NATIONAL MARINE FISHERIES SERVICE
ENDANGERED SPECIES ACT SECTION 7
BIOLOGICAL OPINION

Title: Biological Opinion on the Issuance of Permit No. 14604 for Research on Shortnose Sturgeon in the Delaware River

Action Agency: Permits, Conservation and Education Division, Office of Protected Resources, National Marine Fisheries Service

Consultation Conducted By: Endangered Species Division, Office of Protected Resources, National Marine Fisheries Services

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NOAA's National Marine Fisheries Service
Endangered Species Act Section 7 Consultation

Biological Opinion

Agency: Permits, Conservation and Education Division of the Office of Protected Resources, NOAA's National Marine Fisheries Service

Activity Considered: Biological Opinion on the Permits, Conservation and Education Division's proposal to issue a Permit (Number 14604) to Harold Brundage, Environmental Research and Consulting, Inc., for research on shortnose sturgeon in the Delaware River pursuant to section 10(a)(1)(A) of the Endangered Species Act of 1973.

Consultation Conducted by: Endangered Species Division of the Office of Protected Resources, NOAA's National Marine Fisheries Service

Approved by: _____

Date: APR 13 2010

Section 7(a)(2) of the Endangered Species Act (ESA) (16 U.S.C. 1531 *et seq.*) requires that each federal agency shall ensure that any action authorized, funded, or carried out by such agency is not likely to jeopardize the continued existence of any endangered or threatened species or result in the destruction or adverse modification of critical habitat of such species. When the action of a federal agency "may affect" a listed species or critical habitat that has been designated for them, that agency is required to consult with either NOAA's National Marine Fisheries Service (NMFS) or the U.S. Fish and Wildlife Service (USFWS), depending upon the listed resources that may be affected. For the action described in this document, the action agency is NMFS' Office of Protected Resources – Permits, Conservation and Education Division. The consulting agency is NMFS' Office of Protected Resources – Endangered Species Division.

This document represents NMFS' biological opinion (Opinion) on the effects of the proposed studies on endangered and threatened species and designated critical habitat, and has been prepared in accordance with section 7 of the ESA. This Opinion is based on our review of the Permits, Conservation and Education Division's draft Environmental Assessment, draft amendment to Permit Number 14604, the most current shortnose sturgeon stock assessment reports, recovery plan, scientific and technical reports from government agencies and the peer-reviewed literature, biological opinions on similar research, and other sources of information.

A complete administrative record for this consultation is on file at NMFS' Office of Protected Resources (OPR).

CONSULTATION HISTORY

The Office of Protected Resources – Permits, Conservation and Education Division (PR1) has previously authorized three permits to Harold Brundage, Environmental Research and Consulting, Inc. (ERC) for scientific research on shortnose sturgeon prior to the instant proposed research permit. These are permits 374, 1174, and 1486. The Office of Protected Resources – Endangered Species Division (PR3) produced Opinions for each of those permits concluding that their authorization was not likely to jeopardize the continued existence of shortnose sturgeon.

In December 2009, PR1 began discussions with PR3 regarding the proposed methodology under the instant permit 14604, namely anesthesia methodology. PR1 and PR3 conducted a series of phone calls to a number of shortnose sturgeon biologists to discuss the proposed anesthesia techniques under the 14604 permit application.

On December 30, 2009, PR3 received a consultation request from PR1 on permit 14604, which incorporated all of the above discussions. Since PR1 and PR3 had participated together in these discussions, no additional information was requested of PR1 beyond what was in the initiation package. On January 11, 2010, PR3 initiated consultation on permit 14604.

BIOLOGICAL OPINION

I. DESCRIPTION OF THE PROPOSED ACTION

The proposed action addressed in this Opinion is PR1's authorization of permit 14604 to ERC. The authority for PR1's permit issuance is section 10(a)(1)(A) of the Endangered Species Act of 1973, as amended (ESA; 16 U.S.C. 1531 *et seq*.). The proposed activities involve purposeful harassment, harm, wounding, trapping, capture, or collection ("take[1]") of endangered shortnose sturgeon (*Acipenser brevirostrum*) for scientific purposes.

The activities proposed under permit 14604 are to characterize habitat use, relative abundance, reproduction, juvenile recruitment, temporal and spatial distributions, and reproductive health of the shortnose sturgeon population in the Delaware River and Estuary. The proposed permit requests annual authorization for non-lethal sampling methods on up to 1,000 adult and juvenile shortnose sturgeon (this is on a per year basis, with no more than 3,600 fish sampled over five years). Research activities would include: capturing via gill net, trammel net, and trawl net; measuring and weighing; tagging with PIT and Floy T-bar tags; and sampling tissue for genetic analysis. A subset of 30 adults and 30 juveniles would be anesthetized tagged with acoustic transmitters. Another subset of 24 adults would be examined internally using laparoscopic techniques and each would potentially also have gonad biopsy and blood sample taken for analysis. Another subset of 20 adults per year would be included in hydroacoustic gear testing. Additionally, lethal collection of up to 300 eggs and larvae each year (not to exceed 900 over

[1] The ESA defines "take" as "to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect, or to attempt to engage in any such conduct." The term "harm" is further defined by regulations (50 CFR §222.102) as "an act which actually kills or injures fish or wildlife. Such an act may include significant habitat modification or degradation which actually kills or injures fish or wildlife by significantly impairing essential behavioral patterns including breeding, spawning, rearing, migrating, feeding, or sheltering."

five years) would take place during seasonal spawning activity by artificial substrate, D-frame ichthyoplankton net, and/or epibenthic sled. Finally, one unintentional mortality or serious harm event resulting from research is requested annually, not to exceed three over the life of the permit. The proposed take is summarized in Table 1 as follows.

Table 1. Activities proposed to be authorized for ERC's research on endangered shortnose sturgeon (*Acipenser brevirostrum*) in the Delaware River under Permit 14604.

Species	**Life Stage**	**Sex**	**Expected Annual Take**	**Take Action**	**Location**
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile & adult	Male & female	Up to 896 annually; total of 3,600 over 5yrs	Capture, hold, measure, weigh, photograph, scan (for tags), Floy T-bar tag, PIT tag, & tissue sample	Delaware River (netting area=rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 30 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222) & implant acoustic tag	Delaware River (netting area=rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile	Male & female	Up to 30 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222) & implant acoustic tag,	Delaware River (netting area=rkm 79-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 24 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222), laparoscopically evaluate (coelomic cavity, collect blood, & collect biopsy of gonads (if sex unclear)	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 20 annually	Capture, hold, measure, weigh, photograph, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, & hydroacoustic testing (tethered in nylon sock)	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)

Species	Life Stage	Sex	Expected Annual Take	Take Action	Location
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile & adult	Male & female	1 unintentional mortality or serious harm* annually, not to exceed 3 over the life of the permit	Unintentional mortality, storage, measure, weigh, photograph, fin clip, freeze, transport arrangements made with NMFS for further sampling and disposal	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	eggs & larvae	Male & female	300 annually (lethal take), not to exceed 900 over the life of the permit	Intentional (directed mortality)	Delaware River (netting area=rkm 215-245)

* This permit would allow for unintentional mortality or serious injury caused by the presence or actions of researchers. A serious injury is considered any injury that will likely result in mortality. This includes, but is not limited to: deaths or injuries resulting from infections related to sampling procedures; and deaths or injuries sustained by animals during capture and handling, or while attempting to avoid researchers or escape capture.

Capture by Gill and Trammel Net. Gill nets of 12.7 cm (5-inch) or 15.2 cm (6-inch) stretched mesh would be used to sample for adult shortnose sturgeon. Gill nets of 2.5 cm (1-inch) to 10.2 cm (4-inch) stretched mesh would be used to sample for juveniles. Gill nets would typically be 100 m in length and 1.8 m deep, although shorter nets may sometimes be used. Trammel nets 50 m in length and 1.8 m deep, consisting of two outer panels of 60.8-cm (24-inch) stretched multifilament nylon mesh and an inner panel of 2.5-cm (1-inch) stretched multifilament nylon mesh, may also be used to sample for juveniles.

The proposed net-set protocol is summarized in Table 2. Gill nets and trammel nets would be set in waters having minimum dissolved oxygen (D.O.) concentrations no less than 5.0 mg/L, with one exception (i.e., net-set duration would be reduced to the next lower duration when D.O. measured between 4 and 5 mg/L). Netting would cease in waters above 28ºC until consulting with the National Marine Fisheries Service - Office of Protected Resources (NMFS-PR). The maximum net set duration is 14 hours, but is limited to between river kilometer (rkm) 186 and 215 when temperature is 15 ºC or below and D.O. is 5 mg/L or greater. Outside of this range, nets could be set for 10 hours in daylight hours only.

Table 2. Summary of Netting Conditions

Water Temperature (ºC)	Minimum D.O. Level (mg/L)[1]	Maximum Net Set Duration (hr)
< 15	5	14[2]
< 15	5	10
15 < 25	5	4
25 < 28	5	2
>28		Cease netting until consulting with NMFS

1 If DO concentration is between 4 and 5 mg/L at any temperature range, netting may occur, but only at the next lower net set duration indicated.

2 Net set duration for 14 hours is limited to the river range of rkm 186 to 215 and would be subject to temperature and dissolved oxygen levels indicated.

Gill and trammel netting to mark and recapture adult shortnose sturgeon for population estimation would be performed in two distinct phases each year of the study. Overwintering shortnose sturgeon would be sampled between November and March in the tidal Delaware River between Roebling and Trenton, New Jersey (approximately rkm 186-215). Dispersed-phase sampling would be performed between May and October throughout the tidal Delaware River from Artificial Island to Trenton (rkm 79-215). It is anticipated the overwintering areas would be sampled most intensively to develop population estimates using the Chapman modification of the Schnabel method (Ricker 1975) which assumes a closed population due to the confined overwintering area of shortnose sturgeon in the Delaware River.

Gill/trammel and trawl netting for juvenile shortnose sturgeon would be conducted within the tidal Delaware River from Artificial Island, NJ to Trenton (approximately rkm 79-215). Sampling for juveniles would be conducted year-round, subject to water temperature limitations given in Table 2 above.

Capture by Trawl. Trawling would be performed year round expressly to sample juvenile shortnose sturgeon, subject to the netting environmental conditions in Table 2. Dovel and Berggren (1983) found small trawls effective while collecting juvenile shortnose sturgeon in the Hudson River. Trawling for juvenile shortnose sturgeon would similarly be performed in the tidal Delaware River from Artificial Island to Trenton (rkm 79-215) using a 4.9 m otter trawl and/or a 14.6 m yankee trawl. Specifications for these trawls are provided in Table 3.

Table 3. Description of Proposed Trawling Gear

	4.9 m Otter Trawl	14.6 m Yankee Trawl
Headrope (m)	5.2	14.6
Footrope (m)	6.4	21
Net body mesh (mm)	38	50 and 80
Codend mesh (mm)	32	50
Innerliner mesh (mm)	13	14

Trawls would be towed at a maximum speed of approximately 2.5 knots (5 mph) for 10 to 15 minutes with an outboard powered boat. To lessen benthic disturbances, a global positioning system (GPS) would be used to direct trawls so that nets would not be towed over the same exact

location more than once in a 24-hour period. Further, trawling would be conducted primarily over sand substrates avoiding hard bottoms, vegetated areas, organic material, or woody debris. If a trawl became snagged on bottom substrate and debris, it would be untangled immediately to reduce stress on captured animals.

General Handling (e.g., holding, measuring, weighing). Once captured, sturgeon would be held temporary in boat-side net pens measuring approximately 200 cm long x 150 cm wide x 200 cm deep. Additional net pens would be onboard to accommodate excess holding of sturgeon and/or bycatch. Handling of fish would be kept to a minimum and fish would not be held for more than two hours after removed from capture gear, typically less than 30 minutes. Once recovered, sturgeon would be transferred to an onboard holding tank, they would be weighed, measured, fin clipped, tagged, photographed, and further processed. To minimize handling, sturgeon would be moved and handled by researchers using latex gloves and, when in onboard holding tanks, sturgeon would be immersed in a continuous stream of water supplied by a pump-hose assembly mounted over the side of the research vessel. Dissolved oxygen (D.O.) would be supplemented with compressed oxygen to ensure the D.O. concentration does not fall below saturation. Sturgeon would be weighed on a platform scale fitted with a small waterproof cushion attached to the surface of weighing platform. Total length of each sturgeon would be measured with a standard measuring board, and, by using calipers, mouth width and interorbital width would be measured to confirm species (Moser *et al*. 2000).

The time required to complete routine, non-invasive methods (*i.e.,* PIT tagging, measuring, weighing) would be less than one minute per fish. The cumulative time required for procedures such as anesthesia induction, telemetry tagging, and genetic tissue sampling would vary, but would typically average less than 15 to 20 minutes per fish, not accounting for recovery time from anesthesia. During processing, all fish would be treated with a slime coat restorative in the onboard live well, and, if anesthetized or otherwise necessary, placed in a separate net pen to ensure full recovery prior to release.

Hydroacoustic Testing. Hydroacoustic/sonar would remotely scan and identify sturgeon while still in nets. The additional time required for this scanning would conform to netting summary of conditions in Table 2. Additionally, up to 20 shortnose sturgeon would be captured and tethered at different depths in soft nylon or cotton mesh sleeves for periods not exceeding two hours. The objectives of this investigation would be to: 1) determine if adult shortnose sturgeon can readily be detected with hydroacoustic/sonar systems under varying field conditions; 2) determine how close to the bottom the species can be resolved; and 3) determine the efficiency and characteristics of the technology which would best enable sturgeon to be remotely identified and enumerated in a mixed-species environment.

Methods proposed would follow those of Brundage and Jung (2009) and Nealson and Brundage (2007) where shortnose sturgeon and three other fish species for hydroacoustic data collection were captured using anchored bottom-set gill nets fished parallel to the current at navigation channel depth (approximately 10–12 m). The nets would be 100 m long by 1.8 m deep and consisted of either 12.7-cm or 15.2-cm stretched monofilament mesh. Hydroacoustic measurements would be collected by passing over the netted fish with a downward looking broadband transducer. Following acoustic data collection, the netted fish would be recovered,

identified, and measured for total length. Acoustic data would be collected using a SciFish 2100 broadband sonar system. Data would typically be collected over a frequency range of 110-220 kHz, using a pulse length of 1 meter and an acoustic pulse repetition rate of 3 pings per second. However, the current proposal also includes collection of data at a ping rate higher than that used in previous investigations, using both broadband and narrowband sonar at ping rates of over 30 pings per second alternating between broadband single-beam and narrowband split-beam signals. Narrowband split-beam processing allows locating a target with an accurate bearing angle with the broadband spectrum capable of being adjusted according to transducer sensitivity and the beam plot across the band. For that reason, it is also proposed to collect data from fish tethered in a specially designed frame (or sock) where the aspect angles could be controlled allowing better detail and specification, (Jung *et al.* 2004).

Genetic Tissue Sample. Genetic information would be obtained from tissue samples of sturgeon to help characterize the genetic "uniqueness" of the Delaware River population and would also help quantify the current level of genetic diversity within the population. Immediately prior to release, a small (1.0 cm^2) soft tissue sample would be collected from the trailing margin of soft tissue of one of the pectoral fins or caudal fin using sharp sterilized scissors. Tissue samples would be preserved in individually labeled vials containing 95% ethanol. Genetic tissue samples collected from shortnose sturgeon for archival purposes would be provided to the NOAA/NOS Tissue Archive in Charleston, South Carolina, or to Co-investigators identified in the permit. Proper certification, identity, and chain of custody of samples would be maintained during transfer of tissue samples.

Additionally, some ELS samples would be preserved in 95% ETOH for genetic tissue analysis to develop a population estimates based on the level of genetic diversity discovered in subsequent year classes. In this instance, genetic diversity measurements of DNA samples taken from both the parental generation and the ELS would be compared to make inferences on the minimum number of adults required to achieve the level of genetic diversity found. Genetic tissue samples from other matched pairs of year classes (i.e., 2007 and 2009) would also be compared for similar objectives.

PIT Tag. Prior to PIT tagging, the entire dorsal surface of captured sturgeon would be scanned using a PIT tag reader to detect PIT tags of previously captured fish. All unmarked shortnose sturgeon (≥330 mm TL) would be tagged using 11.9 mm x 2.1 mm PIT tags injected using a 12 gauge needle at an angle of 60 to 80° in the dorsal musculature (left and just anterior to the dorsal fin). No fish would be double-tagged with PIT tags. The last step after injecting PIT tags would be to verify and record the PIT tag code with a tag reader. During the study, the rate of PIT tag retention would be documented and reported to NMFS in annual reports.

Floy (T-bar Anchor) Tag. The researcher proposes to tag shortnose sturgeon with Floy (T-bar anchor) tags to incorporate incidental recaptures by commercial or recreational fishermen and other researchers to make possible collection of information useful for the assessment of the sturgeon population. In all captured shortnose sturgeon, Floy tags would be anchored in the dorsal fin musculature base and inserted forwardly and slightly downward from the left side to the right through the dorsal pterygiophores. After removing the injecting needle, the tag would be spun between the fingers and gently tugged to be certain it is locked in place. During the

study, the rate of Floy tag retention would be documented and reported to NMFS in annual reports.

Collecting Eggs/Larvae with Mats, D-nets, and Epibenthic Sleds. Three hundred eggs and or larvae (ELS) are requested by the applicant annually (not to exceed 900 over five years) to meet the following research objectives: 1) determine the upstream limit of shortnose sturgeon spawning in the Delaware River; 2) document habitat characteristics at shortnose sturgeon spawning locations; 3) estimate the relative intensity and periodicity of spawning by sampling region; and 4) document the genetic diversity component of the population derived in-part from analyses of early life stage nDNA and/or mtDNA.

Sampling ELS would be performed using egg mats, D-nets, or epibenthic sleds (described below) in previously documented spawning habitat for shortnose sturgeon between Trenton and five kilometers upstream of Lambertville, New Jersey (rkm 215-245). Sampling would be performed between late March and May, but could also be extended into June if shortnose sturgeon ELS are still being taken and the authorized take has not been exceeded. The positions and early spring movements of previously telemetered sturgeon would be monitored to aid in documenting spawning runs (i.e., locate spawning areas and document the spawning activity) at various locations in the river. Positions of tagged fish would be identified and recorded using portable GPS units and measures of key habitat attributes (water temperature, depth, current velocity, substrate, etc.) would be obtained. Once the location of spawning activity is suspected (typically between 10 ^{O}C and 18 ^{O}C) sampling devices would be deployed just downstream. Egg density, distribution, and spawning periodicity would be closely monitored throughout the spawning season so that annual egg deposition could be estimated for all suspected spawning areas.

ELS samples would be placed in container(s) filled with a 10 percent aqueous formalin solution and stored for laboratory analysis to make positive species identification. However, some samples also would be preserved in 95% ETOH for genetic testing. In this instance, genetic tissue samples from both the parents and the larvae/eggs would be compared to develop a population estimate based on the level of genetic diversity discovered in subsequent year classes. Once a total of 500 shortnose sturgeon eggs or larvae have been taken annually, artificial substrates, d-nets, or epibenthic sleds would be removed from the river.

Egg mat samplers proposed to sample ELS are 56cm diameter circular polyester floor-buffing pads anchored to the river bottom with concrete pavers and marked with a float. They are designed to passively collect eggs adrift deployed in a stratified fashion to cover likely spawning habitats (McCabe and Beckman 1990). They would be checked and reset at least once daily during the spawning season and collected eggs would immediately be transported to shore, photographed, removed from artificial substrates, and preserved for later laboratory analysis. Mats would then be returned to the river in their anchored position and any excess eggs would be allowed to remain on the mat to potentially hatch in the river.

To document spawning success and periodicity, D-frame ichthyoplankton nets (76 cm across the base, 54 cm high, fitted with a knotless 1600 µm mesh 317.5 cm nylon bag with a detachable cod end) would be bottom set within and just downstream of various suspected locations for up to 1-

3 hours (Taubert 1980, Auer and Baker 2002). The float and anchor configuration would hold the net upright in currents up to 1 m s^{-1}. A calibrated digital flow meter mounted in the center of the net mouth would be used to calculate the volume of water filtered for each sample to develop an index of abundance and spawning success (# ELS/ volume of water sampled). Ichthyoplankton net samples would be examined in the field if possible, or preserved and processed in the laboratory.

A similarly constructed D-frame ichthyoplankton net (as described above) would be fitted to an epibenthic sled towed slowly near documented spawning areas to collect eggs and larval samples. The sled is designed to be towed against the prevailing current for 5 minutes averaging approximately 1.0 m/second speed through water. A calibrated mouth would be used. Following deployment and retrieval of the sampling gear, net rinsing would be performed to concentrate the sample into the cod end bucket. The samples would then be examined to observe sturgeon egg/or larval species and the contents preserved for laboratory analysis and stored.

Implanting Acoustic Transmitter Tags. A maximum of 30 adult ($\geq$ 600 mm) and 30 juvenile (<600 mm) shortnose sturgeon annually would be surgically implanted with an internal acoustic transmitter. The total weight of tags would not exceed 2 percent of the fish's total body weight. Adult sturgeon would be tagged with VEMCO V16-5H acoustic tags, and juvenile sturgeon would be tagged with either VEMCO V7-4L, V9-6L, or V13-1H tags, depending on the weight of the individual sturgeon. Specifications for these transmitters are reported in Table 4.

Table 4. Proposed Vemco Acoustic Tag Models and Specifications

Model	Length	Diameter	Weight (H^20)	Weight (O^2)
V7-4L	22.5 mm	7 mm	1.0 g	1.8 g
V9-6L	21.0 mm	9 mm	1.6 g	2.9 g
V13-1H	36.0 mm	13 mm	6.0 g	11.0 g
V16-5H	95.0 mm	16 mm	16.0g	36.0 g

Anesthesia for Implanting Acoustic Tags. Shortnose sturgeon selected for transmitter implantation would be netted at temperatures 27 ^{O}C or below and 9 ^{O}C or above. Each sturgeon prepared for surgery would be anaesthetized using a solution of up to 150 mg/L of tricaine methane sulfonate (MS-222) buffered to neutral pH with sodium bicarbonate. A low volume pump would deliver the anesthetic over the fish's gills through a tube placed within the sturgeon's mouth until reaching a sedated to deep state of anesthesia (i.e., loss of equilibrium, some reaction to touch stimuli, opercula movement). The anesthetic's induction and recovery time would vary between 5 and 9 minutes, but would be appropriate for shortnose sturgeon under the specific water temperature and oxygen conditions present (Fox *et al*. 2000).

Surgery for Implanting Acoustic Tags. The following 5 to 8 minute transmitter implantation surgery under anesthesia (Coyle *et al.* 2004) would be used. Just prior to the surgical procedure, the tube supplying the anesthetic would be removed and the sturgeon would be placed on a moist surgery rack where respiration would be maintained by directing fresh

ambient water pumped across the gills with tube inserted in the animals' mouth. The incision site for implanting the tag (40 to 60 mm anterior to the pelvic fins, although the specific location would vary with fish size) would be disinfected with povidone iodine (10 percent solution). A sterile surgical packet containing all surgical instruments and supplies, would be used to make a 10 mm incision in individual fish selected for surgery. A sterilized sonic transmitter coated with an inert polymer compound would be inserted into the surgical openings of sturgeon and the incision closed with interrupted sutures of 3-0 polydioxanone (PDS) and treated with povidone iodine to prevent infection. Post-surgery fish would be held in an aerated holding tank and released upon recovery from anesthesia. Based on the implantation of over 175 acoustic tags in shortnose sturgeon under previous permits, the applicant estimated the surgical procedure would require approximately 5 to 8 minutes to complete, with a total holding time (anesthesia induction, surgery, and recovery) of 20 minutes or less. Internal tags would not be implanted in unhealthy or stressed fish or pre-spawning fish in the spring.

During processing, while in the onboard live well, all fish would be treated with a slime coat restorative and after surgery placed in a separate net pen to ensure full recovery prior to release. Any fish not responding readily would be recovered further in the net pen by holding the fish upright and immersed in river water and gently moved in a frontward motion to aid freshwater passage over the gills to stimulate the fish. When showing signs of being able to swim away strongly, the fish would be released and a spotter would watch to make sure the fish remains down and fully recovered.

Anesthesia for Laparoscopic Surgery. The proposed anesthesia protocol calls for a rapid induction of surgical anesthesia using a buffered solution of 250 mg/L MS-222 followed immediately by an 87.5 mg/L maintenance solution of MS-222 during surgery. Each animal chosen for laparoscopic examination (up to 24 proposed annually) would be selected in excellent, non-stressed condition when netted. When removed from the net, each fish would be immediately transported (two to three minute transport) to a near-by field laboratory providing a 110-v electrical outlet to operate the lab and surgical equipment. Upon arrival, the animal would be anesthetized with a 250 mg/L solution of buffered tricaine while and fitted with a heart rate monitor to assist determining when a state of surgical anesthesia has been reached

The researcher's goal would be to rapidly achieve the desired plane of surgical anesthesia while minimizing the stressful effects on animals during laparoscopy (Summerfelt and Smith 1990). Surgical anesthesia would be reached when the fish exhibits complete loss of equilibrium, decreased muscle tone and reaction to massive stimulation, while maintaining a depressed ventilation rate and a regular heart rate (Ross and Ross 1999, Summerfelt and Smith 1990). Just prior to laparoscopy, the animal would be positioned in lateral recumbence within a recirculating anesthesia machine delivering a maintenance dose of 87.5 mg/L buffered tricaine over the fish's gills. The time required to reach the proper plane of anesthesia would average 2 to 7 minutes (M. Matsche, Maryland Department of Natural Resources, unpublished).

The following anesthesia precautions are planned for both transmitter implantation and laparoscopy. Researchers performing anesthesia on shortnose sturgeon would have first received supervised training on shortnose sturgeon or another surrogate species before doing so, and the Permit Holder would report this training to NMFS prior to the activity. Researchers would use

MS-222 at concentrations up to 150 mg/L when anesthetizing shortnose sturgeon to implant acoustic transmitters and up to 250 mg/L when anesthetizing shortnose sturgeon for laparoscopic examinations. Such solutions would be made fresh daily. Only researchers designated in the permit are authorized to direct anesthesia using MS-222 at concentrations above 150 mg/L MS-222. While performing a surgical procedure, upon encountering a sudden reflex reaction from an anesthetized fish, the Researcher would stop the procedure and evaluate the level of anesthesia before proceeding. Prior to anesthetizing shortnose sturgeon with MS-222, researchers would saturate the solution with dissolved oxygen and buffer it to a neutral pH with sodium bicarbonate. Only visually non-stressed animals in excellent health would be anesthetized. To avoid injury while anesthetizing sturgeon in bath treatments, researchers would use restraint (e.g., netting) to prevent animals from jumping or falling out of the container. When anesthetizing shortnose sturgeon, researchers would observe animals closely to establish when the proper level of anesthesia is reached; further, a heart monitor would be used when using concentrations of MS-222 above 150 mg/L to induce surgical anesthesia. Researchers would observe shortnose sturgeon closely during recovery from anesthesia, ensuring full recovery prior to release. All researchers would wear protective clothing, gloves, and goggles when handling MS-222 powder and the MS-222 solution would be disposed of by using state adopted procedures.

Laparoscopic Surgery. Laparoscopic examinations have been used extensively in fisheries research (Murray 1998, Moccia *et al*. 1984, Ortenberger *et al*. 1996, and Stoskopf 1993) and refined for sturgeon work by Hernandez-Divers *et al*. (2004). Minimally invasive procedures (such as examining internal organs, determining sex, and performing biopsies), have been used by permit co-investigators (CIs, namely Mark Matsche) on the Delaware River for the past four years (Permit 1486) who have also conducted training courses on the same procedures for other researchers. The CIs now propose to continue these same techniques determining the sex and reproductive health of 24 adult shortnose sturgeon annually.

Using sterile technique, a small (~5 mm) incision would be made in the ventral body wall slightly off midline at a level midway between the pectoral girdle and the cloaca through which a 5-mm trocar would be inserted. A 5-mm rigid laparoscope would then be inserted through the trocar to allow visualization of gonads to determine sex and reproductive health of the animal. If necessary, the body cavity would be insufflated with ambient air by attaching a battery-powered air pump to the insufflation port of the trocar increasing the working space within the body cavity. On rare occasions, the swim bladder would be punctured with a hypodermic needle guided by the laparoscope in order to gain a general visual assessment of all internal organs. For each animal, a modified version of a quantitative health assessment index for rapid evaluation of fish (Adams *et al*. 1993) would be made in a standardized fashion so results could be compared between individuals. Determination of the sex and reproductive status of the animal would be made and recorded. In those instances where the sex of the animal is not readily apparent, a biopsy of the gonad would be taken.

Biopsy Procedure. In some instances where the sexes of the animal are not readily apparent, biopsies of the gonad material would be taken and preserved for histological evaluation and sex determination. To accomplish the biopsy, a second small (~5mm) incision would be made midway between the first incision and the pectoral girdle on the lateral aspect of the body

approximately 1 cm dorsal to the ventral scutes. A second 5 mm trocar would then be inserted through the new incision, followed by a laparoscopic biopsy instrument to biopsy the gonad material. The sample would be approximately 5 mm in size (2-3g sample) and would be placed in 10% neutral buffered formalin for preservation. Upon completion of the biopsy, the body cavity and biopsy site would again be visually assessed to ensure that there was no obvious hemorrhaged or herniated tissue requiring additional attention. The laparoscope and the two trocars would then be removed from the body and the incisions would be closed with a single suture in a cruciate pattern using polydioxanone (PDS) suture material.

Blood Collection. Blood would be collected from the caudal veins of 24 shortnose sturgeon adults annually to ascertain if estrogenic compounds might be adversely affecting the Delaware River shortnose sturgeon population. This would be achieved by inserting a hypodermic needle perpendicular to the ventral midline at a point immediately caudal to the anal fin. The needle would be slowly advanced while applying gentle negative pressure with the syringe until blood freely flows into the syringe. Once a blood sample is collected, direct pressure would be applied to the site of to ensure clotting and prevent further blood loss (Stoskopf 1993). Needle and syringe size, as well as blood volume collected, would be dependent on the fish size as presented in Table 5.

Table 5. Needle and Syringe Sizes Proposed Based on Fish Weight

Weight (g)	Sample Size (ml.)	Needle Size (Gauge x Length)	Syringe Size (ml.)
$\leq$ 1000	2	22g x 5/8”	3
1000 - 2000	3	22g x 5/8”	3
> 2000	6	20g x 1”	6

Each blood sample would be divided equally between two tubes, one tube containing the anti-coagulant lithium heparin and one tube containing none. Blood samples would then be centrifuged and placed in a cool dry place until they could be transferred by common carrier to Co-investigators identified in the permit for diagnostic work. In addition, a blood smear would be made at this time.

Unintentional Mortality of Shortnose Sturgeon. The researcher has requested one annual unintended mortality or serious injury annually, not to exceed three over the life of the permit. If a greater incidence of mortality or serious injury should occur, research would cease and NMFS-PR would need to be consulted to determine the cause of mortality and to discuss any remedial changes in research methods before a decision could be made to resume research. The Permits Division could grant authorization to resume permitted activities based on review of the incident depending on the circumstances, or else suspend activities.

II. PERMIT CONDITIONS

Terms and Conditions

The activities authorized herein must occur by the means, in the areas, and for the purposes set forth in the permit application, and as limited by the Terms and Conditions specified in this

permit, including all attachments and appendices. Any permit noncompliance constitutes a violation and is grounds for permit modification, suspension, or revocation, and for enforcement action.

A. Duration of Permit

1. Personnel listed in Condition C.1 of this permit (hereinafter "Researchers") may conduct activities authorized by this permit up to five years from the date of issuance. This permit expires on the date indicated and is non-renewable. This permit may be extended by the Director, NMFS Office of Protected Resources, pursuant to applicable regulations and the requirements of ESA.

2. Researchers must suspend all permitted activities in the event serious injury or mortality[2] of protected species reaches that specified in the Table 1 ("take table", Table 1 above in the Biological Opinion; Appendix 1 of the Permit). The Permit Holder must contact the Chief, NMFS Permits, Conservation and Education Division (hereinafter "Permits Division") by phone (301-713-2289) within two business days. The Permit Holder must also submit a written incident report as described in Condition E.2. The Permits Division may grant authorization to resume permitted activities based on review of the incident report and in consideration of the Terms and Conditions of this permit.

3. If authorized take[3] is exceeded, Researchers must cease all permitted activities and notify the Chief, NMFS Permits, Conservation and Education Division (hereinafter "Permits Division") by phone (301-713-2289) as soon as possible, but no later than within two business days. The Permit Holder must also submit a written incident report as described in Condition E.2. The Permits Division may grant authorization to resume permitted activities based on review of the incident report and in consideration of the Terms and Conditions of this permit, which includes initiation of consultation, as stated in the Modification, Suspension, and Revocation section of this permit.

Researchers must comply with the following conditions related to the manner of taking:

a. *Netting, Holding, and Handling Conditions*

i. The Permit Holder must take all necessary precautions to ensure sturgeon are not harmed during capture, including use of appropriate net mesh size and twine preventing shutting gill opercula, restricting gill netting activities, and decreasing the time of net sets.

[2] This permit allows for unintentional mortality or serious injury caused by the presence or actions of researchers. A serious injury is defined by regulation as any injury that will likely result in mortality. This includes, but is not limited to: deaths or injuries resulting from infections related to sampling procedures; and deaths or injuries sustained by animals during capture and handling, or while attempting to avoid researchers or escape capture.
[3] Under the ESA, a take means to harass, harm, pursue, hunt, shoot, wound, kill, trap, capture, or collect, or attempt to do any of the preceding.

ii. Location (GPS), temperature, dissolved oxygen (D.O.), capture gear used (e.g., mesh size, gillnet, trammel, trawl), soak time, species captured, and mortalities must be measured and recorded (at the depth fished) each time nets are set to ensure appropriate environmental netting conditions are adhered to. This data must be made available to NMFS in annual reports or upon request (Appendix 4).

iii. After removal from capture gear, sturgeon must be allow to recover in floating net pens or in onboard live wells while shielding them from direct sunlight.

iv. Researchers must carry secondary net pen(s) in the research vessel to accommodate larger catches; overcrowded fish must be transferred to the spare net pens or else released.

v. Any shortnose sturgeon overly stressed from capture must be resuscitated and allowed to recover inside net pen or live well; prior to release, it may only be PIT tagged, weighed, and measured.

vi. When water temperatures are below 15^{O}C, nets may be set in daylight hours only, or for a total of 10 hours, with one exception; that is, 14 hours net sets may take place overnight between rkm 186 and 215.

vii. Nets may not be set more than 4 hours in water between 15^{O}C and 25^{O}C.

viii. Nets may not be set more than 2 hours in water between 25^{O}C and 28^{O}C.

ix. Netting activities must cease above 28^{O}C until consulting with NMFS.

Table 1: Summary of Netting Conditions

Water Temperature (ºC)	Minimum D.O. Level (mg/L)[1]	Maximum Net Set Duration (hr)
< 15	5	14[2]
< 15	5	10
15 ≤ 25	5	4
25 ≤ 28	5	2
>28		Cease netting until consulting with NMFS

1. If DO concentration is between 4 and 5 mg/L at any temperature range, netting may still occur, but only at the next lower net-set duration indicated.

2. Net-set duration for 14 hours is limited to the river range of rkm 186 to 215 below 15ºC.

x. Gear must be deployed only in waters with having $\geq$ 5 mg/L D.O. at the deepest depth sampled by the gear for the duration of deployment, with one exception; that is, if D.O. is between 4 and 5 mg/L, netting may still occur, but at the next lower net set duration indicated in Table 1 above. All netting in waters with D.O. concentrations less than 4 mg/L must cease.

xi. A depth sounder/global positioning system must be used to monitor bottom characteristics to limit disturbance of substrate while netting or trawling.

xii. If a net or trawl becomes snagged (on bottom substrate, debris, etc.), it must be untangled immediately to reduce stress on the animals.

xiii. Trawls may be towed at a maximum speed of 2.5 knots and for no more than 10 minutes per trawl.

xiv. When fish are onboard the research vessel for processing, the flow-through holding tank must allow for total replacement of water volume every 15 minutes. Backup oxygenation of holding tanks with compressed oxygen is necessary to ensure sturgeon do not become stressed and D.O. levels remain at or above 5 mg/L.

xv. The total holding time of shortnose sturgeon after removal from capture gear must not exceed two hours unless fish have not recovered from anesthesia.

xvi. When water temperature exceeds 27^{o}C, the total holding time of shortnose sturgeon after removal from the capture gear must never be longer than 30 minutes.

xvii. The total handling time during research procedures must not exceed 20 minutes.

xviii. If water temperature exceeds 27^{O}C, or is less than 7 ^{O}C, research procedures must be non-invasive only (e.g., PIT and Floy tagging, measuring, weighing, and genetic tissue clipping).

xix. Hydroacoustic/sonar testing may take place when temperatures are below or equal to 15^{O}C and when D.O. is above 5 mg/L concentration; however, the time required for testing must not exceed two hours.

xx. Onboard handling of sturgeon should be minimized, keeping fish in water as much as possible and supported with a sling or net.

xxi. Smooth rubber gloves must be worn to reduce abrasion of skin and removal of mucus on fish.

xxii. Shortnose sturgeon (and bycatch) must be allowed to recover before they are released to ensure full recovery, and they must be treated with an electrolyte bath (e.g., salt) prior to release to help reduce stress and restore slime coat.

xxiii. Sturgeon are extremely sensitive to chlorine; therefore, a thorough flushing of holding tanks with bleach would be required between sampling periods.

b. *Larval Sampling with Artificial Substrates, D-nets and Epibenthic Sleds*

i. The total number of shortnose sturgeon eggs or larvae collected annually by artificial substrates, d-nets or epibenthic sleds must not exceed 300 (up to 900 during over five years).

ii. Once a total of 300 shortnose sturgeon eggs or larvae have been taken annually (up to 900 during over five years), artificial substrates, d-nets, or epibenthic sleds must be removed from the river.

iii. All artificial substrates, d-nets, or epibenthic sleds must be removed from the river upon completion of this project, or by the expiration date of this permit, whichever comes first.

iv. Ichthyoplankton net samples must be examined at least every three hours after deployed.

v. The epibenthic sled should be towed against the prevailing current for 5 minutes averaging approximately 1.0 m/second speed through water.

c. *Genetic Tissue Sampling*

i. Care must be used when collecting genetic tissue samples from the soft fin rays of sturgeon (pectoral or tail fins). Instruments should be changed or disinfected and gloves changed between each fish sampled to avoid possible disease transmission or cross contamination of genetic material.

ii. Submission and archival of genetic tissue samples must be coordinated with Julie Carter at the NOAA-NOS tissue archive in Charleston, SC (843)762-8547. Samples must be submitted between six and twelve months after collection.

iii. The Permit Holder may not transfer biological samples to anyone not listed in the application without obtaining prior written approval from NMFS. Any such transfer will be subject to such conditions as NMFS deems appropriate.

iv. The terms and conditions concerning samples collected under this authorization will remain in effect as long as the material taken is maintained under the authority and responsibility of the Permit Holder.

d. *Tagging Conditions*

i. PIT tags must be used to individually identify all captured fish not previously tagged. Prior to placement of PIT tags, the entire dorsal surface of each fish must be scanned with a waterproof PIT tag reader and visually inspected to ensure detection of fish tagged in other studies. Previously PIT-tagged fish must not be retagged.

ii. Researchers must not insert PIT tags or perform other surgical procedures on juvenile shortnose sturgeon less than 330 mm in length.

iii. PIT tags must be injected in the left, dorsal musculature just anterior to the dorsal fin with the copper antenna oriented up for maximum signal strength and scanned after implantation to ensure proper tag function.

iv. Numbered Floy tags must be anchored in the dorsal fin musculature base, inserted forwardly and slightly downward from the left side to the right through the dorsal pterygiophores.

v. The rate of PIT tag and Floy tag retention and the condition of fish at the site of tag injection must be documented during the study and results reported to NMFS in annual and final reports.

vi. The total weight of tags must not exceed 2% of the sturgeon's total body weight unless otherwise authorized by NMFS-PR.

vii. Surgical implantation of internal tags must only be attempted when fish are in excellent condition, and must not be attempted on pre-spawning fish in spring or fish on the spawning ground.

viii. During surgical procedures, instruments must be sterilized or

changed between uses.

ix. To ensure proper closure of surgical incisions, a single interrupted suturing technique should be applied.

e. *Anesthetization*

i. Researchers performing anesthesia on shortnose sturgeon must have first received supervised training on shortnose sturgeon or another surrogate species before doing so. The Permit Holder must report this training to NMFS prior to the activity.

ii. Researchers may use MS-222 at concentrations up to 150 mg/L when anesthetizing shortnose sturgeon to implant acoustic transmitters; and up to 250 mg/L when anesthetizing shortnose sturgeon for laparoscopic examinations; such solutions should be made fresh daily.

iii. Only researchers designated in the permit are authorized to direct anesthesia using MS-222 at concentrations above 150 mg/L MS-222.

iv. While performing a surgical procedure, upon encountering a sudden reflex reaction from an anesthetized fish, the Researcher must stop the procedure and evaluate the level of anesthesia before proceeding.

v. Prior to anesthetizing shortnose sturgeon with MS-222, researchers must saturate the solution with dissolved oxygen and buffer it to a neutral pH with sodium bicarbonate.

vi. Only visually non-stressed animals in excellent health can be anesthetized.

vii. To avoid injury while anesthetizing sturgeon in bath treatments, researchers must use restraint (e.g., netting) to prevent animals from jumping or falling out of the container.

viii. When anesthetizing shortnose sturgeon, researchers must observe animals closely to establish when the proper level of anesthesia is reached; further, a heart monitor must be used when using concentrations of MS-222 above 150 mg/L to induce surgical anesthesia.

x. Researchers must observe shortnose sturgeon closely during recovery from anesthesia, ensuring full recovery prior to release.

xi. All researchers are required to wear protective clothing, gloves, and goggles when handling MS-222 powder.

xii. MS-222 solution must be disposed of by using state adopted procedures.

f. *Laparoscopic Examination, Gonad Biopsy and Blood Collection*

i. Only designated CIs are authorized to direct laparoscopy, blood sampling, or biopsy procedures.

ii. Should an uncontrolled hemorrhage occur while performing a surgical procedure, the procedure should be stopped and the bleeding stabilized before proceeding.

iii. Blood and biopsy samples may be transported for analyses to the Maryland DNR, Oxford Maryland Laboratory. Blood samples may also be sent to the Antech Diagnostics Laboratory, Lake Success, New York.

iv. Blood and biopsy samples, not consumed during testing, must be properly disposed of after all testing is completed.

Reporting Requirements

1. The Permit Holder must submit annual, final, and incident reports, and any papers or publications resulting from the research authorized herein to the Permits Division. Reports may be submitted:

- through the online system at https://apps.nmfs.noaa.gov
- by email attachment to the permit analyst for this permit
- by hard copy mailed or faxed to the Chief, Permits Division, Office of Protected Resources, NMFS, 1315 East-West Highway, Suite 13705, Silver Spring, MD 20910; phone (301)713-2289; fax (301)713-0376.

2. Written incident reports related to serious injury and mortality events or to exceeding authorized takes, must be submitted to the Chief, Permits Division within two weeks of the incident. The incident report must include a complete description of the events and identification of steps that will be taken to reduce the potential for additional research-related mortality or exceedence of authorized take.

3. An annual report must be submitted to the Chief, Permits Division by December 30 for each year the permit is valid. The annual report describing activities conducted during the previous permit year must follow the format in Appendix 2.

4. A final report must be submitted to the Chief, Permits Division within 180 days after expiration of the permit, or five years after its date of issuance, or, if the research concludes prior to permit expiration, within 180 days of completion of the research. The final report must follow the format in Appendix 2.

5. Careful and detailed records must be kept on the time of recovery and other responses from anesthesia, handling, tissue sampling, tag retention and healing, and condition and health of any shortnose sturgeon.

6. A *Biological Sample Certification, Identification and Chain of Custody Form* (Appendix 3a) must accompany shipments of genetic tissue samples to the NOAA-NOS archive in Charleston, South Carolina. Samples must be submitted between six and twelve months after collection.

7. A *Field Collection Report* appearing in Appendix 3b should also accompany multiple genetic tissue samples (hard copy or spreadsheet) when shipping to the NOAA-NOS archive.

8. Environmental sampling data (e.g., dissolved oxygen, temperature, net set duration, and other data associated with capture) must be recorded (See Appendix 4) and made available to NMFS in annual reports or when requested periodically.

9. Specimens or body parts of dead shortnose sturgeon should be individually preserved — preferably on ice or refrigeration — until sampling and disposal procedures are discussed with NMFS. The take should be documented by completing the sturgeon salvage form (Appendix 5).

10. NMFS requests Atlantic sturgeon interactions are reported to Lynn Lankshear, NMFS-PR (Lynn.Lankshear@noaa.gov or 978-281-9300 x 6535). This report should be documented by completing the sturgeon salvage form (Appendix 5). Specimens or body parts of dead Atlantic sturgeon should be preserved — preferably on ice or refrigeration — until sampling and disposal procedures are discussed with NMFS.

11. Research results must be published or made available to the scientific community in a reasonable period of time, or to NMFS when requested periodically.

Notification and Coordination

1. The Permit Holder must provide written notification of planned field work to the Assistant Regional Administrator for Protected Resources at the address listed below. Such notification must be made at least two weeks prior to initiation of any field trip/season and must include the locations of the intended field study and/or survey routes, estimated dates of research, and number and roles (for example: PI, CI, veterinarian, boat driver, safety diver, animal restrainer, Research Assistant "in training") of participants.

   Northeast Regional Office, NMFS, Protected Resources Division, 55 Great Republic Drive, Gloucester, MA 01930-2298; phone (978) 281-9300; fax (978) 281-9394.

2. To the maximum extent practical, the Permit Holder must coordinate permitted activities with activities of other Permit Holders conducting the same or similar activities on the same species, in the same locations, or at the same times of year to avoid unnecessary disturbance of animals. The appropriate Regional Office may be contacted at the address listed above for information about coordinating with other Permit Holders.

III. APPROACH TO THE ASSESSMENT

NMFS approaches its section 7 analyses of research permits through a series of steps. The first step identifies those aspects of proposed actions that are likely to have direct and indirect physical, chemical, and biotic effects on listed species or on the physical, chemical, and biotic environment of an action area. As part of this step, we identify the spatial extent of these direct and indirect effects, including changes in that spatial extent over time. The results of this step define the action area for the consultation. The second step of our analyses identifies the listed resources that are likely to co-occur with these effects in space and time and the nature of that co-occurrence (these represent our exposure analyses). In this step of our analyses, we try to identify the number, age (or life stage), and gender of the individuals that are likely to be exposed to an action's effects and the populations or subpopulations those individuals represent. Once we identify which listed resources are likely to be exposed to an action's effects and the nature of that exposure, we examine the scientific and commercial data available to determine whether and how those listed resources are likely to respond given their exposure (these represent our response analyses).

The final steps of our analyses – establishing the risks those responses pose to listed resources – are different for listed species and designated critical habitat (these represent our risk analyses). Our jeopardy determinations must be based on an action's effects on the continued existence of threatened or endangered species as those "species" have been listed, which can include true biological species, subspecies, or distinct populations of vertebrate species. Because the continued existence of species depends on the fate of the populations that comprise them, the continued existence of these "species" depends on the fate of the populations that comprise them. Similarly, the continued existence of populations are determined by the fate of the individuals that comprise them; populations grow or decline as the individuals that comprise the population live, die, grow, mature, migrate, and reproduce (or fail to do so).

Our risk analyses reflect these relationships between listed species, the populations that comprise that species, and the individuals that comprise those populations. Our risk analyses begin by identifying the probable risks actions pose to listed individuals that are likely to be exposed to an action's effects. Our analyses then integrate those individual risks to identify consequences to the populations those individuals represent. Our analyses conclude by determining the consequences of those population level risks to the species those populations comprise.

We measure risks to listed individuals using the individuals' "fitness," or the individual's growth, survival, annual reproductive success, and lifetime reproductive success. In particular, we examine the scientific and commercial data available to determine if an individual's probable lethal, sub-lethal, or behavioral responses to an action's effect on the environment (which we

identify during our response analyses) are likely to have consequences for the individual's fitness.

When individual, listed plants or animals are expected to experience reductions in fitness in response to an action, those fitness reductions are likely to reduce the abundance, reproduction, or growth rates (or increase the variance in these measures) of the populations those individuals represent (*see* Stearns 1992). Reductions in at least one of these variables (or one of the variables we derive from them) is a necessary condition for reductions in a population's viability, which is itself a necessary condition for reductions in a species' viability. As a result, when listed plants or animals exposed to an action's effects are not expected to experience reductions in fitness, we would not expect the action to have adverse consequences on the viability of the populations those individuals represent or the species those populations comprise (e.g., Brandon 1978, Mills and Beatty 1979, Stearns 1992, Anderson 2000). As a result, if we conclude that listed plants or animals are not likely to experience reductions in their fitness, we would conclude our assessment.

Although reductions in fitness of individuals are a necessary condition for reductions in a population's viability, reducing the fitness of individuals in a population is not always sufficient to reduce the viability of the population(s) those individuals represent. Therefore, if we conclude that listed plants or animals are likely to experience reductions in their fitness, we determine whether those fitness reductions are likely to reduce the viability of the populations the individuals represent (measured using changes in the populations' abundance, reproduction, spatial structure and connectivity, growth rates, variance in these measures, or measures of extinction risk). In this step of our analyses, we use the population's base condition (established in the *Environmental Baseline* and *Status of the Species* sections of this Opinion) as our point of reference. If we conclude that reductions in individual fitness are not likely to reduce the viability of the populations those individuals represent, we would conclude our assessment.

Reducing the viability of a population is not always sufficient to reduce the viability of the species those populations comprise. Therefore, in the final step of our analyses, we determine if reductions in a population's viability are likely to reduce the viability of the species those populations comprise using changes in a species' reproduction, numbers, distribution, estimates of extinction risk, or probability of being conserved. In this step of our analyses, we use the species' status (established in the *Status of the Species* section of this Opinion) as our point of reference. Our final determinations are based on whether threatened or endangered species are likely to experience reductions in their viability and whether such reductions are likely to be appreciable.

To conduct these analyses, we rely on all of the evidence available to us. This evidence might consist of monitoring reports submitted by past and present permit holders; reports from NMFS Science Centers; reports prepared by natural resource agencies in states, and other countries; reports from foreign and domestic nongovernmental organizations involved in marine conservation issues; the information provided by PR1 when it initiates formal consultation; information from commercial interests; and the general scientific literature.

During each consultation, we conduct electronic searches of the general scientific literature using *American Fisheries Society*, *Google Scholar*, *ScienceDirect*, *BioOne*, *Conference Papers Index, JSTOR,* and *Aquatic Sciences and Fisheries Abstracts* search engines. We supplement these searches with electronic searches of doctoral dissertations and master's theses. These searches specifically try to identify data or other information that supports a particular conclusion (for example, a study that suggests shortnose sturgeon will exhibit a particular response to DO concentrations) as well as data that does not support that conclusion. When data are equivocal, or in the face of substantial uncertainty, our decisions are designed to avoid the risks of incorrectly concluding that an action would not have an adverse effect on listed species when, in fact, such adverse effects are likely.

We rank the results of these searches based on the quality of their study design, sample sizes, level of scrutiny prior to and during publication, and study results. Carefully designed field experiments (for example, experiments that control potentially confounding variables) are rated higher than field experiments that are not designed to control those variables. Carefully designed field experiments are generally ranked higher than computer simulations. Studies that produce large sample sizes with small variances are generally ranked higher than studies with small sample sizes or large variances.

IV. DESCRIPTION OF THE ACTION AREA

The proposed action area consists of the Delaware Bay, tidal river, and freshwater sections of the Delaware River extending from the mouth of the Delaware Bay (rkm 0) to just upstream of Lambertville, New Jersey (rkm 245) (Figure 1). The Delaware River Estuary and mixed tidal river extends from Cape May (NJ) and Cape Henlopen, Delaware (rkm 0) to the fall line at Trenton, New Jersey (rkm 215). The Delaware Bay region of the estuary is 72 kilometers extending from the Capes to a line between stone markers at Liston Point, Delaware and Hope Creek, New Jersey (Polis *et al*. 1973).

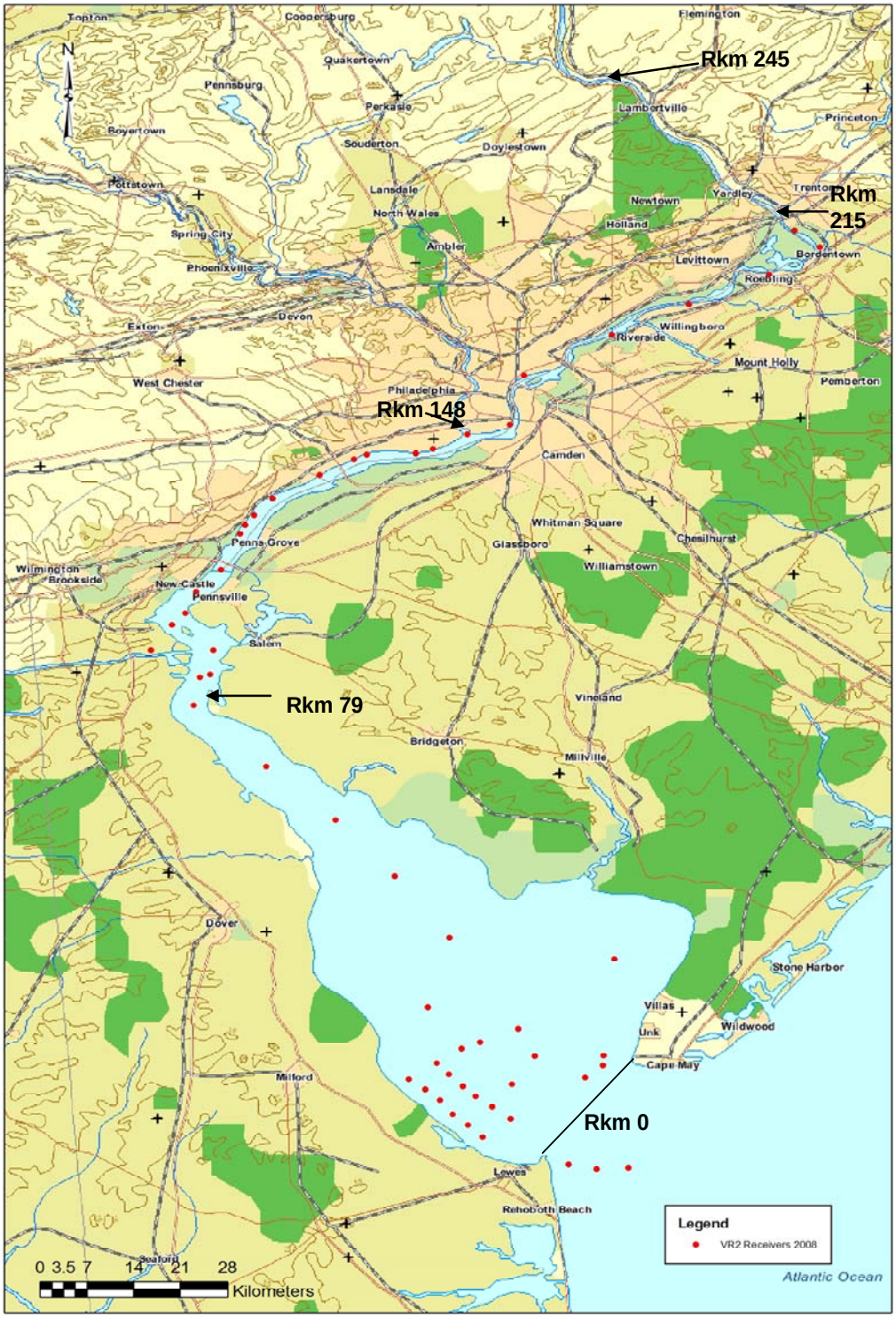
The Delaware Estuary varies in width from 18 kilometers at the Capes; to 43 kilometers at its widest point (near Miah Maull Shoals). Water depth in the bay is less than 9 meters in 80 percent of the bay, excluding the dredged channel, and is less than 3 meters deep in much of the tidal river area. Artificial Island (rkm 79) is located approximately seven kilometers upstream of the hypothetical line demarking the head of Delaware Bay. The tidal river upstream of this area narrows makes a northwesterly 60 degree bend accentuated by Artificial Island on the New Jersey shore. More than half of the typical river width in this area is relatively shallow — less than 5.5 meters — while the deeper part, including the dredged channel has depths of up to 12.2 meters. The Delaware River between Philadelphia (rkm 161) and Trenton (rkm 215) is tidal freshwater with semidiurnal tides. Mean tidal range at Philadelphia, Pennsylvania is 1.8 meters (USACOE, Philadelphia District 2009), and water pH generally is about 6-8.

Average tidal flow, as measured near the Delaware Memorial Bridge (rkm 108) approximately 32 kilometers above Artificial Island, amounts to 11,320 cubic meters per second (NMFS 2009). At this point, tidal flow of this magnitude is 17 times greater than the total average freshwater flow rate flowing into the estuary. Proceeding south (toward the mouth of the estuary), tidal

flow increasingly dominates freshwater downstream flow; proceeding upstream, the ratio of tidal flow to net downstream flow becomes smaller as tidal influence decreases.

The freshwater portion of the action area extends above the fall line at Trenton, New Jersey (rkm 215) to just north of Lambertville, New Jersey (rkm 245), and is characterized by bottom substrate consisting of rocky shoals and cobble substrate suitable for shortnose sturgeon spawning habitat.

Figure 1: Map of Action Area



V. Status of the Species/Critical Habitat

NMFS has determined that the action being considered in this Opinion may affect the following species protected under the ESA:

Shortnose sturgeon	*Acipenser brevirostrum*	Endangered

No critical habitat has been designated for shortnose sturgeon; therefore, none will be affected by the proposed action.

The following summarizes the biology and ecology of the endangered species in the action area that are relevant to the effects analysis in this Opinion. For more comprehensive treatments of the biology, ecology, and management of shortnose sturgeon, refer to Dadswell *et al.* (1984), Gilbert (1989), the Final Recovery Plan for Shortnose Sturgeon (NMFS 1998), and the Canadian Assessment and Update Status Report on the Shortnose Sturgeon (COSEWIC 2005).

A. Listed Resources Not Considered Further in this Opinion

The following ESA listed resources occur in the action area, but we believe they are either not likely to be exposed to the proposed research or are not likely to be adversely affected.

Green sea turtle	*Chelonia mydas*	Endangered
Hawksbill sea turtle	*Eretmochelys imbricata*	Endangered
Kemp's ridley sea turtle	*Lepidochelys kempii*	Endangered
Leatherback sea turtle	*Dermochelys coriacea*	Endangered
Loggerhead sea turtle	*Caretta caretta*	Threatened
Humpback whale	*Megaptera novaeangliae*	Endangered
Fin whale	*Balaenoptera physalus*	Endangered
North Atlantic right whale	*Balaena glacialis*	Endangered

The authorized activities would include netting in freshwater-tidally mixed areas between rkm 79 to 215. Although sea turtles and some listed whales occur within the action area, we believe that it is highly unlikely that these animals would swim upriver to where nets will be set (rkm 79 to 215). Therefore, entanglement in nets is expected to be unlikely. There have been reports of sea turtles being seen as far up-river as Artificial Island, NJ (rkm 79), so netting safeguards have been incorporated into the permit. These safeguards are similar to those for marine mammals and include: 1) continual, complete, and thorough visual net checks; 2) netting time restricted between 30 minutes after sunrise to 30 minutes before sunset; and 3) no deployment of nets if other listed species are found in the action area. Sea turtles and listed whales, if at all, would most likely be found in Delaware Bay and possibly the lower river.

B. Status of Species Considered in this Opinion

Species Description, Range-wide Distribution, and Population Structure. Shortnose sturgeon occur along the Atlantic Coast of North America, from the St. John River in Canada to the St. Johns River in Florida. The Shortnose Sturgeon Recovery Plan describes 20 shortnose

sturgeon population segments that exist in the wild. Two additional geographically distinct populations occur behind dams in the Connecticut River (above the Holyoke Dam) and in Lake Marion on the Santee-Cooper River system in South Carolina (above the Wilson and Pinopolis Dams). Although these populations are geographically isolated, genetic analyses suggest that individual shortnose sturgeon move between some of these populations each generation (Quattro *et al.* 2002, Wirgin *et al.* 2005).

At the northern end of the species' distribution, the highest rate of gene flow (which suggests migration) occurs between the Kennebec and Androscoggin Rivers. At the southern end of the species' distribution, populations south of the Pee Dee River appear to exchange between 1 and 10 individuals per generation, with the highest rates of exchange between the Ogeechee and Delaware Rivers (Wirgin *et al.* 2005). Wirgin *et al.* (2005) concluded that rivers separated by more than 400km were connected by very little migration while rivers separated by no more than 20km (such as the rivers flowing into coastal South Carolina) would experience high migration rates. Coincidentally, at the geographic center of the shortnose sturgeon range, there is a 400km stretch of river with no known populations occurring from the Delaware River, New Jersey to Cape Fear River, North Carolina (Kynard 1997). However, shortnose sturgeon are known to occur in the Chesapeake Bay, and may be transients from the Delaware River via the Chesapeake and Delaware Canal (Skjeveland *et al.* 2000, Welsh *et al.* 2002) or remnants of a population in the Potomac River.

Several authors have concluded that shortnose sturgeon populations in the southern end of the species geographic range are extinct. Rogers and Weber (1994), Kahnle *et al.* (1998), and Collins *et al.* (2000) concluded that shortnose sturgeon are extinct from the St. Johns River in Florida and the St. Marys River along the Florida and Georgia border. Rogers and Weber (1995b) also concluded that shortnose sturgeon have become extinct in Georgia's Satilla River.

Table 6. Estimated shortnose sturgeon population densities.

Population/Subpopulation	Distribution	Datum	Estimate	Confidence Interval	Authority
Saint John River	New Brunswick, Canada	1973/1977	18,000	30%	Dadswell 1979
Kennebecasis River	Canada	1998 – 2005	2,068	801 - 11,277	COSEWIC 2005
Penobscot River	ME	2006 - 2007	1,049	673 – 6,939	Univ. Maine, 2008 SJ Fernandes - 2008
Kennebec River	ME	1977/1981	7,200	5,046 - 10,765	Squiers *et al.* 1982
		2003	9,500	6,942 - 13,358	Squiers 2003
Androscoggin River	ME		7200	5000 - 10,800	Squiers *et al.* 1993
Merrimack River	MA	1989 – 1990	33	18 - 89	NMFS 1998
Connecticut River	MA, CT	2003	-	1,500 - 1,800	Connecticut DEP 2003
		1998-2002	-	1,042 - 1,580	Savoy 2004
Above Holyoke Dam		1976 – 1977	515	317 - 898	Taubert 1980, NMFS 1998

Population/Subpopulation	Distribution	Datum	Estimate	Confidence Interval	Authority
		1977 – 1978	370	235 - 623	Taubert 1980, NMFS 1998
		1976 – 1978	714	280 – 2,856	Taubert 1980, NMFS 1998
		1976 – 1978	297	267 - 618	Taubert 1980, NMFS 1998
Below Holyoke Dam		1988 – 1993	895	799 – 1,018	Savoy and Shake 1992,
Hudson River	NY	1980	30,311		Dovel 1979, NMFS 1998
		1995	38,000	26,427 - 55,072	Bain *et al.* 1995, NMFS 1998
		1997	61,000	52,898 - 72,191	Bain *et al.* 2000
Delaware River	NJ, DE, PA	1981/1984	12,796	10,288 - 16,367	Hastings *et al.* 1987
		1999/2003	12,047	10,757 - 13,589	Brundage and O'Herron 2003
Chesapeake Bay	MD, VA	no data	-	-	
Potomac River	MD, VA	no data	-	-	
Neuse River	NC	2001-2002	extirpated		Oakley 2003, Oakley and Hightower 2007
Cape Fear River	NC	1997	>100		Kynard 1997, NMFS 1998
Winyah Bay	NC, SC	no data	-	-	
Waccamaw - Pee Dee River	SC	no data	-	-	
Santee River	SC	no data	-	-	
Lake Marion (dam-locked)	SC	no data	-	-	
Cooper River	SC	no data	-	-	
ACE Basin	SC	no data	-	-	
Savannah River	SC, GA		1,000 - 3,000		Bill Post, SCDNR 2003
Ogeechee River	GA	1990s	266		Bryce *et al.* 2002
		1993	266	236 - 300	Kirk *et al.* 2005
		1993	361	326 - 400	Rogers and Weber 1994
		1999/2000	195	-	Bryce *et al.* 2002
		2000	147	105 - 249	Kirk *et al.* 2005
		2004	174	97 - 874	Kirk *et al.* 2005
		2008	368	244-745	Kirk 2008 NMFS Ann. Report
Delaware River	GA	1988	2,862	1,069 - 4,226	NMFS 1998
		1990	798	645 – 1,045	NMFS 1998
		1993	468	315 - 903	NMFS 1998
		2003-2005	6,320	4,387-9,249	DeVries 2006
Satilla River	GA		unknown	-	Kahnle *et al.* 1998
Saint Marys River	FL		unknown	-	Kahnle *et al.* 1998, Rogers and Weber 1994
Saint Johns River	FL	2002	1	-	FFWCC 2007

In addition to these wild populations there are several captive populations of shortnose sturgeon (Table 7). One captive population of shortnose sturgeon is maintained at the Conte Anadromous Fish Research Center in Massachusetts, which is operated by the United States Fish and Wildlife Service (USFWS). These sturgeon were taken from the Connecticut River population and are currently held by Dr. Boyd Kynard under Permit Number 1239. Captive populations of shortnose sturgeon captured from the Savannah River population are housed at three USFWS hatcheries: Bear's Bluff (South Carolina), Orangeburg (South Carolina), and Warm Springs (Georgia). The USFWS provides progeny of these captive shortnose sturgeon to other facilities for research, educational purposes, and public display. The University of Florida (Gainesville) recently acquired shortnose sturgeon from these hatcheries for research purposes.

Smaller, captive populations that have been developed from these USFWS facilities are maintained in several facilities for educational purposes. The South Carolina Aquarium in Charleston, South Carolina, maintains a population of eight juvenile shortnose sturgeon. The Springfield Science Museum in Springfield, Massachusetts, maintains a population of about five juvenile shortnose sturgeon. Captive populations are also held in the North Carolina Zoo in Asheboro, North Carolina; National Aquarium in Baltimore, Maryland; and the Riverbanks Zoological Park in Columbia, South Carolina.

Table 7. Populations reared in captivity

Conte Fish Research Center	MA
Bear's Bluff hatchery	SC
Orangeburg hatchery	SC
Warm Springs hatchery	GA

Life History Information. Shortnose sturgeon are anadromous fish that live primarily in slower moving rivers or nearshore estuaries near large river systems. They are benthic omnivores that feed on crustaceans, insect larvae, worms, and molluscs (Moser and Ross 1995, NMFS 1998) but they have also been observed feeding off plant surfaces and on fish bait (Dadswell *et al.* 1984).

During the summer and winter, adult shortnose sturgeon occur in freshwater reaches of rivers or river reaches that are influenced by tides; as a result, they often occupy only a few short reaches of a river's entire length (Buckley and Kynard 1985). During the summer, at the southern end of their range, shortnose sturgeon congregate in cool, deep, areas of rivers where adult and juvenile sturgeon can take refuge from high temperatures (Flournoy *et al.* 1992, Rogers and Weber 1994, Rogers and Weber 1995b, Weber 1996). Juvenile shortnose sturgeon generally move upstream for the spring and summer seasons and downstream for fall and winter; however, these movements usually occur above the salt- and freshwater interface of the rivers they inhabit (Dadswell *et al.* 1984, Hall *et al.* 1991). Adult shortnose sturgeon prefer deep, downstream areas with soft substrate and vegetated bottoms, if present. Because they rarely leave their natal rivers, Kieffer and Kynard (1993) considered shortnose sturgeon to be freshwater amphidromous (*i.e.* adults spawn in freshwater but regularly enter saltwater habitats during their life).

Shortnose sturgeon in the northern portion of the species' range live longer than individuals in the southern portion of the species' range (Gilbert 1989). The maximum age reported for a shortnose sturgeon in the St. John River in New Brunswick is 67 years (for a female), 40 years

for the Kennebec River, 37 years for the Hudson River, 34 years in the Connecticut River, 20 years in the Pee Dee River, and 10 years in the Delaware River (Gilbert 1989 using data presented in Dadswell *et al.* 1984). Male shortnose sturgeon appear to have shorter life spans than females (Gilbert 1989).

Listing Status. Shortnose sturgeon were listed as endangered on March 11, 1967 (32 FR 4001) pursuant to the Endangered Species Preservation Act of 1966. Shortnose sturgeon remained on the list as endangered with the enactment of the ESA in 1973. Shortnose sturgeon were first listed on the International Union for Conservation of Nature and Natural Resources Red List in 1986 where it is still listed as Vulnerable and facing a high risk of extinction based in part on: an estimated range reduction of greater than 30% over the past three generations, irreversible habitat losses, effects of habitat alteration and degradation, degraded water quality and extreme fluctuations in the number of mature individuals between rivers.

Status and Trends of Shortnose Sturgeon Populations. Despite the longevity of adult sturgeon, the viability of sturgeon populations are highly sensitive to juvenile mortality that result in reductions in the number of sub-adults that recruit into the adult breeding population (Anders *et al.* 2002, Gross *et al.* 2002, Secor *et al.* 2002). This relationship caused Secor *et al.* (2002) to conclude that sturgeon populations can be grouped into two demographic categories: populations that have reliable (albeit periodic) natural recruitment and those that do not. The shortnose sturgeon populations without reliable natural recruitment are at the greatest risk.

Several authors have also demonstrated that sturgeon populations generally, and shortnose sturgeon populations in particular, are much more sensitive to adult mortality than other species of fish (Boreman 1997, Gross *et al.* 2002, Secor *et al.* 2002). These authors concluded that sturgeon populations cannot survive fishing related mortalities that exceed five percent of an adult spawning run and they are vulnerable to declines and local extinction if juveniles die from fishing related mortalities.

Based on the information available, most shortnose sturgeon populations in the northern portion of the species range, from Delaware River north to the St. John River in Canada, appear to have sufficient juvenile survival to provide at least periodic recruitment into the adult age classes combined with relatively low adult mortality rates sufficient to maintain the viability of most of these populations. As a result, most of these populations appear to be relatively large and stable, except for shortnose sturgeon populations in the Merrimack and Connecticut Rivers (Table 6).

Delaware River Shortnose Sturgeon Population. Shortnose sturgeon occur throughout the Delaware River estuary and occasionally enter the nearshore ocean off Delaware Bay (Brundange and Meadows 1982). Tagging studies by O'Herron *et al.* (1993) found that the most heavily used portion of the river appears to be between river mile 118 below Burlington Island and river mile 137 at the Trenton Rapids. In spring, spawning adults migrate up-river in the non-tidal river in freshwater, and are common at least as far upstream as Scudders Falls (rkm 225). According to Dadswell *et al*. (1984), ripe adults have been captured as far upstream as Lambertville (rkm 240). The farthest upstream confirmed account of a shortnose sturgeon in the Delaware River is from 1998. A fish was captured during electrofishing for American shad below the lower tip of Old Sow Island near Raubsville, Pennsylvania (rkm 287).

Hastings *et al*. (1987) estimated a modified Schnabel estimate of adult shortnose sturgeon in the Delaware River at 12,796 (95% confidence interval – 10,228 to 16,367) based on mark recapture data collected during 1981-1984. Environmental Research and Consulting, Inc. (2006b) later estimated the population at 12,047 – 13,580. A Chapman modification of the Schnabel estimate was used based on mark-recapture data collected from January 1999 through March 2003.

Similarity between the two estimates suggests that the Delaware River shortnose sturgeon population is stable but has not increased in the 20+ years between studies. The recapture of 168 shortnose sturgeon during the later study, tagged as adults by Hastings *et al.* (1987), suggests that older fish comprise a substantial portion of the Delaware River population (ERC, Inc. 2006b).

Delaware shortnose sturgeon are documented to spawn from late March through early May. Spawning occurs primarily between Scudders Falls and the Trenton rapids (rkm approximately 223-215) in Mercer County (Hoff 1965, O'Herron *et al*. 1993). The capture of early life stages (eggs and larvae) in this region in the spring of 2008 confirms that this area of the river is used for spawning and as a nursery area. Shortnose sturgeon eggs have also been collected upstream of Titusville, NJ (rkm 229) in spring 2008. The river in the nontidal area, beginning at the fall line at Trenton Rapids, is relatively shallow (<3 meters in summer) characterized by pools, riffles and rapids (O'Herron *et al*. 1993) and the substrate is composed primarily of sand, gravel, and cobble, with soft sediments found in areas of weaker currents. Spawning can occur between 8 and 25^{o}C, with most spawning occurring within the 10-18^{o}C range. Recent surveys by ERC, Inc. for early life stages, as well as observations from impingement/entrainment studies, confirm the presence of shortnose sturgeon larvae and/or eggs between Scudders Falls (rkm 223) and Trenton (rkm 215). Larvae collected at Fairless Hills, PA, cogeneration plant (approximately rkm 191) (well south of the spawning/rearing area), may have been carried there during a one day flood event.

After spawning, most adult shortnose sturgeon spend the summer and early fall foraging throughout the river, between the vicinity of Trenton south to Artificial Island (rkm 79) (J. O'Herron, *pers. comm.* 2008). Some foraging may also occur in winter, though sturgeon are not feeding heavily at this time (J. O'Herron, *pers. comm*. 2008). Predominate substrates in the tidal river include fine grain sediments (silt, sand and clay). Larger substrates ranging from gravel to bedrock can be found in certain areas, however (ERC, Inc. 2006b). Though gut analysis has not been performed on Delaware River shortnose sturgeon, according to J. O'Herron (*pers. comm.* 2008), oligochaetes, Asian clams and chironomids were observed over occupied sturgeon habitats during macroinvertebrate sampling conducted in the early 1980's.

Shortnose sturgeon were found to overwinter in the Roebling (rkm 199), Bordentown, (rkm 207), or Trenton reaches from December-March. The channel off Duck Island (rkm 208) is known to be used heavily by overwintering shortnose sturgeon (O'Herron 1993). Recent acoustic tagging studies indicate the existence of an overwintering area in the lower portion of the river, below Wilmington, DE (ERC, Inc. 2006a). Wintering adults are normally observed in tight aggregations and movement at this time appears to be minimal. In addition, results from a preliminary tracking study of juvenile shortnose sturgeon suggest that the entire lower Delaware River from Philadelphia (approx. rkm 161) to below Artificial Island (rkm 79) may be utilized as an overwintering area by juvenile shortnose sturgeon (ERC, Inc. 2007b). According to ERC,

Inc. (2007b), juvenile sturgeon in the Delaware River appear to overwinter in a dispersed fashion rather than in dense aggregations like adults.

Acoustic tagging studies by ERC, Inc. (2006a) indicate that adult shortnose sturgeon demonstrate one of two generalized movement patterns, either making long excursions from the upper to the lower tidal river (Pattern A) or remaining in and utilizing the upper tidal river (Pattern B) (ERC, Inc. 2006a). Fish with Pattern A movements made long distance excursions, often moving between the upper tidal river and the area of the Chesapeake and Delaware Canal (C&D Canal) (rkm 95) or farther downstream. Movements were often rapid, with one fish swimming 121 kilometers in six days. The long distance excursions often occurred in spring, after the spawning period (likely movement to summer foraging areas), and in early to mid-winter (likely moving to overwintering areas) (ERC, Inc. 2006a). Most of the tagged shortnose sturgeon occupied known overwintering areas in the Roebling, Bordentown and Trenton reaches of the upper tidal river during December through March. Three fish, however, appear to have overwintered in the downriver, below Wilmington (rkm 113). This suggests the existence of an overwintering area in the lower river. Downriver overwintering areas are known to occur in other river systems, but previously there had been no evidence of such in the Delaware River (ERC, Inc. 2006a). Movement patterns observed in the ERC study indicate some, but not all, of the adult shortnose sturgeon overwintering in the upper tidal Delaware River move to the spawning area in the lower non-tidal river in late March and April (ERC, Inc. 2006a).

Preliminary tracking studies of juvenile shortnose sturgeon showed different patterns of movements in the winter (n=3), indicating that the entire lower Delaware River (Philadelphia to below Artificial Island; approx. rkm 161-79) may be utilized for overwintering (ERC, Inc. 2007b). One fish, whose tag was active in late spring and summer, showed movement spanning approximately 25 kilometers between Chester and Deepwater Point ranges (rkm 130-101), spending much of its time in the vicinity of Marcus Hook (rkm 128; ERC, Inc. 2007b).

VI. ENVIRONMENTAL BASELINE

By regulation, environmental baselines for biological opinions include the past and present impacts of all state, Federal or private actions and other human activities in the action area, the anticipated impacts of all proposed Federal projects in the action area that have already undergone formal or early section 7 consultation, and the impact of State or private actions which are contemporaneous with the consultation in process (50 CFR '402.02). The environmental baseline for this Opinion includes the effects of several activities that affect the survival and recovery of the listed species in the action area. The following information summarizes the primary human and natural phenomena in the Delaware River that are believed to affect the status and trends of endangered shortnose sturgeon and the probable responses of the sturgeon to these phenomena.

Dams and Water Diversion. Dams are used to impound water for water resource projects such as hydropower generation, irrigation, navigation, flood control, industrial and municipal water supply, and recreation. Dams can have profound effects on diadromous fish species by fragmenting populations, eliminating or impeding access to historic habitat, modifying free-flowing rivers to reservoirs and altering downstream flows and water

temperatures. Direct physical damage and mortality can occur to diadromous fish that migrate through the turbines of traditional hydropower facilities or as they attempt to move upstream using fish passage devices. The construction of dams throughout the shortnose sturgeon's range is probably the main factor reducing their reproductive success which, in turn, could be the primary reason for the reduction in population size for shortnose sturgeon.

Although there are dams located on other rivers where other shortnose sturgeon populations are found, the Delaware River is the longest undammed river east of the Mississippi (DRBC 2009). This is due, in large part, to the National Wild and Scenic (16 U.S.C. 1271 *et seq.*) designations of portions of the river. Historically, dams have been proposed for the Delaware River. Tocks Island Dam was a huge multi-purpose reservoir project proposed for the Delaware River six miles upstream of the Delaware Water Gap. The dam would have created a 40-mile long lake with depths up to 140 feet. Almost 250 billion gallons of water were to be stored behind the dam with ample "dry storage" for floodwaters. The project was to be the U.S. Army Corps of Engineers' eighth largest U.S. dam project and its largest east of the Mississippi River. Today, there are various wing dams currently located on the Delaware, but no dams on the scale of having turbines and hydropower facilities. However, there are dams located among the Delaware River's tributaries. Located on the upper west branch of the Delaware River, the Cannonsville Reservoir exists as an impounding reservoir to supply 50% of the drinking water to New York, New York.

Bycatch. Directed harvest of shortnose sturgeon is prohibited. In 1998, the Atlantic States Marine Fisheries Commission (ASMFC) imposed a coast-wide fishing moratorium on Atlantic sturgeon until 20 year classes of adult females could be established (ASMFC 1998). NMFS followed this action by closing the Exclusive Economic Zone (EEZ) to Atlantic sturgeon take in 1999. Shortnose sturgeon has likely benefitted from this closure as any bycatch in the fishery targeting Atlantic sturgeon has been eliminated.

Although directed harvest of shortnose sturgeon has been prohibited since 1967, bycatch of this species has been documented in other fisheries throughout its range. Adults are believed to be especially vulnerable to fishing gears for other anadromous species (such as shad, striped bass and herring) during times of extensive migration – particularly the spawning migration upstream, followed by movement back downstream (Litwiler 2001). The shad fishery on the Altamaha River was reported to have a 2.3% shortnose sturgeon mortality rate from 2007-2009 (Bahn and Peterson 2009). Additionally, bycatch in the southern trawl fishery for shrimp *Penaeus* spp. was eliminated at 8% in one study (Collins *et al.* 1996).

The 1998 Recovery Plan for shortnose sturgeon lists commercial and recreational shad fisheries as a source of shortnose bycatch. Although shortnose sturgeon are primarily captured in gill nets, they have also been documented in the following gears: pound nets, fyke/hoop nets, catfish traps, shrimp trawls, and hook and line fisheries (recreational).

Bycatch in the gill net fisheries can be quite substantial and is believed to be a significant threat to the species. The catch rates in drift gill nets are believed to be lower than for fixed nets; longer soak times of the fixed nets appear to be correlated with higher rates of mortalities. In an

American shad gill net fishery in South Carolina, of 51 fish caught, 16% were bycatch mortality and another 20% of the fish were visibly injured (Collins *et al.* 1996).

Poaching. There is evidence of shortnose sturgeon targeted by poachers throughout their range, and particularly where they appear in abundance (such as on spawning grounds) but the extent of the poaching is difficult to assess (Dadswell 1979, Dovel *et al.* 1992, Collins *et al.* 1996). There have been several documented cases of shortnose sturgeon caught by recreational anglers. One shortnose sturgeon illegally taken on the Delaware River was documented by a NJ DFW conservation officer in Trenton New Jersey (NJCOA 2006). Additionally, citations have been issued for illegal recreational fishing of shortnose in the vicinity of Troy, New York on the Hudson River and on the Cooper River in South Carolina.

Poaching has also been documented for other sturgeon species in the United States. Cohen (1997) documented poaching of Columbia River white sturgeon sold to buyers on the U.S. east coast. Poaching of Atlantic sturgeon has also been documented by law enforcement agencies in Virginia, South Carolina, and New York, and is considered a potentially significant threat to the species, but the present extent and magnitude is largely unknown (ASPRT 2008).

Dredging. Many rivers and estuaries are periodically dredged for flood control or to support commercial and recreational boating. Dredging also aids in construction of infrastructure and in marine mining. Dredging may have adverse impacts on aquatic ecosystems including direct removal/burial of organisms, turbidity, contaminant resuspension, noise/disturbance, alterations due to hydrodynamic regime and physical habitat and actual loss of riparian habitat (Chytalo 1996, Winger *et al.* 2000).

Dredges are generally either mechanical or hydraulic. Mechanical dredges are used to scoop or grab bottom substrate and are capable of removing hard-packed materials and debris. Mechanical dredge types are clamshell buckets, endless bucket conveyor, or single backhoe or scoop bucket types, however, these dredge types often have difficulty retaining fine materials in the buckets and do not dredge continuously. Material excavated from mechanical dredging is often loaded onto barges for transport to a designated placement site (USACOE 2008).

Hydraulic dredges are used principally to dredge silt, sand and small gravel. Hydraulic dredges include cutterhead pipeline dredges and self-propelled hopper dredges. Hydraulic dredges remove material from the bottom by suction, producing slurry of dredged material and water, either pumped directly to a placement site, or in the case of a hopper dredge, into a hopper and later transported to a dredge spoil site. Cutterhead pipeline dredges can excavate most materials including some rock without blasting and can dredge almost continuously (USACOE 2008).

The impacts of dredging operations on sturgeon are often difficult to assess. Hydraulic dredges can lethally take sturgeon by entraining sturgeon in dredge drag arms and impeller pumps (NMFS 1998). Mechanical dredges have also been documented to lethally take shortnose sturgeon (Dickerson 2006). In addition to direct effects, indirect effects from either mechanical or hydraulic dredging include destruction of benthic feeding areas, disruption of spawning migrations, and deposition of resuspended fine sediments in spawning habitat (NMFS 1998).

Another critical impact of dredging is the encroachment of low D.O. and high salinities upriver after channelization (Collins *et al*. 2001). Adult shortnose sturgeon can tolerate at least short periods of low D.O. and high salinities, but juveniles are less tolerant of these conditions in laboratory studies. Collins *et al.* (2001) concluded harbor modifications in the lower Savannah River have altered hydrographic conditions for juvenile sturgeon by extending high salinities and low D.O. upriver.

In addition to the impacts of dredging noted above, Smith and Clugston (1997) reported that dredging and filling eliminates deep holes, and alters rock substrates. Nellis *et al*. (2007) documented that dredge spoil drifted 12 km downstream over a 10 year period in the Saint Lawrence River, and that those spoils have significantly less macrobenthic biomass compared to control sites. Using an acoustic trawl survey, researchers found that Atlantic and lake sturgeon were substrate dependent and avoided spoil dumping grounds (McQuinn and Nellis 2007). Similarly, Hatin *et al.* (2007) tested whether dredging operations affected Atlantic sturgeon behavior by comparing CPUE before and after dredging events in 1999 and 2000. The authors documented a three to seven-fold reduction in Atlantic sturgeon presence after dredging operations began, indicating that sturgeon avoid these areas during operations.

Current, Ongoing Dredging Projects in the Delaware River. The Delaware River is a vital commercial and recreational waterway. It is navigable by large, oceangoing vessels as far inland as Philadelphia, Pennsylvania, and by smaller vessels to Trenton, New Jersey. The Chesapeake and Delaware Canal is navigable by oceangoing vessels, connecting the Delaware River below Wilmington, Delaware, with the Chesapeake Bay. The Delaware River Basin Commission, the federal government, and the four Delaware Basin states-New York, Pennsylvania, New Jersey, and Delaware-jointly manage assets and concerns of the Basin. The U.S. Army, Corps of Engineers has responsibility for maintaining navigation on the river and has historically dredged the Delaware River's federal shipping channel since the late 1800s when the controlling depth of the Delaware River was 18 feet (USACOE 2009).

Current and ongoing channelization plans for the Delaware River Main Channel include appropriated construction funds to deepen and maintain the existing shipping channel from 40 feet to 45 feet from Philadelphia Harbor, Pennsylvania and Beckett Street Terminal, Camden, New Jersey to the mouth of the Delaware Bay. Although this is a total distance of 165 kilometers, the lower portion of the river (53 km), mostly in the Delaware Bay, is already at 45 feet or deeper (USACOE 2009). A Biological Opinion has been completed on the Delaware River Main Channel dredging project, which concluded that the dredging project is likely to adversely affect but is not likely to jeopardize the continued existence of shortnose sturgeon.

The existing authorized widths in the straight portions of the channel, ranging from 400 feet in Philadelphia to 1,000 feet in the bay, would not change. However, 12 of the existing 16 bends in the channel would be widened for safer navigation. In addition, the Marcus Hook area, within the proposed action area of the sturgeon research, would be deepened to 45 feet. Approximately 16.4 million cubic yards of material must be removed to deepen the channel during initial construction phases of the project. Of that amount, approximately 12.3 million cubic yards of sand, silt, and clay would be taken from the river portion of the project – the area from Philadelphia, Pennsylvania, and Camden, New Jersey, to the Upper Delaware Bay. About

77,000 cubic yards of rock would also be removed from the Marcus Hook area of the river. The bulk of dredging will be performed by hopper and hydraulic pipeline dredges with a bucket dredge used for rock removal in the Marcus Hook area (USACOE 2009).

Blasting. Bridge demolition, dredge, and other projects may include plans for blasting with powerful explosives. For example, the dredging of the Delaware River referenced above includes blasting. Fish are particularly susceptible to the effects of underwater explosions and are killed over a greater range than other organisms (Lewis 1996). Unless appropriate precautions are made to mitigate the potentially harmful effects of shock wave transmission to physostomous (i.e., air-bladder connected to the gut) fish like shortnose sturgeon, internal damage and/or death may result (NMFS 1998). A study testing the effects of underwater blasting on juvenile shortnose sturgeon and striped bass was conducted in Wilmington Harbor, NC in December of 1998 and January of 1999 (Moser 1999). There were seven test runs that included 23-33 blasts (3 rows with 10-11 blast holes per row and each hole 10 ft apart) with about 24-28 kg explosives per hole. For each blast 50 hatchery reared shortnose sturgeon and striped bass were placed in cages three feet from the bottom at distances of 35, 70, 140, 280 and 560 feet upstream and downstream of the blast area. A control group of 200 fish was held 0.5 miles from the blast site (Moser 1999). Test blasting was conducted with (3) and without (4) an air curtain placed 50 ft from the blast area. External assessments of impacts to the caged fish were conducted immediately after the blasts and 24 hours after the blasts. After the 24 hour period, a subsample of the caged fish, primarily from those cages nearest the blast at 35 feet and some from 70 feet, were sacrificed for necropsy.

Shortnose sturgeon selected for necropsy all appeared to be in good condition externally and behaviorally. Results of the tests, including necropsies, indicated the fish that had survived the blast, lived through the 24 hour observation period, and appeared outwardly fine. However, they may have had substantial internal injuries. Moser concluded that many of the injuries would have resulted in eventual mortality (Moser 1999). The necropsy results also indicated in the fish held in cages at 70 feet were less seriously injured by test blasting than those held at 35 feet from the blast. Finally, shortnose sturgeon juveniles suffered fewer, less severe internal injuries than juvenile striped bass tested, and there appeared to be no reduction of injury in fish experiencing blasts while the air curtain was in place (Moser 1999).

Bridge Construction/Demolition. Bridge construction and demolition projects may interfere with normal shortnose sturgeon migratory movements and disturb areas of sturgeon concentrations. Bridge demolition projects may include plans for blasting piers with powerful explosives. Unless appropriate precautions are made to mitigate the potentially harmful effects of shock wave transmission to physostomous (i.e., airbladder connected to the gut) fish like shortnose sturgeon, internal damage and/or death may result. From 1993 through 1994, NMFS consulted with the Federal Highway Administration to assess the potential impacts of demolishing bridge piers to shortnose sturgeon. NMFS advised the Federal Highway Administration to employ several conservation measures designed to minimize the transmission of harmful shock waves. These measures included restricting the work to seasonal "work windows," installing double-walled cofferdams around each pier to be blasted, and dewatering the outer cofferdams. The use of an air gap (e.g., double-wall cofferdam, bubble screen) to attenuate shock waves is likely to reduce adverse effects to shortnose sturgeon and other

swimbladder fish (Sonolysts 1994). Blast pressures below which negative impacts to shortnose sturgeon are unlikely to occur are not known. Wright (1982) determined that detonations producing instantaneous pressure changes greater than 100kPa (14.5psi) in the swimbladder of a fish will cause serious injury or death.

Water Quality and Contaminants. The quality of water in river/estuary systems is affected by human activities conducted in the riparian zone and those conducted more remotely in the upland portion of the watershed. Industrial activities can result in discharge of pollutants, changes in water temperature and levels of D.O., and the addition of nutrients. In addition, forestry and agricultural practices can result in erosion, run-off of fertilizers, herbicides, insecticides or other chemicals, nutrient enrichment, and alteration of water flow. Coastal and riparian areas are also heavily impacted by real estate development and urbanization resulting in storm water discharges, non-point source pollution, and erosion. The Clean Water Act regulates pollution discharges into waters of the United States from point sources, however, it does not regulate non-point source pollution.

The water quality over the range of shortnose sturgeon varies by watershed but is notably poorer in the north than in the south. The U.S. Environmental Protection Agency (EPA) published its third edition of the National Coastal Condition Report (NCCR III) in 2008, a "report card" summarizing the status of coastal environments along the coast of the United States (USEPA 2008; See Table 8 below). The report analyzes water quality, sediment, coastal habitat, benthos, and fish contaminant indices to determine status. The northeast region of the U.S. (Virginia to Maine) received an overall rating of "fair to poor." The southeast region of the U.S. (Florida to North Carolina) received an overall rating of "fair," the best rating in the nation. Areas of concern having poor index scores for the Delaware River were water quality and tissue contaminants.

Table 8. Summary of the EPA NCCR III for the U.S. east coast published by the EPA (2008) grading coastal environments. (Northeast region=VA to ME; southeast region=FL to NC)

	Region	
Status Index	**Northeast**	**Southeast**
Water quality	Fair	Fair
Sediment	Fair to poor	Fair
Coastal Habitat	Good to fair	Fair
Benthos	Poor	Good
Fish Tissue	Poor	Good to fair
Overall	**Fair to poor**	**Fair**

Chemicals such as chlordane, dichlorodiphenyl dichloroethylene (DDE), DDT, dieldrin, PCBs, cadmium, mercury, and selenium settle to the river bottom and are later consumed by benthic feeders, such as macroinvertebrates, and then work their way higher into the food web (e.g., to sturgeon). Some of these compounds may affect physiological processes and impede a fish's ability to withstand stress, while simultaneously increasing the stress of the surrounding

environment by reducing DO, altering pH, and altering other physical properties of the water body.

Life history of shortnose sturgeon (i.e., long lifespan, extended residence in estuarine habitats, benthic foraging) predispose sturgeon to long-term, repeated exposure to environmental contamination and potential bioaccumulation of heavy metals and other toxicants (Dadswell 1979, NMFS 1998). However, there has been little work on the effects of contaminants on shortnose sturgeon to date. Shortnose sturgeon collected from the Delaware and Kennebec Rivers had total toxicity equivalent concentrations of polychlorinated dibenzo-p-dioxins (PCDDs), polychlorinated dibenzofurans (PCDFs), PCBs, DDE, aluminum, cadmium, and copper above adverse effect concentration levels reported in the literature (ERC 2002, 2003).

Heavy metals and organochlorine compounds accumulate in sturgeon tissue, but their long-term effects are not known (Ruelle and Henry 1992, Ruelle and Keenlyne 1993). High levels of contaminants, including chlorinated hydrocarbons, in several other fish species are associated with reproductive impairment (Cameron *et al*. 1992, Longwell *et al*. 1992, Hammerschmidt *et al.* 2002, Giesy *et al.* 1986, Mac and Edsall 1991, Matta *et al*. 1998, Billsson *et al.* 1998), reduced survival of larval fish (Berlin *et al.* 1981, Giesy *et al*. 1986), delayed maturity (Jorgensen *et al.* 2003) and posterior malformations (Billsson e*t al.* 1998). Pesticide exposure in fish may affect anti-predator and homing behavior, reproductive function, physiological maturity, swimming speed, and distance (Beauvais *et al*. 2000, Scholz *et al*. 2000, Moore and Waring 2001, Waring and Moore 2004).

Sensitivity to environmental contaminants also varies by life stage. Early life stages of fish appear to be more susceptible to environmental and pollutant stress than older life stages (Rosenthal and Alderdice 1976). Dwyer *et al*. (2005) compared the relative sensitivities of common surrogate species used in contaminant studies to 17 listed species including shortnose and Atlantic sturgeons. The study examined 96-hour acute water exposures using early life stages where mortality is an endpoint. Chemicals tested were carbaryl, copper, 4-nonphenol, pentachlorophenal (PCP) and permethrin. Of the listed species, Atlantic and shortnose sturgeon were ranked the two most sensitive species tested (Dwyer *et al.* 2005). Additionally, a study examining the effects of coal tar, a byproduct of the process of destructive distillation of bituminous coal, indicated that components of coal tar are toxic to shortnose sturgeon embryos and larvae in whole sediment flow-through and coal tar elutriate static renewal (Kocan *et al.* 1993).

Land Use Practices. In all, the Delaware River basin contains 13,539 square miles, draining parts of Pennsylvania (6,422 square miles or 50.3 percent of the basin's total land area); New Jersey (2,969 square miles, or 23.3%); New York (2,362 square miles, 18.5%); and Delaware (1,004 square miles, 7.9%) (DRBC 2009). Included in the total area number is the 782 square-mile Delaware Bay, which lies roughly half in New Jersey and half in Delaware.

The Delaware River Basin Commission published the first *State of the Delaware River Basin Report* in December 2008 which stated the Delaware River basin population as of 2000 to be 7,758,675 and it is expected to approach 9 million by 2030. The Delaware River watershed is primarily divided between developed, agriculture, forest, wetlands and water, and "other" including mining uses (DRBC 2008). The major rivers draining into the Delaware are the

Lehigh and Schuylkill Rivers. The most heavily urbanized areas are at the lower extent of the watershed region, where large industrialized cities such as Philadelphia, Pennsylvania, Wilmington, Delaware, Camden, New Jersey, and Trenton, New Jersey are found.

Power Plant Operations. Shortnose sturgeon are susceptible to impingement on cooling water intake screens at power plants. Electric power and nuclear power generating plants can affect sturgeon by impinging larger fish on cooling water intake screens and entraining larval fish. The operation of power plants can have unforeseen and extremely detrimental impacts to water quality which can affect shortnose sturgeon.

Public Service Enterprise Group Nuclear operates two nuclear power plants pursuant to licenses issued by the U.S. Nuclear Regulatory Commission (NRC). These facilities are the Salem and Hope Creek Generating Stations (Salem and HCGS), which are located on adjacent sites within a 740-acre parcel of property at the southern end of Artificial Island in Lower Alloways Creek Township, Salem County, New Jersey. Consultation pursuant to Section 7 of the ESA between NRC and NMFS on the effects of the operation of these facilities has been ongoing since 1979. An Opinion was issued by NMFS in April 1980 in which NMFS concluded that the ongoing operation of the facilities was not likely to jeopardize the continued existence of shortnose sturgeon. Consultation was reinitiated in 1988 due to the documentation of impingement of sea turtles at the Salem facility. An Opinion was issued on January 2, 1991 in which NMFS concluded that the ongoing operation was not likely to jeopardize shortnose sturgeon, Kemp's ridley, green, or loggerhead sea turtles. Consultation was reinitiated in 1992 and a new Opinion was issued on August 4, 1992 and again on May 14, 1993. In 1998 the NRC requested that NMFS modify the Reasonable and Prudent Measures and Terms and Conditions of the ITS, and, specifically, remove a sea turtle study requirement. NMFS responded to this request in a letter dated January 21, 1999 and also with a revised ITS which served to amend the May 14, 1993 Opinion. The 1999 ITS exempts the annual take (capture at intake with injury or mortality) of 5 shortnose sturgeon, 30 loggerhead sea turtles, 5 green sea turtles, and 5 Kemp's ridleys. Since monitoring of the intakes was initiated in 1978, 18 shortnose sturgeon have been recovered from the Salem intakes which are located in Delaware Bay. No shortnose sturgeon or have been observed at the HCGS intakes.

Research. Research activities could also pose a threat to shortnose sturgeon. Excluding the proposed permit detailed in this Opinion, there are 15 permits (Table 9) authorizing take of shortnose sturgeon on the east coast of the United States. All of the permits authorize the sampling of adult or juvenile shortnose sturgeon. There is one permit currently authorizing shortnose sturgeon research on the Delaware River (permit 1486) which applicants were currently operating under until its expiration on 1/31/10.

Table 9. Existing shortnose sturgeon research permits similar to the proposed action.			
Permit No.	**Location**	**Authorized Take**	**Research Activity**
10115 Expires: 8/3/2013	Saltilla & Saint Marys Rivers, GA & FL	85 adult/juv 20 ELS	Capture, handle, measure, weigh, PIT tag, tissue sample, collect ELS
14394 Expires: 9/30/14	Altamaha River and Estuary, GA	500 adult/juv. (1 lethal), 100 ELS	Capture, handle, weigh, measure, PIT tag, transmitter tag, tissue sample, anesthetize, laparoscopy, blood collection, fin ray section, collect ELS
10037 Expires: 4/30/2013	Ogeechee River and Estuary, GA	150 adult/juv. (2 lethal), 40 ELS	Capture, handle, measure, weigh, PIT tag, tissue sample, fin-ray section, anesthetize, laparoscopy, blood collection, radio tag, collect ELS
1447 Expires: 2/28/2012	S. Carolina Rivers and Estuaries	100 adult/juv. (2 lethal), 100 ELS	Capture, handle, measure, weigh, PIT and DART tag, transmitter tag, anesthetize, tissue sample, gastric lavage, collect ELS
1505 Expires: 5/15/2011	S. Carolina Rivers and Estuaries	98 adult/juv. (2 lethal), 200 ELS	Capture, handle, measure, weigh, PIT and DART tag, transmitter tag, anesthetize, laparoscopy, blood collection, tissue sample, gastric lavage, collect ELS
1542 Expires: 7/31/2011	Upper Santee River Basin, SC	5 adult/juv.; 100 ELS	Capture, handle, weigh, measure, PIT and dart tag, tissue sample, ELS collection
1543 Expires:11/30/2011	Upper Santee River Basin, SC	3 adult/juv.	Capture, handle, weigh, measure, tissue sample
1486 * Expires: 1/31/2010	**Delaware River and Estuary NJ & DE**	**1,750 adult/juv. (10 lethal), 1000 ELS**	**Capture, handle, measure, weigh, Floy & T-bar tag, PIT tag, tissue sample, anesthetize, ultrasonic tag, laparoscopy, blood collection, collect ELS**
1547 Expires:10/31/2011	Hudson River, (Haverstraw & Newburgh), NY	500 adults/juv.	Capture, handle, weigh, measure, PIT & Carlin tag, tissue sample
1575 Expires11/30/2011	Hudson River (Tappan-Zee), NY	250 adult/juv.	Capture, handle, measure
1580 Expires: 3/31/2012	Hudson River and Estuary, NY	82 adult/juv.; 40 ELS	Capture, handle, measure, weigh, PIT tag, Carlin tag, photograph, tissue sample, collect ELS
1449 Expires: 3/31/2010	Upper Conn. River, MA	80 adult/juv.; 200 ELS	Capture, handle, measure, weigh, PIT tag, external radio tag, collect ELS
1549 Expires: 1/31/2012	Upper Conn. River, MA	673 adult/juv (5 lethal), 1,430 ELS from East Coast rivers	Capture, handle, measure, weigh, anesthetize, PIT tag, TIRIS tag, radio tag, temperature/depth tag, tissue sample, borescope, laboratory tests, photographs, collect ELS
1516 Expires: 5/15/2011	Lower Conn. River & Estuary., CT	500 adult/juv (2 lethal); 300 ELS	Capture, handle, measure, weigh, PIT tag, sonic/radio tag, gastric lavage, fin ray section, collect ELS
1578 Expires: 11/30/2011	Kennebec River and Estuary, ME	500 adult/juv.; 30 ELS	Capture, handle, measure, weigh, tissue sample, PIT tag, acoustic tag, anesthetize, collect ELS
1595-03 Expires: 3/31/2012	Penobscot River and Estuary, ME	200 adult/juv. (2 lethal); 50 ELS	Capture, handle, measure, weigh, borescope, photograph, tissue sample, blood sample, Carlin tag, PIT tag, anesthetize, transmitter tag, collect ELS

* Applicant's current permit in the Delaware River expiring January 31, 2010.
ELS = early life stages

Conservation. The Delaware River Basin Commission was established in 1961 through concurrent compact legislation into law which created a regional body with the force of law to oversee management of the Delaware River system. Members include the four basin state governors and the Division Engineer, North Atlantic Division, U.S. Army Corps of Engineers, who serves as the federal representative. Commission programs include water quality protection, water supply allocation, regulatory review (permitting), watershed planning, drought management, flood loss reduction, and recreation. Furthermore, the Delaware River Basin Commission has embarked on a water conservation program which adopts policies to reduce the demand for water.

The National Wild and Scenic Rivers System was created by Congress in 1968 to preserve certain rivers with outstanding natural, cultural, and recreational values in a free-flowing condition for the enjoyment of present and future generations. Portions of the upper, middle, and lower Delaware River are part of the National Wild and Scenic Rivers System. This designation is significant, because it keeps the Delaware free of large dams and hydroelectric projects. The upper portion flows between Hancock and Sparrow Bush, New York, along the Pennsylvania border and stretches for 73 miles. The middle portion flows for 35 miles through the Delaware Water Gap National Recreation Area and cuts an "S" curve through Kittatinny Ridge. The lower portion contains several segments of the Delaware River and its tributaries: 1) from river mile 193.8 to the northern border of the city of Easton, Pennsylvania; 2) from just south of the Gilbert Generating Station to just north of the Point Pleasant Pumping Station; 3) from just south of the Point Pleasant Pumping Station to a point 1,000 feet north of the Route 202 Bridge; 4) from 1,750 feet south of the Route 202 Bridge to the southern boundary of the town of New Hope, Pennsylvania, to the town of Washington Crossing, Pennsylvania; 5) all of Tinicum Creek; 6) Tohickon Creek from the Lake Nockamixon Dam to the Delaware River; and 7) Paunacussing Creek in Solebury Township.

Section 303(d) of the Federal Clean Water Act (CWA) requires States to develop a list (303(d) List) of waterbodies for which existing pollution control activities are not sufficient to attain applicable water quality standards and to develop Total Maximum Daily Loads (TMDLs) for pollutants of concern. A TMDL sets a limit on the amount of a pollutant that can be discharged into a waterbody such that water quality standards are met. The states of Delaware, Pennsylvania, New Jersey, and New York are responsible for implementing TMDLs for the Delaware River.

All of the states along the Delaware River – Pennsylvania, New Jersey, New York, and Delaware – each have State Departments of Conservation managing programs which impact the Delaware River Basin such as air, waste, soil, water, fish, and wildlife. The Delaware Department of Natural Resources, Division of Fish and Wildlife conducts biological surveys and studies of living resources throughout the state, manages approximately 60,000 acres including ponds, wildlife and water access areas and facilities for public use and enjoyment, and improves the public's understanding and interest in the state's fish and wildlife resources through information and outreach programs. The Pennsylvania Department of Conservation and Natural Resources (PDCNR) also manages many conservation programs. The Pennsylvania Rivers Conservation Program was developed by PDCNR to conserve and enhance river resources through preparation and accomplishment of locally initiated plans. The program provides technical and financial

assistance to municipalities and river support groups to carry out planning, implementation, acquisition and development activities. The Pennsylvania Natural Heritage Program is a member of NatureServe, an international network of natural heritage programs that gather and provide information on the location and status of important ecological resources, including threatened and endangered species. The Natural Resources and Conservation Service of New Jersey has a conservation stewardship program, awards multiple conservation grants, leads a wetlands reserve program and a wildlife habitat incentives program. The New York Department of Environmental Conservation has an Endangered Species Program, State Wildlife Grants Program, and a Natural Heritage Program.

Integration of the Environmental Baseline. The above activities along the Delaware River pose threats to its shortnose sturgeon population in the following ways. Many activities cause *death* – definite removal of individual fish from the Delaware River population – at the adult, juvenile, and larval stages. Other activities cause *injury* to shortnose, increasing stress levels and decreasing their survival potential. Still, other activities *alter habitat*, potentially changing spawning and survival patterns of these fish.

Delaware River activities potentially causing death to individual shortnose sturgeon are bycatch in commercial and recreational fishing, cooling water intakes and power plants, dredging, blasting, bridge construction, and research. De Vries (2006) hypothesized that, on the Delaware, mortality between shortnose juvenile and adult stages is unusually high possibly as a result of incidental mortality associated with the commercial shad fishery and the simultaneous spawning migration. Hydroelectric or nuclear power plants must use rivers or lakes as sources of running turbines or as cooling mechanisms. Adult and larval shortnose sturgeon are known to be killed or impinged on the screens that cover the cooling water intake screens (Hoff and Klauda 1979, Dadswell *et al.* 1984, NMFS 1993). Dadswell *et al.* (1984) reported that larval and juvenile shortnose sturgeon in the different populations along the Atlantic have been killed after being impinged on the intake screens or entrained in the intake structures of power plants on the Delaware, Hudson, Connecticut, Savannah and Santee rivers. During dredging activities, hydraulic dredges can kill sturgeon by entraining sturgeon in dredge dragarms and impeller pumps. Mechanical dredges have also been documented to kill shortnose sturgeon. Finally, research permits, such as the one being considered in this Opinion, authorize the sometimes lethal take of shortnose sturgeon early life stages on the Delaware.

All of the Delaware River activities identified in the Environmental Baseline section have the potential to injure individual shortnose sturgeon. Commercial and recreational fishing industries that catch shortnose incidentally might return living fish to the river, presumably unharmed, however each fish might have sustained injury in the process. The operation of power plants can also have unforeseen and detrimental impacts to water quality which can injure shortnose sturgeon.

Water quality changes from dredging, shipping, land use practices, point and non-point source pollution could also injure shortnose sturgeon by way of changes in DO concentration or introduction of waterborne contaminants. DO concentrations can be affected by maintenance dredging of Federal navigation channels and other waters. Apart from entrainment, dredging can also change DO and salinity gradients in, and around, the channels (Jenkins *et al.* 1993,

Campbell and Goodman 2004, Secor and Niklitschek 2001). Dredging operations may pose risks to shortnose sturgeon by destroying or adversely modifying their benthic feeding areas, disrupting spawning migrations, and filling spawning habitat with resuspended fine sediments. Since shortnose sturgeon are benthic omnivores, the modification of the benthos could affect the quality, quantity, and availability of sturgeon prey species.

Along with fluctuations in the DO and salinity concentrations, other waterborne contaminants may affect the aquatic environment, causing injury to shortnose sturgeon. These contaminants may come from land use practices, or point and non-point source pollution. Issues such as raised fecal coliform and estradiol concentrations affect all of the wildlife using the river as a habitat. The impact of many of these waterborne contaminants on shortnose sturgeon is unknown, but they are known to affect other species of fish in rivers and streams. These compounds may enter the aquatic environment via wastewater treatment plants, agricultural facilities, as well as runoff from farms (Folmar *et al.* 1996, Culp *et al.* 2000, Wildhaber *et al.* 2000, Wallin *et al.* 2002). For instance, estrogenic compounds are known to affect the male-female sex ratio in streams and rivers via decreased gonadal development, physical feminization, and sex reversal (Folmar *et al.* 1996). Although the effects of these contaminants are unknown in shortnose sturgeon, Omoto *et al.* (2002) found that by varying the oral doses of estradiol-17β or 17α-methyltestosterone given to captive hybrid (*Huso huso* female × *Acipenser ruthenus* male) "bester" sturgeon they could induce abnormal ovarian development or a lack of masculinization. These compounds, along with high or low DO concentrations, can result in sub-lethal effects that may have long-term consequences for small populations.

Other NMFS-permitted research activities could also injure shortnose sturgeon in the Delaware River. The applicant is currently operating under an existing permit, permit 1486, in which they are authorized to capture, handle, weigh, measure, passive integrated transponder (PIT) tag, tissue sample, anesthetize, laparoscopy, collect blood, and collect early life stages (ELS). Permit 1486 also authorizes lethal take of ten shortnose sturgeon. Thus far, mortality as a result of this permit has not been reported. However, fish captured may have been injured in a way that is not quantified. There are no other research permits on the Delaware River at this time.

Delaware River activities potentially altering the habitat of shortnose sturgeon are dredging and land use activities. Due to their benthic nature, dredging for shipping and other activities destroys shortnose feeding areas, disrupts spawning migrations, and fills spawning habitat with resuspended fine sediments. Land use activities also have the capacity to fill spawning habitat with sediments if those activities release sand and silt into the Delaware River.

In conclusion, the Delaware River adult shortnose sturgeon population is estimated at 12,796 (95% confidence interval – 10,228 to 16,367) based on mark recapture data collected during 1981-1984 (Hastings *et al*. 1987). A later study estimated the population at 12,047 – 13,580 (ERC, Inc. 2006b). Similarity between the two estimates suggests that the Delaware River shortnose sturgeon population is stable but has not increased in the 20+ years between studies. The current impact of human activities and recovery of shortnose sturgeon in the Delaware River population is somewhat uncertain.

VII. Effects of the Proposed Action

Pursuant to Section 7(a)(2) of the ESA, federal agencies are directed to ensure that their activities are not likely to jeopardize the continued existence of any listed species or result in the destruction or adverse modification of critical habitat. The proposed activities authorized by permit 14604 would expose shortnose sturgeon to capture, handling, genetic tissue sampling, PIT and Floy tags, genetic tissue sampling, sonic transmitter implantation, anesthesia, laparoscopy, hydroacoustic testing, blood collection, and possible biopsy. In this section, we describe the potential physical, chemical, or biotic stressors associated with the proposed action, the probability of individuals of listed species being exposed to these stressors based on the best scientific and commercial evidence available, and the probable responses of those individuals (given probable exposures) based on the available evidence. As described in the *Approach to the Assessment* section, for any responses that would be expected to reduce an individual's fitness (i.e., growth, survival, annual reproductive success, and lifetime reproductive success), the assessment would consider the risk posed to the viability of the population(s) those individuals comprise and to the listed species those populations represent. The purpose of this assessment is to determine if it is reasonable to expect the proposed studies to have effects on listed species that could appreciably reduce their likelihood of surviving and recovering in the wild.

A. Potential Stressors

The assessment for this consultation identified thirteen possible stressors associated with the proposed permitted activities. These include: 1) capture by gill, trammel, and trawl net; 2) handling for procedures and measurements; 3) PIT tagging; 4) Floy tagging; 5) genetic tissue sampling; 6) anesthesia for implantation/surgery; 7) transmitter implantation/surgery; 8) anesthesia for laparoscopy; 9) laparoscopy; 10) collection of ELS; 11) biopsy procedure; 12) hydroacoustic testing; and 13) blood collection. All captured shortnose sturgeon would be handled, weighed, measured, photographed, PIT tagged, Floy tagged, and genetic tissue sampled. Smaller subsets of these fish could receive some combination of transmitter implantation, laparoscopy, biopsy, hydroacoustic testing (*see* Table 1). Activities are expected to occur in the Delaware River until the permit's expiration. Based on a review of available information, we determined that all fourteen of the possible stressors could pose a risk to shortnose sturgeon. Accordingly, the effects analysis of this consultation focused on all of the potential stressors.

B. Exposure Analysis

Exposure analyses identify the co-occurrence of ESA-listed species with the actions' effects in space and time, and identify the nature of that co-occurrence. The Exposure Analysis identifies, as possible, the number, age or life stage, and gender of the individuals likely to be exposed to the actions' effects and the population(s) or subpopulation(s) those individuals represent.

Table 10 identifies the numbers of shortnose sturgeon that are expected to be exposed annually for five years in the Delaware River under the proposed permit 14604. All captured shortnose sturgeon would be handled, weighed, measured, PIT and Floy tagged, and genetic tissue sampled. Smaller subsets of these fish would be anesthetized for sonic transmitter implantation

or some combination of laparoscopy, blood sample, and biopsy. One incidental mortality (or serious harm) would be authorized per year not to exceed three throughout the permit's duration.

Table 10. Activities proposed to be authorized for ERC's research on endangered shortnose sturgeon (*Acipenser brevirostrum*) in the Delaware River under Permit 14604.

Species	Life Stage	Sex	Expected Annual Take	Take Action	Location
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile & adult	Male & female	Up to 896 annually; total of 3,600 over 5yrs	Capture, hold, measure, weigh, photograph, scan (for tags), Floy T-bar tag, PIT tag, & tissue sample	Delaware River (netting area=rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 30 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222) & implant acoustic tag	Delaware River (netting area=rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile	Male & female	Up to 30 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222) & implant acoustic tag,	Delaware River (netting area=rkm 79-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 24 annually	Capture, measure, weigh, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, anesthetize (MS-222), laparoscopically evaluate (coelomic cavity, collect blood, & collect biopsy of gonads (if sex unclear)	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Adult	Male & female	Up to 20 annually	Capture, hold, measure, weigh, photograph, scan (for tags), Floy T-bar tag, PIT tag, tissue sample, & hydroacoustic testing (tethered in nylon sock)	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)
Shortnose sturgeon (*Acipenser brevirostrum*)	Juvenile & adult	Male & female	1 unintentional mortality or serious harm* annually, not to exceed 3 over the life of the permit	Unintentional mortality, storage, measure, weigh, photograph, fin clip, freeze, transport arrangements made with NMFS for further sampling and disposal	Delaware River (netting area =rkm 79-215; secondary sampling area= rkm 0-215)

Species	Life Stage	Sex	Expected Annual Take	Take Action	Location
Shortnose sturgeon (*Acipenser brevirostrum*)	eggs & larvae	Male & female	300 annually (lethal take), not to exceed 900 over the life of the permit	Intentional (directed mortality)	Delaware River (netting area=rkm 215-245)

The time required to complete routine, non-invasive methods (*i.e.*, PIT and Floy tagging, measuring, weighing) would be less than one minute per fish. The cumulative time required for procedures such as anesthetizing, telemetry tagging, and genetic tissue sampling would vary, but would average less than 20 minutes per fish. Total holding time of shortnose sturgeon, after removal from the capture gear, holding, and scientific procedures would not exceed two hours, unless the fish has not recovered from anesthesia (if anesthetized). Following processing, all fish would be treated with slime coat restorative and, depending on procedure, placed in a separate net pen to ensure full recovery prior to release.

C. Response Analysis

As discussed in the *Approach to the Assessment* section of this Opinion, response analyses determine how listed resources are likely to respond after being exposed to an action's effects on the environment or directly on listed species themselves. For the purposes of consultation, our assessments try to detect potential lethal, sub-lethal (or physiological), or behavioral responses that might reduce the fitness of individuals. Ideally, response analyses would consider and weigh evidence of adverse consequences as well as evidence suggesting the absence of such consequences.

Effects of Capture. The applicant proposes to use bottom-set gill nets, trammel nets, and trawl nets to capture up to 1,000 annually, or totaling 3,600 shortnose sturgeon over five years. Entanglement in nets or damage suffered in trawls could result in injury and mortality, reduced fecundity, and delayed or aborted spawning migrations of sturgeon (Moser and Ross 1995, Collins *et al.* 2000, Moser *et al.* 2000). The majority of shortnose sturgeon that are known to have died during scientific investigations have been killed by gillnets. Historically, the majority of shortnose sturgeon mortality during scientific investigations using nets or trawls has been related to such factors as water temperature, low D.O concentration, netting duration, meshes size, net composition, and netting experience of the researcher (Table 11).

Table 11. The number and percentage of shortnose sturgeon killed by gill nets associated with scientific research permits prior to 2005

	Permit Number					
	1051	1174	1189	1226	1239	1247
Time Interval	1997, 1999 – 2004	1999– 2004	1999, 2001 – 2004	2003– 2004	2000 – 2004	1988 – 2004
Sturgeon captured	126	3262	113	134	1206	1068
Sturgeon mortality	1	7	0	0	5	13
Percentage	0.79	0.22	0	0	0.41	1.22

In 2005, PR1 began analyzing the results of previous research and updating permit conditions to reduce the chances of stress and mortality to shortnose sturgeon during capture. Since that time, there have been no mortalities caused during their capture (Table 12). The primary causes of mortality identified during a review of permits issued prior to 2005 were high temperatures, low dissolved oxygen, and long net set durations. Despite the permit modifications reducing mortality of sturgeon in nets, there is a chance of delayed mortality occurring without being reported. There is no way to estimate the rate of delayed mortality, but NMFS believes it would be less than one percent based on reports of various species of sturgeon captured and transported to rearing facilities.

Table 12. Number of shortnose sturgeon killed during capture under existing scientific research permits

Permit Number	Shortnose sturgeon captured	Shortnose sturgeon mortalities
1420 (2005-2009)	1472	0
1447 (2006-2009)	107	0
1449 (2007-2008)	50	0
1486* (2006-2009)	416	0
1505 (2006-2009)	276	0
1516 (2007-2009)	160	0
1547 (2006-2009)	112	0
1549 (2006-2009)	390	0
1575 (2007-2009)	12	0
1580 (2007-2008)	66	0
1595 (2007-2009)	505	0
10037 (2007-2009)	235	0
10115 (2008-2009)	1	0
Totals	3802	0

* NMFS Permit No. 1486 is the current permit proposed to be replaced by Permit File 14604.

Harold Brundage has maintained a record of verifiable mortality while engaged in other authorized research on shortnose sturgeon within the same proposed action area on the Delaware River under 1486; he has reported zero mortality to PR1 in an annual report. Annual reports from 1999 to 2004 (under NMFS Permit 1174) documented a mortality rate of 0.22%, or 7 fish out of a total of 3,286 captured. However, while working under more conservative sampling effort since 2005 (Permit No. 1486), the researcher has reported no shortnose sturgeon deaths while capturing 311 in four years of research. Nevertheless, in the current application, which requests authorization for a total of 3,600 captures over five years, the applicant outlines goals requiring increased netting activity ahead of an anticipated dredging action proposed by the U.S. Army Corps of Engineers (USACE) on the Delaware River.

Expected Response to Capture. As demonstrated above, there is a chance that shortnose sturgeon could die in gill nets, but mitigation measures included in the proposed activities should reduce the risk associated with sturgeon capture. To limit stress and mortality of sturgeon due to capture efforts, the proposed methodologies would utilize more conservative netting conditions than is suggested by the Moser *et al*. (2000) protocol. The proposed methodology also limits the soak times of gill nets to 2 hours or less at water temperatures in the range of 25^{o}C to 28°C and prohibits netting at temperatures above 28°C unless first contacting NMFS. Dissolved oxygen would also be measured prior to each net set and each time the net is

checked to ensure that at least 5.0 mg/L concentration is maintained. Also, to minimize injury, heavy multifilament nylon (5 or 6 cm stretch for adults; 1 to 4 cm stretch for juveniles) mesh would be used instead of monofilament or light twine, which is more capable of cutting into the fish causing injury. Due to the low ventilation rate and open operculum, the use of trammel nets is encouraged, as they allow the fish to become entangled rather than gilled. Based on the results of fish captures in recent years, the previous research conducted by the applicant, and the thorough mitigation measures included with these proposed activities, we expect the chances of a shortnose sturgeon being killed during capture to be very low. However, NMFS would anticipate some mortality and/or delayed mortality associated with capture due to the volume of netting activity and numbers of fish targeted.

Effects of Handling. Up to 1,000 adult/juvenile sturgeon annually (not exceeding 3,600 total for the entire permit duration) would be handled for length and weight measurements and the other proposed methods under this proposed research authorization. Handling and restraining shortnose sturgeon may cause short term stress responses, but those responses are not likely to result in pathologies because of the short duration of handling. Sturgeon are sensitive to handling stress when water temperatures are high or D.O. is low. Handling stress can escalate if sturgeon are held for long periods after capture. Conversely, stress is reduced the sooner fish are returned to their natural environment to recover. Signs of handling stress are redness around the neck and fins and soft fleshy areas, excess mucus production on the skin, and a rapid flaring of the gills. Additionally, sturgeon tend to inflate their swim bladder when stressed or when handled in air (Moser *et al*. 2000). If not returned to neutral buoyancy prior to release, sturgeon tend to float and would be susceptible to sunburn and bird attacks. In some cases, if pre-spawning adults are captured and handled, it is possible that they would interrupt or abandon their spawning migrations after being handled (Moser and Ross 1995).

Expected Response to Handling. Although sturgeon are sensitive to handling stress, the proposed methods of handling fish are consistent with the best management practices recommended by Moser *et al*. (2000) and endorsed by NMFS and, as such, should minimize the potential handling stress and therefore minimize indirect effects resulting from handling in the proposed research. Mitigation measures described in the environmental assessment, such as wearing rubber gloves to reduce skin abrasions, short handling times, recovering in pens, total holding time of less than 2 hours, and an electrolyte bath prior to release, should lessen the chance of injury or mortality during handling and restraint in any of the river systems. To minimize capture and handling stress, the proposed research plans to hold shortnose sturgeon in net pens until they are processed, at which time they would be transferred to a processing station on board the research vessel. During processing, each fish would be immersed in a continuous stream of water supplied by a pump/hose assembly mounted to over the side of the research vessel. For most procedures planned, the total time required to complete routine handling and tagging would be no more than 15 to 20 minutes. Moreover, following processing, fish would be returned to the net pen for observation to ensure full (return to equilibrium, reaction to touch stimuli, return of full movement) recovery prior to release.

Effects of Passive Integrated Transponder (PIT) Tags. All shortnose sturgeon captured would be marked with PIT tags (up to 1,000 fish per year; no more than 3,600 fish over the life of the permit). Prior to PIT tagging, the entire dorsal surface of each fish would be

scanned to detect previous PIT tags. Unmarked shortnose sturgeon would receive PIT tags by injection using a 12 gauge needle at an angle of 60 ^{o} to 80^{o} in the dorsal musculature (anterior to the dorsal fin).

PIT tags have been used with a wide variety of animal species that include fish (Clugston 1996, Skalski *et al.* 1998, Dare 2003), amphibians (Thompson 2004), reptiles (Cheatwood *et al.* 2003, Germano and Williams 2005), birds (Boisvert and Sherry 2000, Green *et al.* 2004), and mammals (Wright *et al.* 1998, Hilpert and Jones 2005). When PIT tags are inserted into animals that have large body sizes relative to the size of the tag, empirical studies have generally demonstrated that the tags have no adverse effect on the growth, survival, reproductive success, or behavior of individual animals (Brännäs *et al.* 1994, Elbin and Burger 1994, Keck 1994, Jemison *et al.* 1995, Clugston 1996, Skalski *et al.* 1998, Hockersmith *et al.* 2003). However, some fish, particularly juvenile fish, could die within 24 hours after tag insertion, others could die after several days or months, and some could have sub-lethal reactions to the tags.

If mortality of fish occurs, they often die within the first 24 hours, usually as a result of inserting the tags too deeply or from pathogen infection. About 1.3% of the yearling Chinook salmon (*Oncorhynchus tshawytscha*) and 0.3% of the yearling steelhead (*O. mykiss*) studied by Muir *et al.* (2001) died from PIT tag insertions after 24 hours. In the only study conducted on sturgeon mortality and PIT tags, Henne *et al.* (unpublished) found that 14 mm tags inserted into shortnose sturgeon under 330 mm causes 40% mortality after 48 hours, but no additional mortalities after 28 days. Henne *et al.* (unpublished) also show that there is no mortality to sturgeon under 330mm after 28 days if 11.5mm PIT tags are used. Gries and Letcher (2002) found that 0.7% of age-0 Atlantic salmon (*Salmo salar*) died within 12 hours of having PIT tags surgically implanted posterior to their pectoral fins, but nine months later, 5.7% of the 3,000 tagged fish had died. At the conclusion of a month long study by Dare (2003), 325 out of 144,450 tagged juvenile spring Chinook salmon died, but only 42 died in the first 24 hours.

Studies on a variety of fish species suggest that attachment of tags, both internal and external, can result in a variety of sub-lethal effects including delayed growth and reduced swimming performance (Morgan and Roberts 1976, Isaksson and Bergman 1978, Bergman *et al.* 1992, Strand *et al.* 2002, Bégout Anras *et al.* 2003, Robertson *et al.* 2003, Sutton and Benson 2003, Brattey and Cadigan 2004, Lacroix *et al.* 2005). Larger tags and external tags have more adverse consequences, such as impaired swimming, than smaller tags (Bégout Anras *et al.* 2003, Sutton and Benson 2003).

Expected Response to PIT Tags. PIT tags would be used for permanently marking and identifying individual fish by injecting the tags intramuscularly anterior to the dorsal fin. These biologically inert tags have been shown not to cause problems associated with some other methods of tagging fish, that is, scarring and damaging tissue or otherwise adversely affecting growth or survival (Brännäs *et al*. 1994). As such, the proposed tagging of shortnose sturgeon with PIT tags is unlikely to have significant impact on the reproduction, numbers, or distribution of shortnose sturgeon. However, there is one record of young sturgeon mortality within the first 24-48 hours of PIT tag insertion as a result of the tags being inserted too deeply. Henne *et al*. (unpublished) found 14 mm tags injected into smaller shortnose sturgeon caused mortality after 48 hours; also he inferred from his results, either 11.5 or 14 mm PIT tags would not cause

mortality in sturgeon equal to or longer than 330 mm (TL). To address this concern, the applicant would use PIT tags that are 11.9 mm x 2.1 mm using a 12 gauge needle on shortnose sturgeon that are ≥330 mm TL.

Effects of Floy (T-bar Anchor) Tags. All shortnose sturgeon captured would also be marked with Floy tags (up to 1,000 fish per year; no more than 3,600 fish over the life of the permit). Applicants propose to tag shortnose sturgeon by this method to incorporate the incidental recapture of sturgeon by fishermen, researchers, and managers to enable collection of information useful to the assessment of the sturgeon population.

In all captured sturgeon, Floy tags would be anchored in the dorsal fin musculature base and inserted forwardly and slightly downward from the left side to the right through dorsal pterygiophores. After removing the injecting needle, the tag would be spun between the fingers and gently tugged to be certain it is locked in place. During the study, the rate of Floy tag retention would be documented and reported in NMFS annual reports.

Smith *et al.* (1990) compared the effectiveness of dart tags with nylon T-bars, anchor tags, and Carlin tags in shortnose and Atlantic sturgeon. Carlin tags applied at the dorsal fin and anchor tags in the abdomen showed the best retention, and it was noted that anchor tags resulted in lesions and eventual breakdown of the body wall if fish entered brackish water prior to their wounds healing. However, Collins et al. (1994) found no significant difference in healing rates (with T-bar tags) between fish tagged in freshwater or brackish water. Clugston (1996) also looked at T-bar anchor tags placed at the base of the pectoral fins and found that beyond two years, retention rates were about 60%. Collins et al. (1994) compared T-bar tags inserted near the dorsal fin, T-anchor tags implanted abdominally, dart tags attached near the dorsal fin, and disk anchor tags implanted abdominally. They found that for the long-term, T-bar anchor tags were most effective (92%), but also noted that all of the insertion points healed slowly or not at all, and, in many cases, minor lesions developed.

Expected Response to Floy (T-bar Anchor) Tags. The use of Floy tags and PIT tags to mark shortnose sturgeon are duplicative means to identify captured fish. However, we believe that the practice is not expected to significantly impact sturgeon health. The attachment of tags may cause some discomfort and pain to shortnose sturgeon. Generally, there is little observable reaction to the injection of PIT tags. However, the injection of Floy tags may result in more noticeable reactions than the injection of PIT tags. There is also a greater potential for injury from the insertion of Floy tags than PIT tags because the tag is typically interlocked between interneural cartilage. Injury may result during attachment, although the potential for this is seriously reduced when tags are applied by experienced biologists and technicians. Mortality is unlikely for either tag type (PIT or Floy).

Injection of Floy tags into the dorsal musculature, however, may result in raw sores that may enlarge overtime with tag movement (Collins *et al.* 1994, Guy *et al.* 1996). Beyond the insertion site, it is unknown what effects the on fish the attachment of Floy tags may have. We know of no long-term studies evaluating the effect of these tags on the growth or mortality of tagged shortnose sturgeon. Anecdotal evidence recounted in NOAA's protocol (Moser *et al*. 2000) suggests that Floy tags have little impact on the fish because a number of shortnose were

recovered about 10-years after tagging although no data are available to evaluate any effects on growth rate. Studies on other species suggest that the long-term effect of injecting anchor tags into the muscle may be variable. Researchers have observed reduced growth rates in lemon sharks and northern pike from tagging, whereas studies of largemouth bass did not depict changes in growth rates (Tranquilli and Childers 1982, Manire and Gruber 1991, Scheirer and Coble 1991).

To lessen known negative impacts described above using the Floy tag, sterile tagging technique would be used and methods would require to subsequently monitor dorsal fin tag sites of recaptured sturgeon for any lesions. Additionally, results of tag retention and fish health would be reported to NMFS in annual reports and as requested by NMFS. If impacts of the Floy tags are other than insignificant, NMFS would reevaluate their use in the permit.

Effects of Tissue Sampling. All shortnose sturgeon captured would be tissue-sampled. Tissue sampling does not appear to impair the sturgeon's ability to swim and is not thought to have any long-term adverse impact. Many researchers have removed tissue samples according to this same protocol with no mortalities (Wydoski and Emery 1983).

Expected Response to Tissue Sampling. We expect shortnose sturgeon tissue sampled under the proposed permit to respond similarly to that reported above. Therefore, we do not anticipate any long-term adverse effects to the sturgeon from this activity.

Effects of Collection of ELS. The proposed research would document spawning activity by collecting up to 300 shortnose sturgeon ELS annually (not to exceed 900 over five years) using a combination of egg mats, D-nets, or epibenthic sleds in previously documented spawning habitat for shortnose sturgeon between Trenton and five kilometers upstream of Lambertville, New Jersey. The artificial substrates used to collect ELS are polyester floor-buffing pads that passively collect eggs or larvae adrift in the water during spawning activity. Drifting or dislodged embryos would settle on the pads, be identified, and preserved. D-frame ichthyoplankton nets would be bottom set and would hold upright in the current. Epibenthic sleds would have D-frame ichthyoplankton nets fitted to them and would be towed slowly near these documented spawning areas.

Each adult female sturgeon produces between 94,000 and 200,000 eggs every 3 years (COSEWIC 2005). The survival from egg to juvenile is likely the most critical aspect in determining the strength of the year class (COSEWIC 2005). However, as the five-year proposed take of 900 eggs or larvae is small compared to the potential total release of eggs, such take would be considered to have minimal impact on the shortnose sturgeon population of the river.

Expected Response to Collection of ELS. As stated above, we believe that the annual proposed take of up to 300 eggs or larvae per year (for a total cap of 900 over five years) is small compared to the potential total release of eggs and such take would be considered to have minimal impact on the shortnose sturgeon population of the Delaware River. Should there be an excess of the authorized take, they would immediately be returned to the river. Due to their relatively small size, these pads or nets would not disrupt the flow water flow or habitat.

Effects of Anesthetic. During the five years of research authorized by the proposed permit 14604, up to 30 shortnose sturgeon annually in the Delaware River could be anesthetized with MS-222 at concentrations up to 150 mg/L in order to sedate the fish for transmitter implantation. In addition, up to 24 shortnose sturgeon annually could be anesthetized with MS-222 at concentrations up to 250 mg/L for laparoscopy. MS-222's mode of action prevents the generation and conduction of nerve impulses directly affecting the central nervous system, cardiovascular system, neuromuscular junctions, and ganglion synapses (Brown 1988). It is rapidly absorbed through the gills. However, because MS-222 is acidic and poorly absorbed, resulting in a prolonged induction time, Sodium bicarbonate (NaHCO3) would be used to buffer the water to a neutral pH.

The decision on whether or not to use an anesthetic must be considered in terms of the animal's welfare, the physiological effects of the anesthetic, and the consequences of not using the anesthetic (Summerfelt and Smith 1990). Furthermore, there are two important parameters to consider when deciding how to use the anesthetic. These are: 1) the level of sedation needed for the given procedure to be used (referred to as anesthetic stage, or plane); 2) and the effective concentration needed to reach that particular level of sedation.

Anesthetics may be classified as general, local, and regional. Anesthetic stages include normal, light, deep, partial or total loss of equilibrium, loss of reflex activity, and medullary collapse (Summerfelt and Smith 1990). Summerfelt and Smith (1990) modified these stages from McFarland (1959) and Jolly *et al*. (1972) to indicate the various planes in which a fish may be anesthetized. A given researcher must consider the wellbeing of the fish to be anesthetized when choosing which stage of anesthesia is required for their particular procedure. For example, some localized implantation surgeries or fin clips may require a light sedation, while a much more invasive procedure, such as laparoscopy with biopsy, could require a deeper plane of anesthesia. This is because body movements during surgery (such as tail flexions) increases the likelihood of additional trauma to the fish. Noxious stimuli induced by incision, organ manipulation, and tissue biopsy could result in an immediate reaction by fish that receive no anesthesia or are only lightly sedated.

The effective concentration of anesthetic is that which produces a total loss of equilibrium in 50% of the particular fish species in a specified time (Schoettger and Julin 1967). There is no unequivocally defined effective concentration for shortnose sturgeon, because various researchers have used differing concentrations depending on the research procedure they have chosen to perform. Again, some localized implantation surgeries or fin clips may require a light anesthesia, whereas a more invasive surgery might require deep anesthesia.

MS-222 is a recommended anesthetic for sturgeon research when used at safe concentrations (Moser *et al.* 2000, USFWS 2008; *but see* Henyey *et al*. 2002 and Jennings and Looney 1998 (striped bass), preferring electronarcosis to MS-222). It is rapidly absorbed through the gills and its mode of action is to prevent the generation and conduction of nerve impulses with direct actions on the central nervous system and cardiovascular system. As previously described, different planes/states of anesthesia can be achieved with different doses of MS-222 - lower doses tranquilize and sedate fish while higher doses fully anesthetize them (Taylor and Roberts

1999). In 2002, MS-222 was FDA-approved for use in aquaculture as a sedative and anesthetic in food fish (FDA 2002).

One risk associated with employing MS-222 to anesthetize sturgeon is using concentrations at harmful or lethal levels, therefore it is imperative that the proper effective concentration is chosen for the methods proposed under a given study. Studies show short-term risks of using MS-222 to anesthetize sturgeon other than shortnose, but show no evidence of irreversible damage when concentrations are used at levels recommended for particular procedures. A study on steelhead and white sturgeon revealed deleterious effects to gametes at concentrations of 2,250 to 22,500 mg/L MS-222, while no such effects occurred at 250 mg/L and below (Holcomb *et al.* 2004). Another study did not find MS-222 to cause irreversible damage in Siberian sturgeon, but found MS-222 to severely influence blood constituents when currently absorbed (Gomulka *et al*. 2008; *see also* Cataldi *et al.* 1998 for Adriatic sturgeon).

The above studies show use risks of MS-222 to other sturgeon species, but also show that irreversible damage could be avoided if researchers use proper effective concentrations for their particular technique. Pertaining to shortnose sturgeon specifically, studies conducted by Haley 1998, Moser *et al.* 2000, Collins *et al.* 2006, 2008 show no obvious deleterious effects with MS-222 at concentrations up to 150 mg/L when used for laparoscopy and transmitter implantation. Mark Matsche conducts anesthetization training (250 mg/L) and laparoscopy with biopsy at the Warm Springs National Fish Hatchery Center (Fish and Wildlife Service) and has produced a video on this technique. The use of concentrations up to 250 mg/L is carefully monitored via heart monitor to produce deep narcosis for laparoscopy and possible biopsy. Many researchers have been trained in this method (Mark Matsche, Maryland Department of Natural Resources and Bill Post, South Carolina Department of Natural Resources, pers. comm). This technique has not produced evident complications or observed shortnose sturgeon mortality, but one other fish of a different species was observed not to recover quickly from sedation.

Effects of MS-222 would be short-term and only affect the target species. MS-222 is excreted in fish urine within 24 hours and tissue levels decline to near zero in the same amount of time (Coyle *et al.* 2004). To increase absorption time and ensure a fast anesthesia process, the applicant will add sodium bicarbonate to buffer the acidic MS-222 to a more neutral pH. Therefore, at the proposed rates of anesthesia, narcosis would take one minute and complete recovery time would range from three to five minutes (Brown 1988).

Studies show that recovery from anesthetic stress is more of a concern than the anesthetic itself, which leaves the body in 24 hours. Scientists have examined physiological responses of other fish species to MS-222. MS-222 has increased stress response in rainbow trout (Wagner *et al*. 2003), channel catfish (Small 2003), and steelhead trout (Pirhonen and Schreck 2003), as indicated by elevated plasma cortisol levels (Coyle et al. 2004). Additionally, a comparison of steelhead trout controls to MS-222-treated steelhead revealed an anesthetic stress response regarding feed. Steelhead sampled at 4, 24, and 48 hours after MS-222 exposure fed less than their controlled counterparts (Pirhonen and Schreck 2003). Furthermore, initial exposure to MS-222 may cause stress or, less likely, have an inherent hemodilution effect. The effect of MS-222 on the microhematocrit of freshwater fish blood was examined by Hattingh (1977) and showed that blood containing MS-222 had a higher hematocrit value than blood without anesthetic.

MacAvoy and Zaepfel (1997) also studied the effect of MS-222 on blacknose dace hematocrit and found elevations in fish exposed to 500 mg/L for three minutes. Laitinen *et al*. (1981) revealed that MS-222 induced modification on liver metabolism in splake (*Salvelinus fontinalis*). Puceat *et al.* (1989) compared the hepatic contents (glycogen, glucose, lactate, ATP, ADP, AMP) of rainbow trout exposed to MS-222 versus 2-phenoxyethanol and found no significant difference regarding each anesthetic's effect on glucose release. Soivio *et al.* (1977) studied the effects of anesthesia with MS-222, neutralized MS-222, and benzocaine on the blood constituents of rainbow trout. All anesthetics raised Mg++ concentrations and neutralized MS-222 and benzocaine elevated the plasma K+ concentration more than unbuffered MS-222. Spotte *et al.* (1991) found that deep anesthesia of mummichogs resulted in the increase in plasma cortisol, indicating a primary neuroendocrine stress response. In contrast, Strange and Schreck (1978) briefly anesthetized yearling Chinook salmon with MS-222 (50 mg/L) and found this to cause no change in plasma cortisol concentrations. These studies indicate sublethal physiological concerns if duration of exposure is not limited and the fish carefully monitored.

Expected Response to Anesthetic for Implanting Acoustic Tags. Due to the fact that the applicant aims to use a concentration up to 150 mg/L of MS-222 and ensure that fish are anesthetized for a short period of time, NMFS believes that most shortnose sturgeon sedated by MS-222 at 150 mg/L would be exposed only to minimal short-term risk and should recover to normal. The applicant aims to avoid the possibility of irreversible effects by following concentration recommendations and recovery procedures used in successful shortnose sturgeon diet studies with similar methodologies (Haley 1998, Moser *et al.* 2000, Collins *et al.* 2006, 2008). The applicants are experienced in using MS-222, having used it to perform surgical procedures many times on Atlantic and shortnose sturgeon. Because MS-222 is acidic and poorly absorbed, resulting in a prolonged induction time, Sodium bicarbonate (NaHCO3) would be used to buffer the water to a neutral pH. At the proposed rate, induction time would be approximately three to five minutes and complete recovery times would range from five to six minutes (Brown 1988). MS-222 would be excreted in fish urine within 24 hours and tissue levels would decline to near zero in the same amount of time (Coyle *et al.* 2004). The applicant would limit duration of anesthesia and monitor recovery in net pens before releasing fish.

However, due to the sublethal physiological concerns surrounding the use of MS-222 on fish, buffered by the success of its use in similar shortnose sturgeon studies, it is reasonable to anticipate possible anesthesia mortality throughout the life of the permit. None of the previous NMFS-permitted studies conducting anesthesia have experienced mortality using the techniques proposed by the applicant. The permit would authorize successful methodologies set forth by leading researchers of shortnose sturgeon. Probable ill effects aside from mortality might be sublethal stress responses, however, the permit incorporates precautions to shorten anesthetic time and monitor recovery in pens. The permit also incorporates many other precautions which were explained in the Description of the Proposed Action section. Therefore, the anesthesia methodology as proposed for transmitter implantation is reasonably anticipated to cause possible unintentional mortality, but is not likely to reduce the viability of the Delaware River shortnose sturgeon population. By extension, MS-222 anesthesia proposed for transmitter implantation is not likely to reduce the viability of shortnose sturgeon as listed under the ESA. This conclusion can be reached as long as the stated concentration of MS-222 are used and proposed duration exposure and procedures are closely followed.

Expected Response to Anesthetic for Laparoscopy. Due to the expertise that the applicant has developed in using a concentration up to 250 mg/L of MS-222 to anesthetize shortnose sturgeon to a deep narcosis for laparoscopy and biopsy, NMFS believes that most shortnose sturgeon sedated by MS-222 at 250 mg/L would be exposed only to minimal short-term risk and should recover to normal. The Co-Investigator, Mark Matsche, who is performing this technique under the proposed permit has developed this procedure and trained others on how to perform it effectively. He would use all precautions as explained in the Description of the Proposed Action Section, including the use of a heart monitor.

NMFS believes that, due to the invasiveness of the proposed laparoscopic method and accompanying biopsy, an effective concentration of 250 mg/L (to reach deep narcosis) is proper to reduce reactions that each fish could potentially have under a lower concentration (lighter plane) of anesthesia. Mark Matsche (CI) reports that, when conducting laparoscopic technique under a light plane of anesthesia, shortnose sturgeon can have a violent reaction (which might or might not be pain-induced) to the scope insertion. A violent reaction or even light twitch response to the laparoscope could severely injure the fish. Therefore, NMFS is convinced that it is safer for the fish to be in a deep narcosis, where this type of potentially injurious reaction is not observed.

Due to potential unknown physiological effects or sublethal stress responses, the anesthesia for laparoscopy as proposed is reasonably anticipated to cause possible unintentional mortality, but is not likely to reduce the viability of the Delaware River shortnose sturgeon population. By extension, MS-222 anesthesia for laparoscopy is not likely to reduce the viability of shortnose sturgeon as listed under the ESA. This conclusion can be reached as long as the stated concentration of MS-222 are used and proposed duration exposure and procedures are closely followed.

Effects of Internal Sonic Transmitters. For the proposed activities, a maximum of 30 adult (≥600 mm) and 30 juvenile (<600 mm) shortnose sturgeon would be surgically implanted with an internal acoustic transmitter using the protocol measures presented in Coyle *et al.* (2004). Applicants would use Vemco V16-5H for adults and either V7-4L, V9-6L, or V13-1H tags for juveniles, depending on the weight of the individual fish. Sonic transmitter devices would be limited in size to no more than 2% of a given fish's body weight. These same fish will have also been tagged with PIT and Floy tags and will have been tissue sampled, captured, measured, weighed, scanned for tags, and anesthetized. Although more invasive surgical procedures are required for this internal implantation, these tags provide greater retention rates than external attachment (Collins *et al*. 2002, Counihan and Frost 1999).

In general, adverse effects of these proposed tagging procedures could include pain, handling discomfort, hemorrhage at the site of incision, risk of infection from surgery, affected swimming ability, and/or abandonment of spawning runs. Choice of surgical procedure, fish size, morphology, behavior and environmental conditions can affect the success of telemetry transmitter implantation in fish (Jepsen *et al*. 2002).

Survival rates after implanting transmitters in shortnose sturgeon are high. Collins *et al*. (2002) evaluated four methods of radio transmitter attachment on shortnose sturgeon. They found 100% survival and retention over their study period for ventral implantation of a transmitter with internally-coiled antenna. Their necropsies indicated there were no effects on internal organs. Dr. Collins in South Carolina (M. Collins, *pers. comm.* 2006) has also more recently reported no mortality due to surgical implantation of internal transmitters. Devries (2006) reported movements of 8 male and 4 female (≥ 768 mm TL) shortnose sturgeon internally radio-tagged between November 14, 2004 and January 14, 2005 in the Delaware River. Eleven of these fish were relocated a total 115 times. Nine of these fish were tracked until the end of 2005. The remaining individuals were censored after movement was not detected, or they were not relocated, after a period of 4 months. Periodic checks for an additional 2 months also showed no movement. Although there were no known mortalities directly attributable to the implantation procedure; the status of the 3 unrelocated individuals was unknown (Devries 2006).

Growth rates after transmitter implantation are reported to decrease for steelhead trout. Welch *et al*. (2007) report results from a study to examine the retention of surgically-implanted dummy acoustic tags over a 7 month period in steelhead trout pre-smolts and the effects of implantation on growth and survival. Although there was some influence in growth to week 12, survival was high for animals > 13 cm FL. In the following 16 week period growth of surgically implanted pre-smolts was the same as the control population and there was little tag loss from mortality or shedding. By 14 cm FL, combined rates of tag loss (mortality plus shedding) for surgically implanted tags dropped to < 15% and growth following surgery was close to that of the controls.

Tag weight relative to fish body weight is an important factor in determining the effects of a tag (Jepsen *et al*. 2002). The two factors directly affecting a tagged fish are tag weight in water (excess mass) and tag volume. Perry *et al*. (2001) studied buoyancy compensation of Chinook salmon smolts tagged with surgical implanted dummy tags. The results from their study showed that even fish with a tag representing 10% of the body weight were able to compensate for the transmitter by filling their air bladders, but the following increase in air bladder volume affected the ability of the fish to adjust buoyancy to changes in pressure. Winter (1996) recommended that the tag/body weight ratio in air should not exceed 2%. Tags of greater sized implants produced more mortality of juvenile Atlantic salmon. There was 60% mortality (3 of 5 fish) with a 32-mm implant and 20% mortality (1 of 5 fish) with a 28-mm implant and 20% mortality (1 of 5 fish) with a 24-mm implant (Lacroix *et al*. 2004). Fish with medium and large external transmitters exhibited lower growth than fish with small transmitters or the control group (Sutton and Benson 2003).

Implanted transmitters could affect fish swimming performance. Thorstad *et al*. (2000) studied the effects of telemetry transmitters on swimming performance of adult farmed Atlantic salmon. These researchers found that swimming performance and blood physiology of adult Atlantic salmon (1021-2338 g, total body length 45-59 cm) were not affected when equipped with external or implanted telemetry transmitters compared with untagged controls. There was no difference in endurance among untagged salmon, salmon with small external transmitters, large external transmitters and small body-implanted transmitters at any swimming speed. Authors cautioned that results of wild versus farmed salmon may be different (Peake *et al*. 1997). However, a similar study using sea-ranched Atlantic salmon found no difference in endurance,

similar to the farmed salmon study (Thorstad *et al.* 2000). On the other hand, juvenile Chinook salmon < 120 mm FL with either gastrically or surgically implanted transmitters had significantly lower critical swimming speeds than control fish 1 and 19-23 days after tagging (Adams *et al.* 1998).

Implanted transmitters could effect fish growth. Juvenile Chinook salmon with transmitters in their stomachs (gastrically implanted) consistently grew more slowly than fish with surgically implanted transmitters, fish with surgery but no implanted transmitter, or fish exposed only to handling (Adams *et al*. 1998).

Water temperature has been shown to affect rainbow trout implanted with simulated transmitters. 80 rainbow trout were implanted with simulated transmitters and held at various temperatures for 50 days (10, 15, 20 degrees) (Bunnell and Isely 1999). Transmitter expulsion ranged from 12% to 27% and was significantly higher at 20 degrees C than at 10 degrees C. Mortality ranged from 7 – 25% and was not related to temperature.

Since implantation requires surgery, healing is frequently described in the relevant scientific literature. Several factors can affect obstacles to wound healing in fish including secondary infection and inflammation. Fish epidermal cells at all levels are capable of mitotic division, and during wound healing there is a loss of the intracellular attachments and cells migrate rapidly to cover the defect and provide some waterproof integrity (Wildgoose 2000). This leads to a reduction in the thickness of the surrounding epidermis and produces a thin layer of epidermis at least one cell thick over the wound, however the process can be inhibited by infection (Wildgoose 2000). Thorstad *et al*. (2000) state that incisions were not fully-healed in 13 of the farmed Atlantic salmon with implanted transmitters; two of these had signs of inflammation. Juvenile largemouth bass implanted with microradio transmitters exhibited short-term (5 days) inflammation around the incision and suture insertion points for both non-absorbable braided silk and non-absorbable polypropylene monofilament, but in the longer term (20 days) almost all sutures were shed and the incisions were completely healed (Cooke *et al*. 2003). Chapman and Park (2005) examined suture healing following a gonad biopsy of Gulf of Mexico sturgeon and found both the absorbable and nonabsorbable sutures to effectively sew the skin after biopsy with all sturgeons surviving surgery and incisions healing 30 days after the intervention. Dummy radio transmitters compounded the inflammatory effect silk sutures had on healing incisions compared with inflammation without transmitters (Wagner *et al*. 2000).

The expulsion or rejection of surgically implanted transmitters has been reported from a number of studies, therefore, expulsion could be an argument for using externally attached transmitters. It does not appear that expulsion causes further complications or death in fish that manifest this occurrence. Such expulsions often occur shortly after tagging and can lead to premature end of studies. Rates of tag shedding and ways of implant exits depend on species, fish condition, tag weight and environmental conditions (Jepsen *et al*. 2002). There are basically three ways for an implant to exit; through the incision, through an intact part of the body wall and through the intestine. Trans-intestinal expulsion is rare but has been occasionally reported in rainbow trout (Chisholm and Hubert 1985). Five months after tagging, 20% of juvenile Atlantic salmon had expelled their tags through the body wall, adjacent to the healed incision (Moore *et al*. 1990). No mortality or infection occurred as a result of tag expulsion, and fish continued to mature and

behave like the control fish. Expulsion occurred in 13 of 22 rainbow trout tagged with dummy tags coated with paraffin wax within 42-175 days after tagging (Chisholm and Hubert 1985). In another study of rainbow trout, three of 21 fish expelled their tags via body wall without subsequent mortality (Lucas 1989). Tag expulsion by juvenile Atlantic salmon during Lacroix *et al.*'s study occurred but was not a cause of death (2004). Two surgically implanted transmitters were also apparently expelled by Atlantic sturgeon (Moser and Ross 1995). In Kieffer and Kynard's (1993) study, one shortnose sturgeon implanted with a sonic tag rejected its internal tag.

Coating the transmitters has been suggested to vary the rate of expulsion. It has been hypothesized that paraffin coating of the transmitter increases expulsion rate (Chisholm and Hubert 1985). Moser and Ross (1995) reported that retention of surgically implanted tags could be improved for Atlantic sturgeon when the transmitters were coated with a biologically inert polymer, Dupont Sylastic. Additionally, Kieffer and Kynard (*In press*) report that tag rejection internally is reduced by coating tags with an inert elastomer and by anchoring tags to the body wall with internal sutures. Kieffer and Kynard's fish retained tags for their operational life, and in most cases, lasted much longer (mean, 1,370.7 days).

Expected Response to Internal Sonic Transmitters. We expect that shortnose sturgeon exposed to internal sonic transmitter implantation would respond similar to the available information presented above. Survival rates are expected to be high with no ill effects on internal organs expected as a result of the transmitters. We do not expect mortality to occur as a result of this procedure, although a few tagged fish from studies reported above have disappeared and their fate is unknown. We expect that growth rates or swimming performance could be affected and that expulsion of the transmitter could occur. There have been no mortalities or infections reported to be associated with expulsion. We expect that the surgical wound would heal normally, but acknowledge that adverse effects of these proposed tagging procedures could include pain, handling discomfort, hemorrhage at the site of incision, risk of infection from surgery, affected swimming ability, and/or abandonment of spawning runs. The research methodologies will minimize these risks, as choice of surgical procedure, fish size, morphology, behavior and environmental conditions can affect the success of telemetry transmitter implantation in fish (Jepsen *et al*. 2002).

PR1 proposes to authorize the use standardized protocols endorsed by NMFS (Moser *et al*. 2000) which aim to minimize the effects caused by internally implanting transmitter tags. To ensure the sturgeon can endure the weight of these tags, a condition would be imposed stating that the total weight of all transmitters and tags would not exceed 2% of the fish's body weight. Surgical implantation of internal tags would only be attempted when fish are in excellent condition, and would not be attempted on pre-spawning fish in spring or fish on the spawning ground, nor in water temperatures greater than 27^{o}C or less than 7^{o}C. By using proper anesthesia, sterilized conditions, precautions, and the surgical techniques described above, these procedures would not be expected to have a significant impact on the normal behavior, reproduction, numbers, distribution or survival of shortnose sturgeon.

Effects of Laparoscopy. Up to 24 shortnose sturgeon annually would be exposed to laparoscopy. Laparoscopy is a minimally invasive surgery (MIS), or an operation performed

through small incision(s) compared to larger incisions needed for traditional surgeries. In comparison to most traditional surgical procedures, MIS induces relatively minor tissue trauma, which, in most cases, results in shorter postoperative recovery periods, decreased postoperative care, and fewer postoperative complications (Cook and Stoloff 1999). Laparoscopy is used in fish species to qualitatively assess morphological health and to visually identify the sex and maturity status of study fish accurately. Laparoscopy can begin in two different ways. The procedure could be done by cutting a small incision in the fish's body cavity and inserting an endoscope to view gonads or other internal organs (Hernandez-Divers *et al*. 2004, Moccia *et al*. 1984, Swenson *et al*. 2007, Wildhaber *et al*. 2005). The endoscope can also be inserted through the urogenital pore, which avoids having to make an incision (Kynard and Kieffer 2002, Ortenburger *et al*. 1996). For laparoscopy using an incision entry point, a trocar is sometimes used. The trocar acts to "guide" the endoscope into the fish through the incision, and protects the incision from further tear. Endoscopes may also be flexible or rigid. The rigid 42 endoscope always requires a straight path to the organs being examined whereas the flexible endoscope may give and bend (Dover and Van Bonn 2001). Finally, insufflation with a gas is used to provide the visual internal space needed for effective examination with the endoscope.

Permit 14604 would authorize researchers to conduct a 1 – 3 minute laparoscopy by incision on up to 24 captured shortnose sturgeon annually. After immobilized with MS-222 (effects of anesthesia are analyzed in a separate section), animals would be positioned in lateral recumbency on a portable surgical table. Researchers would make a 5 mm incision in the ventral body wall slightly off midline at a level midway between the pectoral girdle and the cloaca. A 5 mm trocar would then be inserted through the incision and a 5 mm rigid endoscope would be inserted through the trocar. If necessary, the body cavity would be insufflated with ambient air by attaching a battery-powered air pump to an insufflation port on the trocar.

When compared to other methods, laparoscopy has been shown as an accurate method for determining the sex of fish from the Acipenseridae and Salmonidae families. Swenson *et al*. (2007) utilized laparoscopy to correctly determine the maturity status and sex of mature individuals for 96% of the eastern brook trout examined in their study. The percentage was determined by euthanizing trout after laparoscopy for dissection and comparing results of the two methods. Wildhaber *et al*. (2005) assessed the effectiveness of ultrasound versus laparoscopy for sex determination of shovelnose sturgeon by verifying results with histological analysis. These researchers found that the success of the method used for sex determination was dependent upon its invasiveness, whereby laparoscopy was more effective than ultrasound.

Within laparoscopy technique, inserting the endoscope through the urogenital pore for sex determination is not as consistent as sex determination of endoscopy through incision (shovelnose sturgeon; Wilhaber *et al*. 2005). Introduction of the endoscope through the urogenital pore was not difficult in female arctic char, but resulted in accidental rupture of the spermatic duct in some of the males (Ortenburger *et al*. 1996). Furthermore, Kynard and Kieffer (various sturgeon species; 2002) observed an unpredictability of urogenital opening size based on length of fish. They recommended choosing an endoscope with small rather than large diameter. To avoid this unpredictability, it could be prudent to utilize an incision, rather than urogenital pore insertion, to create a predictable opening and therefore the endoscope diameter could properly be chosen.

Many studies comment on the absence of injury or other evident damage from laparoscopic procedure and report it to be a relatively safe procedure when carried out carefully. It is reported that laparoscopy does not harm reproductive structures, does not cause internal damage such as bruising or infection, and does not cause hemorrhage or buoyancy problems. Kynard and Kieffer (2002) reported that careful use of an endoscope will not harm reproductive structures and is suitable for all sturgeon species. They also report that endoscopes inserted through the urogenital pore will not damage the female oviduct valve. Moccia *et al*. (1984) noted that necropsy of rainbow trout maintained under controlled lab conditions revealed no evidence of internal damage from a previous endoscopic procedure, such as internal bruising or infection. They also note that gross healing of the surgical incision is 70% complete in 7 – 10 days, without signs of inflammation or other damage even without antibiotics. Hernandez-Divers *et al*. (2004) reported that no morbidity or mortality occurred as a result of laparoscopy to Gulf of Mexico sturgeon as there was no significant hemorrhage or trauma associated with any fish. Furthermore, they also noted that no postoperative swimming or buoyancy problems (i.e. swim bladder injury) were observed in their study.

Laparoscopy post-procedure mortality is reported in the literature for Salmonidae, and has been attributed to small fish size and coincident chronic gill disease infection. Swenson *et al*. (2007) reported a 3.3% post-procedure mortality for laparoscopy of eastern brook trout, which was limited to trout smaller than 70 mm FL. These fish were from the smallest class size Swenson *et al.* (2007) examined for their study and therefore they hypothesized that smaller individuals may be at greater risk from laparoscopy than larger fish. They suggested this could be due to the fact that the procedure may have taken longer for small fish because it was more difficult to view internal organs. Orgenburter *et al*. (1996) reported that 2 of the arctic char that underwent laparoscopy in their study died compared to none in the control group. This was attributed to severe chronic gill disease and no signs of peritonitis or inflammation of the coelomic viscera were found on necropsy and subsequent histological examination of deceased fish (Ortenburger *et al*. 1996). These researchers were ultimately unable to definitively relate deaths to the procedure described – because both deceased fish had survived for more than 5 days following procedure and were diagnosed as having severe degenerative gill disease at the time of death (Ortenburger *et al*. 1996).

It has also been suggested that stresses incurred during the procedure and delayed complications, as well as increased susceptibility to predation after release, could also contribute to mortality. Moccia *et al*. (1984) suggested that incidental loss of epidermal mucus, increases in body temperature, drying of the skin, or a combination of these factors could contribute to eventual mortality in fish that undergo laparoscopy, but their previous laboratory studies indicate this is unlikely. Although immediate mortality may be low post-laparoscopy, we should not rule out the possibility of delayed complications from laparoscopy, such as reopening of the incision, infection, and injury to internal organs (Swenson *et al.* 2007). Accidental perforation of the caudal air bladder was known to have occurred in 3 of the 70 arctic char evaluated by Ortenburger *et al*. (1996). Ecchymotic hemorrhages were seen on microscopic evaluation of the tissue surrounding the genital pore only in female arctic char that had ovulated and hemorrhage appeared to be associated with oviposition rather than introduction of the endoscope (Ortenburger *et al*. 1996). Inflammatory infiltrates were only seen surrounding the genital pore in male arctic char, and may indicate disruption of the normal communication of the vas deferens

(Ortenburger *et al*. 1996). The blind and forced puncture of an endoscopic cannula and trocar into the coelom can potentially cause visceral bruising or perforation and researchers used a threaded design for gradual advancement by rotation to avoid bruising (Hernandez-Divers *et al*. 2004). The use of insufflation pressure greater than 4-8 mm Hg could compromise circulation, especially venous return, in fish with lower arterial and venous blood pressures (Hernandez-Divers *et al*. 2004). Fish released into wild settings after laparoscopy may be more susceptible to these and other sources of related mortality, such as subsequent predation (Swenson *et al*. 2007).

Further study is needed to evaluate the long-term lethal and sublethal effects of endoscopy in natural settings and there is still a need to document the continued fertility of fish subjected to endoscopy. However, studies of radio tagging, a procedure that is more invasive than endoscopy, suggest that these problems are minimal. For example, radio tags in largemouth bass and dummy acoustic transmitters in juvenile Atlantic salmon had few long-term effects on fish in the wild (Cooke *et al*. 2003, Lacroix *et al*. 2004).

Expected response to laparoscopy. We expect that the shortnose sturgeon exposed to procedures under this action would respond similarly as revealed in the literature above. Since the proposed methodologies would be using the laparoscopy technique by incision rather than urogenital pore, we expect to see less complications than are reported for inserting an endoscope through a urogenital pore. In addition, we do not expect to see significant hemorrhage or trauma associated with the procedure. We also do not expect postoperative swimming problems. Finally, the post-procedure mortality seen in Salmonidae has been attributed to small fish size and gill infection. Laparoscopy would only be conducted on adults. We expect that a large majority of the shortnose sturgeon that undergo this procedure would not have fin infections.

Available information reports that laparoscopy is safe when carried out carefully. NMFS' evaluation of the laparoscopy under this action reveals that the CIs under the proposed permit have trained many other researchers at the Warm Springs National Fish Health Center and have routinely performed similar procedures on shortnose sturgeon without complication (Permit 1486).

The procedures would increase the risk of complications associated with the added stress of the surgical procedures and the extended time under anesthesia. Because the sutures used to close the laparoscopy sites penetrate the body wall, they would also provide a route of possible infection. To combat this, as small an incision as possible would be used, which would minimize the amount of suture necessary and decrease the healing time. Finally, suture ties would be kept as short as possible and disinfectant would be applied to the sutures prior to recovering the animal from anesthesia. This treatment would help prevent fungal growth on the sutures that could possibly infect the animal prior to healing of the incision wounds. We expect that the small incision and insertion of the laparoscope would have little probability of killing or producing sub-lethal effects as healing is rapid, although delayed complications are possible.

Effects of Biopsy. In some cases during laparoscopy, the sex of the fish is not readily apparent, so biopsies of the gonadal material could be taken for a definite sex determination.

This would only happen if sex is unclear, so an undetermined portion of the 24 shortnose sturgeon that undergo laparoscopy could also be biopsied. A 5 mm sized sample (2-3g) would be removed from each fish and placed in buffered formalin for preservation. Upon completion of the biopsy, the body cavity and biopsy site would be visually assessed to ensure that there was no obvious hemorrhaged or herniated tissue requiring additional attention. The incision would be sutured with a single suture in a cruciate pattern using PDS suture material.

Gonad samples do not cause disruptive hemorrhaging of the sampled site because of the lack of blood vessels in the vicinity of the sampled site. Further, sturgeon seem to return to completely normal behavior within a day or 2 after surgery (Jefferies 2005). Hernandez-Divers *et al.* (2004) conducted laparoscopic sex determinations, gonadal biopsies (5 mm sample taken) and various reproductive surgeries on hatchery-reared Gulf of Mexico sturgeon. The five male sturgeon that received gonadal biopsies survived the surgery and the authors concluded that the surgery was minimally invasive, safe and effective.

Because no formal studies of sublethal effects of gonadal biopsies on sturgeon exist we looked to similar studies for insight. Studies conducted by Ritchie (1965, 1970) evaluated the effects of gonadal biopsies on the survival and the survival and recapture rates of striped bass, respectively. Ritchie (1965) conducted biopsies on 20 wild fish (10 age 2 fish and 10 age 3 fish) while in the lab. The gonads were accessed through the urogenital pore and fish were not fed for the duration of the experiment to produce uniform stress and magnify any effects caused by the biopsies. Fish were also not anesthetized. All fish were sacrificed and received necropsies at the end of the experiment. At necropsy, 15% of the fish had unhealing gonad wounds (three fish), one of which was considered to be serious. Tests to determine the survival between the groups of fish were inconclusive. Ritchie (1970) conducted field tests of the effects of gonadal biopsy using the same procedures as in Ritchie (1965) on the survival of tagged striped bass. The author concluded that the biopsies did not alter the survival rate or the travel habits of the striped bass.

Expected Response to Biopsy. We believe that gonad biopsy would have a minimal impact on the shortnose sturgeon population of the Delaware River. Previous researchers have indicated that biopsy surgery is safe and effective and gonad samples do not have potential to cause disruptive hemorrhaging at the sample site. Studies conducted on other fish species reveal that survival rate after biopsy is either not altered, or has a very low incidence of chronic unhealed wound sites.

Effects of Blood Collection. Upon completion of laparoscopy to the 24 sturgeon each year, blood will be collected from the caudal vein. This is achieved by inserting a needle, attached to a syringe, perpendicular to the ventral midline at a point immediately caudal to the anal fin. The needle is slowly advanced while applying gentle negative pressure to the syringe until blood freely flows into the syringe. Once the blood is collected, direct pressure is applied to the site of venipuncture to ensure clotting and prevent subsequent blood loss (Stoskopf 1993).

Venipuncture is a simple way of drawing blood from shortnose sturgeon while they are undergoing a laparoscopy to perform blood analysis later. Venipuncture is non-lethal and is not expected to have any sub-lethal effects (Klinger *et al.* 2003). Effects of drawing blood samples with syringes from the caudal vein of shortnose sturgeon, could include pain, handling

discomfort, possible hemorrhage at the site or risk of infection. To mitigate these effects, the needle would be slowly advanced while applying gentle negative pressure to the syringe until blood freely flows into the syringe. Once the blood is collected, direct pressure would be applied to the site of venipuncture to ensure clotting and prevent subsequent blood hemorrhaging (Stoskopf 1993). The site would then be disinfected and checked again after recovery prior to release. Additionally, all of the researchers responsible for obtaining these samples will have received extensive experience in the procedure.

Expected Response to Blood Collection. As stated above, venipuncture is non-lethal and we do not expect this method to have sub-lethal effects. We acknowledge that pain, handling discomfort, possible hemorrhage at the site or risk of infection could occur, but procedure mitigation efforts (such as pressure and disinfection) lessen those possibilities. We believe that drawing blood in the manner described appears to have little probability of killing shortnose sturgeon or producing sub-lethal effects as long as the procedure is conducted by a qualified veterinarian or experienced biologist.

Effects of Hydroacoustic Testing. Hydroacoustic/sonar testing is requested to remotely scan and identify sturgeon while still in nets. Additionally, up to 20 shortnose sturgeon would be captured and tethered at different depths in soft nylon or cotton mesh sleeves for periods not exceeding two hours. There are two stressors we examined for this type of testing and methodology. The first hydroacoustic testing stressor we examined was possible effects to the sturgeon from sonar. The second hydroacoustic testing stressor we examined was the potential stress caused by tethering 20 sturgeon in the soft nylon or cotton mesh sleeves for up to two hours.

Hyrdoacoustics are frequently used for remotely locating fish for research and fishing, and it has been demonstrated that fish do not hear within common ranges used. Many studies have explored the use of sonar technology to identify sturgeon species underwater (Nealson and Brundage 2007, Auer and Baker 2007, Hartman and Nagy 2006, Tang *et al*. 2006, Auer *et al*. 2003, Grady *et al*. 2000). Studies show that, with few exceptions, fish cannot hear sounds above about 3-4 kHz, and the majority of species are only able to detect sounds to 1 kHz or even below (Popper 2008).

Tethering shortnose sturgeon in mesh sleeves is similar to what the fish would experience for net capture. They will be held underwater for no more than two hours and NMFS netting protocols would be adhered to.

Expected Response to Hydroacoustic Testing. We expect that shortnose sturgeon response to hydroacoustic testing would be similar to what has been reported above.
The proposed permit calls for the use of a broadband sonar system operating at 110-220 kHz. We believe that, due to the studies which show that most fish cannot hear sounds above 3-4 kHz, we do not believe that shortnose sturgeon will be affected by this frequency. Since tethering of shortnose sturgeon under the permit will be conducted according to netting conditions, we believe that tethering effects would be similar to netting effects as explained in the netting response section.

VIII. Cumulative Effects

Cumulative effects include the effects of future state, tribal, local or private actions that are reasonably certain to occur in the action area considered by this Opinion. Future federal actions that are unrelated to the proposed action are not considered in this section because they require separate consultation pursuant to section 7 of the ESA.

NMFS expects the natural and human-induced phenomena in the action area will continue to influence shortnose sturgeon as described in the Environmental Baseline. However, it is the combination and extent to which these phenomena will affect shortnose sturgeon that remains unknown.

Future federal actions as well as scientific studies contributing to conservation or recovery of shortnose sturgeon will require consultation under the ESA and such studies are not included in the *Cumulative Effects* section of this Opinion. Sources queried for the information on non-federal activities include the U.S. Census Bureau and Lexis-Nexis news and law online search engine. On Nexis, we reviewed bills passed from 2007-2010 and pending bills under consideration were included as further evidence that actions are reasonably certain to occur. In addition, statutes already in place that continue to provide the authority of state agencies to regulate anthropogenic effects were reviewed. State regulation is critical for future anthropogenic impacts in a region. Pending and existing legislation for the states of Delaware, Pennsylvania, New Jersey, and New York addresses oil spill prevention and response; water supply and water quality concerns; riparian and coastal development; ecosystem, natural resource, and endangered species recovery and protection; and regulation of fisheries and invasive species.

IX. Integration and Synthesis of Effects

As explained in the *Approach to the Assessment* section, risks to listed individuals are measured using changes to an individual's "fitness" – i.e., the individual's growth, survival, annual reproductive success, and lifetime reproductive success. When listed plants or animals exposed to an action's effects are not expected to experience reductions in fitness, we would not expect the action to have adverse consequences on the viability of the population(s) those individuals represent or the species those populations comprise (Brandon 1978, Mills and Beatty 1979, Stearns 1992, Anderson 2000). As a result, if the assessment indicates that listed plants or animals are not likely to experience reductions in their fitness, we conclude our assessment.

The narrative that follows integrates and synthesizes the information contained in the *Status of the Species*, the *Environmental Baseline*, and the *Effects of the Action* sections of this Opinion to assess the risk the proposed activities pose to shortnose sturgeon. There are known cumulative effects (i.e., from future state, local, tribal, or private actions) that fold into our risk assessment for this species.

The proposed issuance by PR1 of scientific research Permit No. 14604 to Harold Brundage, ERC, would authorize directed take of shortnose sturgeon in the Delaware River. The proposed activities under this permit include capture and handling, PIT and Floy tags, genetic tissue

sampling, anesthetization, laparoscopy, transmitter implantation, blood sample, and biopsy. One incidental mortality (or serious harm) would be authorized per year not to exceed three throughout the permit's duration. The *Status of listed resources* section identified the construction of dams throughout the shortnose sturgeon's range as the main factor that probably reduced their reproductive success which, in turn, could be the primary reason for the reduction in population size for shortnose sturgeon. However, dams are not the main factor reducing population size of the Delaware River population, due to the river's Wild and Scenic designation. Other threats to the survival and recovery of shortnose sturgeon include habitat fragmentation and loss, siltation, water pollution, decreased water quality (low DO, salinity alterations), bridge construction, dredging and blasting, incidental capture in coastal fisheries, impingement on intake screens of power plant operations, and land use practices. Reasonably likely future actions described in the *Cumulative effects* section include state legislation to address oil spill prevention and response; water supply and water quality concerns; riparian and coastal development; ecosystem, natural resource, and endangered species recovery and protection; and regulation of fisheries and invasive species. Recent population studies suggest that the Delaware River system shortnose sturgeon population is one of the larger stable stocks within its range, having an estimated spawning population of 12,000 adults (Brundage 2003).

The Delaware River population of shortnose sturgeon is one of the larger and healthier stocks within its range. As stated above, the most recent shortnose sturgeon population estimate on the Delaware River is an estimated spawning population of 12,000 adults (Brundage and O'Herron 2003). The anticipated impact of one sturgeon mortality (or serious harm) annually on the population, therefore, would be small based on the 2003 abundance estimate, or 0.008%. Taking into consideration that the Delaware River shortnose sturgeon population is one of the larger and healthier stocks within its range, NMFS believes that the allowance of one incidental mortality annually, not to exceed three throughout the life of the permit, when considering other external incidental mortalities, is unlikely to reduce the viability of the Delaware River population. Therefore, it is unlikely to reduce the viability of shortnose sturgeon as listed under the ESA.

Permit 14604 would be valid for five years from the date of issuance and would authorize non-lethal sampling methods on up to 1,000 adult and juvenile shortnose sturgeon (this is on a per year basis, with no more than 3,600 fish sampled over the life of the permit). Research activities would include: capturing via gill net, trammel net, and trawl net; measuring and weighing; tagging with PIT and Floy T-bar tags; and sampling tissue for genetic analysis. A subset of 30 adults and 30 juveniles would be anesthetized tagged with acoustic transmitters. Another subset of 24 adults would be examined internally using laparoscopic techniques and each would potentially also have gonad biopsy and blood sample taken for analysis. Another subset of 20 adults per year would be included in hydroacoustic gear testing. Additionally, lethal collection of up to 300 eggs and larvae each year (not to exceed 900 over five years) would take place during seasonal spawning activity by artificial substrate, D-frame ichthyoplankton net, and/or epibenthic sled. Finally, one unintentional mortality or serious harm event resulting from research is requested annually, not to exceed three over the life of the permit.

Although some degree of stress or pain is likely for individual fish captured, handled and tagged, and while tagging and tissue sampling will result in tissue injuries, none of the research procedures are expected to result in mortality or reduced fitness of individuals. However, due to

netting complications or anesthesia complications, one incidental mortality could occur per year with a cap of three throughout the life of the permit. Delayed or aborted spawning for some individual fish is a possibility, but the likelihood is remote given the mitigation measures proposed. The proposed permit is not expected to affect the population's reproduction, distribution, or numbers. Because the proposed action is not likely to reduce the Delaware River population's likelihood of surviving and recovering in the wild, it is not likely to reduce the species' likelihood of surviving and recovering in the wild.

IX. Conclusion

After reviewing the current status of endangered shortnose sturgeon, the environmental baseline for the action area, the effects of the proposed research program, and the cumulative effects, it is NMFS's biological opinion that the issuance of permit 14604 to Harold Brundage, ERC, is not likely to jeopardize the continued existence of the endangered shortnose sturgeon. Critical habitat has not been designated for shortnose sturgeon.

CONSERVATION RECOMMENDATIONS

Section 7(a)(1) of the Act directs Federal agencies to utilize their authorities to further the purposes of the Act by carrying out conservation programs for the benefit of endangered and threatened species. Conservation recommendations are discretionary agency activities to minimize or avoid adverse effects of a proposed action on listed species or critical habitat, to help implement recovery plans, or to develop information.

The following conservation recommendations would provide information that would improve the level of protections afforded in future consultations involving proposals to issue permits for research on the endangered shortnose sturgeon:

1. *Cumulative Impact Analysis.* Before authorizing any additional permits for activities similar to those contained in the proposed permits, F/PR1 should work with the shortnose sturgeon recovery team and the research community to develop protocols that would have sufficient power to determine the cumulative impacts (that is, includes the cumulative lethal, sub-lethal, and behavioral consequences) of existing levels of research on individuals populations of shortnose sturgeon.

2. *Estimation of actual levels of "take."* Before authorizing any additional permits for activities similar to those contained in the proposed permits, F/PR1 should review the annual reports and final reports submitted by researchers that have conducted research on shortnose sturgeon as well as any data and results that can be obtained from the permit holders. This should be used to estimate the numbers of shortnose sturgeon killed and harassed by these investigations, and how the harassment affects the life history of individual animals. The results of the study should be provided to F/PR3 for use in the consultations of future research activities.

REINITIATION NOTICE

This concludes formal consultation on the proposed permit to Harold Brundage, ERC (Permit No. 14604) pursuant to the provisions of section 10 of the Endangered Species Act. Reinitiation of formal consultation is required where discretionary Federal agency involvement or control over the action has been retained (or is authorized by law) and if: (1) the amount or extent of allowable take is exceeded; (2) new information reveals effects of the agency action that may affect listed species or critical habitat in a manner or to an extent not considered in this Opinion; (3) the identified action is subsequently modified in a manner that causes an effect to the listed species or critical habitat not considered in this Opinion; or (4) a new species is listed or critical habitat designated that may be affected by the action.

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