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TECHNICAL NOTES

Induced Spermiation in Wild Atlantic Sturgeons Held Captive Up to Six Years

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Abstract.—The ability to obtain gametes from male Atlantic sturgeons *Acipenser oxyrinchus* brought into captivity may be important to the success of future restoration efforts for this species. Atlantic sturgeons obtained as juveniles from the Delaware River and as sexually mature males from the Hudson River were held in freshwater at the Northeast Fishery Center, Lamar, Pennsylvania, from 1991 to 1997. After 4 years in captivity, three of three Hudson River fish were induced to spermiate by injection of common carp pituitary solution (CCP), luteinizing hormone-releasing hormone analogue (LHRHa), or a combination of both. Hatch rates were similar for eggs fertilized with a captive male's milt (3.1%) versus a wild male's milt (mean ± SD, 3.1 ± 0.5%). At 58 d posthatch, survival of the captive male's offspring (74.3%) was comparable to that of wild male's (61.9 ± 7.8%). After 6 years in captivity, two of five fish from the Delaware River were induced to spermiate by injection of CCP, and nearly all spermatozoa exhibited rapid forward motion. Results showed that feral male Atlantic sturgeons can be held in freshwater over a period of years for use as captive broodstock.

Once plentiful on the eastern coast of North America, populations of Atlantic sturgeon *Acipenser oxyrinchus oxyrinchus* have undergone severe declines (Murawski and Pacheco 1977). Currently, the Hudson River and estuary still remain one of the major spawning and nursery grounds for this species (Van Eenennaam et al. 1996). In the Delaware River, subadult Atlantic sturgeons have recently been captured and tagged, but numbers taken and catch per unit effort declined during a 1991–1997 study (C. Shirey, Delaware Division of Fish and Wildlife, personal communication). Waldman et al. (1996) indicated that the annual occurrence of these individuals largely reflects that the Delaware River estuary is one of a series of estuaries seasonally inhabited by subadult Atlantic sturgeons. In response to the decline of Atlantic sturgeon populations, the Northeast Fishery Center (NEFC), a U.S. Fish and Wildlife Service Technology Center, initiated studies involving capture, transport, spawning, and culture of Atlantic sturgeons. The ability to obtain gametes in subsequent years from individuals brought into captivity in various stages of maturity is an important step towards synchronizing spawning times of both sexes. In this study, we document hormone-induced spermiation in feral male Atlantic sturgeons after long-term captivity (4–6 years). Published reports of induced spermiation in captive sturgeons include the following species: white sturgeon *A. transmontanus* (Doroshov et al. 1983), shortnose sturgeon *A. brevirostrum* (Smith et al. 1985), and Gulf sturgeon *A. oxyrinchus desotoi* (Parauka et al. 1991); however, fish were held for only 7 months or less. Unpublished sources indicated that induced spermiation was achieved in pallid sturgeon *Scaphirhynchus albus* and shovelnose sturgeon *S. platorynchus* held in captivity for 2 years or more (H. Bollig, U.S. Fish and Wildlife Service, personal communication) and in shortnose sturgeon (K. Ware, U.S. Fish and Wildlife Service, personal communication). Hormone-induced spermiation and successful use of milt from Atlantic sturgeons held in short-term captivity (13 d) was reported by Smith et al. (1980), but documented evidence of longer periods of captivity and subsequent induced spermiation for this species was not found.

In this study, juveniles from the Delaware River and sexually mature males from the Hudson River were captured in gill nets and transported to NEFC in 1991. The captured sturgeons were maintained in 6-m-diameter circular fiberglass tanks located in NEFC's intensive culture building. Constant flow-through water was supplied to the fish via underground piping from a reservoir maintained with water from nearby Fishing Creek. Culture tank water temperatures were ambient and ranged annually from less than 1°C to 19°C. Water flows were maintained at about 100 L/min in a circular pattern. Daily illumination was not measured but consisted of varying combinations of ambient light, supplemented with incandescent lighting

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from 0800 to 1600 hours. Over the study period (1991–1997), sturgeons were given daily prophylactic salt treatments (salinity was <1‰) using either 22.6-kg blocks or bags of loose, noniodized salt. However, to treat outbreaks of fungus, which typically occurred a few days after capture, additional salt was added to create a daily salinity peak of about 10‰. In addition, salt treatments for fungal infection were accompanied by daily flush-type formalin treatments of 150 mg/L for as many as seven consecutive days. Under these conditions, the sturgeons were not exposed to the prolonged periods of salinity normally experienced in the natural ocean migrations of the species.

Hudson River mature males.—In 1991, four sexually mature males flowing with milt were captured in gill nets during spawning season (June) at Catskill, New York, and transported to NEFC. All captives were offered a variety of fresh, live, frozen, and formulated feeds, but feeding behavior was minimal or not observed for a number of months. Eventually, all fish began to eat trout pellets (GR7–30, 6.3-mm diameter; Zeigler Brothers, Inc., Gardners, Pennsylvania). Once feeding commenced, Atlantic sturgeons were offered pellets at a rate of 0.5% body weight daily by the manual broadcast of feed and the use of 12-h-duration automatic belt feeders (Zeigler Brothers, Inc.). Through the study period (1991–1997), three out of four animals survived. Only one of the individuals began to gain weight in 1991; the remaining two lost weight during the first 2 years of captivity. By 1995, condition factors of all three individuals were judged to be favorable for experimental injection of spawning hormones. Underwater injections were administered into the dorsal musculature on June 17 and 18, 1995, using either a 1-cm^3 or 5-cm^3 syringe outfitted with a 20-gauge, 3.8-cm needle. Hormones selected for use were common carp pituitary (CCP; Argent Chemical Laboratories, Redmond, Washington) and luteinizing hormone-releasing hormone analogue (LHRHa; Sigma Chemical Co., St. Louis, Missouri). One fish (HDW-021) was given a primer dose of LHRHa 12 h before the final CCP dose; the other two fish (HDW-024 and HDW-023) were given only one injection of either LHRHa or CCP (Table 1).

Twenty-four hours after injection (June 19, 1995), swimming activity increased. Each individual was captured and placed on a stretcher for examination. Milt was extracted from the genital opening with a 60-mL syringe outfitted with a short length of clear, flexible plastic tubing. Milt was obtained from all three males at a mean volume (±SD) of 247 ± 81 mL (Table 1). Some milt was expelled into the water during handling. Sperm motility was estimated by viewing water-activated milt under a microscope with 100× magnification. All milt samples were judged to have sperm motility with a rapid forward motion value of 4 points, as described by Persov (1941). Milt from sturgeon HDW-021 was used to fertilize one-fourth (16,960) of the eggs taken from a Hudson River female, which resulted in a hatch rate of about 3.1%. Similar hatch rates (3.0, 2.6, and 3.8%) occurred in the eggs that were fertilized with milt from three freshly caught (wild) Hudson River males (3.1 ± 0.5%). Milt from wild males was transported from the Hudson River to NEFC in oxygenated plastic bags and kept cool with crushed ice in an insulated cooler for 2 d. Motility of sperm from wild males was comparable to that of HDW-021. Offspring of each male were reared in separate 60-L circular tanks (one tank per family) at densities ranging from 6.6 to 8.8 fish/L; HDW-021 progeny were at the highest density. At 58 d posthatch, survival of captive male progeny was higher (74.3%) than those of wild males (67.6, 50.8, and 67.3%). Measurement of 20 individuals from each family showed mean total length of HDW-021 progeny was less (57.5 ± 9.7 mm) than the progeny of wild sturgeons (60.3 ± 8.2, 65.8 ± 9.4, and 66.6 ± 10.7). Average weight of HDW-021 progeny was also less than those of wild males (0.8 versus 1.02, 1.38, and 1.18 g), as determined by the collective weight of 20 offspring from each family.

TABLE 1.—Hormone treatments and milt yield (Jun 19) of male Hudson River Atlantic sturgeons after 4 years of captivity at the Northeast Fishery Center in 1995. Hormones used were common carp pituitary (CCP) and luteinizing hormone-releasing hormone analogue (LHRHa).

Fish identification	Hormone	Dosage (mg/kg)	Injection schedule		Milt yield (mL)
			Time (hours)	Date	
HDW-024	CCP	1.0	0930	Jun 18	320
HDW-021	LHRHa	0.01	1600	Jun 17	260
	CCP	1.0	0930	Jun 18	
HDW-023	LHRHa	0.03	0930	Jun 18	160

Delaware River juveniles.—Sixteen juvenile Atlantic sturgeons captured in gill nets near Artificial Island in the lower Delaware River in 1991 were taken into captivity at NEFC. Mean total length and weight at capture were 101 ± 10 cm and 5.3 ± 1.6 kg, respectively. Based on length data from Murawski and Pacheco (1977) and Dovel and

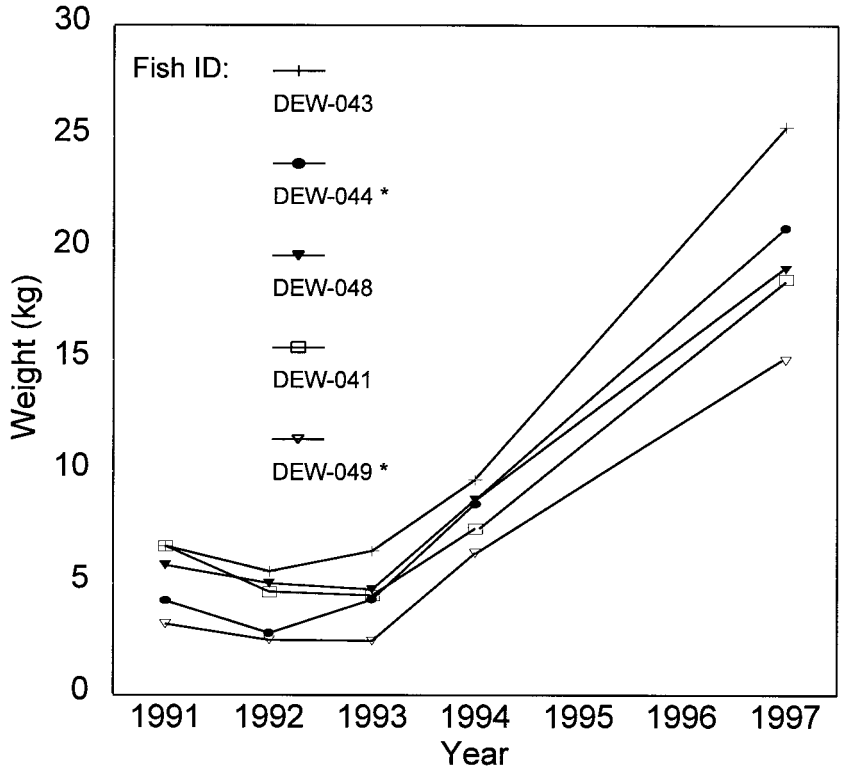

FIGURE 1.—Weight changes in Atlantic sturgeons captured as juveniles in the Delaware River and held in captivity at the Northeast Fishery Center from 1991 to 1997. Asterisks denote fish that produced milt after hormone injection.

Berggren (1983), the age of these fish upon capture was estimated to be 7 or 8 years; however, they could have been as young as 3 years based on recovery of tagged juveniles of known ages (C. Shirey, Delaware Division of Fish and Wildlife, personal communication). Although a wide variety of fresh, live, frozen, and formulated feeds were offered, initial acceptance of food items was poor. Eventually most fish began feeding regularly on the trout feed offered at a rate of 0.5% of body weight daily. Through the study period (1991–1997), there were five survivors, and some weight gains became evident in 1993 (Figure 1). Mean length increased from 101 ± 10 cm to 150 ± 11 cm over the study period, and mean weight increased from 5.3 ± 1.6 kg to 19.8 ± 3.8 kg.

On July 11, 1996, gonadatropins were injected to determine if the males were sexually mature. Fish were captured and restrained in a net, and CCP was injected into the dorsal musculature of all five captives at a rate of 1 mg/kg of body weight. Twenty-four hours later, each sturgeon was netted and placed on a stretcher for examination. No milt was collected from any of the five sturgeons. The following year, on June 19, 1997, the same fish were administered CCP injections with dosage rates equivalent to those of the previous year. Milt was collected after 24 h from two of five individuals (DEW-044 and DEW-049; Figure 1). Milt volumes were not measured, but samples were judged to have a good sperm motility value (4 points, as described above). Ages of the captive Delaware River Atlantic sturgeons at the time of milt expression was about 9–14 years, which was calculated by adding 6 years of captivity to the ages estimated upon capture in 1991. Gender or degree of gonadal development in the three individuals that did not produce milt was not determined.

Previous to this study, male Atlantic sturgeons have been induced to spermiate during short-term captivity (13 d) under the influence of exogenous hormones (Smith et al. 1980). However, this study demonstrated that both sexually mature and immature feral male Atlantic sturgeons reared in captivity for a number of years can be induced to spermiate by injection of gonadotropins. Percent hatch was low (3.1) for eggs fertilized by milt from

the captive Hudson River sturgeon (HDW-021), but it was similar to the hatch rates obtained using milt from wild males (3.0, 2.6, and 3.8%). The authors suggest that these low hatch rates were due to poor egg quality because the ovaries of the female used for spawning were only partially ovulated. At 58 d posthatch, progeny of HDW-021 had higher survival than progeny of wild males but were smaller, most likely due to higher rearing density. It is of interest to note that Atlantic sturgeons described in this study were induced to spermiate even though they were held continuously in salinities less than 1‰.

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