5.8%
	2008	794	748	46	6.1%
	2009	321	262	45	17.2%
	2010	243	219	12	5.5%
	2011	2308	212	29	13.7%
	2012	3025	2908	119	4.1%
	2013	650	618	32	5.2%
	2014	329	248	8	3.2%
	2015	1057	221	22	10.0%
	2016	857	802	55	6.9%
	2017	261	186	43	23.1%
	2018	534	233	60	25.8%
	2019	1567	1290	227	17.6%
	2020	789	784	100	12.8%
	2021	422	363	20	5.5%

Table 3-26. Modified proportional stock density (PSD) of White Perch in Head-of-Bay subestuaries, Bohemia, Bush, Gunpowder, Middle, Northeast, and Sassafras Rivers, are the proportion of 4.9m trawl and beach seine catches with quality length or greater White Perch. N_{TOTAL} is the total number of White Perch (all juveniles and adults) captured in trawl and seine catches. Number of L_{STOCK} is the number of all adult White Perch (adults age +1). Number of $L_{QUALITY}$ is the number of harvestable adults (≥ 200 mm).

Subestuary	Years	N_{TOTAL}	N L_{STOCK}	N $L_{QUALITY}$	Modified PSD
Bohemia River	2006	1725	745	0	0%
Bush River	2006	1543	1133	0	0%
	2007	5651	477	0	0%
	2008	3263	1929	0	0%
	2009	4695	1130	0	0%
	2010	7392	1905	0	0%
Gunpowder River	2009	6076	743	0	0%
	2010	3913	917	0	0%
	2011	6164	254	0	0%
	2012	4013	1776	6	0.3%
	2013	1645	789	5	0.6%
	2014	2946	182	0	0%
	2015	4859	290	0	0%
	2016	1254	890	7	0.8%
Middle River	2009	5851	1664	0	0%
	2010	4586	866	0	0%
	2011	11978	801	0	0%
	2012	2830	2267	2	0.1%
	2013	1679	1137	2	0.2%
	2014	3098	336	4	1.2%
	2015	7108	283	4	1.4%
	2016	1248	744	2	0.3%
	2017	1439	374	3	0.8%
Northeast River	2007	2961	1222	0	0%
	2008	2967	1468	0	0%
	2009	4681	2841	0	0%
	2010	7929	2104	0	0%
	2011	6692	1485	20	1.3%
	2012	6699	4962	63	1.3%
	2013	4781	3875	13	0.3%
	2014	6929	2254	5	0.2%
	2015	3828	2247	4	0.2%
	2016	2073	968	2	0.2%
	2017	3123	557	4	0.7%
Sassafras River	2020	873	722	4	0.6%
	2021	1146	683	1	0.1%

Table 3-27. Geometric mean (GM) and 95% confidence intervals of all finfish species for FHEP 4.9 m bottom trawl, FHEP beach seine, and for Juvenile Index (JI) – NRMA (haul 1) in the Sassafras River, by sampling year.

			95% Confidence Intervals	
Year	Sassafras River Sample	GM	Upper	Lower
2020	FHEP Trawl	25	45	14
	FHEP Seine	.	.	.
	JI Seine - NRMA (Haul 1)	88	166	47
2021	FHEP Trawl	11	21	8
	FHEP Seine	78	108	58
	JI Seine - NRMA (Haul 1)	59	272	13

*No FHEP seines were conducted in 2020 due to HABs present.

Figure 3-1. Map illustrating subestuaries sampled in summer 2021: Sassafras River (1), Tred Avon River (2), and their land use categories. Land use data is based on Maryland Department of Planning (DOP) 2010 land use land cover data. Figure includes previously sampled subestuaries referenced throughout this report (marked by white stars; Broad Creek, Harris Creek, Bohemia River, Northeast River, Gunpowder River, Middle River, and Bush River).

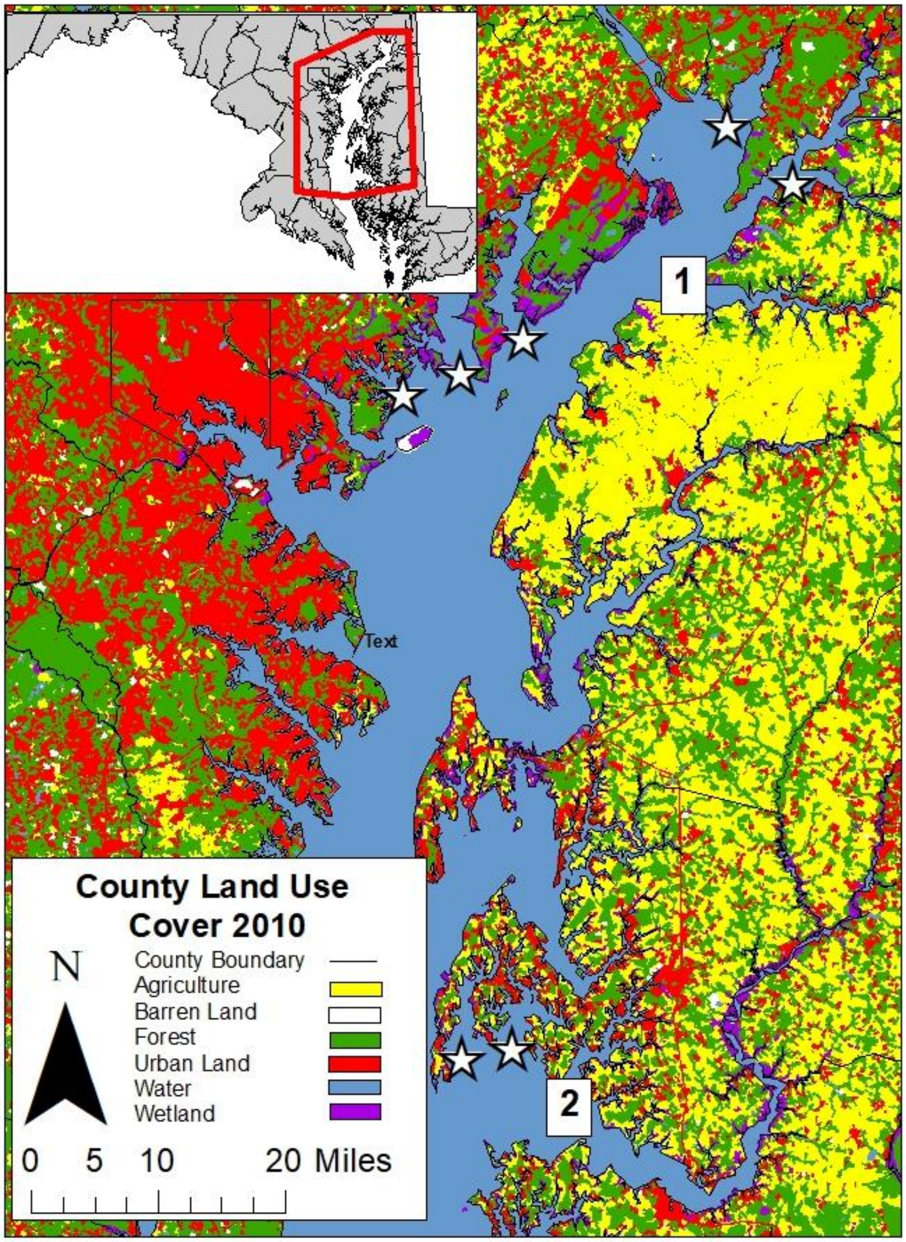
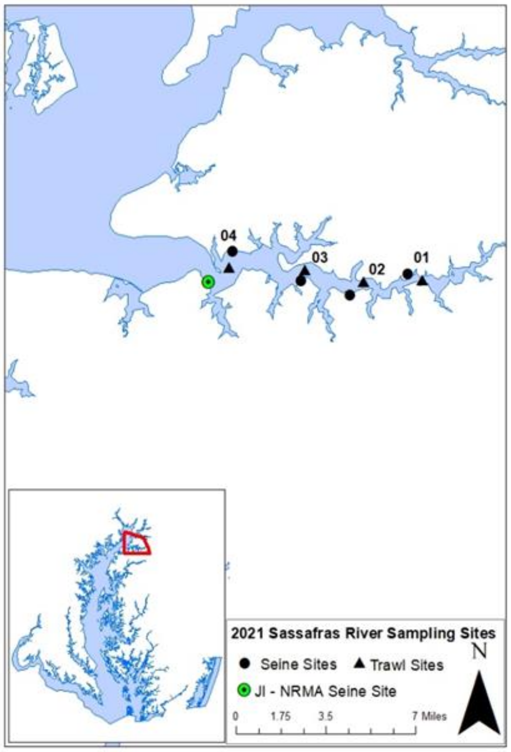


Figure 3-2. Map indicating 2021 locations of sampling sites for subestuaries, Sassafras River and Tred Avon River.



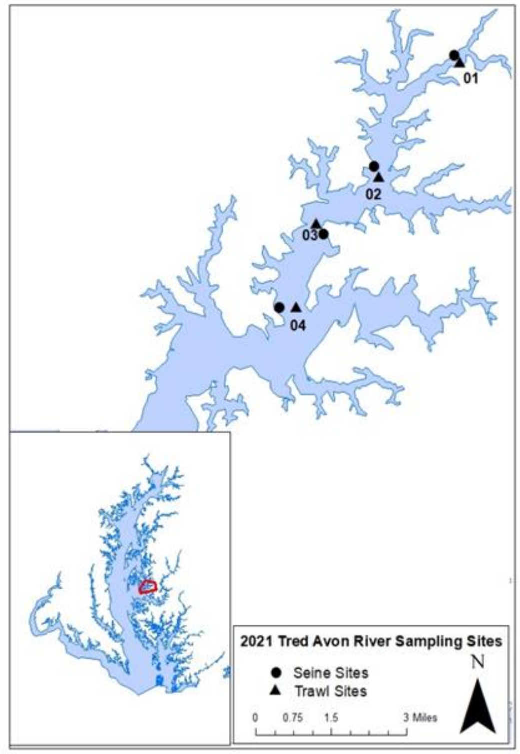


Figure 3-3. Map indicating the locations of seine and bottom trawl sites for the lower Choptank River tributaries, Broad Creek (2012 – 2017, 2020), Harris Creek (2012 – 2016), and Tred Avon River (2006 – 2020).

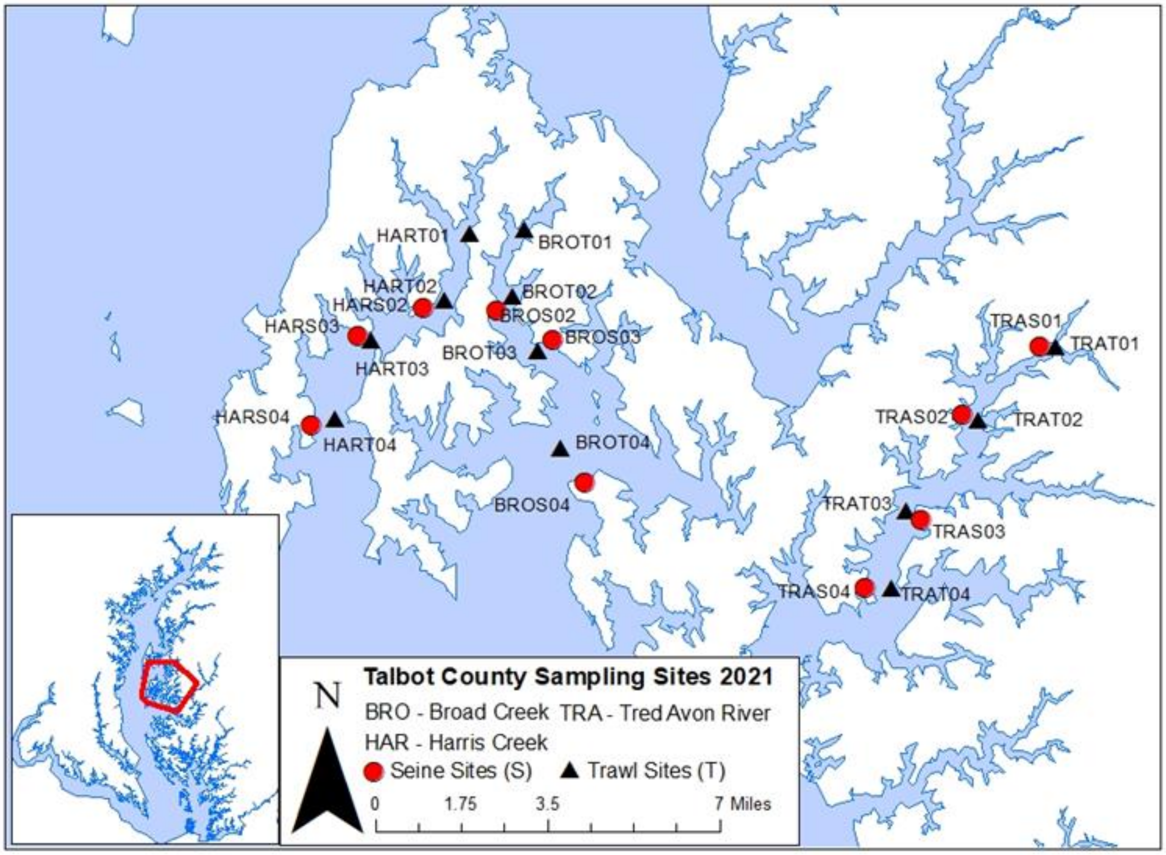


Figure 3-4. Map indicating the locations of seine and bottom trawl sites for Head-of-Bay subestuaries , Bohemia River (2006), Bush River (2006 – 2010), Gunpowder River (2009 – 2016), Middle River (2009 – 2017), Northeast River (2007 – 2017), and Sassafras River (2020 – 2021); including juvenile index (JI) seine site at Sassafras Natural Resource Management Area (NRMA).

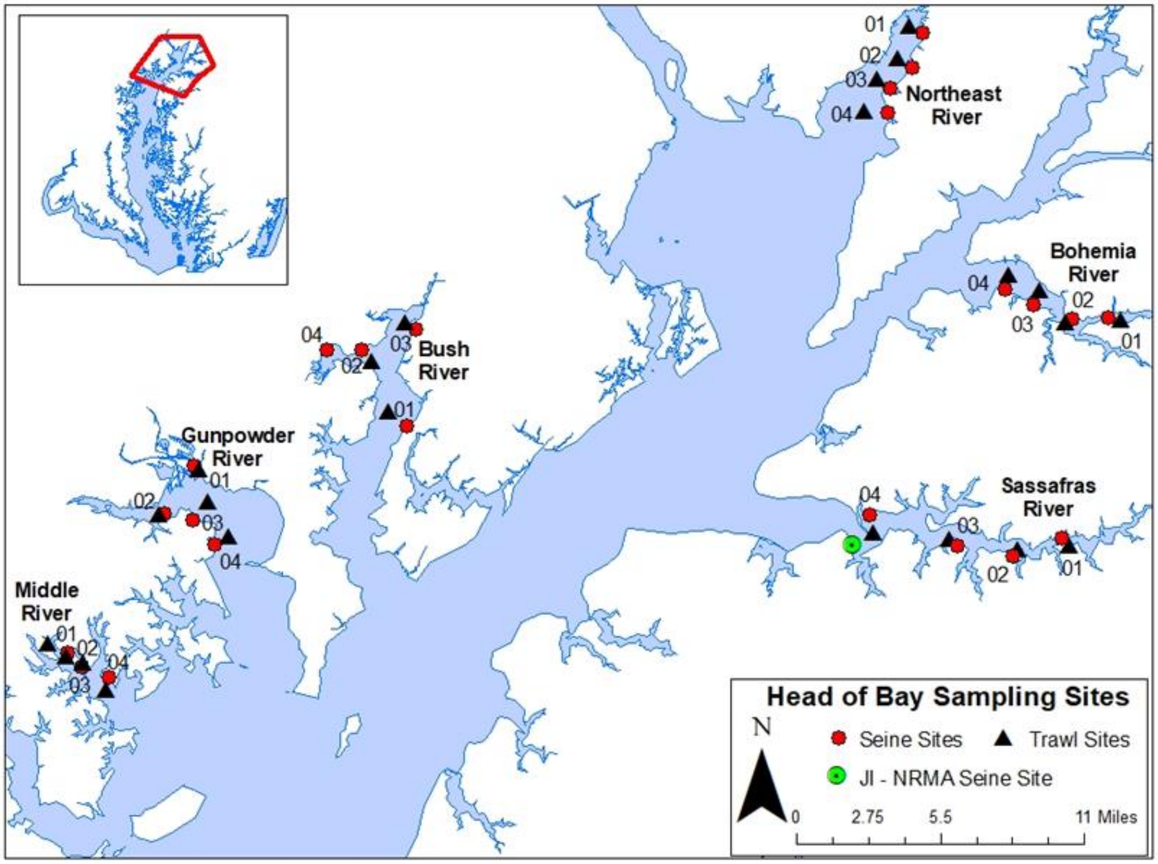


Figure 3-5. Mean subestuary bottom dissolved oxygen during summer sampling, 2003 – 2021, plotted against level of development (C / ha or structures per hectare).

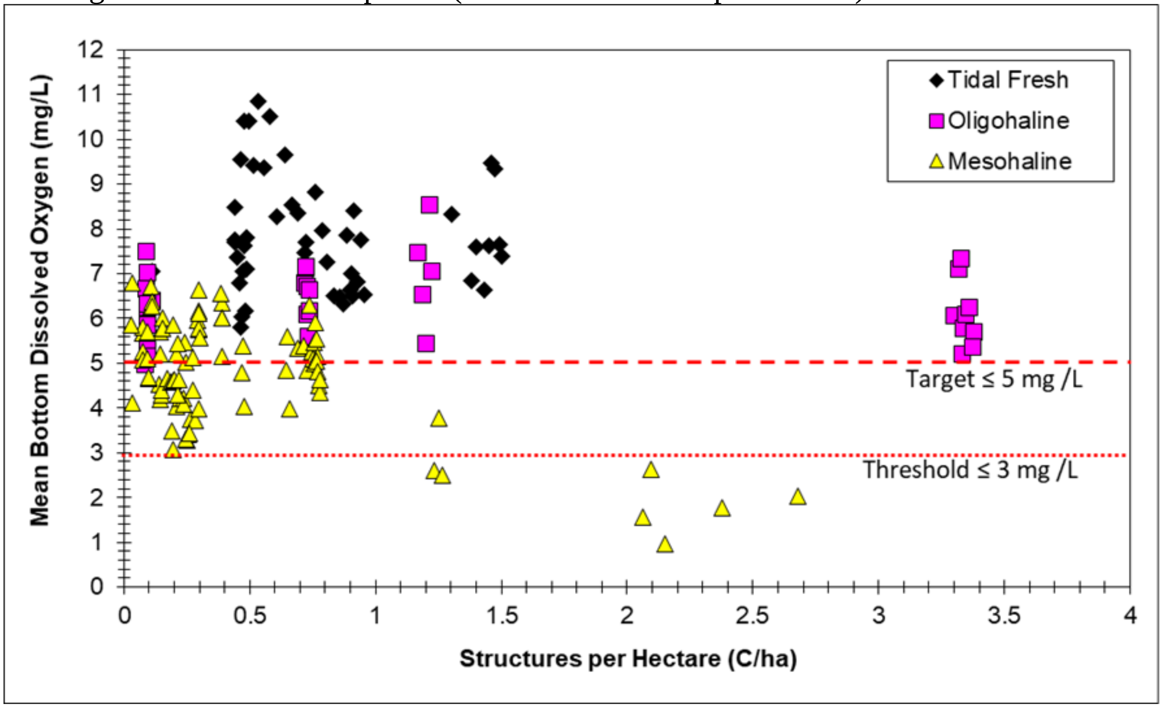


Figure 3-6. Mean subestuary surface dissolved oxygen during summer (July – October) sampling, 2003 – 2021, plotted against level of development (C / ha or structures per hectare).

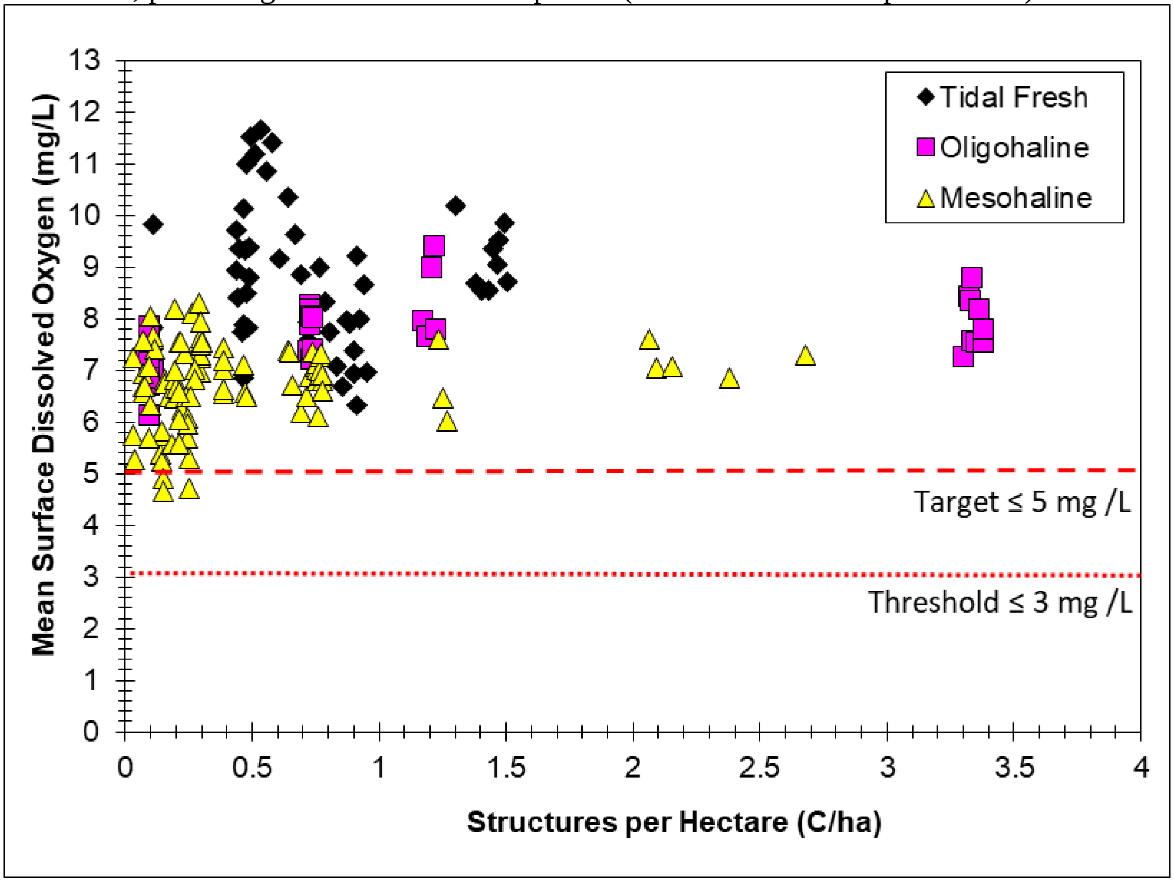


Figure 3-7. Maryland Department of Planning (DOP) estimates agricultural land coverage (% watershed land area) by region (western or eastern shore) versus median bottom dissolved oxygen (DO) in mesohaline subestuaries within major drainages (2003 – 2021). Quadratic model predicts median bottom DO and agricultural coverage (%) using data from both regions.

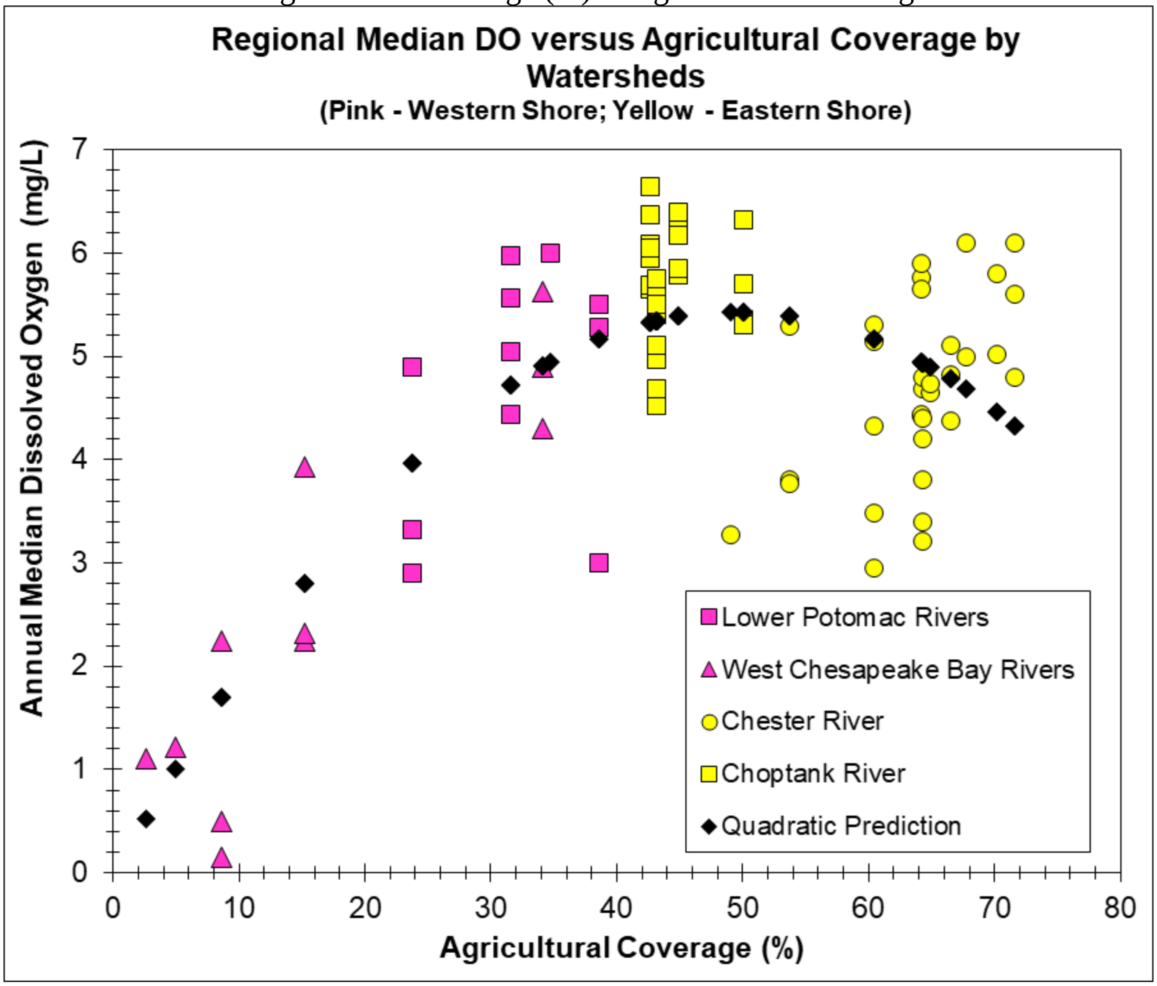


Figure 3-8. Trends in development (structures per hectare = C / ha) from 1950 to 2021 of watersheds of three subestuaries surveyed in the Choptank River, Broad Creek, Harris Creek, and Tred Avon River. Black diamond markers indicate the years that subestuaries were sampled. Development data was not available for 2021 and 2020 was used for this year.

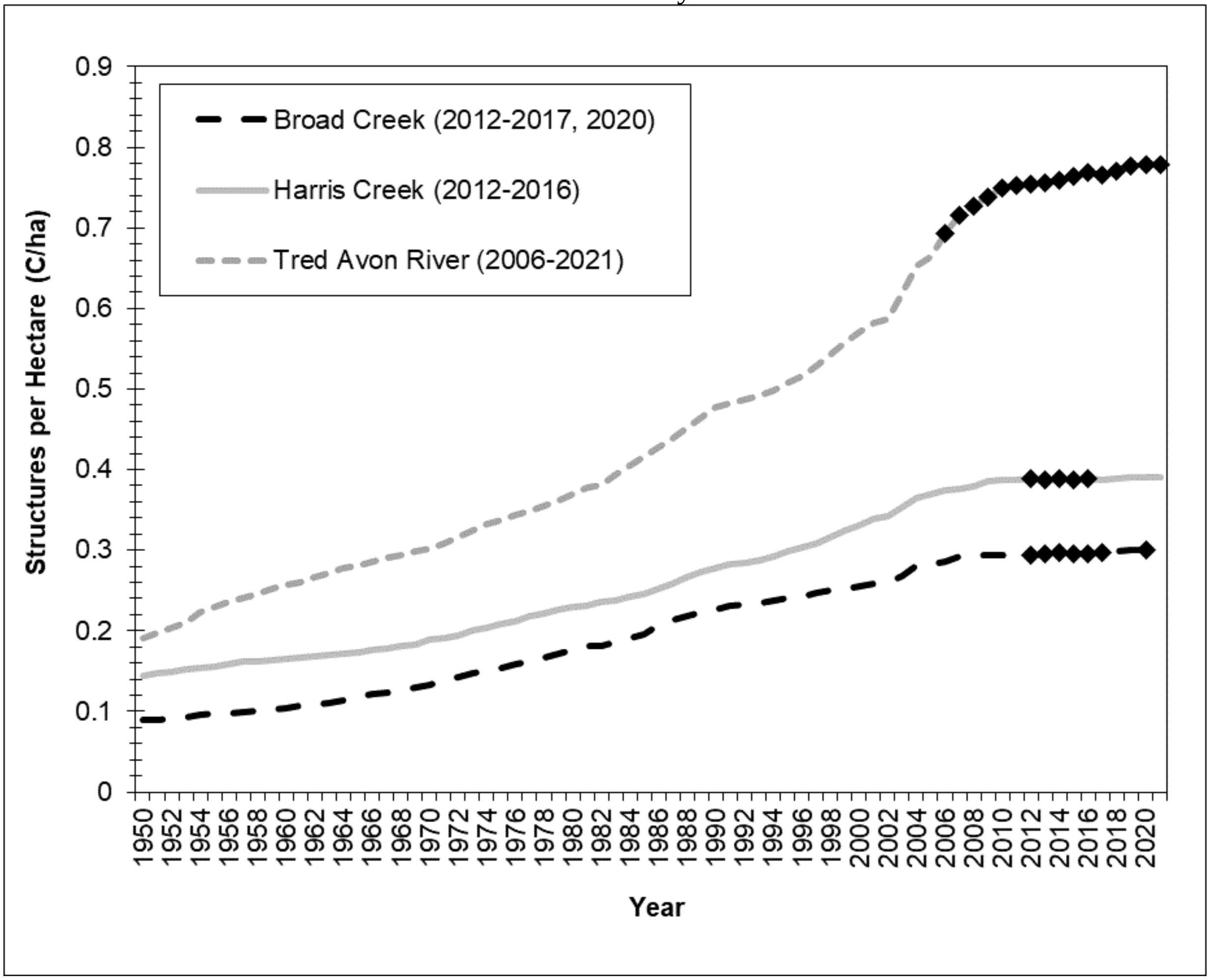


Figure 3-9. Bottom dissolved oxygen (DO; mg / L) readings (2003 – 2021) in Choptank River subestuaries, Broad Creek, Harris Creek, and Tred Avon River, versus intensity of development (C / ha = structures per hectare) in Talbot County. Target (5 mg / L) and threshold (3 mg / L) boundaries are indicated by red dashed lines. See legend for years subestuaries were sampled.

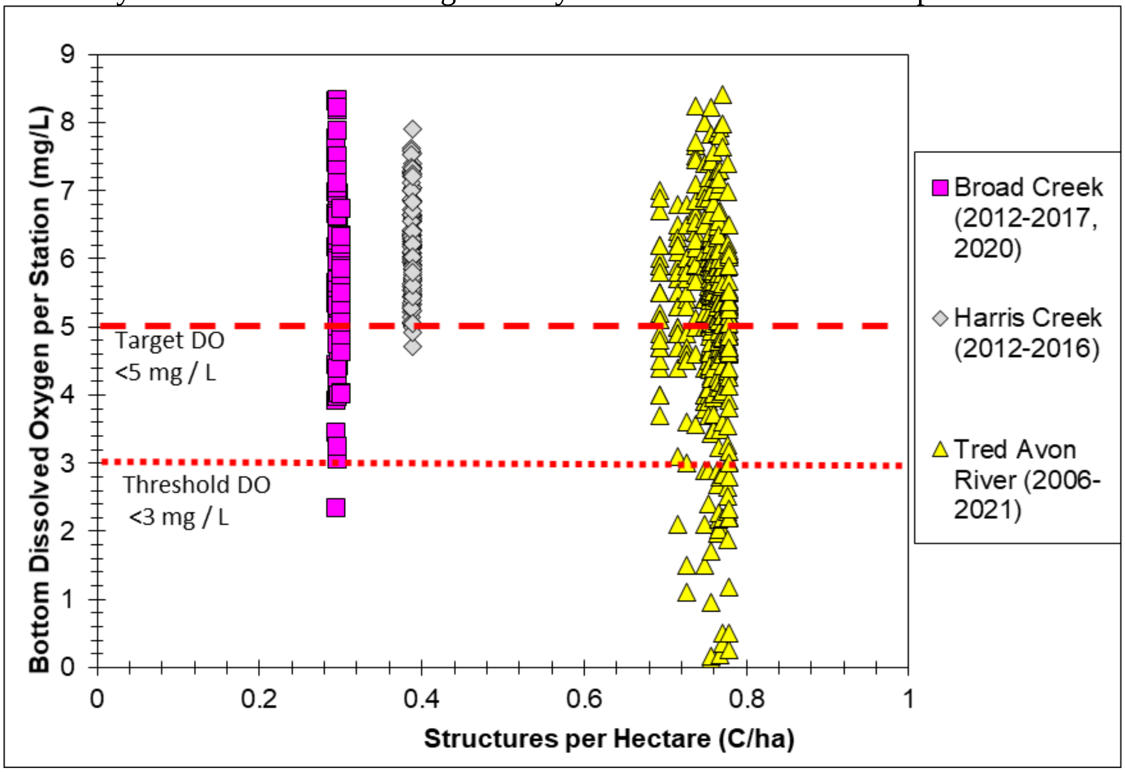


Figure 3-10. Median bottom dissolved oxygen (DO; red squares; mg / L) by year sampled for Broad Creek, Harris Creek, and Tred Avon River. Solid black bars indicate range of all bottom DO measurements for that year. The y-axes range from 0 to 9 mg / L; x-axes range are years from 2005 to 2022.

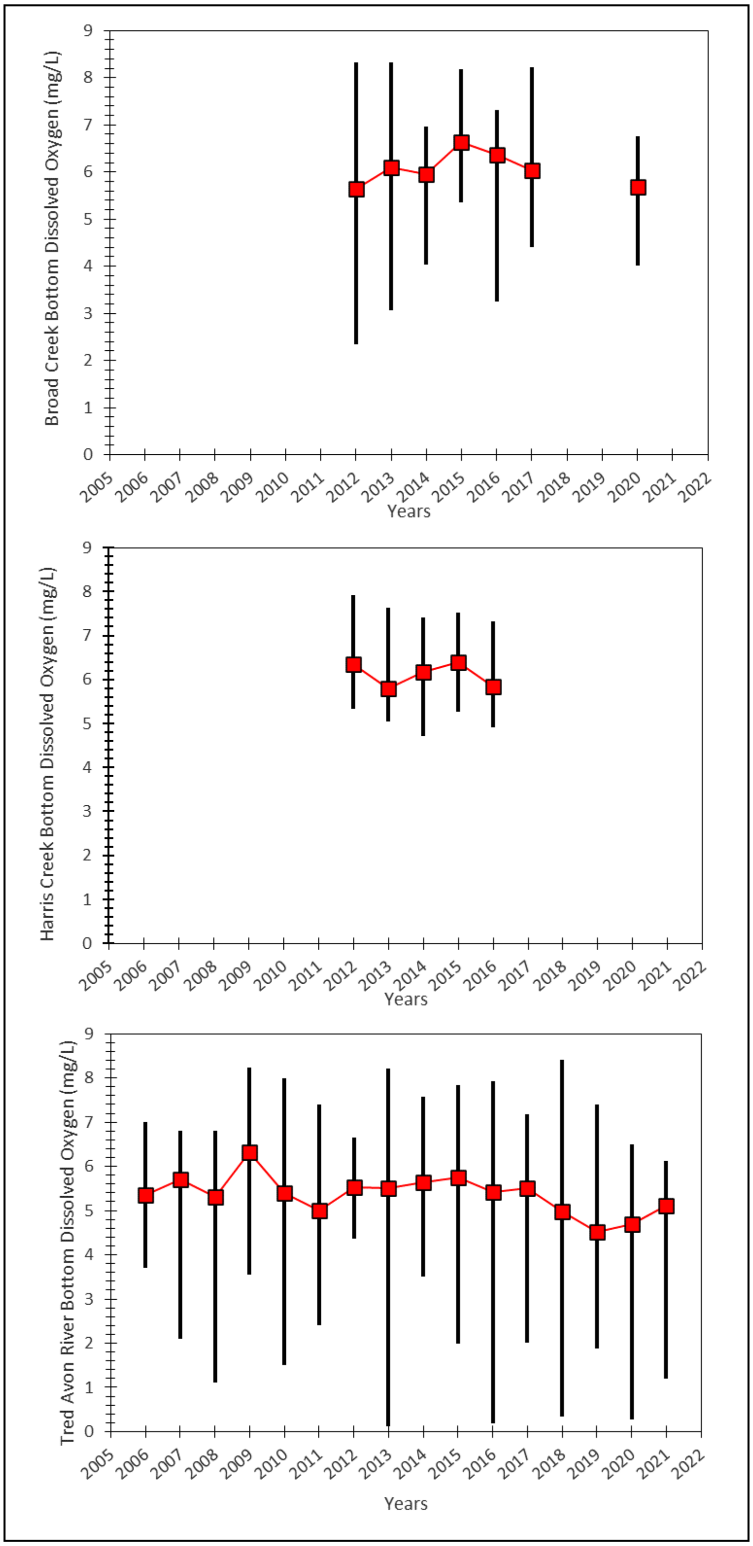


Figure 3-11. Mean bottom dissolved oxygen (DO; mg / L) for all years surveyed for Broad Creek, Harris Creek, and Tred Avon River, by sampling station. Dotted line indicates the median of all DO measurement data for the time-series. The y-axes range from 0 to 8 mg / L; x-axes range are years from 2005 to 2022.

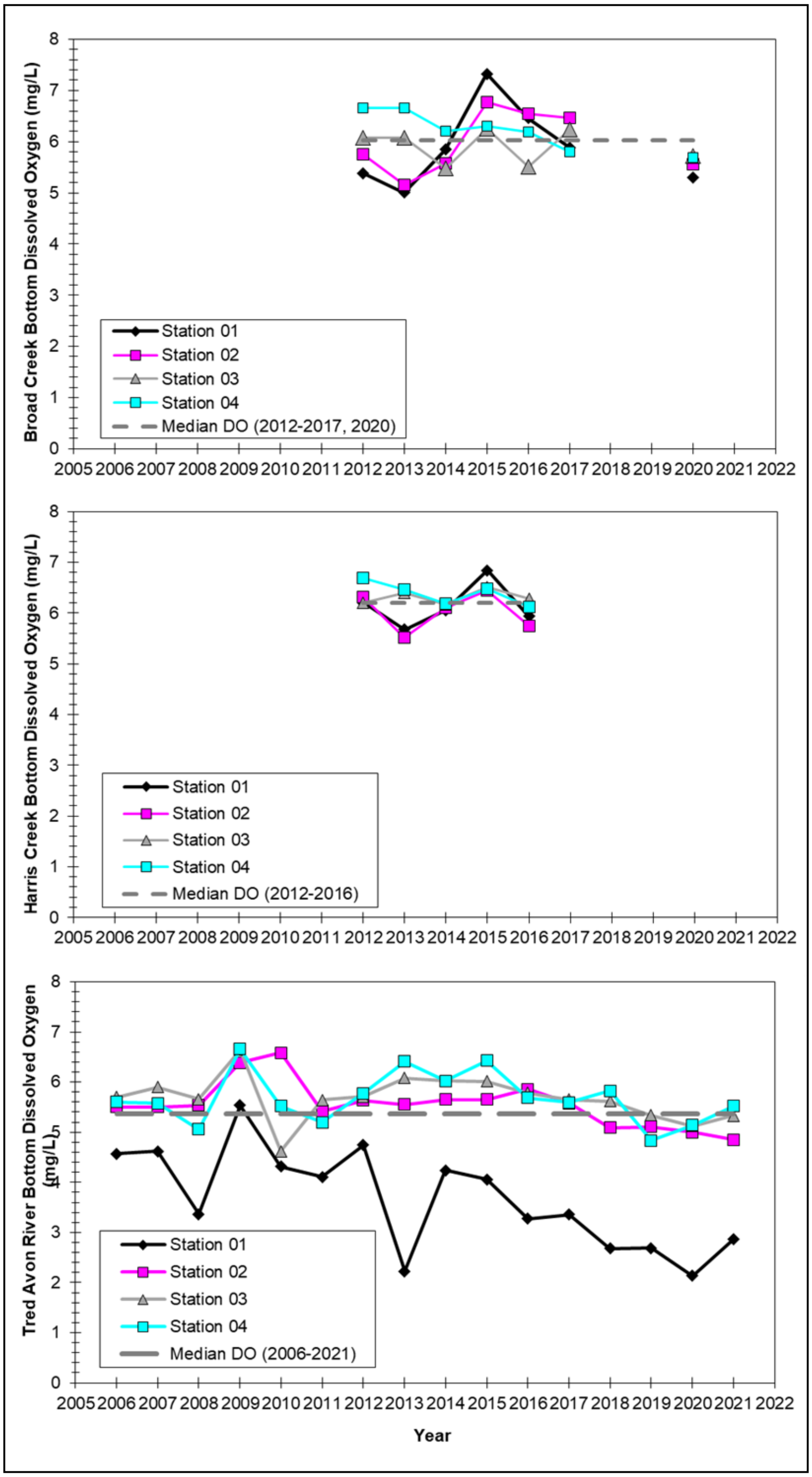


Figure 3-12. Median Secchi depth (m) for Broad Creek, Harris Creek, and Tred Avon River (red squares), by year. Solid black bars indicate the range of Secchi depth (m) measurements by year. The y-axes range from 0 to 2.5m; x-axes range are years from 2005 to 2022.

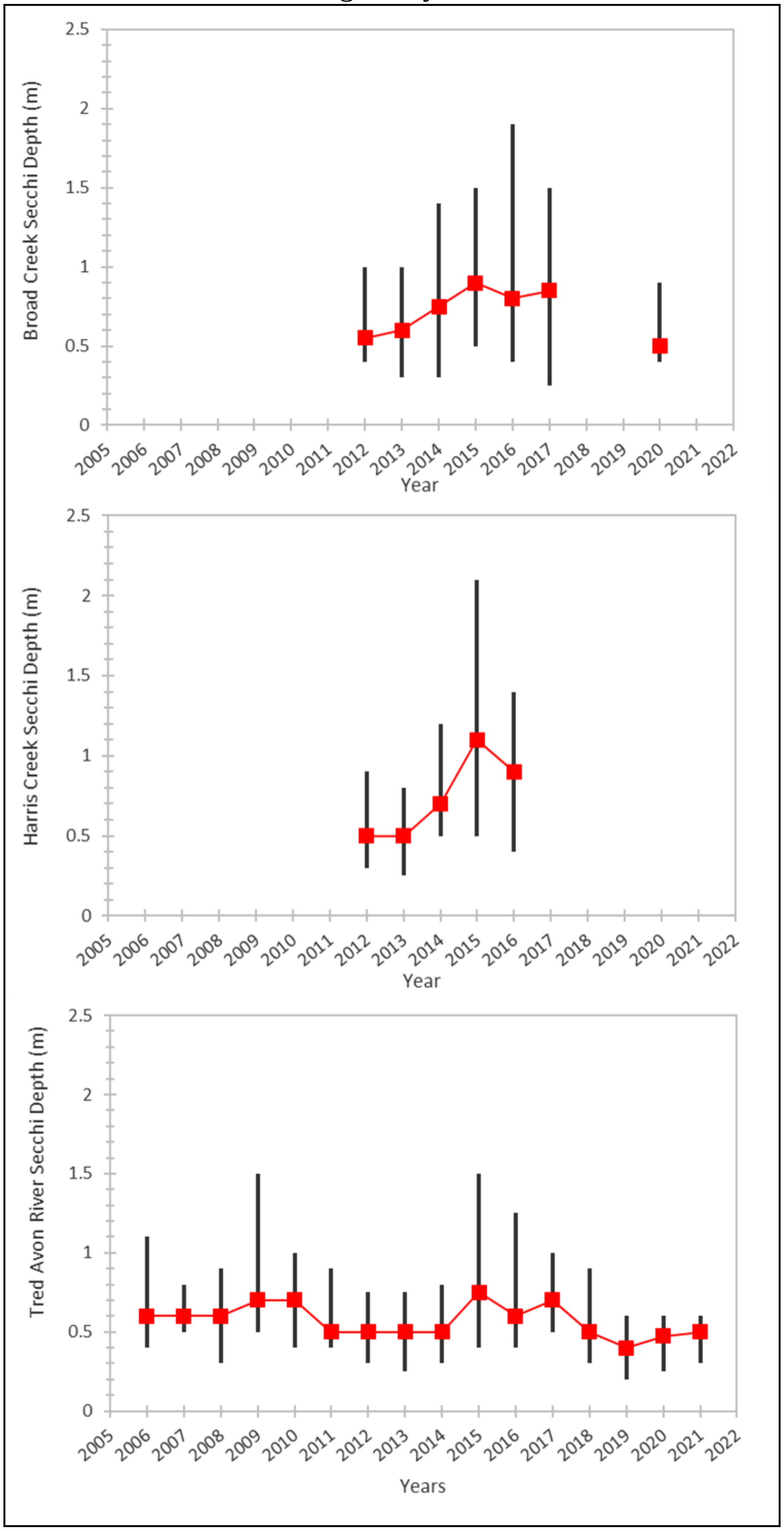


Figure 3-13. Coverage of SAV (percent of coverage in water area) for the mouth of the Choptank River (containing Broad Creek, Harris Creek, and Tred Avon River) during 1989 – 2020. Median of only fully mapped years (1989 – 2017, 2019-2020) for the time-series is indicated by the dashed line. Data for 2021 was not available at the time of this report.

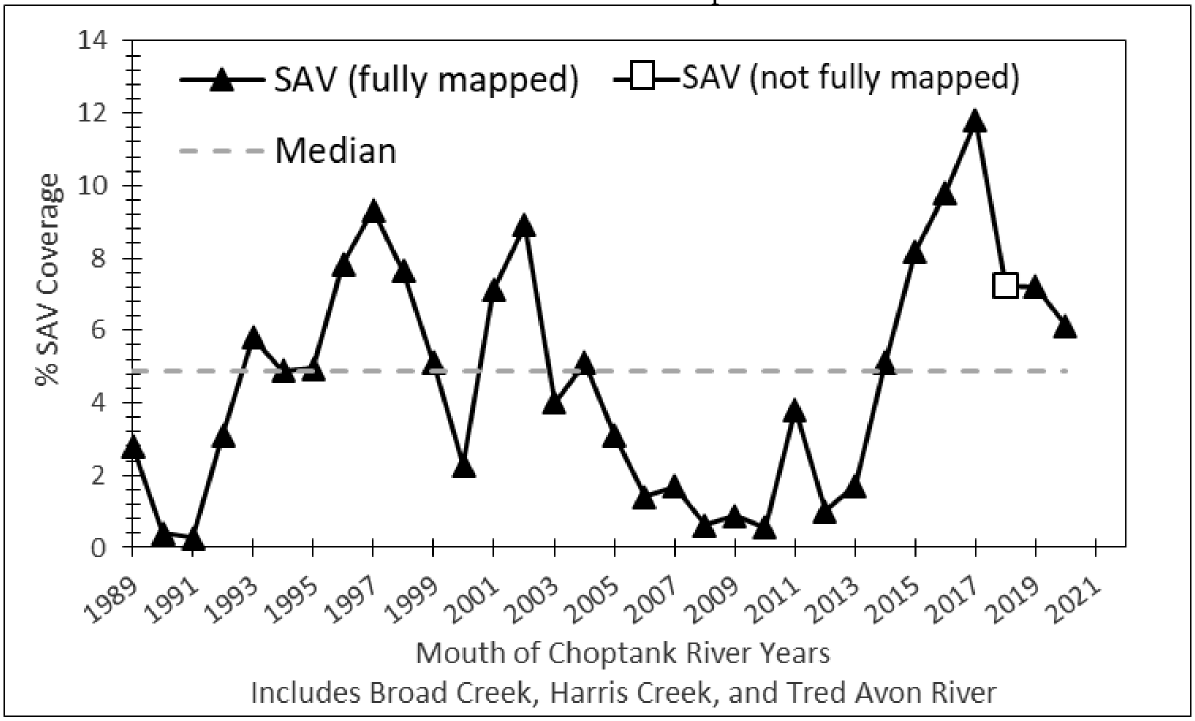


Figure 3-14. Median bottom pH (red squares) for Broad Creek, Harris Creek, and Tred Avon River, by sampling year. Solid black bars indicate the range of pH measurements by year. The y-axes range from 5.5 to 9.5; x-axes range are years from 2005 to 2022.

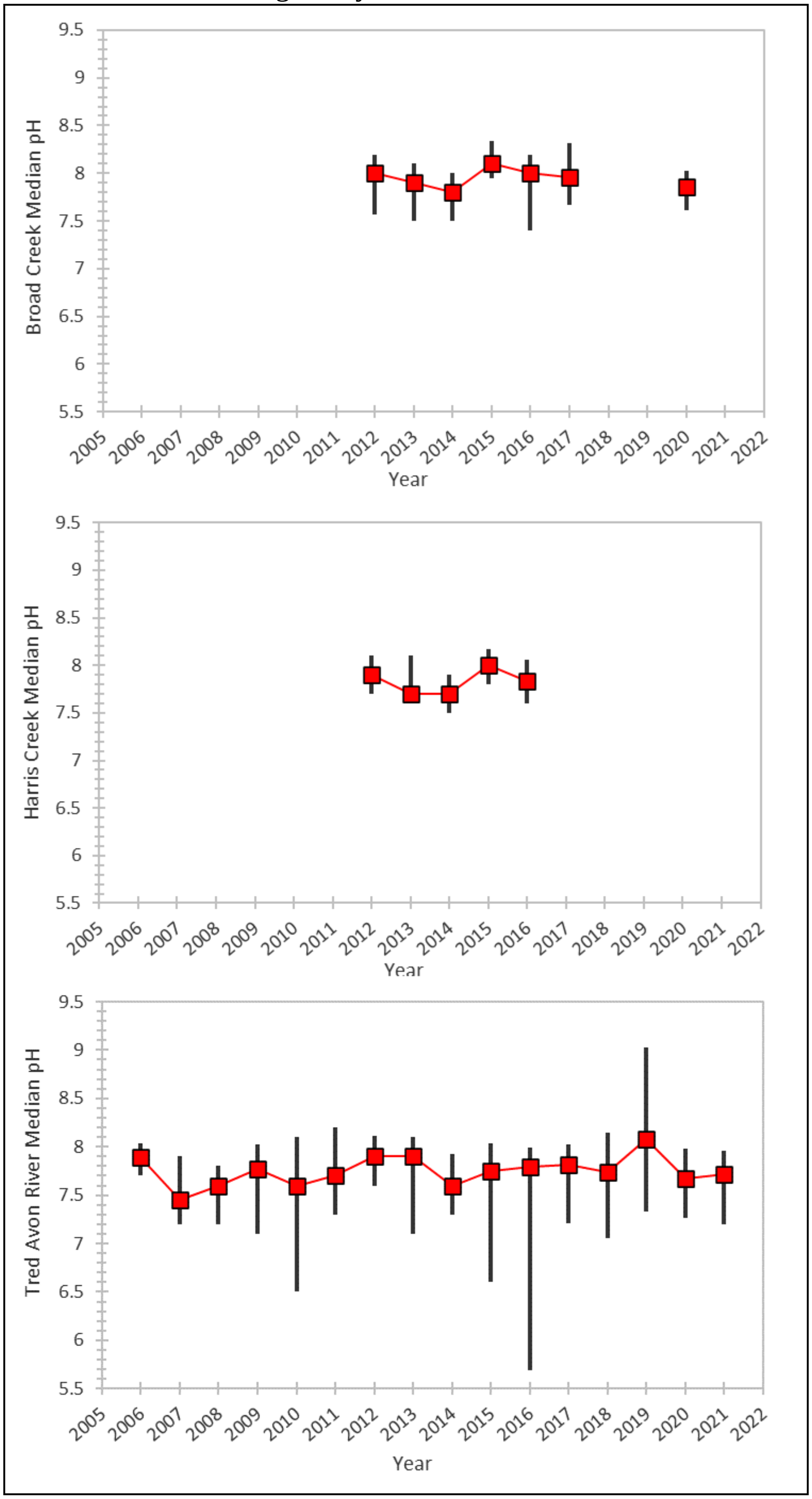


Figure 3-15. Median bottom salinity (red squares; ppt = ‰) for Broad Creek, Harris Creek, and Tred Avon River, by sampling year. Solid black bars indicate the range of salinity measurements by year. The y-axes range from 0 to 18 ppt; x-axes range are years from 2005 to 2022.

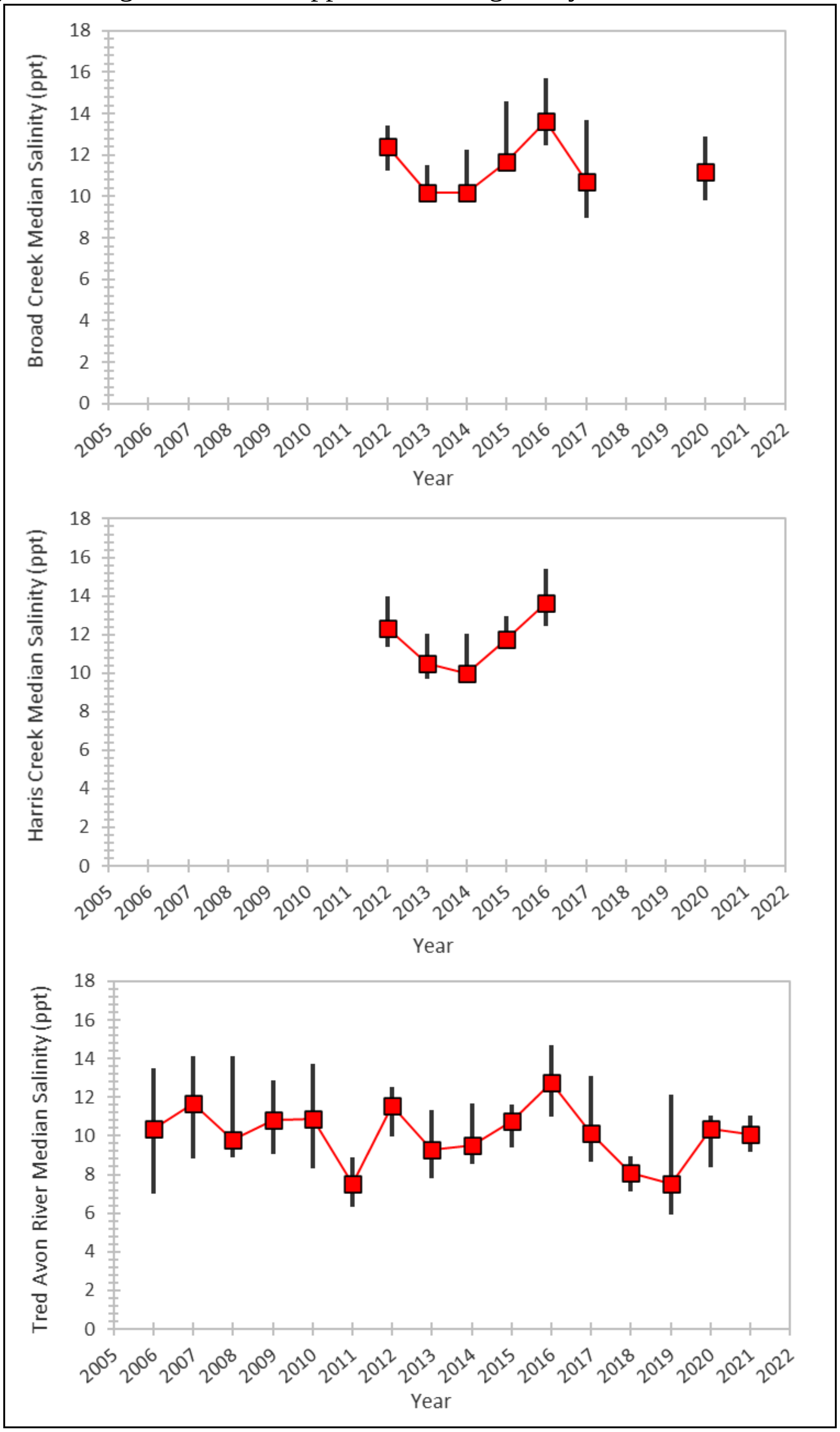


Figure 3-16. Trends in levels of development (structures per hectare = C / ha) during 1950 – 2019 in the Head-of-Bay subestuaries, Bohemia River, Bush River, Gunpowder River, Middle River, Northeast River, and Sassafras River. Black diamond markers indicate the years that subestuaries were sampled. Tax map data were not available for 2020 and 2021.

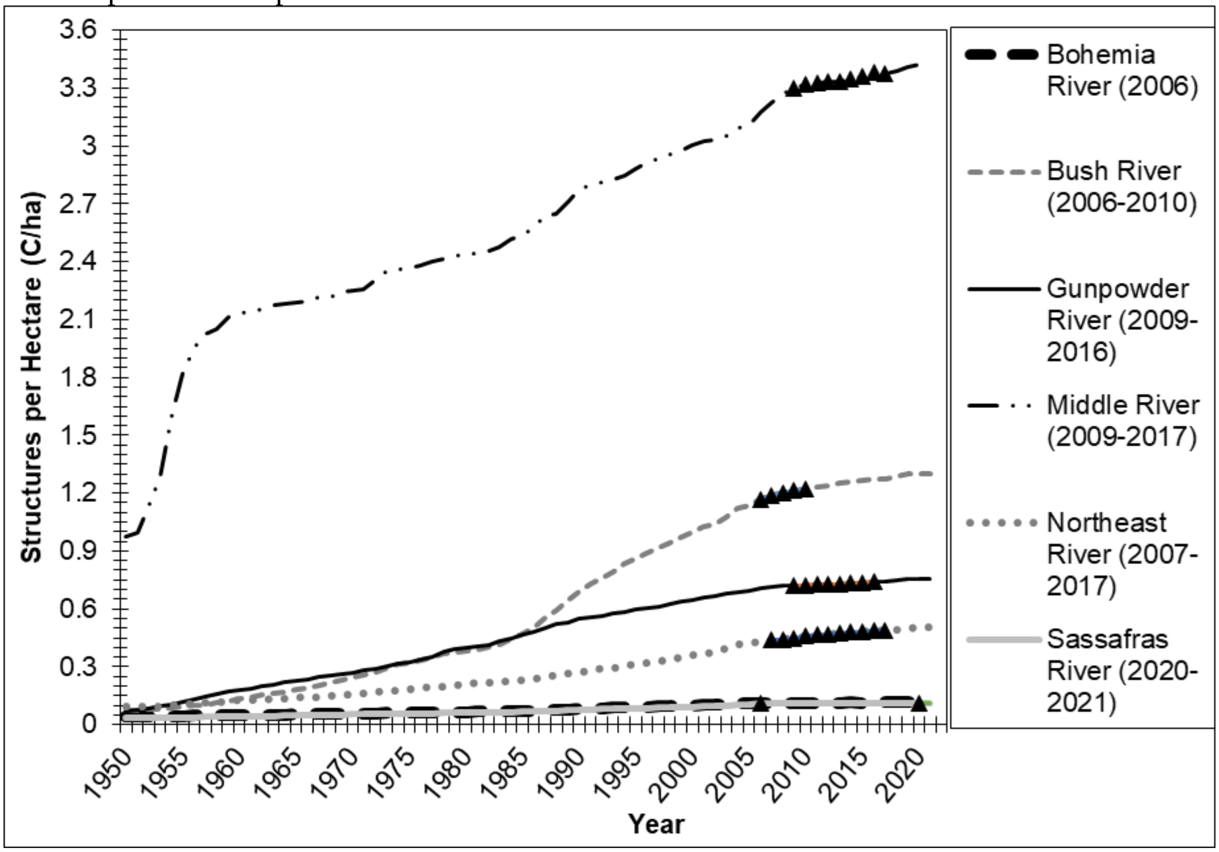


Figure 3-17. Summer (July – September) median bottom dissolved oxygen (DO; red squares; mg/L) for Chesapeake Bay Program Sassafras River (CBP ET3.1) and Head-of-Bay (CBP CB1.1) monitoring stations from 1989 to 2021. Solid black bars indicate range of bottom DO measurements for each year. The y-axes range from 0 to 12 mg/L; x-axes range are years from 1988 to 2022.

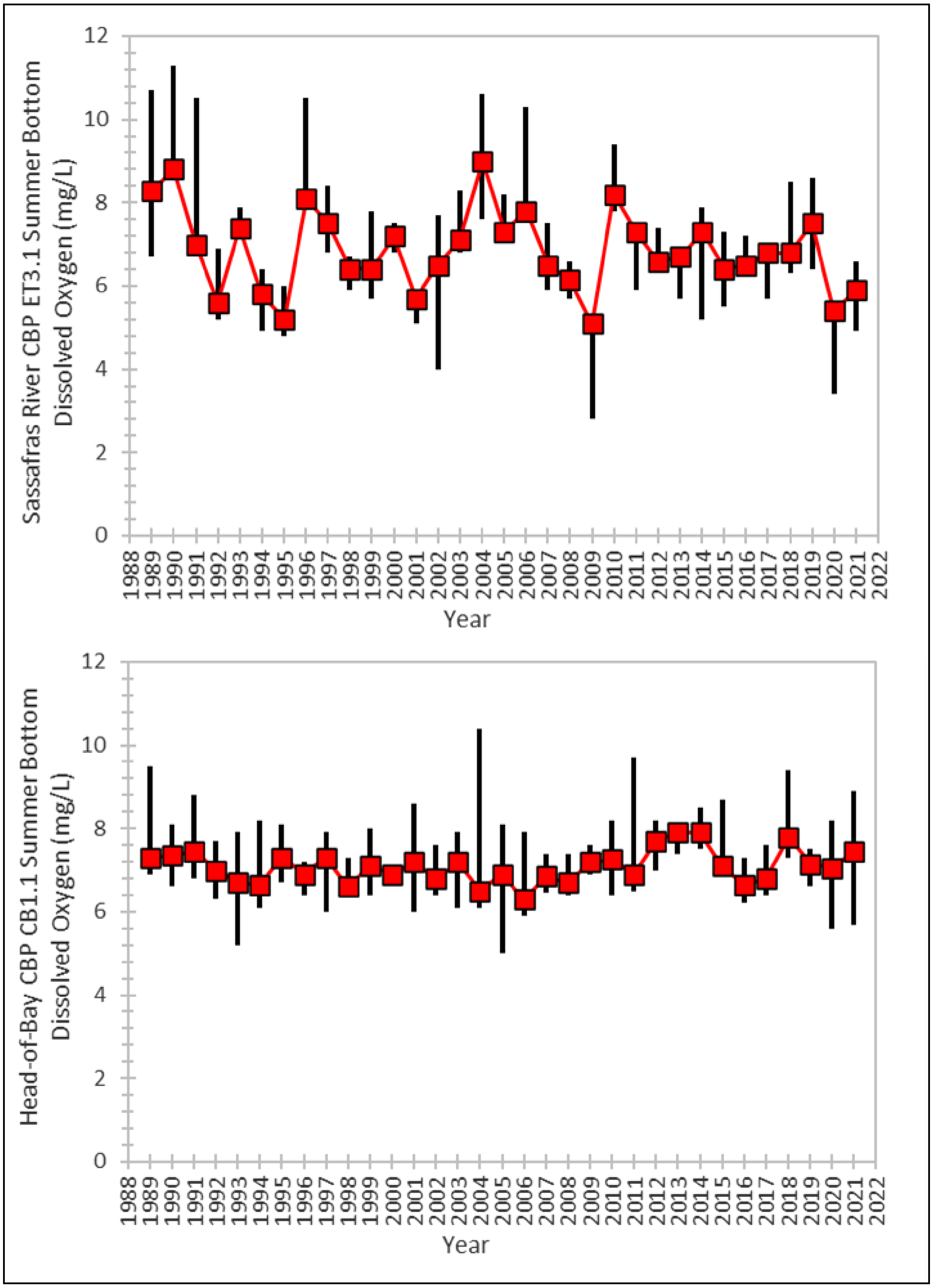


Figure 3-18. Bottom dissolved oxygen (DO; mg/L) versus intensity of development (C/ha = structures per hectare) in the Head-of-Bay subestuaries. Target (= 5 mg/L) and threshold (= 3 mg/L) boundaries are indicated (red dashed lines). The y-axes range from 0 to 16 mg/L; x-axes range from 0 to 3.6 structures per hectare (C/ha).

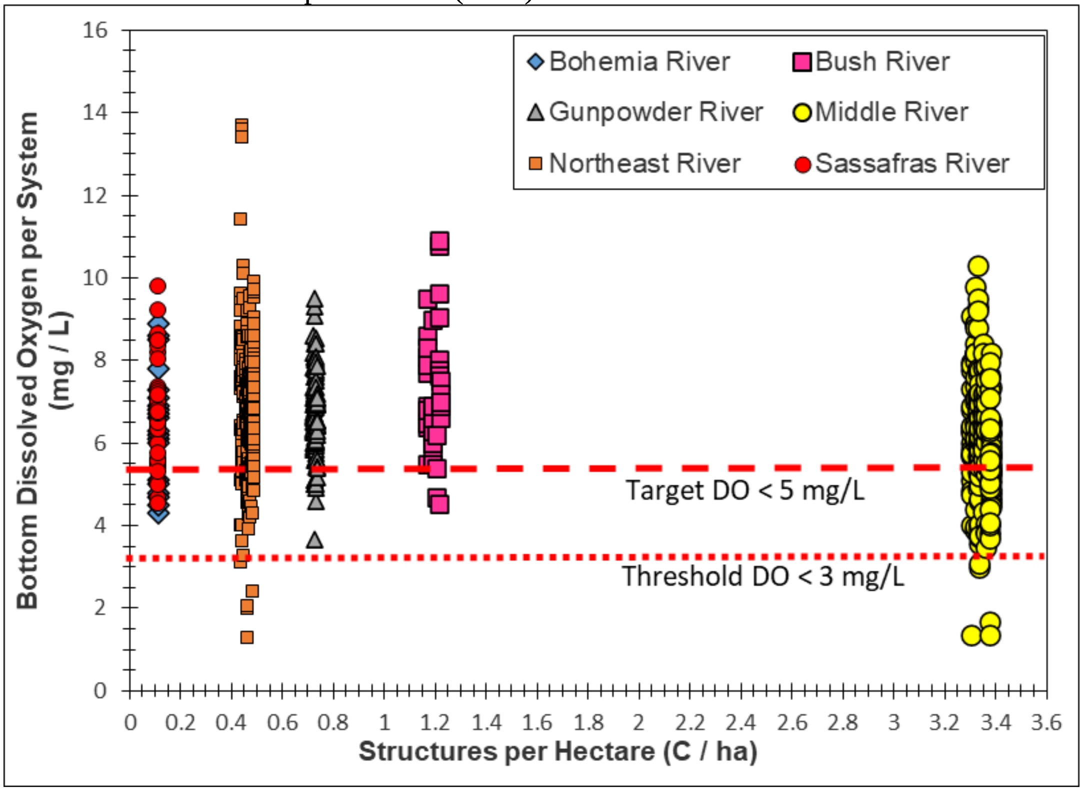


Figure 3-19. Median bottom dissolved oxygen (DO; red squares; mg/L) for Head-of-Bay subestuaries for each year sampled. Solid black bars indicate range of bottom DO measurements for that year. The y-axes range from 0 to 16 mg/L; x-axes range are years from 2005 to 2022.

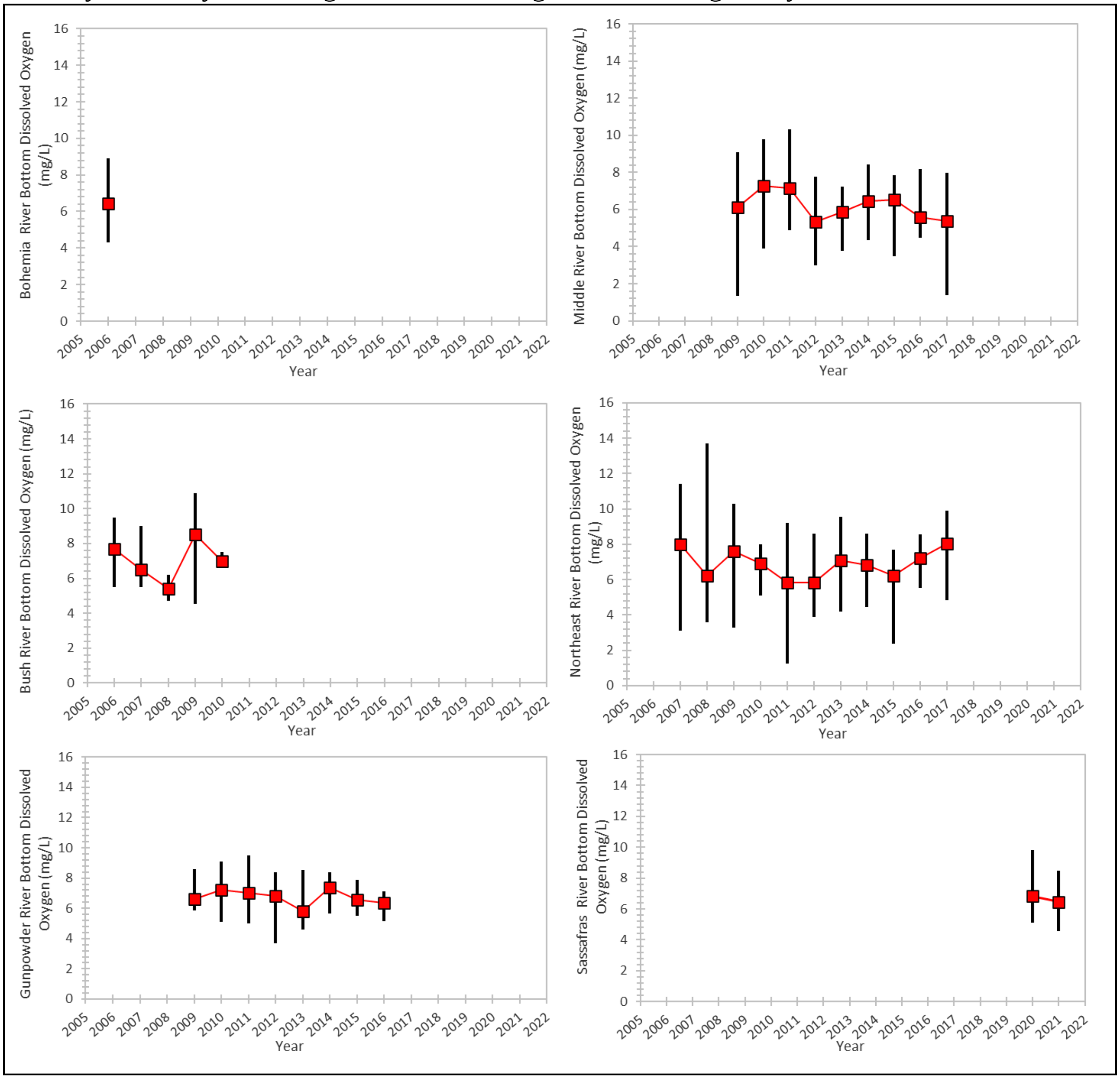


Figure 3-20. Mean bottom dissolved oxygen (DO; mg/L) for all years surveyed for Head-of-Bay subestuaries, by sampling station. Dotted line indicates the median of all DO measurement data for the time-series available. The y-axes range from 3 to 11 mg/L; x-axes range are years from 2005 to 2022.

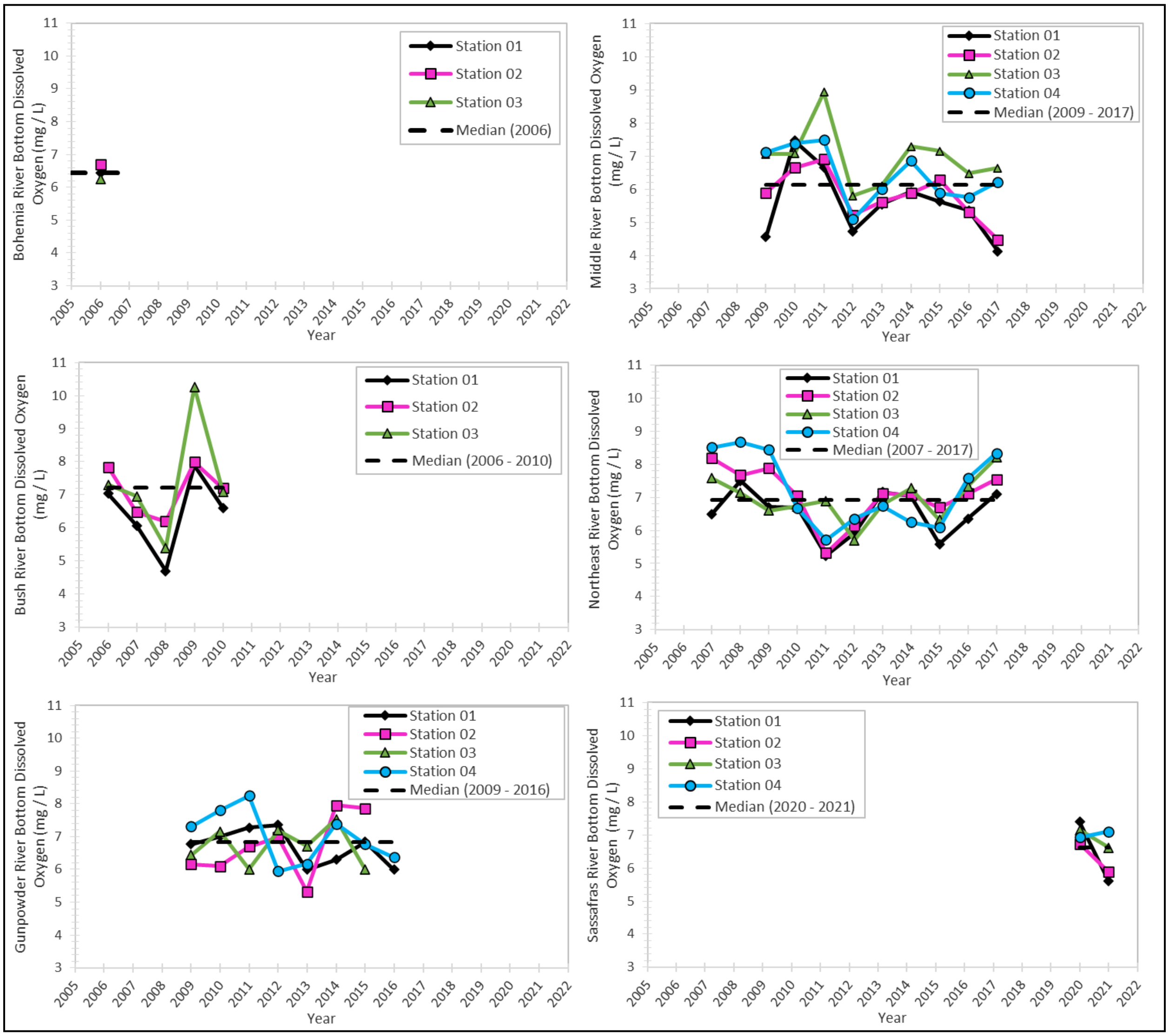


Figure 3-21. Median Secchi depth (m) for Bohemia River, Bush River, Gunpowder River, Middle River, Northeast River, and Sassafras River (red squares), by year. Solid black bars indicate the range of Secchi depth (m) measurements by year. The y-axes range from 0 to 2.0m; x-axes range are years from 2005 to 2022.

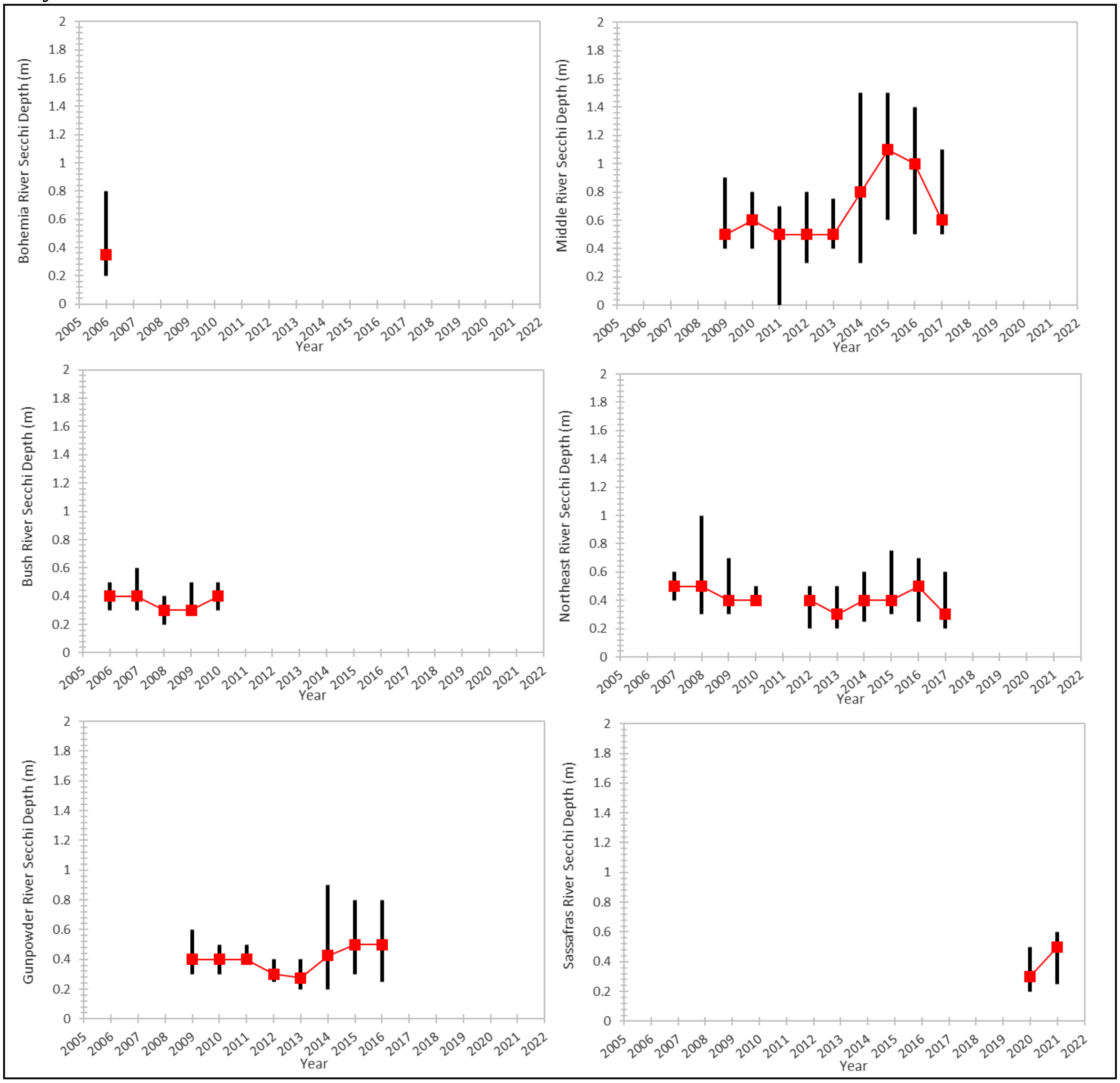


Figure 3-22. Median bottom pH (red squares) for Bohemia River, Bush River, Gunpowder River, Middle River, Middle River, Northeast River, and Sassafras River, by sampling year. Solid black bars indicate the range of pH measurements by year. The y-axes range from 6 to 10; x-axes range are years from 2005 to 2022.

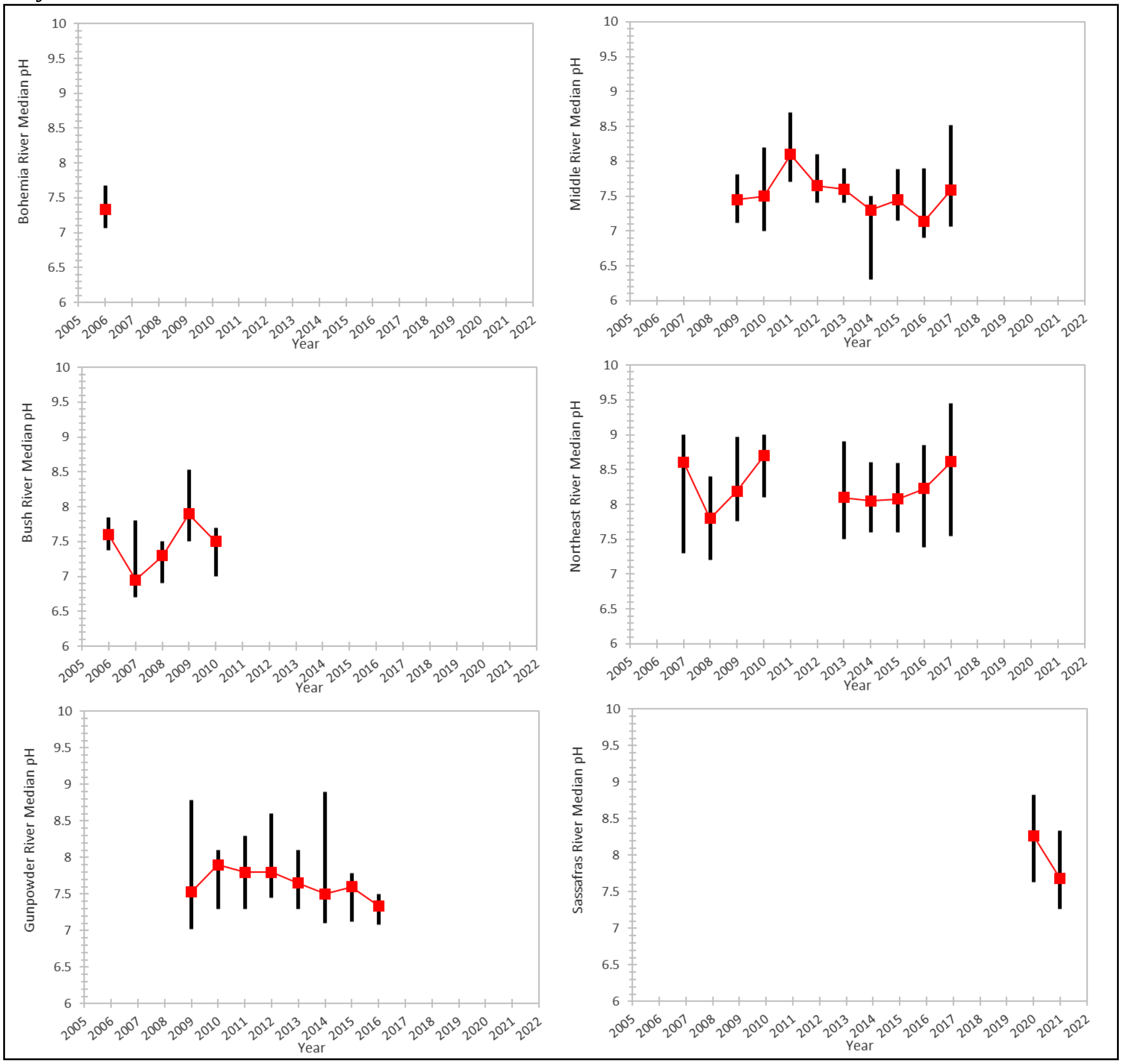


Figure 3-23. Median bottom salinity (red squares; ppt = ‰) for Bohemia River, Bush River, Gunpowder River, Middle River, Northeast River, and Sassafras River, by sampling year. Solid black bars indicate the range of salinity measurements by year. The y-axes range from 0 to 9 ppt; x-axes range are years from 2005 to 2022.

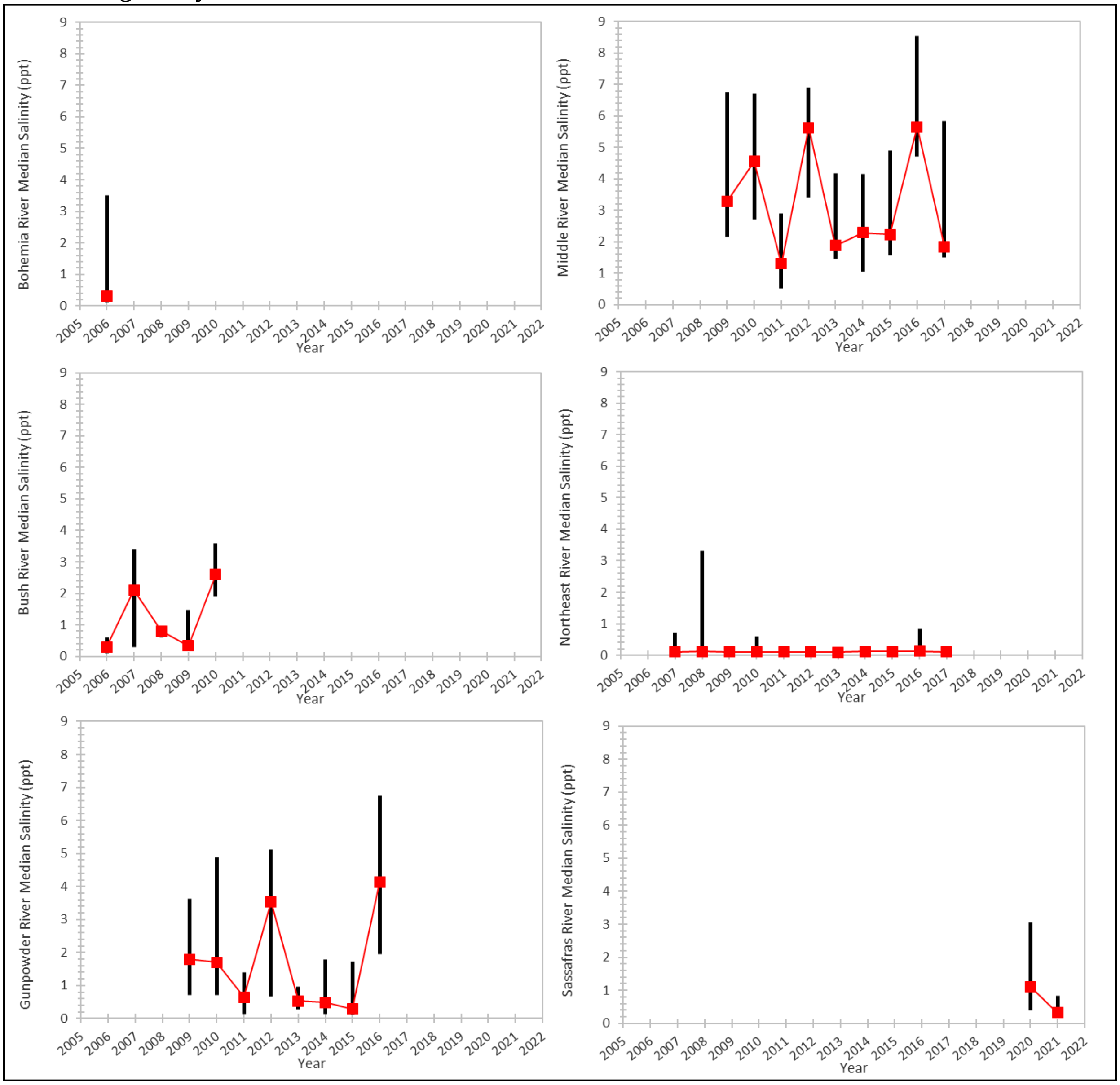


Figure 3-24. Annual number of finfish species (richness) collected by beach seines in tidal-fresh, oligohaline, and mesohaline subestuaries versus intensity of watershed development (C/ha = structures per hectare) from 2003 to 2021. Points were omitted if beach seine effort (number of samples) < 15 samples.

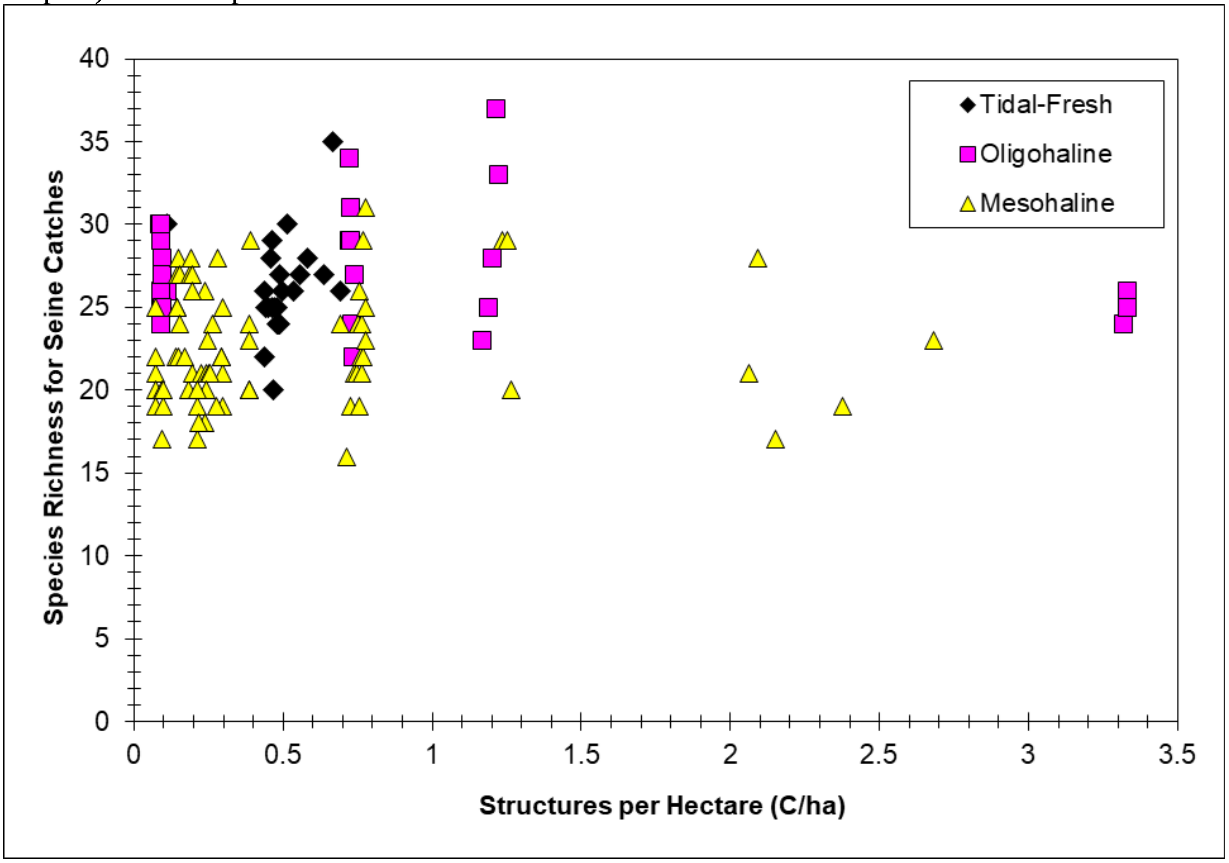


Figure 3-25. Annual 4.9m trawl geometric mean (GM) catches plotted against C/ha subestuaries sampled during 2003 – 2021, separated by salinity class.

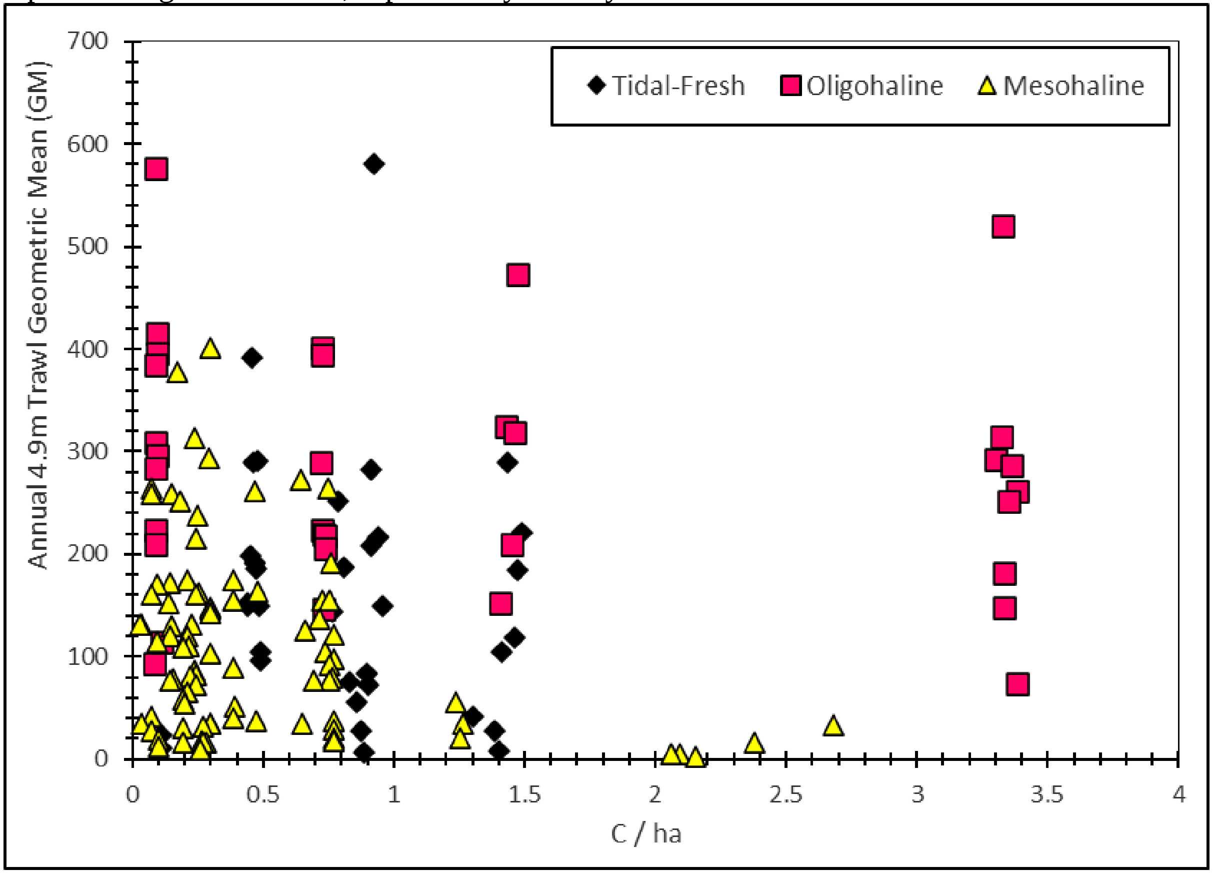


Figure 3-26. Number of finfish species (richness) collected by 4.9m bottom trawl in tidal-fresh, oligohaline, and mesohaline subestuaries versus intensity of development (C/ha = structures per hectare) from 2003 to 2021. Points were omitted if number of samples was less than 15.

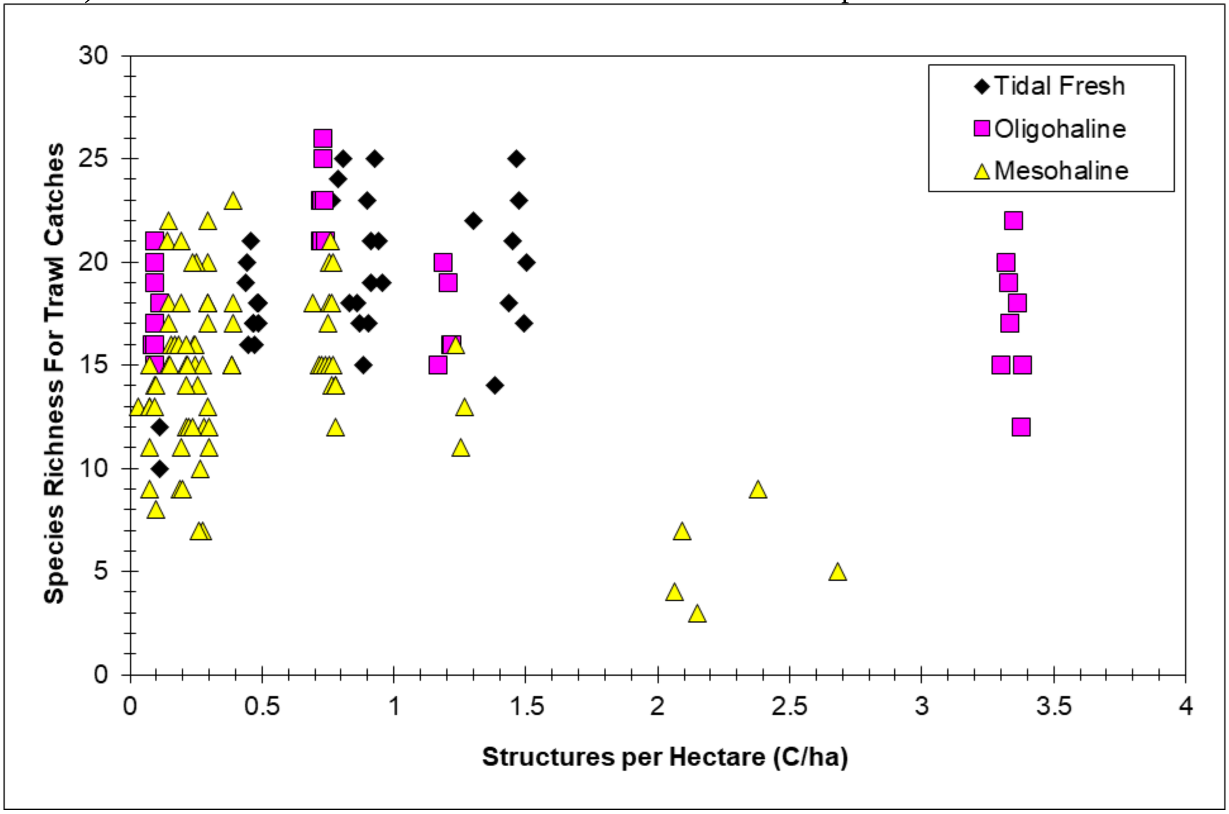


Figure 3-27. Annual 4.9 m bottom trawl catch geometric mean (GM) of all finfish species (red squares) for Broad Creek, Harris Creek, and Tred Avon River, by sampling year. Black bars indicate the 95% confidence intervals. The y-axes range from 0 to 600; x-axes range are years from 2005 to 2022.

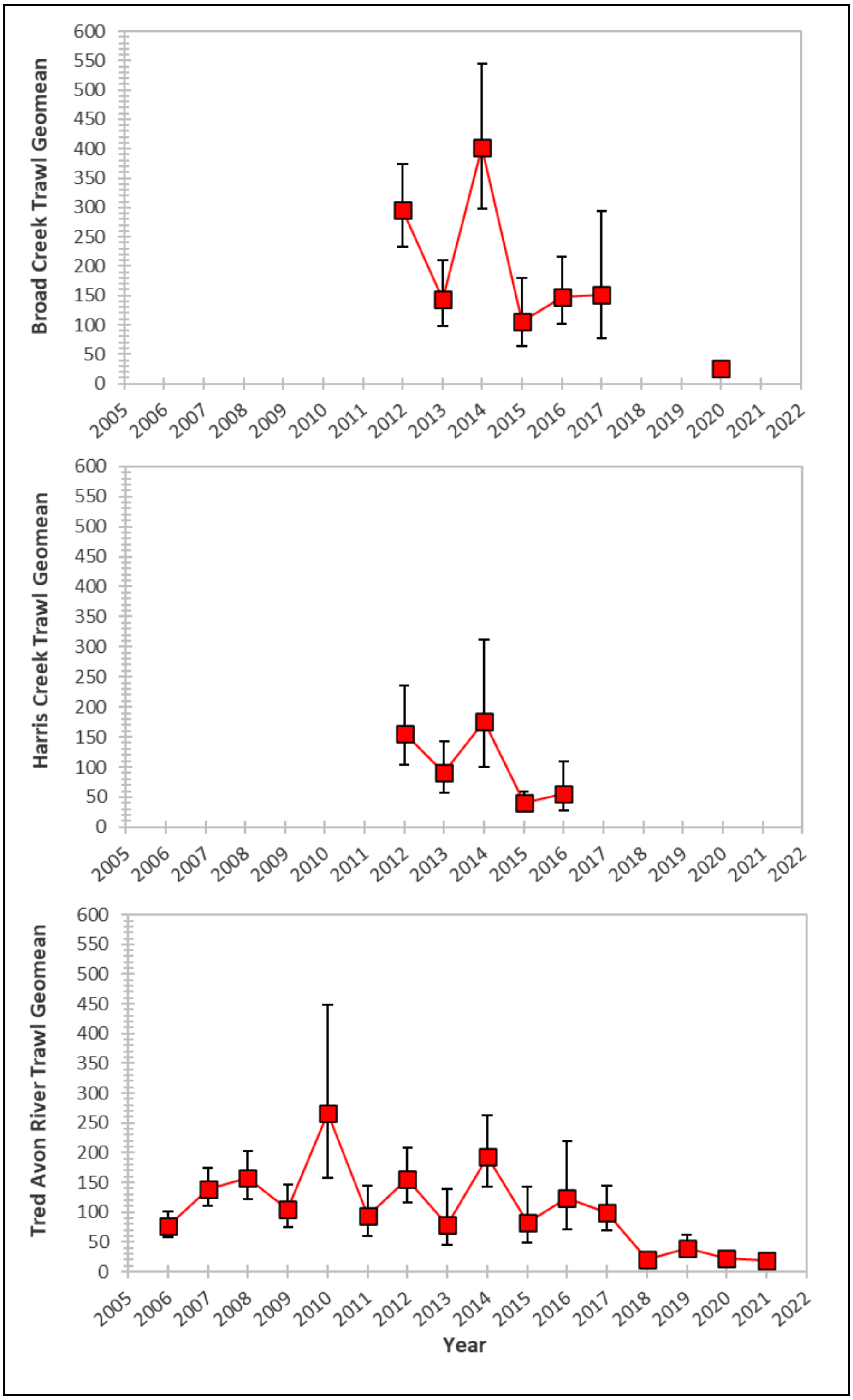


Figure 3-28. Finfish species composition for 4.9m bottom trawl catch in Broad Creek, Harris Creek, and Tred Avon River for all sampling years combined. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”.

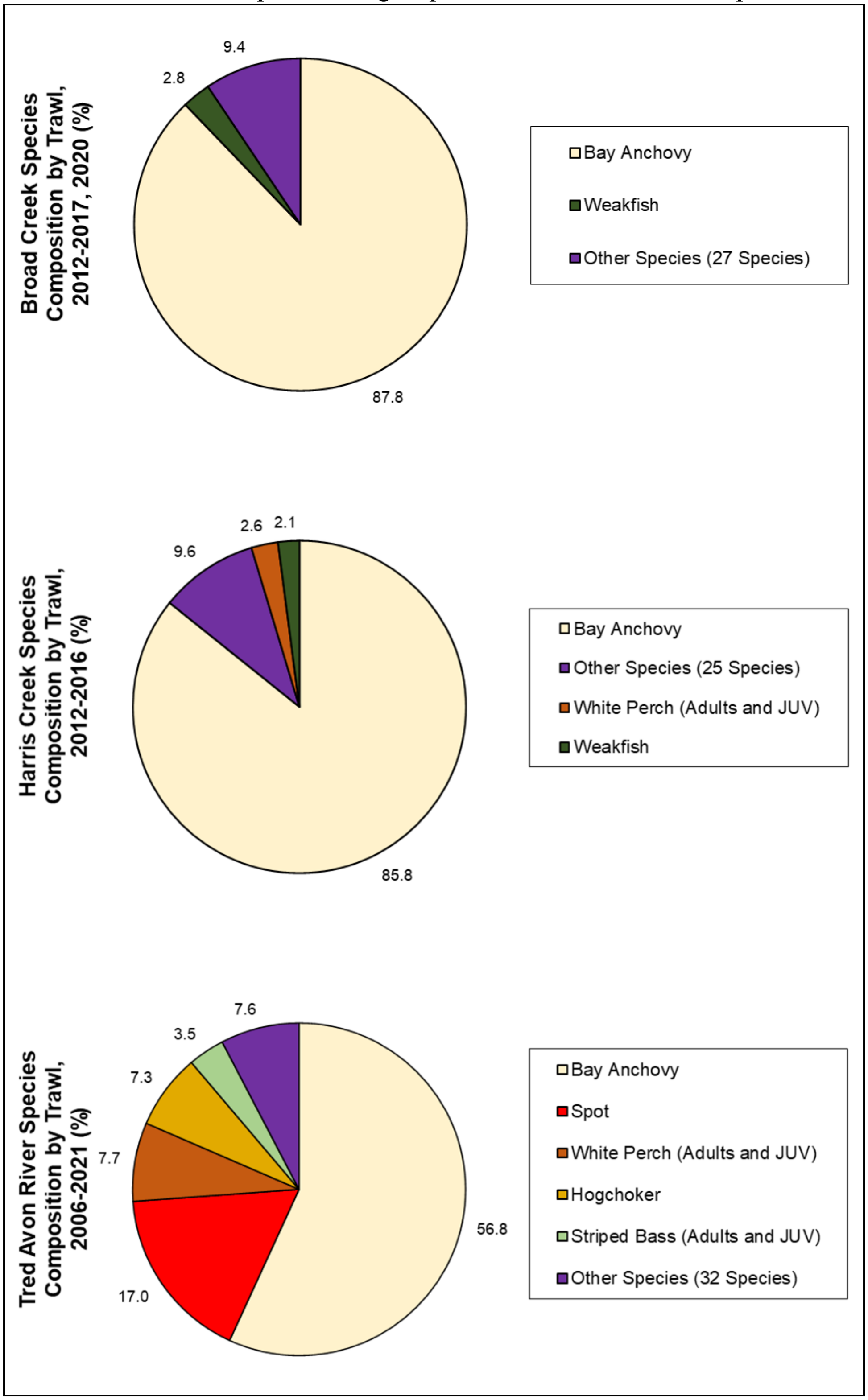


Figure 3-29. Finfish species composition for 4.9 m bottom trawl catch in Broad Creek, Harris Creek, and Tred Avon River for each year sampled. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”. The y-axes range from 0 to 100%; x-axes vary based on years sampled.

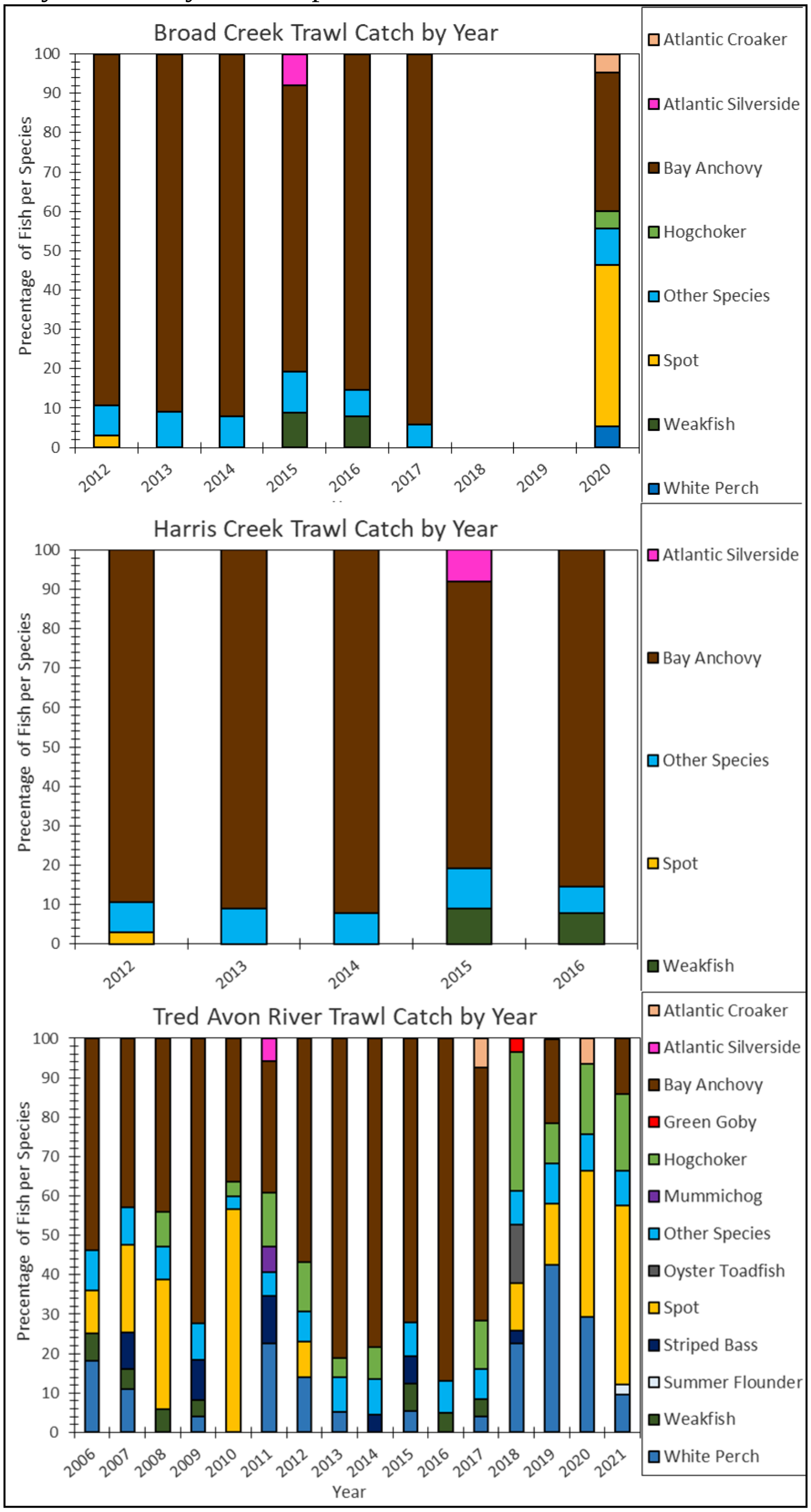


Figure 3-30. Percent similarity index (%) for 4.9m bottom trawl stations 01 – 04 in Choptank River tributaries, Broad Creek, Harris Creek, and Tred Avon River, by year. The greater the similarity value, the more finfish species there are in common throughout all bottom trawl stations (01 – 04). The y-axes range from 0 to 100%; x-axes range are years from 2005 to 2022.

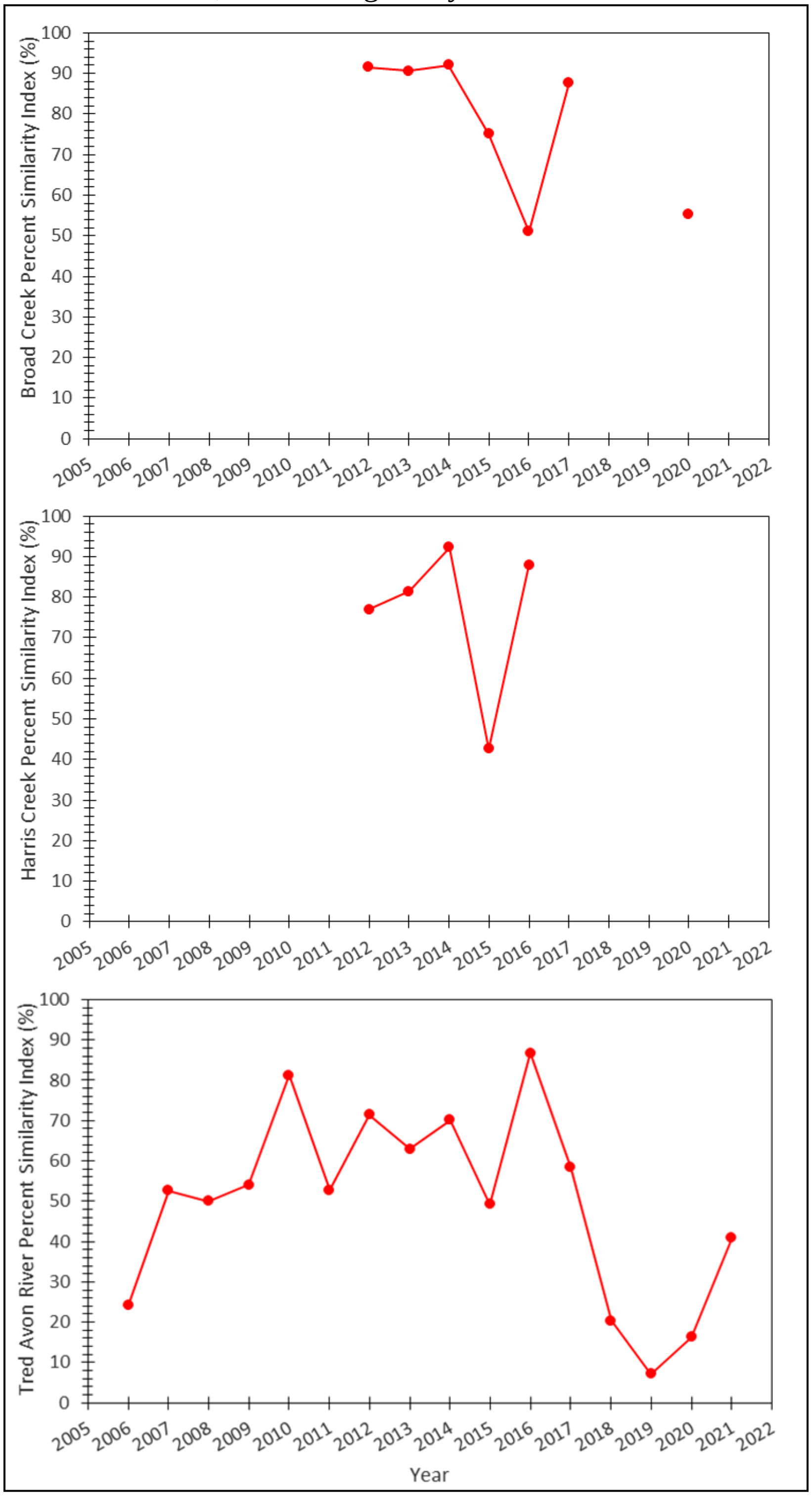


Figure 3-31. Finfish species composition for 4.9m bottom trawl catch in all mesohaline subestuaries sampled during 2003 – 2021, by year. Finfish species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”.

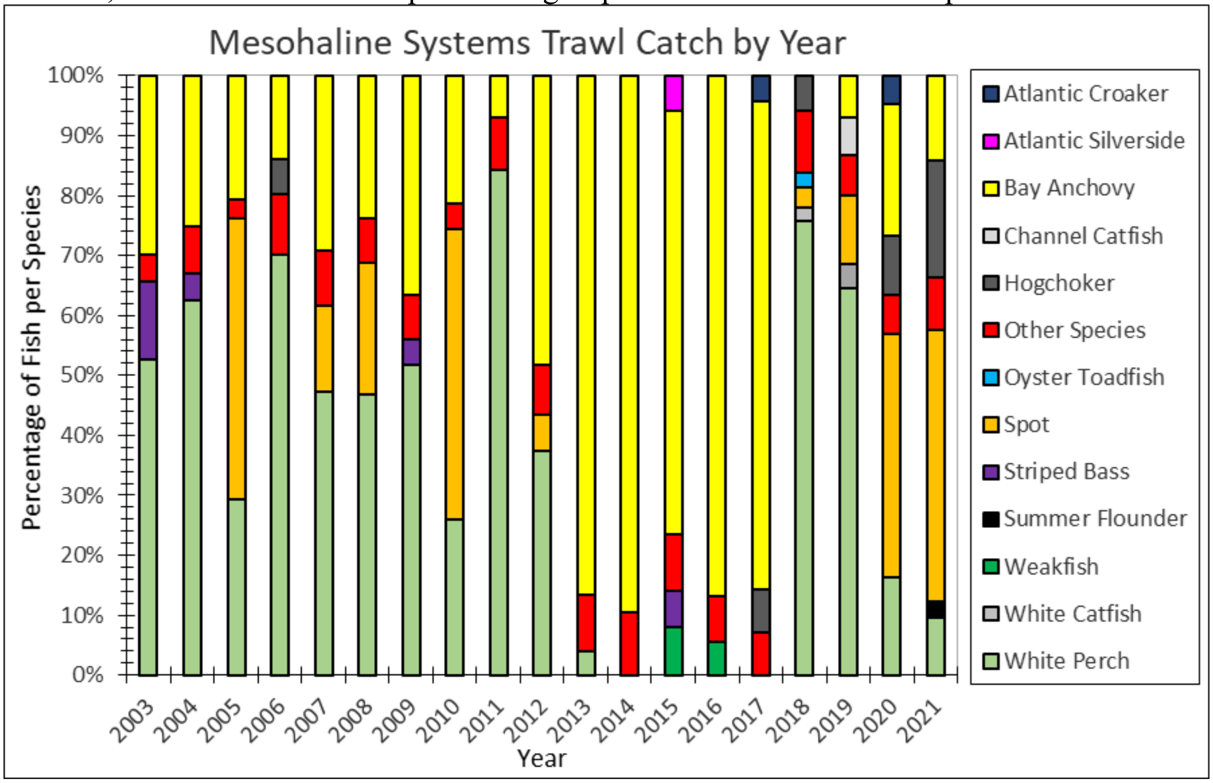


Figure 3-32. Geometric mean (GM) per 4.9m bottom trawl catch for adult White Perch in Broad Creek, Harris Creek, and Tred Avon River, by sampling year. Black bars indicate the 95% confidence intervals. The y-axes range from 0 to 50; x-axes range are years from 2005 to 2022.

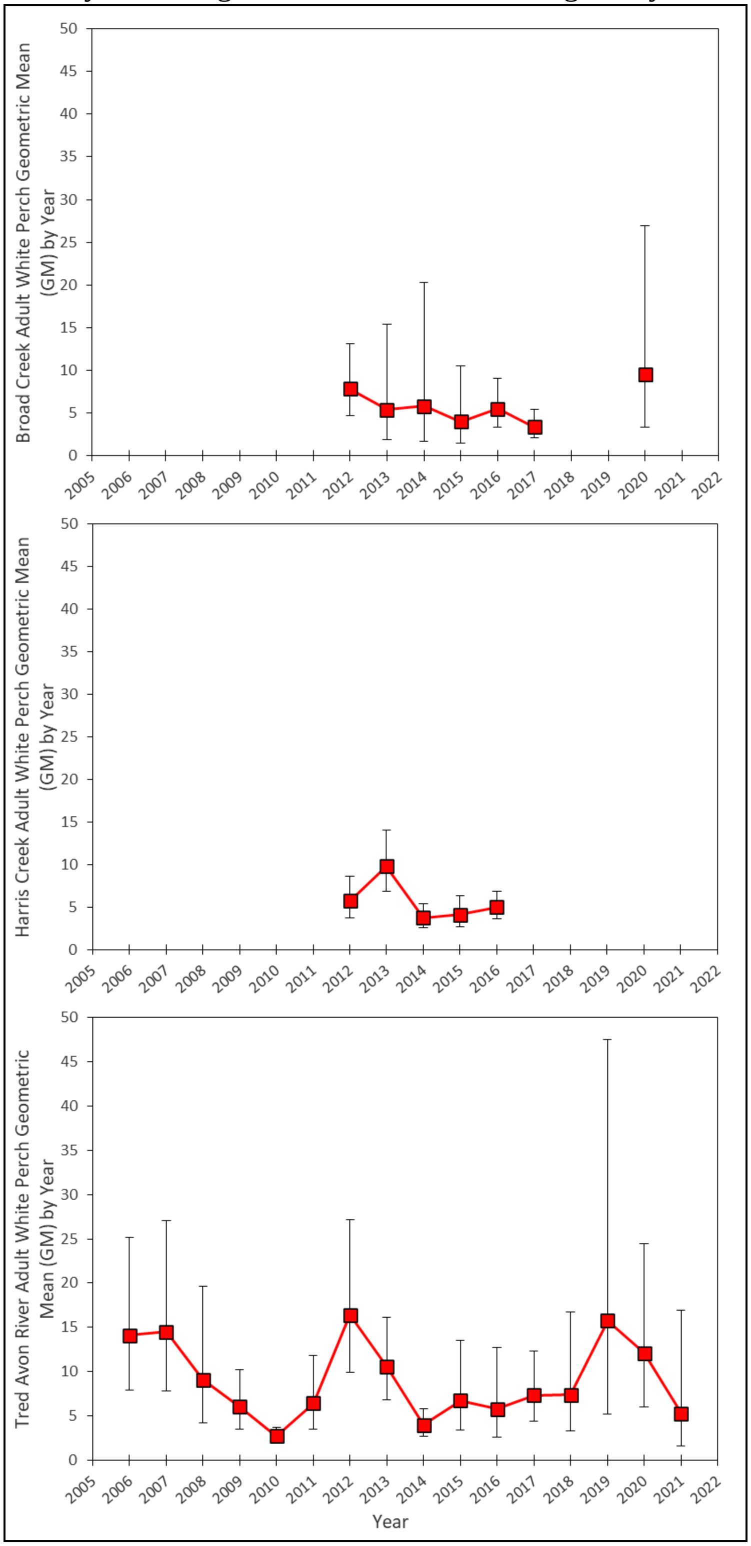


Figure 3-33. Modified proportional stock density (PSD) of White Perch in Choptank River tributaries, Broad Creek, Harris Creek, and Tred Avon River, is the proportion of 4.9m trawl and beach seine catches with quality length or greater White Perch.

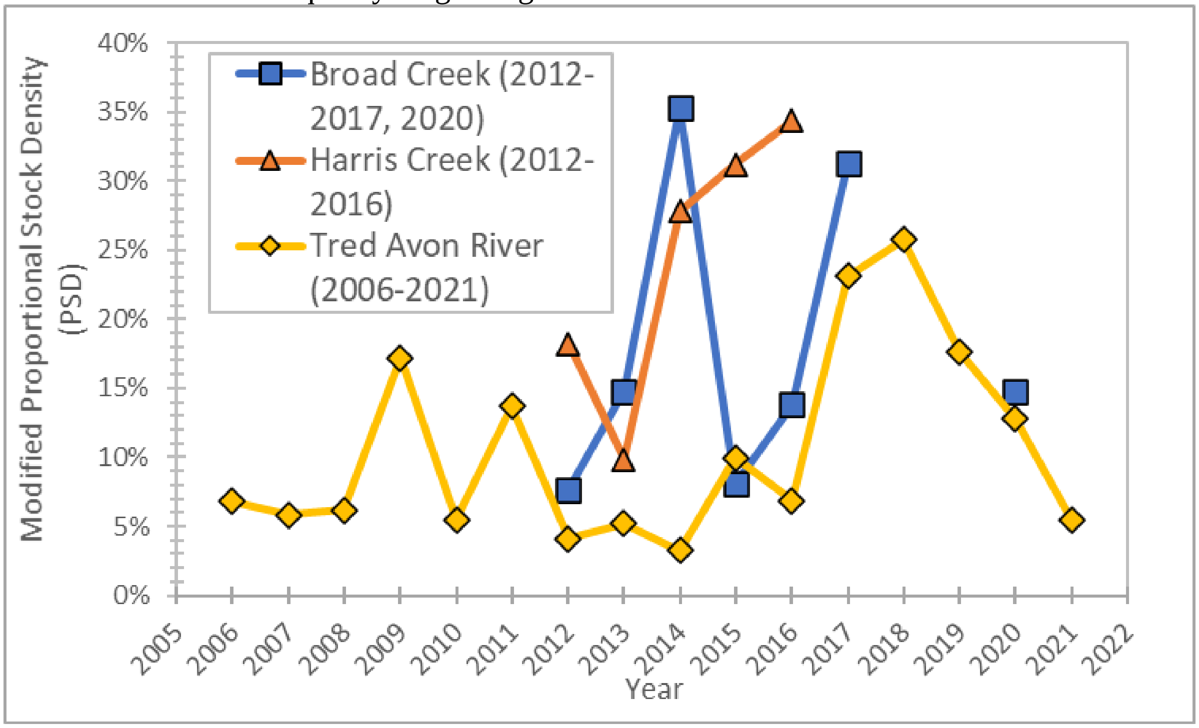


Figure 3-34. Annual beach seine catches geometric mean (GM) per of all finfish species (red squares) for Broad Creek, Harris Creek, and Tred Avon River, by sampling year. Black bars indicate the 95 % confidence intervals. The y-axes range from 0 to 700; x-axes range are years from 2005 to 2022.

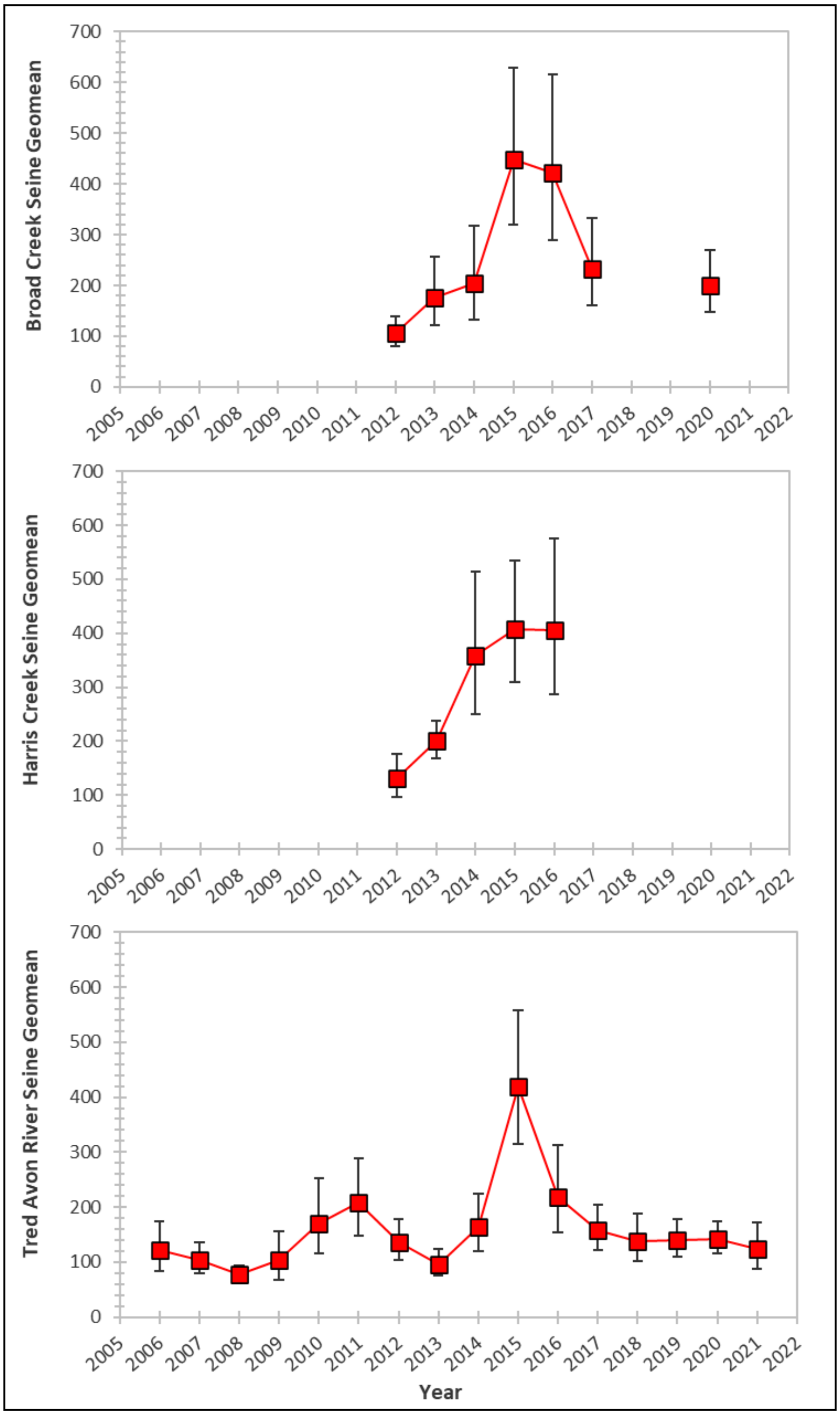


Figure 3-35. Finfish species composition for beach seine catch in Broad Creek, Harris Creek, and Tred Avon River for all sampling years combined. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”.

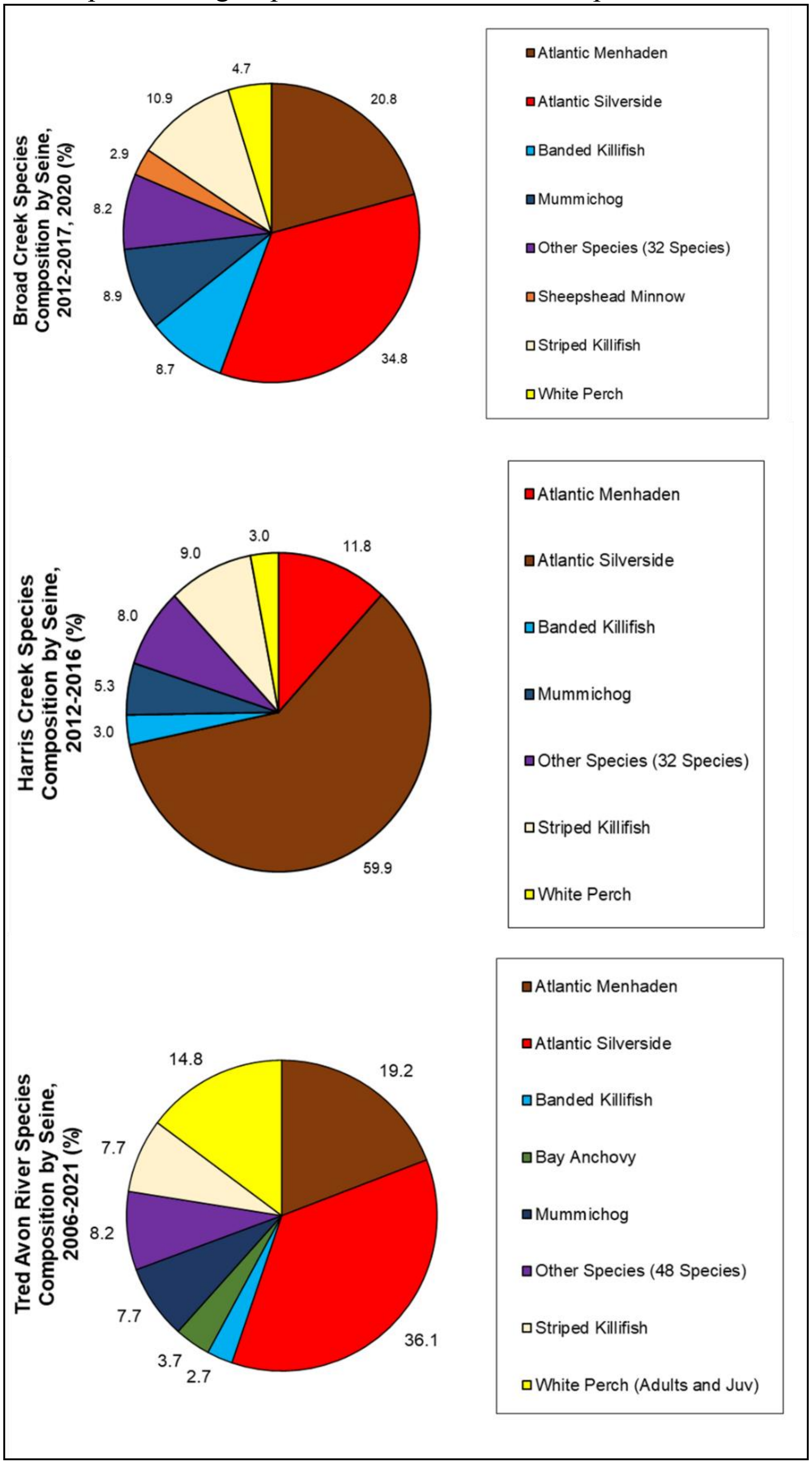


Figure 3-36. Annual 4.m bottom trawl catch geometric mean (GM) per of all finfish species (red squares) for Bohemia, Bush, Gunpowder, Middle, Northeast, and Sassafras Rivers, by sampling year. Black bars indicate the 95% confidence intervals. The y-axes range from 0 to 700; x-axes range are years from 2005 to 2022.

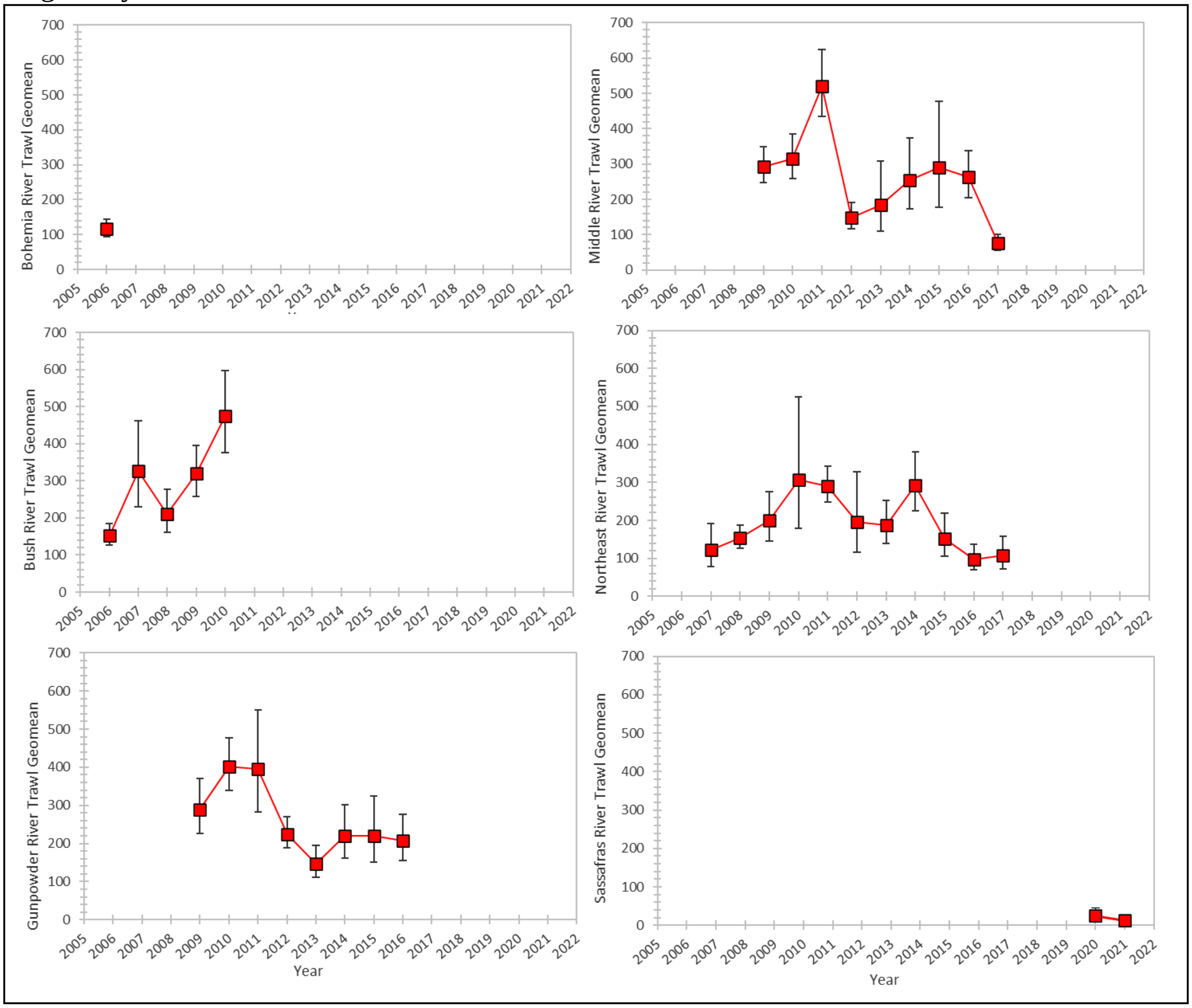


Figure 3-37. Finfish species composition for 4.9m bottom trawl catch in Head-of-Bay subesestuaries for all sampling years combined. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”.

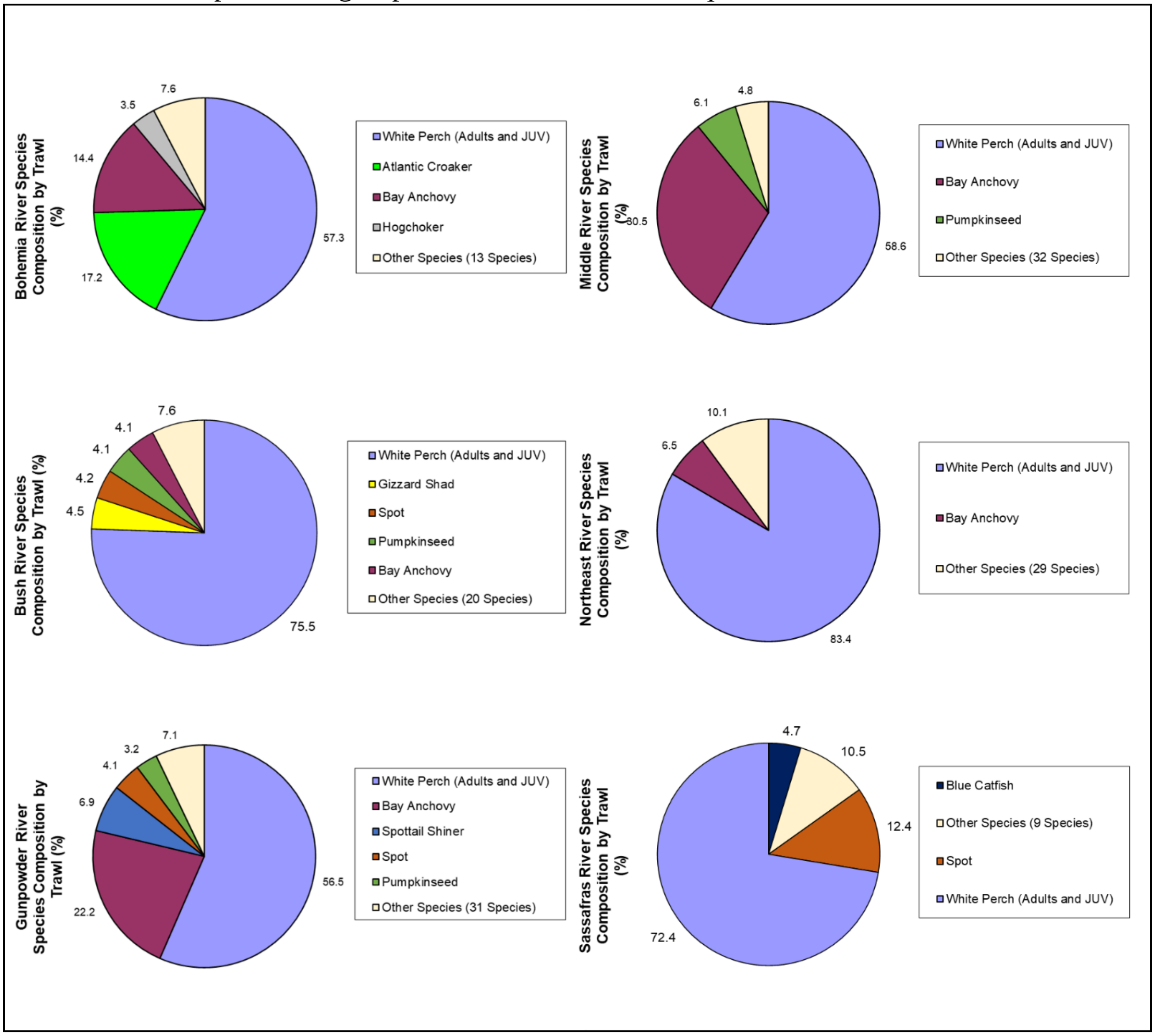


Figure 3-38. Finfish species composition for 4.9m bottom trawl catch in Head-of-Bay subestuaries, by year. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”. The y-axes range from 0 to 100%; x-axes vary based on years systems were sampled.

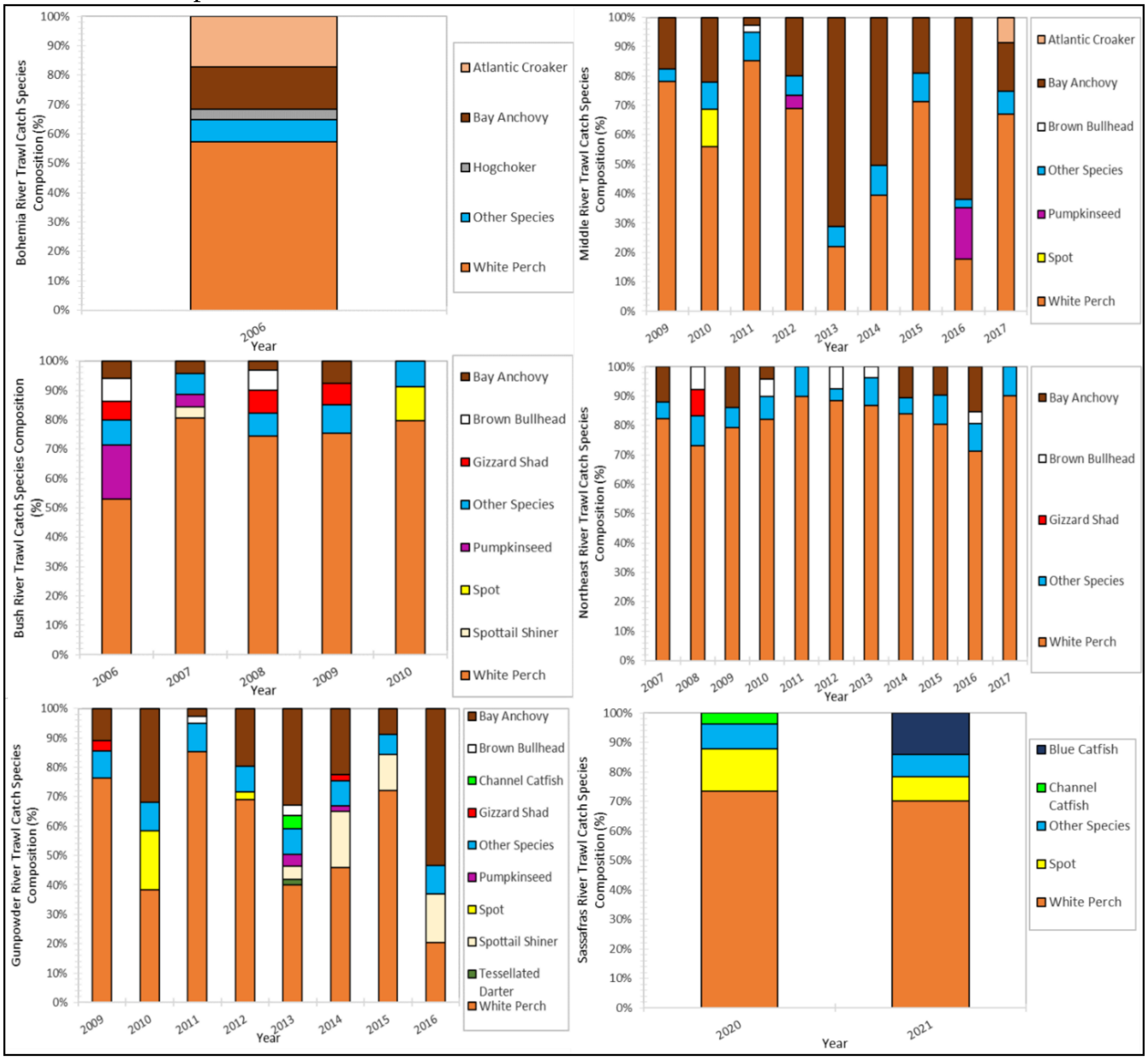


Figure 3-39. Finfish species composition for beach seine catch Sassafras River at FHEP beach seine stations 01 – 04 and at Juvenile Index (JI) – NRMA seine site during 2020 – 2021 for all years combined. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as “other species”.

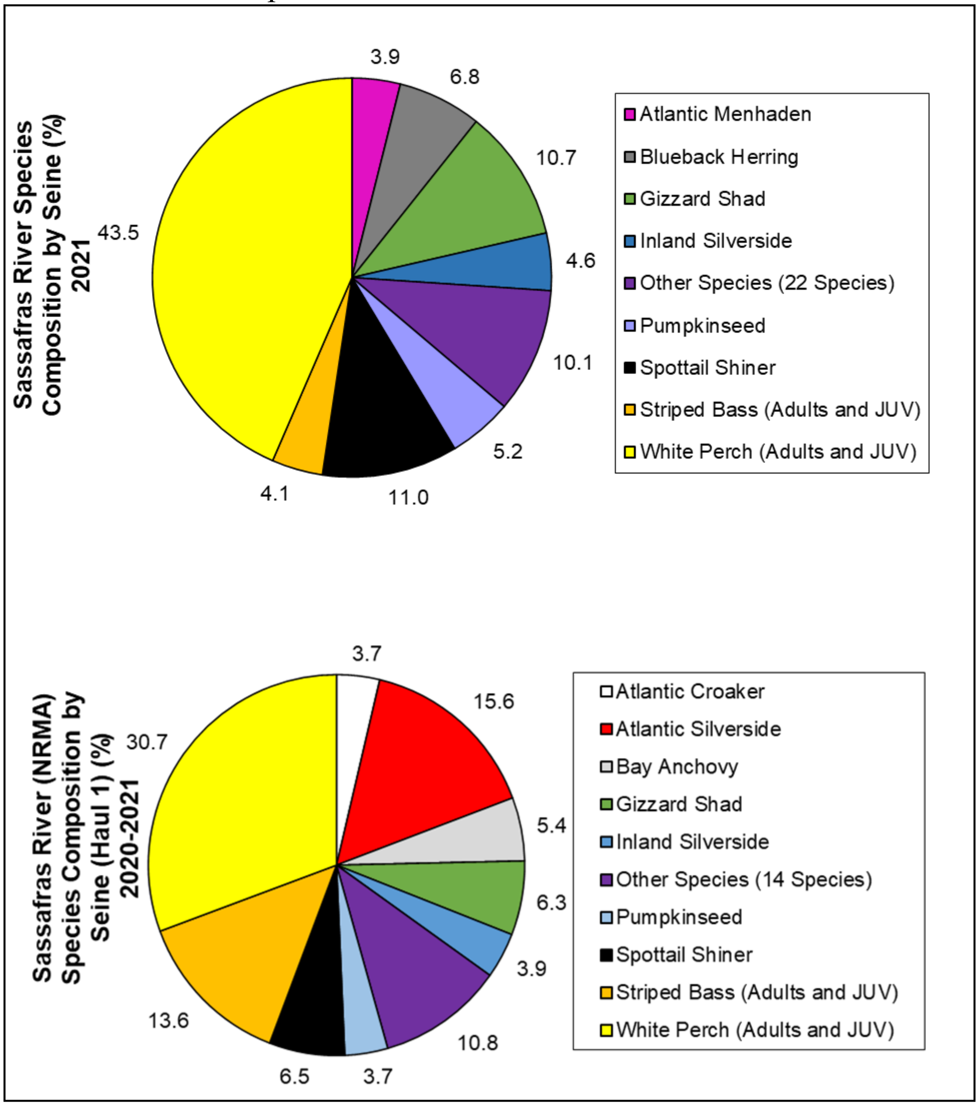
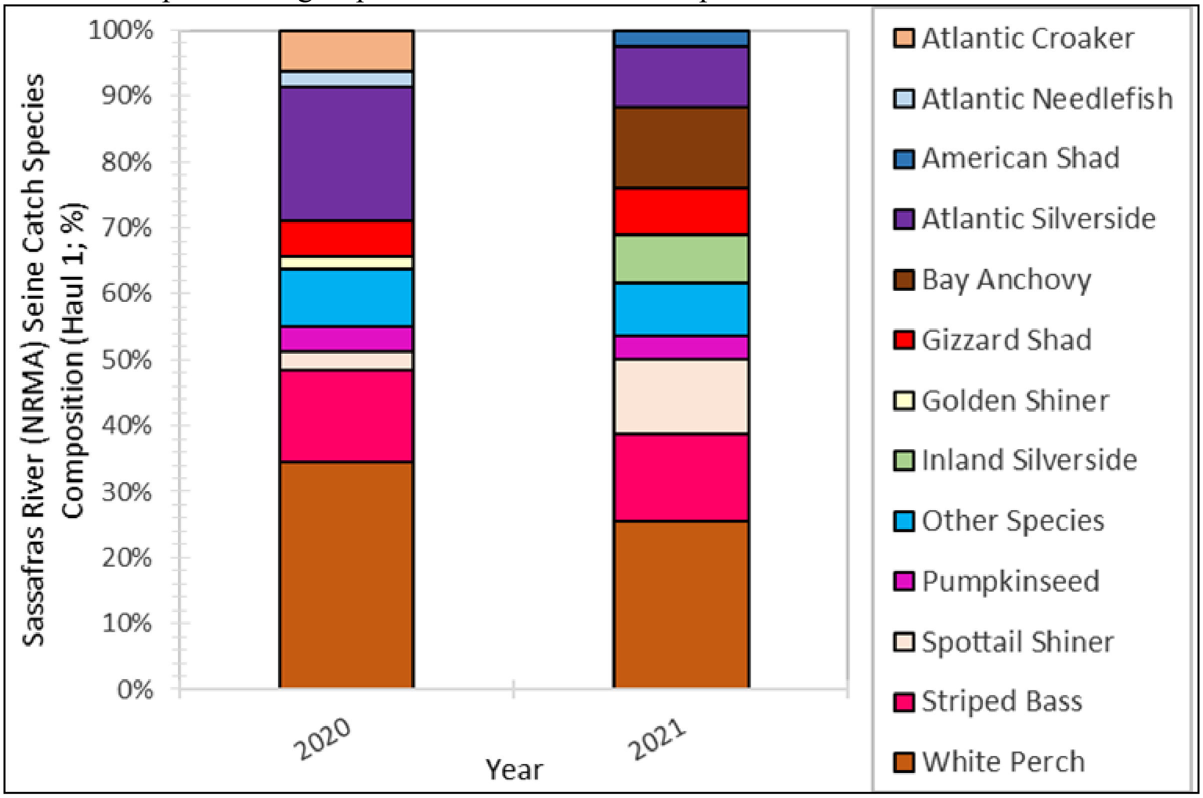


Figure 3-40. Finfish species composition for beach seine in Sassafras River at Juvenile Index (JI) – NRMA seine site during 2020 – 2021. Species that define the top 90% are identified, and the remainder of species are grouped and labeled as "other species".



MD - Marine and estuarine finfish ecological and habitat investigations
Project 2: Support multi-agency efforts to assess finfish habitat and implement ecosystem-based fisheries management.

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

Introduction

Project 2 documents participation by the Fisheries Habitat and Ecosystem Program (FHEP) in habitat, multispecies, and ecosystem-based management forums that relate to recreationally important finfish in Maryland's Chesapeake Bay and Atlantic coast during July 1, 2021 - June 30, 2022. Activities used information generated by F-63 or were consistent with the goals of F-63.

Changes to Project 2 Planned Activities due to Coronavirus Pandemic - Activities under Project 2 were altered at times due to the Pandemic. Virtual meetings and email provided additional opportunities when in-person activities were cancelled.

Fisheries Habitat and Ecosystem Program Website – We continued to update the website with project developments and publications. The website can be found at https://dnr.maryland.gov/fisheries/pages/fhep/index.aspx .

Environmental Review Unit Bibliography Database – We maintain an Environmental Review Unit database, adding additional literature as it becomes available. Older reports that are not in electronic format are scanned and saved. Program staff continue to track research and literature regarding restoration effectiveness.

Review of County Comprehensive Growth Plans – We reviewed the comprehensive plan for Queen Anne's County and provided recommendations consistent with maintaining viable fish habitat. At the urging of engaged citizen groups, Queen Anne's County acknowledged the need to consider impervious surface limits to conserve high quality areas. We commended this strategy and recommended adopting zoning that is consistent with the targets and thresholds. We provided updated impervious surface estimates of Queen Anne's County watersheds for the County's comprehensive growth plan.

We reviewed an amendment to Charles County's comprehensive plan to expand of the industrial park district within Indianhead Airport, providing recommendations consistent with full support of maintaining the Watershed Conservation District where the airport is located. We supported the recommendations found in the Ecosystem-Based Management Plan to conserve the full suite of unique resources found in the Mattawoman Creek watershed, including high quality terrestrial and aquatic habitat.

Cooperative Activities – J. Uphoff continued to work with Resource Assessment Service (RAS) on the impact of dissolved oxygen (DO) and water temperature on resident Striped Bass in summer. Temperature and DO criteria developed under F-63-R-11 are being compared to historic water quality measurements and Chesapeake Bay Program (CBP) projections based on the extent of success in reducing nutrients and climate warming. Progress on results have been presented to the CPB *Ad Hoc* Modeling Workgroup.

J. Uphoff assisted RAS in developing a proposal for the CPB to resuscitate a modest zooplankton monitoring program. Zooplankton was monitored during 1985-2002 and again in 2011. It has been identified repeatedly as a critical need for an ecosystem-based approach to fisheries management. This proposal was circulated with the Bay Program Forage Team for comments. Comments were integrated into the proposal, and it was sent to the Bay Program's Principal's Staff Committee. It has not been funded as of this date.

M. McGinty obtained historical oyster fouling data from MD DNR Shellfish Division for the hard bottom benthic forage index (HBBI). The intent is to examine any gross changes (primarily losses or gains in organisms since this is presence absence data) by updating the HBBI to include recent years of data.

J. Uphoff and M. McGinty responded e Magothy River Association inquiries about protecting Yellow Perch eggs from raccoon predation. M. McGinty assisted the Magothy River Association in interpretation of some of their habitat data.

J. Uphoff and M. McGinty, as members of the steering committee of a Virginia Institute of Marine Science doctoral student, attended several meetings on developing habitat suitability models and habitat suitable indicators for ages 0-4 Striped Bass in Chesapeake Bay. Maps were presented that were based on predictions from hydrodynamic model output such as dissolved oxygen, stratification, current speed, and data on depth and bottom type. J. Uphoff and M. McGinty provided suggestions for refinements that may or may not make predictions more relevant.

J. Uphoff responded to an inquiry from East Carolina University for the North Carolina Division of Marine Fisheries on Maryland's dissolved oxygen attainment criteria for migratory (i.e., anadromous) fish spawning.

J. Uphoff contributed comments to a DNR review of MDE's draft comments regarding the potential impact of discharges from an Atlantic salmon aquaculture facility proposed for Federalsburg. Several potential issues were identified that could impact spawning and habitat of anadromous fishes in Marshyhope Creek.

M. McGinty participated in a meeting to discuss updates of the Green Infrastructure tool. She will work with the team to provide updated anadromous spawning data and other data as needed.

J. Uphoff drafted metrics for a Chesapeake Bay Atlantic Menhaden stoplight index for review and discussion by a Fishing and Boating Services workgroup. This index uses stoplight colors to represent status of available indicators. This index is in response to long wait times for spatial modeling or an aerial survey that would depict status of Atlantic Menhaden in the Bay. Anglers have concerns about Bay Menhaden status and a stoplight index could provide a useful communication tool.

J. Uphoff and M. McGinty met with Alex MacCleod about his PhD research on yellow perch reproductive success in Maryland subestuaries. His work indicated that physical habitat issues (high salinity, sediment, etc.) play a role in addition to endocrine disruptors identified earlier. Alex hopes to publish his work.

M. McGinty provided finalized map updates for an online environmental review GIS tool that has anadromous fish data layers representing historical spawning sites and stream segments where spawning was likely to occur. The maps include stream segments that flow into the segment (contributing to the water quality of the segment) and impervious surface estimates of watersheds at various scales. This tool was built to answer requests from state and federal environmental review units.

J. Uphoff provided comments on a USGS/NOAA proposal to conduct a Patuxent River fish habitat assessment.

J. Uphoff attended an environmental flow workshop for the upper Potomac River (Seneca Breaks to Little Falls).

Presentations and Outreach – Uploads of our 2015 annual report (Federal Aid Grant F63-R, Segment 6, 2015, Marine and Estuarine Finfish Ecological and Habitat Investigations) reached a milestone on ResearchGate of 100 reads.

J. Uphoff, M. McGinty, A. Park, and C. Hoover participated in various webinars, including seminars on sharks, sea level rise, jellyfish, comprehensive planning, salt intrusion, precipitation, toxic contaminants, fisheries engineering and science, stock assessment, and ecosystem-based fisheries management.

J. Uphoff, and A. Park attended the National American Fisheries Society (AFS) meeting held in Baltimore on Nov. 6th – 10th. J. Uphoff presented *What Do Simple Long-Term Egg and Juvenile Indices Say About Chesapeake Bay Striped Bass Productivity?* M. McGinty virtually presented *Applying Impervious Surface Thresholds to Guide Fish Habitat Management.* J. Uphoff moderated two symposia. Papers from the Atlantic coast Striped Bass Symposium, including the paper above, may be published in a themed issue of Marine and Coastal Fisheries. J. Uphoff, M. McGinty, A. Park, and C. Hoover reviewed virtual AFS presentations from the meeting. J. Uphoff and C. Hoover also provided striped bass spawning season temperature and egg abundance / presence data during 1955-2021 to assist an additional presentation given during the American Fisheries meeting in Baltimore.

A. Park presented on the Bush River estuarine fish community sampling conducted by volunteers from 2011 to 2020 for Anita C. Leight Estuary Center (ACLEC) via a webinar. A presentation on ACLEC's data was made annually during the volunteer training workshop. Sampling was conducted by FHEP during 2006-2010 and was turned over to ACLEC volunteers in 2011. However, data collected by ACLEC declined in consistency and accuracy over the years to the point that the data was unacceptable to FHEP staff. Shortage of staff precluded FHEP from committing to training and supervising volunteers.

J. Uphoff described the results of FHEP monitoring of changes in fish habitat in Corsica River to the Corsica Implementers Team (Queen Anne's County).

J. Uphoff assisted a producer with Maryland Public Television in putting together a panel discussion about how development impacts the Chesapeake Bay watershed.

J. Uphoff participated in a three-person panel for *Past, Present and Future of Striped Bass, A Chesapeake Perspective* that was streamed virtually. This forum was produced by the Coastal Conservation Association and Fishtalk Media. It dealt with the past of Striped Bass management and covered habitat and harvest. It drew a record audience (around 325 attended live) for a Fishtalk feature. It was recorded and is available online.

J. Uphoff provided answers to questions about Striped Bass and Atlantic Menhaden in Maryland's portion of Chesapeake Bay forwarded by a legislative librarian. Links to the multitude information in F-63 reports and PDFs of journal articles were provided.

J. Uphoff presented on Maryland's progress in monitoring forage for Striped Bass in Maryland's portion of the Chesapeake Bay for DNR's Bay, Lands, and Tributaries group, the Midshore Anglers' Club, and the Izaak Walton League.

J. Uphoff responded to an inquiry by Office of Communications about whether fish harvest was negatively impacting control of excessive phantom midge hatches in Back River. This has been an ongoing issue and is possibly related to the upgrades of the sewage treatment plant have inadvertently provided a very productive habitat for phantom midges. This high productivity has overwhelmed the ability of the fish population to control the midges.

Chesapeake Bay Program – M. McGinty and A. Park participated in Fish Habitat Action Team meetings and J. Uphoff participated in Forage Action Team meetings. J. Uphoff and M.

McGinty participated in the Sustainable Fisheries Goal Implementation winter and summer meetings.

J. Uphoff and M. McGinty provided comments on Bay Program fisheries research proposals and a climate change document.

J. Uphoff participated in a CBP workshop entitled Rising Bay Water Temperature. Comments on climate warming issues for fisheries were forwarded to the steering committee.

Envision the Choptank – Envision the Choptank (https://www.envisionthechoptank.org/) is a collaboration of conservation organizations, government agencies (town, county, state, and federal), and local citizens that work to maintain and improve the viability of the Choptank River's water quality and natural resources. J. Uphoff and M. McGinty participated in virtual meetings of the Envision the Choptank Working with Local Government workgroup. Engaging in county and town comprehensive plan updates to strengthen natural resource components is a priority of this workgroup. We hope the Envision effort will provide an entry into to more consideration of fish habitat in county planning.

Project 3: Developing Priority Fish Habitat Spatial Tools
Updating the Provisional Index of Hard Bottom Forage Taxa for Recreationally Important Finfish in Maryland's Portion of Chesapeake Bay

Margaret McGinty, Jim Uphoff, and Mitch Tarnowski
Note: Mitch Tarnowski contributed to this project at no cost to the grant.

Abstract

Presence-absence of benthic Oyster community taxa were combined with Spot, Atlantic Croaker, and Striped Bass (target gamefish) diet composition estimates into hard bottom benthic indices (HBBIs) of forage available on Oyster bars in three salinity regions of Maryland's portion of Chesapeake Bay. Declines in Spot, Atlantic Croaker, and Striped Bass HBBI's since 1995 were suggested for high (> 15 ppt) and moderate (10-15 ppt) salinity regions of Chesapeake Bay, but not in the low (< 10 ppt) salinity region. Correlation analysis did not suggest strong, consistent associations of target species' HBBIs with Susquehanna River annual discharge, hypoxic volume, or duration of hypoxia among all salinity regions. When we plotted time-series means for the HBBI and taxa categories for a station with the mean for their salinity category, we saw a general pattern of stations in the eastern shore of the Chesapeake Bay performing better than stations on the western shore. There has been considerable work done to assess forage of soft bottom habitats, but the HBBI is the first assessment of hard bottom communities in Maryland's portion of the Bay.

Introduction

A workshop convened in 2015 to develop an assessment of Chesapeake Bay forage reported a paucity of data for forage on hard bottom habitat (Ihde et al. 2015). Natural hard bottom habitat in Chesapeake Bay would consist of primarily of Oyster bars and some rock substrate. Studies examining the importance of Oyster bars to finfish concluded that this habitat attracts gamefish such as Spot, Atlantic Croaker, and Striped Bass (Breitburg 1999; Harding and Mann 2001). Though not solely dependent on Oyster fouling organisms, these three gamefish opportunistically forage on these highly complex and productive habitats (Harding and Mann 2001).

Concerns about poor catches of Spot and Atlantic Croaker prompted exploration of untapped data on presence-absence of epibenthic taxa (fouling organisms) collected by the Maryland Fall Oyster Survey to develop a hard bottom benthic index (HBBI) as an indicator of benthic forage availability for these two species on Oyster bottom (Uphoff et al. 2019). These HBBIs for Spot and Atlantic Croaker used presence-absence of Oyster fouling taxa weighted by their contribution to diet weight to track changes in the epibenthic community potentially available as forage in Maryland's portion of Chesapeake Bay. Long-term declines were found and a need to explore potential stressors such as hypoxia and flow was identified. This report updates Uphoff et al. (2019), adding three recent years of data to update the HBBI time-series, adding a Striped Bass HBBI, and evaluating the associations of hypoxia and discharge with each species' HBBI.

Methods

We obtained 2019-2021 fouling data from the Maryland Oyster Fall Survey (M. Tarnowski MD DNR, personal communication) to update the time-series; a description of the Maryland Oyster Fall Survey can be found in Tarnowski (2022). The updated dataset covered

1995-2021, a total of 27 years of data for 208 sites (Figure 1). We analyzed data from sites with at least 20 years of data (Table 1). Salinity was a limiting factor in the distribution of most of the taxa we observed and needed to be accounted for (See Table 4 on page 213 in Uphoff et al. 2019 for a list of taxa by salinity limits). We estimated mean salinity by station during 1995-2021 and assigned each station into one of three salinity regions (categories). Stations with mean salinity less than 10 ppt were classified as low salinity; stations with means between 10 and 15 ppt, moderate salinity; and stations with mean salinity greater than 15 ppt, high salinity. Moderate salinity habitat was strongly represented with 123-148 stations per year; low and high salinity stations were moderately represented, 30-32 and 19-31 stations per year, respectively (Table 1).

Ihde et al. (2015) reported Striped Bass, Spot, and Atlantic Croaker diet summaries from a Chesapeake Bay fishery-independent trawl survey conducted during 2002-2012, ChesMMAP (The Chesapeake Bay Multispecies Monitoring and Assessment Program, www.vims.edu/research/departments/fisheries/programs/multispecies_fisheries_research/chesmmap/). Diet items were classified into four general invertebrate prey categories: crustaceans, mollusks, worms, and miscellaneous. These diet groupings accounted for practically all of the weight of the diet for Spot (98.6%) and Atlantic Croaker (98.3%), and a lower percentage for Striped Bass (51.7%). For Spot, crustaceans accounted for 7.6% of diet weight; mollusks, 11.3%; worms, 32.6%; and the miscellaneous category accounted for 47.1%. Crustaceans accounted for 15.4% of diet weight for Atlantic Croaker; mollusks, 14.3%; worms, 41.3%; and the miscellaneous category accounted for 27.3%. Crustaceans accounted for 25.6% of diet weight for Striped Bass; mollusks, 2.6%; worms, 14.4%; and the miscellaneous category accounted for 9.1%. We applied these categories to sort hard bottom taxa to a diet taxa category (Ihde et al. 2015). We examined the individual taxa under each taxa category (crustaceans, mollusks, worms, miscellaneous) and identified those accounting for 90% of presence in each salinity category.

The annual HBBI for each target species and salinity category was estimated by multiplying the mean of the number of taxa for stations in that salinity category by the diet proportion for a species and summing these across diet categories:

(1) $HBBI = [(PC \cdot XC)) + (PM \cdot (XM)) + (PW \cdot (XW)) + (PO \cdot (XO))]$; where

P_ = proportion of diet comprised of a given taxonomic group C (crustacean), M (mollusk), W (worm), or O (other or miscellaneous);

X_ = mean number of taxa of diet category C, M, W, or O present for a given salinity category within a year;

C = crustaceans;

M = mollusks;

W = worms; and

O = miscellaneous or other taxa.

We used Pearson correlations to assess the associations of year, discharge, and hypoxia with each species HBBI and taxa diet groupings. Correlations of $\pm$ 0.50 were considered of interest to management and strong correlations were indicated by $r \geq 0.80$ (Ricker 1975). Bonferonni corrections to levels of significance (P) equivalent to 0.05 and 0.10 were used to judge associations. We used Susquehanna River annual discharge at Conowingo Dam (https://nwis.waterdata.usgs.gov) estimates of annual Chesapeake Bay hypoxic volume (average hypoxic volume in km^3) and annual hypoxia duration (in total number of days) from Virginia Institute of Marine Science (VIMS; https://www.vims.edu/research/products/cbefs/hypoxic-volume/index.php; Table 2).

Finally, we mapped individual station HBBIs over the entire time-series relative to their respective salinity region time-series mean HBBI to visualize whether stations were at or above the time-series means or fell below. This mapping exercise was also conducted for each of the major diet categories.

Results

Twenty-eight taxa were identified and eight taxa were frequently encountered (present in 36% or more of samples; Table 2). This dominant grouping consisted of barnacles and mud crabs (crustaceans); bryozoa, anemone, *Molgula*, and hydrozoa (miscellaneous); *Ishadium* (mollusk); and mud tubes (polychaete worms). Remaining taxa were encountered less than 10% of the time (Table 2). Barnacles, bryozoa, Mud Crab, *Ishadium*, and mud tubes were dominant taxa in all three salinity regions (Table 3). There were 10 dominant taxa in the high salinity region and 8 each in moderate and low salinity regions (Table 3).

After adding data from 2019-2021, we estimated annual HBBIs for each salinity category for Spot (Figure 2; Table 4), Atlantic Croaker (Figure 3; Table 4), and Striped Bass (Figure 4; Table 4), providing time-series spanning 1995-2021. Appendix 1 provides an estimate of the annual mean of the number of taxa for stations in a salinity category prior to application of target species diet weight percentages.

There were three basic time trends suggested by correlation analysis of species' HBBIs by salinity category. There were nine comparisons and the adjusted P for $P \leq 0.05$ was 0.0056 and for $P < 0.10$ it was 0.011. Moderate declines were indicated for all three species in the high salinity region and Striped Bass in the medium salinity category ($r = -0.53$ to -0.62, adjusted $P < 0.05$; Table 5). A decline may have been indicated in medium salinity waters by borderline correlations for Atlantic Croaker and Spot ($r = -0.49$ and -0.50, respectively; adjusted $P < 0.10$). Change over time was not suggested for all three target species' HBBIs in low salinity waters; correlations ($r = -0.22$ to -0.32) were too low to be of interest (Table 5).

The three species-specific HBBIs were strongly correlated with one another within all three salinity categories (Table 5). There were 16 comparisons and the adjusted P for $P < 0.05$ was 0.0012 and for $P < 0.10$ it was 0.0063. Correlations among species were strongest in the high salinity region ($r = 0.97$-0.98, adjusted $P < 0.05$), followed by mid-salinity ($r = 0.91$-0.98, adjusted $P < 0.05$), and low salinity ($r = 0.84$-0.93, adjusted $P < 0.05$). Adjacent salinity categories were moderately to strongly correlated among all three species. Correlations among species between high and medium salinity categories were all considered moderate and ranged from 0.61-0.72 and were significant at an adjusted $P < 0.05$. Correlations among species between low and medium salinity categories were considered moderate to strong and ranged from 0.71-0.81 and were significant at an adjusted $P < 0.05$. Correlations among target species between high and low salinity categories were poor to moderate and ranged from 0.42-0.53 and some were significant at an adjusted $P < 0.10$ (Table 5).

Discharge and hypoxia metrics were strongly associated: hypoxic volume and discharge $r = 0.88$, $P < 0.0001$; hypoxic duration and discharge $r = 0.82$, $P < 0.0001$; and hypoxic volume and duration $r = 0.83$, $P < 0.0001$). All three species' high salinity HBBIs were poorly correlated with discharge, volume of hypoxia, or extent of hypoxia (Table 6). The Striped Bass HBBI for moderate salinity waters was moderately and negatively correlated with discharge, but poorly correlated with measures of hypoxia. Spot and Atlantic Croaker HBBIs for moderate salinity waters were moderately and negatively correlated with discharge, hypoxic volume, and hypoxic duration. In low salinity waters, all three species indices exhibited moderate negative correlations with discharge and both hypoxia metrics (Table 6).

When we examined taxa groupings by salinity, crustaceans exhibited moderate declines over time in moderate and low salinity regions (Table 7). Mollusks exhibited a moderate increasing trend in low salinity areas. Taxa within the miscellaneous category exhibited moderate negative correlations with discharge, hypoxic volume, and extent of hypoxia in the moderate and low salinity regions. Remaining taxa were weakly correlated with discharge and hypoxia metrics (Table 7).

When Spot (Figure 5), Atlantic Croaker (Figure 6), and Striped Bass station HBBIs (Figure 7) were compared to the time-series average for each salinity category, western shore and mid-Bay stations generally fell below the time-series mean, while eastern shore stations were usually at or above the time-series mean. Crustaceans followed a similar pattern to the HBBI; eastern shore stations appeared to meet or exceed the time-series average more frequently than the western shore (Figure 8). Mollusks were different, with the upper stations in the mainstem of the Bay and Potomac River showing a higher frequency of meeting or exceeding the time-series mean (Figure 9). Stations that met or exceeded the time-series average for worms appeared to be more evenly dispersed with clusters of stations below the average, particularly in the upper main Bay (Figure 10). Stations with miscellaneous taxa meeting or exceeding the time-series mean tend to be more prevalent on the eastern shore than the western shore with the exception of the lower Patuxent River (Figure 11).

Discussion

Presence-absence of benthic Oyster community taxa were combined with target gamefish diet composition estimates into HBBIs of forage available on hard bottom habitat (Oyster bars) in three salinity regions of Maryland’s portion of Chesapeake Bay. Declines in Spot, Atlantic Croaker, and Striped Bass HBBI’s since 1995 were suggested for high and moderate salinity regions of Chesapeake Bay, but not in the low salinity region. While correlation analyses indicated declines in Spot, Atlantic Croaker, and Striped Bass HBBI’s in the high salinity region, we did not find associations of discharge or hypoxia indicators with HBBIs in that region strong enough to be of interest. Examination of maps of the extent of hypoxic waters in the Bay indicated that high salinity sites did not routinely overlap hypoxic habitat. Discharge was modestly and negatively associated with Striped Bass HBBIs in moderate and low salinity waters, but hypoxia indicators were only associated with the Striped Bass HBBI in low salinity waters. Correlations of discharge or the two hypoxia indicators were moderate and negative for low and moderate salinity regions.

The strong correlations among discharge, hypoxic volume, and duration of hypoxia make it difficult to sort out specifically which factor or factors have the potential to influence HBBIs. Variations in river discharge to the Chesapeake Bay set up stratification, drive estuarine circulation, and cause fluctuations in inputs of freshwater, sediments, and nutrients - processes that greatly influence hypoxia (Hagy et al. 2004; Kemp et al. 2005; Maryland Sea Grant 2009). Invertebrate forage indices were related (in taxon-specific ways) to freshwater discharge and winter–spring chlorophyll concentration, and three summer water quality variables: dissolved oxygen, salinity, and water temperature (Woodland et al. 2021). Additional factors may influence changes in HBBIs. Woodland et al. (2021) found a negative relationship between annual abundance indices of invertebrate forage taxa and intensity of spring warming. The Atlantic multidecadal Oscillation, a climate index, was also identified as an influence (Woodland et al. 2021). It may be possible to compare changes in Oyster abundance and harvest in the 2020

Oyster stock assessment update covering 1999-2019 (MD DNR 2020) with HBBI's to evaluate the possibility that substrate disturbance from exploitation is a factor. However, based on past harvests, Oysters abundance during the periods available for estimating HBBIs and covered by the stock assessment would have been much lower than in the past.

A soft bottom benthic index (i.e., an index of invertebrates living in the sediment) covering 1995-2021 has been developed from a component of a Chesapeake Bay benthic index of biotic integrity (BIBI; Uphoff et al. 2022); the BIBI provides an accessible summary of benthic habitat status in relation to water quality (Weisburg et al. 1997; Versar Inc 2022; Uphoff et al. 2022; see Project 4 as well). This soft bottom benthic invertebrate biomass index for Maryland tidal waters (grams / m^2 of benthic invertebrates) has been used as a Striped Bass benthic forage index (Uphoff et al. 2022; see Project 4). Benthic biomass in Maryland's portion of the Bay has generally been lower since 2010, similar to changes in HBBIs in high and moderate salinity regions for all three species (Uphoff et al. 2022). Moderate relationships were indicated between the soft bottom biomass index and HBBIs for Atlantic Croaker and Spot (Uphoff et al. 2019). However, there was little indication of correspondence of the soft bottom benthic index to condition of Striped Bass (Uphoff et al. 2022). Invertebrates are generally important to smaller Striped Bass (< 300 mm TL) in spring, while polychaetes contributed to the production of larger Striped Bass in spring and summer (Hartman and Brandt 1995; Overton et al. 2009).

When we plotted comparisons of time-series means for target species' HBBIs for a station with the mean for their salinity category, we saw a general pattern of stations in the eastern shore of the Bay performing better than stations on the western shore. Taxa means for crustaceans and miscellaneous fouling organisms followed this pattern as well, but worms and mollusks were somewhat different. This east-west pattern may indicate greater habitat stress on the western side of the Bay or that the eastern shore is naturally more productive than the western shore. Some species of worms resist hypoxic conditions or quickly repopulate when the stress subsides. Jewett et al. (2005) reported an increase in Serpulids in response to increased hypoxia.

The diet information we used to classify taxa and estimate diet contribution for these species were adopted from Ihde et al. (2015) and based on fish sampled by trawl that could have limited ability to sample rough, hard bottom. Simonsen and Cowan (2013) found diet variations in Atlantic Croaker in comparisons of restored Oyster Reef to adjacent mud habitat. Predators caught in trawls relied more heavily on invertebrate prey because they were generally smaller than those captured by gill nets in Pamlico Sound, North Carolina (Binion-Rock et al. 2019). Diet samples may be biased by the location within the water column where the sample was taken and ontogenetic changes in diet composition (Binion-Rock et al. 2019). However, strong correlations of HBBIs among the three species within each salinity region indicated the diet proportions were not having a major effect on HBBI trends.

We attempted to mine historical data from 1977 to establish a reference point to compare present conditions. However, after entering the data, we found there were few fall survey sites that could be compared to present data. Most of the stations were in higher salinity areas and few stations that matched present data. There is potential to examine regional change in taxa presence in high salinity areas.

Stakeholders have asked for explanations of what may be driving some recreational fishery issues in the Bay such as the "disappearance" of Striped Bass from the lower Bay during summer while the upper Bay fishery remained robust. These issues cannot be entirely addressed through single species stock assessment and a broader set of hypotheses should be addressed. Angler concerns regarding reduced catch of Striped Bass in the lower Potomac River and lower Chesapeake Bay prompted Fishing and Boating Services, in concert with DNR's Resource Assessment Service, to apply an ecosystem approach to examine potential causes of declining catch rates (i.e., consider water quality, temperature, soft bottom benthic forage, etc.). They investigated water quality and benthic infauna data to determine if these parameters could explain declines in Striped Bass catch rates in the lower Potomac River (Uphoff et al. 2016). This investigation found declining dissolved oxygen and polychaete abundance were concurrent with declining presence of Striped Bass in the lower Bay (Uphoff et al. 2019).

The pattern of HBBI declines among salinity regions (declines suggested for high and moderate salinity regions and not for low salinity) correspond to the perceived movements of Striped Bass that were of concern. This general correspondence should not be interpreted as cause and effect based on correlations alone. 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).

Stakeholders and Maryland's fishery managers want to know whether there is enough forage to support gamefish in Maryland's portion of the Bay. Development of HBBIs may help address this question. We believe that this work demonstrates the value in mining presence-absence data from the fall Oyster survey to examine hard bottom forage conditions. There has been considerable work done to assess forage of soft bottom habitats, but the HBBI is the first assessment of hard bottom communities in Maryland's portion of the Bay.

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Table 1. Number of samples by year at sites with at least twenty years of data.

Year	N	High Salinity	Moderate Salinty	Low Salinity
1995	192	30	130	32
1996	182	26	126	30
1997	175	22	123	30
1998	187	24	132	31
1999	206	31	143	32
2000	201	30	140	31
2001	197	28	138	31
2002	209	29	148	32
2003	197	27	138	32
2004	206	29	145	32
2005	205	29	144	32
2006	205	29	144	32
2007	209	30	147	32
2008	208	30	146	32
2009	209	30	147	32
2010	189	19	138	32
2011	209	30	147	32
2012	204	29	144	31
2013	205	30	144	31
2014	206	30	144	32
2015	194	28	135	31
2016	204	30	143	31
2017	203	30	141	32
2018	202	28	142	32
2019	206	30	144	32
2020	203	30	143	30
2021	201	29	142	30

Table 2. Susquehanna River discharge (cubic feet per second or cfs), duration of hypoxia (days), and hypoxic volume (km^3) used in correlation analyses.

Year	Susquhanna R. Discharge (cfs)	Hypoxia duration (days)	Hypoxic volume km^3
1995	27,860	90	3.3
1996	51,870	133	9.7
1997	42,910	133	6.8
1998	46,420	123	9.4
1999	22,850	57	1.9
2000	35,580	115	5.5
2001	24,470	102	3.5
2002	28,210	76	1.9
2003	51,120	146	10.7
2004	66,560	154	9.5
2005	50,890	121	8.8
2006	46,630	131	5.6
2007	40,390	116	5.0
2008	40,850	106	6.6
2009	31,150	96	3.9
2010	34,030	122	5.4
2011	65,730	147	8.9
2012	40,920	130	4.8
2013	34,750	92	3.5
2014	36,510	117	5.9
2015	30,190	92	4.2
2016	24,480	90	3.8
2017	39,620	96	5.3
2018	55,110	137	7.1
2019	62,040	131	9.8
2020	37,900	95	5.0
2021	38,960	141	6.6

Table 3. Proportion of samples where each taxa was observed and its assigned diet category. All salinity regions and years were combined.

Taxa	Proportion Present	Diet Category
Barnacle	0.955	Crustacean
Mud Crab	0.686	Crustacean
Bryozoa	0.878	Miscellaneous
Anemone	0.560	Miscellaneous
Molgula	0.378	Miscellaneous
Hydrozoa	0.092	Miscellaneous
Boring Sponge	0.077	Miscellaneous
Other Sponge	0.034	Miscellaneous
Microcionia	0.021	Miscellaneous
Lissosedrix	0.001	Miscellaneous
Ischadium	0.834	Mollusks
Mytilopsis	0.086	Mollusks
Crepidula	0.067	Mollusks
Mytilis	0.035	Mollusks
Mercenaria	0.007	Mollusks
Anomia	0.006	Mollusks
Geukensia	0.006	Mollusks
Rangia	0.006	Mollusks
Mya	0.004	Mollusks
Mud Snail	0.004	Mollusks
Mulinia	0.004	Mollusks
Macoma	0.003	Mollusks
Urosalpinx	0.001	Mollusks
Petricola	0.001	Mollusks
Mud Tube	0.583	Worm (polychaete)
Serpulids	0.063	Worm (polychaete)
Stylochus	0.047	Worm (flatworm)
Sabellaria	0.042	Worm (polychaete)

Table 4. Taxa representing 90% presence by salinity zones in order of highest to lowest.

High Salinity	Moderate Salinity	Low Salinity
Barnacles	Barnacles	Barnacles
Bryozoan	Bryozoan	Ischadium
Molgula	Ischadium	Bryozoan
Mud Crab	MudCrab	MudCrab
Anemone	Mud Tube	Mytilopsis
Ischadium	Anemone	Mud Tube
Mud Tubes	Molgula	Anemone
Crepidula		
Serpuilids		
Boring Sponge		

Table 5. Annual Hard Bottom Index (HBBI) for Striped Bass, Spot and Atlantic Croaker, by salinity category.

	High Salinity (>15ppt)			Moderate Salinity (10-15ppt)			Low Salinity (<10 ppt)		
Year	Striped Bass	Spot	Atlantic Croaker	Striped Bass	Spot	Atlantic Croaker	Striped Bass	Spot	Atlantic Croaker
1995	1.09	2.72	2.25	0.85	1.87	1.55	0.74	1.29	1.20
1996	0.80	1.63	1.38	0.69	1.17	1.01	0.62	0.95	0.87
1997	0.86	2.03	1.64	0.73	1.37	1.21	0.70	1.08	1.01
1998	0.94	2.17	1.79	0.84	1.60	1.42	0.79	1.28	1.21
1999	0.96	2.21	1.85	0.87	1.76	1.52	0.77	1.34	1.20
2000	0.95	2.11	1.81	0.82	1.53	1.39	0.75	1.26	1.21
2001	0.85	1.93	1.65	0.88	1.85	1.54	0.76	1.29	1.15
2002	0.84	1.80	1.57	0.86	1.78	1.49	0.76	1.38	1.25
2003	0.94	2.07	1.76	0.73	1.29	1.18	0.70	1.17	1.10
2004	1.00	2.24	1.91	0.69	1.24	1.11	0.61	1.11	1.03
2005	0.85	1.96	1.64	0.70	1.38	1.24	0.60	1.01	0.96
2006	0.81	1.88	1.55	0.78	1.58	1.38	0.71	1.26	1.19
2007	0.97	2.25	1.92	0.85	1.66	1.44	0.76	1.28	1.26
2008	0.77	1.72	1.50	0.73	1.49	1.31	0.68	1.24	1.18
2009	0.87	1.99	1.72	0.67	1.44	1.21	0.70	1.20	1.16
2010	0.77	1.69	1.48	0.73	1.47	1.31	0.65	1.23	1.11
2011	0.67	1.27	1.13	0.57	0.95	0.85	0.61	0.94	0.89
2012	0.64	1.27	1.04	0.61	1.08	0.94	0.61	1.07	0.93
2013	0.80	1.73	1.40	0.71	1.37	1.23	0.76	1.40	1.34
2014	0.75	1.79	1.43	0.61	1.24	1.04	0.65	1.07	0.97
2015	0.76	1.64	1.39	0.70	1.47	1.22	0.69	1.27	1.11
2016	0.80	1.87	1.55	0.71	1.56	1.29	0.70	1.32	1.12
2017	0.90	2.05	1.72	0.73	1.54	1.31	0.78	1.34	1.26
2018	0.81	1.80	1.52	0.65	1.18	1.05	0.59	0.88	0.90
2019	0.72	1.64	1.34	0.64	1.04	0.98	0.66	0.96	0.98
2020	0.78	1.63	1.35	0.66	1.11	1.05	0.65	0.91	0.92
2021	0.82	1.84	1.55	0.71	1.30	1.22	0.69	1.08	1.08

Table 6. Correlations of salinity region HBBIs of Striped Bass, Spot, and Atlantic Croaker with year and among target species' salinity region HBBIs. Bold indicates significance at a Bonferroni method adjusted P ≤ 0.05.

				High salinity			Moderate salinity			Low salinity	
Salinity	Species	Statistic	Year	Striped Bass	Spot	Atlantic Croaker	Striped Bass	Spot	Atlantic Croaker	Striped Bass	Spot
High	Striped Bass	r	**-0.578**								
		P	**0.002**								
High	Spot	r	**-0.526**	**0.967**							
		P	**0.005**	**<.0001**							
High	Atlantic Croaker	r	**-0.545**	**0.977**	**0.988**						
		P	**0.003**	**<.0001**	**<.0001**						
Moderate	Striped Bass	r	**-0.619**	**0.681**	**0.677**	**0.701**					
		P	**0.001**	**<.0001**	**0.000**	**<.0001**					
Moderate	Spot	r	-0.498	**0.614**	**0.663**	**0.685**	**0.911**				
		P	0.008	**0.001**	**0.000**	**<.0001**	**<.0001**				
Moderate	Atlantic Croaker	r	-0.494	**0.655**	**0.693**	**0.716**	**0.951**	**0.978**			
		P	0.009	**0.000**	**<.0001**	**<.0001**	**<.0001**	**<.0001**			
Low	Striped Bass	r	-0.319	0.516	0.531	0.524	**0.787**	**0.774**	**0.802**		
		P	0.105	0.006	0.004	0.005	**<.0001**	**<.0001**	**<.0001**		
Low	Spot	r	-0.289	0.421	0.443	0.458	**0.707**	**0.829**	**0.811**	**0.837**	
		P	0.144	0.029	0.021	0.016	**<.0001**	**<.0001**	**<.0001**	**<.0001**	
Low	Atlantic Croaker	r	-0.217	0.500	0.516	0.532	**0.709**	**0.773**	**0.803**	**0.890**	**0.935**
		P	0.278	0.008	0.006	0.004	**<.0001**	**<.0001**	**<.0001**	**<.0001**	**<.0001**

Table 7. Results of Pearson correlation analysis to examine associations of discharge (cfs) and hypoxia (volume as km^3 and duration in number of days) on HBBI by salinity class. Bold indicates significance at a Bonferroni method adjusted P ≤ 0.05.

		High Salinity			Moderate Salinity			Low Salinity		
Variable	Statistic	HBBI Striped Bass	HBBI Spot	HBBI Atlantic Croaker	HBBI Striped Bass	HBBI Spot	HBBI Atlantic Croaker	HBBI Striped Bass	HBBI Spot	HBBI Atlantic Croaker
Discharge	r	-0.18	-0.24	-0.24	**-0.54**	**-0.71**	**-0.66**	**-0.59**	**-0.68**	**-0.55**
	P	0.3792	0.2210	0.2314	**0.0037**	**<.0001**	**0.0002**	**0.0011**	**0.0001**	**0.0028**
Hypoxic Volume	r	-0.08	-0.13	-0.13	-0.43	**-0.62**	**-0.55**	**-0.50**	**-0.62**	**-0.53**
	P	0.6766	0.5034	0.5077	0.0248	**0.0005**	**0.0028**	**0.0085**	**0.0006**	**0.0045**
Hypoxic Duration	r	-0.19	-0.23	-0.22	-0.48	**-0.64**	**-0.58**	**-0.59**	**-0.63**	**-0.55**
	P	0.3401	0.2517	0.2634	0.0115	**0.0003**	**0.0014**	**0.0012**	**0.0005**	**0.0030**

Table 8. Results of Pearson correlation analysis to examine associations of year, discharge (cfs), and hypoxia (volume in km^3 and duration in number of days) on taxa groupings by salinity class. Bold indicates significance at a Bonferroni method adjusted $P \leq 0.05$.

Variable	Statistic	Crustaceans	Worms	Mollusks	Miscellaneous
		High Salinity			
Year	r	**-0.51**	-0.45	-0.38	-0.47
	p	**0.0017**	0.0045	0.0120	0.0032
Discharge	r	0.0463	-0.2155	-0.2004	-0.2275
	p	0.2046	0.0701	0.0791	0.0635
Hypoxic Volume	r	0.06	-0.10	-0.24	-0.12
	p	0.1951	0.1534	0.0592	0.1365
Hypoxic Duration	r	-0.05	-0.18	-0.19	-0.22
	p	0.1996	0.0937	0.0832	0.0677
		Moderate Salinity			
Year	r	**-0.67**	-0.23	0.07	-0.50
	p	**<0.0001**	0.0635	0.1828	0.0021
Discharge	r	-0.13	-0.41	0.07	**-0.73**
	p	0.1334	0.0081	0.1842	**<.0001**
Hypoxic Volume	r	-0.05	-0.31	0.05	**-0.66**
	p	0.2042	0.0300	0.2017	**0.0001**
Hypoxic Duration	r	-0.11	-0.34	0.02	**-0.67**
	p	0.1432	0.0209	0.2332	**<0.0001**
		Low Salinity			
Year	r	-0.22	-0.22	**0.60**	-0.34
	p	0.0702	0.0665	**0.0003**	0.0211
Discharge	r	-0.36	-0.23	0.27	**-0.71**
	p	0.0168	0.0637	0.0454	**<.0001**
Hypoxic Volume	r	-0.22	-0.23	0.09	**-0.61**
	p	0.0696	0.0620	0.1674	**0.0002**
Hypoxic Duration	r	-0.36	-0.27	0.19	**-0.61**
	p	0.0153	0.0448	0.8280	**0.0002**

Appendix 1. Estimates of the annual mean of the number of taxa for stations in a salinity category prior to application of target species diet weight percentages. These means correspond to XC, XM, XW, or XO in equation 1. Appendix 1a = high salinity region; 1b = moderate salinity region; and 1c = low salinity region.

1a.			High Salinity		
YEAR	N	Crustacean	Worm	Mollusk	Miscellaneous
1995	30	1.77	1.70	1.27	4.00
1996	26	1.73	0.81	0.85	2.42
1997	22	1.59	0.95	1.05	3.14
1998	24	1.79	1.00	1.42	3.29
1999	31	1.77	1.19	1.35	3.26
2000	30	1.73	1.43	1.00	2.97
2001	28	1.57	1.14	1.39	2.71
2002	29	1.66	1.10	1.17	2.52
2003	27	1.78	1.33	0.89	2.96
2004	29	1.90	1.34	1.28	3.21
2005	29	1.55	1.21	0.86	2.86
2006	29	1.52	0.93	1.14	2.83
2007	30	1.73	1.40	1.47	3.17
2008	30	1.40	1.17	1.17	2.33
2009	30	1.53	1.43	0.93	2.77
2010	19	1.37	1.37	0.63	2.26
2011	30	1.50	0.67	0.97	1.77
2012	29	1.52	0.28	0.93	2.03
2013	30	1.70	0.60	0.93	2.77
2014	30	1.43	0.70	1.03	2.83
2015	28	1.50	0.89	0.96	2.39
2016	30	1.47	1.03	0.93	2.80
2017	30	1.70	1.13	1.20	3.00
2018	28	1.57	1.00	1.00	2.64
2019	30	1.37	0.80	0.77	2.53
2020	30	1.67	0.73	0.77	2.50
2021	29	1.59	0.93	1.21	2.72

Appendix 1 continued.

1b.		Moderate Salinity			
	N	Crustacean	Worm	Mollusk	Miscellaneous
1995	130	1.73	0.87	0.97	2.86
1996	126	1.83	0.25	0.93	1.79
1997	123	1.67	0.68	1.00	1.92
1998	132	1.84	0.96	0.99	2.20
1999	143	1.85	0.99	0.99	2.51
2000	140	1.80	1.04	0.96	2.01
2001	138	1.89	0.81	1.01	2.82
2002	148	1.86	0.79	1.01	2.70
2003	138	1.76	0.70	1.01	1.72
2004	145	1.63	0.60	1.02	1.70
2005	144	1.48	0.90	0.93	1.85
2006	144	1.65	0.95	0.90	2.21
2007	147	1.86	0.87	1.05	2.37
2008	146	1.51	0.91	0.92	2.08
2009	147	1.37	0.64	0.96	2.17
2010	138	1.49	0.98	0.90	1.99
2011	147	1.46	0.30	0.96	1.35
2012	144	1.56	0.31	0.92	1.61
2013	144	1.53	0.84	1.02	1.84
2014	144	1.41	0.36	1.04	1.91
2015	135	1.53	0.55	0.99	2.26
2016	143	1.45	0.70	0.90	2.38
2017	141	1.49	0.80	0.97	2.23
2018	142	1.55	0.54	0.96	1.65
2019	144	1.61	0.55	1.03	1.33
2020	143	1.62	0.66	1.00	1.40
2021	142	1.57	0.97	1.03	1.58

Appendix 1 continued.

1c.		Low Salinity			
	N	Crustacean	Worm	Mollusk	Miscellaneous
1995	32	1.81	0.53	1.66	1.69
1996	30	1.70	0.30	0.83	1.33
1997	30	1.87	0.40	1.13	1.43
1998	31	2.00	0.68	1.19	1.65
1999	32	1.88	0.63	1.00	1.88
2000	31	1.81	0.84	1.23	1.52
2001	31	1.94	0.45	1.16	1.84
2002	32	1.78	0.75	1.06	1.88
2003	32	1.78	0.53	1.47	1.47
2004	32	1.47	0.44	1.69	1.41
2005	32	1.53	0.41	1.44	1.28
2006	32	1.69	0.69	1.53	1.56
2007	32	1.78	0.88	1.66	1.44
2008	32	1.53	0.84	1.38	1.47
2009	32	1.69	0.66	1.72	1.41
2010	32	1.47	0.63	1.22	1.66
2011	32	1.69	0.16	1.66	1.22
2012	31	1.61	0.10	1.42	1.61
2013	31	1.71	0.90	1.81	1.65
2014	32	1.78	0.06	1.84	1.50
2015	31	1.74	0.19	1.74	1.87
2016	31	1.71	0.29	1.26	2.03
2017	32	1.88	0.78	1.34	1.69
2018	32	1.59	0.41	1.63	0.94
2019	32	1.78	0.41	1.81	1.03
2020	30	1.87	0.27	1.70	1.03
2021	30	1.73	0.63	1.60	1.20

Figure 1. Sites used to assess HBBI indices by salinity classification: high > 15ppt, moderate = 10-15 ppt, and low < 10ppt.

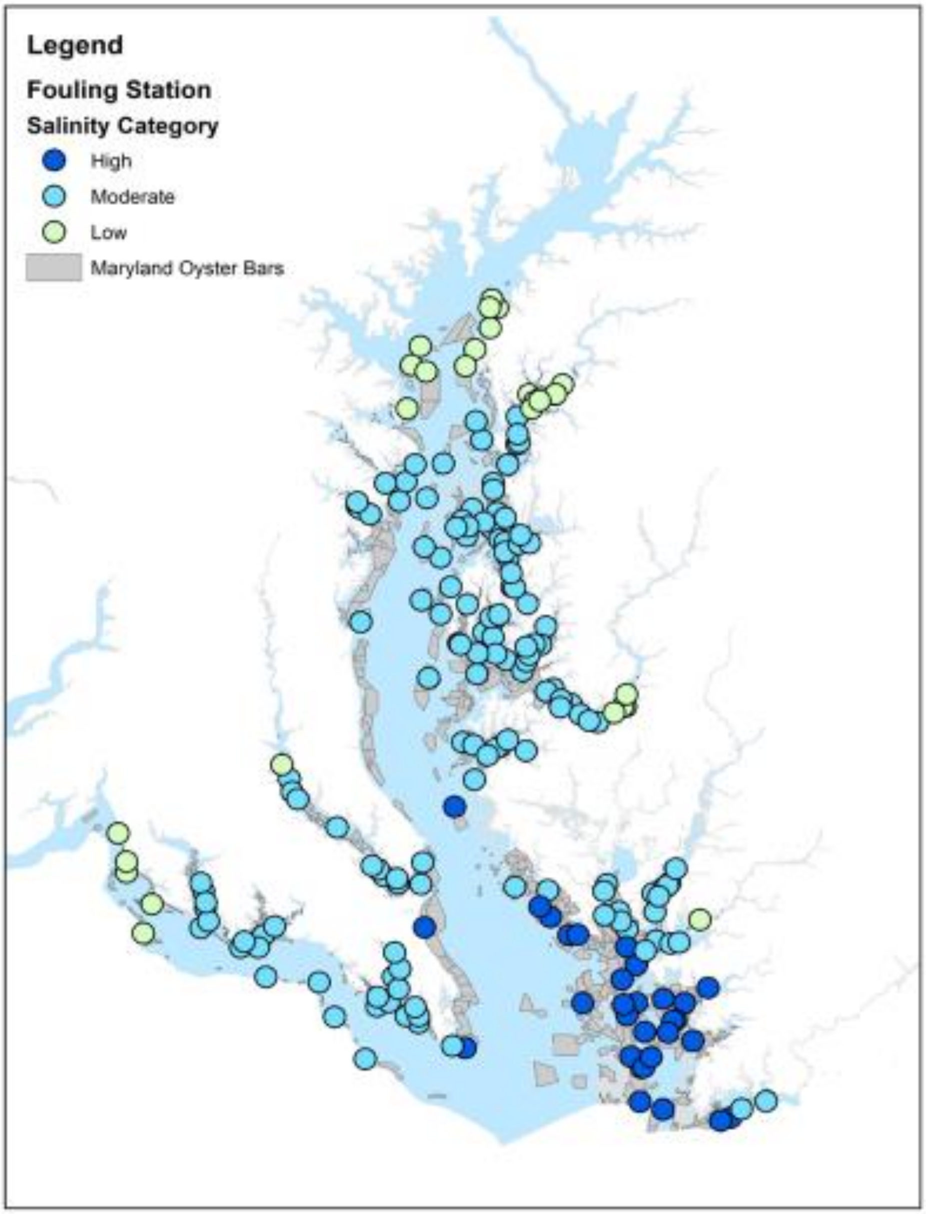


Figure 2. Spot hard bottom benthic indices (HBBI) by salinity region.

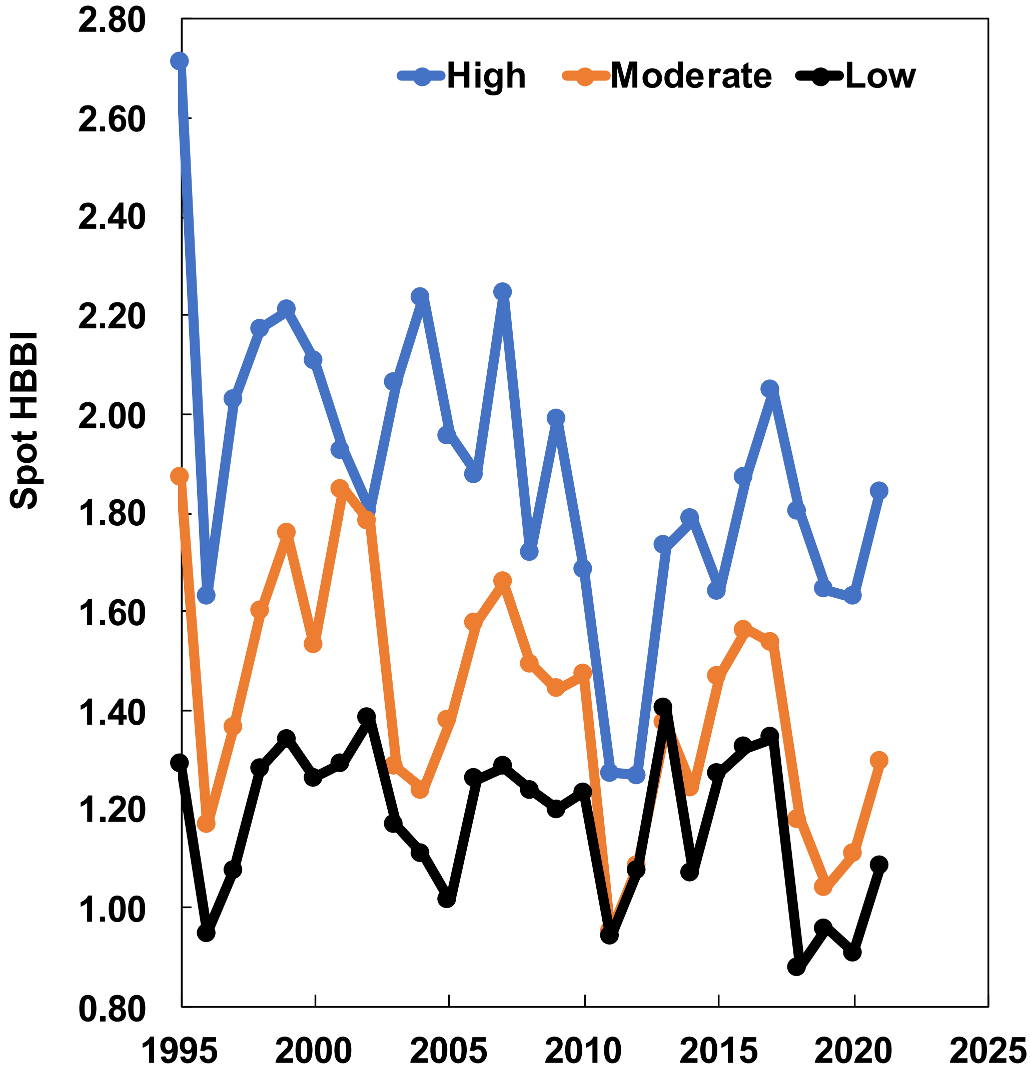


Figure 3. Atlantic Croaker hard bottom benthic indices (HBBI) by salinity region.

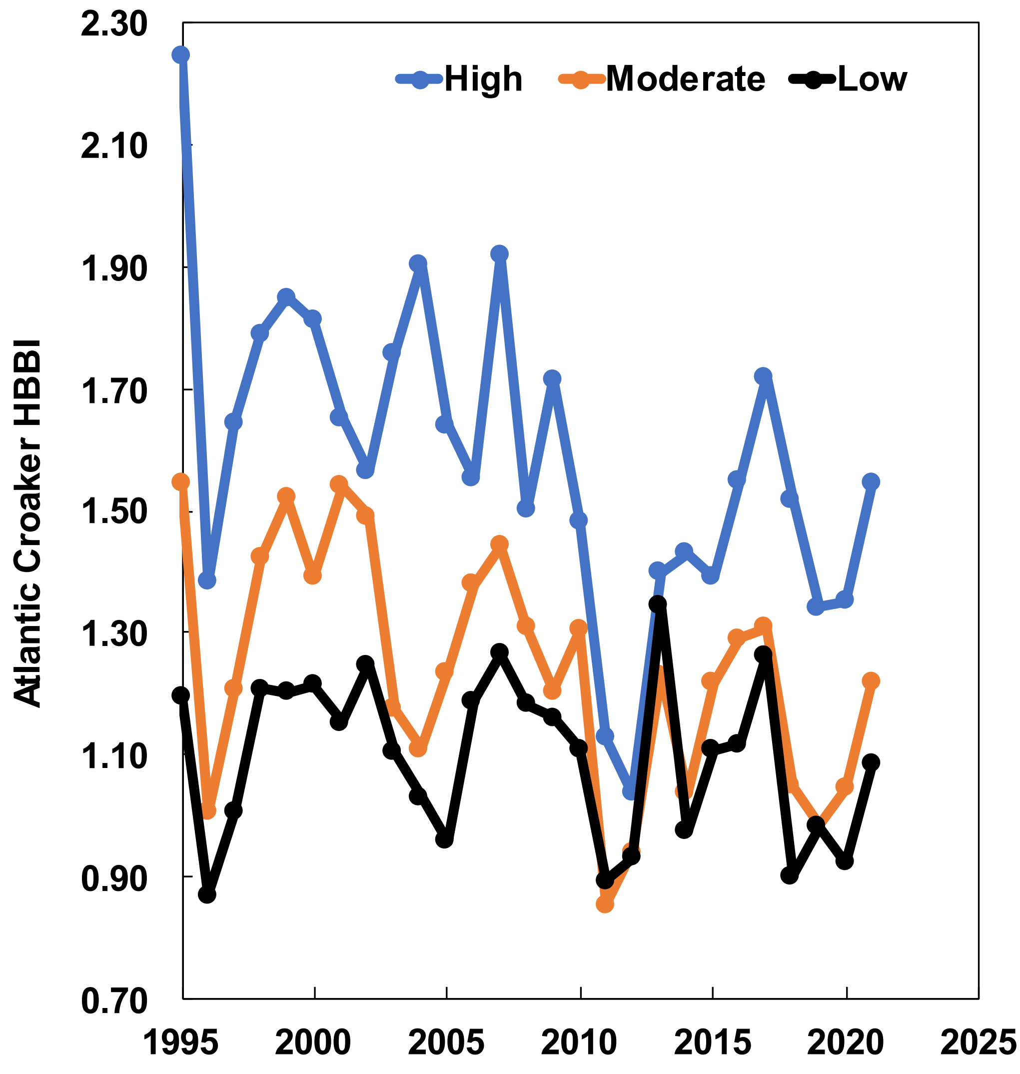


Figure 4. Striped Bass hard bottom benthic indices (HBBI) by salinity region.

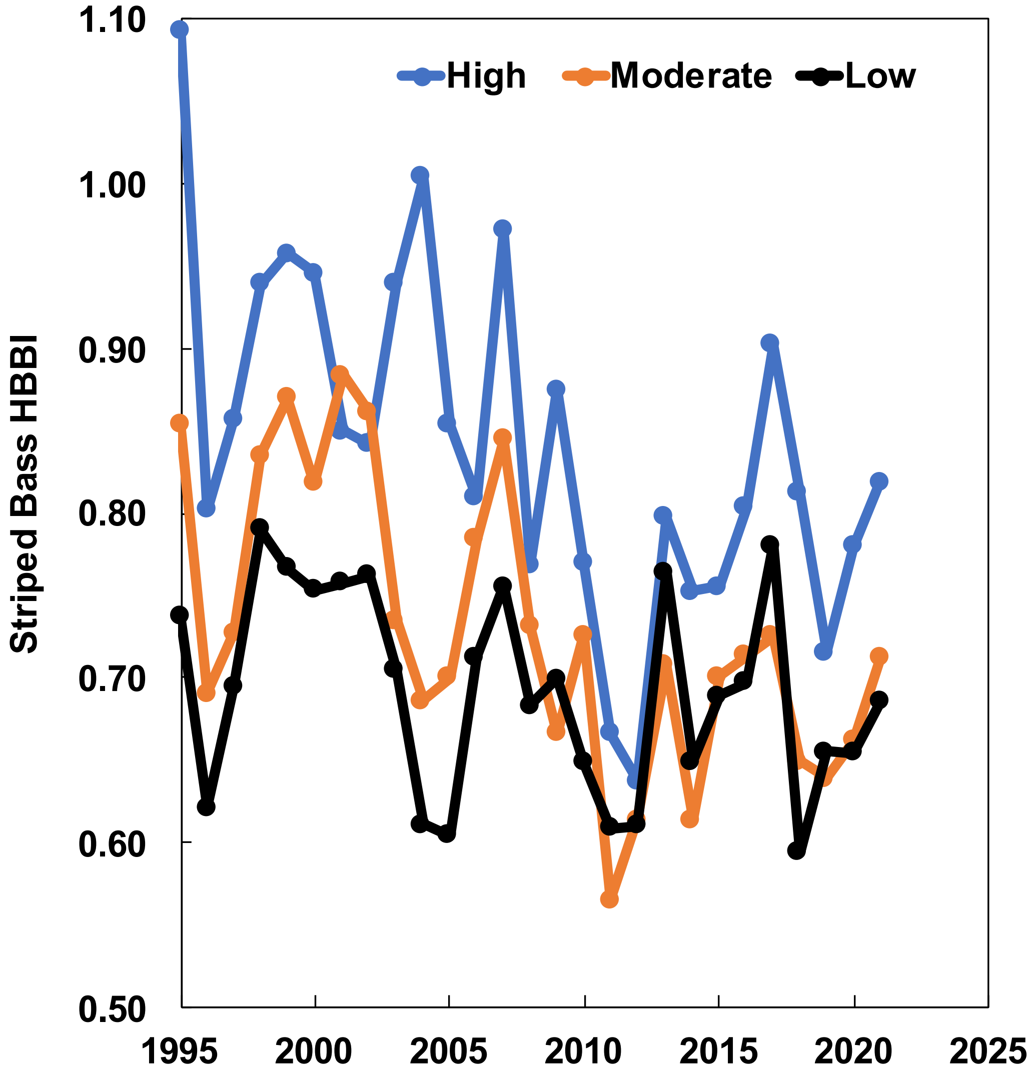


Figure 5. Station HBBI relative status for Spot calculated by pooling data for each station over the time-series, compared to the time-series average HBBI of its corresponding salinity category. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

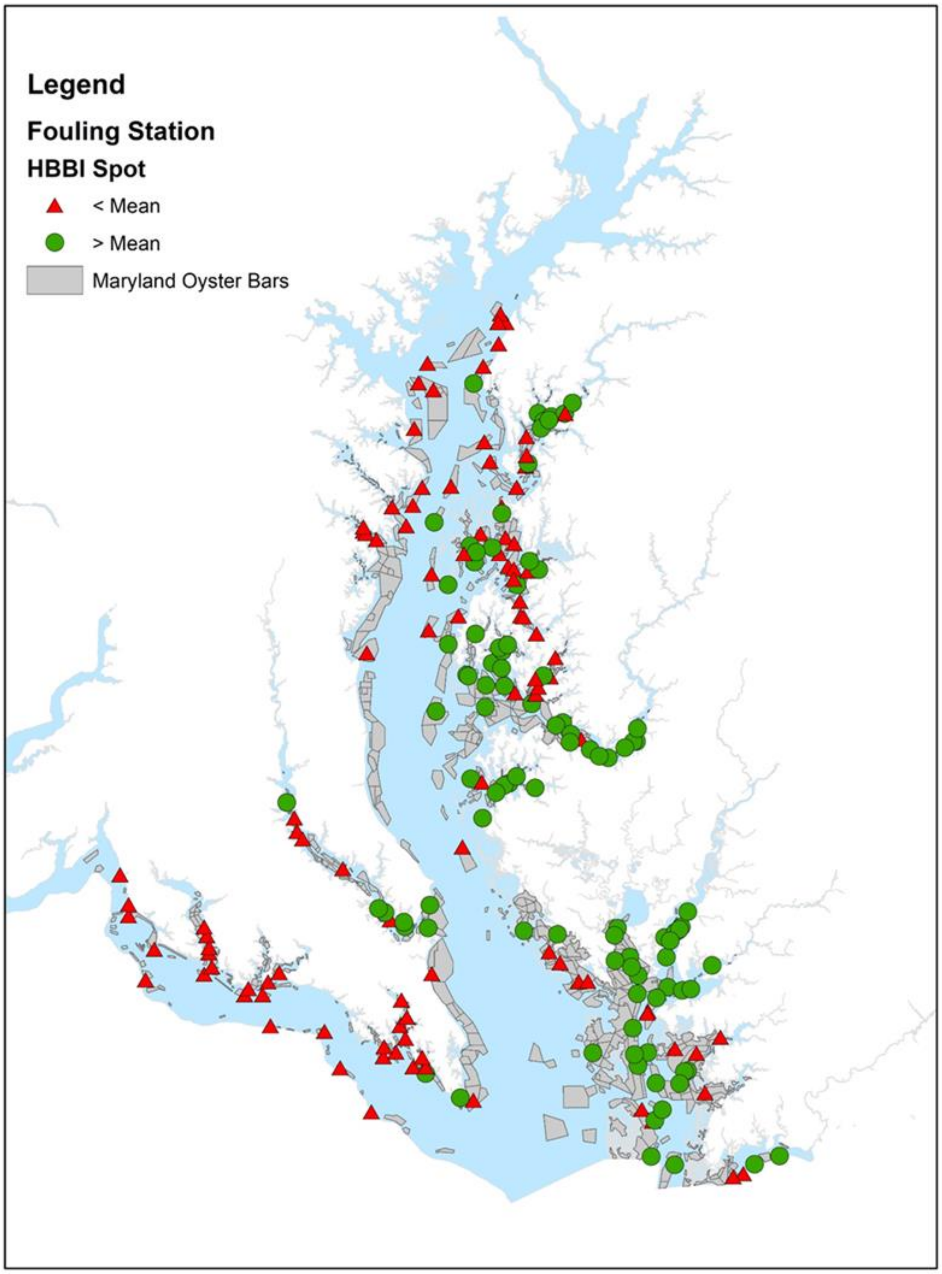


Figure 6. Station HBBI relative status for Atlantic Croaker calculated by pooling data for each station over the time-series, compared to the time-series average HBBI of its corresponding salinity category. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

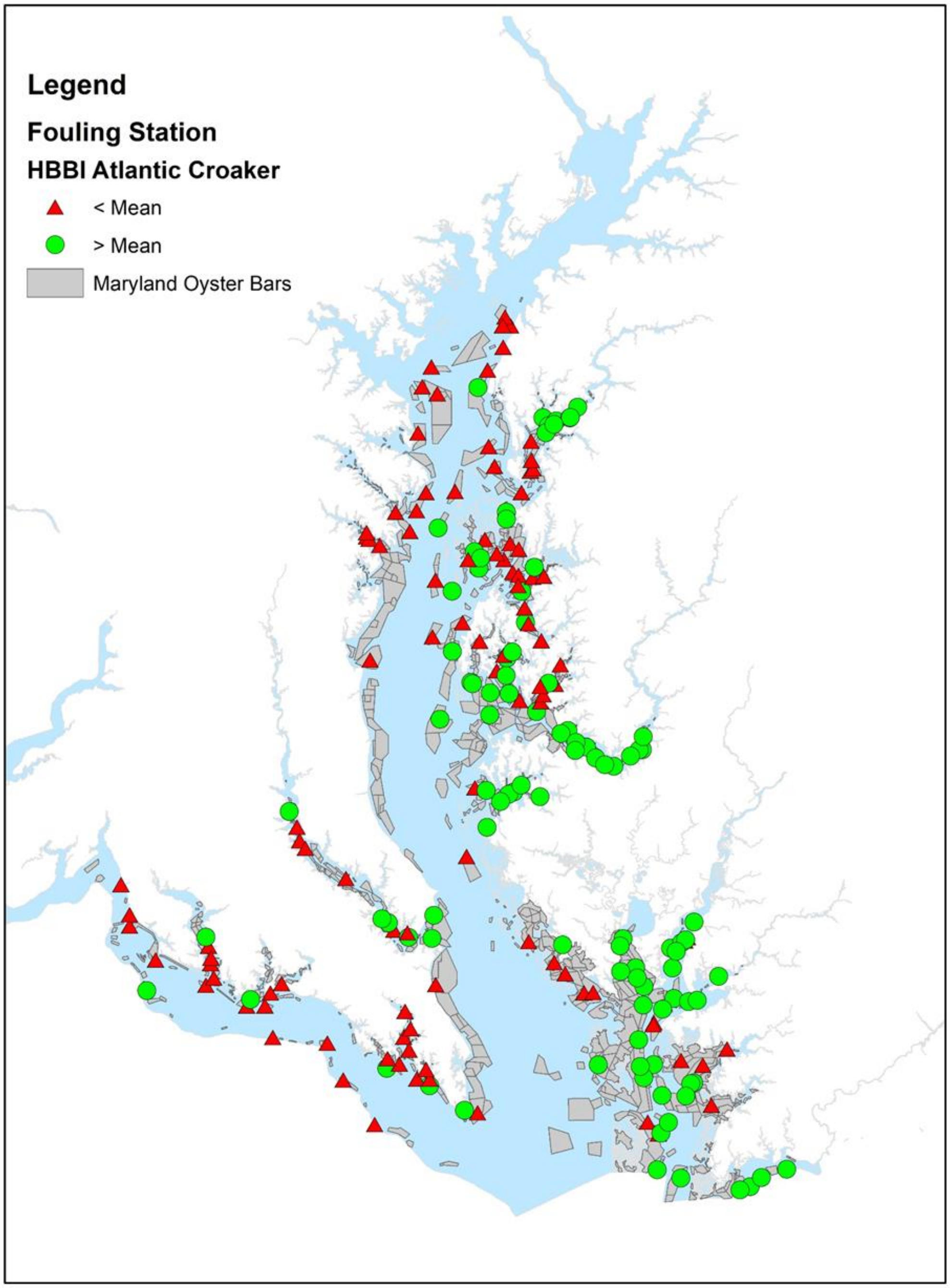


Figure 7. Station HBBI relative status for Striped Bass calculated by pooling data for each station over the time-series, compared to the time-series average HBBI of its corresponding salinity category. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

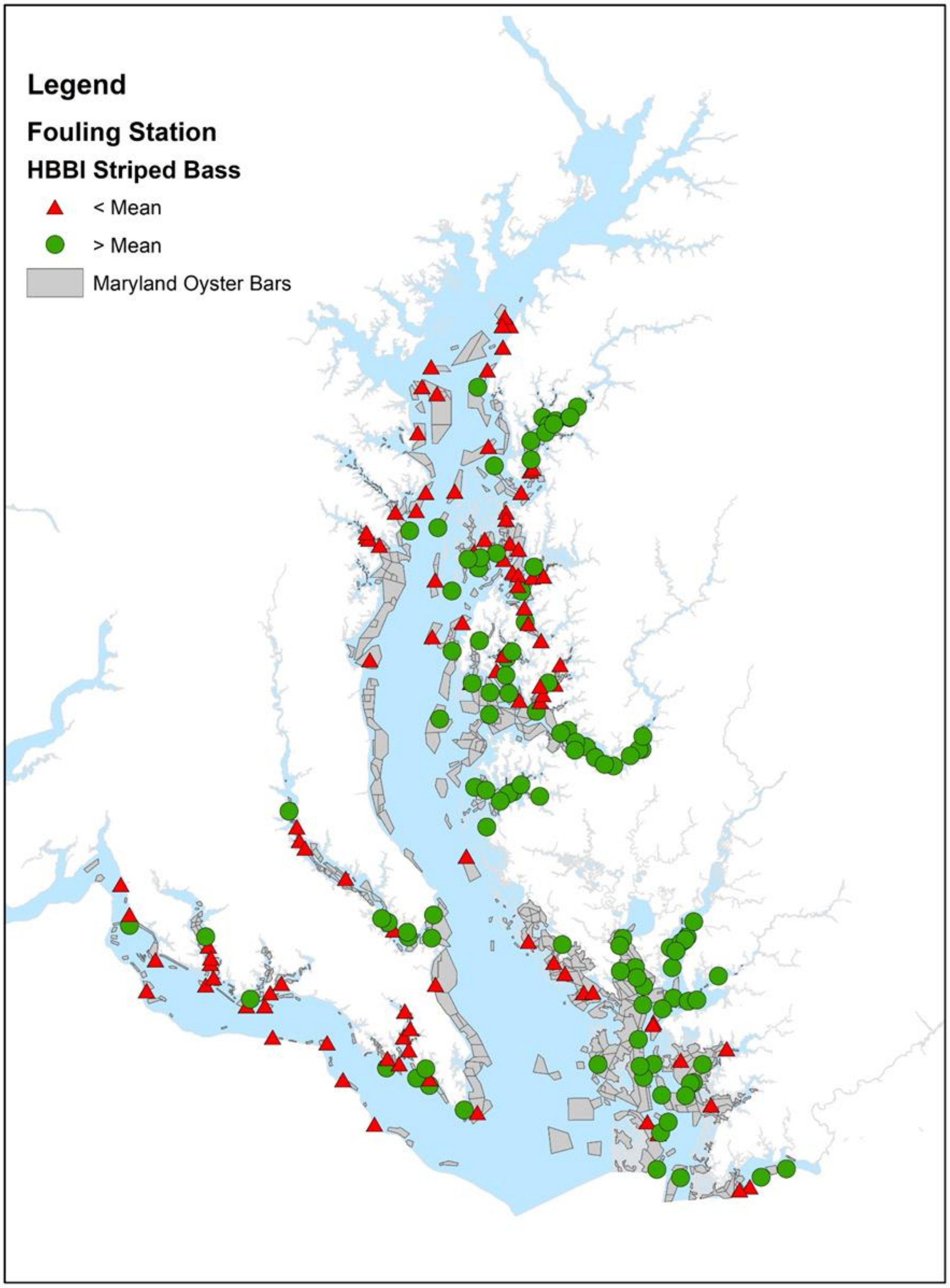


Figure 8. Mean presence of crustaceans for each station over the time- series compared to its salinity region time-series mean presence. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

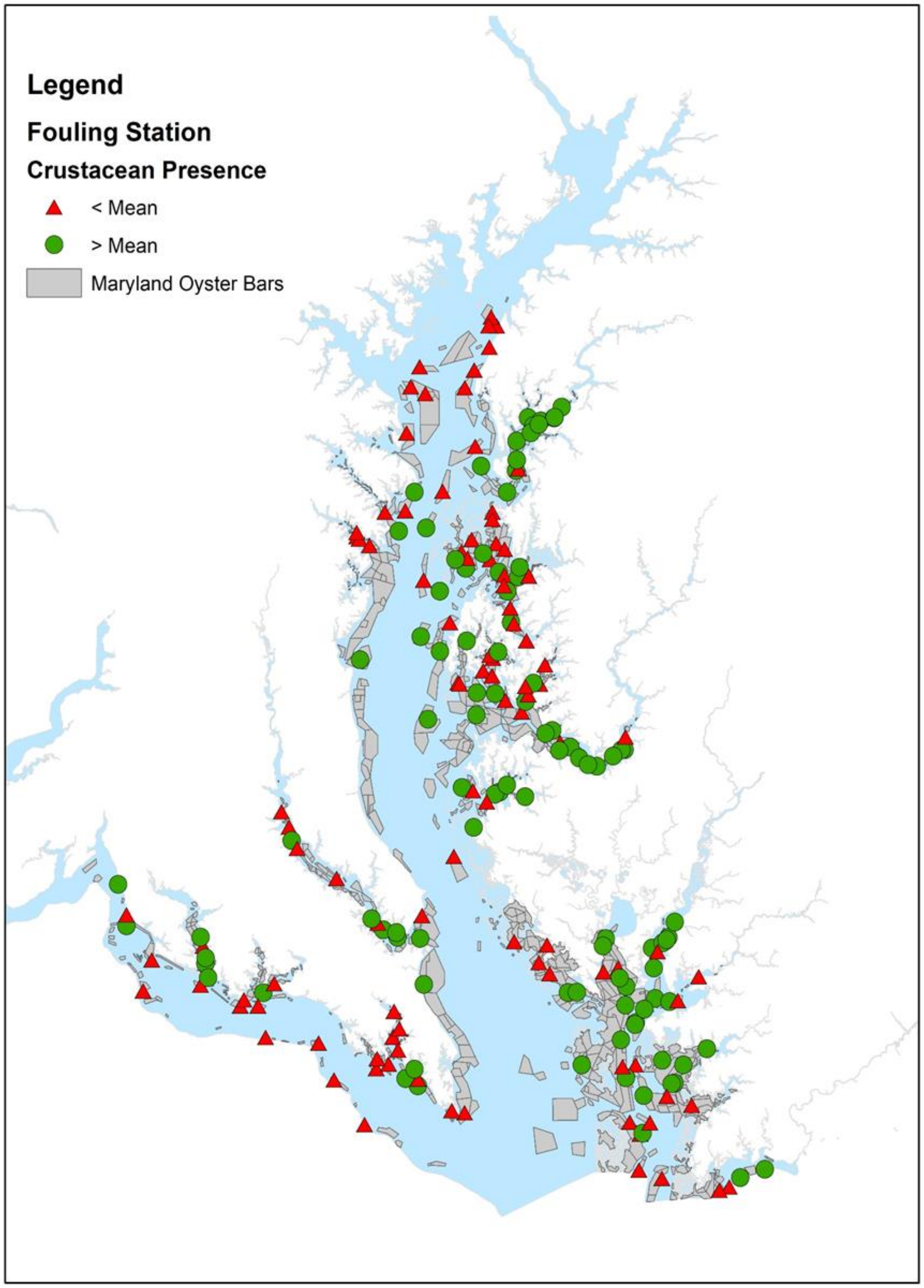


Figure 9. Mean presence of mollusks for each station over the time-series compared to its salinity region time-series mean presence. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

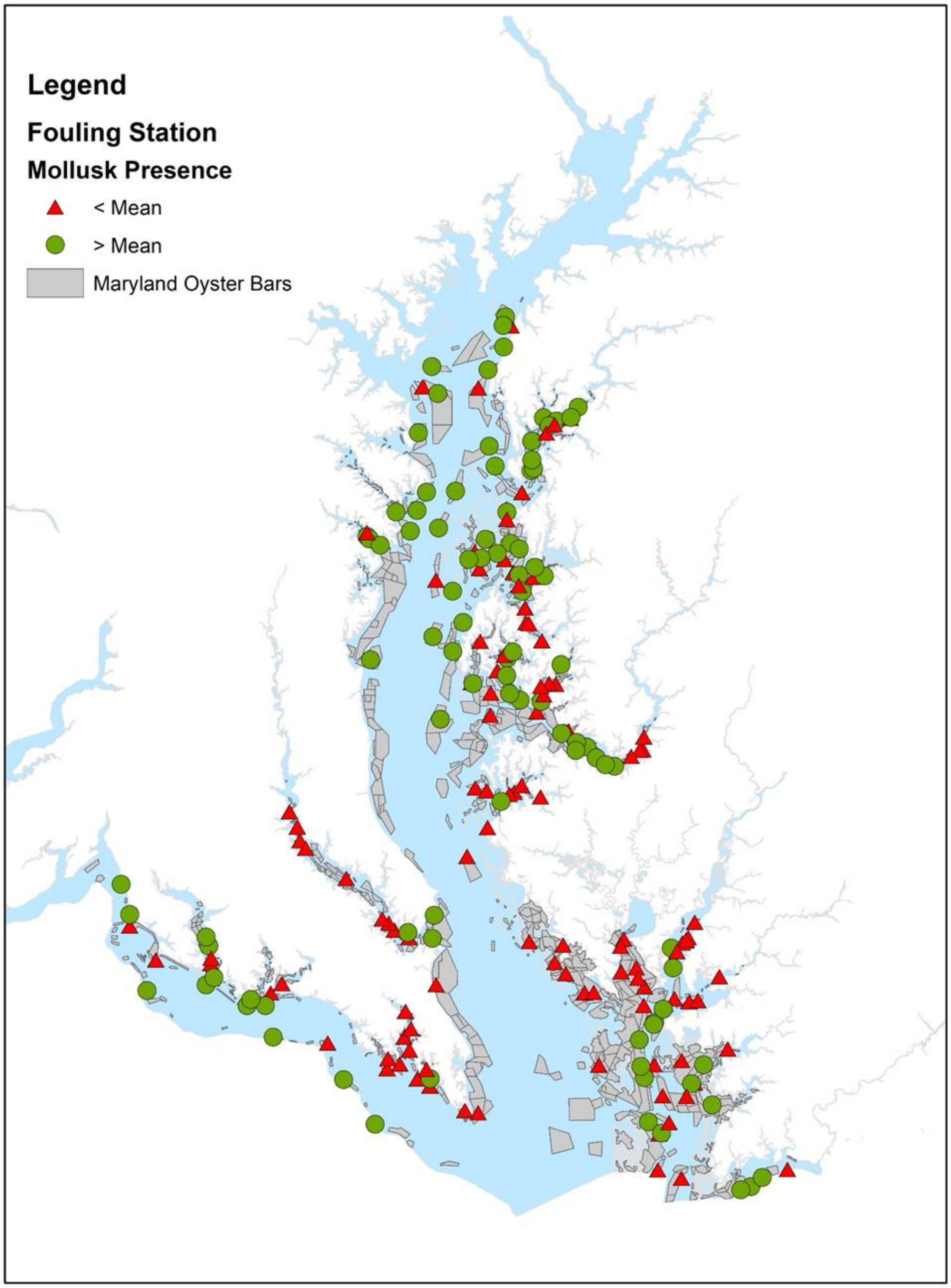


Figure 10. Mean presence of worms for each station over the time-series compared to its salinity region time-series mean presence. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.

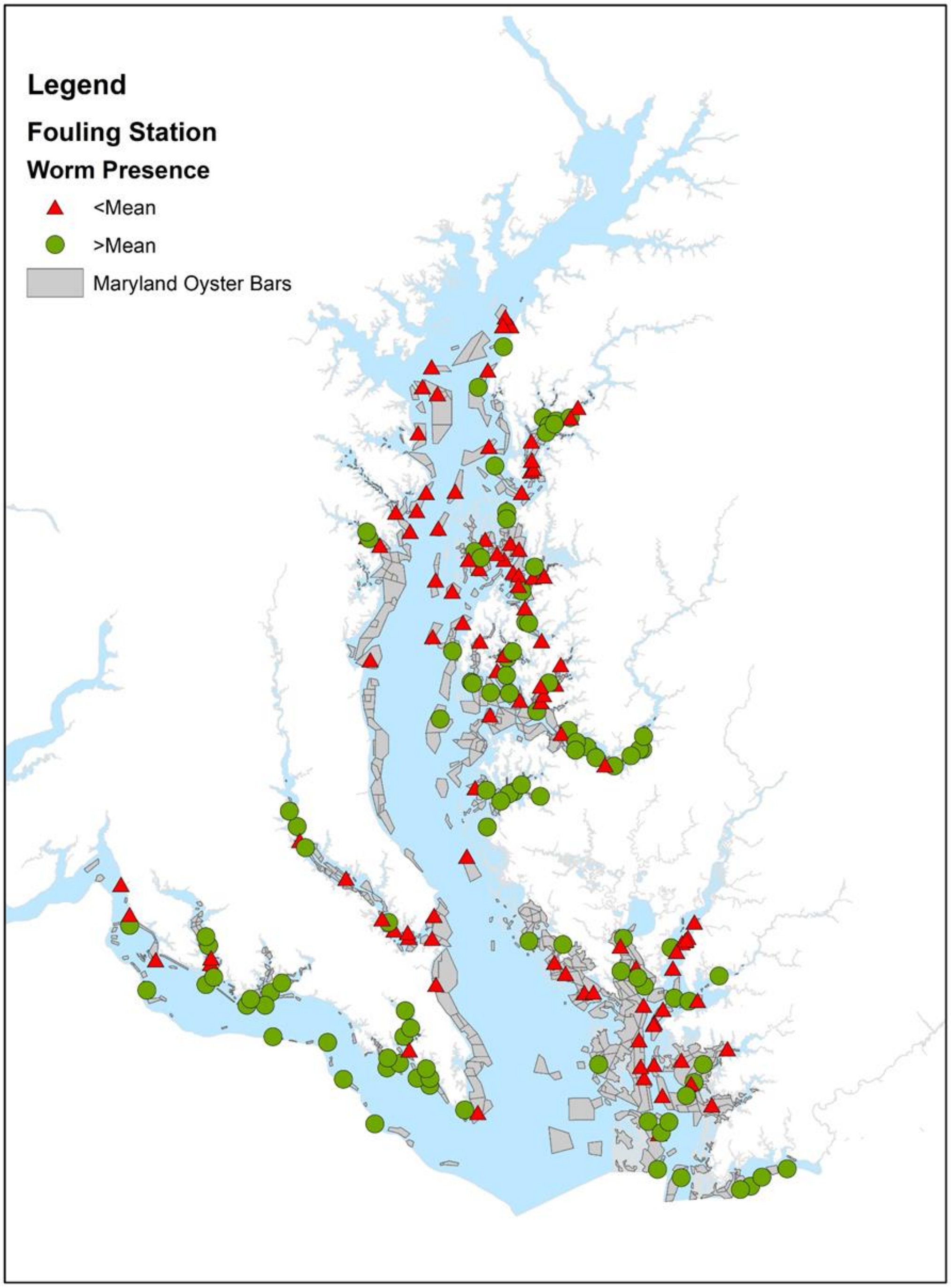
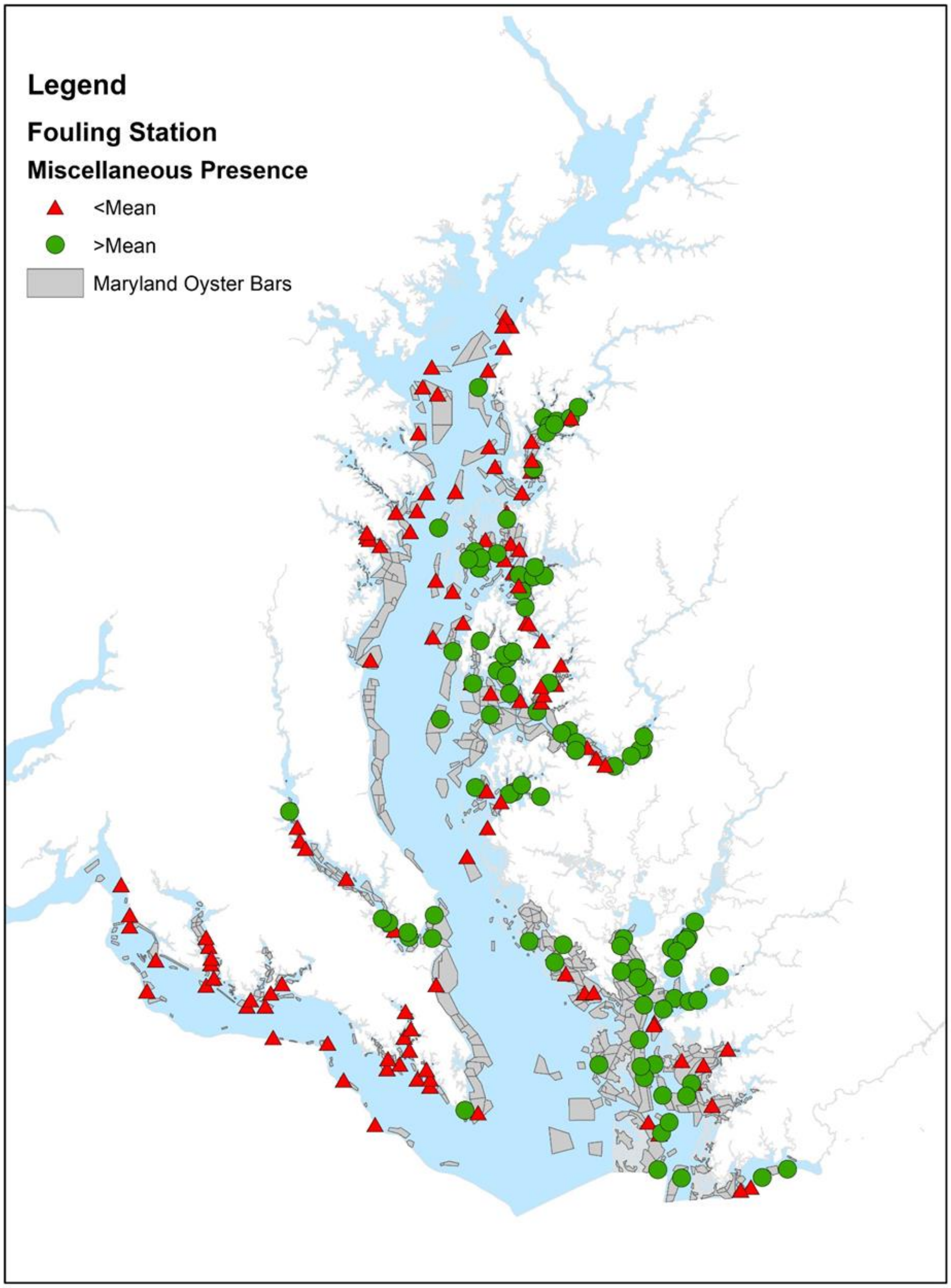


Figure 11. Mean presence of miscellaneous benthic invertebrates for each station over the time-series compared to its salinity region time-series mean presence. Red triangle indicates below salinity region average for a site and a green circle indicates above average for a site.



MD – Marine and estuarine finfish ecological and habitat investigations
Project 4: Resident Striped Bass forage benchmarks

Jim Uphoff, Alexis Park, and Carrie Hoover

Changes to Project 4 Activities due to Coronavirus

Sampling of Striped Bass condition and diets for Job 4 was not affected by the Pandemic.

Executive Summary

Indices of Striped Bass condition, relative abundance, and natural mortality, and forage relative abundance from surveys and fall diets provided metrics (indicators) to assess forage status and Striped Bass well-being in Maryland's portion of Chesapeake Bay. In addition to providing insight on forage status, these indicators were inexpensive and tractable for staff. The proportion of Striped Bass without body fat (P0), anchored our approach, providing a measure of condition and potential for starvation that was well-related to feeding. Proportion of Striped Bass in fall with empty guts (PE) provided trends in prey supply relative to predator demand based on relative abundance and diet sampling, respectively. The proportion of diet items by number and weight of prey per weight of Striped Bass (C) supplemented PE. Metrics based on examination of individual Striped Bass (P0, PE, and C) were split into two size classes (small, <457 mm TL and large, $\geq$ 457 mm TL) due to sampling considerations and recent divergence in trends in P0 between the size classes. The P0 and PE metrics had targets and thresholds and remaining metrics were considered supplemental. An index of survival (SR) that reflected natural mortality (M) was developed for small Striped Bass and trends could be compared with published estimates for large fish. Remaining metrics could not be split for size classes. A Striped Bass recreational catch per trip index (RI) provided an index of relative abundance. Species specific forage-to-Striped Bass ratios were developed from relative abundance indices of major prey (FRs; focal prey species are Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab).

In 2021, the P0 and PE indicators for both size classes did not breach their thresholds; the three indicators with target values met them. Small Striped Bass condition was consistently poor (breaching the threshold) during 1998-2007 and shifted to a mix afterward. Condition of large Striped Bass was at its threshold in 6 of 7 years during 1998-2004 and has improved to only slightly missing its target once since 2014. Estimates of PE for both size classes in 2021 met their target values and were the best in their time-series. Estimates of PE for large and small Striped Bass have improved from threshold conditions prior to 2007. Large Striped Bass have been mostly at target PE since 2014. A target was not readily suggested for PE of small fish. Atlantic Menhaden dominated small and large Striped Bass diets by weight during fall; C has been higher since 2013, more frequently ranking in the top half of estimates. Bay Anchovy were dominant by number in small Striped Bass diets, but made up a low fraction of fall diet weight in all but the worst years. Small Blue Crabs were a minor component by weight as well, but were numerically abundant in some years. Spot, a major prey that had contributed to achievement of target P0 and PE for small fish in 2010, have been largely absent in fall diets of both size classes since 2015. Bay Anchovy were consistently present in fall diets of both size classes of Striped Bass during 2006-2014, but have fallen substantially as a percent of large fish diet since 2015 as Atlantic Menhaden became more frequent. Bay anchovy represented a variable percentage of small fish diets during 2006-2015 and had a steadier, higher frequency afterwards. Diet changes

since 2015 suggest the pelagic pathway is making a larger contribution to fall diets in recent years.

A rapid rise in Striped Bass abundance in upper Bay during the mid-1990s, followed by a dozen more years at high abundance after recovery was declared in 1995, coincided with declines in relative abundance of Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab (i.e., major pelagic and benthic prey) to low levels. Changes in FRs largely reflected decreasing prey during 1983-1994 since RI was low. After 1995, prey indices stayed relatively low and FR changes usually reflected fluctuations in RI. It appears that slightly higher (but not statistically different) Atlantic Menhaden indices since 2007 may have biological significance based on improvement in recent body fat and fall diet metrics.

We did not estimate relative survival (SR) for 2021 due to concerns about the validity of the gill net index for that year. An outbreak of Covid in the Head-of-Bay crew caused two weeks during the main spawning period to be missed and it was feared the estimate would be negatively biased. Multiple lines of evidence suggest that survival of both small (past SR estimates) and large (published estimates) Striped Bass have fluctuated due to changing natural mortality in Chesapeake Bay since the late 1990s.

Introduction

The Chesapeake Bay stock of Striped Bass *Morone saxatilis* 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). A large contingent of Chesapeake Bay Striped Bass that do not participate in the Atlantic coast migration (hereafter, resident Striped Bass) constitute a year-round population of predators, and provide Maryland's major saltwater recreational fishery and an important commercial fishery; they are mostly males along with some young, immature females (Setzler et al. 1980; Kohlenstein 1981; Dorazio et al. 1994; Secor and Piccoli 2007; Maryland Sea Grant 2009).

Striped Bass, fueled by a series of strong year-classes in Chesapeake Bay, were abundant in the 1960s and early 1970s, then declined as recruitment faltered and fishing mortality rates increased (Richards and Rago 1999). Moratoria were imposed in several Mid-Atlantic States in the mid-to-late 1980s and conservative regulations were put in place elsewhere (Uphoff 1997; Richards and Rago 1999). Recovery of Atlantic coast Striped Bass was declared in 1995 after rapid Chesapeake Bay stock growth (Richards and Rago 1999; ASMFC 2021). Management since recovery has been based on much lower fishing mortality and much higher size limits than were in place into the early 1980s (Richards and Rago 1999; ASMFC 2021). An Atlantic Menhaden consumption per Striped Bass recruit analysis indicated that conservative regulatory changes could have increased demand by approximately 2- to 5-times (Uphoff 2003).

Concern emerged about the impact of high Striped Bass population size on its prey-base shortly after recovery from severe depletion in 1995 (Hartman 2003; Hartman and Margraf 2003; Uphoff 2003; Savoy and Crecco 2004; Heimbuch 2008; Davis et al. 2012; Overton et al. 2015; Uphoff and Sharov 2018). Major declines in abundance of important prey (Bay Anchovy *Anchoa mitchilli*, Atlantic Menhaden *Brevoortia tyrannus*, and Spot *Leiostomus xanthurus*) in Maryland's portion of Chesapeake Bay (hereafter upper Bay) coincided with Striped Bass recovery (Uphoff 2003; Overton et al. 2015). Reports of Striped Bass in poor condition and with ulcerative lesions increased in Chesapeake Bay shortly after recovery; linkage of these phenomena with poor feeding success on Atlantic Menhaden and other prey was considered plausible (Overton et al. 2003; Uphoff 2003; Gauthier et al. 2008; Overton et al. 2015; Uphoff and Sharov 2018). Mycobacteriosis, a chronic wasting disease, became widespread in Chesapeake Bay in the late 1990s and was concurrent with lesions and poor condition (Overton et al. 2003; Jiang et al. 2007; Gauthier et al. 2008; Jacobs et al. 2009b). Challenge experiments with Striped Bass linked nutrition with progression and severity of the disease, and reduced survival (Jacobs et al. 2009a). Tagging models indicated that annual instantaneous natural mortality rates (M) of legal sized Striped Bass in Chesapeake Bay increased substantially during the mid-1990s while annual instantaneous fishing mortality rates (F) remained low (Kahn and Crecco 2006; Jiang et al. 2007; ASMFC 2013; NEFSC 2019). Prevalence of mycobacteriosis and M appeared to be lower outside Chesapeake Bay (Matsche et al. 2010; NEFSC 2019), but abundance, condition, and M of the coastal migratory contingent has been linked to abundance of ages 1+ Atlantic Menhaden (Buccheister et al. 2017; Uphoff and Sharov 2018; ASMFC 2020; Chagaris et al. 2020)

Maryland's fisheries managers and stakeholders want to know whether there is enough forage to support Striped Bass in Maryland's portion of the Bay. Maintaining a stable predator-prey base is a challenge for managing Striped Bass in lakes (Axon and Whitehurst 1985; Matthews et al. 1988; Cyterski and Ney 2005; Raborn et al. 2007; Sutton et al. 2013; Wilson et al. 2013). Formal assessments of abundance and biomass of Striped Bass and most forage species in upper Bay are lacking due to cost and difficulty in mathematically separating migration from mortality. The Atlantic States Marine Fisheries Commission (ASMFC) has adopted ecological (forage) reference points for Atlantic Menhaden along the Atlantic coast and Striped Bass is a predator of concern because of its high sensitivity to Atlantic Menhaden population size (ASMFC 2020; Chagaris et al. 2020; Drew et al. 2021; Anstead et al. 2021). In 2014, a forage fish outcome was included in the Chesapeake Bay Agreement (Chesapeake Bay Program): "By 2016, develop a strategy for assessing the forage fish base available as food for predatory species in the Chesapeake Bay." Project 4 is a direct response by MD DNR to this outcome.

Indicators based on monitoring, such as forage indices, prey-predator ratios, condition indices, and prey abundance in diet samples have been suggested as a basis for forage assessment for Striped Bass in Chesapeake Bay (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; Dettmers et al. 2012; Fogarty 2014).

The approach used here is based on a suite of indicators (metrics) that are inexpensively and easily developed from existing MD DNR sampling programs. This report provides indicators through 2021. In addition to providing information for judging whether the forage base is adequate to support Striped Bass in Maryland's portion of Chesapeake Bay, two additional objectives were low cost and tractability for available staff.

During 2014-2019, we developed an integrated index of forage or IF that was comprised of five metrics covering all sizes of Striped Bass within a defined size range (286-864 mm TL or 11.3-34.0 inches). Forage status was judged by whether target (indicating good forage conditions) or threshold (indicating poor forage conditions) reference points were met for each metric. Time periods where body fat indicators were at target or threshold levels provided a time frame for developing targets and thresholds for other metrics.

Uphoff et al. (2020) expressed concern that divergences of some metrics between small (<457 mm TL; < 18 inches) and large ($\geq$ 457 mm TL) Striped Bass were masked by the IF approach. In this report, we have split metrics developed from sampling individual Striped Bass (condition and feeding metrics) between large and small fish where possible. Targets and thresholds were possible for a reduced number of metrics that could be split into the two size classes. Results in this report will be organized into sections that describe metrics for small Striped Bass, metrics for large fish, and metrics for both sizes combined.

Poor condition is a common problem for Striped Bass in lakes when prey supply is inadequate (Axon and Whitehurst 1985; Matthews et al. 1988; Cyterski and Ney 2005; Raborn et al. 2007; Sutton et al. 2013; Wilson et al. 2013). The proportion of Striped Bass without body fat (P0), a nutritional indicator, anchors our approach, providing a measure of condition and potential for starvation for each size class that was well-related to proximate composition and feeding of Striped Bass in the laboratory (Jacobs et al. 2013). The target developed by Jacobs et al. (2013) has been retained for both size classes and thresholds developed in previous years were revisited in Uphoff et al. 2022). Lipids are the source of metabolic energy for growth, reproduction, and swimming for fish and energy reserves relate strongly to foraging success, reproductive success, potential prey density, habitat conditions, environmental stressors, and subsequent fish health and survival (Tocher 2003; Jacobs et al. 2013); P0 integrates these factors into a single measure. A reliable and easily applied indicator of nutritional state is critical for evaluating hypotheses related to nutrition, prey abundance, density, and the outcome of the management measures that may follow (Jacobs et al. 2013).

Proportion of empty guts (PE) was used as a consumption-based indicator of major prey availability for each size class. Supplemental metrics on weight of prey consumed per weight of Striped Bass that consumed them (C), and composition of prey consumed (by number) could be estimated for each size class as well.

While upper Bay Striped Bass feed on a wide range of prey, Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab *Callinectes sapidus* have consistently accounted for most annual diet biomass in Chesapeake Bay studies (Hartman and Brandt 1995c; Griffin and Margraf 2003; Walter et al. 2003; Overton et al. 2009; Overton et al. 2015; Buccheister and Houde 2016). We selected these species as focal prey (major prey) for forage indices. Forage ratios of species-specific indices of major prey relative abundance from fishery-independent surveys to an indicator of resident Striped Bass relative abundance were examined for each focal prey as an indicator of potential attack success. These forage ratios could not be split into size categories. Forage species indices alone would not consider the possibility of predator interference or the vulnerability exchange process of foraging arena theory (Ginzburg and Akçakaya 1992; Yodzis 1994; Ulltang 1996; Uphoff 2003; Walters and Martell 2004; Walters et al. 2016).

Benthic invertebrate indices (invertebrates other than Blue Crabs) are included in this report even though benthic invertebrates have not contributed much to fall diets. Uphoff et al. (2018) found that P0 the previous summer and the previous fall could influence P0; condition of Striped Bass in summer may be influenced by benthic invertebrates since they can be a significant component of their spring diet (Overton et al. 2015). The utility of estimates of biomass of invertebrates comprising a benthic IBI in Maryland’s portion of the Bay used for water quality monitoring was explored in Uphoff et al. (2018). A complementary index for hard (oyster) bottom was developed by M. McGinty (Uphoff et al. 2018).

The ratio of age-3 relative abundance of male Striped Bass in spring spawning ground gill net surveys (Versak 2021) to their year-class-specific juvenile indices (Durell and Weedon 2021) during 1985-2021 was used as an indicator of change in relative survival of small fish (SR) due to M prior to recruitment to the fishery. The SR was an index for small fish since it tracked survival trends between young-of-year and age 3. Martino and Houde (2012) detected density-dependent mortality of age 0 Striped Bass in Chesapeake Bay, supporting a hypothesis that density dependence in the juvenile stage can contribute significantly to regulation of year-class strength. We expected SR to vary without trend if M remained constant. Very general trends in the SR, an index of the effect of M on small Striped Bass, could be compared with trends in estimates of M for large fish developed from conventional (NEFSC 2019) and acoustic tags (Secor et al. 2020).

Methods

Abbreviations and definitions - Table 1 contains important abbreviations and definitions.

Striped Bass condition, feeding success, and diet composition indices – Indicators of condition, feeding success, and diet composition during October-November were developed for Striped Bass caught by hook-and-line. A citizen-science based Striped Bass diet monitoring program was conducted by Chesapeake Bay Ecological Foundation (CBEF) during 2006-2015 and 2006-2013 collections were used to estimate feeding success and diet composition. Diet samples from a Fish and Wildlife Health Program (FWHP) Striped Bass health survey were used after 2013. Methods for CBEF and FWHP collections have been described in Uphoff et al. (2014; 2015; 2016) and will be briefly repeated below.

The collector's permit issued to CBEF allowed for samples of up to 15 Striped Bass less than 457 mm total length (or TL; small Striped Bass or fish; the minimum length limit for Striped Bass was 457 mm or 18 inches when the permit was issued) and 15 fish 457 mm TL or larger (large Striped Bass or fish) per trip during 2006-2014. The small and large designations replace sublegal and legal sized designations used in previous reports; this change was made to prevent confusion that may arise due to length limit changes (the length limit was 457 mm TL during 1998-2014; it was raised to 508 mm TL in 2015, lowered to 483 mm TL in 2018 and has remained there through 2021).

Striped Bass diet collections by CBEF were made in a portion of upper Bay bounded by the William Preston Lane Bridge to the north, the mouth of Patuxent River to the south, and into the lower Choptank River (Figure 1). Most active trips by CBEF occurred in Choptank River, but some occurred in the mainstem Chesapeake Bay. Active trips were our source of small sized fish, but large sized fish were caught as well. Striped Bass kept as samples during active trips were placed in a cooler and either processed upon return to shore or held on ice for processing the next day. Collections of large Striped Bass were supplemented by sampling charter boat hook-and-line catches at a fish cleaning business. These fish were predominately from the mainstem Chesapeake Bay; they were iced immediately and cleaned 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 by CBEF at the check station.

Striped Bass collected for health samples by Fish and Wildlife Health Program (FWHP) have been processed since 2014 by Fish Habitat and Ecosystem Program (FHEP) biologists for diet information. Collections by FWHP were not constrained by collector's permit conditions like CBEF collections. Fish have been collected by hook-and-line from varying locations during fall since 1998 between Baltimore, Maryland (northern boundary) and the Maryland-Virginia state line (southern boundary; Figure 1). Sampling by FWHP was designed to fill size class categories corresponding to age-classes in an age-length key to assess Striped Bass health. Some trips occurred where fish in filled out length classes were discarded (typically small fish). Samples were usually obtained by fishing on a charter boat using the techniques considered most effective by the captain (bait or artificial lures). Bait was excluded from diet data.

Condition was estimated from an existing FWHP Striped Bass health survey that began in 1998. Nutritional status (condition) for upper Bay Striped Bass was estimated as the proportion of fish without visible body fat (P0) during October-November in FWHP samples (Jacobs et al. 2013). Estimates of P0 were made for the two size classes of Striped Bass. Estimates of P0 for 1998–2013 were provided by FWHP and remaining years were estimated from FWHP data by FHEP. Standard deviations and confidence intervals (90%) 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 condition target based on 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 (high FRs). 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 (Jacobs et al. 2013). A level of P0 of 0.30 or less was used to judge whether Striped Bass were in good condition. Variation of tissue lipids estimated from body fat indices was greater than for moisture and the higher P0 target accounted for this additional variation plus a buffer for misjudging status (J. Jacobs, NOAA, personal communication). 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. Uphoff et al. (2014) estimated the P0 threshold as 0.68 (average of the lower 95% CI of high P0 estimates for both size classes during 1998-2004, a period of consistently poor condition). Other indicators of condition were described in Jacobs et al. (2013), but P0 was chosen because it could be applied to data collected by CBEF; P0 estimates from CBEF collections were similar to those estimated for FWHP collections for years in common (Uphoff et al. 2018).

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. Collections by CBEF were processed by James Price with aid on occasion from J. Uphoff and Joseph Boone (a retired MD DNR fisheries biologist). Guts were removed from the Striped Bass and emptied. Total length of intact fish and shrimp, carapace width of crabs, and shell length of intact bivalves were measured; some food items were weighed with a calibrated digital scale. Non-linear allometry equations for converting diet item length to weight (Hartman and Brandt 1995a) were used for items that were only measured. In a few cases, equations for a similar species were substituted when an equation was not available. These equations, originally developed and used by Hartman and Brandt (1995a), had been used to reconstruct diets for Overton et al. (2009) and Griffin and Margraf (2003).

Fish Habitat and Ecosystem Program staff identified, measured, and weighed diet items from FWHP sampling (2014 to present) as FWHP staff processed Striped Bass in the lab. All organisms were blotted as dry as possible before weighing. Three broad data categories of diet data were formed for processing. The first category was composed of fish and invertebrates where information from individual organisms was desired. Lengths (TL for fish, CW or carapace width for crabs, and maximum length of shell for intact bivalves) and weights were measured. Bay Anchovy were a special case since Striped Bass sometimes consumed large numbers. Up to ten Bay Anchovies were measured and weighed per Striped Bass and the remainder were weighed together. Total weight of partially intact fish in a gut was recorded. The second category were data from larger invertebrates that may be present as whole individuals or identifiable with inspection as parts. If these items were in good condition, they were recorded as counts and individual lengths and mass recorded with the same procedure as Bay Anchovy. Otherwise, a count and combined mass were recorded. In some cases, it was only possible to record that these organisms were present (lots of parts, not many whole). The third category was soft invertebrates such as amphipods or polychaetes that were likely to be broken up or digested. Presence was the only numerical descriptor possible. Empirical relationships developed by Stobberup et al. (2009) for general taxonomic categories were used to estimate relative weight from frequency of occurrence of these soft invertebrates. These soft items were uncommon in our fall collections, but were more common during other seasons (J. Uphoff, personal observation).

Diets were analyzed separately for small and large Striped Bass for both CBEF and FWHP collections. These categories accounted for ontogenic changes in Striped Bass diet, but also reflected unbalanced sample availability to CBEF (small fish could only be collected by fishing for them directly, while large sized fish were supplemented by cleaning station samples). The lower limit of fish analyzed in the small category, 286 mm, was the minimum length in common among years during 2006-2013. An upper limit of 864 mm avoided inclusion of very 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 without flesh were excluded. 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.

Percentage of food represented by an item in numbers was estimated for each Striped Bass size class based on fish with stomach contents for each year since 2006 (Pope et al. 2001). Estimates included both counts of whole items and presence of partially intact prey (portions that were intact enough to identify a prey, but not intact enough to measure and weigh as individuals). The latter could include multiple individuals, so percent by number was negatively biased to some extent.

Relative availability of prey biomass (biomass consumed or 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 an indicator of total prey availability (Hyslop 1980). Standard deviations and 90% CI's of PE were estimated using the normal distribution approximation of the binomial distribution (Ott 1977). Estimates of PE from Overton et al. (2009) were available to estimate threshold conditions during 1998-2000 (Uphoff et al. 2017). In addition, this indicator could be derived from published diet information from the 1930s (Hollis 1952) and the 1950s (Griffin and Margraf 2003) for comparisons within our small fish category.

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. We converted these estimates into PE; PE was 0.54 for fish between 301 and 500 mm, TL (approximating our small class) and 0.57 for Striped Bass between 501 and 700 mm (approximating our large class; Overton et al. 2009). These 1998-2000 estimates were comparable to our highest estimates of PE and were concurrent with high P0, high abundance of Striped Bass, and a nadir in major prey indices (except the Bay Anchovy trawl index). Target PE was estimated for small or large fish from periods when PE corresponded with target estimates of P0.

To aid interpretation of PE, we examined prey-predator length ratios (PPLR) of the two size classes of Striped Bass. For this analysis we determined PPLRs for the two largest major prey in fall diets: Spot and Atlantic Menhaden. This analysis was based on ratios for whole prey and was split for small and large Striped Bass. We determined median PPLR for each year and size class of Striped Bass; we compared these estimates to optimum PPLR for Striped Bass (0.21; Overton et al. 2009).

Relative abundance indices of prey and Striped Bass - We used geometric mean catches from fixed station seine and trawl surveys as indicators of relative abundance of major prey in upper Bay. A shoreline seine survey targeting age-0 Striped Bass provided indices since 1959 for Atlantic Menhaden, Bay Anchovy, and Spot (Durell and Weedon 2021). Additional indices for Spot and Bay Anchovy since 1989 were estimated from a Blue Crab trawl survey conducted during summer (Uphoff 1998; Rickabaugh and Messer 2020; MD DNR 2022a; the most current estimates were provided by H. Rickabaugh, MD DNR, personal communication). These surveys sampled major and minor tributaries, sounds adjacent to the mainstem upper Bay, but not the mainstem (Figure 1). Sampling occurred during May-October. Density of juvenile Blue Crabs in a stratified random winter dredge survey that has sampled Chesapeake Bay-wide (Maryland and Virginia) since 1989 was our indicator of Blue Crab relative abundance (Sharov et al. 2003; Jensen et al. 2005; MD DNR 2022b). 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). Each forage index was divided by its mean for years in common among all surveys (1989-current) to place their time-series on the same scale for graphical comparisons of trends among surveys.

A soft bottom benthic biomass index (invertebrates living in the sediment) has been a component of a Chesapeake Bay benthic index of biotic integrity (BIBI); the BIBI provides an accessible summary of benthic habitat status (Weisburg et al. 1997). We used the biomass (grams / m^2) of benthic invertebrates component for Maryland tidal waters as our index (Figure 3-37 in Llansó and Zaveta 2019; Versar Inc 2022). The BIBI has been employed to monitor water quality since 1995 and the latest indices are for 2021. The benthic biomass component consists of 7 polychaetes, 10 mollusks, 1 isopod, 2 amphipods, and 2 ribbon worms (see Table 2-5 in Llansó and Zaveta 2019). Uphoff et al. (2018) explored the relationship of this benthic biomass index on resident Striped Bass condition. This index was not incorporated into a forage ratio (described below for major prey).

A fishery-independent index of relative abundance of upper Bay resident Striped Bass was not available and we used estimates of Maryland Striped Bass catch-per-private boat trip (released and harvested fish; RI) from the National Marine Fisheries Service's (NMFS) Marine

Recreational Information Program (MRIP; NMFS Fisheries Statistics Division 2022) database as an index. Online estimates of catch and effort are available for 1981 and onwards. 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* (NEFSC 2019; NEFSC 2012; ASMFC 2013). Our RI estimates were based on revised MRIP estimates, but these estimates varied little from those estimated by the previous recreational survey (Marine Recreational Fishing Statistics Survey or MRFSS; J. Uphoff, MD DNR, unpublished analysis).

The RI 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 2022). The RI equaled September-October recreational private and rental boat catch of Striped Bass divided by estimates of trips for all species 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 2022). This wave was chosen because portions or the whole wave were continuously open for harvest of 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 as could be obtained. Migratory fish were unlikely to have been present during this wave. The RI was related to juvenile indices 2-5 years earlier (determined by multiple regression) and to Atlantic coast abundance estimates (Uphoff et al. 2014). Trends in the RI compared favorably to the abundance estimates for 2–5-year-old Striped Bass estimated by the statistical catch at age model used in the recent stock assessment (NEFSC 2019; Uphoff et al. 2020).

We used forage indices divided by RI (forage index-to-Striped Bass index ratios, i.e., forage ratio or FR) as indicators of forage supply of major prey relative to Striped Bass demand, an index of potential attack success. Ratios were standardized by dividing each year's FR estimate by the mean of FR during 1989 to the present, a time-period in common among all data; FR estimates were available for every year since 1983 except 1987 (RI was not estimated).

We did not estimate relative survival (SR) for 2021 due to concerns about the validity of the spring gill net index for that year. An outbreak of Covid in the Head-of-Bay crew caused two weeks during the main spawning period to be missed and it was feared the estimate would be negatively biased (B. Versak, MD DNR, personal communication).

Confidence intervals (90%) were developed for ratio-based metrics using an Excel add-in, @Risk, to simulate distributions reported for numerators and denominators. Each annual set of estimates was simulated 5,000-times. Ratio metrics simulated were RI and FR for Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab. Annual means and standard errors reported for these indices were used to generate simulations. Numerators and denominators of the RI and the Blue Crab index were considered normally distributed since their annual distributions were characterized by a means and SE in their respective sources (NMFS Fisheries Statistics Division 2022; MD DNR 2022b; SE for Blue Crab is a personal communication from G. Davis, MD DNR). Remaining indices for Atlantic Menhaden (seine), Bay Anchovy (seine and trawl), and Spot (seine and trawl) and the JI for Striped Bass were based on geometric means (Durell and Weedon 2021). Geometric mean indices were back-transformed into the mean of $\log_e$-transformed catches (+1) and its standard error was derived from the 95% CI. The $\log_e$-

transformation normalized the original catch data. Geometric means were recreated by exponentiating the simulated mean of $\log_e$-transformed catches (+1).

@Risk used Latin Hypercube sampling to recreate input distributions by stratifying their cumulative curves into equal intervals and then sampled each interval without replacement (Palisade Corporation 2016). Sampling was forced to represent values in each interval and recreated the original input distribution. Latin Hypercube sampling uses fewer iterations compared to random sampling employed by Monte Carlo simulations and is more effective when low probability outcomes are present (Palisade Corporation 2016).

Results

Sample Size Summary - During 1998-2021, 2,114 small and 2,922 large Striped Bass were sampled during October-November (Table 2). Annual sample sizes for small fish in October-November ranged from 24 to 271 with a median of 120. Annual sample sizes for large fish ranged from 49 to 327 with a median of 194. Fewer dates were sampled within similar time spans after the FWHP became the platform for sampling in 2014 because numbers collected per trip were not confined by the terms of the CBEF collector's permit (6-12 per trips in fall by FWHP during 2014-2019 versus 11-22 trips by CBEF during 2006-2013). In most years, starting dates for surveys analyzed were similar between those conducted by CBEF and FWHP (October 1-9), but samples taken on September 24, 2015, were included in that year's analysis because the earliest date sampled in October would have been October 21, 2015. The late start date for 2021 reflected a dearth of fish available until mid-October (J. Uphoff, MD DNR, personal observation). End dates tended to be earlier in November for FWHP surveys, reflecting when size categories were filled out (Table 2).

Small Striped Bass Condition, feeding success, and diet composition indices - Condition of small Striped Bass has transitioned from consistently poor during 1998-2007 to a mix of at or near target P0 interspersed with scattered years of poor P0 afterward (Figure 2). Small Striped Bass were at the target level of condition ($P0 \leq 0.30$) during 2008, 2015, 2017, and 2021 (best of the time-series). Small fish in the upper Bay during fall were in poorest condition during 1998-2007, 2011-2012, 2016, and 2019; we adopted P0 = 0.67 (minimum during 1998-2007) as this size group's threshold (Uphoff et al. 2022). Estimates of P0 (0.36-0.46) were between the target and threshold during 2009-2010, 2013-2014, 2018, and 2020. The 90% confidence intervals of P0 allowed for separation of years at or near the threshold from remaining estimates (Figure 2).

Estimates of PE of small Striped Bass during fall, 2006-2021, ranged between 0.10 and 0.57 (Figure 3). Estimates of PE during 2006-2007, 2012, and 2015 could not be clearly separated from the threshold based on 90% CI overlap; PE during 1998-2000 (Overton et al. 2009), was the threshold for small fish (PE = 0.54; Uphoff et al. 2016). Lowest estimates of PE for small fish (2009-2011, 2014, 2017, and 2019) could be separated from remaining higher estimates (except 2008) based on 90% confidence interval overlap. Estimates of PE during 2008-2011, 2014, and 2016-2021 were clearly lower than the 90% CIs of years that breached the threshold. Estimated PE in 2021 (0.26) was below the threshold and time-series median (0.31; Figure 3).

In combination and by number, Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab accounted for 96.1% of diet items encountered in small Striped Bass collected from upper Bay during fall, 2006-2021 (Figure 4). Bay Anchovy accounted for the highest percentage by number when all years were combined (62.5%, annual range = 19.1-87.9%); Atlantic Menhaden, 19.1% (annual range = 0-69.9%); Spot 5.4% (annual range = 0-70.7%); Blue Crab, 12.2%

(annual range = 0.8-34.6%); and other items accounted for 3.9% (annual range = 0-24.5%; Figure 4). During 2021, Atlantic Menhaden accounted for 69.9% of the diet items; Bay Anchovy, 29%; Spot and Blue Crab, 0%; and other items accounted for 1.1%. The vast majority of major prey in small Striped Bass diet samples during fall fell within young-of-year length cut-offs (Uphoff et al. 2016).

By weight, small Striped Bass diets in fall 2006-2021 (combined) were comprised of Atlantic Menhaden (71.7%), Bay Anchovy (13.4%), Spot (8.7%), Blue Crab (1.9%) and other items (2.7%; Figure 5). Estimates of C (total grams of prey consumed per gram of Striped Bass) for small Striped Bass varied as much as 8.7-times during 2006-2021. During years of lowest C (2007, 2011, 2016, and 2017), varying items contributed to the diet of small fish. During years of when C was high (more than twice the 2006-2021 median) either Spot (2010) or Atlantic Menhaden (2013-2014) dominated diet mass. The 2021 estimate of C of small fish (0.016) was above the median (0.013) of the year time-series (Figure 5).

Median PPLRs of large prey of small Striped Bass (Spot and Atlantic Menhaden combined) were 0.20-0.38 during 2006-2021 (Figure 6). Median PPLRs for small fish were particularly high (0.34-0.38) during 2012 and 2015-2019. They were close to the optimum (0.21) described by Overton et al. (2009) in 2010 (2010 PPLR = 0.199) when Spot constituted a large fraction of their diet. The median PPLR was 0.30 for 2021 (Figure 6). High estimates of C (defined previously) coincided with three of the four lowest PPLRs.

Large Striped Bass condition, feeding success, and diet composition indices - Condition of large Striped Bass has transitioned from mostly poor during 1998-2004 to a mix of at or near target P0 after 2013. Large Striped Bass were at the target level of condition (P0 $\leq$ 0.30) during 2008-2010, 2014-2015, and 2017-2021 (Figure 7). Estimated P0 (0.011) in 2021 was the lowest of the time-series. Large fish during fall were usually in poorest condition (P0 $\geq$ 0.70) during 1998-2004 (except 2002) and we adopted P0 = 0.70 as this size group's threshold. The 90% confidence intervals of P0 allowed for separation of years at the target from remaining estimates and estimates at the threshold from those at the target. Five of six estimates were above the threshold during 1998-2001 and 2004, and could be separated from most (7 of 8) P0 estimates that fell between the target and threshold (Figure 7).

Overton et al. (2009) provided an estimate of the percent of Striped Bass in their large size class (501-700 mm, TL) with food during 1998-2000 (within the period of threshold P0) and we used this estimate (0.58) as a threshold PE for large sized fish (Figure 8). Estimates of PE of large Striped Bass during fall were at the threshold level in 2006, 2012, and 2017 based on 90% CI overlap. There was a modest association of PE and P0 ($r = 0.52$, $P = 0.047$) during 2006-2020 (Uphoff et al. 2022); review of the plot of these variables (not shown) indicated that P0 at the target level was more likely when PE was 0.34 or less (7 of 9 points) than above it (1 of 4). The PE target for large fish was set at 0.34 and was met during 2014-2015 and 2018-2021 (Figure 8).

Major prey accounted for 92.6% of diet items, by number, encountered in large Striped Bass diet samples during fall 2006-2021 (Figure 9). Atlantic Menhaden accounted for 48.7% when all years were combined (annual range =12.4-97.0%); Bay Anchovy, 14.9% (annual range = 0-32.5%); Spot, 7.6% (annual range = 0-52.4%); Blue Crab, 21.4% (annual range = 0-59.4%); and other items, 7.4% (annual range = 0-40.0%). Spot have represented a noticeably lower percentage of fall diet items since 2014. The "Other" category accounted for a higher fraction of large Striped Bass diets by number in 2012 and 2017 (36.2% and 40.0%, respectively) than remaining years (< 9.7%). During 2021, Atlantic Menhaden accounted for 89.7% of the diet items; Bay Anchovy, 3.8%; Spot, 0%; Blue Crab, 2.2%; and other items accounted for 4.3% (Figure 9). The vast majority of major prey fell within young-of-year length cut-offs (Uphoff et al. 2016).

By weight, Atlantic Menhaden predominated in large fish sampled (88.2% of diet weight during fall, 2006-2021, combined); Bay Anchovy accounted for 1.1%; Spot, 3.2%; Blue Crab, 3.5%; and other items, 4.3% (Figure 10). Estimates of C for large Striped Bass varied as much as 3.8-times among years sampled. The 2021 estimate of C of large fish (0.018) was above the time-series median (0.015; Figure 10).

Median PPLRs of large prey (Spot and Atlantic Menhaden) for large Striped Bass were 0.19-0.30 during 2006-2020 (Figure 11). The median PPLR was 0.24 for 2021 (Figure 11). Median PPLRs for large Striped Bass were much closer to the optimum (0.21 based on Overton et al. 2009) than for small fish.

Relative abundance indices of Striped Bass and major prey – Relative abundance of Striped Bass (RI) during 1981-2020 was lowest prior to 1994 (mean RI < 0.4 fish per trip; Figure 12). Estimates of RI then rose abruptly to a high level and remained there during 1995-2006 (mean = 2.6). Estimates of RI fell by about a third of the 1995-2006 mean during 2008-2013 (mean = 1.8), rose to 2.4-3.6 during 2014-2019 (2019 was the second highest of the time-series), before falling to 1.8 in 2020 and 1.4 in 2021. The 90% confidence intervals indicated that RI was much lower during 1981-1993 than afterward and that there was some chance that RI during 2008-2013 was lower than other years during 1995-2019. Ninety percent CIs of the 2021 estimate were similar to those for 2009 and 2011-2013 (Figure 12). The trend in RI compared favorably to the trend in estimated aggregate abundance of 2- to 5-year-old Striped Bass along the Atlantic Coast, particularly in the years after recovery was declared (Uphoff et al. 2020).

Major pelagic prey were generally much more abundant during 1959-1994 than afterward (Figure 13). Bay Anchovy seine indices following the early to mid-1990s were typically at or below the bottom quartile of indices during 1959-1993. Highest Bay Anchovy trawl indices occurred in 1989-1992, 2001-2002, and 2020-2021, while lowest indices occurred during 2006-2011 and 2015-2019. There was little agreement between the two sets of Bay Anchovy indices; however, there were few data points representing years of higher abundance in the years in common and contrast may have been an issue (comparisons were of mostly low abundance points). Atlantic Menhaden seine indices were high during 1971-1994 and much lower during 1959-1970 and 1995-2021. There has been a slight recent upward shift in Atlantic Menhaden seine indices from mostly their lowest sustained level during 1995-2012 (Figure 13)

Major benthic forage indices were low after the 1990s, but years of higher relative abundance were interspersed during the 2000s (Figure 14). Seine (1959-2021) and trawl (1989-2021) indices for Spot had similar trends that indicated high abundance during 1971-1994 and low abundance during 1959-1970 and after 1995 (with 3 or 4 years of higher indices interspersed). Spot indices in 2020 and 2021 were much better than the previous nine years. Blue Crab densities (1989-2019) were generally at or above the time-series median during 1989-1998, and 2009-2015. Blue Crab densities in 2020-2021 were the lowest of the time-series (Figure 14).

Most of the annual indices of biomass of soft bottom benthic invertebrates during 1995-2018 were well above the time-series median during 2000-2009 (Figure 15). Indices well below the median indices occurred during 1996, 1998, 2003-2004, 2012 and 2021. Biomass indices have been mostly below the time-series median since 2010 (Figure 15).

Species-specific standardized FRs exhibited similar general patterns during 1983-2021 (Figure 16). Indices were at their highest in the early1980s when Chesapeake Striped Bass were at their lowest level and fell steadily in the early 1990s as Striped Bass recovered and forage indices declined. A nadir in the ratios appeared during 1995-2004 (Striped Bass recovery was declared in 1995), followed by occasional "spikes" of Spot and Blue Crab ratios and a slight elevation in Atlantic Menhaden ratios after 2004. Forage ratios in 2021, with the exception of Blue Crab, were above their 1995-2021 medians (Figure 16). The Atlantic Menhaden FR has been generally elevated during 2005-2021 from its nadir during 1997-2004, but has been well below levels prior to the early 1990s (Figure 17). The Bay Anchovy seine FR was similar to years of higher FRs since 1995 (2006-2009 and 2010-2013; Figure 18). The Spot seine FR during 2020-2021 was in the higher portion of the range exhibited since 1995 (Figure 20).

Relative survival of small Striped Bass – Relative survival could not be estimated for 2021. Results for 1985-2020 are reported in Uphoff et al. (2022).

Discussion

Average condition of small and large Striped Bass was good (met target conditions) during 2021 and represented the best body fat indices for both size classes for the whole time-series. Small Striped Bass condition was consistently poor (breaching the threshold) during 1998-2007 and shifted to a mix afterward. During 2008-2021, there were four years where P0 of small fish met the target, four years that the threshold was exceeded, and six years in between. Condition of large Striped Bass was at its threshold in 6 of 7 years during 1998-2004 and has improved, only slightly missing its target once since 2014.

The P0 metric represents an integration of multiple factors that affect condition into a single measure. Lipids are the source of metabolic energy for growth, reproduction, and swimming for fish and energy reserves relate strongly to foraging success, reproductive success, potential prey density, habitat conditions, environmental stressors, and subsequent fish health and survival (Tocher 2003; Jacobs et al. 2013). It is important to note that our condition and diet samples are mostly from survivors of two to five years (depending on size and age) of some combination of feeding success, growth, environmental conditions, mycobacteriosis, and catch-and-release and harvest mortality that reduce abundance and intraspecific competition among Striped Bass. The summer preceding our fall monitoring may be particularly stressful and potentially lethal. Summer represented a period of no to negative growth in weight for ages 3-6 during 1990-1992 (Hartman and Brandt 1995b), higher mortality of diseased and healthy Striped Bass (Groner et al. 2018), hypoxia and temperature stress (Constantini et a. 2008; Maryland Sea Grant 2009; Coutant 2013; LaPointe et al. 2014; Kraus et al. 2015; Itakura et al. 2021), and high catch-and-release mortality (Lukacovic and Uphoff 2007). Condition of Striped Bass in summer was a good predictor of fall condition, and condition in fall of the previous year appeared related to condition in the next fall (Uphoff et al. 2017). If fewer fish make it through these hurdles, the survivors may benefit from reduced intraspecific competition for forage. The RI is a rather blunt indicator of resident abundance since it aggregates both large and small size groups and seems likely to be dominated by the small size class. Improvement in condition due to greatly reduced abundance of Striped Bass is not likely to be comforting to fishermen or managers.

Large Striped Bass have been mostly at target PE associated with target P0 since 2014. A target was not readily suggested for PE of small fish, but PE was clearly below the threshold during 2008-2010, 2014, and 2016-2021. Estimates of PE for large and small Striped Bass were modestly correlated (Uphoff et al. 2022) and both have generally improved from threshold conditions since 2006.

The PE metric is a simple and robust indicator of overall feeding success (Baker et al. 2014), but it can be biased by high frequency of small items that may not have much nutritional value or low frequency of large items with higher nutritional value and digestion times (Hyslop 1980). Additional information (numeric frequency of diet items and estimates of C) aids interpretation of PE.

Atlantic Menhaden dominated small and large Striped Bass diets by weight during fall; C has been higher since 2013, more frequently ranking in the top half of estimates. Bay Anchovy were dominant by number in small Striped Bass diets, but made up a low fraction of fall diet weight in all but the worst years. Small Blue Crabs were a minor component by weight as well, but were abundant in some years. Spot, a major prey that had contributed to lower PPLR of large major prey and achievement of target P0 and PE for small fish in 2010, have been largely absent in fall diets of both size classes. The seine and trawl FRs for Spot during 2010 were much higher than other years in either the body fat or consumption time-series and were within the range estimated for 1990 (year used as a target for P0; Jacobs et al. 2013).

Small Striped Bass condition has improved since the mid-2000s, but not as consistently as for large fish. The transition from small to large major prey may represent a bottleneck for small Striped Bass. Small Striped Bass would have more difficulty in catching and handling the same sized large major prey than large Striped Bass in any given year. Animal feeding in nature is composed of two distinct activities: searching for prey and handling prey (Yodzis 1994). Both can be influenced by prey size, with larger prey obtaining higher swimming speeds (typically a function of body length) that enable them to evade a smaller predator and larger size makes prey more difficult to retain if caught (Lundvall et al. 1999). With high size limits and low fishing mortality in place for Striped Bass since restoration, intraspecific competition for limited forage should be greater for small Striped Bass because they compete with one another and large Striped Bass. Striped Bass in our large category were uncommon in Maryland's Bay prior to restoration because of higher F and lower length limits; pound net length-frequencies in the 1960s-1970s rarely contained large fish (J. Uphoff, MD DNR, personal observation). In addition to being able to handle a wider size range of prey, large striped bass should forage more efficiently and outcompete small fish through greater vision, swimming speed, and experience (Ward et al. 2006). Below threshold P0 of small fish in 2016 and 2019 coincided with two large year-classes of Striped Bass having approached or reached the large size category (2011 year-class in 2016 and 2015 year-class in 2019).

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 benthic invertebrates other than Blue Crab are important diet items during winter and spring-early summer, respectively (Walter et al. 2003; Hartman and Brandt 1995c; Overton et al. 2009; 2015). These prey did not usually make a large contribution to diet mass during fall, but on occasion White Perch made a contribution to large Striped Bass C. The effect of other items consumed in other seasons would be incorporated into P0, but their contribution to P0 would be unknown, although it might be suspected from high P0 that seemed anomalous.

A rapid rise in Striped Bass abundance in Maryland's portion of the Bay during the mid-1990s, followed by a dozen more years at high abundance after recovery was declared in 1995, coincided with declines in relative abundance of Atlantic Menhaden, Bay Anchovy, Spot, and Blue Crab (i.e., major pelagic and benthic prey) to low levels. Changes in FRs largely reflected decreasing prey during 1983-1994 since RI was low. After 1995, prey indices stayed relatively low and RI increased; FR changes usually reflected fluctuations in RI. Striped Bass were often in poor condition during fall, 1998-2004, and vulnerable to starvation. Improvements in condition after 2007 coincided with lower Striped Bass abundance, spikes or slight increases in some major forage indices, and higher consumption of larger major prey (Spot and Atlantic Menhaden) in fall diets. A return of Striped Bass to high abundance after 2014 was not accompanied by greatly increased major forage, but it appears that slightly higher Atlantic Menhaden seine indices since 2007, while not always statistically distinguishable from indices during the 1998-2004 when threshold P0 was predominant, may have biological significance based on improvement in recent body fat and fall diet metrics.

Forage to Striped Bass ratios indexed potential attack success on major prey (Uphoff 2003; MD Sea Grant 2009). Atlantic Menhaden FR reached its nadir during 1995-2004 and has risen just above it since. The FRs for Atlantic Menhaden, Spot, and Bay Anchovy since 2005 have been well below those that occurred in 1990, the year used to set target conditions for P0 (Jacobs et al. 2013). Condition of both size classes improved after 2004, but improvement was steadier and more pronounced for large Striped Bass. Bay Anchovy were consistently present in fall diets of both size classes of Striped Bass during 2006-2014, but have fallen substantially as a percent of large fish diet since 2015-2018 (10-29%) to 0-4% in 2019-2021 as Atlantic Menhaden became frequent in their fall diet. Bay anchovy represented a variable percentage (22.7-87.9%) of small fish diets during fall 2006-2015 and had a steadier, higher frequency (65-90%) afterwards. Spot have made an insignificant contribution to fall diets of both size classes of Striped Bass since 2011 and Blue Crab have made a consistently smaller contribution to small Striped Bass diets since 2015. These changes since 2015 suggest the pelagic pathway is making a larger contribution to fall diets. Overton et al. (2015) described shifting prey dependence over time in Chesapeake Bay based on bioenergetics analyses of annual Striped Bass diets in the late 1950s, early 1990s, and early 2000s. By the early 2000s, there was a greater dependence on Bay Anchovy by all ages of Striped Bass and older fish had a greater dependence on the benthic component as Atlantic Menhaden declined in the diet (Overton et al. 2015). Stable isotope analyses of archived Striped Bass scales from Maryland's portion of Chesapeake Bay indicated an increasing shift from pelagic to benthic food sources during 1982-1997 (Pruell et al. 2003).

The soft bottom benthic index time-series covered 1995-2021 and changes prior to Striped Bass recovery and in the recent years could not be addressed. Benthic biomass has generally been lower since 2010. Changes in benthic invertebrate populations have the potential to affect Striped Bass directly or through reductions in benthic major prey. There was little indication of correspondence of the soft bottom benthic index to P0 of either size class of Striped Bass, but an exploratory analysis indicated a weak positive correlation of the two standardized Spot indices (combined into a single analysis) with the soft bottom index (1995-2018; $r = 0.31$, $P = 0.033$; Uphoff et al. 2022).

While top-down control of forage is suggested by opposing trends of major forage and Striped Bass, bottom-up processes may also be in play. A long-term decline Bay Anchovy in Maryland's portion of Chesapeake Bay (based on the seine index) was linked to declining abundance of the common calanoid copepod *Acartia tonsa* that, in turn, was linked to rising long-term water temperatures, eutrophication, and hypoxia (Kimmel et al. 2012; Roman et al. 2019; Slater et al. 2020). Non-predatory copepod mortality was higher under hypoxic conditions and implied a direct linkage between low dissolved oxygen and reduced copepod abundances (Slater et al. 2020). Houde et al. (2016) found Chl a and variables associated with freshwater flow, e.g. Secchi disk depth and zooplankton assemblages, were correlated with age-0 Menhaden abundance in the upper Bay. Variations in river flows to the Chesapeake Bay set up stratification, drive estuarine circulation, and cause fluctuations in inputs of freshwater, sediments, and nutrients, processes that greatly influence hypoxia (Hagy et al. 2004; Kemp et al. 2005; Maryland Sea Grant 2009). Woodland et al. (2021) demonstrated that bottom–up processes influenced fish and invertebrate forage in Chesapeake Bay (including our major forage species and benthic invertebrates included the BIBI based index; Blue Crabs were not examined). Annual abundance indices of many forage taxa were higher in years when spring water temperatures warmed slowly. Forage indices also were related (in taxon-specific ways) to winter–spring chlorophyll concentration and freshwater discharge, and to three summer water quality variables: dissolved oxygen, salinity, and water temperature, in addition to a broad-scale climate indicator (Atlantic Multidecadal Oscillation or AMO; Woodland et al. 2021). The AMO was the best single predictor of recruitment patterns of Atlantic Menhaden in Chesapeake Bay and along the Atlantic coast, suggesting that broad-scale climate forcing was an important controller of recruitment dynamics, although the specific mechanisms were not identified (Buccheister et al. 2016). The MD Spot seine index was negatively and weakly correlated with the AMO (January-April mean; r = -0.41, P = 0.0012, 1957-2017; J. Uphoff, unpublished analysis).

A hypoxia-based hypothesis, originally formed to explain die-offs of large adult Striped Bass in southeastern reservoirs, links increased M and deteriorating condition in Chesapeake Bay through a temperature-oxygen squeeze (mismatch of water column regions of desirable temperature and dissolved oxygen in stratified Chesapeake Bay during summer; Coutant 1985; Price et al. 1985; Coutant 1990; Coutant 2013). Constantini et al. (2008); Kraus et al. 2015; Itakura et al. 2021) examined the impact of hypoxia on 2-year-old and older Striped Bass in Chesapeake Bay through bioenergetics modeling and acoustic tagging and concluded that a temperature-oxygen squeeze by itself was not limiting for Striped Bass. However, Groner et al (2018) suggested that Striped Bass are living at their maximum thermal tolerance and that this is driving increased mycobacteriosis and associated mortality. Adequate levels of Striped Bass prey can offset negative effects of warm temperatures and suboptimal dissolved oxygen in reservoirs (Thompson et al. 2010; Coutant 2013).

Multiple lines of evidence suggest that survival of both small and large Striped Bass decreased in Chesapeake Bay due to higher M since the late 1990s. Higher frequency of below time-series (1985-2020) median SR between ages 0 and 3 after 1996 was concurrent with declines in conventional tag-based estimates of survival of 457-711 mm of Striped Bass in Chesapeake Bay (based on time varying estimates of M; Uphoff et al. 2022). Annual survival decreased from 77% during 1987-1996 to 44% during 1997-2017, a 43% reduction (based on Table B8.25 in NEFSC 2019); estimates of F in Chesapeake Bay from tagging have been low and estimates of M have been high (NEFSC 2019). Secor et al. (2020) implanted a size-stratified sample of Potomac River Striped Bass with acoustic transmitters and recorded their migrations during 2014-2018 with telemetry receivers throughout the Mid-Atlantic Bight and Southern New England. Analysis of the last day of transmission indicated that Chesapeake Bay resident Striped Bass experienced lower survival (30% per year) than coastal shelf emigrants (63% per year; Secor et al. 2020).

Decreased survival of large Striped Bass estimated from conventional tags during 1987-1996 and 1997-2017 in NEFSC (2019) was attributed to mycobacteriosis. Mycobacteriosis alone would not necessarily be the only source of increased M of Chesapeake Bay Striped Bass.

Jacobs et al. (2009b) were able to experimentally link the progression of mycobacterial disease in Striped Bass to their diet: inadequate diet led to more severe disease progression compared with a higher ration. In addition, abundant individuals competing for limited prey may hinder one another's feeding activities, leading directly to starvation (Yodzis 1994). Shifts from high survival during 1987-1996 to lower survival afterwards (Kahn and Crecco 2006; Jiang et al. 2007; ASMFC 2013; NEFSC 2019) lagged two years behind downward shifts in forage-to-Striped Bass ratios. Dutil and Lambert (2000) found that the response of M of Atlantic Cod (*Gadus morhua*) 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). Recovery of the northern stock of Atlantic Cod has paralleled recovery of Capelin (*Mallotus villosus*), its main prey (Rose and Rowe 2015); increases in size composition and fish condition and apparent declines in mortality followed. Condition of both size classes of resident Striped Bass has improved since the mid-2000s in concert with slight improvement in Atlantic Menhaden FR and consumption. No other major prey FR (or benthic invertebrate biomass) matches this timing. 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).

The fall in survival in the mid-to-late 1990s 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, may not be 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 may have serious implications for interstate management since Chesapeake Bay is the main contributor to Atlantic coast fisheries (Richards and Rago 1999; NEFSC 2019). 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 F, and compensatory M would undercut both objectives.

Long-term analyses of M based on conventional tags indicated survival of large Striped Bass decreased after stock recovery (NEFSC 2019), but the time blocks analyzed were large and only differentiated two periods (pre- and post-1997), the former of low M and latter of high M. A finer temporal resolution of M estimates is needed to relate forage or other conditions to survival of large fish. Survival of small Striped Bass in Chesapeake Bay has not been explored with conventional or acoustic tags.

Catch-and-release mortality different from that assumed in NEFSC (2019) could have confounded estimation of M from tagging experiments. Increases in conventional tag-based estimates of M of legal-sized fish over time could also reflect misspecification of parameters such as tag reporting rates that make absolute estimates less reliable (NEFSC 2019); however, M estimates based on acoustic tags (not subject to reporting rates) produced similar differences in mortality of coastal migrants and Chesapeake Bay residents (Secor et al. 2020).

Hook-and-line samples collected by CBEF (2006-2013) and FWHP (2014-2021) were treated as a single time-series. Sampling by CBEF stopped in 2015 due to failing health of Mr. Price (CBEF President and organizer of the CBEF diet survey). Samples were collected by both programs during 2014, providing an opportunity for comparison (Uphoff et al. 2018). Sizes of Striped Bass sampled by the two programs were comparable and estimates of P0 were similar. Fall diets were dominated by Atlantic Menhaden and Spot were absent in both cases. Differences arose in smaller major prey, particularly Bay Anchovy, and in the importance of "Other" prey (Uphoff et al. 2018). There has not been a readily discernable shift in patterns of PE, C, and frequency of diet items by number that would be readily attributed to changes from CBEF to FWHP sampling programs.

The CBEF conducted a year-round diet sampling program useful to MD DNR free of charge, but this level of sampling could not be maintained by FHEP staff due to existing duties. Piggybacking diet sampling onto the existing fall FWHP Striped Bass health survey provided a low-cost alternative that would provide information on Striped Bass condition and relative availability of major prey, but would not characterize the annual diet or condition changes within a year. Consumption based indices of prey availability in fall (PE and C) appeared to be more sensitive and biologically significant (i.e., were reflected by P0) than FRs based on relative abundance indices (Uphoff et al. 2022).

We treated hook-and-line samples in fall as random samples (Chipps and Garvey 2007) rather than as cluster samples (Rudershausen et al. 2005; Hansen et al. 2007; Overton 2009; Nelson 2014), i.e., individual fish rather than a school were considered the sampling unit. This choice reflected changing feeding behavior of Striped Bass in fall and the nature of hook-and-line fishing for them. 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). Striped Bass leave the structures they occupied during summer-early fall and begin mobile, aggressive, open water feeding. Forage begins to migrate out of the Bay and its tributaries (and refuges therein) or to deeper water at this time and are much more vulnerable to predation. Both major forage and Striped Bass schools are constantly moving and changing. Schools of Striped Bass and their prey no longer have a fixed location, presenting well mixed populations (J. Uphoff, MD DNR, personal observation) that made a random sampling assumption reasonable. Treating hook-and-line samples as a cluster required a broad definition of a cluster in Overton et al. (2009), i.e., an entire day's effort that assumed fish caught that day represented a non-independent sample. Neither assumption (random or cluster) provided a complete description of how hook-and-line sampling works and we believed that random sampling was a better fit.

Two additional objectives of this forage assessment are low cost and tractability for available staff. Ecosystem based fisheries management has been criticized for poor tractability, high cost, and difficulty in integrating ecosystem considerations into tactical fisheries management (Fogarty 2014). It has been the principal investigator's unfortunate experience that complex and comprehensive ecosystem-based approaches to fisheries management for the entire Chesapeake Bay i.e., Chesapeake Bay Ecopath with Ecosim and Maryland Sea Grant's Ecosystem Based Fisheries Management for Chesapeake Bay (Christensen et al. 2009; MD Sea Grant 2009) have not gained a foothold in Chesapeake Bay's fisheries management. This is not surprising. While policy documents welcome ecosystem-based approaches to fisheries management and a large number of studies that have pointed out the deficiencies of single-species management, a review of 1,250 marine fish stocks worldwide found that only 2% had included ecosystem drivers in tactical management (Skern-Mauritzen et al. 2016).

The index-based forage assessment approach represents a less complex, low-cost attempt to integrate forage into Maryland's Striped Bass management. Given the high cost of implementing new programs, we have used information from existing sampling programs and indices (i.e., convenience sampling and proxies for population level estimates, respectively; Falcy et al. 2016). This trade-off is very common in fisheries and wildlife management (Falcy et al. 2016).

We used available estimates of central tendency and variability for ratio simulations. We did not attempt to standardize indices to account for influences such as latitude, date, and temperature. Use of standardizing techniques that "account" for other influences have increased, but they require additional staff time and often barely have a detectable effect on trends. Maunder and Punt (2004) described that their effect "can be disappointingly low" and they do not guarantee removal of biases.

Forage indices and forage to Striped Bass ratios were placed on the same scale by dividing them by arithmetic means over a common time period (ratio of means). Conn (2009) noted in several scenarios that arithmetic mean of scaled indices performed as well as the single index estimated by a hierarchal Bayesian technique. Falcy et al. (2016) found that ratios of means provided a reasonable method for combining indices into a composite index to be calibrated with population estimates of Chinook Salmon *Oncorhynchus tshawytscha*, but there was no one optimal method among the four techniques applied.

We have revised our original approach that integrated information for both size classes to one where each size class is evaluated separately. We felt important differences in forage dynamics between size classes were being lost by integrating them. The switch to size specific metrics complicated interpretation of other metrics that encompassed both size classes and could not be split. At this point, it is not apparent how to integrate these metrics, but they are reported and available for review. For this report, the two metrics with targets and thresholds (P0 and PE), hopefully, can alert busy fisheries managers and stakeholders about the status of forage and whether forage concerns merit further attention.

By splitting into small and large fish size classes, the P0 and PE metrics represent four pieces of information. The science of decision making has shown that too much information can lead to objectively poorer choices (Begley 2011). The brain's working memory can hold roughly seven items and any more causes the brain to struggle with retention. Proliferation of choices can create paralysis when the stakes are high and information is complex (Begley 2011).

The P0 and PE targets and thresholds represent a framework for condensing complex ecological information so that it can be communicated simply to decision makers and stakeholders. The target, threshold, or in-between status approach for P0 and PE was similar to traffic light style representations (but without the colors) for applying the precautionary approach to fisheries management (Caddy 1998; Halliday et al. 2001). Traffic light representations can be adapted to ecosystem-based fisheries management (Fogarty 2014). The strength of the traffic light method is its ability to take into account a broad spectrum of information, qualitative as well as quantitative, which might be relevant to an issue (Halliday et al. 2001). It has three elements – a reference point system for categorization of indicators, an integration algorithm, and a decision rule structure based on the integrated score (Halliday et al. 2001). In the case of P0 and PE, it contains the first two elements, but not the last. Decision rules would need input and acceptance from managers and stakeholders.

Some form of integration of indicator values is required in the traffic light method to support decision making and simplicity and communicability are issues of over-riding importance (Halliday et al. 2001). Integration has two aspects, scaling the indicators to make them comparable (target, threshold, or in-between status in our case) and applying an operation to summarize the results from many indicators. Caddy (1998) presented the simplest case for single-species management where indicators were scaled by converting their values to traffic lights (red, yellow, and green), and decisions were made based on the proportion of the indicators that were red. In 2021, the P0 and PE indicators for both size classes would not have been red; the three indicators with target and limit values would have been green.

Recent discussions with DNR fisheries managers and stock assessment scientists have indicated a preference for a stoplight approach for forage assessment in Maryland's portion of Chesapeake Bay based on time-series lower quartiles and medians. This approach is currently being developed for portraying the status of ages 1+ Atlantic Menhaden in Maryland's portion of the Bay and the P0 metric for large Striped Bass is part of this assessment. We will report on progress on applying this approach to some or all of the metrics reported for F-63 in the next annual report.

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Table 1. Important abbreviations and definitions.

Abbreviation	Definition
@Risk	Software used to simulate confidence intervals of ratios
C	Grams of prey consumed per gram of Striped Bass, an indicator of feeding success and prey availability.
CBEF	Chesapeake Bay Ecological Foundation.
CI	Confidence interval.
CPUE3	Unmodified gill net index of relative abundance of age 3 male Striped Bass.
CV	Coefficient of variation.
F	Instantaneous annual fishing mortality rate.
FR	Mean major forage ratio score (mean of scores assigned to standardized major prey to Striped Bass ratio
FWHP	Fish and Wildlife Health Program
HI	Hybrid gill net index of relative abundance of age-3 male Striped Bass that has been adjusted for catchability change with population size.
IF	Forage index. Mean score for five indicators of forage status (FR, PE, P0, RI, and SR)
JI	Juvenile index of relative abundance of a species.
M	Instantaneous annual natural mortality rate.
MRIP	Marine Recreational Information Program
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).
PPLR	Ratio of prey length to predator length.
q	Catchability (efficiency of a gear).
RI	Catch (number harvested and released) of Striped Bass per private and rental boat trip, a measure of relative abundance.
SR	Relative survival index for small sized resident Striped Bass to age-3.

Table 2. Number of dates sampled and number of small (<457 mm, TL) and large sized Striped Bass collected for October-November diet information in each size category, by year. Diet collections were made by Chesapeake Bay Ecological Foundation (CBEF) during 2006-2013 and MD DNR Fish and Wildlife Health Program (FWHP) after 2013. Start date indicates first date included in estimates of P0, PE, C, and diet composition and end date indicates the last.

Year	N dates	Small N	Large N	1st date	Last date	Source
2006	19	118	49	2-Oct	26-Nov	CBEF
2007	20	76	203	4-Oct	29-Nov	CBEF
2008	15	29	207	4-Oct	25-Nov	CBEF
2009	17	99	240	3-Oct	25-Nov	CBEF
2010	22	112	317	9-Oct	29-Nov	CBEF
2011	19	74	327	1-Oct	26-Nov	CBEF
2012	11	47	300	7-Oct	30-Nov	CBEF
2013	14	191	228	3-Oct	18-Nov	CBEF
2014	7	121	84	2-Oct	12-Nov	FWHP
2015	8	174	173	24-Sep	17-Nov	FWHP
2016	12	165	260	3-Oct	16-Nov	FWHP
2017	9	271	52	2-Oct	13-Nov	FWHP
2018	6	260	87	3-Oct	28-Nov	FWHP
2019	8	135	90	1-Oct	19-Nov	FWHP
2020	10	116	120	7-Oct	19-Nov	FWHP
2021	8	126	185	14-Oct	30-Nov	FWHP

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 during 2006-2013 (these regions were one in the same after 2013. Patuxent River seine stations are not included in analyses.

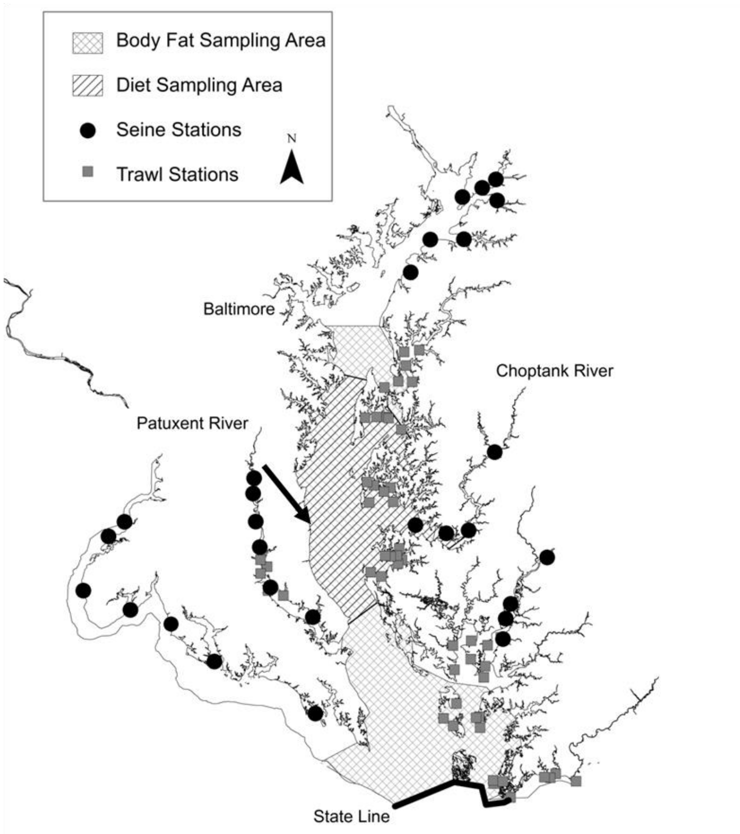

Figure 2. Proportion of small Striped Bass without body fat (P0) during October-November (MD DNR Fish and Wildlife Health Program monitoring) and its 90% confidence interval, with body fat targets (best condition) and thresholds (poorest condition).

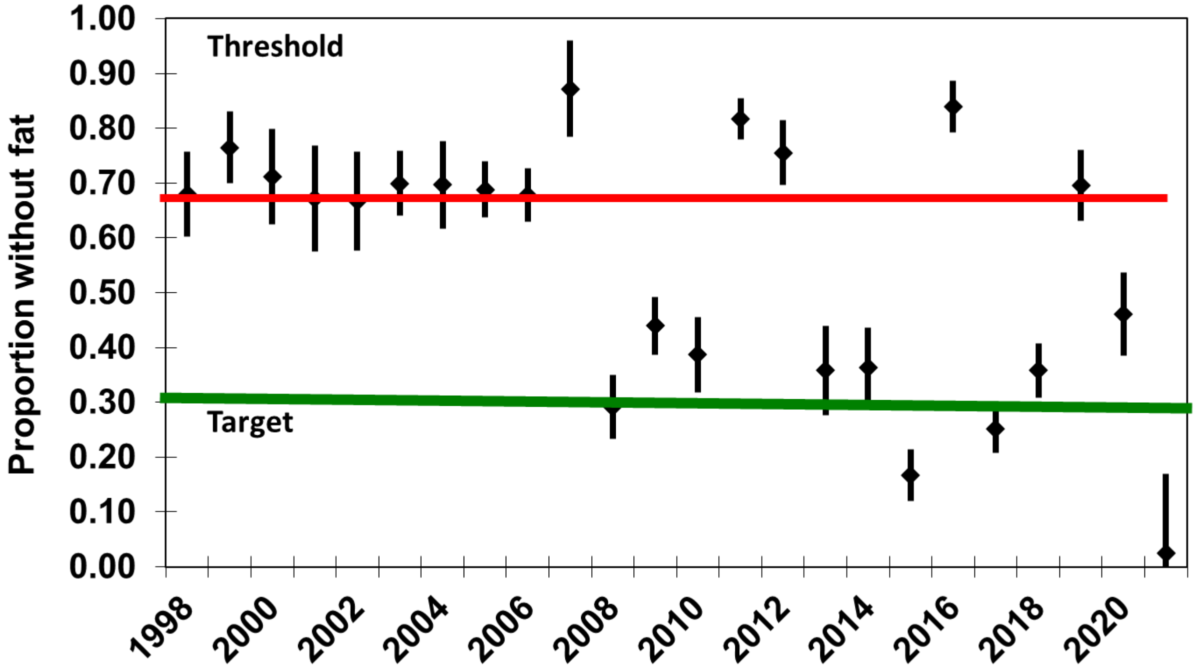

Figure 3. Proportion of small Striped Bass guts without food (PE) in fall and its 90% confidence interval. Red diamond represents threshold PE.

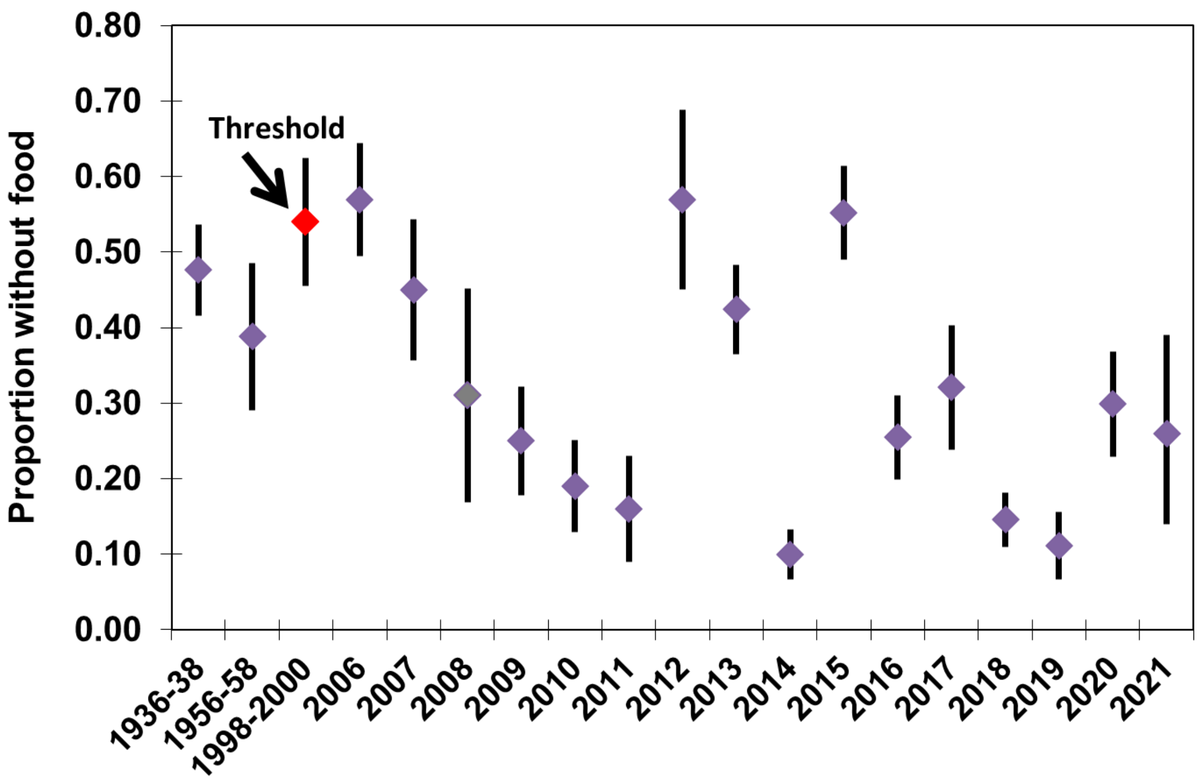

Figure 4. Percent, by number (counts of individuals plus presence of parts), of identifiable (excludes unknown) major forage groups in small Striped Bass (< 457 mm TL) guts, in fall.

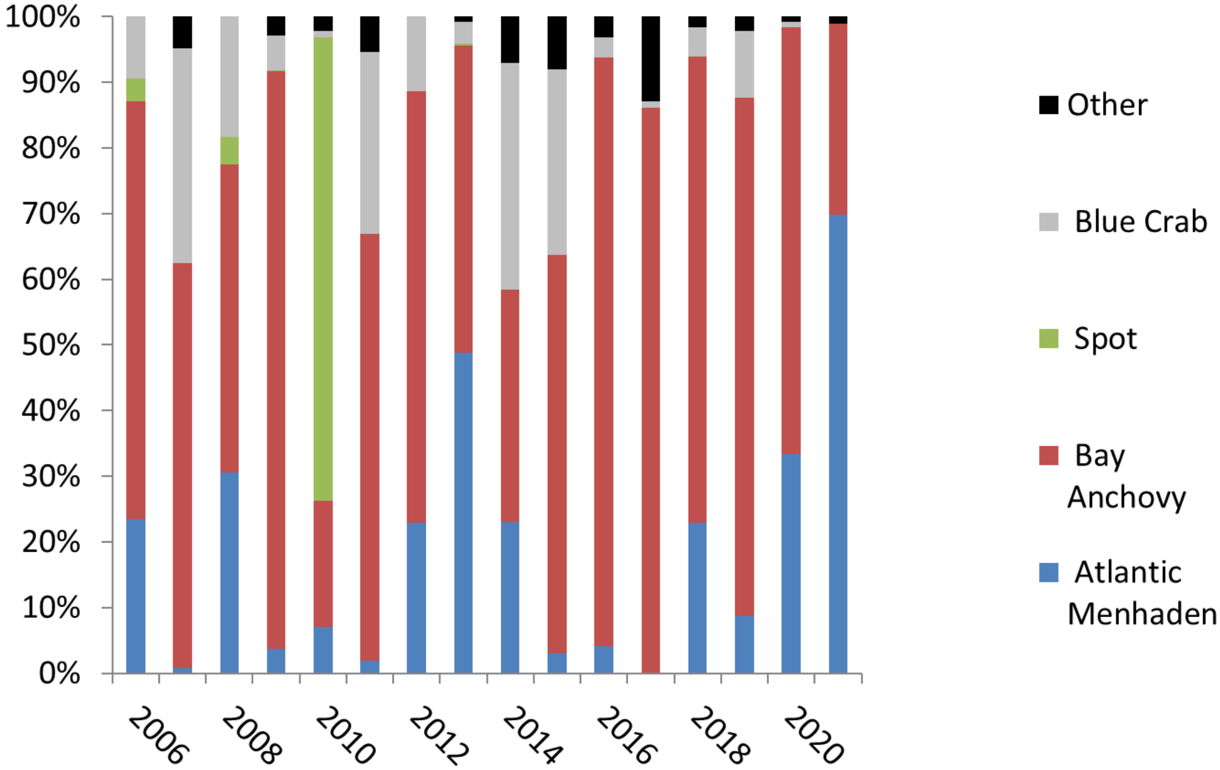

Figure 5. Gram prey consumed per gram (C) of small (< 457 mm TL) Striped Bass in fall hook-and-line samples. Age-0 forage dominate the diet. Arrow indicates color representing Atlantic Menhaden which disappeared on the figure legend.

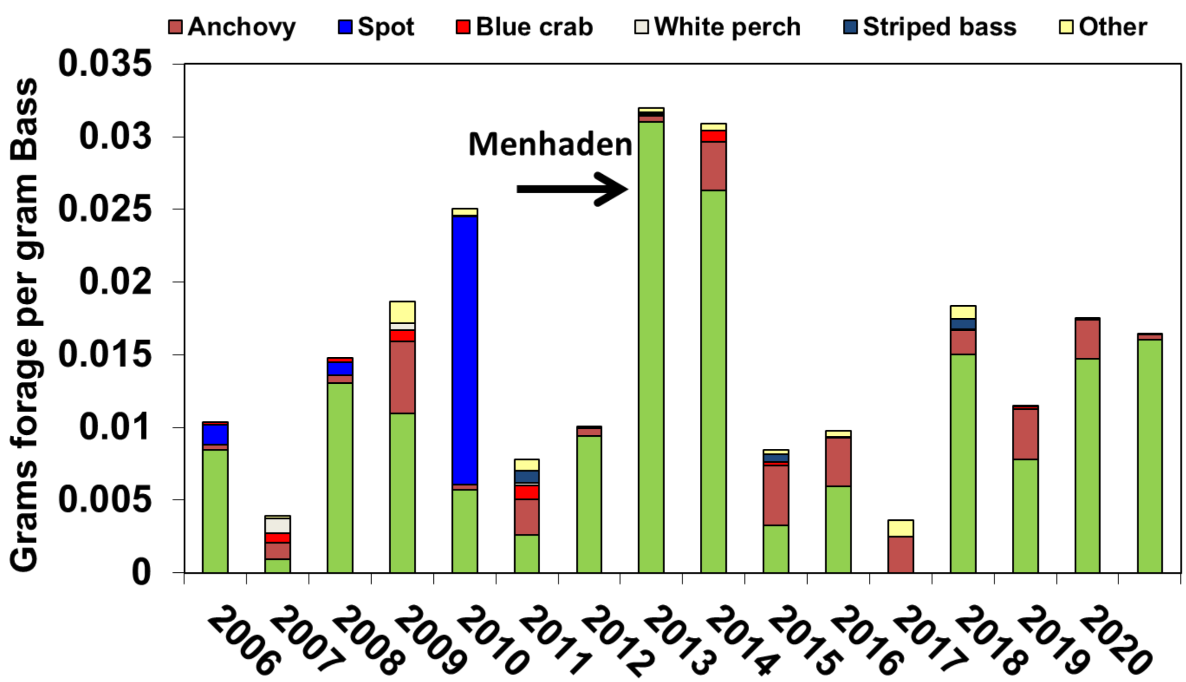

Figure 6. Median prey-predator length ratios (PPLR) for large major prey (Spot and Atlantic Menhaden) for small (< 457 mm) Striped Bass. Optimum ratio was estimated by Overton et al. (2009).

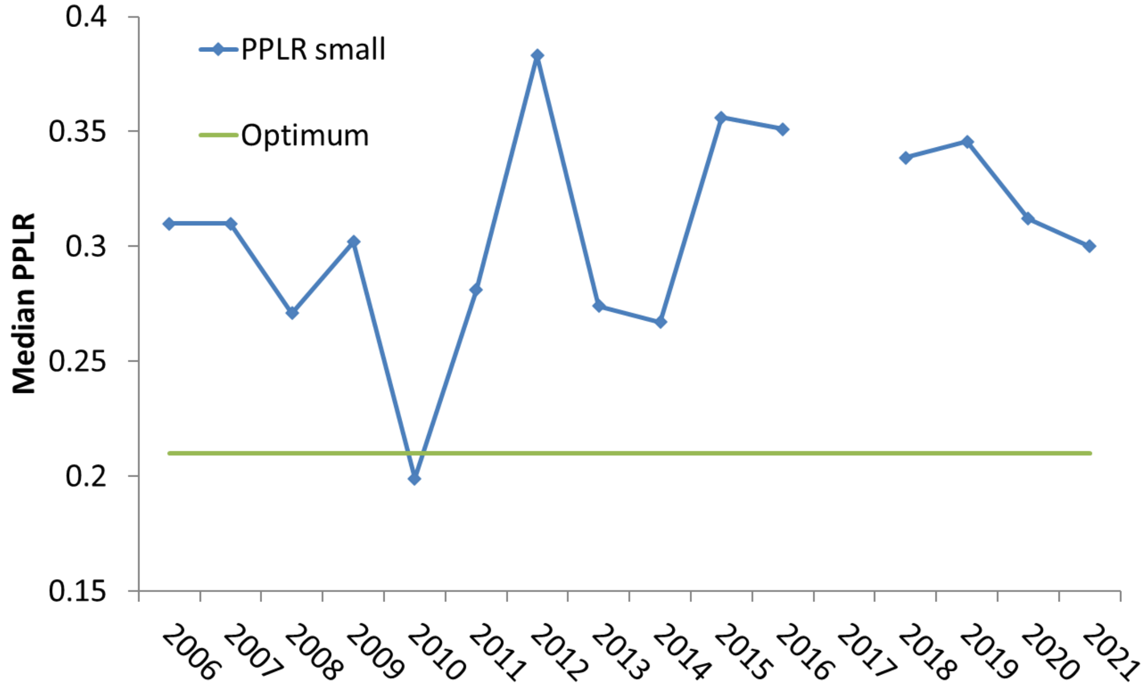

Figure 7. Proportion of large Striped Bass without body fat (P0) during October-November (MD DNR Fish and Wildlife Health Program monitoring) and its 90% confidence interval, with body fat targets (best condition) and thresholds (poorest condition).

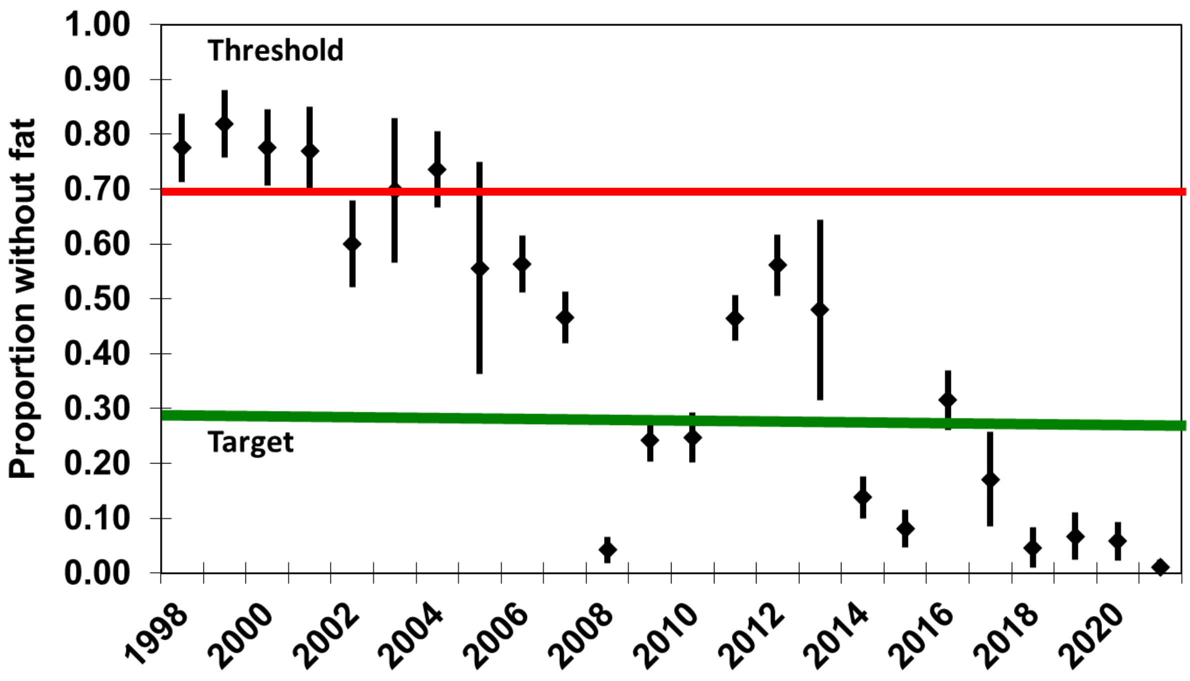

Figure 8. Proportion of large Striped Bass (≥ 457 mm or 18 in, TL) guts without food (PE) in fall and its 90% confidence interval, with body fat targets (best condition) and thresholds (poorest condition).

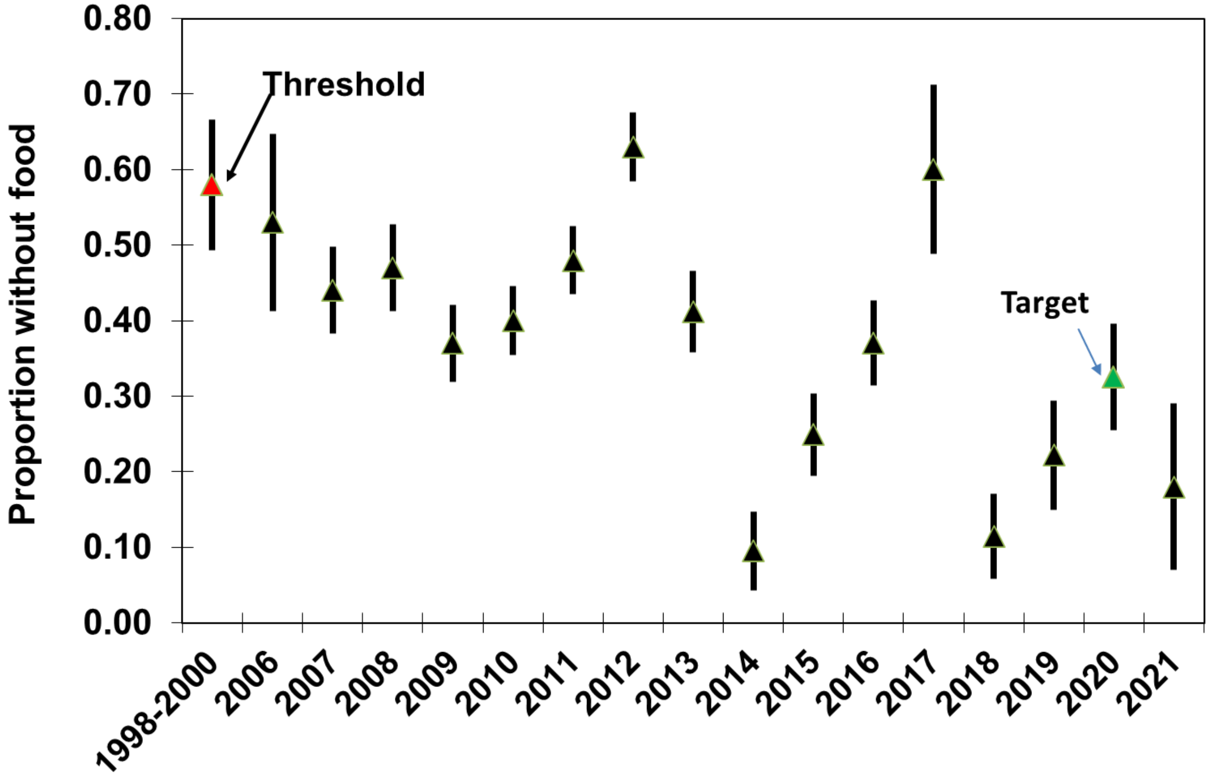

Figure 9. Percent of large Striped Bass (≥ 457 mm TL) identifiable diet represented by major forage groups, by number, in fall.

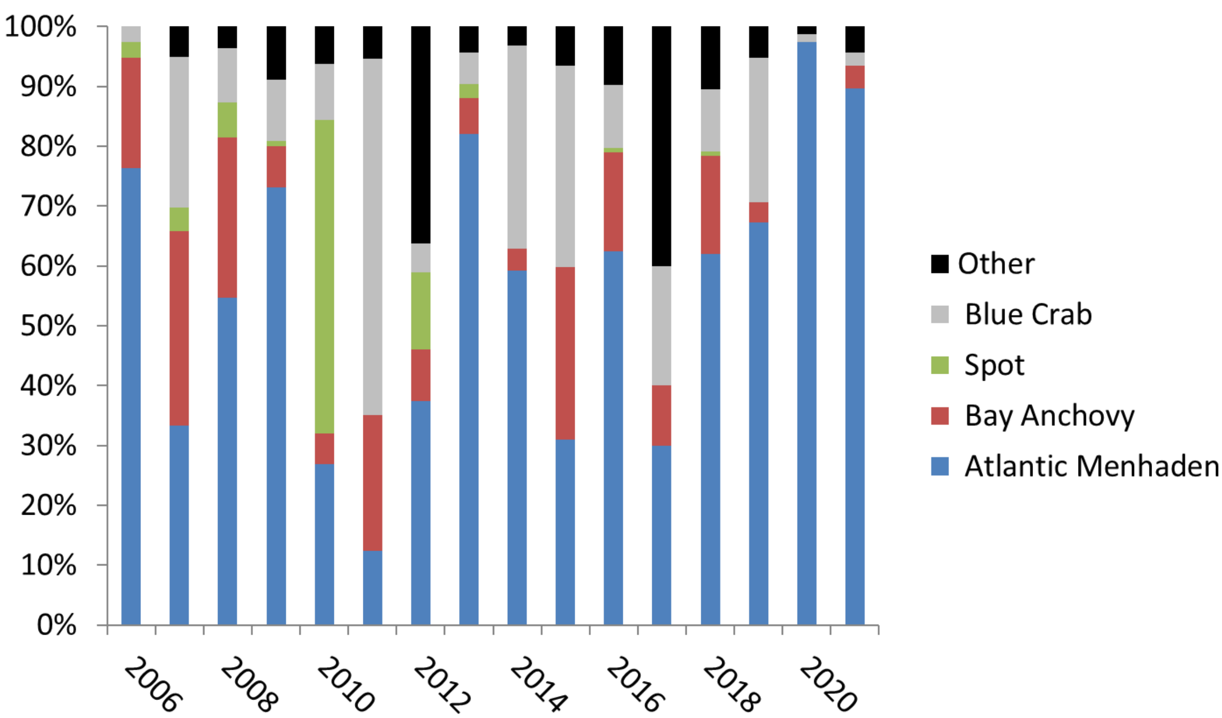

Figure 10. Grams of prey consumed per gram (C) of large (≥ 457 mm TL) Striped Bass during October-November. Fall consumption dominated by age 0 forage. Arrow indicates color representing Atlantic Menhaden which disappeared on the figure legend.

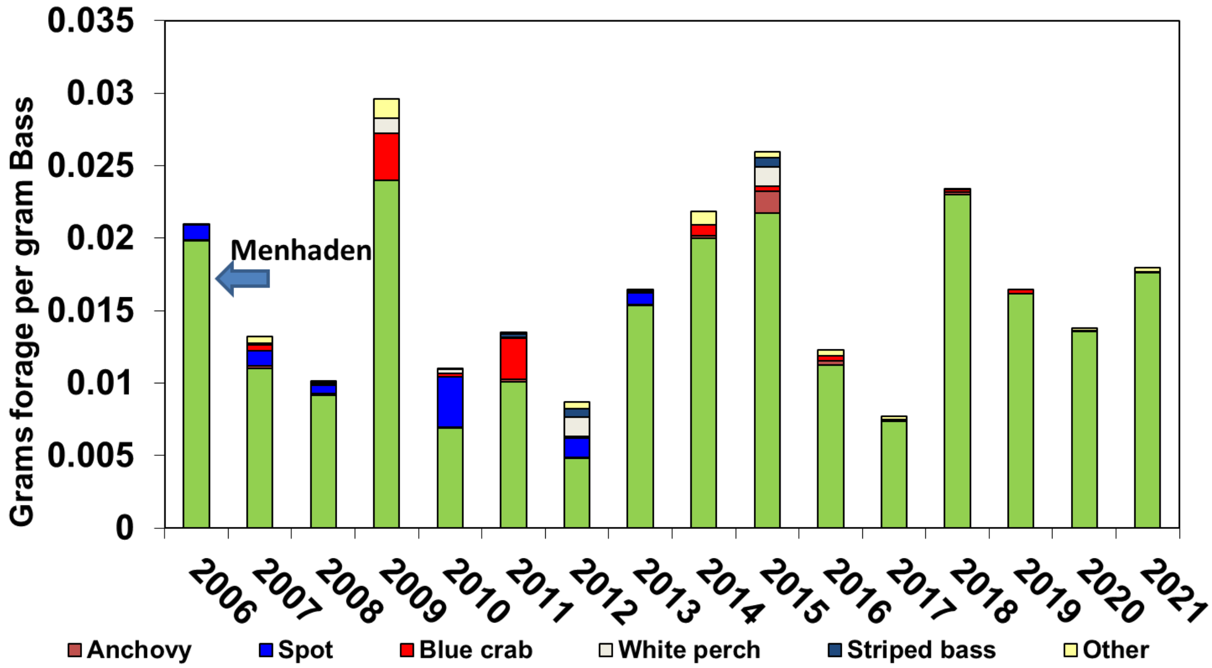

Figure 11. Median prey-predator length ratios (PPLR) for large major prey (Spot and Atlantic Menhaden) for large Striped Bass (≥ 457 mm). Optimum ratio was estimated by Overton et al. (2009).

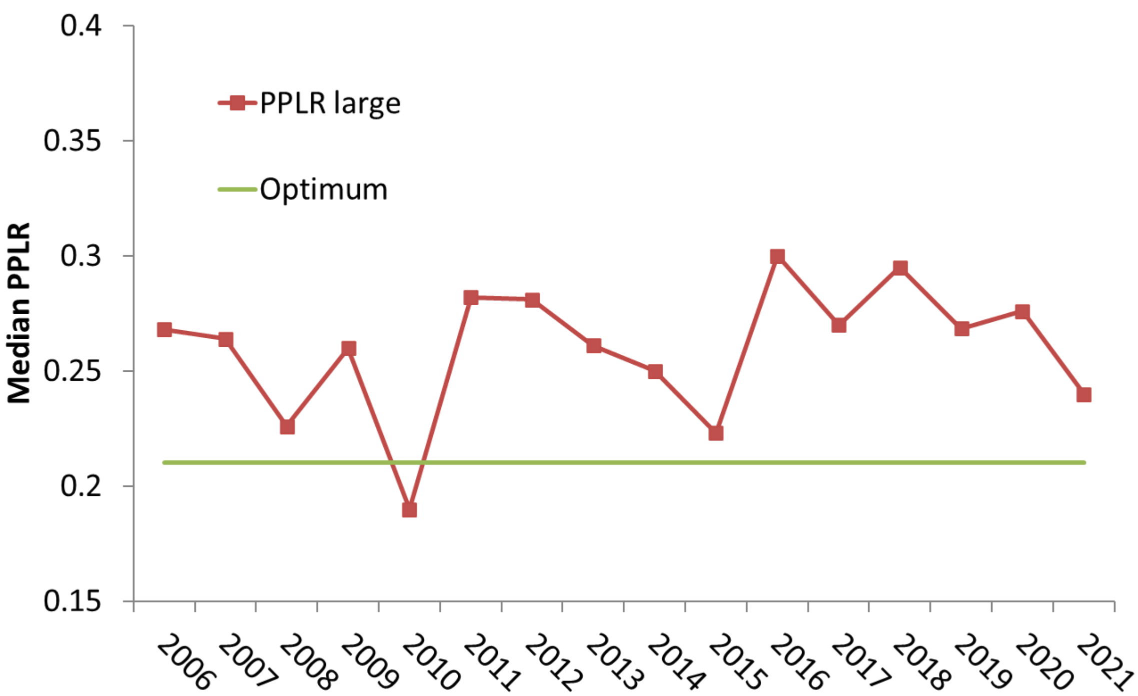

Figure 12. Maryland resident Bay Striped Bass annual abundance index (RI; MD MRIP inshore recreational catch per private boat trip during September-October; mean = black line) during 1981-2020 and its 90% confidence intervals based on @Risk simulations of catch and effort distributions. Catch = number harvested and released.

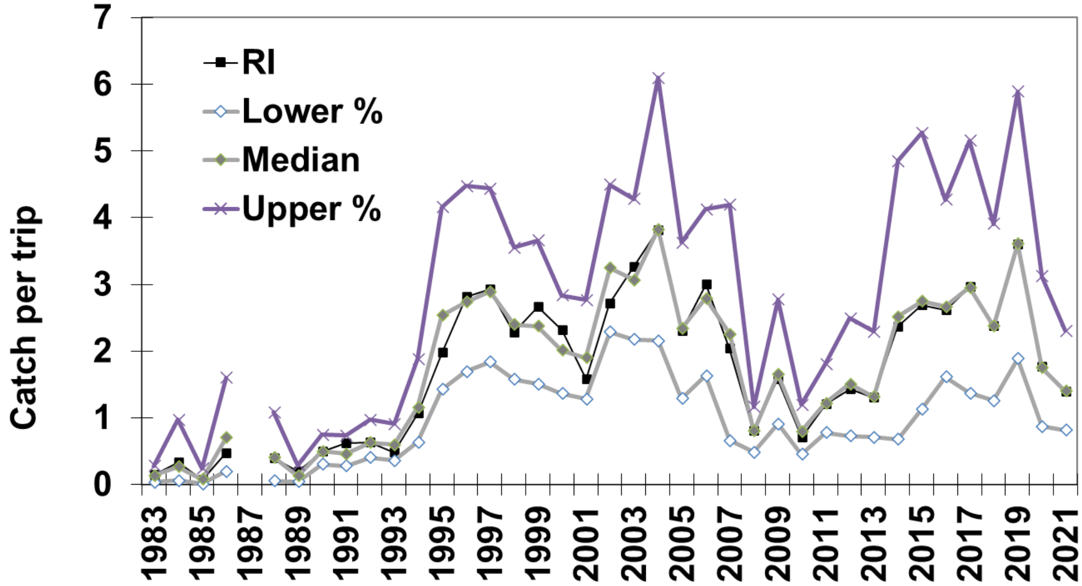

Figure 13. Trends in major pelagic prey of Striped Bass in Maryland Chesapeake Bay surveys since 1959. Indices were standardized to their 1989-2021 means (years in common). Menhaden = Atlantic Menhaden and Anchovy = Bay Anchovy.

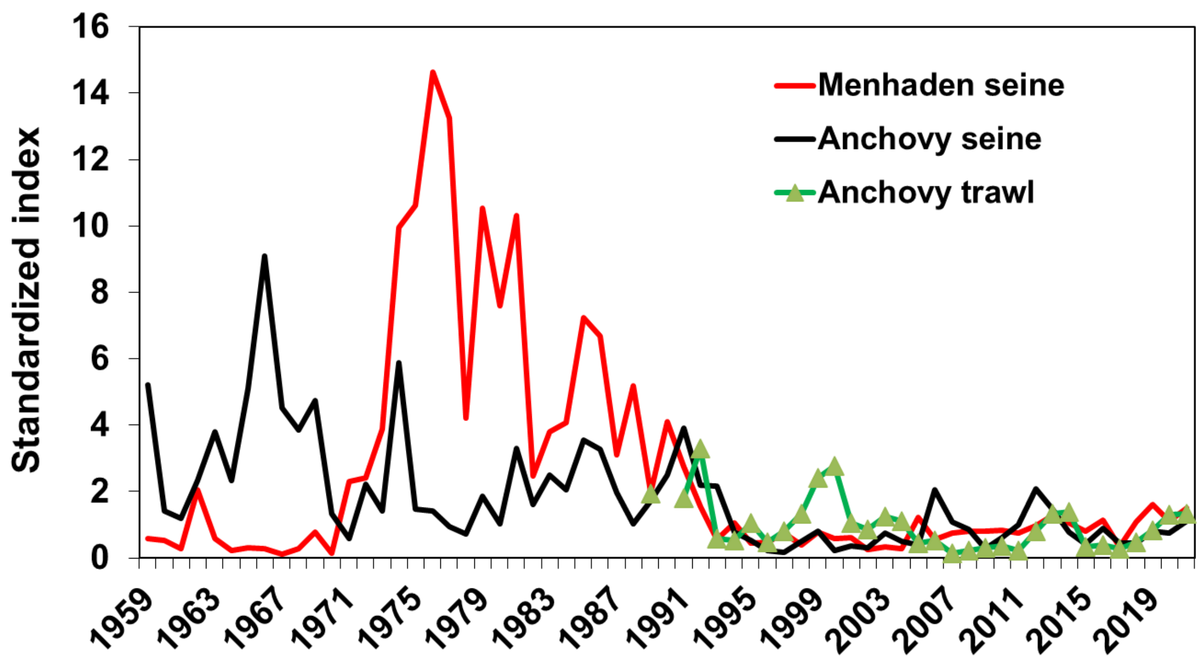

Figure 14. Trends in major benthic prey of Striped Bass in Maryland Chesapeake Bay surveys, since 1959. Indices were standardized to their 1989-2021 means (years in common).

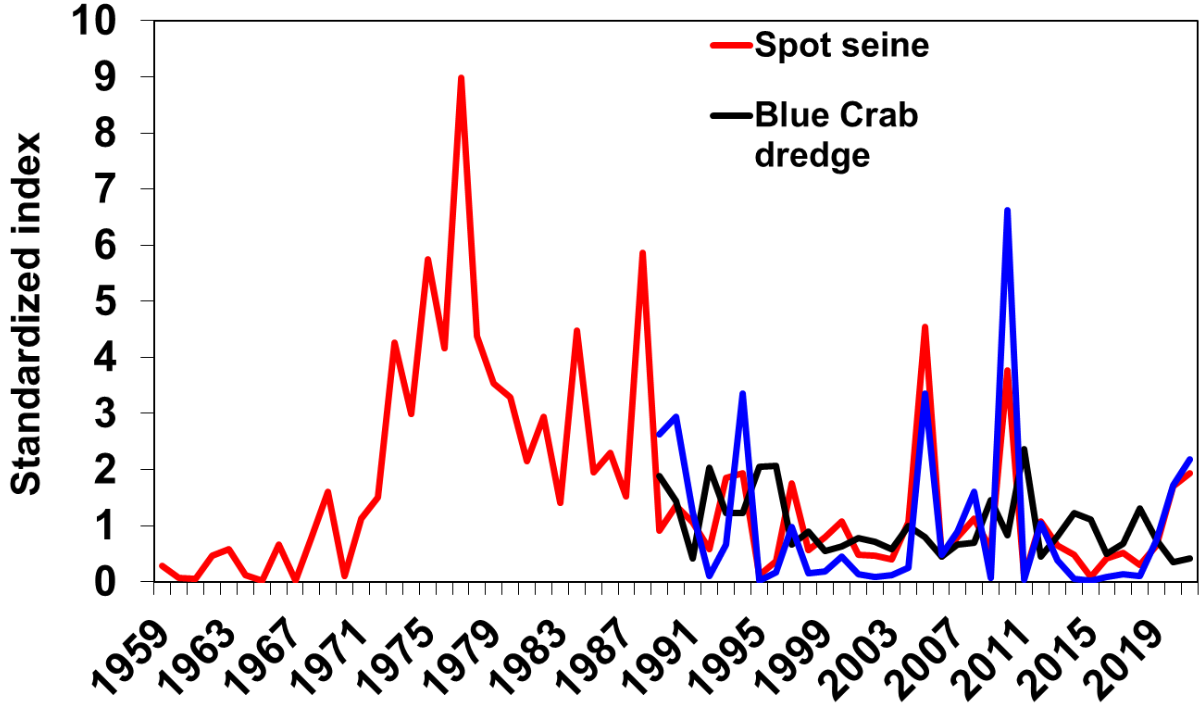

Figure 15. Trends in soft bottom benthic invertebrate biomass in Maryland waters (grams / m^2) and its median during 1995-2021 (based on Figure 3-37 in Versar Inc 2022).

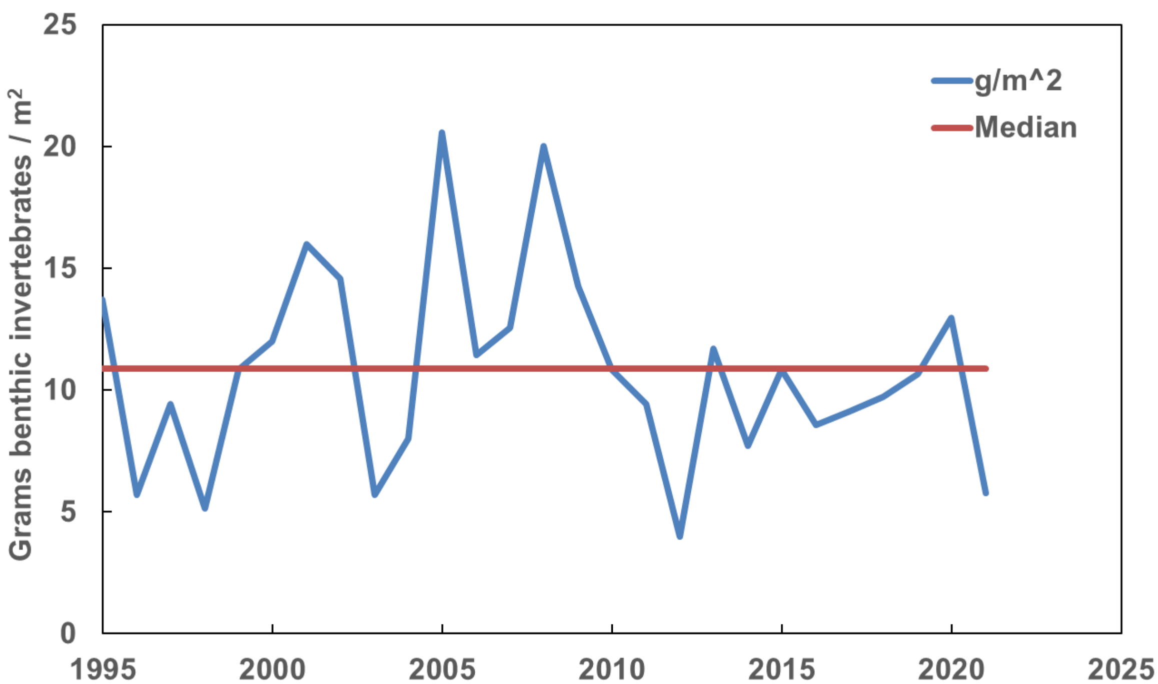

Figure 16. Trends of standardized ratios of major upper Bay forage species indices to Striped Bass relative abundance (RI). Forage ratios have been standardized to their 1989-2021 mean to place them on the same scale. S indicates a seine survey index; T indicates a trawl survey index; and D indicates a dredge index. Note the $\log_{10}$ scale on Y-axis.

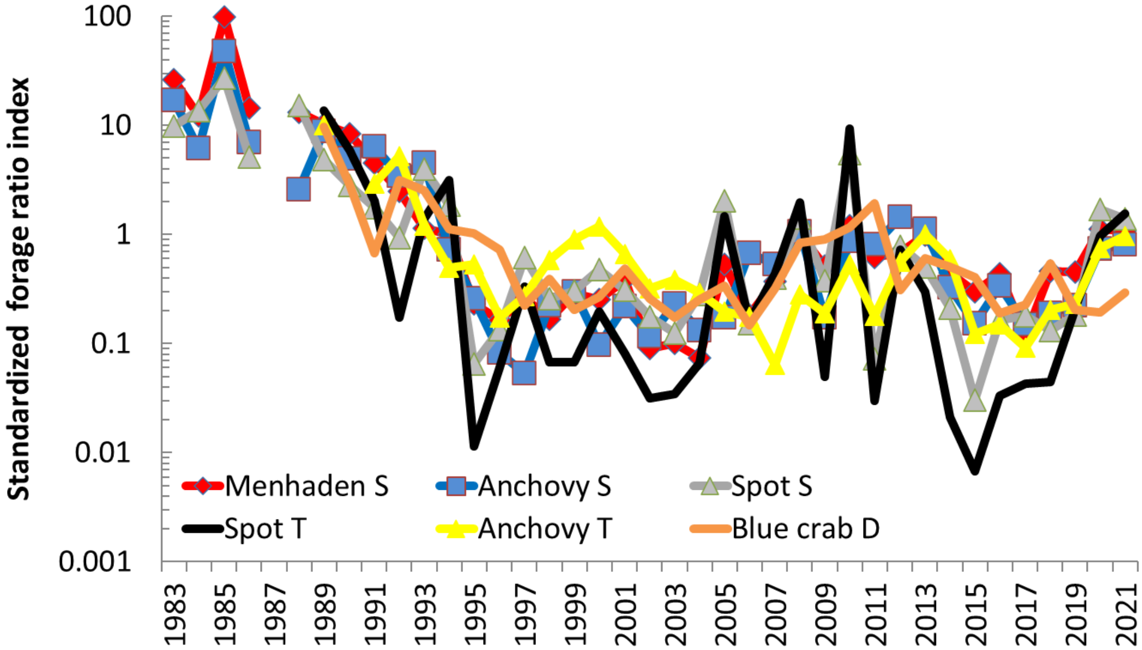

Figure 17. Atlantic Menhaden index to Striped Bass index (RI) ratios (Atlantic Menhaden FR) since 1983 and their 90% confidence intervals based on @Risk simulations of Atlantic Menhaden seine indices and RI distributions. Note $\log_{10}$ scale on the Y-axis.

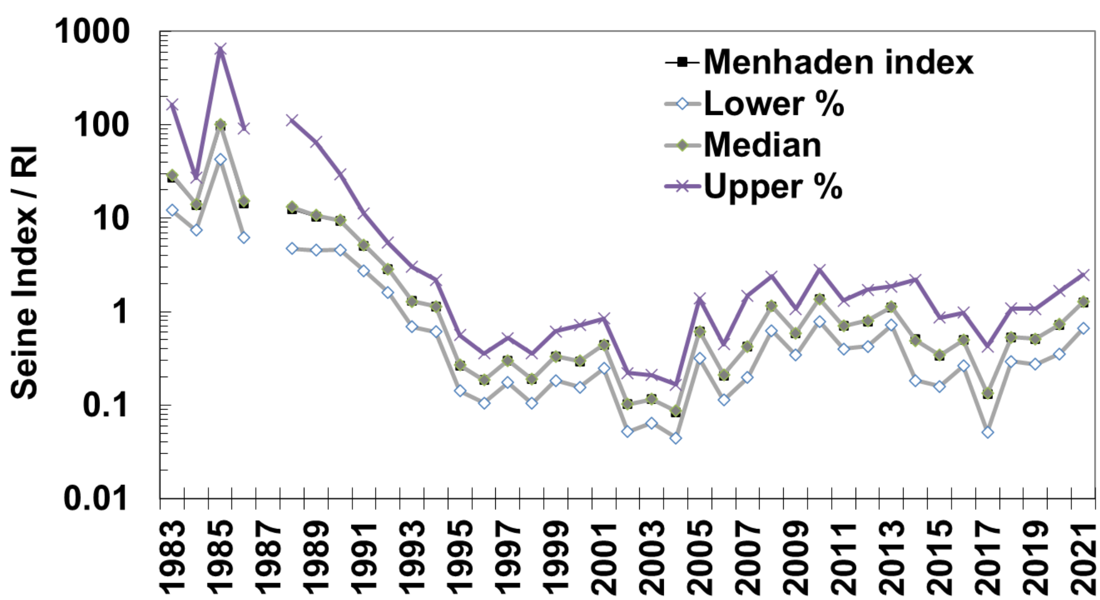

Figure 18. Bay Anchovy seine index to Striped Bass index (RI) ratios (Bay Anchovy seine FR) since 1983 and their 90% confidence intervals based on @Risk simulations of Bay Anchovy seine indices and RI distributions. Note $\log_{10}$ scale on the Y-axis.

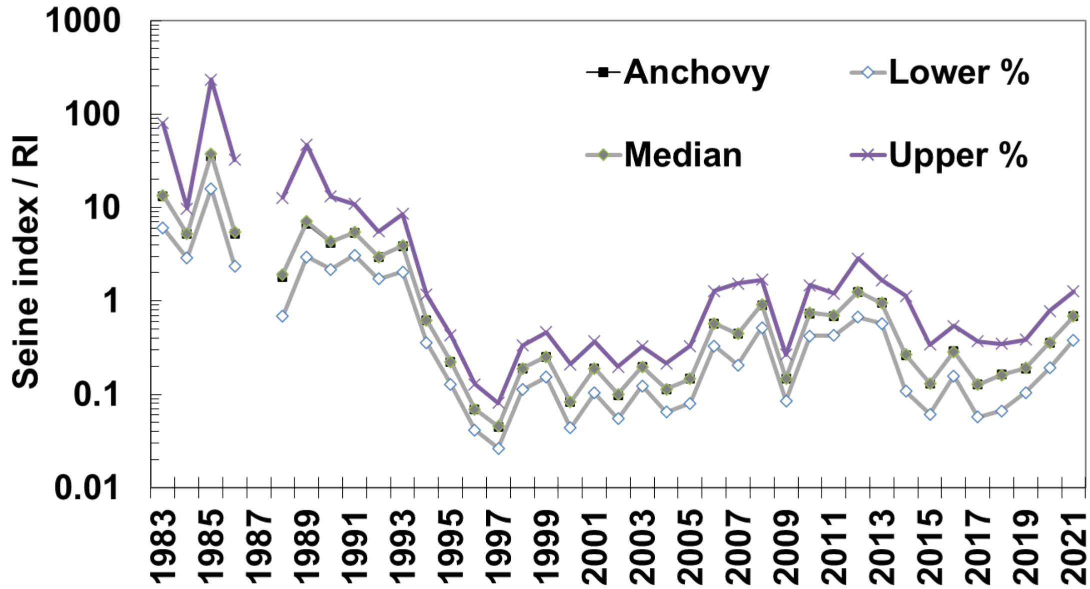

Figure 19. Bay Anchovy trawl index to Striped Bass index (RI) ratios (Bay Anchovy trawl FR) since 1989 and their 90% confidence intervals based on @Risk simulations of central tendency and estimated dispersion of data of trawl indices and RI. Note $\log_{10}$ scale on the Y-axis.

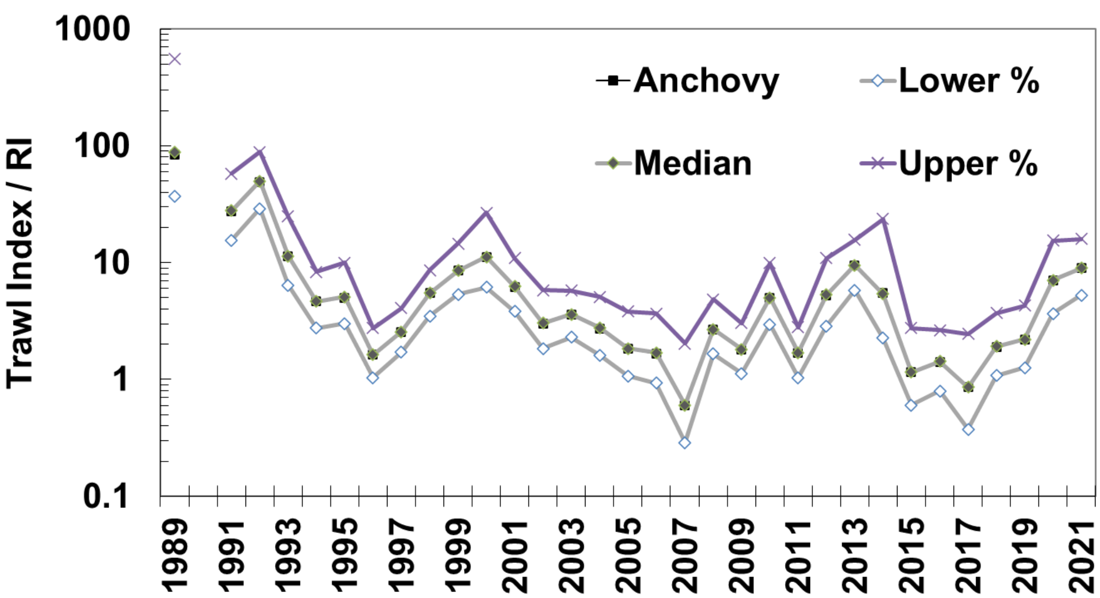

Figure 20. Spot seine index to Striped Bass index (RI) ratios (Spot seine FR) since 1983 and their 90% confidence intervals based on @Risk simulations of central tendency and estimated dispersion of data of Spot seine indices and RI. Note $\log_{10}$ scale on Y-axis

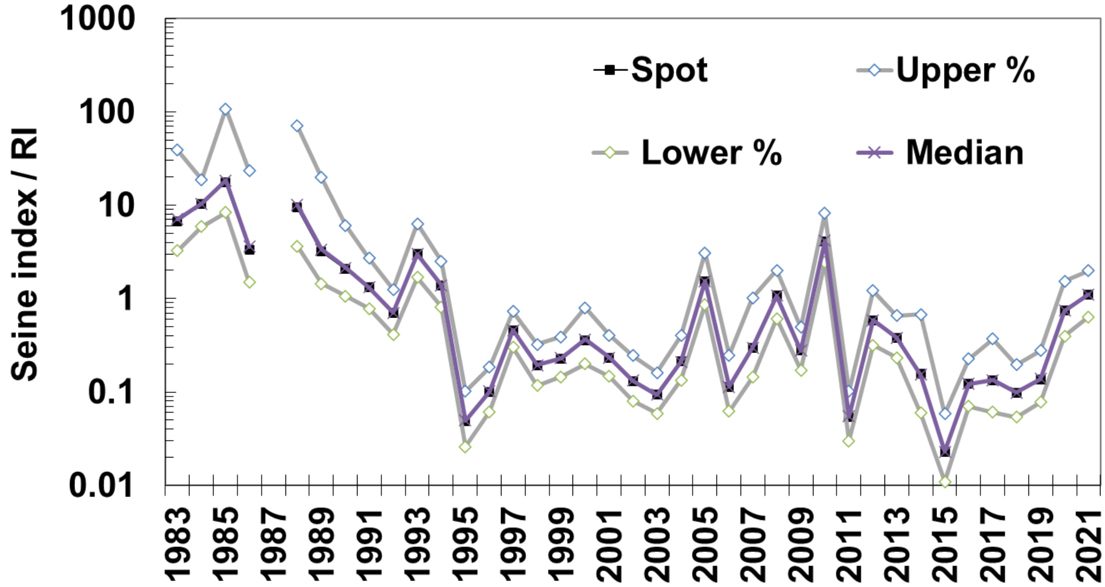

Figure 21. Spot trawl index to Striped Bass index (RI) ratios (Spot trawl FR) since 1989 and their 90% confidence intervals based on @Risk simulations of central tendency and estimated dispersion of data of trawl indices and RI. Note $\log_{10}$ scale on Y-axis.

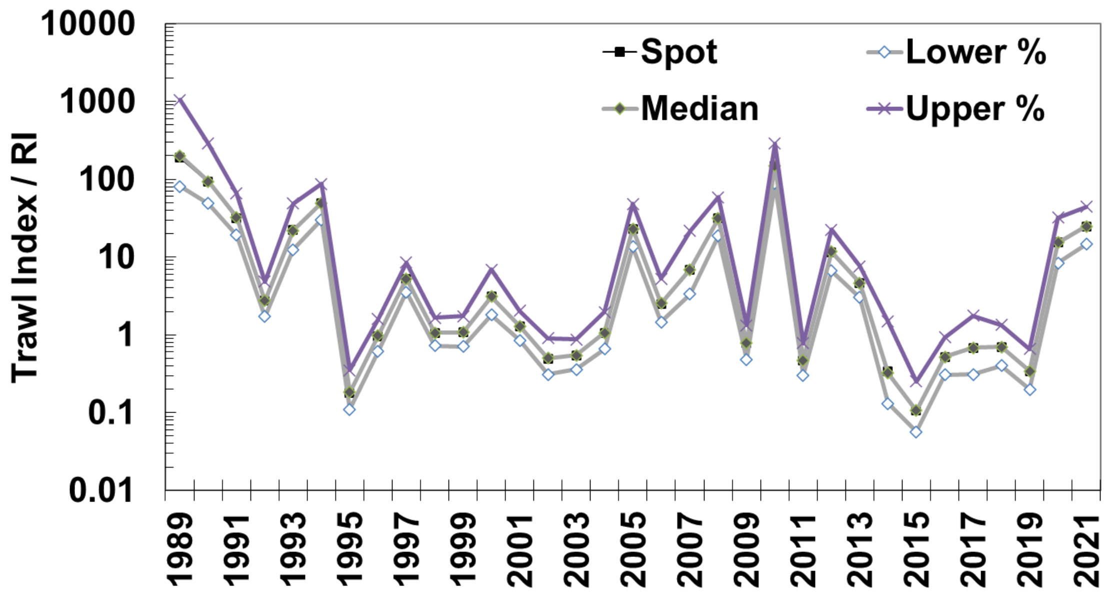

Figure 22. Blue Crab index to Striped Bass index (RI) ratios (Blue Crab FR) since 1989 and their 90% confidence intervals based on @Risk simulations of central tendency and estimated dispersion of data of Blue Crab (age 0) winter dredge densities and RI. Note the $\log_{10}$ scale on Y-axis.