Population and DPS Origin of Subadult Atlantic Sturgeon in the Hudson River

Final Report Submitted to the Water Resources Institute
May 27, 2016

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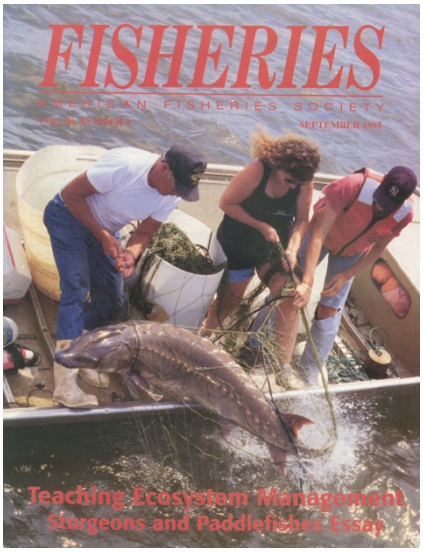

Abstract: At one time, Atlantic sturgeon supported a signature fishery in the Hudson River Estuary and identification of its migratory patterns is listed as a priority under Long Range Target 1 of the Actions Planned for 2010-2014 (Effectively Managing Migratory Fish). This study provided important new information that will be used by the NYSDEC and NOAA's Office of Protected Resources to manage Atlantic sturgeon in the Hudson River ecosystem and coastwide. Atlantic sturgeon is federally listed under the U.S. Endangered Species Act (ESA) as five Distinct Population Segments (DPS), of which four were designated as "endangered" and one as "threatened." The New York Bight DPS is comprised of the Hudson and Delaware River populations and is listed as "endangered." Subadult Atlantic sturgeon are known to exit their natal estuaries to coastal waters and non-natal estuaries where they are vulnerable to distant anthropogenic threats. In fact, during the warmer months, the Hudson River hosts large numbers of subadults, but their population and DPS origin is largely unknown although Section 7 of the ESA demands that origin of individual specimens be determined. We used microsatellite DNA analysis at 11 loci and sequence analysis of the mitochondrial DNA (mtDNA) control region to determine the DPS and population origin of 106 subadult Atlantic sturgeon collected in the lower tidal Hudson River estuary. We found that 101 of the 106 subadults assigned to the Hudson River with at least 95% and usually 100% probability. Of those 5 specimens that did not assign to the Hudson, 2 assigned to the James River, VA, 2 assigned to the Kennebec River, ME, and 1 assigned to the Saint John River, NB. Thus, four specimens assigned to DPS other than the New York Bight DPS and one to the Canadian Management Unit. This analysis will permit the quantification of the effects of anthropogenic threats in different locales or across seasons in the Hudson River Estuary on individual populations or DPS of Atlantic sturgeon and will serve as a model for similar population composition analysis for other estuaries coastwide.

Summary Points of Interest

A. Greater than 95% of subadult Atlantic sturgeon in the Hudson River are of Hudson River origin.
B. However, the Hudson River is host to a number of subadult Atlantic sturgeon that were spawned in other populations and sometimes other Distinct Population Segments.
C. The Hudson River harbors the population of Atlantic sturgeon with the largest Effective Population Size (N_e) coastwide.

Keywords

Microsatellite DNA analysis, mitochondrial DNA control region, Individual Based Assignment Testing, Mixed Stock Analysis, Distinct Population Segments

Background: Atlantic sturgeon *Acipenser oxyrinchus oxyrinchus* is the poster child for the Hudson River estuary with its image serving as the logo for the Hudson River prominently displayed on bridges crossing the main-stem river and its major tributaries. Historically, Atlantic sturgeon supported one of the three signature fisheries within the Hudson River Estuary. Spawning populations of Atlantic sturgeon extend from the St. Lawrence River, Quebec, to at least the Altamaha River, Georgia. Historically, there were close to 30 spawning populations coastwide (ASSRT 2007), but that number has dwindled in recent years to 15-20 rivers (Wirgin et al. 2015b). Atlantic sturgeon are anadromous and their spawning locations within natal rivers are above the salt front and usually over gravel, cobble, or boulder bottom. In the Hudson River, they are known to spawn from early June to early July in deep water in an area extending from Hyde Park to Catskill, New York and perhaps even further upriver. Their eggs are demersal and hatch within 4-6 days post-fertilization; the exact duration is temperature dependent. Juvenile Atlantic sturgeon are resident within their natal rivers for 2-6 years before migrating as subadults into coastal waters. Their duration of river residency is population dependent and is shorter in southern compared to northern rivers.

Subadult Atlantic sturgeon are highly migratory in coastal waters, the duration of migration can be prolonged, is population dependent, and utilizes unknown migratory corridors. Mature adult Atlantic sturgeon return to natal rivers to spawn, their age at maturity is once again highly variable and population dependent. For example, females in South Carolina spawn at 7-19 years (Smith et al. 1982), age 15 and older in the Hudson River (Bain, 1997), and at 27-28 years in the St. Lawrence River (Scott and Crossman 1973). In comparison, males spawn in the Suwanee River, Florida, at 7-9 years (Huff 1975), in the Hudson River at age 12 and older (Bain 1997), and 16-24 years in the St. Lawrence River (Caron et al. 2002). Post spawning adults exit their natal estuaries, weeks to months after spawning, and resume their coastal movements. The absence of adults from spawning rivers for many years and the difficulty in collecting early life-stages make censusing of populations and evaluation of temporal trends in their abundances problematic.

At varying times, many rivers coastwide, including the Hudson and particularly the proximal Delaware, hosted large fisheries for Atlantic sturgeon primarily targeting caviar-laden females. Many of these fisheries crashed, including that in the Hudson, in the late 1890s, to levels that were less than 10% of their historical highs. As fisheries in northern and mid-Atlantic rivers declined, the fisheries shifted to more southern rivers, particularly in South Carolina and Georgia, but these too suffered a similar fate as those in the Hudson and Delaware. By 1998 a federal coastwide 40-year harvest moratorium was imposed on the fisheries. This was followed in 2012 by U.S. federal listing of the species under the Endangered Species Act (ESA) as five Distinct Population Segments (DPS) of which four (New York Bight, Chesapeake Bay, Carolinas, and South Atlantic) were designated as “Endangered” and the fifth (Gulf of Maine) as “Threatened” (Federal Register 2012ab) As a result, federal management of the species under ESA is on a DPS basis, rather than as a single coastwide entity. However, because the abundance of the individual spawning populations is believed to vary by at least an order of magnitude, it is also important to consider the vulnerabilities of individual populations to the variety of anthropogenic stressors identified in the listing document. For example, the Hudson River population is considered to be the largest coastwide and the Delaware River population one of the smallest. Thus, the Delaware

River population is considered to be more vulnerable to extinction from anthropogenic stressors than the Hudson River population despite both of their listings within the New York Bight DPS.

As mentioned previously, subadult Atlantic sturgeon from all spawning populations migrate into coastal waters for extended durations. Besides coastal waters, it is known that subadults migrate seasonally into non-natal estuaries such as Long Island Sound, the Connecticut River (Waldman et al. 2013), and the inner Minas Basin of the Bay of Fundy (Wirgin et al. 2012). These may include estuaries that do not support spawning such as the Connecticut River-Long Island Sound and estuaries that do support contemporary spawning such as Delaware Bay and Chesapeake Bay. Conversely, subadults spawned in estuaries other than the Hudson are believed to seasonally move into the lower Hudson River Estuary. Because of their highly migratory behavior outside of their natal estuaries, specimens from multiple populations and DPS are likely to co-aggregate in coastal waters (Wirgin et al. 2015ab) or other estuaries distant from the river in which they were spawned (Waldman et al. 2013).

It is sometimes important to determine the DPS and population origin of individual specimens in these mixed coastal and estuarine aggregations because of their vulnerabilities to anthropogenic stressors at locales distant from their natal estuaries. For example, it has been documented that bycatch of Atlantic sturgeon in coastal fisheries targeted to other species may be an important contributor to the decline of some more vulnerable populations or their failure to rebuild (Wirgin et al. 2015b). Similarly, vessel strike mortalities of Atlantic sturgeon have been shown to frequently occur in the Delaware River (Brown and Murphy 2010) and James River (Balazik 2012) and have been proposed to be a significant factor in the decline of those populations. Migratory subadult Atlantic sturgeon may also be vulnerable to a variety of anthropogenic stressors in the Hudson River, including exposure to toxic chemicals such as PCBs and a perceived recent increased frequency of vessel strike mortalities. Thus, there is a need to identify the origin of individual sturgeon specimens to quantify the vulnerabilities of individual populations and DPS to stressors at locales distant from their natal estuaries.

Because they are highly migratory outside of their natal estuaries, determination of the abundance of subadults and adults of individual populations and DPS and tracking their movements in coastal waters and non-natal estuaries is problematic. Genetic analysis has proven to be an effective tool to identify the population and DPS origin of individual Atlantic sturgeon and their mixed aggregations. Briefly, the genotypes of fish of unknown origin are compared to those in reference collections from known spawning populations. The origin of individual specimens of unknown ancestry is then assigned to the reference collection whose genotypes best match those of the unknown individuals or their aggregations. In practice, this has involved using microsatellite DNA analysis at 11 independent loci and sequence analysis of the mitochondrial DNA (mtDNA) control region to characterize spawning adults and pre-migratory juveniles from reference spawning populations (Wirgin et al. 2012; Waldman et al. 2013; Wirgin et al. 2015ab). Collections of specimens of unknown origin are then characterized at the same 11 microsatellite loci and mtDNA sequence and compared to those in the reference collections. Using an approach termed, Individual Based Assignment (IBA) testing, the population and DPS origin of each individual specimen in a mixed aggregation can be assigned with determined probabilities of accuracy. A second approach, Mixed Stock Analysis (MSA), can be used to determine the proportion of

individuals in a mixed aggregation that assign to each reference population and DPS. In this and past studies, we have genetically characterized 1,3497 individuals from 11 reference spawning populations of Atlantic sturgeon at these 11 microsatellite loci and the mtDNA control region. This reference data allows us to determine the origin of subadult Atlantic sturgeon of unknown origin in the current study.

Our objectives in this study as described in our proposal were several fold:

1- Estimate the overall proportion of non-natal subadult Atlantic sturgeon within the lower tidal Hudson River estuary seasonally and identify their population and DPS of origin.
2- Define the overall spatial boundaries of the incursion of non-natal subadult Atlantic sturgeon within the tidal Hudson River estuary and their minimum and maximum length range.

Although not identified in the original proposal, we also felt that it was prudent to address two additional objectives with this data

1- Increase the number of samples in our reference Hudson River collection by adding additional years of juvenile and adult collections.
2- Identify effective population size (N_e) of Hudson River Atlantic sturgeon based on 3 years of juvenile collections.

Methods

Sample collections

In total, we were able to secure 106 subadult juvenile samples from the Hudson River that were collected between early June and mid-November. We targeted specimens that were >600 mm total length (TL) and <1300 mm (TL). Specimens were collected between 2009 and 2014, with the vast majority being collected in 2014. Additionally, almost all of the samples were collected by Normandeau Associates by gill nets with a smaller number coming from trawling. All samples were deposited by Normandeau in the tissue repository housed by the National Ocean Service in Charleston, SC. Unfortunately, a number of samples that Normandeau records showed were deposited with the NOS repository were never located decreasing the number of samples that could be analyzed in this study. Also, during this time (2015), the tissue repository was moved from Charleston, SC to the USGS facility in Leetown, WV which exacerbated the problem.

Additionally, 111 specimens were analyzed from three year-classes of juvenile Hudson River specimens (<500 mm TL) (2011 (n=30), 2013 (n=35), 2014 (n=46)) to bolster our Hudson River reference collection sample size. This would provide us with more confidence in our assignment testing and mixed stock analysis. These reference samples were obtained from the NYSDEC springtime collections from the Haverstraw Bay, NY area.

DNA Isolations

Fin clips were the source of DNA from all samples analyzed in this study. Fin clips were washed with phosphate-buffered saline, and incubated in cetyltrimethyl ammonium bromide (C-Tab) buffer (Saghai-Maroof et al. 1984) and digested at 65^{o} C with proteinase K (Roche Diagnostics, Indianapolis, IN). DNAs

were purified by phenol-chloroform extractions, alcohol precipitated, air dried and resuspended in TE buffer. Concentrations and purities of DNAs were evaluated using a Nanodrop ND-1000 Spectrophotometer (NanoDrop Technologies, Wilmington, DE). DNA concentrations were adjusted to 50 ng/μl for standardization of subsequent analyses.

Mitochondrial DNA Control Region Sequence Analysis

A 560 base pair (bp) portion of the mtDNA control region was amplified with derived Atlantic Sturgeon-specific primers S1 (5'- ACATTAAACTATTCTCTGGC- 3') and G1 (5'- GAATGATATACTGTTCTACC- 3') (Ong et al. 1996). The same primers were used to sequence a portion of the 560 bp amplicon. We report here data on only 205 bp of the amplicon to allow for comparison of haplotypes in subadult Hudson River specimens to previously characterized reference collections from other rivers (Wirgin et al. 2000; Wirgin et al. 2007; Peterson et al. 2008; Grunwald et al. 2008; Fritts et al. 2016).

Polymerase chain reactions (PCRs) were in 50 μl volumes that contained 50 ng of template DNA, 5 μl of 10 x Roche Applied Science (Indianapolis, IN) reaction buffer, 0.25 μl of each dNTP (25 mM stocks) (GE Healthcare, Piscataway, NJ), 0.07 μl of S1 primer (0.1 μM stock), 0.05 μl of G1 primer (0.1 μM stock) (Integrated DNA Technologies, Coralville, IA), 1 unit of Taq DNA Polymerase (Roche Applied Science) and 43.9 μl of H_20. Amplification conditions were 94^O C for 5 min followed by 40 cycles at 94^OC for 45 s, 56^O C for 45 s, 72^O C for 60 s, followed by a final extension at 72^O C for 10 min in MJ Research PTC-100™ thermal cyclers. Amplicons were purified with QIAquick PCR Purification kits (Qiagen, Valencia, CA).

Purified PCR products were Dye-Terminator Cycle Sequenced as recommended in GenomeLab Methods Development kits by the manufacturer (Beckman Coulter, Inc., Fullerton, CA). Sequencing conditions were 30 cycles at 96^o C for 20 s, 50^o C for 20 s, and 60^o C for 240 s. Sequencing products were EtOH precipitated, re-suspended in 40 μl of Beckman Coulter CEQ Sample Loading Buffer, loaded into a Beckman Coulter CEQ™ 8000 automated capillary-based DNA sequencer, run using the standard long fast read method (LFR-1), and analyzed with the Sequence Analysis Module of the CEQ™ 8000 Genetic Analysis System.

Microsatellite Analysis

Eleven microsatellite loci were scored that were previously shown to be effective in distinguishing reference specimens from spawning populations (King et al. 2001; Wirgin et al. 2015ab). These loci included LS19, LS39, LS54, LS68 (May et al. 1997), Aox23, AoxD45 (King et al. 2001), and Aox44, AoxD165, AoxD170, AoxD188, AoxD24 (Henderson-Arzapalo and King 2002).

Microsatellite genotypes were determined using the Beckman Coulter sequencer. Individual PCR reactions were multi-pooled, diluted up to 1:3 with Sample Loading Solution (Beckman Coulter), 0.5- 2.0 μl of reactions were loaded onto 96 well plates along with 0.5 μl of CEQ DNA Size Standard-400 and 40 μl of Sample Loading Solution (Beckman Coulter), and run with the FRAG 1 program (Beckman Coulter).

Statistical Analyses

Microsatellite data was initially examined using MicroChecker (Van Oosterhout et al. 2004) to identify the presence of null alleles, scoring errors, and/or large allele drop-out. Individual-based assignment (IBA) tests and mixed-stock analysis (MSA) were used to estimate the DPS and population origin of Atlantic Sturgeon in our collection of Hudson River subadults using the ONCOR program (Kalinowski et al. 2008). ONCOR used genetic data to estimate the population of origin of individuals by performing data analysis and simulations for mixture analysis and assignment tests. In mixture analysis, a reference baseline genetic data set was used to estimate the population composition of a mixed collection using conditional maximum likelihood to estimate mixture proportions. Individual-based assignment tests, using multi-locus likelihood functions, were used to assign individuals in a mixed collection to the reference collection that would have the highest probability of producing the given genotype in the mixed collection. ONCOR used the methods of Rannala and Mountain (1997) to estimate the probability. It should be noted that our analysis of a combination of diploid and haploid mtDNA data violates an assumption of this Monte Carlo resampling method. We estimated mixture proportions with 95% confidence limits based on 10,000 bootstraps. Results were reported for each population in the reference baseline collection as well as for each DPS.

Additionally, leave-one-out- tests were performed in ONCOR to evaluate how well individual specimens could be assigned to the DPS or population from which they were collected. In this test, each individual in each reference collection was sequentially removed from the baseline and its origin estimated using the rest of that reference collection. This test provides a quantitative measure of the accuracy of assignments to each reference or DPS collection.

NeEstimator (Do et al. 2014) was used to estimate effective population size (Ne) using a single-sample method—a bias-corrected version of the method based on linkage disequilibrium (Waples and Do, 2010).

Results

In total, we were able to obtain 106 samples from the NOS and USGS tissue repository for our analysis. Mean total length of the specimens was 990.6 mm (range 595 to 1720 mm). Collection sites in the Hudson River ranged from River Mile (RM) 7 to RM 77 with the vast majority of the specimens being taken between RM 48 and RM 49. Collection dates ranged between mid-June and mid-November with the vast majority of specimens taken in mid-June and early to mid-September. Similarly, most (73%) of the specimens were collected in 2014, but some dated back to as early as 2009.

We were successful in using a combination of microsatellite DNA and mtDNA control region sequence analyses to accurately assign DPS and population origin to all of the specimens using the ONCOR program with our reference data set. Our reference collections used to make these assignments consisted of 1,347 specimens from 11 spawning populations coastwide (Table 1). As indicated in Table 2, our assignment accuracy using leave-one-out tests was very high to the five individual DPS (and Canadian populations) and less so to the individual populations (Table 3). For example, we were 92.1% accurate in assignments at the DPS level and our mean accuracy in assignments at the population level

was 85.8%. Specifically, we were 90.3% accurate in assigning Hudson River collected specimens back to the Hudson with the vast majority of the misassignments going to the Delaware River (6.5%)

Using Mixed Stock Analysis in ONCOR, we initially determined the proportions of specimens from the 11 reference collections contributing to our collection of subadults of unknown origin from the Hudson River (Table 4). As expected, the vast majority of specimens were contributed by the Hudson River (95%) with smaller proportions noted from the Kennebec River (2%), James River (1.8%), and Saint John River (<1%).

Given the assignment accuracies described above, we felt confident in using these reference collections to assign our juvenile Hudson River collection to individual DPS and spawning populations. In total, 101 (95.3%) specimens assigned to the Hudson River, in most cases with 100% probability (see Appendix for data on each individual specimen). However, 9 of the 101 Hudson River-assigned specimens did so with less than 100% probability, but in all cases these specimens were assigned to the Hudson with $\geq$ 95% probability. Five specimens assigned to spawning populations other than the Hudson. These included 2 specimens that assigned to the James River, VA, 2 specimens that assigned to the Kennebec River, ME, and 1 specimen that assigned to the Saint John River, NB, Canada. Of these 5 non-Hudson assigned specimens, only one of the James River specimen assigned with 100% probability with the other 4 specimens' assignment probabilities ranging between 82.7% to 94.8%. Surprisingly, these 5 specimens assigned to populations in other than the New York Bight DPS, with 2 assigning to the Chesapeake Bay DPS, 2 assigning to the Gulf of Maine DPS, and one assigning to the Canadian management unit.

Given the new reference collection data from three years collection of Hudson River juveniles generated for this study, we calculated effective population size (*Ne*) estimates for the Hudson River and compared these to our other reference collections coastwide (Table 5). Not surprisingly, we found that the Hudson River had the largest *Ne* coastwide (217.4; 95% CI 156.8-337) followed by the Altamaha River, GA (138.7; 95% CI 103.5-201), and the Savannah River, SC-GA (138.1; 95% CI 109.3-182.7). Also, the Delaware River, the second population in the New York Bight DPS, had one of the smallest *Ne* (41.6; 95% CI 36.6-47.5).

Discussion

Subadult Atlantic sturgeon are known to migrate into coastal waters (Wirgin et al. 2015ab) and subsequently into non-natal estuaries, some of which host natural reproduction and others which do not (Waldman et al. 2013). During these seasonal forays, subadults may be exposed to a variety of anthropogenic stressors in these non-natal estuaries which may acutely jeopardize their survival or cause sublethal effects. Estuarine stressors that were identified in the U.S. federal listing documents (Federal Register 2012ab) included vessel strikes, bycatch, dredging, chemical pollution, compromised water quality, and other environmental perturbations. Many of these stressors to sturgeons are known to occur regularly in the tidal Hudson River estuary. Because Atlantic sturgeon are federally listed and managed as 5 DPS, it is important for Protected Resources managers to evaluate and quantify the potential effects of these stressors on representatives of the individual DPS and perhaps populations that may have migrated to non-natal estuaries (Damon-Randall et al. 2013). However, there was an

absence of empirical quantitative data to address the DPS and population origin of subadult Atlantic sturgeon in any non-natal estuary. Therefore, this study was designed to fill this void and determine the DPS and population origin of subadults in the tidal Hudson River estuary. Our overall hypothesis in this project was that all subadult Atlantic sturgeon in the Hudson River seasonally were spawned in the Hudson River. We tested this hypothesis using two DNA approaches, microsatellite and mtDNA analyses, that played a major role in the initial delineation of the 5 DPS and in their subsequent management by NOAA (Federal Register 2012ab).

This study was designed to optimize the likelihood of detecting non-natal specimens in the Hudson River by focusing our analysis on; 1) subadults that ranged in size from 600 to 1300 mm TL, 2) specimens that were collected in summer and fall after the completion of spawning, and 3) were sampled in the lower river where subadults from elsewhere were most likely to aggregate. Although, these were our goals, they were not met as stringently as we would have hoped. That is because our analysis was restricted to specimens that had been previously collected in programs designed for other objectives. Thus, we feel that may have underestimated the proportion of non-natal subadults that seasonally migrate into the Hudson River.

Our major finding was that the Hudson River estuary does seasonally host subadult Atlantic sturgeon that were spawned elsewhere. In fact, approximately 5% of our subadult specimens were spawned in other populations-in all cases not even within the New York Bight DPS. Of the five specimens not spawned in the Hudson River, 2 assigned to the Chesapeake Bay DPS (James River, VA), 2 assigned to the Gulf of Maine DPS (Kennebec River, ME), and one assigned to the Saint John River within the Canadian management unit. Thus, migrations of subadults from other DPS and populations may subject them to a number of stressors that are common in the Hudson River and may not be encountered in their natal rivers. We feel that our assignments of these subadults to other than the New York Bight DPS is accurate given the results of our leave-one-out tests. These tests demonstrated that across all reference collections coastwide, our mean assignment accuracy to the 5 DPS was 92.1% with very few misassignments of Hudson River specimens to the Gulf of Maine or Chesapeake Bay DPS. For example, only 2.2% of Hudson River collected reference specimens misassigned to the Gulf of Maine DPS.

One additional outgrowth of our study was our ability to estimate effective population size (N_e) of Atlantic sturgeon from the Hudson River as well as other populations coastwide. Although the ratio of N_e to census size for Atlantic sturgeon is unknown, it can provide a relative measure of the sizes of individual populations and overall trends in their abundances. Not surprisingly, N_e of the Hudson River population was by far the largest coastwide, far exceeding that of the Delaware River, the second population in the New York Bight DPS. Our N_e results are consistent with thoughts expressed in the most recent Atlantic sturgeon review (ASSRT 2007) in which the Hudson River population was viewed as the most robust coastwide. However, for the first time we provide a quantitative comparative index of the size of Atlantic sturgeon populations coastwide.

Policy Implications

Our results will inform NOAA managers for their Section 7 consultations to evaluate the likelihood of proposed projects to negatively impact Atlantic sturgeon from each of the 5 DPS. Prior to our study, there was an absence of empirical quantitative data on the movements of subadult Atlantic sturgeon to non-natal estuaries and the likelihood of their encountering stressors there. Our estimates of N_e also provided resource managers with the first relative measures of population abundance for each of these spawning populations coastwide, including the Hudson River.

Outreach Comments

Results from this study were recently presented by Dr. Wirgin on May 17, 2016, to USGS and NOAA Office of Protected Resources Managers at the *Atlantic and Shortnose Sturgeon Research and Management: Past, Present, and Future* workshop in Leetown, West Virginia. The title of his talk was: *Use of Individual Based Assignment Tests in the Coastwide Management of Atlantic Sturgeon* by Wirgin, I., D. Fox, T. Savoy, and M. Stokesbury.

Dr. Wirgin also plans to discuss his results and their implications for sturgeon management locally with Amanda Higgs, Robert Adams, and Greg Kinney of the NYSDEC Hudson River Fisheries Unit at their New Paltz, NY office.

As usual, Dr. Wirgin intends to publish these results in a peer reviewed journal as part of a larger manuscript on the use of DNA analysis in the management of Atlantic sturgeon.

Student Training

Ms. Melissa Della Torre, a spring 2016 graduate of the MS program in the Department of the Environmental Medicine of the NYU School of Medicine, participated in conducting research for this project. In it, she was trained in DNA isolations, PCR, mtDNA sequencing, microsatellite DNA analysis, and statistical analysis of population genetics data. She presented a poster on her studies of Atlantic and shortnose sturgeon biology at New York Marine Sciences Consortium annual meeting (Oct, 2015) at which she was awarded a prize for best graduate student poster.

Table 1

Locations where reference Atlantic sturgeon were collected, sample size (N), sampling date, and total lengths (mean TL)

Sampling Location	N	Sampling Date	Total Length Range (cm) (mean TL)	Maturity Status
St. Lawrence River	50	May-June 2014-5	All spawning adults	(A)
Saint. John River	66	July-Aug 1992	All spawning adults	(A)
		Aug 1993	127-244	(A)
	161	May-Aug 2014	162.6-248.9 (199.7)	(A)
Kennebec River	43	June 2010-Aug 2011	133-197.4 (171.6)	(A)
Hudson River	67	May-June 1993-4	160.8-244 (189.3)	(A)
	50	June 2010	170-198 (198)	(A)
	30	March-April 2011	43.2-54 (49.8)	(J)
	35	March-April 2013	41.1-52.8 (46.2)	(J)
	46	April-May 2014	28.7-48.9 (43.9	(J)
Delaware River	33	Sept-Nov 2009	22.0-34.9 (28.4)	(J)
	26	Sept-Nov 2009	22.3-36.7 (30.4)	(J)
	49	Sept-Nov 2011	23.5-36.3 (28.9)	(J)
James River	58	Unknown	26.0-49.5 (45.7)	(J)
	58	July-Sept 2014	All spawning adults	(A)
Albemarle Sound	41	May-Sept 1998	28.6-48.5 (38.8)	(J)
	31	Dec 2006-Jan 2011	27.0-49.9 (40.3)	(J)
	16	Jan 2013-Mar 2014	31.5-49.4 (43.0)	(J)
Edisto River	53	April-Oct 1996	27.7-50 (39.9)	(J)
	52	May-Sept 2005	32.6-48.5 (42.4)	(J)
Savannah River	50	May-June 2013	31.6-44.7 (39.1)	(J)
	50	May-2014	27.4-47.9 (37.0)	(J)
Ogeechee River	26	June 2007-Aug. 2009	19.9-52.0 (28.6)	(J)
	45	July-Aug. 2014	22.7-31.0 (26.0)	(J)
	67	May-July 2015	16.6-44.2 (33.2)	(J)
Altamaha River	49	June-July 2005	31.9-40.4 (37.9)	(J)
	40	July-Aug. 2011	32.7-48.1 (38.6)	(J)
	55	May-June 2014	28.1-48.7 (37.2)	(J)
TOTAL	1,347			

Table 2

Proportion of Reference Individuals Correctly Assigned to the DPS from which They Collected

DPS	N	% Correctly Assigned	Largest Misassignment	DPS
Canada	272	97.4%	1.5%	GOM
GOM	41	83.0%	7.3%	Canada
NYB	323	95.7%	2.2%	GOM
CB	113	91.2%	3.3%	SA
CAR	82	87.8%	11.0%	SA
SA	464	97.0%	2.6	CAR
Mean		92.1%		

Table 3

Proportion of Reference Individuals Correctly Assigned to the Population from which They Were Collected

Population	N	% Correctly Assigned	Largest Misassignment	Population
St. Lawrence	49	95.9%	2.0%	Saint John
Saint John	223	97.8%	1.3%	Kennebec
Kennebec	41	85.4%	4.9%	Saint John
Hudson	216	90.3%	6.5%	Delaware
Delaware	107	86.0%	13.1%	Hudson
James	113	90.3%	2.7%	Altamaha
Albemarle Sound	82	85.4%	4.9%	Altamaha
Edisto	94	91.5%	4.3%	Altamaha
Savannah	99	71.7%	15.2	Altamaha
Ogeechee	135	71.1%	13.3%	Altamaha
Altamaha	136	78.7%	12.5%	Savannah
		X=85.8%		

Table 4

Mixed stock analysis of reference population proportions of subadult Atlantic sturgeon in the Hudson River

Population Estimates	**%**	**95% Confidence Intervals**
St. Lawrence	0.000	(0.000, 0.013)
Saint John	0.008	(0.000, 0.030)
Kennebec	0.020	(0.000, 0.075)
Hudson	0.954	(0.880, 0.991)
Delaware	0.000	(0.000, 0.025)
James	0.018	(0.000, 0.047)
Albemarle	0.000	(0.000, 0.020)
Edisto	0.000	(0.000, 0.000)
Savannah	0.000	(0.000, 0.019)
Ogeechee	0.000	(0.000, 0.000)
Altamaha	0.000	(0.000, 0.000)

Table 5

N_e estimates based on 2 or 3 years (except Kennebec) of collections combined using the linkage disequilibrium method

Population	N_e	95% CI	Rank
Saint John*	51.5	46.6-57.1	6
Kennebec	53.0	40.5-73.2	4
Hudson*	217.4	156.8-337	1
Delaware	41.6	36.6-47.5	8
James	45.5	41.1-50.5	7
Albemarle*	21.2	19.1-23.6	10
Edisto	52.8	45.1-62.4	5
Savannah	138.1	109.3-182.7	3
Ogeechee	34	29.7-39.1	9
Altamaha	138.7	103.5-201	2

*based on three years of collections

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