American Shad Restoration in Three Maryland Rivers

F-57-R Segment 20 Progress Report
Reporting period January 1, 2018 to June 30, 2019

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*****Reporting Timeline*****

This progress report will cover calendar year 2018 sub-projects one, two, three and overall restoration progress. Elements of the project initiated in 2019 will be briefly reported. A comprehensive progress report for 2019 will be submitted in 2020.

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Need

American Shad *Alosa sapidissima* was once the most important commercial and recreational fish species in the Chesapeake Bay. In response to severe population declines from 1900 to the 1970s, Maryland closed its fishery in 1980. Various factors that contributed to the decline include over-fishing, stream blockages and poor water quality (Hildebrand and Schroeder 1928). Severely depressed or extirpated native adult stocks do not presently utilize most Chesapeake Bay tributaries, including the Choptank River (Klauda et al. 1991) and Patapsco River (USFWS 2013). The Choptank River historically supported commercially fished spawning stocks (Mansueti and Kolb 1953). Improvements in water quality, sustained fishing moratorium, and removal of many stream blockages has reopened potential shad spawning habitat in the Chesapeake Bay. Since shad populations indicate evidence of density dependent spawning behavior, self-sustaining shad populations are not likely to return to tributaries without hatchery stocking. Development of spawning, culture, marking and stocking techniques could restore spawning populations of American Shad to these target tributaries.

Objective

The overall objective for this proposed scope of work is to restore self-sustaining American Shad populations to the Choptank River and Patapsco River. Prior to project inception, the depressed native stocks in the Choptank River did not exhibit any evidence of spawning activity, according to exploratory sampling efforts conducted by the Maryland Department of Natural Resources in the early 1990s. This tributary supported spawning runs and active commercial and recreational fisheries in the past (Mansueti and Kolb 1953). Sampling conducted by the United States Fish and Wildlife Service (USFWS) on the Patapsco River in 2013 indicated that only a small remnant population of American Shad remained in the river (USFWS 2013).

Expected Results and Benefits

Hatchery stocked larvae and early juveniles are intended to provide adult spawners that will produce self-sustaining populations in the target tributary. These fish have tremendous value for stock assessment purposes at the larval, juvenile and adult life stages since all stocked shad receive a unique otolith mark to identify either the larval or juvenile life stage stocking. Natural

spawn and strip spawn culture techniques allow for the production of large numbers of larval and juvenile shad for stocking and assessment purposes.

Upper Bay and Potomac River shad populations currently support active catch and release recreational fishing. Restoring shad stocks to tributaries that historically supported runs will increase fishing opportunities for anglers. Recreational fishing that targets American Shad now occurs in the Patuxent River (previously stocked) and Choptank River (currently stocked). Angling groups have expressed interest in future angling opportunities in the Patapsco River. An indirect benefit of restoring shad populations to self-sustainable levels is the increased prey availability provided by both adult and juvenile shad for larger, more economically important recreational species such as Striped Bass *Morone saxatilis*, Bluefish *Pomatomus saltatrix,* and Weakfish *Cynoscion regalis.*

Approach

Maryland Department of Natural Resources began a pilot project in 1993 to assess the response of American Shad adult broodstock during collection, handling and captive holding. In 1994, experimental spawning was conducted using timed-release hormone implants. The success of these trials encouraged development of a long-term spawning, culture, stocking and assessment program. In 1995, a non-funded, full-scale hatchery production effort was conducted with positive results. The project continued over the next three years through various short-term funding sources. In 1998, it was determined that a long term funding source would be required, since it would take years of additional stocking and assessment to successfully support restoration. Federal Aid in Sport Fish Restoration funds has been utilized to conduct this long-term effort.

The project consists of three sub-projects:

1. *Produce, mark and stock cultured American Shad in the Choptank and Patapsco rivers.*
2. *A. Assess the contribution of hatchery-produced fish on the resident/pre-migratory stock in the Choptank and Patapsco rivers.*

   B. Monitor the abundance and mortality of larval and juvenile shad using marked hatchery-produced fish.
3. *Analyze the contribution of hatchery origin American Shad to the adult spawning*

population and monitor the recovery of naturally produced stocks.

Location

Restoration efforts will occur in the Choptank River (Figure 1). The Choptank River watershed is rural-impacted by agricultural activities and low urban development. Choptank River efforts include the tributary Tuckahoe Creek.

Restoration efforts will also occur in the Patapsco River (Figure 2). The Patapsco River watershed is heavily urban-impacted, but has been the subject of numerous mitigation efforts due to its designation as a targeted watershed (e.g. sewage treatment upgrades, dam removal).

2018 Sub-Project 1

Produce, mark and stock cultured American Shad in the Choptank and Patapsco rivers.

Objectives

In 2018, the Maryland Department of Natural Resources produced, marked, and stocked American Shad larvae and juveniles. American Shad production needs were met by strip spawning broodstock from the Potomac River. American Shad larvae and early juveniles were cultured, marked, and stocked into the Choptank and Patapsco rivers (Figure 1; Figure 2, respectively). Larval fish were stocked into the target tributary immediately after being marked with a day-3 oxytetracycline (OTC) mark. Early juvenile fish were marked with OTC on day-3,6, then were stocked as larvae into hatchery ponds, cultured until approximately 30 days of age, then transported to the Choptank River for stocking.

Materials and Methods

Broodstock Collection

American Shad were originally produced utilizing tank spawn culture methods developed by the project. Declining production success of American Shad from tank spawn operations dictated that an additional source of larvae be developed.

In 2001, the decision was made to collect ripe fish on the spawning grounds and manually strip eggs and milt from mature broodstock. The Potomac River was chosen as the

source population due to its healthy American Shad spawning population. The project hired a commercial fisherman to assist in egg collections that year. In 2002, it was determined that project personnel could perform these collections more efficiently and economically than the commercial fisherman. This is the method that is now utilized. The channel in front of Fort Belvoir concentrates a significant amount of American Shad (Figure 3). Collection of broodstock is carried out aboard a 7.0 m flat-bottom, center console skiff equipped with an outboard motor.

Weather and temperature conditions in late March and early April greatly influence the timing of American Shad spawning on the Potomac River. It is essential to begin sampling in early April to ensure that collections occur during peak shad spawning period. Sampling begins when water temperatures are 14°C to 16°C. In early April, the majority of captured American Shad females are gravid, but not yet ripe for egg collection, and are considered green. In early May, most captured females are ripe and suitable for egg collection. After a female has released her eggs, it is considered spent. Gradually, over a period of a month or two, the composition shifts from predominantly green females to mostly spent females. Once the shift in ratios occurs, contribution to hatchery production is low and broodstock collections cease.

Gill nets were set parallel to the channel edge at depths varying between approximately 7.0 and 15.0 m. Nets were set during a time window between 1730 and 2030 hours, close to sunset. Nets were checked for catch after 10-20 minutes soak time. Nets were fished in this manner for approximately 2-3 hours.

Catch per unit effort is used as an index of relative abundance. Gill net catch per unit effort (CPUE) is established by dividing the number of fish caught per net, by the square footage of net fished per soak time. A hand tally counter (tallycounterstore.com) is used to keep accurate count of all American Shad and bycatch caught from each net. Although trends in overall American Shad catch rates can be monitored using CPUE, the use of non-standardized gear and the change in gill net fishing techniques through the years makes it difficult to establish an accurate relative abundance over time.

Egg Fertilization and Culture

Egg fertilization was conducted aboard the skiff on the Potomac River. Ripe females and males were removed from gill nets and placed into separate holding tanks on the boat. Eggs were manually stripped into clean, dry, stainless steel bowls and milt was deposited over the eggs

using the dry method described by Howey (1985). River water was then added to activate the sperm. The eggs and milt were mixed together with a turkey feather, and then set aside for 10 minutes to water harden. Fertilized eggs were rinsed clean of any blood and ovarian tissue, and carefully poured into a floating egg box for at least one hour to further water-harden. This minimizes egg damage during transport to the hatchery. Pure oxygen was delivered to the egg boxes during transport. The floating egg boxes were secured to the deck of the boat and transported to Joseph Manning Hatchery (Brandywine, Maryland) for culture (Figure 4).

Eggs were placed into modified McDonald hatching jars supplied by approximately 2.0 L/min water flow. Prophylactic treatments of formalin were administered in the morning and afternoon to control fungi. Eggs were exposed to a 600:1 treatment of formalin for approximately 17 min. Eggs were volumetrically measured at the hatchery and percent fertilization was determined 24 hours post-fertilization.

American Shad eggs hatch on day six at Joseph Manning Hatchery water temperatures. In order to stimulate a simultaneous hatch, jars were removed from the egg bank, placed outdoors in sunlight for ten minutes and stirred occasionally. The increased temperature, lower oxygen content, concentrated hormonal influence and agitation stimulates simultaneous hatching. Jars were then placed around 1.5 m circular, flow-through larval culture tanks. Water was again supplied at approximately 2.0 L/min. Larvae flowed out of the hatching jar into circular culture tanks after they hatched.

Food was introduced to American Shad at day three. American Shad larvae were fed live *Artemia sp*. (www.brineshrimpdirect.com) and 100µm AP100 larval fish food (Zeigler Bros, Gardners, PA) three times daily during daylight hours.

Prior to stocking, larvae were enumerated using a volumetric direct proportion procedure in which a columnar sample of water is collected with a 25.0 mm diameter PVC tube at random locations in the larval tank. Larvae were enumerated in this sample and the total number of larvae in the tank was estimated by extrapolation to the total tank volume. In addition to this enumeration method, eggs were volumetrically measured and counted while performing the fertilization procedure prior to hatching.

Marking

All fish stocked into target tributaries were given an OTC mark through larval immersion. Oxytetracycline marking is a valuable assessment tool to determine hatchery origin, larval survival and juvenile abundance and mortality estimates. Larval marks were produced by immersion in a 300 ppm buffered OTC bath for six hours. Dissolved oxygen (DO) content was monitored and regulated (>5.0 ppm) by a high pressure/low volume air stone connected to a liquid oxygen delivery system. All water used at Manning Hatchery for OTC marking was softened before use (Culligan ion exchange system). Reliable marking can only take place in water with hardness below 20 mg/L and well water hardness at Manning Hatchery routinely exceeds 200 mg/L. Samples analyzed from each group of OTC-marked fish indicated that all fish stocked were successfully marked. Marks were verified by viewing larval otoliths under ultraviolet microscopy (Zeiss Axioskop 20).

In 2009, a three-year rotating, year-specific mark for larval stocked American Shad was implemented (Table 1). This procedure will validate current shad ageing protocols for adult, hatchery-origin American Shad collected. This research protocol was recommended by the Atlantic States Marine Fisheries Commission (ASMFC) American Shad and River Herring Technical Committee. Larvae designated for early juvenile stocking are given a day-3,6 mark.

Larval Stocking

In 2018, fish intended for larval stocking received a larval immersion mark at day-3 after hatch. Stocking was accomplished by placing OTC-marked larvae into boxes designed for shipping tropical fish. These containers consisted of an outer shell cardboard box, an inner insulating foam box, a black plastic trash bag to reduce stress of bright sunlight and a double thickness plastic fish bag. Larval culture tanks were drawn down to crowd the fish. Larvae were scooped out of the tanks using a modified milk jug and placed into the shipping bags/boxes, which were supplemented with approximately 1.0 ppt salt to mitigate stress. Each bag was filled with pure oxygen and sealed with electrician's tape. Boxes were transported to either the Choptank River at Greensboro, Stoney Point on the Tuckahoe Creek, or to the Patapsco River at the Rt. 648 Bridge or South West Area Park boat ramp (Figure 5; Figure 6, respectively).

The bags were placed in the water long enough for temperature to acclimate. The bags were then opened and river water was slowly introduced to further acclimate larvae to river water conditions. Bags were then emptied into flowing water to minimize predation.

Early Juvenile Stocking

Fish intended for early juvenile stocking received immersion marks at days three and six after hatch. After the second mark was administered, larvae were stocked into culture ponds for approximately thirty days. Manning Hatchery, GenOn Energy, and University of Maryland Center for Environmental Science (UMCES) Aquaculture and Restoration Ecology Laboratory (AREL) provided grow out ponds to hold fish for the restoration effort (Figure 4). The decision to take juveniles out of the ponds was based on zooplankton density. Food availability in grow out ponds was evaluated with a plankton net. Early juveniles were removed from culture ponds when food availability declined significantly and fish began prowling the edges of the pond.

Juvenile fish tend to stress easily and direct netting from hatchery ponds into transport tanks results in unnecessary mortality. To prevent loss, juvenile fish were concentrated within the grow out ponds, using a seine net 61.0 m long, 3.1 m deep, with 6.4 mm stretch mesh. They were then effectively removed by scooping the concentrated schools of fish out with buckets and were transferred into the transport tanks. A one-horsepower water pump was used to create current within the seine net to orient shad into the water flow. This current serves two purposes. Shad are concentrated in the flow for ease of bucketing, and it separates the fish from the algae and detritus. Early juvenile survival increased in recent years due to the reduction of algae and detritus in the transport tanks. Early juveniles were transported to Choptank and Patapsco river stocking locations in culture pond water that has salinity of 6.0-8.0 ppt.and DO saturated to mitigate stress. Ponds at GenOn Energy and UMCES-AREL already have elevated salinity of 6.0-8.0 ppt.

A one-horsepower trash pump was carried on the stocking truck to temper juvenile shad before stocking. Fish were tempered until temperature and salinity in the tank were within one degree Celsius and 1.0 ppt salinity of the river value. Although this adds a considerable amount of time that fish are aboard the transport tank, it is assumed this procedure increases the survival of early juvenile stocked shad by reducing stress. Juvenile stocking was accomplished by quick-dumping marked juveniles through a quick release drain hose with a diameter of 15.0 cm, directly from the transport vehicle into the river.

Stocking Goals

The project developed stocking goals (Table 2; Table 3) based on past experience with juvenile collections. Stocking multiple life stages gives fisheries managers the ability to assess larval survival and estimate juvenile mortality and abundance of each life stage.

Larval stocked fish can efficiently contribute large numbers of juveniles if larval survival is high. Fish stocked as early juveniles survive extremely well and are young enough to successfully imprint to the stocked tributary. Stocking early juveniles can also mitigate the impacts of poor larval survival since post-stocking survival of this life stage is high. Early juvenile production is limited by grow out pond availability and space.

Results and Discussion

American Shad Strip Spawn Production Summary

Since the inception of gill netting in 2001, a normal distribution of egg production is observed most years. American Shad are collected from the Potomac River spawning area when temperatures range from 15°C to 22°C. In 2018, a steady increase in water temperature was observed from the middle of April to the middle of May, and then a slight temperature decrease was observed until the collection season ended on 21 May (Figure 7). The reduction in egg collection numbers from the start of the second week of May to the end of the third week, can be attributed in part to the water temperature decline (Figure 7). However, after collecting 90 liters on 8 May, inclement weather was the primary factor for the reduction of egg collection. Thunderstorms occurred every evening and there were sustained periods of rain throughout each day. The thunderstorms prevented collection or ended collection nights early. The additional precipitation in the watershed washed many logs and debris into the water, increased turbidity, and created hazardous boating and fishing conditions, which hindered or completely prevented egg collection. Coinciding with the deteriorating conditions, the ratio of spent females greatly increased during the third week of May, at which point collection ended (Figure 8). In 2018, staff observed the second largest egg collection year (483 L) since the project's inception in 2001 (Figure 9). Increased broodstock collection was attributed to favorable weather conditions during the peak of the American Shad spawning season. Fifteen or more liters of eggs were collected on

twelve consecutive nights. During that stretch, a record high of 90 liters of eggs were collected in one night, which contributed to the above average production year (Table 4; Figure 7).

In 2016, gill net fishing techniques were altered half way through the collection season to try to reduce bycatch and maximize American Shad broodstock collection. Project biologists began experimenting with net fishing duration time. American Shad spawn near, or just after sundown (Mansueti and Kolb 1953). Starting 4 May 2016, nets were fished just before sunset, without taking slack tide into consideration. Only one to two nets were fished, to prevent fishing too much gear at one time. The nets were fished more frequently (10-15 minutes) and re-set. Nets were fished this way for approximately two hours per night. Bycatch was greatly reduced with the change in fishing technique (Figure 10). 2018 continued to use the modified fishing technique. Bycatch collection numbers were low (Figure 10). Staff fished two different stretch mesh sizes (117.5mm and 127.0mm). Gill nets with smaller mesh size have the tendency to catch smaller fish while nets with larger mesh sizes have a tendency to catch larger fish.

The Maryland Department of Natural Resources collected 2,559 adult American Shad by gill net on the Potomac River in 2018. One thousand one hundred and sixty four ripe females produced 483 L of eggs. Overall fertilization was 50.7%. The estimated number of fertilized eggs produced was 7,812,943 (Table 4).

Stocking Summary

American Shad were stocked as larvae and early juveniles at various locations in the Choptank River and Patapsco River (Tables 5-8, Figures 5-6). A summary of 2018 American Shad stocking production separated by event appears in Table 5 and Table 6. Historical American Shad stocking production summaries for all years are contained in Tables 7 through 11.

Despite collecting a large number of eggs, an electrical malfunction with the well at Joseph Manning Hatchery during peak production resulted in the mortality of 2,700,000 fertilized eggs, which reduced the overall numbers of stocked larvae. As a result, American Shad larvae stocking goals for the Choptank River were not met for 2018. However 1,550,000 larvae were stocked (Table 2; Table 10). The project was able to exceed early juvenile stocking goals, which are valuable to assess hatchery contribution (Table 2). Patapsco River larvae and early juvenile stocking goals were both met (Table 3).

2019 Sub-Project 1 – Preliminary Results – Work in Progress

Analysis of the data for 2019 is currently in progress. Adult American Shad were caught by gillnet on the Potomac River from 17 April to 19 May 2019 for broodstock collection. A total of 1,693 total American Shad were caught, which produced 542 liters of eggs. More than 2.34 million larvae and 465,000 early juveniles were stocked into the Choptank River. Approximately 200,000 larvae and 135,000 early juveniles were stocked into Patapsco River.

The complete analyses and summary of the data collected in 2019 to produce, mark and stock cultured American Shad in the Choptank and Patapsco rivers will appear in the 2020 F-57-R progress report.

2018 Sub-Project 2

Objectives

Restorative stocking of American Shad in the Choptank and Patapsco rivers began in 1994 and 2012, respectively. Choptank River summer juvenile seine survey commenced in 1996 and the Patapsco River summer seine surveys began in 2013. Both surveys had a goal to collect juvenile American Shad to determine the success of the stocking program in each river. From 2013 to 2017, and MPA funded grant supported restoration activities in the Patapsco River and all fisheries monitoring was conducted by USFWS Maryland Fish and Wildlife Conservation Office (MDFWCO). In 2018, the Patapsco River was added to the Maryland Department of Natural Resources' Wildlife and Sport Fish Restoration grant, along with the Choptank River, to continue the project for stocking and assessment.

Two quantifiable population variables were identified to evaluate restoration progression of juvenile American Shad in the targeted rivers.

A. "Assess the contribution of hatchery-produced fish to the resident/pre-migratory stock in the Choptank River and Patapsco River."

B. "Monitor the abundance and mortality of larval and juvenile shad using marked hatchery-produced fish".

Materials and Methods

Juvenile American Shad were collected by seine from the Choptank River from late summer to early fall. A seine 61.0 meters long, 3.1 meters deep, with 6.4mm stretch mesh, was deployed by boat from shore into deep water and back to shore at established seine sites. The net was retrieved by hand. Juvenile American Shad were collected from the seine, placed into plastic bags, labeled, and stored on ice. Upon return to the lab, the samples were frozen to -9 °C. All bycatch species data were recorded.

Due to the size of the river and the number of underwater trees encountered, the seine net used on the Patapsco River is smaller than the net used on the Choptank River. The net used to sample the Patapsco River was a seine 30.5 meters long, 1.2 meters deep, with 6.4mm stretch mesh. The net was deployed by hand from shore into deep water and back to shore at established seine sites. Juvenile American Shad were collected from the seine, placed into plastic bags, labeled, and stored on ice. Upon return to the lab, the samples were frozen to -9 °C. All bycatch species data was recorded.

Sagittal otoliths were removed from each American Shad captured from the Choptank River. Otoliths were mounted on 76.2 mm x 25.4 mm glass slides with Crystalbond 509 (Aremco Products, Ossining, NY).

Mounted otoliths were lightly ground on 600 grit silicon carbide wet sandpaper and viewed under an LED epifluorescent light at 400X magnification at 50-100 watts with a Zeiss Axioskop 20 microscope. The presence and location of OTC mark epifluorescence was recorded. Epifluorescence is a technique in which transmitted light in the wavelength of 490-515 nm is allowed to strike the specimen. The specimen then absorbs this light energy and reflects light of a longer wavelength back through the microscope objective.

CPUE and Geometric Mean

The juvenile index is described by calculation of a CPUE. It is defined as the number of captured juvenile American Shad divided by the number of seine hauls completed. Indices of relative abundance are presented as the total arithmetic mean (AM) catch per haul and geometric mean (GM) catch per haul. The GM has been adopted by the ASMFC as the preferred index of relative abundance. The GM is a more precise statistical tool for handling these data because it is not as sensitive to a single large sample value. American Shad are schooling fish and subject to

these types of captures with a large seine net. Since the inception of this project, CPUE has been used to quantify indices of relative abundance. As a bridge between previous F-57-R progress reports, the 2008 report indicated AM and GM. All indices of relative abundance from 2009 on are now reported as GM only.

Mortality and Abundance Estimates

In addition to providing future broodstock, juvenile stocking is valuable as a pre-migratory stock assessment tool through the use of a multiple marking technique. Hatchery stocking is also used to evaluate the efficacy of stocking different life stages and the eventual impact to the returning adult population.

There are assumptions made when using these types of estimates as described by (Ricker 1975):

- The marked fish suffer the same natural mortality as the unmarked fish.
- The marked fish are as vulnerable to capture as are the unmarked fish.
- The marked fish do not lose their mark.
- The marked fish become randomly mixed with the unmarked; or the distribution of fishing effort (in subsequent sampling) is proportional to the number of fish present in different parts of the body of water.
- All marks are recognized and reported on recovery.
- There is only a negligible amount of recruitment to the catchable population during the time of recoveries are being made.

Estimates of survival, instantaneous mortality and abundance were calculated for American Shad in the Choptank River and Patapsco River.

Estimates of juvenile abundance, mortality and survival were derived from the following formulas.

Larval survival to juvenile stocking was calculated by (Ricker 1975):

$$S_1 = \frac{(R_{12})\ M_2}{(M_1)\ R_{22}}$$

$$\text{Variance } S_1 = {S_1}^2\{\left(\frac{1}{R_{12}}\right) + \left(\frac{1}{R_{22}}\right) - \left(\frac{1}{M_1}\right) - \left(\frac{1}{M_2}\right)\}$$

where M_1 is the number of fish marked at the start of the first interval (larval stocking), M_2 is the number of fish marked at the start of the second interval (early juvenile stocking), R_{12} is recaptures of larval marked fish in the second interval (after early juvenile stocking), R_{22} is recaptures of early juvenile interval marked fish in the second interval or (after early juvenile stocking), and S_1 is the survival rate of larvae during interval one (from the time of marking larvae in interval one to time of marking early juveniles in interval two).

Instantaneous mortality is derived from survival estimates and is used in conjunction with stocking data to calculate juvenile abundance:

$$Z = \frac{-\,ln\,ln\,S_1}{interval}$$

Where Z is instantaneous mortality rate and S_1 is survival rate.

Abundance of juvenile shad prior to out migration was also calculated by Chapman's modification to the Peterson estimate (Ricker 1975):

$$N = \frac{\{(C+1)(M+1)\}}{R+1}$$

where N is the population estimate, M is the number of marked fish stocked, C is the number of fish examined for tags (total captures) and R is the number of marked fish that were recaptured (larval or early juveniles).

From Ricker (1975): Calculation of 95% confidence limits based on sampling error using the number of recaptures in conjunction with Poisson distribution approximation.

Chapman's modification (1951):

$$N^* = \frac{\{(C+1)(M+1)\}}{(R+1)}$$

Where R_1 is from Pearson's formula to calculate upper and lower limits:

$$R_1 = R + 1.92 \pm 1.960\sqrt{R + 1.0}$$

Results and Discussion

Choptank River

The Choptank River (Figure 11) was sampled weekly nine times from 1 August through 25 September. The study collected 496 American Shad juveniles from the Choptank River in 2018. A total of 483 samples were successfully analyzed. The remaining 13 samples were lost or damaged during processing. Among 2018 samples, 91% were of hatchery origin and 9% were wild origin. Of hatchery origin captures, 49% were day-3 marked larvae and 51% were day-3,6 marked early juveniles (Table 12).

Juvenile American Shad were collected at all seven of the established seine sites on the Choptank River, and three additional seine sites on Tuckahoe Creek in 2018 (Table 13). The sample sites in the Choptank River and Tuckahoe Creek did not include the upper range of the juveniles in these tributaries. The area of greatest juvenile abundance was most likely sampled, but the lack of acceptable seining sites precludes collections upstream from Depue Landing on the Choptank River and Stoney Point on Tuckahoe Creek (Figure 11). Downstream juvenile habitat is historically limited by salinity. The salinity parameters during the 2018 summer seine survey were considerably lower than the long-term average of 4.1 ppt at the lowest seine site Fossil Cliff (Figure 12).

Choptank River Geometric Mean

During the 2018 juvenile seine survey, 490 American Shad were captured from the Choptank River and Tuckahoe Creek. The 2018 GM was calculated to 3.07, which is well above

the average of 1.48 from the previous eighteen years (Figure 13). The GM has increased significantly during the sample period from 2000 to 2018 with the low GM calculation of 0.06 in 2002 to a high of 4.13 in 2010 ($p < 0.001$; Figure 13).

From 2000 to 2009, GM was calculated exclusively from collection sites on the Choptank River. Data from Tuckahoe Creek were not used to calculate GM from 2000 to 2009 due to inconsistencies in collection seine sites. Beginning in 2010 after the utilization of three consistent seine sites on Tuckahoe Creek, the GMs for the Choptank River and Tuckahoe Creek were combined. A two-sample, two-tailed t-test was performed on the Choptank River GM and the Choptank River/Tuckahoe Creek GM from 2010-2016. This was calculated to determine if there was a significant difference between the GM from the Choptank River alone and the addition of the Tuckahoe Creek tributary. No significant difference was found ($t = 2.179$, $DF = 12$, $p = 0.851$). Since there is no difference between the two GMs, starting in 2017, a single line will depict long-term GMs for the Choptank River from 2000-2009 and the Choptank River and Tuckahoe Creek from 2010 through the current year.

Prior to 1996 stocking efforts, no American Shad were captured in this tributary in 35 years (sampling conducted by other department projects prior to 1996).

Higher transport mortality has been observed when stocking early juveniles in the Choptank River when water temperatures exceed 24^{o}C. It is possible that post-stocking mortality of early juveniles is higher at this distant location. Travel time for stocking early juveniles from culture ponds at GenOn Energy to the Daniel Crouse Memorial Park boat ramp was approximately two hours. Culturing the majority of early juveniles at the UMCES-AREL facility in Cambridge, MD has considerably reduced transport time compared to other cooperative culture facilities (Figure 4; Figure 5). The differences observed between the Patuxent River and Choptank River early juvenile mortality suggest the need to avoid stocking distant rivers with juvenile American Shad during the heat of the day. An effort has been made to transport and stock juveniles during early morning hours. Early juvenile marked fish are recaptured in the summer seine survey, therefore these handling procedures appear to be working. Handling juvenile shad will be conducted in this manner in future years.

Choptank River Mortality and Abundance Estimates

Estimates of larval survival, instantaneous mortality and juvenile abundance were calculated for Choptank River American Shad in 2018 (Table 14). Survival of day 3 marked larval stocked American Shad to early juvenile stocking in the Choptank River was calculated at 0.3256 for the 35-day period, which exceeds the average of the seventeen-year data set (0.205). Daily mortality of larval stocked shad to the time of early juvenile stocking was calculated to be Z = 0.0321, (±2 SE = 0.0618). Juvenile abundance of day 3 larval stocked American Shad was calculated to be 504,600 using survival estimates and stocking data. Traditionally, using early juvenile recaptures to estimate total juvenile abundance is a more accurate measure than larval or late juvenile recaptured fish. Survey recapture rates were used to estimate the composition of the juvenile stock (Table 15). Total juvenile abundance in the Choptank River (20 June 2018) was calculated by Chapman's modification to the Peterson estimate at 1,142,111 (upper limit = 1,299,168; lower limit = 1,003,977). Abundance of wild origin juveniles was estimated at 104,888 in 2018. The 2018 wild abundance estimate was the second highest estimate in the seventeen-year data set (Table 16; Figure 14).

Total juvenile abundance was calculated annually for the Choptank River since 1996 (Table 16). Minkkinen et al. (1997) estimated Choptank River total juvenile American Shad abundance in 1996 at 109,300. No wild fish were collected during that assessment. The population was comprised of 28,600 larval stocked fish and 80,700 fish that had been stocked as juveniles.

In comparison, estimated total abundance at the time of early juvenile stocking was 404,000 in 2002 and 349,800 in 2003. The 2005 and 2006 estimates indicated the highest levels of total juvenile abundance (more than 1 million) observed until 2015 in the Choptank River (Table 16). The 2018 abundance estimate of 1,142,100 is nearly double the seventeen-year average (694,000), and is the second highest estimate on record.

Survival of larval anadromous species can vary widely from year to year. In previous spawning seasons, larval survival ranged from a low of 0.042 in 2011 to a high of 0.621 in 2015 (Table 14). These larvae are sensitive to both biotic and abiotic factors during the first weeks of development (Crecco 1985). Larval-origin juvenile abundance is not correlated with larval stocking effort. This is due to variable larval survival from year to year (Table 14). Total juvenile abundance is variable according to the level of stocking effort and larval survival for each year,

and is positively correlated with larval stocked-origin juvenile abundance. ($r^2 = 0.6175$, P = 0.00091).

Based on prior observations, recruitment to the juvenile population is set by approximately 30 days and mortality is very low past this point. Direct comparisons of Choptank River abundance to other target tributaries, such as the Patuxent River, are not appropriate without consideration of the quality and quantity of juvenile habitat available. Based on historical juvenile recaptures from this project, the Choptank River has much more juvenile habitat than the Patuxent River and Marshyhope Creek, so at this time it is unknown whether an abundance estimate of 1,000,000 is high for this river. Based on the amount of available juvenile nursery habitat, the Choptank River should be able to support at least four times the abundance of the Patuxent River. Considering past abundance estimates of more than 400,000 juveniles in the Patuxent River, it is possible that the Choptank River could support between 1.5 million and 2.0 million juveniles.

Natural recruitment is occurring in the Choptank River according to juvenile otolith analysis. No wild juveniles were captured in the first five years of the restoration effort. Total captures were low (1997-2000), and no wild juveniles were captured until 2001 (Table 12). Poor hatchery production in previous years prevented stocking sufficient numbers of larvae in the Choptank River. At the inception of the project, we estimated that a minimum of 2,750,000 larvae should be stocked into the Choptank River annually to ensure recaptures, juvenile recruitment and subsequent sufficient adult recruitment. The restoration effort decided to suspend stocking Marshyhope Creek and apply all project resources towards stocking and monitoring in the Choptank River. This strategy should enable collection of sufficient information to accurately estimate the survival and abundance of juvenile American Shad in the Choptank River each year.

The Choptank River wild juvenile abundance estimate was calculated using the time interval between larval and early juvenile stocking events. This estimate calculated 104,900 wild juveniles for 2018, which accounted for 9% of the juvenile population. In 2007 and 2008, wild origin juveniles accounted for sixteen and nineteen percent of the captures respectively, two of the highest recorded sample years for wild captures. In 2009, the wild capture percentage substantially declined to seven percent from nineteen percent, which initiated a percentage point

decrease each year through 2011 (Table 12). From 2012 to 2017, the wild captures averaged 8% per year. Wild American Shad in 2018 comprised 9% of the total juvenile population.

Patapsco River

In 2018, the study collected no American Shad juveniles from the Patapsco River. The Patapsco River (Figure 2) was sampled weekly from 31 July through 17 September. The project was unable to collect juvenile shad at sites where they were collected in previous seasons. Flooding from a documented rain event occurred throughout the Patapsco Valley area on 27 May which deposited more than 8" of rain in a two-hour period. It is hypothesized that the flood pushed the majority of juvenile shad out of the system.

Patapsco River Geometric Mean

No American Shad were captured in 2018, therefore a Geometric Mean was not calculated.

Patapsco River Mortality and Abundance Estimates

No American Shad were captured in 2018, therefore abundance and mortality estimates were not calculated.

2019 Sub-Project 2 – Preliminary Results – Work In Progress

Choptank River

During the 2019 juvenile American shad survey, 769 American Shad were sampled from the Choptank River. The Choptank River was sampled weekly ten times from 2 July through 4 September. Juvenile American Shad were collected at all seven of the established seine sites on the Choptank River, and three additional seine sites on the Tuckahoe Creek. Analysis of origin will be presented in the next F-57-R American Shad progress report in 2020.

Patapsco River

During the 2019 juvenile American shad survey, 40 American Shad were sampled from the Patapsco River. The Patapsco River was sampled weekly fourteen times from 25 June

through 26 September. Juvenile American Shad were collected at five of the seven established seine sites on the Patapsco River. Analysis of origin will be presented in the next F-57-R American Shad progress report in 2020.

2018 Sub-Project 3

Analyze the contribution of hatchery origin American Shad to the adult spawning population and monitor the recovery of naturally produced stocks.

Objectives

Patuxent River and Choptank River spawning ground surveys commenced in 1999 to collect adult American Shad. Restorative stocking of American Shad in these two target tributaries began in 1994 and 1996, respectively. In 2018, the Patapsco River was added to the restoration project for stocking and assessment. The Patapsco River was the target of a shad and herring restoration project funded by the Maryland Port Authority (MPA) from 2013-2017 as part of a compensatory mitigation package designed around the Masonville Project. The Masonville Project is a Dredged Material Containment Facility (DMCF) located in Baltimore Harbor. The Masonville Cove Environmental Education Center and a 22-hectare Conservation Area were constructed adjacent to the DMCF in 2009. As a component of the DMCF project, the MPA was required to develop a mitigation project to offset impacts associated with filling approximately 53 hectares of open water in the Patapsco River; a major tributary to the Chesapeake Bay. The Patapsco River restoration project will hopefully add fishing opportunities for the local community by re-establishing anadromous species such as American Shad and Hickory Shad to the Patapsco River. The Patapsco River was the logical choice to continue stocking efforts to assess restoration efforts conducted to date. Maryland Department of Natural Resources restoration work thus far indicates that self-sustaining shad restoration will likely occur over a period of decades, rather than years. The 2018 removal of Bloede Dam will open the river at its most downstream blockage and reintroduce unimpeded access for shad and herring.

Three quantifiable population variables were identified to evaluate restoration progression of adult American Shad spawning stocks in the targeted rivers.

1) *Estimate catch-per-unit effort (CPUE) in each targeted river using geometric mean.*

2) *Estimate the contribution of hatchery produced fish to the adult spawning populations.*
3) *Estimate the age composition and frequency of virgin and repeat-spawning.*

Materials and Methods

Survey Locations

Sampling was conducted at historical American Shad spawning areas described by anecdotal data, and concentrated in river reaches where shad were encountered during previous sampling efforts. In 2018, sampling occurred in the Choptank and Patapsco rivers (Figure 1; Figure 2, respectively). The Patuxent River population was determined to be recovered in 2014 and is sampled on a three year rotation and will not be sampled again until 2020.

Two sample methods utilized this year were gill netting and electrofishing. In the Choptank River, the electrofishing sampling starts 1.5 km downstream from Red Bridges and extends 2.1 km downstream to the Route 313 Bridge in Greensboro, Maryland (Table 17; Figure 15). In the Patapsco River, the electrofishing sampling area is divided in two sections, from the wastewater treatment plant located just west of Rte. 648 (Baltimore Annapolis Road) to approximately 1.43 km downstream. The second section of electrofishing is conducted from the I-895 Bridge that crosses the Patapsco River downstream approximately 1.49 km to West Patapsco Avenue (Table 17, Figure 16).

Electrofishing was conducted with a Smith-Root electrofishing boat model SR18-E (Vancouver, WA) from 10 April to 30 May during daylight hours on the Choptank River, and from 12 April to 31 May on the Patapsco River. Each survey was accomplished with three people; one person piloting the boat and two people netting shad from the bow. Each river was sampled in an upstream to downstream direction with constant voltage applied to the entire reach. Total pedal time (s) was recorded to calculate CPUE.

Gill net surveys were conducted on the Choptank River from 10 April to 30 June. Two gill nets (12.7 cm stretch mesh), one 123 m and one 134 m in length, and 3.05 m deep, were deployed to cover the entire water column. Gill netting attempts were conducted one km upstream of the Denton, Maryland boat ramp (Figure 17). Gill nets remained stationary throughout the duration of the sample day. Gill nets were set and retrieved every hour to check

for American Shad and bycatch. Gill netting was utilized on the Choptank River due to the difficulty of capturing American Shad in the historical electrofishing sampling area. The Patapsco River was not sampled by gill net.

Water quality parameters were recorded at the end of each sampling event while still in the sampling reach. Water temperature (◦C), dissolved oxygen (mg/L), salinity (ppt) and conductivity (µS/cm) were obtained using a YSI Pro 2030 water quality meter (Yellow Springs, OH). Catch per unit effort for gill netting followed procedures described in sub-project one of this report.

In each of the targeted rivers, it is likely that shad utilize tidal freshwater areas downstream of the electrofishing collection sites, but increasing river width and depth reduces capture efficiency with electrofishing gear. Anecdotal evidence indicates that substantial spawning habitat and fish movement also exists upstream of currently sampled stream reaches, but sampling habitat is limited by electrofishing access. The use of gill nets eliminates the depth and width issues brought about by electrofishing by using specific mesh size and gill net length/height that targets American Shad and covers the entire water column.

A sub-sample of 20 American Shad was collected per sample trip for age, otolith, and spawning attempts analyses. All other observed shad were counted to calculate CPUE and released. Fish were measured for total length (TL, mm), fork length (FL, mm) and sex was determined. Scale samples were taken for age estimation and spawning mark interpretations. Otoliths were extracted to identify hatchery OTC marks. All hatchery origin American Shad are marked with OTC, which permits analysis of hatchery contribution to the juvenile abundance estimate and the adult spawning stock composition. Shad scales were cleaned, mounted between glass slides, and age was estimated and spawning attempts were counted using a microfiche reader. Two biologists interpreted the scales independently. In cases where readers disagreed on an age/spawning attempt analysis, a consensus age was used as the final age. Scales were analyzed using methods described by Cating (1953). Otoliths were processed using methods described for juvenile fish in sub-project two.

Catch Per Unit Effort Analysis

Relative abundance was omitted in reports prior to 2008 due to changes in sampling protocol and the overall nature of sampling these highly turbid rivers. Beginning in 2008,

attempts were made to standardize CPUE data and apply those results to evaluate restoration progression. Data were standardized using the number of shad encountered per day divided by the shock time in minutes applied to the river the day of sampling. Since the number of sampling days is different each year, the mean CPUE is calculated to obtain an annual CPUE. Adult sample data are unavailable prior to 1999 and any data prior to 2001 are deficient of the necessary catch and effort data to obtain a standard CPUE. Standardization of CPUE advanced in 2011 with the implementation of bracketing CPUE data. Before 2011, data were collected beginning the first week of April and lasting until the CPUE reached zero at the end of the spawning run. Protocol now calls for a CPUE zero at the beginning and end of the survey season to better understand how long fish remain in the spawning area each year.

The GM has been adopted by this project as the preferred index of relative abundance to evaluate stock status and restoration progress. The GM is calculated from the $\log_e(x+1)$ transformation, where x is the number of American Shad encountered per shock time (min). Beginning and ending zeros are omitted from the analysis. The number one is added to all catches in order to transform zero catches, because the log of zero does not exist (Ricker 1975). Since the $\log_e$-transformation stabilizes the variance of catches (Richards 1992), the GM estimate is more precise than the AM and is not as sensitive to a single large sample value. It is almost always lower than the AM (Ricker 1975). The one was subtracted from the final number after final calculations.

Beginning in 2015 with the use of gill nets, the traditional method of CPUE for electrofishing and the use of (GM) for relative abundance was not applicable. On the Choptank River, two gill nets were used and the nets were all the same mesh size. The nets used in 2017 differed in length by 11m. The CPUE for the Choptank River gill netting was calculated by taking the number of fish caught in each net divided by the hours fished for that specific net. That created a CPUE for each net fished that day. The data were then averaged for a CPUE each day fished. The length of each net was not taken into account when calculating CPUE.

Origin Composition (Hatchery vs. Wild)

The percentage of hatchery versus wild origin American Shad adults sampled on the spawning grounds provides insight into the impact to the adult population of stocking larval and juvenile shad. The presence of adult hatchery origin fish on the spawning grounds early in

restoration may stimulate annual natural reproduction, something that had not occurred in decades prior to the restoration efforts. As restoration efforts continue, a transition from a high proportion of hatchery origin fish to a high proportion of wild fish year after year indicates natural reproduction events leading to successful recruitment to adulthood. Identifying shifts from predominantly hatchery origin adults to a wild origin population indicates a substantial effect upon the adult spawning stock population. This variable is sensitive to small sample sizes.

Virgin and Repeat-Spawning Compositions

A third estimator uses analysis of virgin and repeat-spawning compositions. The number of times a fish embarks on an annual spawning run during its lifetime can be determined through examination of American Shad scales. The composition of virgin and repeat-spawn frequency observed on the spawning grounds provides additional insight to population stability and recruitment. Low levels of virgin-spawners may indicate problems associated with juvenile recruitment to the adult stock or poor spawning success. Conversely, a high level of virgin-spawners usually indicates successful recruitment of individual year classes to the adult spawning stock. A substantial contribution of virgin-spawners and several repeat-spawning classes utilizing the spawning grounds year after year is indicative of a stable spawning stock. In the past, annual spawning stock sample sizes were very low and precluded any virgin and repeat-spawning composition analysis. With the recent increase American Shad captures on the Choptank River a repeat-spawning and age analysis will be conducted.

Results and Discussion

Choptank River Adult American Shad Spawning Stock

Eight American Shad adults were captured by gill net on the Choptank River from 10 April to 30 May 2018 (Figure 18). The water temperature ranged from 9.9°C to 22.0°C. No American Shad were captured by electrofishing. All American Shad encountered during this survey would have been collected to increase sample sizes for scale analysis and spawning stock composition. The eight fish captured in 2018 were captured in the Denton, MD area using 12.7 cm stretch mesh gill net. The survey was located in the same areas as previous surveys. The goal of the study was to determine Choptank River American Shad habitat preference for spawning

and to capture adult American Shad for assessment. A summary of gill net sets and capture data for adult American Shad is located in Table 18. Age and origin data for adult captures on the Choptank River are found in Table 19.

Choptank River American Shad CPUE

During the nine weeks from 10 April to 30 May when American Shad were sampled by gill net on the Choptank River, the average CPUE was 0.11 fish/h. This is very low compared to the average CPUE for 2017, which was 0.31 fish/h. This is only the third year utilizing these sampling methods. More conclusions on the average CPUE can be made once a larger dataset is developed.

Choptank River American Shad Origin Composition (Hatchery vs. Wild)

In 2018, eight adult American Shad from the Choptank River were retained for origin composition analysis using otolith OTC mark interpretations. Of those eight samples, seven otoliths were successfully analyzed and origin was determined. The samples comprised three hatchery stocked larval origin (43%), four hatchery stocked early juvenile origin (57%) and zero wild origin. A larger data set with more captures is needed for robust analysis using origin composition.

Choptank River American Shad Virgin and Repeat-Spawning Compositions

Eight adult American Shad scale samples were collected in 2018. All of the collected samples were successfully analyzed and used to determine the annual spawning attempt composition. The 2018 sample population consisted of 0% virgin-spawners, 50% second-time spawners and 50% third-time spawners (Table 20). American Shad with additional repeat spawn marks were not identified.

Choptank River American Shad Spawning Stock Discussion

In 2018, the Anadromous Restoration program sampled downriver of the historical electrofishing reach in search of Choptank River American Shad adults. This gill net survey was in its third year and has indicated mixed success. Only eight American Shad were captured. This

number is relatively low compared to the 35 adults captured in 2017 and 45 captured in 2016 (Figure 19).

In previous years, the historical electrofishing sampling area of the Patuxent River yielded both American Shad and Hickory Shad in sufficient quantities to calculate CPUE. The historical electrofishing sampling area on the Choptank River produced sufficient numbers of Hickory Shad to calculate CPUE, while very few American Shad adult samples were collected. Traditional analyses (CPUE, origin, and spawning attempt composition) from electrofishing collections do not permit robust assessment of the spawning stock population dynamics due to small sample size. If the trend in data from 2016 and 2017 gill netting continues, it is possible that a more detailed population analysis can be performed in future years.

Project staff will continue to sample different sections of the Denton, Maryland area of the Choptank River using 12.7 cm gill nets to determine where American Shad are staging and spawning in the river. In 2019, project staff will continue to gill net for adult American Shad, but electrofishing in the historical electrofishing reach will be suspended. Staff will survey lower in the system in an attempt to locate additional adult spawning American Shad. Project staff will continue to use the juvenile American Shad component to estimate Choptank River progress. This is due to sufficient captures of juvenile American Shad, which are used to calculate wild juvenile abundance estimates described in sub-project two. The data collected from increased adult recaptures will further aid in Choptank River assessment of restoration success.

Patapsco River Adult American Shad Spawning Stock

Twenty-Five American Shad were captured on the Patapsco River in 2018 from 12 April to 23 May, (Figure 20). Temperature ranged from 9.9°C to 21.3°C. All American Shad encountered during this survey would have been collected to increase sample sizes for scale analysis and spawning stock composition.

This is the first year that the Maryland Department of Natural Resources sampled this area of the Patapsco River. Previous work was subcontracted to USFWS MDFWCO under a grant administered by the MPA. During MDFWCO assessment, adult hatchery origin American Shad began returning to the Patapsco River in 2017. While all data were presented to the department in report format, processing raw data could not be accomplished before the current

report deadline. In subsequent years, data from MDFWCO will be incorporated into the progress report.

Patapsco River American Shad CPUE

During the nine weeks from 12 April to 31 May 2018 when American Shad were surveyed on the Patapsco River, the mean relative abundance (GM) was calculated as 0.063 fish/min. As stocking and sampling continues, more conclusions can be made as to the health of this population based on the GM.

Patapsco River American Shad Origin Composition (Hatchery vs. Wild)

In 2018, 25 adult American Shad from the Patapsco River were retained for origin composition analysis using otolith OTC mark interpretations (Table 21). Of those 25 samples, 24 otoliths were successfully analyzed and origin was determined. The samples comprised 13 larval hatchery origin (54%), eight early juvenile hatchery origin (33%) and three wild origin (13%). A larger data set is needed for robust analysis using origin composition.

Patapsco River American Shad Virgin and Repeat-Spawning Compositions

Twenty-five American Shad scale samples were collected in 2018. All of the scale samples collected were successfully analyzed to determine the annual spawning attempt composition. The 2018 sample population consisted of 16% virgin spawners, 68% second-time spawners and 16% third-time spawners (Table 22). American Shad that have returned more than three times to spawn would be unlikely because fish were not stocked into the Patapsco River prior to 2012.

Patapsco River American Shad Spawning Stock Discussion

Hatchery stockings have been successful. Larvae and early juvenile stocked American Shad are returning to spawn in the Patapsco River as adults. In 2018, 25 American Shad adults were captured on the Patapsco River. In total, 87% of the sampled fish were of hatchery origin. This marks the second year that hatchery stocked American Shad have returned to the Patapsco River. Three of the American Shad caught in 2018 were of wild origin (13%), indicating that there is a very small remnant population remaining.

The American Shad repeat-spawning composition for 2018 is appropriate at this stage in recovery efforts. American Shad stocked in 2012 make up the majority of the spawners in 2018. The relatively low number of virgin spawners could be attributed to stocking locations in 2013. Shad were stocked upstream in the non-tidal portions of the Patapsco River, where American Shad spawning was thought to be taking place. Project biologists determined that there was not sufficient habitat for American Shad larvae and juveniles at these locations. As a result, stocking sites were moved to the tidal fresh portion of the Patapsco River in 2014 (Figure 6).

Few inferences to the health of the American Shad population can be made with a single year of data. As sampling continues, more conclusions can be made as to the success of recovery efforts. The 2018 estimate of GM will serve as the baseline for future estimates.

2019 Sub-Project 3 – Preliminary Results – Work in Progress

Choptank River

Gill net surveys were conducted on the Choptank River for eight weeks from 4 April to 23 May 2019. Eight adult American Shad were captured. All fish were retained for scale age and otolith analysis. A complete analysis of CPUE, origin composition and repeat spawning analysis will be presented in the 2020 F-57-R report.

Patapsco River

Electrofishing surveys were conducted on the Patapsco River for 12 weeks from 21 March to 6 June 2019. Thirty-eight adult American Shad were captured and retained for scale age and otolith analysis. A complete analysis of CPUE, origin composition and repeat spawning analysis will be presented in the 2020 F-57-R report.

2018 Overall Restoration Progress

Choptank River

Determining the progression of the American Shad restoration program through time has been difficult due to low numbers of adult American Shad captures and the annual variability of wild juvenile abundance estimates. Even though the wild juvenile abundance has been variable,

wild juveniles are captured in the summer seine survey. Four-hundred ninety juvenile American shad were analyzed from the summer seine survey. Forty-five wild American Shad juveniles (9% of the catch) were captured during the survey, which indicates that wild spawning is taking place. The goal of the 2018 adult gill netting was to explore the vicinity of Denton, Maryland in attempt to capture adult American Shad returning to the Choptank River. Previous years sampling indicated that the vicinity around Denton was a prime area for capturing adult shad with 46 and 50 captures in 2016 and 2017 respectively. Only eight American Shad adults were captured by gill net on the Choptank River in 2018. The American Shad samples in the vicinity of the Denton, Maryland boat ramp continue to be dominated by female fish. In 2018, No American Shad were captured in the historical electrofishing area. Previous years electrofishing efforts suggest males and females are utilizing different areas of the river, presumptively prior to spawning. Project biologists will continue searching where adult shad are spawning in the river in the future.

Patapsco River

Any conclusions regarding the success of American Shad restoration in the Patapsco River would be premature. At this time, the only conclusions that can be made are that stockings have been successful for juvenile recruitment and that hatchery raised American Shad are beginning to return to the Patapsco River as adults to spawn. In 2018, 25 adult American shad consisting of 11 females and 14 males were captured in the spring electrofishing survey. Of the 25 captures, only three samples were of wild origin. Each of the wild origin captures were age estimated at 5 to 6 years old, suggesting that these fish were from a remnant wild population or were strays from another system. Additional years of stocking and assessment will lead to a more refined analysis of the recovery efforts.

Patuxent River

No sampling occurred on the Patuxent River for American Shad adults in 2018. The American Shad population is sampled every three years to maintain trend data. The Patuxent River American Shad adult population will be assessed again in 2020.

Project-wide Observations 2019

Project wide observations for the 2019 calendar year will be discussed in the next reporting cycle. Sub-project two will be completed for 2019 and all data will be analyzed to give a more complete picture of restoration efforts in the Choptank and Patapsco rivers. These data will be presented in the 2020 reporting period.

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Table 1. *Maryland Department of Natural Resources rotating OTC marks for larval stocked American Shad from 2009-2018.*

Year	Rotating Mark (day)
2009	3
2010	3,9
2011	3,6,10
2012	3
2013	3,9
2014	3,6,10
2015	3
2016	3,9
2017	3,6,10
2018	3

Table 2. *2018 Maryland Department of Natural Resources American Shad stocking goals for the Choptank River.*

Stocking phase	Stocking goal	Stocked
Larvae	2,750,000	1,550,000
Early Juvenile	450,000	460,000

Table 3. *2018 Maryland Department of Natural Resources American Shad stocking goals for the Patapsco River.*

Stocking phase	Stocking goal	Stocked
Larvae	200,000	260,000
Early Juvenile	75,000	75,000

Table 4. *Maryland Department of Natural Resources American Shad broodstock and production data for 2018. Strip spawn collections were conducted on the Potomac River near Fort Belvoir, Virginia. Grey blocks represent data that could not be determined because eggs were released directly back into the Potomac River, or there was an equipment failure at the hatchery, and eggs were unable to be counted. Averages and estimates do not include the days with grey boxes.*

Date	Ripe Females	Total shad	Liters of eggs	Liters of fertilized eggs	Egg fertility	Total viable eggs
4/20/2018	0	10	0	0.0	0.0	0.0
4/22/2018	0	13	0	0.0	0.0	0.0
4/23/2018	17	71	5	4.2	83.40%	145,000
4/24/2018	53	78	15			
4/25/2018	32	70	7	4.0	57.59%	221,200
4/26/2018	88	157	30	12.3	41.12%	1,014,000
4/27/2018	66	150	37	19.8	53.43%	1,239,500
4/29/2018	60	112	24	12.5	52.21%	758,400
4/30/2018	45	98	20	10.3	51.59%	628,000
5/1/2018	54	99	18			
5/2/2018	69	167	36			
5/3/2018	75	208	37	17.2	46.57%	1,317,200
5/4/2018	56	190	21	13.3	63.11%	728,700
5/6/2018	75	170	42	28.1	67.02%	1,566,600
5/7/2018	76	150	37	15.5	41.84%	1520700
5/8/2018	200	362	90	54.1	60.15%	3,546,000
5/10/2018	53	109	15	0.0	0.00%	261,000
5/11/2018	57	125	5	0.0	0.00%	188,000
5/13/2018	25	44	15	9.8	65.27%	540000
5/15/2018	20	25	0	0.0	0.00%	0
5/16/2018	30	67	20	12.6	63.03%	714000
5/17/2018	11	37	7	4.5	64.64%	253400
5/21/2018	2	47	2			
Total	**1164**	**2559**	**483**	**218.3**	**50.69%**	**7,812,943**

Table 5. *2018 Maryland Department of Natural Resources American Shad stocking events in the Choptank River. Mark is the day age of OTC larval immersion.*

Date	Life stage	Mark	Number
5/16/2018	Larvae	Day-3	550,000
5/17/2018	Larvae	Day-3	1,000,000
6/20/2018	Early Juvenile	Day-3,6	335,000
6/25/2018	Early Juvenile	Day-3,6	50,000
6/26/2018	Early Juvenile	Day-3,6	75,000

Table 6. *2018 Maryland Department of Natural Resources American Shad stocking events in the Patapsco River. Mark is the day age of OTC larval immersion.*

Date	Life stage	Mark	Number
5/24/2018	Early Juvenile	Day-3,6	40,000
5/27/2018	Larvae	Day-3	260,000
6/7/2018	Early Juvenile	Day-3,6	35,000

Table 7. *Historical stocking summary for larval and juvenile American Shad in the Choptank River since the inception of the restoration effort (1996-2018).*

Choptank River American Shad

Year	Larvae	Early Juveniles	Late Juveniles
1996	626,000	0	115,110
1997	1,245,000	0	32,612
1998	136,000	0	16,885
1999	442,000	0	64,956
2000	357,000	0	64,369
2001	0	15,000	32,483
2002	1,020,000	100,000	23,118
2003	1,322,000	167,500	0
2004	675,000	125,000	28,898
2005	1,930,000	170,000	41,483
2006	1,720,000	199,800	0
2007	980,000	139,000	0
2008	985,000‡	35,000	0
2009	980,000	139,000	0
2010	3,725,000	530,000	34,272
2011	1,621,922	269,500	45,000
2012	3,692,956	548,000	69,900
2013	3,120,000	441,000	40,000
2014	1,390,000	421,000	50,000
2015	635,000	456,000	38,000
2016	1,905,000	537,000	25,000
2017	2,530,000	403,000	0
2018	1,550,000	535,000	0
Total	**31,602,878**	**5,230,800**	**722,086**

‡ *Stockings include 495,000 day 3,6,9 marked larvae. Only day 3 marked larvae are used in abundance estimates.*

Table 8. *Historical stocking summary for larval and juvenile American Shad in the Patapsco River since the inception of American Shad restoration efforts.*

Patapsco River American Shad

Year	Larvae	Early Juveniles
2012	925,000	0
2013	200,000	95,000
2014	90,000	70,000
2015	220,000	90,000
2016	215,000	75,000
2017	225,000	75,000
2018	260,000	75,000
Total	**2,135,000**	**480,000**

Table 9. *Previously conducted restoration effort for larval and juvenile American Shad in the Patuxent River.*

Patuxent River American Shad

Year	Larvae	Early Juveniles	Late Juveniles
1994	14,000	0	89,760
1995	346,000	0	121,124
1996	655,000	0	173,994
1997	1,345,000	0	60,040
1998	61,000	0	16,726
1999	526,000	0	60,377
2000	349,000	37,250	26,765
2001	364,000	77,500	21,903
2002	472,000	124,750	24,968
2003	717,000	108,000	31,061
2004	537,000	93,000	36,571
2005	708,000	93,000	40,873
2006	720,000	222,300	93,808
2007	431,000	170,500	34,382
2008	490,000	150,000	0
2009	758,000	130,000	25,954
Total	**8,493,000**	**1,206,300**	**832,352**

Table 10. *Previously conducted restoration effort for larval and juvenile American Shad in Marshyhope Creek.*

Marshyhope Creek American Shad

Year	Larvae	Early Juveniles	Late Juveniles
2002	100,000	39,000	9,074
2003	243,000	50,000	0
2004	238,000	33,000	0
2005	205,000	40,000	0
2006	500,000	100,000	0
2007	0	137,000	0
2008	335,000‡	119,500	0
2009	330,000	78,000	0
Total	**1,951,000**	**596,500**	**9,074**

‡ *Stockings include 85,000 day 3,6,9 marked larvae. Only day 3 larvae are used in abundance estimates.*

Table 11. *Historical stocking summary for larval and juvenile American Shad in the Nanticoke River since the inception of the restoration effort (1995-2006). Only fish raised and stocked by the Maryland Department of Natural Resources are included. The state of Delaware also raises and stocks shad for the mainstem Nanticoke River and those figures are not included in these data.*

Nanticoke River American Shad

Year	Larvae	Early Juveniles	Late Juveniles
1995	34,000	0	8,400
1996	0	0	0
1997	152,000	0	0
1998	0	0	0
1999	0	0	0
2000	0	0	0
2001	40,000	0	0
2002	90,000	20,000	13,347
2003	324,000	73,500	0
2004	100,000	60,000	0
2005	275,000	60,000	0
2006	0	40,500	0
Total	**1,015,000**	**254,000**	**21,747**

Table 12. *Juvenile American Shad recaptures in Choptank River from the Maryland Department of Natural Resources summer seine survey since inception of the restoration effort, 1996-2018. Data are percentage of origin composition of all juveniles collected by the survey. n=number of captured juvenile American Shad that were successfully analyzed for origin*.

Sample Year	n	Larval Stocked Origin	Early Juvenile Stocked Origin	Late Juvenile Stocked Origin	Wild Fish
1996	99	37%	NA	63%	0%
1997‡	NA	NA	NA	NA	NA
1998	1	100%	NA	0%	0%
1999	13	36%	NA	62%	0%
2000	8	0%	NA	100%	0%
2001	41	0%	32%	51%	17%
2002	200	58%	25%	8%	9%
2003	188	36%	48%	NA	16%
2004	145	52%	41%	1%	5%
2005	213	76%	14%	1%	9%
2006	290	72%	19%	NA	9%
2007	263	43%	41%	NA	16%
2008	94	43%	38%	NA	19%
2009	151	66%	26%	NA	7%
2010	551	31%	62%	1%	6%
2011	341	19%	75%	2%	5%
2012	550	20%	70%	3%	8%
2013	299	18%	60%	15%	6%
2014	443	21%	66%	5%	8%
2015	531	37%	43%	10%	10%
2016	300	35%	54%	3%	8%
2017	489	46%	44%	NA	10%
2018	483	44%	47%	NA	9%

‡There are no data available for 1997.

Table 13. *Number and location of American Shad juveniles collected in the Maryland Department of Natural Resources 2018 Choptank River seine survey. n=number of juveniles collected before analysis for hatchery marks.*

Site	8/1	8/8	8/15	8/22	8/28	9/5	9/12	9/19	9/25	Total
Depue Landing	2	0	21	0	19	8	18	7	6	**81**
High School	1	1	3	7	7	1	7	11	3	**41**
Railroad Bridge	1	0	0	0	1	0	0	11	1	**14**
Guano Company	19	0	1	4	2	21	7	13	6	**73**
Martinak	2	20	4	7	17	2	7	0	4	**63**
Medfield Lane	0	0	0	4	0	0	1	2	9	**16**
Fallen Trees	19	0	16	6	4	10	4	19	13	**91**
Stoney Point	4	12	8	4	4	6	0	5	12	**55**
Dover Bridge	8	0	2	9	1	0	0	2	4	**26**
Fossil Cliff	13	0	2	9	4	0	0	6	2	**36**
Total	**69**	**33**	**57**	**50**	**55**	**48**	**44**	**76**	**60**	**496**

Table 14. *Estimates of stocked American Shad larval survival and instantaneous mortality to the date of early juvenile stocking in the Choptank River, 2002-2018.*

Year	Instantaneous Mortality (Z)	±2 S.E.	Survival	Interval (days)
2002	0.0677	0.0015	0.2255	22
2003	0.1243	0.0304	0.0943	19
2004	0.0690	0.0810	0.2346	21
2005	0.0290	0.2007	0.4757	24
2006	0.0440	0.1305	0.4335	19
2007	0.0652	0.0407	0.1511	29
2008	0.0459	0.0383	0.0800	55
2009	0.0571	0.1066	0.2850	22
2010	0.0975	0.0135	0.0720	27
2011	0.1444	0.0117	0.0417	22
2012	0.0691	0.0091	0.0416	46
2013	0.1571	0.0133	0.0432	20
2014	0.1380	0.0228	0.0958	17
2015	0.0280	0.1204	0.6209	17
2016	0.0391	0.0464	0.1856	43
2017	0.0418	0.0316	0.1659	43
2018	0.0321	0.0618	0.3256	35

Table 15. *Estimates of American Shad juvenile abundance in the Choptank River on 20 June 2018. Estimates were calculated using Chapman's modification to the Peterson equation (95% confidence interval).*

Life Stage	Peterson Estimate	Upper Limit	Lower Limit
Larval Stocked	503,461	572,695	442,570
Early Juvenile Stocked	533,762	607,162	469,206
Wild Juveniles	104,888	119,311	92,202
Totals	**1,142,111**	**1,299,168**	**1,003,977**

Table 16. *1996-2018 American Shad summer juvenile abundance estimates in the Choptank River. Estimates were calculated using Chapman's modification to the Peterson equation (95% confidence interval, numbers may not add up due to rounding).*

Year	Larval Stocked Origin	Early juvenile Stocked Origin	Late Juvenile Stocked Origin	Wild Origin	Total Juveniles
1996	28,600	80,700	0	0	109,300
1997‡	NA	NA	0	NA	NA
1998‡	NA	NA	0	NA	NA
1999‡	NA	NA	0	NA	NA
2000‡	NA	NA	0	NA	NA
2001‡	NA	NA	0	NA	NA
2002	231,200	100,500	36,200	36,200	404,000
2003	124,000	168,400	0	57,300	349,800
2004	159,400	125,900	4,200	14,700	304,200
2005	922,300	170,800	11,400	108,200	1,212,700
2006	748,300	200,500	0	89,500	1,038,300
2007	148,700	139,500	0	54,800	343,000
2008	48,200	35,400	0	17,200	100,800
2009	377,500	151,000	0	41,500	570,000
2010	268,600	531,000	11,000	50,000	860,400
2011	68,000	270,300	7,400	15,900	361,500
2012	154,000	549,000	20,000	61,300	784,300
2013	135,200	442,500	113,100	44,200	735,000
2014	133,000	420,500	31,500	48,650	633,600
2015	393,300	455,000	103,000	103,000	1,054,300
2016	353,000	535,000	33,000	76,500	998,000
2017	418,700	402,000	NA	89,300	910,000
2018	503,500	533,800	NA	104,900	1,142,100

‡Insufficient sample size to calculate estimate

Table 17. *Maryland Department of Natural Resources adult American Shad electrofishing survey starting and ending coordinates for the Choptank and Patapsco rivers.*

River	Starting latitude/longitude	Ending latitude/longitude
Choptank River	38.984728° N -075.788325° W	38.977021° N -075.801606° W
Patapsco River upstream	39.224738° N -076.640593° W	39.225127° N -076.628076° W
Patapsco River downstream	39.229178° N -076.625855° W	39.239058° N -076.616996° W

Table 18. *Maryland Department of Natural Resources 2018 American Shad gill net sets and captures for American Shad on the Choptank River with associated CPUE.*

Date	Set # (1 net only)	Set Time	Pull Time	# of Fish Caught	Hours Fished	CPUE (Fish/Hour)	Average CPUE	
4/10/2018	1	11:46	15:13	1	3:27	0.290	4/10/2018	0.145
4/10/2018	2	12:04	15:28	0	3:24	0.000		
4/19/2018	2	11:06	15:28	0	4:22	0.000	4/19/2018	0.000
4/19/2018	1	11:16	15:18	0	4:02	0.000		
4/24/2018	2	10:55	15:20	1	4:25	0.226	4/24/2018	0.353
4/24/2018	1	11:23	15:33	2	4:10	0.480		
4/30/2018	1	10:30	15:20	1	4:50	0.207	4/30/2018	0.103
4/30/2018	2	10:38	15:33	0	4:55	0.000		
5/07/2018	2	8:57	14:14	2	5:17	0.379	5/07/2018	0.189
5/07/2018	1	9:06	14:34	0	5:28	0.000		
5/15/2018	1	11:21	14:57	0	3:36	0.000	5/15/2018	0.000
5/15/2018	2	11:31	15:11	0	3:40	0.000		
5/23/2018	1	10:26	15:32	1	5:06	0.196	5/23/2018	0.098
5/23/2018	2	10:32	15:45	0	5:13	0.000		
5/30/2018	1	10:16	15:21	0	5:05	0.000	5/30/2018	0.000
5/30/2018	2	10:22	15:34	0	5:12	0.000		

Table 19. *2018 Maryland Department of Natural Resources American Shad adults caught in the Choptank River.*

Date	Sample #	Sex	Age	Spawn Marks	Origin
4/10/2018	2018-0101	Female	6	2	Larvae
4/24/2018	2018-0120	Female	7	2	Early Juvenile
4/24/2018	2018-0121	Female	6	1	Larval
4/24/2018	2018-0122	Female	6	1	Early Juvenile
4/30/2018	2018-0163	Female	6	2	Early Juvenile
5/07/2018	2018-0169	Female	6	2	Larval
5/07/2018	2018-0170	Female	5	1	No Sample
5/23/2018	2018-0172	Female	5	1	Early Juvenile

Table 20. *2016-2018 Maryland Department of Natural Resources American Shad adult recapture survey spawning composition on the Choptank River. In 2017 adults were sampled with both electrofishing and gillnetting gear.*

		Spawning Attempts			
Year	Sample Size (n)	Virgins	2	3	4
2016	46	0 (0%)	16 (33%)	18 (39%)	11 (24%)
2017	50	32 (64%)	7 (14%)	9 (18%)	2 (4%)
2018	8	0 (0%)	4 (50%)	4 (50%)	0 (0%)

Table 21. *2018 Maryland Department of Natural Resources American Shad adults caught in the Patapsco River.*

Date	Sample #	Sex	Age	Spawn Marks	Origin
4/12/2018	2018-0114	Female	5	0	Wild
4/12/2018	2018-0115	Male	5	1	Early Juvenile
4/23/2018	2018-0117	Female	5	1	Wild
4/23/2018	2018-0118	Male	5	1	Early Juvenile
4/23/2018	2018-0119	Male	6	1	Larval
5/3/2018	2018-0165	Male	5	1	Early Juvenile
5/3/2018	2018-0166	Male	5	1	Early Juvenile
5/3/2018	2018-0167	Male	6	2	Larval
5/3/2018	2018-0168	Female	6	2	Larval
5/23/2018	2018-0173	Female	6	1	Larval
5/23/2018	2018-0174	Male	6	1	Larval
5/23/2018	2018-0175	Male	5	2	Early Juvenile
5/23/2018	2018-0176	Male	6	1	Wild

5/23/2018	2018-0177	Male	6	0	No Sample
5/23/2018	2018-0178	Female	6	1	Larval
5/23/2018	2018-0179	Male	6	2	Larval
5/23/2018	2018-0180	Male	6	1	Larval
5/23/2018	2018-0181	Female	6	1	Larval
5/23/2018	2018-0182	Female	6	1	Larval
5/23/2018	2018-0183	Male	4	1	Early Juvenile
5/23/2018	2018-0184	Female	5	0	Larval
5/23/2018	2018-0185	Male	5	1	Larval
5/23/2018	2018-0186	Female	5	0	Early Juvenile
5/23/2018	2018-0187	Female	5	1	Early Juvenile
5/23/2018	2018-0188	Female	6	1	Larval

Table 22. *Patapsco River American Shad spawning attempt composition for sample year 2018.*

		Spawning Attempts					
Year	**Sample Size (n)**	**Virgins**	**2**	**3**	**4**	**5**	**6**
2018	25	4 (16%)	17 (68%)	4 (16%)	0 (0%)	0 (0%)	0 (0%)

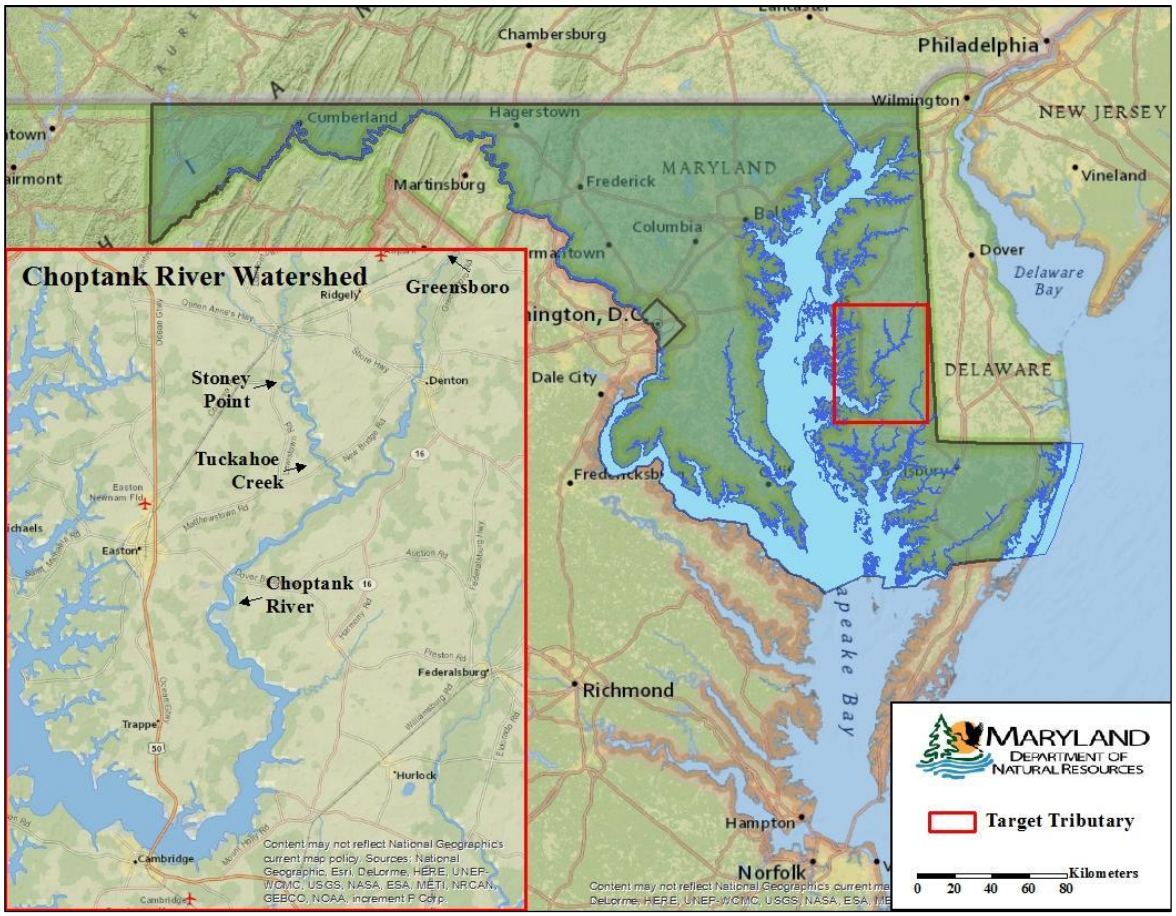

Figure 1. *Choptank River target tributary and culture sites for the Maryland Department of Natural Resources shad restoration project.*

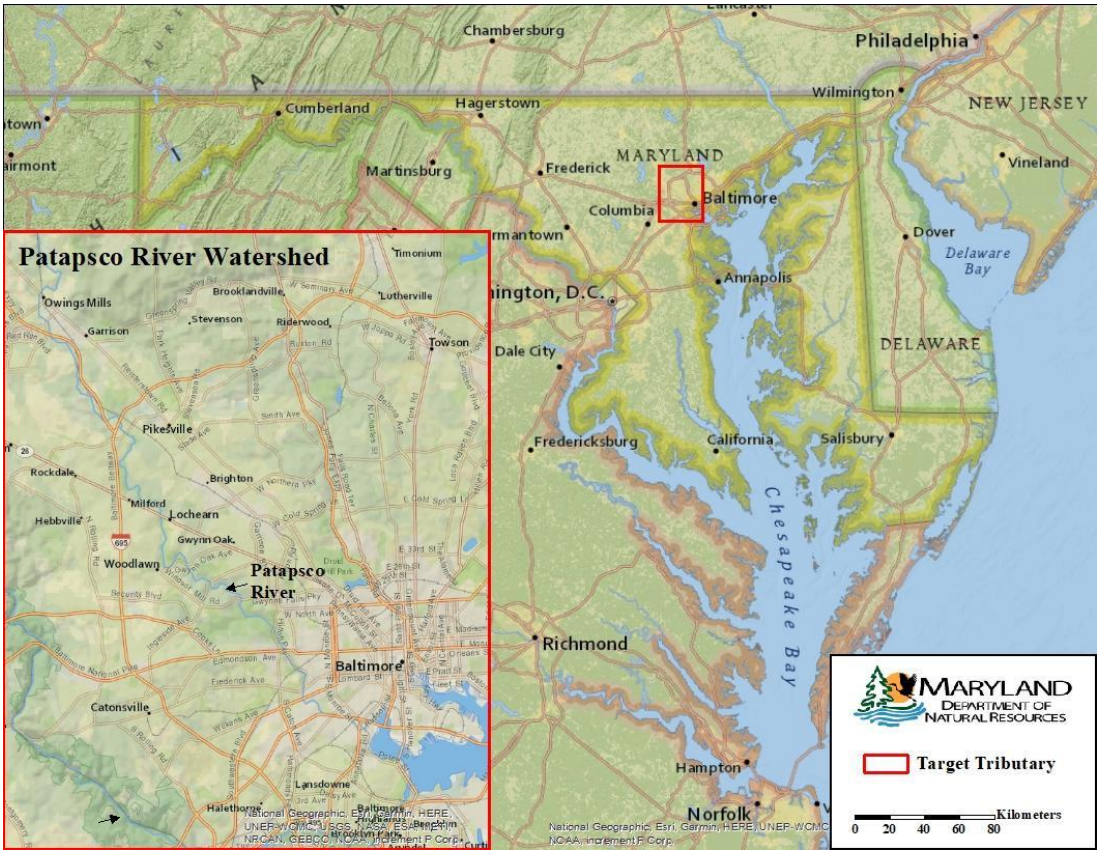

Figure 2. *Patapsco River target tributary and culture sites for the Maryland Department of Natural Resources shad restoration project.*

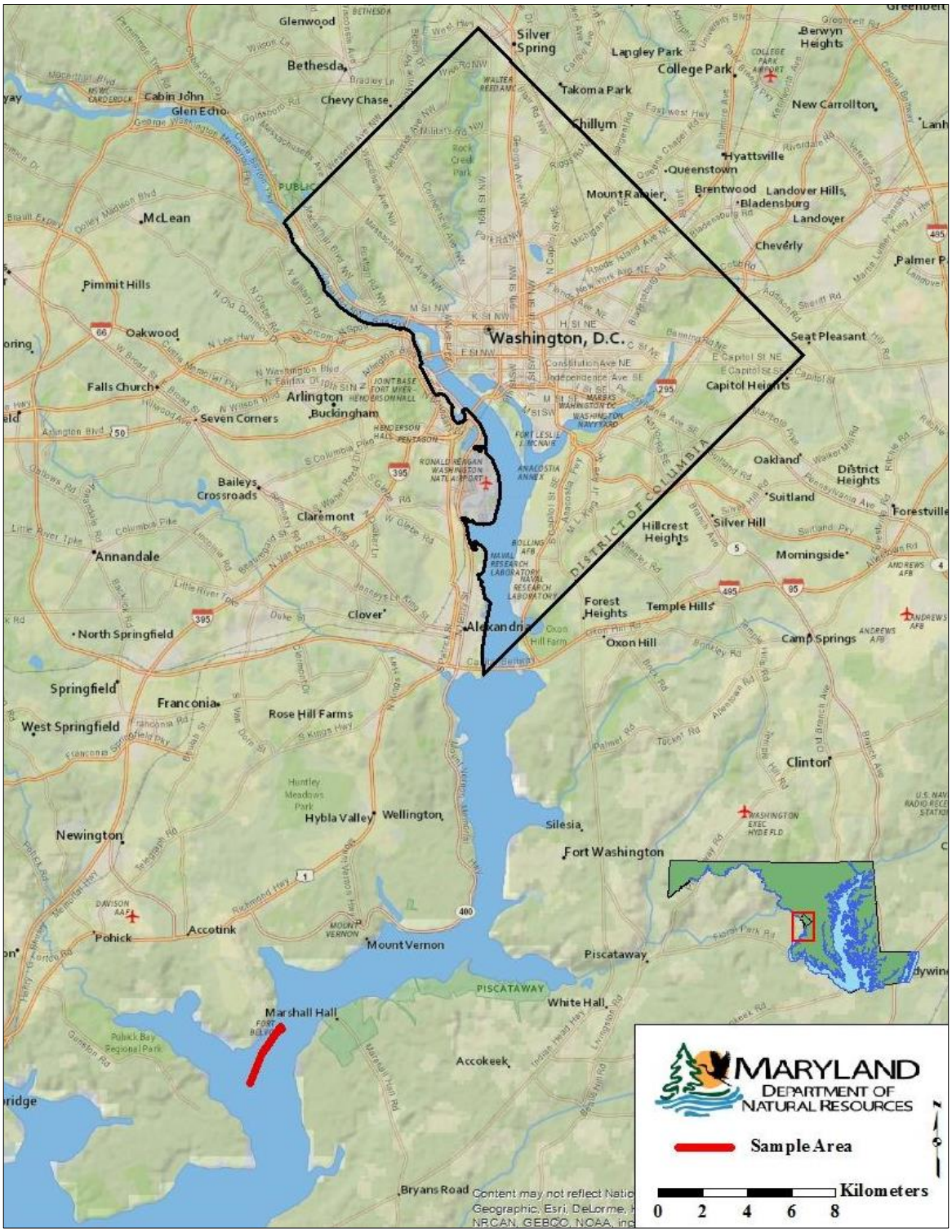

Figure 3. *2018 Maryland Department of Natural Resources American Shad brood stock collection site on the Potomac River.*

Figure 4. *Maryland Department of Natural Resources participating fish culture facilities in the restoration project. GenOn Energy is a power company that allows the project to culture fish for the restoration effort in their onsite ponds. The University of Maryland Center for Environmental Science (UMCES) Horn Point Aquaculture and Restoration Ecology Laboratory (AREL) is a facility that supplies culture ponds for the restoration effort.*

Figure 5. *Maryland Department of Natural Resources Choptank River American Shad stocking sites in 2018*

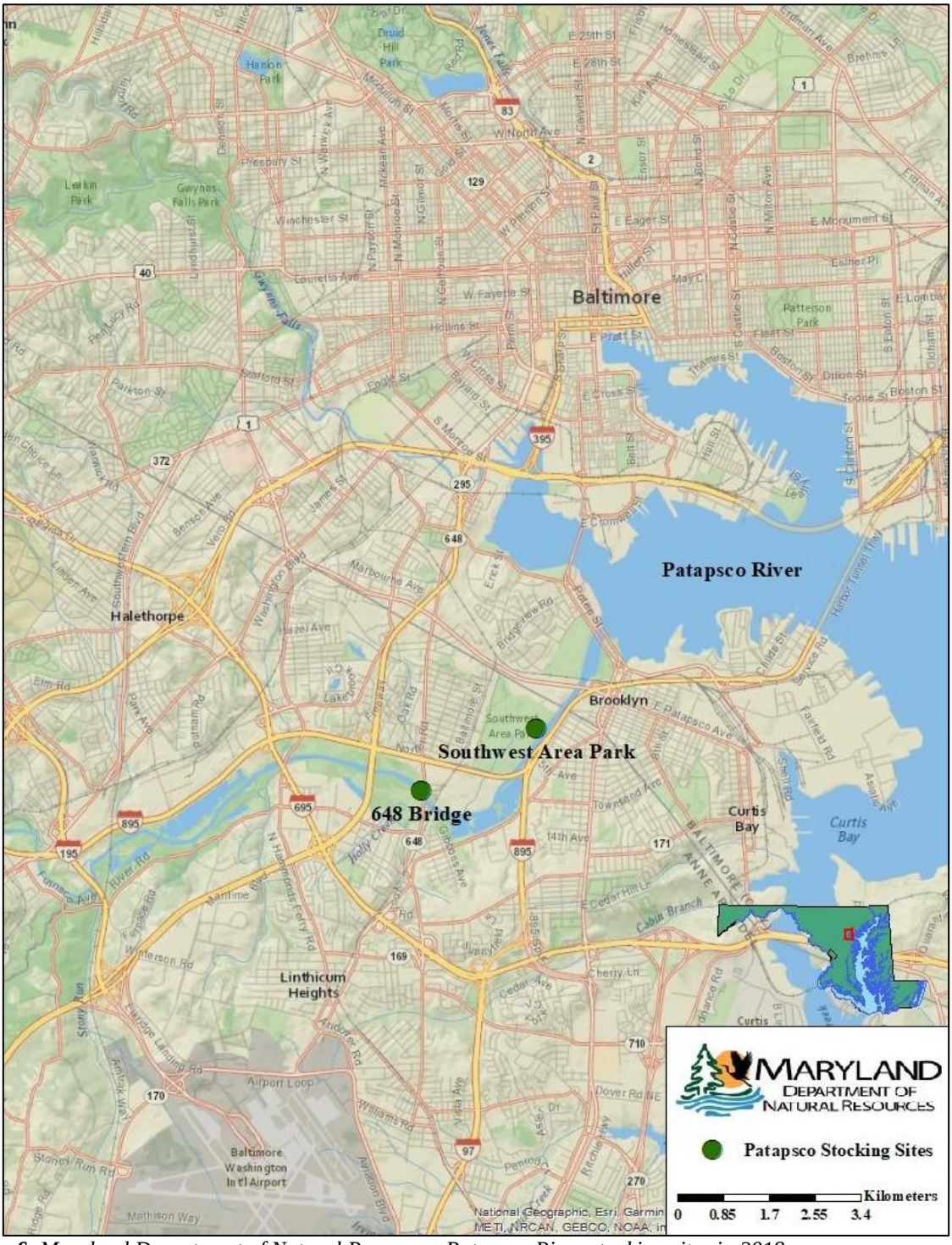

Figure 6. *Maryland Department of Natural Resources Patapsco River stocking sites in 2018.*

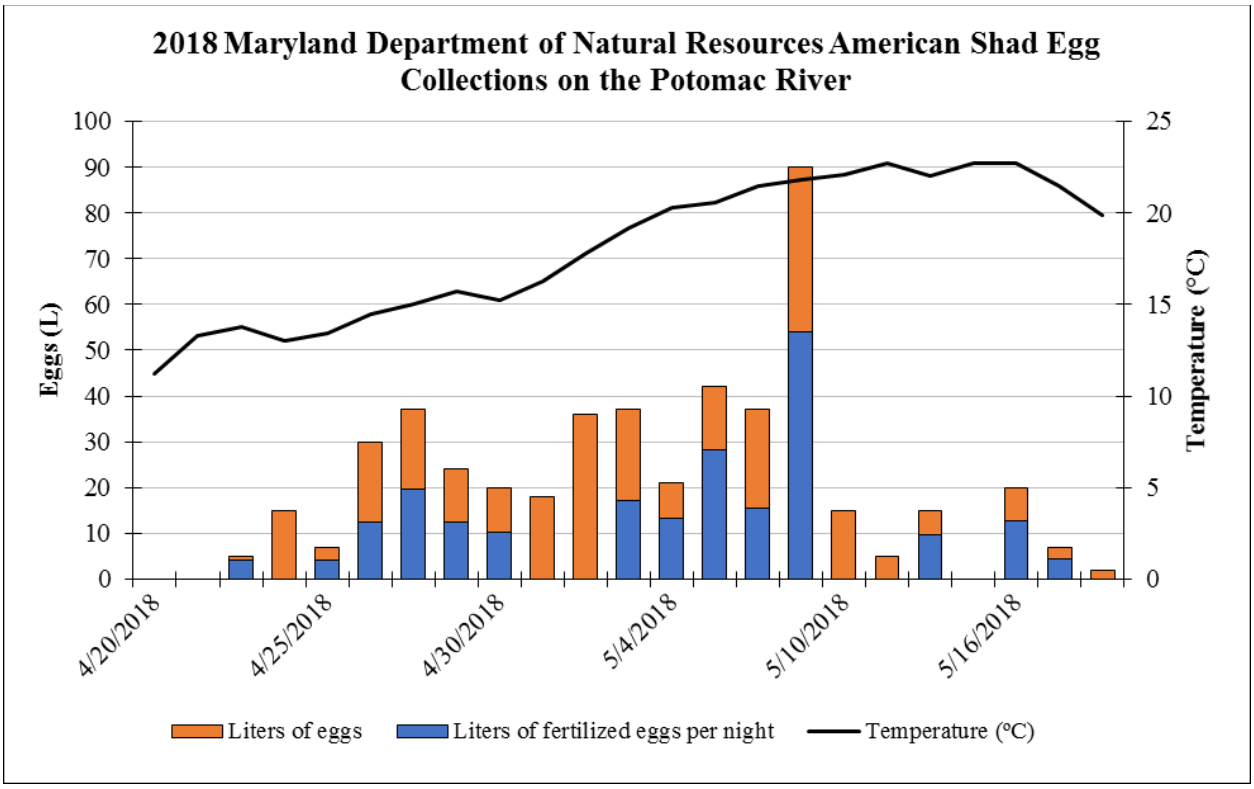

Figure 7. *2018 Maryland Department of Natural Resources volume of total American Shad eggs and viable eggs collected from the Potomac River, along with daily Potomac River temperature.*

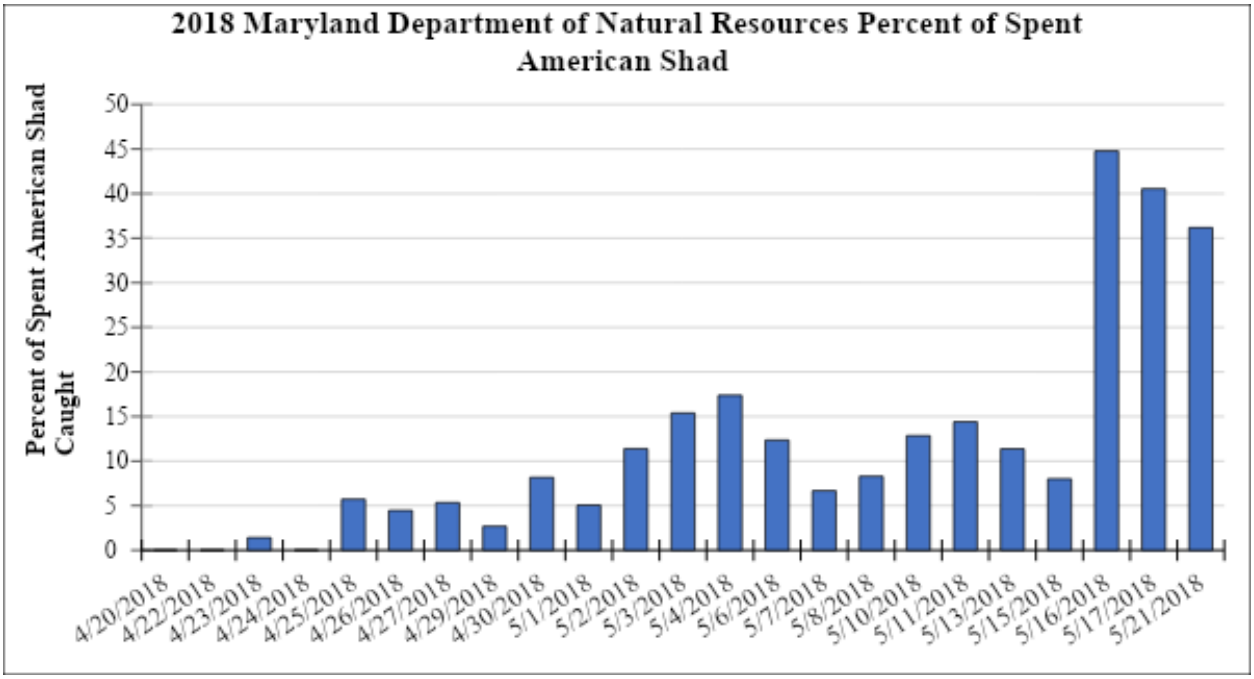

Figure 8. *2018 Maryland Department of Natural Resources percent of spent American Shad collected each day from the Potomac River.*

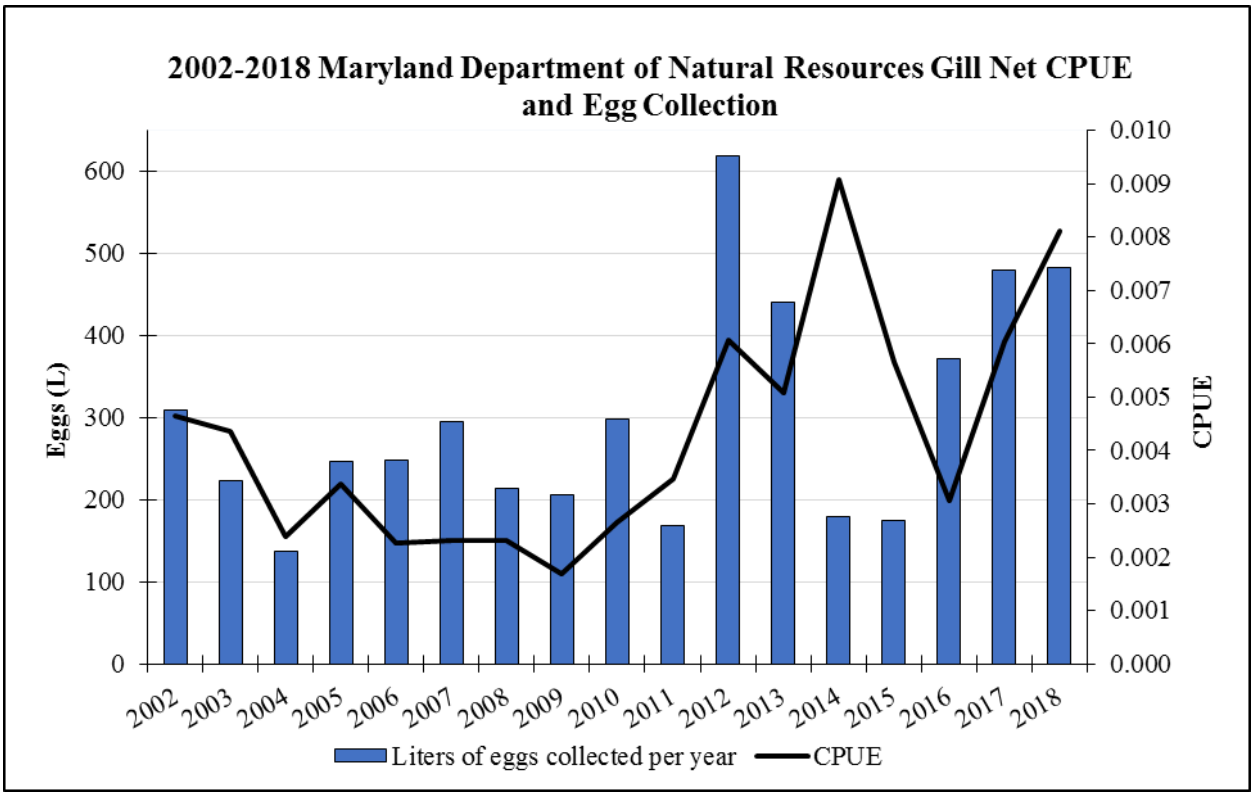

Figure 9. *2002-2018 Maryland Department of Natural Resources gill net CPUE and egg collection on the Potomac River.*

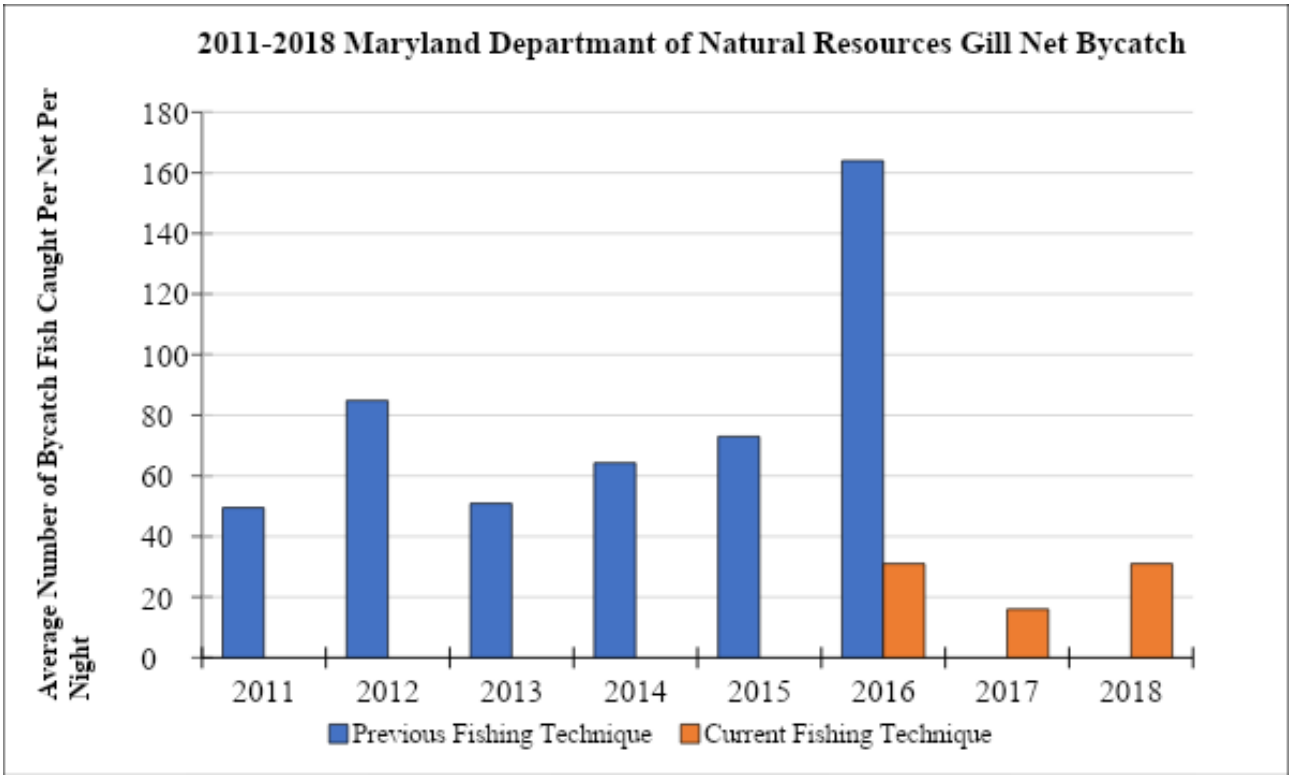

Figure 10. *2011-2018 Maryland Department of Natural Resources gill net bycatch on the Potomac River.*

Figure 11. *Maryland Department of Natural Resources Choptank River juvenile American Shad survey seine sites sampled in 2018.*

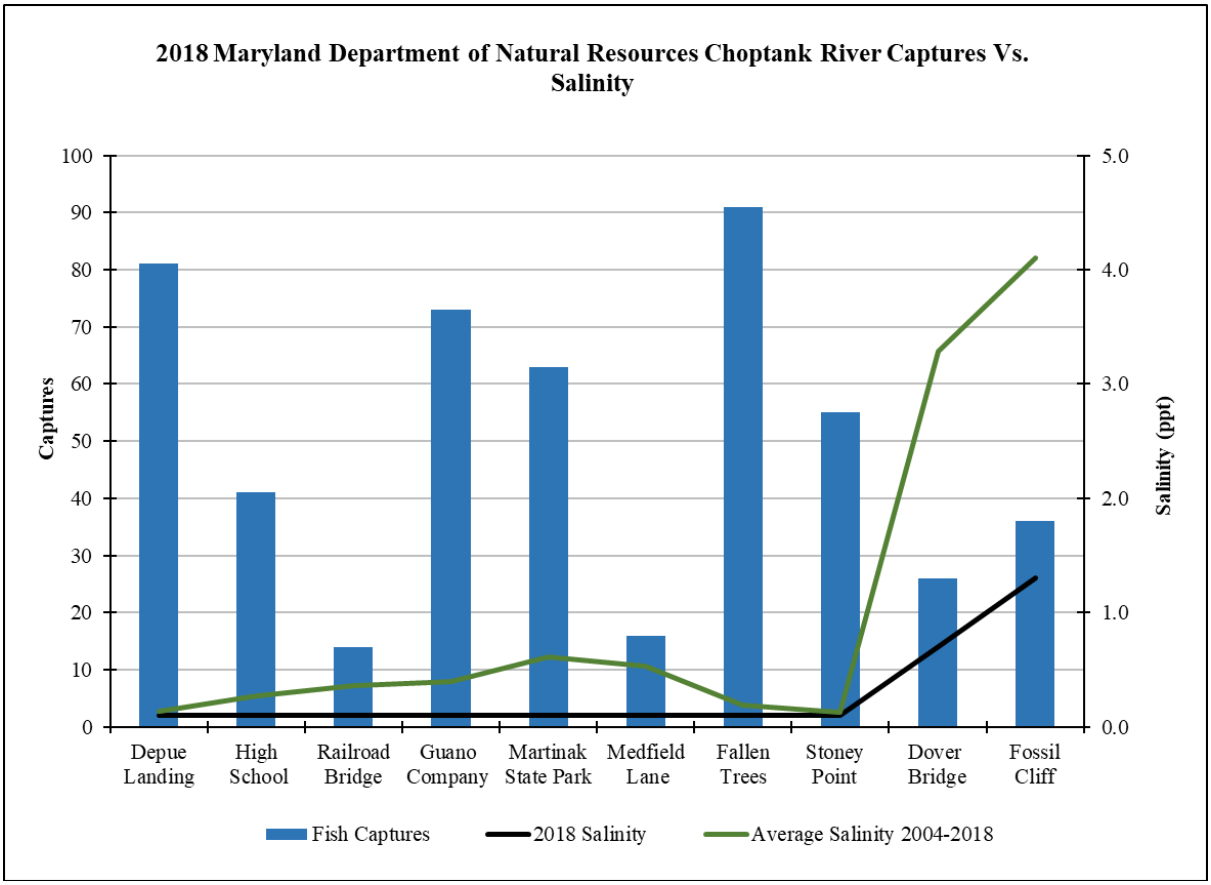

Figure 12. *2018 Maryland Department Natural Resources summer seine survey American Shad juvenile catch composition and salinity by site in the Choptank River and Tuckahoe Creek. Stoney Point, Fallen Trees and Medfield Lane are sample sites on the Tuckahoe Creek.*

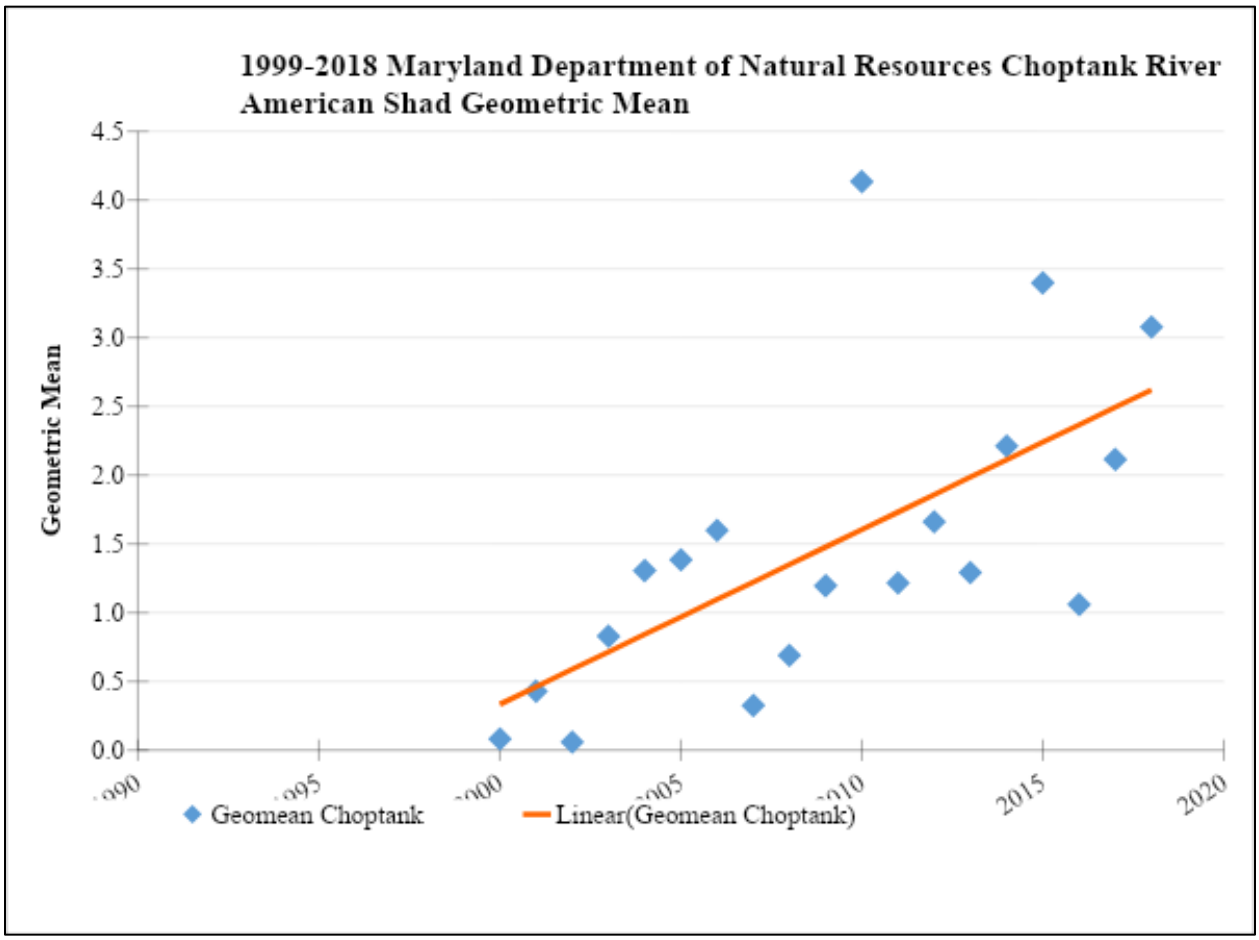

Figure 13. *Maryland Department of Natural Resources Choptank River juvenile American Shad historical geometric mean (GM). Data were generated from the permanent summer seine survey sites conducted on the Choptank River from 1999-2018.*

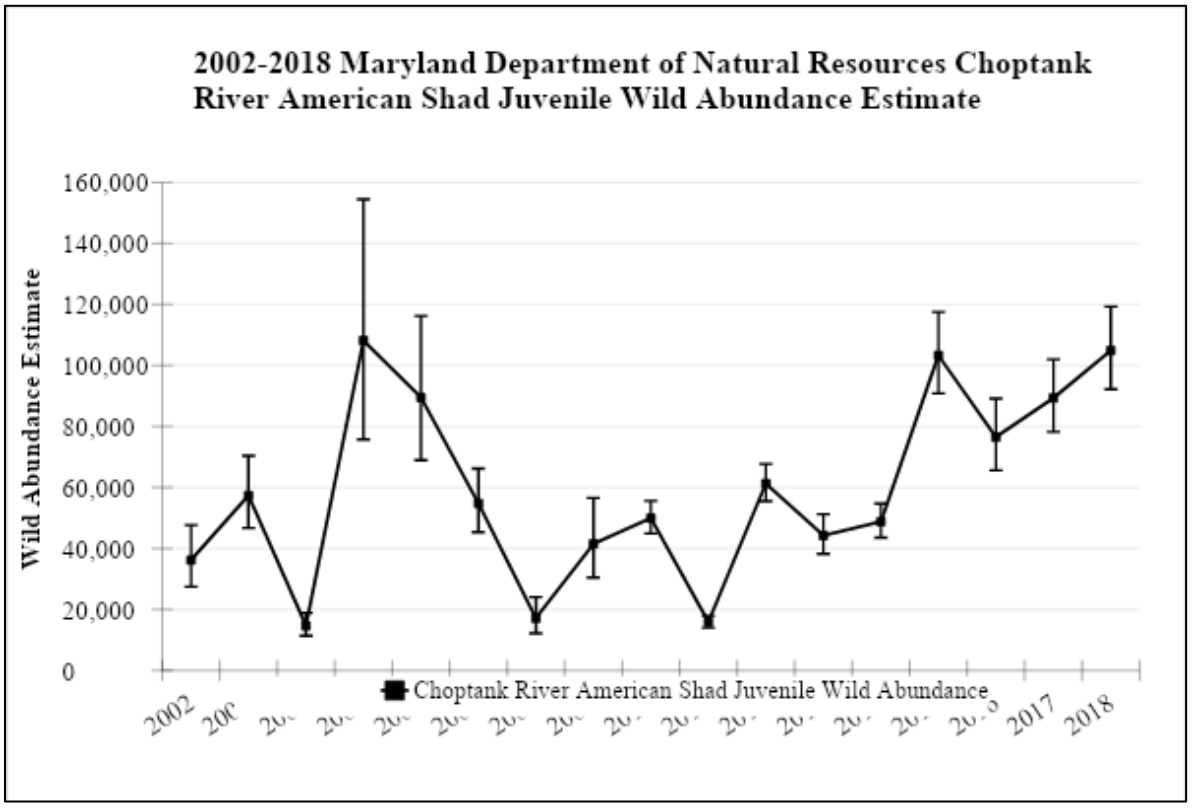

Figure 14. *2002-2018 Maryland Department of Natural Resources American Shad summer juvenile wild abundance estimates in the Choptank River. Estimates were calculated using Chapman's modification to the Peterson equation (95% confidence interval).*

Figure 15. *Maryland Department of Natural Resources American Shad electrofishing survey areas on the Choptank River sampled in 2018.*

Figure 16. *Maryland Department of Natural Resources American Shad electrofishing survey areas on the Patapsco River sampled in 2018.*

Figure 17. *Maryland Department of Natural Resources American Shad survey areas sampled by gillnet in 2018.*

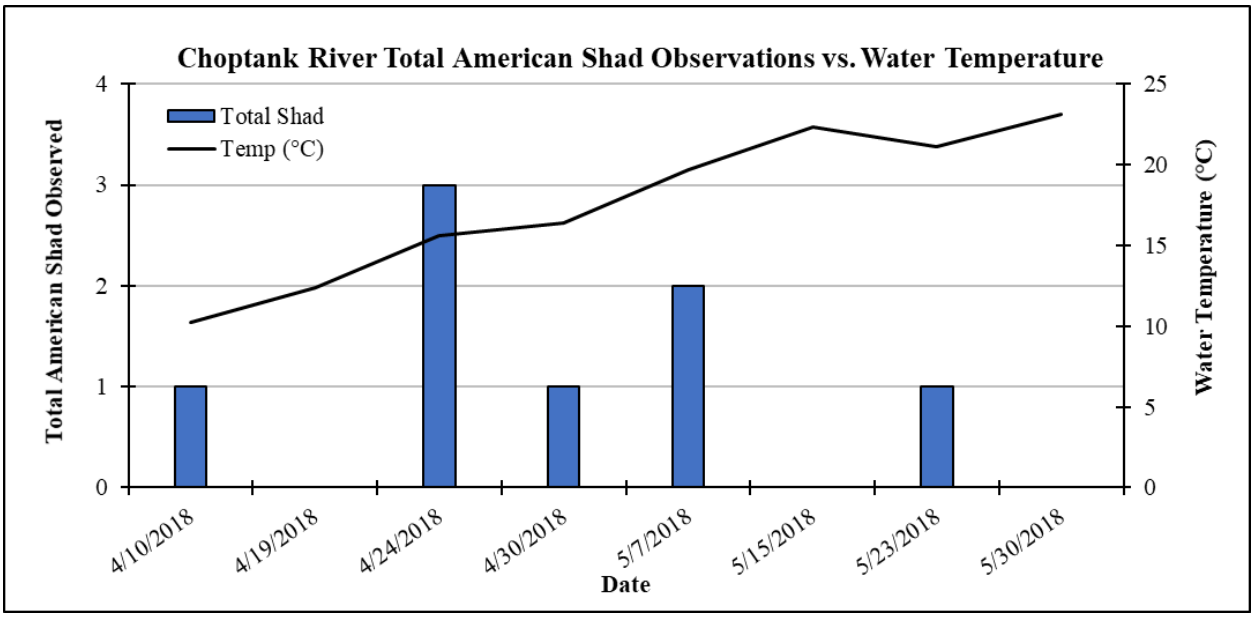

Figure 18. *2018 Maryland Department of Natural Resources gill net collections and observations of adult American Shad on the Choptank River.*

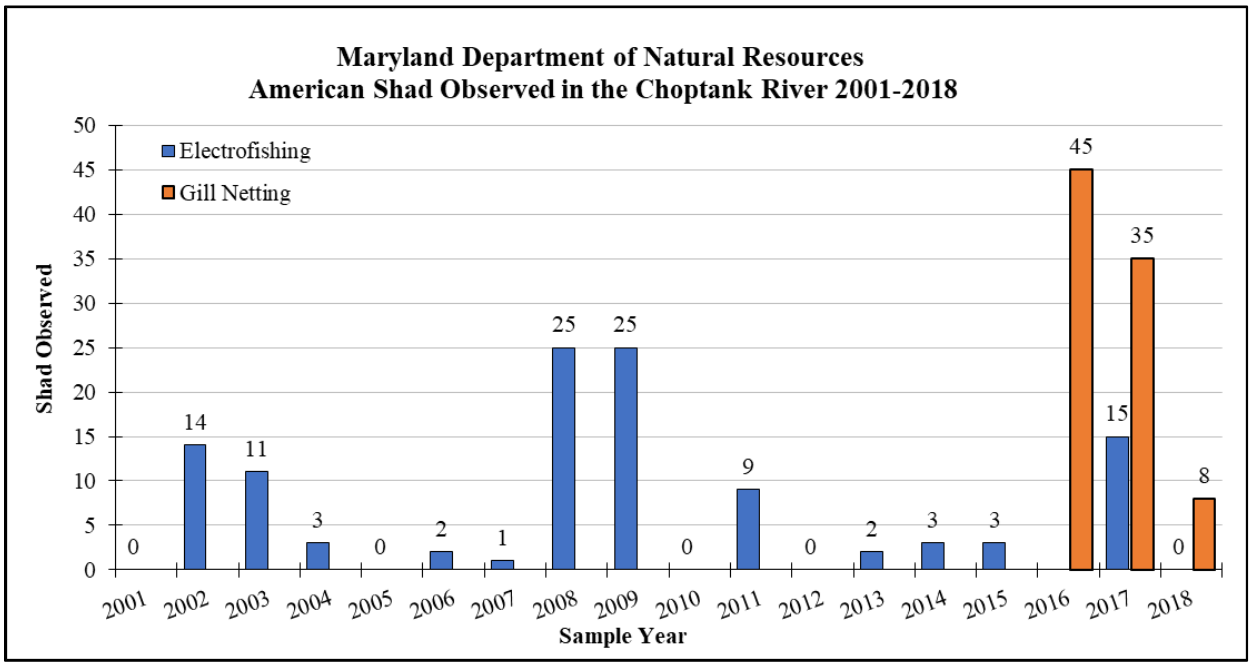

Figure 19. *Maryland Department of Natural Resources observations of adult American Shad in Choptank River 2001-2018. Gill netting for American Shad on the Choptank River began in 2016.*

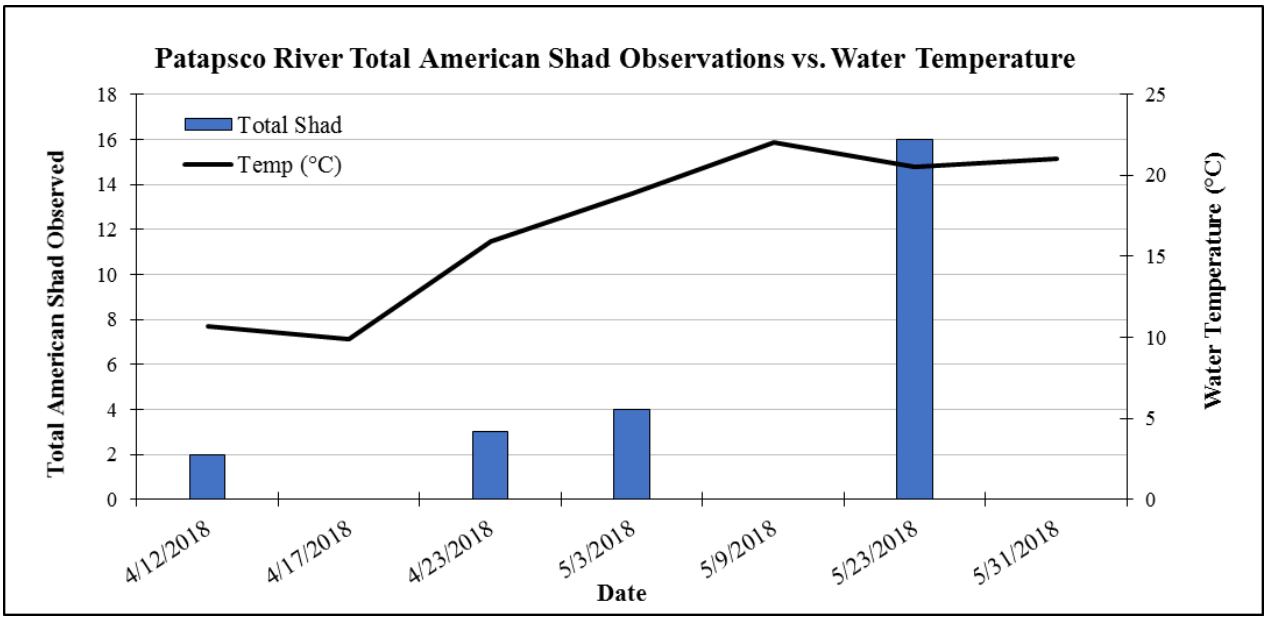

Figure 20. *2018 Maryland Department of Natural Resources electrofishing observations of adult American Shad on the Patapsco River.*