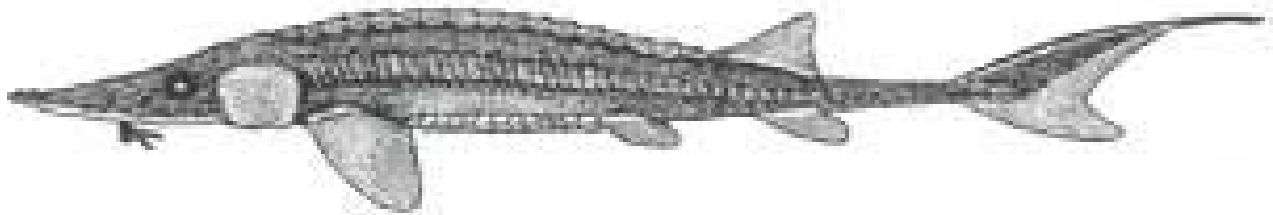


U.S. FISH AND WILDLIFE SERVICE  
DIVISION OF ENVIRONMENTAL QUALITY  
REGION 6

**A HEALTH RISK EVALUATION FOR PALLID STURGEON  
(*SCAPHIRHYNCHUS ALBUS*) IN THE LOWER PLATTE RIVER USING  
SHOVELNOSE STURGEON (*SCAPHIRHYNCHUS PLATORYNCHUS*) AS A  
SURROGATE.**

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U.S. Fish and Wildlife Service  
Nebraska Field Office  
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Grand Island, NE 68801  
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**A HEALTH RISK EVALUATION FOR PALLID STURGEON (*SCAPHIRHYNCHUS ALBUS*)  
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(*SCAPHIRHYNCHUS PLATORYNCHUS*) AS A SURROGATE.**

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## ABSTRACT

Most sturgeon species worldwide have been in steep decline since the 1900s. This research evaluated shovelnose sturgeon health, reproduction, and exposure to environmental contamination in the lower Platte River. Shovelnose sturgeon served as a surrogate species for the endangered pallid sturgeon and their health was assessed by incorporating measurements of general health with hepatic, immune, and reproductive system biomarkers. Environmental contaminants were measured in water, potential pallid sturgeon food items (cyprinid minnows), and shovelnose sturgeon digesta, liver, and blood plasma. Contaminants detected in shovelnose sturgeon at concentrations of concern included PCBs, selenium, and atrazine. Total PCBs in carcasses (n = 8) averaged 0.32 µg/g ww. Selenium averaged 4.8 µg/g dw in carcasses (n = 30) and 80 percent of individuals sampled were within the 4 to 6 µg/g threshold range for reproductive impairment in sensitive fish species. Pallid sturgeon food items had significantly (p < 0.05) greater concentrations of Hg, Se, and Zn than shovelnose sturgeon digesta. Atrazine was detected in all blood plasma samples analyzed (n = 50) at concentrations from 0.24 to 28 µg/L, but was not detected in liver (n = 19). Although the effects of atrazine exposure to shovelnose sturgeon are unknown, results of this study and previous work by others indicate that it may be disrupting steroidogenesis. Gross observations and condition indices seem to indicate that shovelnose sturgeon from the lower Platte River are healthy; however, reproductive biomarkers and histological examination of gonads indicate potential reproductive impairment as indicated by ovarian atresia, abnormal estrogen to testosterone ratios, and high concentrations of vitellogenin in males. Pallid sturgeon may be especially at risk to contaminants in the lower Platte River that bioaccumulate and cause reproductive impairment because they have a more piscivorous diet, greater maximum life-span, and a longer reproductive cycle than shovelnose sturgeon. Strategies to reduce shovelnose sturgeon and pallid sturgeon exposure to environmental contaminants in the lower Platte River are presented.

Key Words: Nebraska, pallid sturgeon, shovelnose sturgeon, *Scaphirhynchus*, Platte River, contaminants, selenium, PCBs, atrazine, reproduction.

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## ACRONYMS AND ABBREVIATIONS

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<	less than
>	greater than
°C	degrees Celsius
µS/cm	microSiemens per centimeter
µg/g	microgram per gram
µg/g	micrograms per gram
µg/L	micrograms per liter
µL	microliters
11KT	11-ketotestosterone
ACF	Analytical Control Facility
Al	aluminum
As	arsenic
B	boron
Ba	barium
Be	beryllium
BEST	Biomonitoring of Environmental Status and Trends program
BMPs	Best Management Practices
Cd	cadmium
CERC	Columbia Environmental Research Center
CF	condition factor
Cr	chromium
CTM	critical thermal maximum
Cu	copper
DDT	dichlorodiphenyltrichloroethane
DO	dissolved oxygen
dw	dry weight
e.g.	example given
E/T	17-beta estradiol to 11-ketotestosterone ratio
E <sub>2</sub>	17 -beta estradiol
ELISA	enzyme-linked immunosorbent assay
ERED	Environmental Residue-Effects Database
EROD	Ethoxyresorufin- <i>O</i> -deethylase
et al.	and others
FCSC	Florida Caribbean Science Center
Fe	iron
FL	fork length

g	gram
GSI	gonado-somatic index
HACs	hormonally active compounds
Hg	mercury
hr	hour
HSI	hepato-somatic index
HSI <sub>GI</sub>	hepato-somatic index (gonad weight included)
Hwy	highway
i.e	in explanation
ID	identification
IUCN	World Conservation Union
kg	kilogram
LOD	limit of detection
LOEC	lowest observed effects concentration
LOQ	limit of quantification
MAs	macrophage aggregates
Mg	magnesium
mg/kg	milligrams per kilogram
mg/L	milligrams per liter
mg/ml	milligrams per ml
ml	milliliter
mm	millimeter
Mn	manganese
Mo	molybdenum
n	sample size
NASS	National Agricultural Statistics Service
NAWQA	National Water Quality Assessment Program
NDEQ	Nebraska Department of Environmental Quality
NE	Nebraska
ng/L	nanograms per liter
Ni	nickel
NPDES	National Pollutant Discharge and Elimination System
NWIS	National Water Information System
NWQL	National Water Quality Laboratory
OC	organochlorine
p	level of significance
PACF	Patuxent Analytical Control Facility
PAHs	polycyclic aromatic hydrocarbons
Pb	lead

PCBs	polychlorinated biphenyls
pers. comm.	personal commentary
pg/g	picograms per gram
pg/ml	picograms per milliliter
pmol/min/mg	picomoles per minute per milligram
QA/QC	quality assurance and quality control
RBCs	red blood cells
SE	standard error
Se	selenium
Service	U.S. Fish and Wildlife Service
SP	<i>Scaphirhynchus platorynchus</i>
Sr	strontium
SSI	spleno-somatic index
TCDD-EQs	dioxin equivalents
UNL	University of Nebraska at Lincoln
USDOI	U.S. Department of the Interior
USEPA	U.S. Environmental Protection Agency
USFDA	U.S. Food and Drug Administration
USGS	U.S. Geological Survey
V	vanadium
VTG	vitellogenin
W	observed body mass
W <sub>G</sub>	gonad weight
W <sub>r</sub>	relative weight
WRD	Water Resources Division
W <sub>s</sub>	standard weight
ww	wet weight
Zn	zinc

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## INTRODUCTION

### Nature and Scope of the Problem

Sturgeon (family Acipenseridae) have been on earth for over 175 million years (Choudhury and Dick, 1998); however, there have been steep declines in most sturgeon species worldwide since the 1900s and currently 23 sturgeon species are listed on the World Conservation Union's (IUCN) Red List of Threatened Species (IUCN, 2004). In North America there are three river sturgeon species (genus *Scaphirhynchus*). They include the federally endangered pallid sturgeon *S. albus*, the federally endangered Alabama sturgeon *S. suttkusi* and the shovelnose sturgeon *S. platyrhynchus*. The Missouri and Platte rivers in Nebraska provide important habitat for two of these three species, the pallid sturgeon and the shovelnose sturgeon.

In 1993, the U.S. Fish and Wildlife Service (Service) Pallid Sturgeon Recovery Plan listed habitat loss, environmental contaminants, commercial fishing, and hybridization as the four suspected primary reasons for pallid sturgeon population declines (Dryer and Sandvol, 1993). Over the last decade, pallid sturgeon catch records have continued to be low with little evidence of reproduction within the last 20 years (Steven Lydick, Service Fisheries Biologist, pers. comm., 2004). Furthermore, the rate of decline in the Missouri River population of pallid sturgeon between Fort Peck Dam and the headwaters of Lake Sakakawea indicates that they are likely to be extirpated from these areas by 2018 (Kapuscinski, 2003). Currently, artificial propagation is being used to supplement wild populations until suitable spawning conditions and habitat in the wild can be restored.

Although shovelnose sturgeon are currently abundant in Nebraska's lower Platte River, their overall population and range has declined both nationally and in Nebraska. A decrease in the range and abundance of shovelnose sturgeon in the Mississippi Valley has been attributed to impoundments, overfishing, and pollution (Bailey and Cross, 1954 as cited by Moos, 1978). Shovelnose sturgeon are no longer found in Pennsylvania, New Mexico, and large parts of Kansas, Kentucky, Tennessee, and other States where they

were once abundant (National Paddlefish and Sturgeon Steering Committee, 1992). In Nebraska, shovelnose sturgeon used to inhabit the entire Platte River and the North Platte River westward into Wyoming (Moos, 1978); however, today they are restricted to the lower Platte River and the Elkhorn river (Steven Lydick, Service Fisheries Biologist, pers. comm., 2004).

Water quality in the lower Platte River is degraded by environmental contaminants. The Nebraska Department of Environmental Quality (NDEQ) listed the entire lower Platte River on Nebraska's 303(d) list of impaired water bodies due to contamination by polychlorinated biphenyls (PCBs), selenium, and fecal coliforms, (NDEQ, 2004). In addition, a national reconnaissance study on potential endocrine disruption in common carp (*Cyprinus carpio*) reported that the lower Platte River had the highest concentrations of dissolved pesticides in water when compared to 24 other sites in the U.S. (Goodbred et al., 1997). The same study reported that the lowest estrogen to testosterone ratios in both male and female carp were found in the Platte River at Louisville, indicating potential endocrine disruption (Goodbred et al., 1997).

The lower Platte River is believed to provide important spawning habitat for both the shovelnose sturgeon and the pallid sturgeon; however, degraded water and habitat quality may be adversely affecting their health and reproduction. This study is the first to evaluate whether environmental contamination in the lower Platte River may be adversely affecting shovelnose sturgeon and pallid sturgeon health and reproduction.

### Study Area

The lower Platte River is a 160 km stretch of the Platte River from Columbus, Nebraska to the confluence of the Missouri River (Figure 1). This stretch of the Platte River supports an abundant population of shovelnose sturgeon that is commonly targeted by anglers. Creel surveys along the lower Platte River during 1992 and 1993 reported that shovelnose sturgeon comprised 4 and 5.3 percent of angler catch and ranked fourth and third in catch abundance, respectively (Holland and Peters, 1994). The number of pallid sturgeon that use the lower Platte River has not been officially estimated, but



anglers reported 4 and 11 captures of pallid sturgeon in 2003 and 2004, respectively (Darrell Feit, Nebraska Game and Parks Commission, pers. comm., 2004). Platte River pallid sturgeon populations have been augmented by the release of 500 to 600 hatchery-propagated fish over a period of three years beginning in 1997 (Lutey, 2001). The lower Platte River from the confluence of the Elkhorn River to the confluence of the Missouri River has been designated as one of six Recovery Priority Management Areas within the historical range of the pallid sturgeon (Dryer and Sandvol, 1993). In addition, radio telemetry tracking of hatchery-reared pallid sturgeon indicates that the lower Platte River below the confluence of the Loup River near Columbus, Nebraska, could also provide conditions necessary for pallid sturgeon survival (Lutey, 2001).

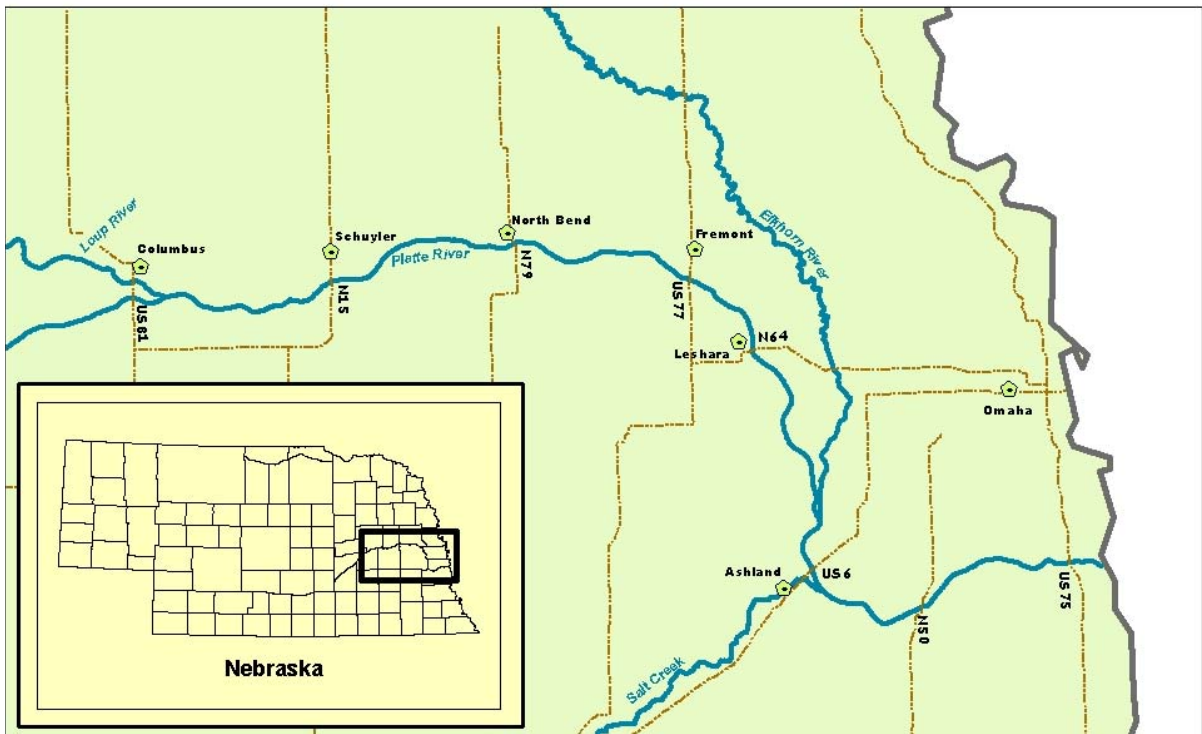


Figure 1. Location of the lower Platte River from the confluence of the Loup River near Columbus, NE, to its mouth at the Missouri River (Nebraska State line).

## Shovelnose Sturgeon as a Surrogate for Pallid Sturgeon

Pallid sturgeon are extremely rare; therefore, the shovelnose sturgeon is frequently used as a surrogate species to evaluate contaminant exposure and effects in areas where the two species overlap (Ruelle and Keenlyne, 1994; Palawski and Olsen, 1996; Conzelmann, 1997; Coffey et al., 2003). The shovelnose sturgeon is closely related to the pallid sturgeon, and the two species are known to hybridize in the wild (Carlson et al., 1985; Ruelle and Keenlyne, 1994). Pallid and shovelnose sturgeon life history characteristics, body condition, physiology, genealogy, and contaminant bioaccumulation have been compared (Ruelle and Keenlyne, 1994). Shovelnose sturgeon serve as a suitable surrogate species for pallid sturgeon in that they both live for 20 years or longer, inhabit the same river basins, spawn at similar intervals, and locations, and accumulate similar inorganic and organic contaminants (Ruelle and Keenlyne, 1994). Limitations of the shovelnose sturgeon as a surrogate species are that pallid sturgeon have a longer life-span, attain a larger size, feed mainly on fish (shovelnose sturgeon feed primarily on invertebrates), and contain a higher percentage of body fat (Ruelle and Keenlyne, 1994).

## Research Objectives

The primary goal of this study was to evaluate environmental risk to pallid sturgeon that occupy the lower Platte River, by using the shovelnose sturgeon as a surrogate species. Shovelnose sturgeon health was evaluated by performing a necropsy-based health assessment according to protocols established by the U.S. Geological Survey's (USGS) Biomonitoring of Environmental Status and Trends (BEST) program (Schmitt et al., 1999; Schmitt and Dethloff, 2000). This health assessment combines measurements of general health with hepatic, immune, and reproductive system biomarkers. Exposure to elemental contaminants and organochlorine contaminants was evaluated by determining their concentrations in shovelnose sturgeon carcasses. Potential differences in elemental contaminant and organochlorine exposure between shovelnose sturgeon and pallid sturgeon were evaluated by comparing residues in shovelnose sturgeon digesta and potential pallid sturgeon food items (i.e., cyprinids < 5

inches long). Shovelnose sturgeon exposure to triazine pesticides was evaluated by measuring concentrations in shovelnose sturgeon liver and blood plasma as well as water samples from the lower Platte River.

We hypothesized that pallid sturgeon food items would have higher concentrations of contaminants than shovelnose sturgeon food items, and that shovelnose sturgeon adverse health effects (e.g., presence of disease, tissue abnormalities, spleen macrophage aggregates, liver enzyme induction, and an abnormal profile of sex steroids and vitellogenin in plasma) would be directly related to concentrations of contaminants in water and/or shovelnose sturgeon tissues and food items.

## METHODS

### Collaboration with Concurrent Research

Shovelnose sturgeon were collected in collaboration with researchers at the University of Nebraska in Lincoln (UNL). During this investigation, UNL was performing a 5-year study (2001 to 2005) titled "Ecology and management of the pallid sturgeon and sturgeon chub in the lower Platte River." Their research objectives in 2002 included monitoring movement of shovelnose sturgeon and pallid sturgeon using radio-telemetry, aging shovelnose sturgeon by collecting and examining pectoral fin rays, and measuring water quality parameters such as temperature, specific conductivity, and dissolved oxygen (DO). Their data on sturgeon movements and age were incorporated into this study to strengthen the assessment of shovelnose sturgeon health and exposure to environmental contaminants.

The USGS National Water Quality Assessment Program (NAWQA) monitored stream flow and water quality parameters at several locations on the lower Platte River in 2002. Daily stream flow data were measured at four sites (North Bend, Leshara, Ashland, and Louisville) and water temperature, DO, and specific conductivity were measured at two sites (Leshara and Louisville). This information is publicly available in the National Water Information System (NWIS) database (NWIS, 2004).

### Sample Collection and Handling

Shovelnose sturgeon were captured by drifting gill nets or by towing a trawl net and were temporarily held in storage tanks (54 quart Rubbermaid containers) before processing. Two to 6 milliliters (ml) of blood were collected from the caudal artery and vein with a 5 ml syringe and a 21 gauge needle. Fish were then euthanized by striking the cranium with a blunt metal rod and body mass and fork length were measured to the nearest kilogram (kg) and millimeter (mm), respectively. Standard measurements for a morphometric character index (Sheehan et al., 1999) were recorded. This was followed by an external examination for lesions, parasites, tumors or other anomalies on the body surface, eyes, opercles, gills, pseudobranches, and fins. An internal necropsy-based assessment was used to evaluate the condition of the liver, gall bladder, kidneys, spleen, gonads, and mesenteric fat. Digital photographs of the ventral side of each fish were taken before and after the opening incision. The right pectoral fin was removed at the point of articulation, placed into a paper envelope and given to UNL for age determination. Liver and gonads were weighed to the nearest 0.1 gram (g) and spleen to the nearest 0.01 g. The right gonad was placed in a Whirl-Pak<sup>®</sup> bag and stored frozen for potential future contaminant analysis. Tissues collected for histology included kidney, liver, spleen, and left gonad (see histology methods below). Three pieces of liver, each approximately 1 cubic centimeter in size, were collected to measure Ethoxyresorufin-O-deethylase (EROD) activity. These samples were contained in a 1.5 ml cryogenic vial and quickly frozen in a mixture of 200 proof ethanol and dry ice. Blood samples were centrifuged for 10 minutes at 3,500 revolutions per minute to form a plasma fraction. The plasma fraction was aspirated into cryogenic vials and quickly frozen in an ethanol/dry ice slush. Liver samples for EROD analysis and blood plasma samples for hormone/atrazine analysis were stored at -80 degrees Celsius (°C) and then shipped overnight to the Columbia Environmental Research Center (CERC) and the Florida Caribbean Science Center (FCSC), respectively. Each shovelnose sturgeon carcass sample (the remaining tissues minus the gonad, kidney, liver, spleen, gill, and pectoral fin samples mentioned above), was wrapped in aluminum and stored in a freezer (-20 °C) at

the Nebraska Ecological Services Field Office. These carcass samples were later shipped frozen to contract labs for homogenate preparation and analytical analysis.

To evaluate dietary uptake of contaminants, stomach contents from shovelnose sturgeon were collected during necropsy. In addition, potential pallid sturgeon food items (i.e., cyprinids less than 5 inches in length) were collected by seining in areas where shovelnose sturgeon were collected. These samples were placed in chemically clean glass containers and were also shipped frozen to contract labs for analytical analysis.

### Age Determination

Shovelnose sturgeon age was estimated at UNL and a detailed method description is provided by Shuman (2003). In brief, cross-sections for each fin-ray were mounted on slides following procedures developed by Rein and Beamesderfer (1994) and annuli were counted following procedures developed by Carlson et al. (1985) and Hurley (1996). Final age estimation for each fish analyzed was determined by averaging the age assessments of three independent research technicians at UNL.

### Hepatic EROD Activity

EROD activity in fish liver has been extensively used as a biomarker of exposure to environmental contaminants including polycyclic aromatic hydrocarbons (PAHs), PCBs, and dioxins (reviewed by Whyte et al., 2000). In this study, shovelnose sturgeon liver microsomal preparation and EROD assay methods followed standard procedures developed by CERC (Nicks et al., 2003). The microsome preparation for each sample was followed by an EROD assay on the same day, and instrument variation among days was checked against positive control samples. EROD activity was measured on a Perkin-Elmer BioSystems Cytofluor 4000 plate reader. The mean specific activity for the positive control samples run in this study was 401 picomoles per minute per milligram (pmol/min/mg) and all positive control samples fell within two standard deviations of the CERC lab mean (Nicks et al., 2003). Detection and quantification limits were calculated for each day samples were run. The limit of detection (LOD) ranged from 0.4 to 0.7 and

the limit of quantification (LOQ) ranged from 0.9 to 1.9. A more detailed description of our study methods for measuring EROD activity in shovelnose sturgeon and quality assurance/quality control (QA/QC) procedures and results are provided by Nicks et al. (2003).

#### H4IIE Bioassay

The H4IIE bioassay measures EROD activity in cultured rat liver cells exposed to an environmental extract and is an accurate and precise bioanalytical screening tool for dioxin-like environmental contaminants (Whyte and Tillitt, 2000; Whyte et al., 2004). For this study, the purpose of the H4IIE bioassay was to quantify the cumulative presence of EROD-inducing compounds in shovelnose sturgeon. Shovelnose sturgeon carcasses were shipped frozen to MSCL where they were homogenized individually. A 20 gram aliquot from each shovelnose sturgeon homogenate was combined with anhydrous sodium sulfate, dehydrated overnight, and then extracted with methylene chloride. Extracts were shipped by Fed Ex overnight to CERC where the H4IIE bioassay was performed. H4IIE cells were seeded in 96-well microtiter plates and grown for 24 hrs before being dosed with serial dilutions of the carcass extracts or a 2,3,7,8-TCDD standard. Cells were then incubated for 72 hrs and assayed for EROD induction using a Perkin-Elmer BioSystems Cytofluor 4000 plate reader. Concentration-response curves for each extract were compared to that of the dioxin (2,3,7,8-TCDD) standard. Dioxin-like extract potencies were measured as 2,3,7,8-TCDD equivalents (TCDD-EQs) in picograms per gram (pg/g) by comparing the extracts' slope values to the slope of the 2,3,7,8-TCDD standard.

QA/QC procedures included replication of assayed samples, comparison of calibration against known standards, proper maintenance and calibration of equipment, accurate sample tracking and chain of custody, proper documentation at all steps of sample processing, and other considerations of Good Laboratory Practices defined in the Code of Federal Regulations Title 40 Part 160. All samples were analyzed on the same day and the LOD and LOQ equaled 0.7 to 1.8 pg/g, respectively. A more detailed

description of our study methods for the H4IIE bioassay and QA/QC procedures and results are provided by Nicks et al. (2005).

### Histopathology

Approximately 1 cubic centimeter sections of liver, spleen, trunk kidney, head kidney, and left gonad were stored in 10 percent buffered formalin and sent via overnight delivery to CERC for analysis. Methods for tissue processing, staining, and analysis are described in more detail elsewhere (Papoulias et al., 2004). In brief, tissues were stained with hematoxylin and eosin and examined using an inverted Nikon Diaphot microscope. Pathology lesions were rated as either 1 = minimal, 2 = mild; 3 = moderate, and 4 = moderately-severe as illustrated by Papoulias (2004). Macrophage aggregates (MAs) in liver were quantified for each fish using Optimas® computer-based image analysis software. A liver sub-sample was stained with Prussian Blue to identify the pigments represented in the MAs.

Male and female gonads were staged according to the classification system of Moos (1978): Stage I = immature; Stage II = developing; Stage III = yolk deposition, or spermatogenesis; Stage IV = pre-spawning; Stage V = spawning; Stage VI = spent.

The six stages for testes developed by Moos (1978) were based on the examination of 309 testes and are briefly summarized herein as follows: Stage I) testes were smaller than other stages and comprised of 85 to 90 percent adipose tissue, Stage II) testes generally had less than 50 percent adipose and were marked by narrow bands of yellowish-white to gray testicular tissue, Stage III) testes had actively dividing spermatocytes and developing spermatozoa, Stage IV) testicular lobules were mostly filled with clusters of spermatozoa, Stage V) testes were creamy white in color and packed with spermatozoa, Stage VI) testes were similar in color to Stage V but were flaccid and typically more than 50 percent smaller in volume.

Ovary stages classified by Moos (1978) were based on examination of 245 females and are briefly summarized herein as follows: Stage I) smaller oocytes compared to other stages and lamellar structure was not obvious, Stage II) yellow adipose tissue contained 25 to 50 percent of the ovarian volume, Stage III) ovaries had larger

oocytes that exhibited vitellogenesis, Stage IV) ovaries filled most of the body cavity and the largest eggs were pigmented, Stage V) eggs larger than other stages and were pigmented, Stage VI) ovaries were flaccid and had ruptured follicles.

#### Plasma Hormones, Vitellogenin, and Atrazine

Blood plasma samples were analyzed by FCSC for two sex hormones (17 beta-estradiol and 11-ketotestosterone), vitellogenin (a precursor protein for yolk synthesis), and atrazine. Hormone concentrations were measured by radioimmunoassay following methods described by Goodbred et al. (1997). Plasma samples that were approximately 50 microliters ( $\mu\text{L}$ ) in volume, were extracted twice with 5 ml of diethyl ether and added to a reaction solution of radiolabeled hormone and a corresponding hormone-specific antibody. After a 24-hr incubation period, the non-antibody bound radiolabeled hormone was removed by adding charcoal dextran followed by centrifugation. The remaining bound radiolabeled hormone was measured using scintillation spectrophotometry. Cross-reactivities of the 17 beta-estradiol ( $E_2$ ) antiserum with other steroids were: 11.2 percent for estrone; 1.7 percent for estradiol; less than 1.0 percent for 17 alpha-estradiol and androstenedione, and less than 0.1 percent for all other steroids examined. Cross-reactivities of the 11-ketotestosterone (11KT) antiserum with other steroids were: 9.7 percent for testosterone, 3.7 percent for androstenedione, and less than 0.1 percent for all other steroids examined. Minimum detection limits were 16.3 and 19.5 picograms per ml (pg/ml) for  $E_2$  and 11KT, respectively. Coefficients of variation were 8.7 percent and 11.2 percent for  $E_2$  and 9.1 percent and 10.5 percent for 11KT for inter and intra-assay variation, respectively.

Vitellogenin (VTG) concentrations in plasma of shovelnose sturgeon were quantified by capture enzyme-linked immunosorbent assay (ELISA) as described by Goodbred et al. (1997). In brief, a purified carp monoclonal antibody (Mab HL 1147 2D3-3A9) was used to capture shovelnose sturgeon VTG. Bound VTG was then disclosed by a rabbit anti-vitellogenin polyclonal antibody (OF114) which was in turn disclosed by a goat anti-rabbit immunoglobulin class G linked to alkaline phosphatase.



An automated ELISA quantified the intensity of yellow color at 405 nm and VTG concentrations were calculated from standard curves.

Atrazine concentrations in blood plasma also were quantified using an ELISA procedure (RaPID atrazine test kit, Strategic Diagnostics Inc., Newark, DE, USA). In brief, samples were mixed with an enzyme conjugate (enzyme labeled atrazine) followed by paramagnetic particles attached with antibodies specific to atrazine. Atrazine and other related triazine herbicides in the sample compete with the enzyme labeled atrazine for antibody binding sites on the magnetic particles. At the end of a 15 minute incubation period, a magnetic field is applied and unbound reagents are decanted. The presence of atrazine is detected by adding a color reagent. The color developed is quantified by a spectrophotometer and is inversely proportional to the concentration of atrazine in the sample. The atrazine ELISA kit was validated for sturgeon blood plasma samples by FCSC by taking a pooled set of sturgeon plasma and running a dilution curve against their standard curve. The method detection limit for this assay was 0.001 nanograms per liter (ng/L). Cross-reactivity data for several triazine compounds are reported by the manufacturer (Strategic Diagnostics Inc., <http://www.sdix.com>). Recoveries were calculated at greater than 95 percent and coefficients of variance were 6.7 percent for results in this report

### Analytical Analyses

Samples collected by Service Personnel for analytical quantification of contaminant residues were submitted to the Patuxent Analytical Control Facility (PACF), since renamed the Analytical Control Facility (ACF) (Appendix Table A.1). Detailed descriptions of lab methods including sample preparation, sample digestion, QA/QC results, and detection limits are provided in the PACF catalogs which are available upon request (ACF phone:304-876-7336). In brief, the analysis of duplicate samples, spiked samples, and standard reference materials indicated acceptable levels of precision and accuracy and limits of detection were within ACF's contract requirements (ACF, 2005). Integrated equal width and depth water samples collected by the USGS Water Resources Division (WRD) in Lincoln, Nebraska, were analyzed by the USGS National Water

Quality Laboratory (NWQL). Field blanks, surrogate samples, spiked samples, and standard reference materials were used to validate acceptable levels of precision and accuracy (Glodt and Pirkey, 1998). Percent recovery for the USGS “Blind Sample Program” also is available from May to June 2002 when samples were collected (<http://bqs.usgs.gov/OBSP/index.html>).

Elemental Contaminants. For elemental contaminants analyses, all samples were freeze dried, percent moisture was determined, and results were provided as wet weight (ww) and dry weight (dw) concentrations. Inductively coupled plasma atomic emission spectrometry was used to determine concentrations of aluminum (Al), boron (B), barium (Ba), beryllium (Be), cadmium (Cd), chromium (Cr), copper (Cu), iron (Fe), magnesium (Mg), manganese (Mn), molybdenum (Mo), nickel (Ni), lead (Pb), strontium (Sr), vanadium (V), and zinc (Zn). Mercury (Hg) concentrations were determined by cold vapor atomic absorption, and graphite furnace atomic absorption was used to measure arsenic (As), selenium (Se), and small concentrations of Pb and Cd.

Organochlorine Chemical Residues. The organochlorine (OC) scan included analysis of hexachlorobenzene, total PCBs (Aroclors 1260, 1254, 1248, and 1242), lindane (alpha, beta, delta, and gamma), chlordane compounds (alpha chlordane, gamma chlordane, cis-nonachlor, trans-nonachlor, and oxychlordane), heptachlor epoxide, dieldrin, endrin, mirex, toxaphene, and dichlorodiphenyltrichloroethane (DDT) p,p' and o,p' isomers and their metabolites (o,p'-DDD, o,p'-DDE, p,p'-DDD, and p,p'-DDE). Quantification methods included electron capture gas chromatography, gas-liquid chromatography, and gas chromatography/mass spectrometry.

Herbicides in Water. Concentrations of triazine herbicides were measured in equal width and depth integrated water-column samples. These samples were collected in May and June by USGS WRD at three bridge locations (NE Highway 50 bridge near Louisville, Highway 64 bridge near Leshara, and Duncan Rd. Bridge near Duncan). Samples were analyzed for atrazine and its metabolites (i.e., desisopropyl atrazine and deethyl atrazine) by the USGS WRD. In addition, water grab samples were collected at each shovelnose sturgeon collection site by Service personnel and submitted to PACF. Concentrations of atrazine, cyanazine, metribuzin, norflurazone, propazine, simazine,

desethyl atrazine, desisopropyl atrazine, and metolachlor were determined by gas chromatography. The limit of detection for all herbicides tested was 0.1 micrograms per liter ( $\mu\text{g/L}$ ). Water grab samples from each sturgeon collection site also were analyzed by Servi-Tech Laboratories in Hastings, Nebraska, for major ions, cations, hardness, alkalinity, pH, and conductivity. Specific conductivity, dissolved oxygen, and temperature were measured with a handheld YSI-85 meter during the collection of water grab samples and while drifting gill nets for shovelnose sturgeon.

Liver Herbicide Scan. The herbicide scan of liver samples included atrazine, cyanazine, metribuzin, norflurazone, propazine, simazine, desethyl atrazine, desisopropyl atrazine and metolachlor. Samples were desiccated overnight, extracted in a PRQ Accelerated Solvent Extractor and then concentrated to dryness for lipid determination by Gel Permeation Chromatography. The final fraction was then concentrated by Turbovap, exchanged into hexane and quantified by a Varian Model 3400 Gas Chromatograph. There was insufficient sample to analyze in duplicate, and small sample masses resulted in the high LODs ( $109 \mu\text{g/g dw}$  and  $0.05\mu\text{g/g ww}$ ).

#### Condition and Organo-somatic Indices

A condition factor (CF), hepato-somatic index (HSI), spleno-somatic index (SSI), and gonado-somatic index (GSI) was calculated for each shovelnose sturgeon collected. These measurements are standard procedures in fish physiology studies and are part of the BEST monitoring program (Schmitt and Dethloff, 2000; Schmitt, 2002).

Condition factor relates fish weight to length and is used to evaluate an organism-level response to factors such as nutritional status, pathogen effects, and toxic chemical exposure (Schmitt and Dethloff, 2000). In this study a Fulton-type CF (Lagler, 1956) was calculated using the equation:  $CF = [(100,000)(W)]/FL^3$ , where W = observed body mass in grams and FL = fork length in mm.

The organo-somatic indices (HSI, SSI, and GSI) are a function of organ weight to body weight and are useful in evaluating response to environmental contaminants, immune function, and reproductive condition. GSI was calculated as a percentage of gonad weight ( $W_G$ ) to body weight using the equation  $(W_G / W)(100)$ . Gonad size can

vary greatly depending on gender and stage; therefore, the gonad paired weight was subtracted from W to calculate the HSI and SSI (e.g.,  $SSI = [\text{spleen weight}/(W - \text{gonad weight})](100)$ ). However, to compare the HSI among shovelnose sturgeon collected from this study to those from the Mississippi River (Coffey et al., 2003), a  $HSI_{GI}$  was calculated by including the gonad weight as part of W (i.e.,  $HSI_{GI} = (\text{liver mass}/W)(100)$ ).

Relative weight ( $W_R$ ) is an alternative method for evaluating fish condition that is available for fish species in which a standard weight equation has been characterized. A standard weight ( $W_S$ ) was calculated for each shovelnose sturgeon in this study using the standard weight equation:  $\log_{10}W_S = -6.287 + 3.330 \log_{10} FL$  (Quist et al., 1998). Relative weight was then calculated as  $W_R = (100)(W)/W_S$  (Wege and Anderson 1978).

### Data Analysis

Research on shovelnose sturgeon movements in the lower Platte River during the course of this study (Swigle, 2003) indicates that shovelnose sturgeon collected from these sites most likely did not move between sites during the collection period (17 days). Therefore, data analysis consisted of comparisons among sites for shovelnose sturgeon parameters likely influenced by recent exposure (i.e., concentrations of atrazine, vitellogenin, and hormones in blood and liver EROD activity). Parameters that are potentially influenced by life-long exposures (e.g., bioaccumulation of elemental contaminants and histological lesions) were pooled from all sites.

A reference population of shovelnose sturgeon not likely exposed to environmental contaminants was not available for comparison. Therefore, data interpretation relied on comparisons with other contaminant studies that included shovelnose sturgeon from the Mississippi River Basin (Coffey et al., 2003; Ruelle and Henry, 1994a), white sturgeon (*Acipenser transmontanus*) from the Columbia River (Foster et al., 2001), and carp from USGS studies (Goodbred et al., 1997; Schmitt 2002). Comparisons between genders, contaminant uptake, and biomarker response were analyzed. Statistical calculations were performed with JMP<sup>®</sup> Version 5 software (JMP, 2002). Where means are provided, the “±” refers to a standard error unless otherwise noted.

## RESULTS and DISCUSSION

### Shovelnose Sturgeon Collections

A total of fifty-three shovelnose sturgeon were collected from five stretches of the lower Platte River between May 28 and June 13, 2002 (Figure 2). All shovelnose sturgeon collected were mature adults (33 males and 20 females) and their body mass averaged  $720 \pm 26$  grams and ranged from 400 to 1200 grams (Appendix Table A.2). Age was estimated for 44 shovelnose sturgeon, averaged  $8.8 \pm 0.4$  years, and ranged from 5 to 14 years.

The sex ratio slant towards males was not unusual as males tend to be captured more frequently than females during the spawning season (Barnickol and Starrett, 1951; Moos, 1978). Morphometric measurements indicate that almost all sturgeon collected were non-hybridized shovelnose sturgeon; however, at least one sturgeon collected (SP-36) had a morphological character index below 0.03, and was possibly a shovelnose sturgeon/pallid sturgeon hybrid (Sheehan et al., 1999).

Gross observations during the external examination found that 29 of 53 fish had no abnormalities whereas the remaining 24 fish had minor visible lesions that included short opercles ( $n = 4$ ), yellow skin discoloration ( $n = 3$ ), an opaque eye ( $n = 1$ ), and damaged or eroded fins ( $n = 17$ ) that were apparently not from recent capture by netting. There were no visible growths, skin ulcers, or skeletal deformities. The yellow skin discoloration was observed on the last three sturgeon collected (two males and one female) from the Platte River near Columbus, and its cause or significance is unknown. No external parasites were observed; however, internal parasites (Class: Nematoda) were found in two fish. Internal gross observations found that 44 of 53 shovelnose sturgeon had little or no gonad fat. A lack of fat reserves can indicate poor nutritional health (Ruelle and Henry, 1994a) or may reflect increased energy use during migration to spawning areas (Griffiths, 2002; Hendry and Beall, 2004).

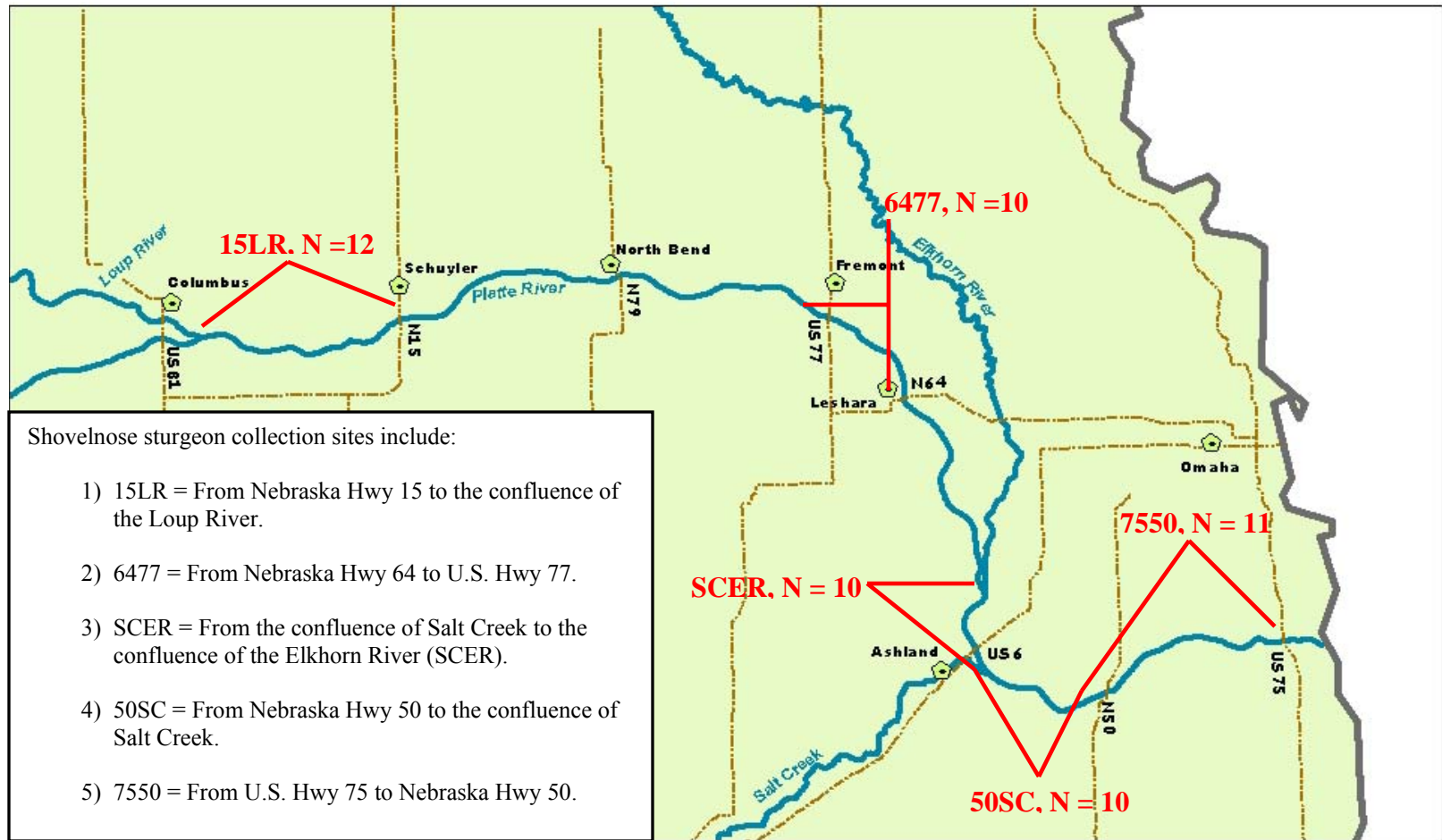


Figure 2. Reaches of the lower Platte River where shovelnose sturgeon were collected and the number of fish captured (n) from each reach.

## Water Quality

Water temperature, DO, and specific conductivity in the lower Platte River were measured by UNL researchers on 11 days between May 21 and June 20, 2002 (Appendix Table A.3). In addition, USGS-NAWQA researchers measured water temperature, specific conductivity, and DO in the lower Platte River at Leshara and Louisville in May and June of 2002 (NWIS, 2004).

Water temperatures measured by UNL averaged  $24 \pm 0.4$  °C and ranged from 15.7 to 29.7 °C (n = 87). At the two NAWQA monitoring stations water temperature averaged  $24 \pm 3$  °C and ranged from 14 to 34 °C (n = 6). The Nebraska water quality standard for warm water aquatic life (a maximum temperature of 32 °C; NDEQ, 2002) was exceeded on one occasion at the Leshara site. These average temperatures were warmer than the historic mean temperatures for the months of May ( $18 \pm 0.5$  °C; n = 74) and June ( $22 \pm 0.4$ ; n = 82) as measured by the NAWQA monitoring station at Louisville from 1973 through 2003 by the National Water Information System (NWIS, 2004).

The results of this study indicate that water temperatures in the lower Platte River during the spawning period of 2002 were warmer than previously reported as ideal for successful shovelnose sturgeon spawning. The spawning period for shovelnose sturgeon is believed to be less than a month during spring when water temperatures are between 16.9 and 21.5 °C (Elser et al., 1977; Christenson, 1975, both as cited by Moos, 1978). Moos (1978) reported that during the shovelnose sturgeon spawning period on the Missouri River, water temperatures remained around 18 - 19 °C for the month of June, and that the summer maximum temperature in both 1968 and 1969 was 24 °C in July, which equaled the average temperature recorded in this study.

Laboratory and field observations of other sturgeon species indicate that minor changes in water temperatures can adversely affect spawning efforts. Gulf sturgeon (*Acipenser oxyrinchus*) stop spawning if water temperature exceeds 21 to 22 °C (Sulak and Clugston, 1997). Lake sturgeon (*Acipenser fulvescens*) left spawning beds in the Gull River when water temperature dropped from 14 to 12 °C (Harkness and Dymond, 1961 as cited by Moos, 1978). White sturgeon cultured to spawn in water at 18 °C had

reduced oocyte development and ovulation and an increased incidence of follicular atresia (Webb et al., 1999).

Specific conductivity ranged from 271 to 964 microSiemens per centimeter ( $\mu\text{S}/\text{cm}$ ) and was greatest below the confluence of Salt Creek. Specific conductivity at the NAWQA monitoring stations averaged  $563 \pm 73 \mu\text{S}/\text{cm}$  and ranged from 408 to 786  $\mu\text{S}/\text{cm}$  ( $n = 6$ ). The high conductivity below Salt Creek is most likely a result of natural saline seeps within the watershed (Farrar and Gersib, 1991). However, municipal wastewater also is high in phosphorus, nitrogen, and other mineral nutrients that increase water conductivity and wastewater treatment plants contribute approximately 32 percent Salt Creek's flow at its confluence with the Platte River (Verstraeten, 1997).

Nebraska does not have a conductivity-based water quality standard for aquatic life. However, studies of inland fresh waters indicate that streams supporting healthy mixed fisheries have a conductivity range between 150 and 500  $\mu\text{S}/\text{cm}$ , whereas a conductivity outside the range of 50 to 1500  $\mu\text{S}/\text{cm}$  may not be suitable for some fish or invertebrates. Important habitat factors for spawning gulf sturgeon include calcium ion concentrations of 6 to 18 mg/L corresponding to a conductivity range of 40 to 110  $\mu\text{S}/\text{cm}$  (Sulak and Clugston, 1997). Calcium ions were measured in five samples from the lower Platte River during this research and averaged  $72 \pm 12 \text{ mg}/\text{L}$ .

Dissolved oxygen concentrations ranged from 6.78 to 15.7 mg/L ( $n = 86$ ) and from 5.5 to 11.2 mg/L ( $n = 6$ ) as measured by researchers from UNL and NAWQA, respectively. These concentrations did not represent hypoxic conditions and low DO during the spawning period for shovelnose sturgeon in the lower Platte River does not appear to be a concern. However, low flow velocity and high water temperatures in the summer can depress DO to harmful levels and have resulted in past sturgeon die-offs in the Platte River near Columbus, NE (NGPC, 2003). Adverse effects to sturgeon exposed to hypoxic conditions (i.e., DO concentrations less than 3 mg/L) can include decreased survival and growth (Secor and Gunderson, 1998).



## Histopathology

Fish health assessments that include histopathology are increasingly being used to evaluate environmental stress as tissue lesions can provide a definitive biological endpoint of historical exposure (Stentiford et al., 2003). The incidence and severity of histopathological lesions in kidney, liver, spleen, and gonadal tissues of shovelnose sturgeon from the lower Platte River (Appendix Table A.4) are summarized below. These results are a reiteration of results initially provided in a report by Papoulias et al. (2004) which includes additional information and digital images.

Livers from all shovelnose sturgeon appeared normal upon gross examination and were similar in appearance. The incidence of liver inflammation as indicated by leukocytes also was low in all shovelnose sturgeon sampled. In addition, most shovelnose sturgeon (42/53) had only minimal to mild reduction in liver fat/glycogen indicating that metabolism and nutrition was normal. Eight of the nine fish that had a moderate reduction in liver fat/glycogen and one fish (SP-29) that had a moderately-severe reduction in reserves were all in the late stage of spawning and reduction in fat reserves would be expected. However, one fish (SP-09, a stage II female) had moderate reduction of fat/glycogen.

No abnormalities were observed in shovelnose sturgeon head kidneys. Most fish (37/49) showed only minimal to mild debris in kidney tubules or Bowman's space. A few fish (9/49) had slightly swollen or shrunken glomeruli, the significance of which is unknown (Figure 3). Fat deposits were observed in the kidney of one fish (SP-08) and may reflect metabolic disturbance. Similar kidney fat deposits were reported in Japanese medaka (*Oryzias latipes*) after long-term exposure to  $\beta$ -hexachlorocyclohexane (Wester and Canton, 1986). Fourteen males that were in the late reproductive stage V were observed to have sperm in their kidney tubules and Bowman's space; however, this may not be unusual because in sturgeon some of the renal tubules drain into the vas deferens (Hoar 1969).

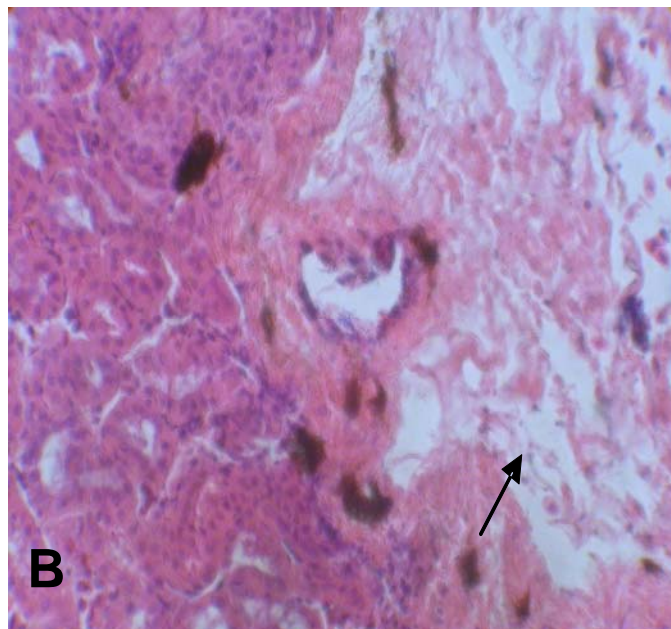
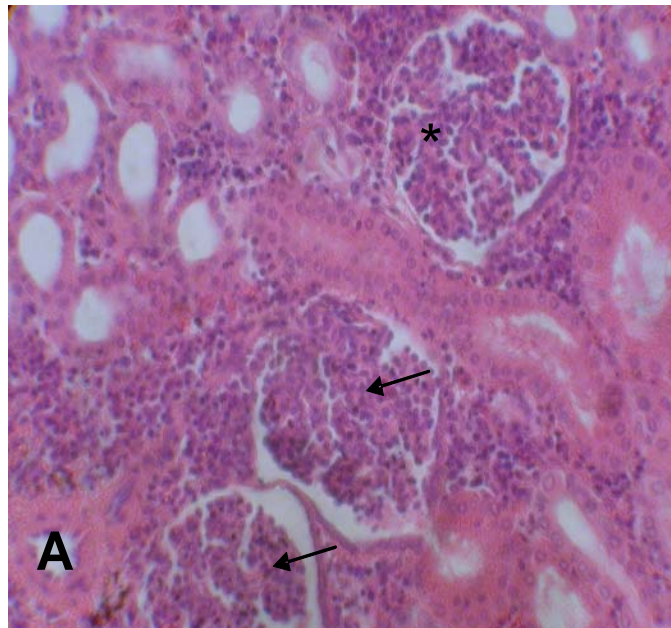


Figure 3. Kidney abnormalities in *Scaphirhynchus platyrhynchus* (SP) from the lower Platte River, Nebraska, 2002.

A = kidney from SP-05 at 400x magnification: \* indicates normal glomeruli, arrows indicate hypotrophic glomeruli.

B = kidney from SP-08 at 100x magnification: arrow indicates fat.

Ninety-one percent of shovelnose sturgeon (48/53) had minimal to mild reductions of red blood cells (RBCs) in the spleen, four individuals had moderate reductions, and one individual (SP-13) had a moderately-severe reduction. When a fish is stressed, RBCs are released from the spleen into the blood (Takashima and Hibiya 1995). There may have been reductions in spleen RBCs as a result of stress from handling. However, *Scaphirhynchus* sp. have been reported to exhibit a low physiological response to acute handling and severe confinement (Barton et al., 2000). Inflammation in the spleen was low in all but three fish, but five individuals showed either large fat deposition (Figure 4), minimal to mild necrosis, or edema.

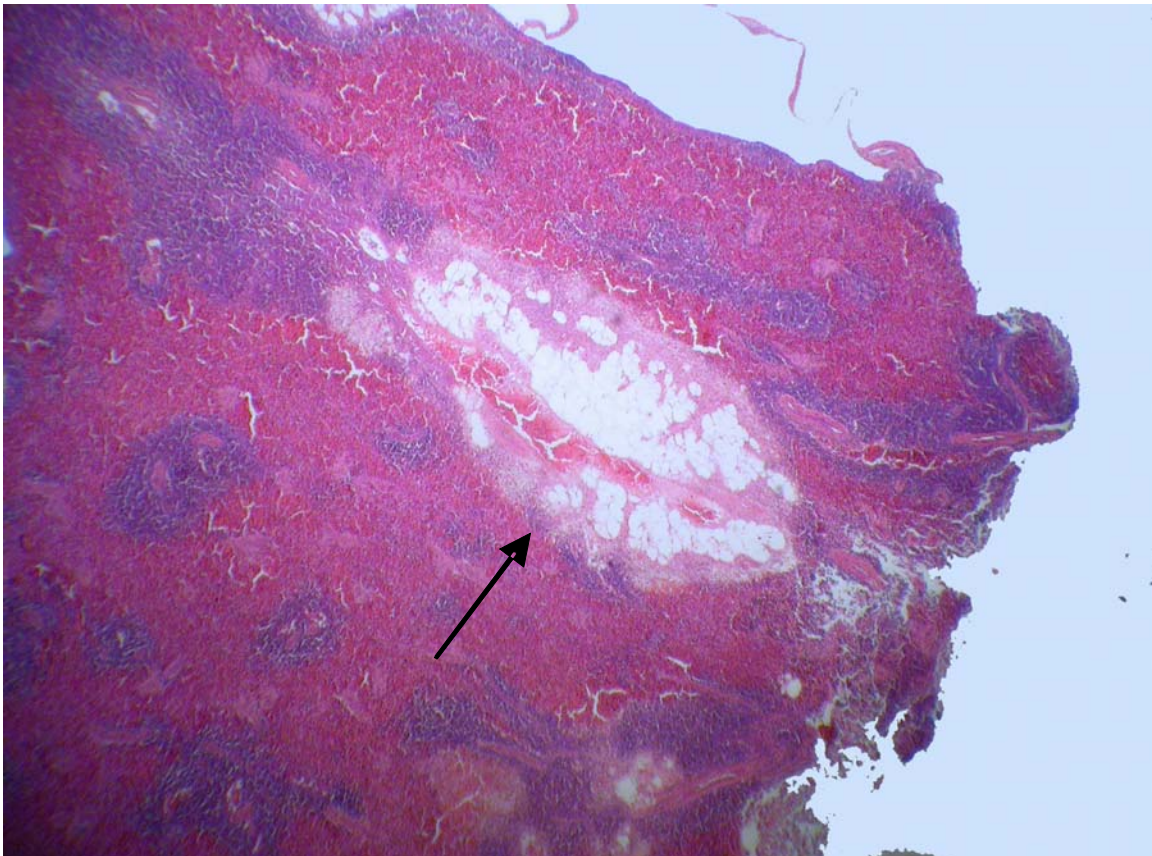


Figure 4. Fat deposition (indicated by arrow) in the spleen of a *Scaphirhynchus platyrhynchus* (SP-07) collected from the lower Platte River, Nebraska, 2002. The image is at 400x magnification.

A variety of reproductive stages were found in shovelnose sturgeon from the lower Platte River (Appendix Table A.2). Most females (11/ 20) were stage II and most males (21/32) were stage V. Only six of 20 adult female shovelnose sturgeon were in spawning condition (stage V). No sturgeon were found in stages I, IV or VI. Three of four stage V females that had empty follicles were collected above the confluence of the Elkhorn River, perhaps indicating that this provides spawning habitat for shovelnose sturgeon.

A lack of stage I and stage IV females during the spawn was previously reported for shovelnose sturgeon from the Missouri River (Moos, 1978). However, a large percentage of stage-II females, as found in this study, was not reported by Moos (1978). Out of 56 females staged in May and June of 1968 and 1969, 20 were stage V, 20 were stage II, and 16 were stage III (Moos, 1978). In this study, there were nearly twice as many stage-II females compared to stage V females (11 and 6, respectively) and only 3 of 20 females were stage III.

Ovaries of several females at stages II, III, and V showed varying degrees of atresia (Appendix Table A.4) (Figure 5). Atresia is a degenerative process commonly observed in post-spawning fish when vitellogenic eggs are not released and are resorbed by the fish's body. A high percentage of vitellogenic eggs with atresia before spawning, as well as atresia of pre-vitellogenic eggs, can indicate a pathological condition. In this study, no pre-vitellogenic eggs with atresia were observed. However, a high percentage (i.e., greater than 25 percent) of atretic bodies in two early-stage females may indicate that these individuals did not spawn completely the previous year and resorbed many of their eggs. Of the six spawning (stage V) females collected, one (SP-49) had a high incidence of atresia and four had slight atresia that may have been at an earlier progressive stage. The presence of eggs undergoing atresia in spawning stage females suggests spawning may not be successful. A variety of stressors can induce atresia including age, changes in hormone concentrations, light, warm water temperature, and nutrition (Guraya, 1986; Webb et al., 1999). Environmental contaminants such as atrazine (Spanó et al., 2004), PCBs (Collier et al., 1992), and mercury (Kirubakaran and Joy, 1988) also have been linked to follicular atresia in fish.

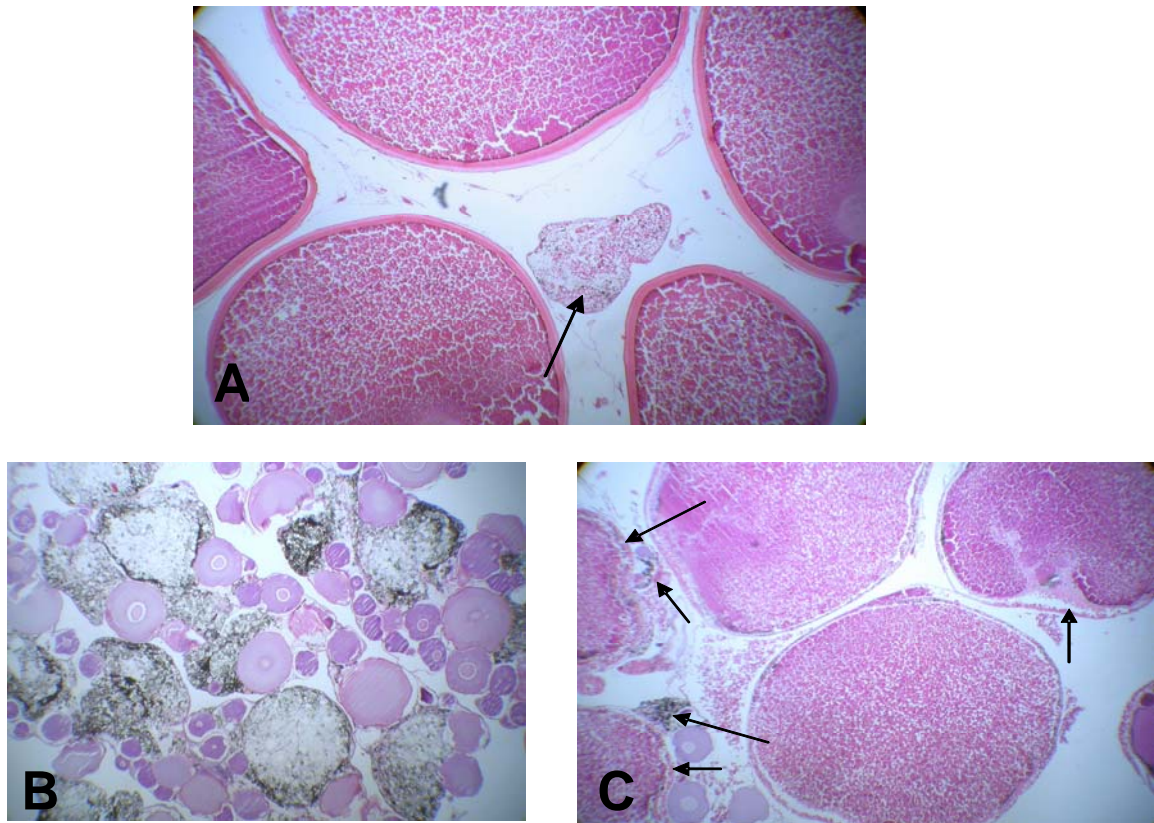


Figure 5. Ovaries with atresia in *Scaphirhynchus platyrhynchus* (SP) from the lower Platte River, Nebraska, 2002. All images are at 40x magnification.

- A. SP-33, stage V ovary with light atresia as indicated by the arrow.
- B. SP-16, stage III ovary with heavy atresia as indicated by black pigment.
- C. SP-49, stage V ovary with heavy atresia as indicated by arrows.

One late stage-III male shovelnose sturgeon (SP-08) was intersex with a testis-ova condition. Grossly, the gonad appeared to be a testis and a few pre-vitellogenic oocytes were observed histologically scattered along the distal margins of the organ (Figure 6). Sturgeon are gonochoristic and hermaphroditism is abnormal in this group of fishes (Van Eenennaam and Doroshov, 1998; Harshbarger et al., 2000).

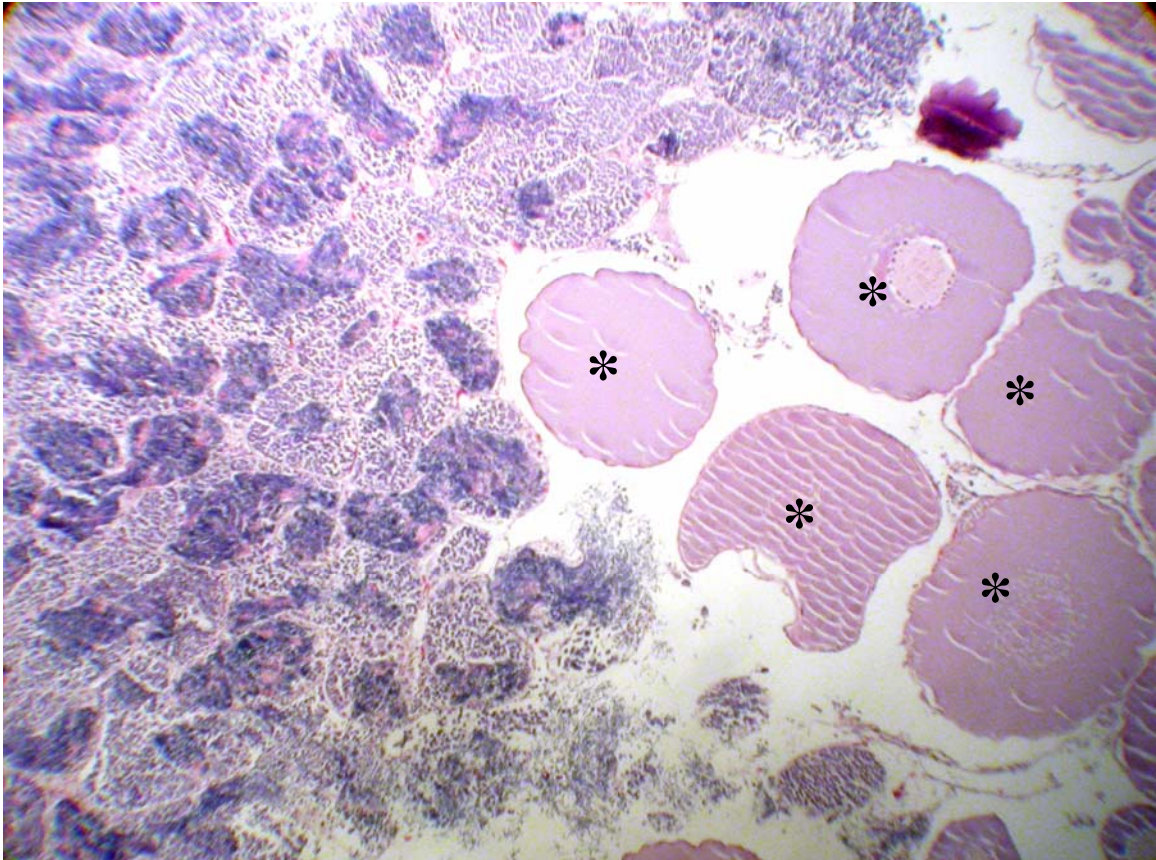


Figure 6. Stage III testis with stage III oocytes in a *Scaphirhynchus platyrhynchus* (Fish ID = SP-08) from the lower Platte River, Nebraska, 2002. The asterisks indicate developing eggs embedded in testis. The image is at 100x magnification.

Hermaphroditism in shovelnose sturgeon from the Missouri River System has been reported previously; however, its incidence and severity appears to be increasing. June (1977) reported finding a 2 percent incidence of testis-ova in shovelnose sturgeon from 1965 to 1971 in Lake Oahe. Moos (1978) reported that 1.6 percent of the fish caught on the Missouri River below Gavins Point Dam from 1968-69 had testis-ova. Twenty-five years later in the same stretch of river, the USGS cooperative unit in South Dakota collected 54 fish, of which 19 percent had testis-ova (Papoulias, unpublished data). Papoulias et al. (2002) reported that 12 percent of male shovelnose sturgeon caught from the lower Missouri River in 2001 had testis-ova. In most fish, the condition was extreme in that both male and female gametes were mature and the intersex condition was macroscopically observable (Papoulias et al., 2002).

Hermaphroditic shovelnose sturgeon from the Mississippi River have also been reported. Harshbarger et al. (2000) found two shovelnose sturgeon with testis-ova out of 17 fish examined (two of seven males) just below the confluence with the Missouri River near St. Louis. Research by Southern Illinois University found five intersex fish out of 59 males collected in 2003 between Alton, IL and Cape Girardeau, MO (Brian Koch, personal communication, 2003).

A high incidence of male feminization has been reported in wild populations of other fish species (De Metrio et al., 2003; Jobling et al., 1998). Out of 162 male swordfish (*Xiphias gladius* L.) collected from the Mediterranean, 25 percent (n = 40) exhibited female pre-vitellogenic oocytes within the testes (De Metrio et al., 2003). The authors concluded that the high rate of intersex males could be a result of exposure to estrogen mimicking substances (De Metrio et al., 2003). A 100 percent incidence of intersex was reported for wild roach (*Rutilus rutilus*) at two sites downstream of sewage treatment works (Jobling et al., 1998). A follow-up study reported that these wild intersex roach had reduced gamete production and sperm motility, which resulted in decreased egg fertilization (Jobling et al., 2002).

Testis-ova have been observed in fish after laboratory exposure to estrogen and estrogenic chemicals (Tabata et al. 2001, Kang et al. 2002). However, in addition to estrogens there are a number of other documented inducers of testis-ova including

senescence, genetic abnormalities, radiation, diet, temperature changes, and hybridization (Atz, 1964; Lam 1983). Shovelnose sturgeon and pallid sturgeon hybridize in the wild and it is believed that their hybridization is a result of anthropogenic changes to the environment that have caused spawning areas and times to overlap (Carlson et al., 1985; Ruelle and Keenlyne, 1994).

#### Liver Macrophage Aggregates (MAs)

Shovelnose sturgeon from the lower Platte River (n = 53) had small but moderate numbers of liver MAs (Appendix Table A.5). Liver MA density averaged  $27 \pm 2$  per  $\text{mm}^2$  and the mean size (area) of MAs averaged  $0.0022 \pm 0.0002 \text{ mm}^2$ . The percent area of hepatic tissue occupied by MAs averaged  $6 \pm 1$  percent and ranged from 0.1 to 28.1 percent.

Macrophage aggregates protect fish by storing, destroying, or detoxifying particulate matter (such as carbon particles or bacteria) and are an important part of immune response in fish (Schmitt and Dethloff, 2000). Macrophage aggregates have been suggested as possible biomarkers of exposure to environmental contaminants, including atrazine (Biagianti-Risbourg and Bastide, 1995). Most studies have noted increases in macrophage aggregates in fish from polluted waters (reviewed in Schmitt and Dethloff, 2000); however, a decreased abundance of liver MAs have also been reported in flounder exposed to hydrocarbon-contaminated sediments (Payne and Fancey, 1998). The formation of MAs may also be influenced by age, diet, and temperature, thereby making cause and effect relationships from environmental contaminants difficult to establish (Blazer et al., 1987). Generally, MAs occur most often in the spleen of teleosts; however, MAs were found only in the livers, not the spleens, of shovelnose sturgeon, which is normal for more primitive fishes (Wolke 1992).

To our knowledge, there are no published data on liver MAs in sturgeon. However, liver MAs have been reported for other fish species including largemouth bass (*Micropterus salmoides*), winter flounder (*Pseudopleuronectes americanus*), white sucker (*Catostomus commersoni*), striped mullet (*Mugil cephalus*), and smallmouth bass (*Micropterus dolomieu*) (Blazer et al., 1987; Murchelano and Wolke, 1991; Couillard and



Hodson, 1996; Frodello et al., 2001; Anderson et al., 2003). Largemouth bass that were stressed had liver MA densities of 9 per mm<sup>2</sup> whereas healthy fish had liver MA densities of 4 MAs per mm<sup>2</sup>; however, average size of MAs were not different at 0.0038 and 0.0058 mm<sup>2</sup> for healthy and stressed fish, respectively (Blazer et al., 1987).

Macrophage aggregates in spleen tissue of non sturgeon species are more responsive to contaminant exposure than liver MAs (Couillard et al., 1999). Therefore, spleen MAs are measured by the BEST program, instead of liver MAs. Monitoring by the BEST program in 1995 found that carp from lower Platte River (n = 9) had a relatively low mean spleen MA density of 9 per mm<sup>2</sup> (Schmitt, 2002). In comparison, carp from 29 of the other 37 stations sampled by BEST had MA densities greater than 10 per mm<sup>2</sup> (Schmitt, 2002). Spleen MA densities for carp and largemouth bass collected by the BEST program at all sites were much lower than liver MA densities in shovelnose sturgeon from this study and did not exceed 40 MA/mm<sup>2</sup>, a reference number reportedly correlated with hypoxic stress, contaminated sediments, or both (Fournie et al., 2001). No such reference value is available for liver MA densities; however, it may be noteworthy that 6 of 53 shovelnose sturgeon from the Platte River had a liver MA density greater than 40.

The percent of tissue occupied by MAs has also been used as an indicator of fish health. Macrophage aggregates in spleens of largemouth bass from a PCB contaminated lake comprised 2.0 percent of the area (Papoulias and Tillitt, 2003), whereas MAs in liver tissue from largemouth bass occupied 2.6 and 1.6 percent of the total area examined in stressed and healthy fish, respectively (Blazer et al., 1987). Percent coverage by MAs in liver of striped mullet from a contaminated costal lagoon in France ranged from 1.5 to 21.5 percent (Frodello et al., 2001).

In this study, there was a statistically significant ( $p < 0.05$ ) but weak ( $r = 0.25$ ) relationship between Platte River shovelnose sturgeon age and the number of MAs in liver (n = 44); however, there were no significant relationships between age and MA size or percent area covered by MAs. Many researches have found significant ( $p < 0.05$ ) positive relationships between MA parameters and fish age (Couillard and Hodson, 1996; Mikaelian et al., 1998; Schmitt, 2002).

Our results indicate that there is a weak but significant ( $p < 0.005$ ) correlation between liver MA density and concentrations of Se in shovelnose sturgeon carcasses (Figure 7). Ongoing research at the University of California-Davis is studying the relationship between liver MAs in white sturgeon exposed to Se. Thus far, qualitative observations indicate that juvenile white sturgeon exposed to 10 to 40  $\mu\text{g/g}$  Se in their diet for six months have greater liver MA densities than controls (Regina Linville, Ph.D. candidate, University of California-Davis, pers. comm., 2004).

Macrophage aggregates can also develop in association with ovarian atresia (Agius and Roberts, 2003). In our study, the four female shovelnose sturgeon with a high to moderate incidence of ovarian atresia also had either a higher percentage of liver comprised of MAs and/or a higher MA density than females with slight to no ovarian atresia indicating that debris from phagocytized ova are perhaps being deposited in MAs.

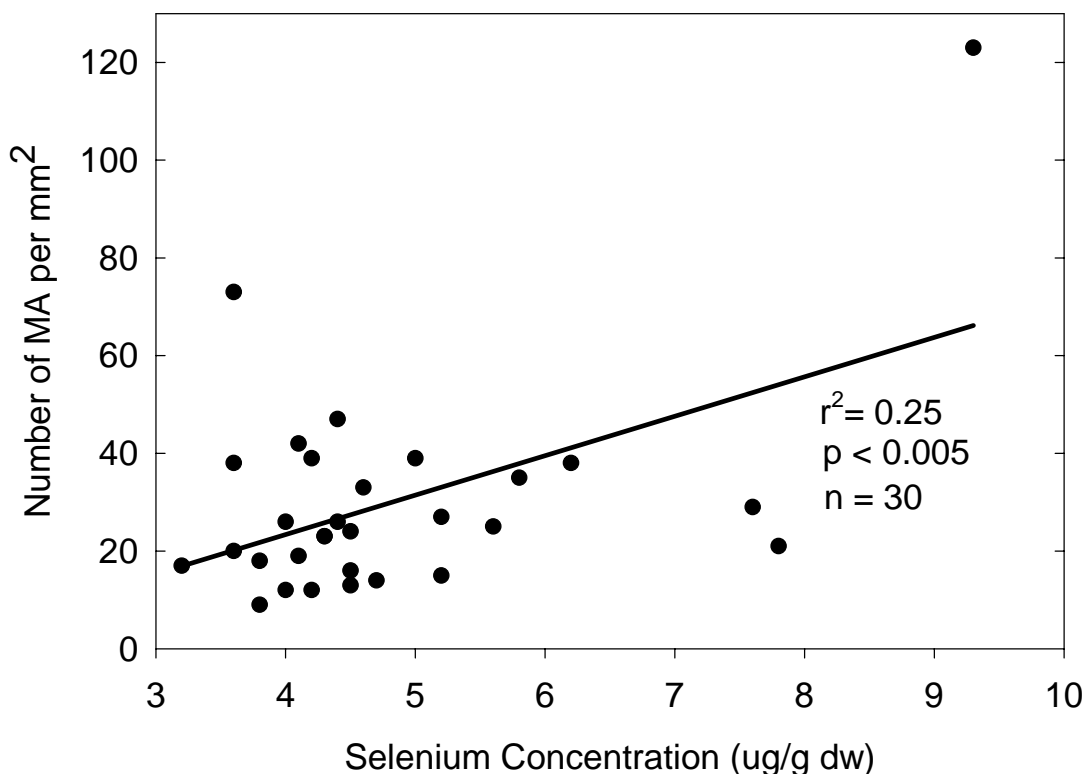


Figure 7. Linear regression between the density of macrophage aggregates (MAs) in spleen and selenium concentrations in carcasses of shovelnose sturgeon from the lower Platte River, Nebraska, 2002.

The presence of hemosiderin, lipofuscin/ceroid, and melanin pigments in MAs may indicate the health of an organism or its exposure to toxic compounds. No melanin stains were observed in shovelnose sturgeon from the Platte River. Hemosiderin and lipofuscin/ceroid pigments were observed in 22 of 53 liver samples analyzed, with hemosiderin being most predominant (Papoulias et al., 2004). Hemosiderin is a by-product of hemoglobin breakdown. Lipofuscin and ceroid are considered to be lipid pigments produced by the peroxidation of unsaturated fatty acids.

#### Reproductive Biomarkers in Blood Plasma

Concentrations of 11-ketotestosterone (11KT), 17 beta-estradiol ( $E_2$ ), and vitellogenin (VTG) in blood plasma were measured for 52 shovelnose sturgeon (Appendix Table A.6). There were no significant differences among collection sites for concentrations of 11KT,  $E_2$ , VTG, or the  $E_2$  to 11KT ratio (E/T) (data not shown). Mean concentrations of hormones in plasma were generally similar to those reported for shovelnose sturgeon from the Mississippi River (Coffey et al., 2003) except for a greater mean concentration of 11KT in female sturgeon from the lower Platte River (Table 1). On average, male shovelnose sturgeon from the lower Platte River also had higher  $E_2$  concentrations and lower KT concentrations than those collected from the Missouri River in 2001 (Papoulias, unpublished data).

Mean concentrations of 11KT and  $E_2$  in blood plasma of shovelnose sturgeon were generally similar between genders and among stages, with the exception of 11KT in stage III fish (Figure 8). Normally, there are clear differences in plasma hormone concentrations between genders as well as differences among stages. For carp collected in the fall, Goodbred et al. (1997) defined abnormal E/T ratios as greater than one for males and less than one for females (Goodbred et al., 1997). By this standard, abnormal E/T ratios for shovelnose sturgeon in our study were observed in 47 percent (9/19) of females and 49 percent (16/33) of males. In comparison, Coffey et al. (2003) reported abnormal E/T ratios in 42 percent of males (8/19) and 5 percent in females (1/22) from two sites on the Mississippi River that are believed to be contaminated by endocrine modulating substances.

Table 1. Mean concentrations of 17-beta estradiol (E<sub>2</sub>), 11-ketotestosterone (11KT), and vitellogenin (VTG) in blood plasma from shovelnose sturgeon collected in the Platte River, Nebraska, 2002, and at two sites on the Mississippi River, 1997.

		Site, Sample Size (N), and Mean Concentration <sup>1</sup> ± Standard Error (SE)					
		<sup>2</sup> Platte River		<sup>3</sup> Mississippi River A		<sup>3</sup> Mississippi River B	
Biomarker	Gender	N	(Mean ± SE)	N	(Mean ± SE)	N	(Mean ± SE)
E <sub>2</sub>	Females	19	698 ± 69	12	726 ± 139	10	526 ± 60
	Males	32	618 ± 25	12	599 ± 77	7	594 ± 119
11-KT	Females	19	538 ± 86	12	353 ± 114	10	292 ± 116
	Males	32	747 ± 84	12	721 ± 113	7	787 ± 206
VTG	Females	19	0.539 ± 0.154	12	4.013 ± 0.969	10	1.635 ± 0.623
	Males	32	0.425 ± 0.095	12	0.005 ± 0.003	7	0.011 ± 0.008

<sup>1</sup>Concentrations are in pg/ml for E<sub>2</sub> and 11-KT and in mg/ml for VTG. <sup>2</sup> Results are from this study, <sup>3</sup> Results are from Coffey et al. (2003) at a reference site (A) and a site contaminated with organochlorines (B).

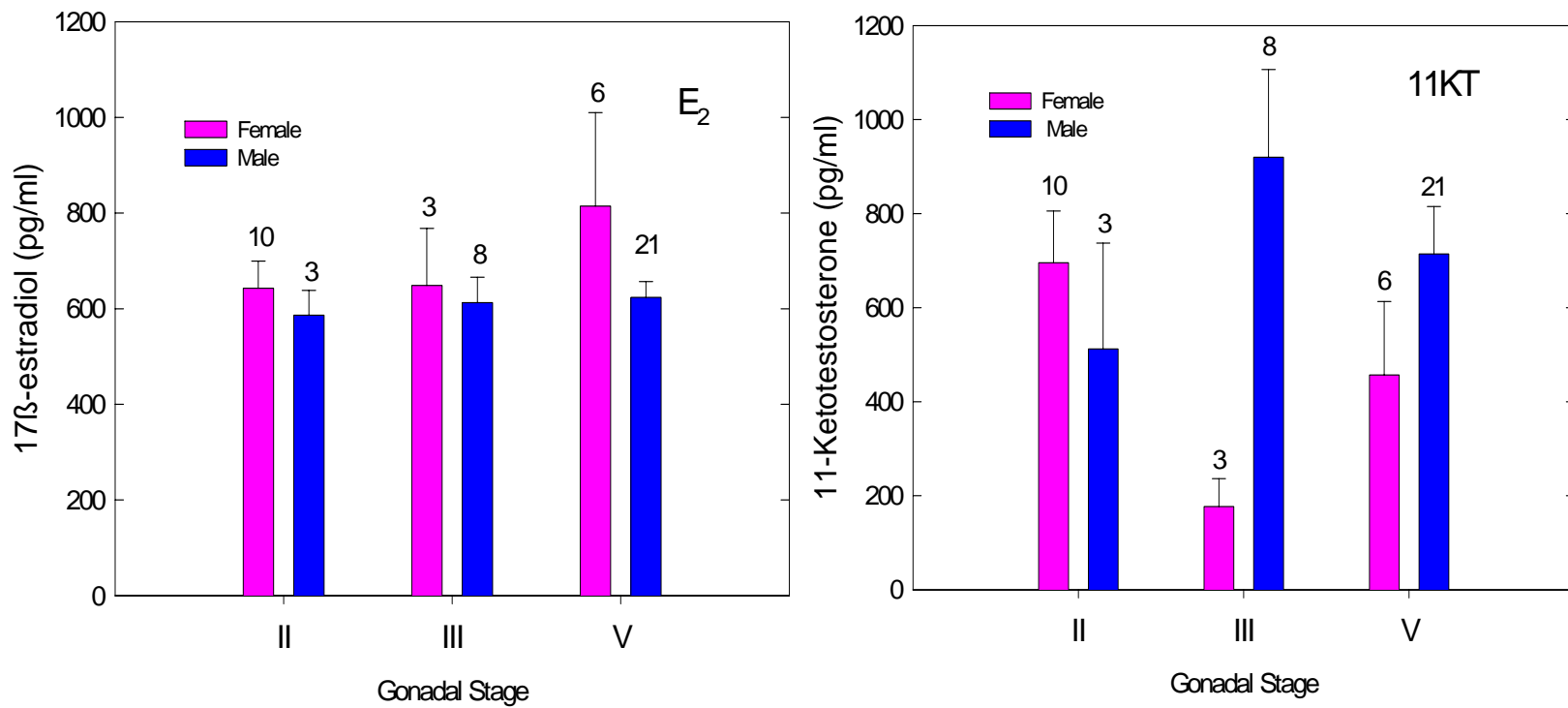


Figure 8. Mean  $\pm$  SE concentration of 17- $\beta$  estradiol and 11-ketotestosterone in shovelnose sturgeon plasma for 3 gonadal stages. Gonadal stages are described by Moos (1978). Sample size is above each standard error bar.

Mean concentrations of VTG in plasma samples were similar between shovelnose sturgeon genders, but were higher in males and lower in females when compared to those collected by Coffey et al. (2003) from the Mississippi River (Table 1). Concentrations of VTG in four males from the lower Platte River exceeded 1 mg/ml, and those with a VTG concentration greater than 0.8 mg/ml also had an abnormal E/T ratio (Figure 9). In comparison, VTG in male shovelnose sturgeon collected by Coffey et al. (2003) never exceeded 0.061 mg/ml. Blood plasma concentrations of VTG in 277 male carp collected nationwide (Goodbred et al., 1997) also never exceeded 1 mg/ml, although differences in VTG expression between shovelnose sturgeon and carp species are unknown.

Vitellogenin production is normally stimulated by circulating estrogens in oviparous fish (Folmar et al., 1996; Schmitt and Dethloff, 2000). However, male fish exposed to hormonally active compounds (HACs) in field or laboratory assessments typically have several fold higher concentrations of VTG in blood plasma than fish from control groups or reference populations (Purdom et al., 1994; Folmar et al., 1996; Jobling et al., 1998; Rodgers-Gray et al., 2000; Ankley et al., 2003; Kirby et al., 2004). Conversely, decreased VTG in females can indicate exposure to anti-estrogenic compounds including synthetic testosterone (e.g., trenbolone, zearalanol, and melengestrol) that are used in the cattle industry (Kime, 1999; Lange et al., 2002; Ankley, 2003). Male fish are genetically capable of producing VTG and it is not unusual for males of some fish species to produce low concentrations without any known exposure to environmental contaminants (Goodwin et al., 1992). However, national monitoring studies have shown that concentrations of VTG in blood plasma of male carp are generally well below 0.1 mg/ml whereas females tend to have concentrations greater than 1.0 mg/ml (Goodbred et al., 1997; Schmitt, 2002). In comparison, concentrations of VTG in male carp (n = 10) collected from a sewage effluent canal had blood serum concentrations greater than 0.1 mg/ml 50 percent of the time and ranged from below detection to 10 mg/ml (Folmar et al., 1996). In our study, 23 of 33 male shovelnose sturgeon had concentrations of VTG greater than 0.1 mg/ml and concentrations ranged from 0.007 - 2.152 mg/ml.

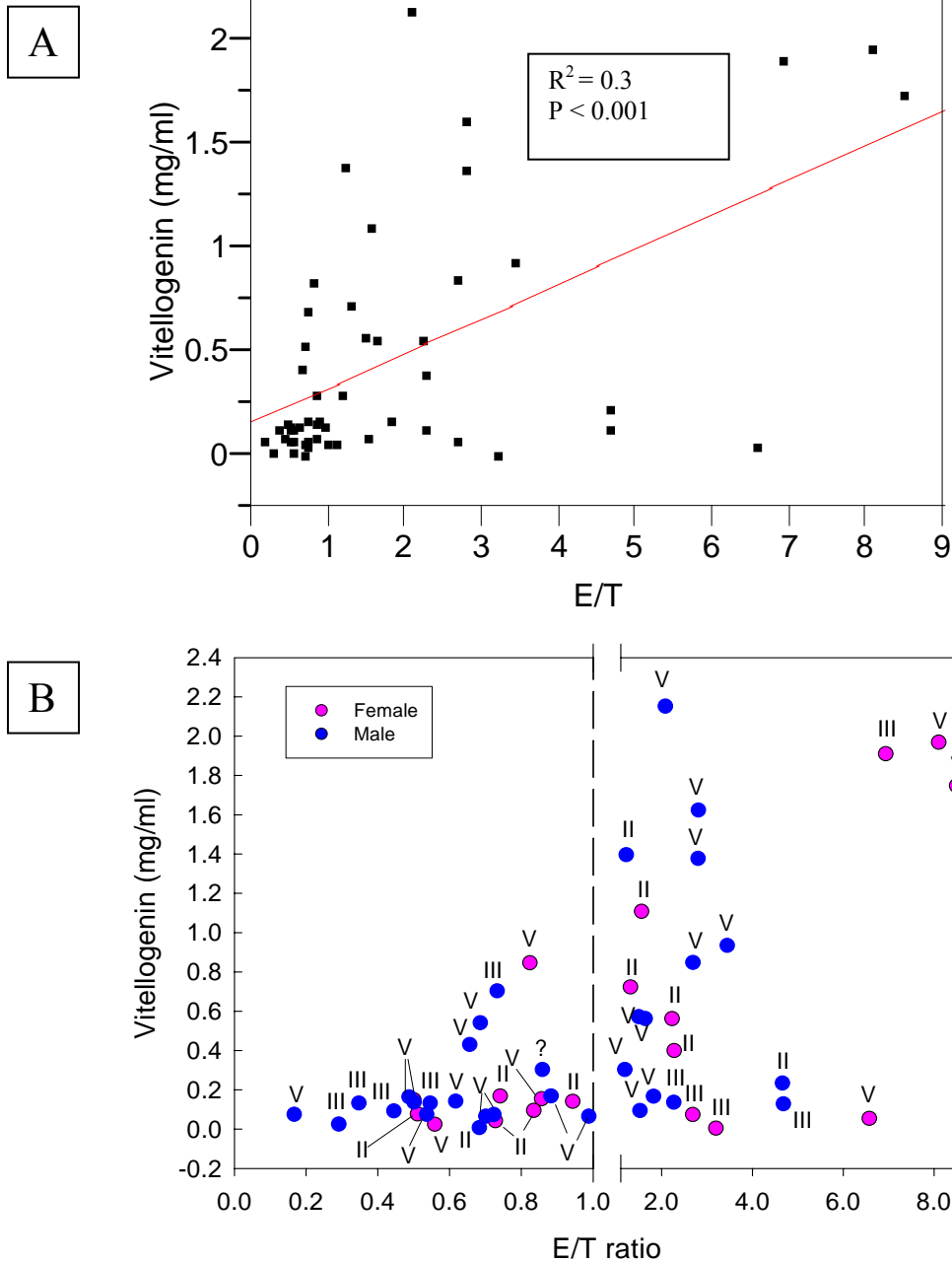


Figure 9. Bivariate plots of 17-β estradiol to 11 ketotestosterone ratio (E/T) versus vitellogenin concentration in shovelnose sturgeon males and females from the lower Platte River, NE. A) Linear regression plot showing a weak but significant relationship between E/T ratio and VTG. B) Roman numerals indicate gonadal stage according to Moos (1978). Note scale break on x-axis at 1.0. Males with an E/T less than one and females with an E/T greater than one are considered normal. ? = the gonadal stage was not determined.

The abnormal E/T ratios and concentrations of VTG in shovelnose sturgeon from this study may reflect natural variation during the breeding season. Plasma samples in this study were collected at a time of peaked spawning activity. Hormone concentrations in shovelnose sturgeon during the spring have not been previously reported; therefore, the data from this study were compared to data from studies that collected carp and shovelnose sturgeon in the fall (Goodbred et al., 1997; Coffey et al., 2003). Hormone concentrations in fish tend to have the greatest variance during spring spawning periods, but also can vary 30-fold in the fall during gonadal recrudescence (Schmitt and Dethloff, 2000).

There also is a concern that the high readings of vitellogenin in male shovelnose sturgeon from this study may be due to the use of a non species-specific vitellogenin antibody. Vitellogenin was measured in this study using an antibody for carp (Goodbred et al., 1997). Although parts of the vitellogenin molecule is thought to be well conserved across species (Heppell et al., 1995), the use of a species-specific VTG antibody is preferable to control for any differences in blood chemistry. The cross reactivity of the carp vitellogenin antibody with non-vitellogenin proteins in shovelnose sturgeon blood is unknown; however, it is notable that Coffey et al. (2003) used the same lab and carp antibody in his study and did not report high VTG concentrations in males.

The combination of elevated blood plasma vitellogenin concentrations and altered E/T ratios in male shovelnose sturgeon from the lower Platte River indicate a likelihood of endocrine disruption and may be a result of exposure to HACs such as atrazine. Adult male goldfish exposed to atrazine had similar effects to those found in male shovelnose sturgeon from this study including increased concentrations of E<sub>2</sub> in blood plasma, decreased concentrations of 11KT in blood plasma, and altered E/T ratios (Spanó et al., 2004). Spanó et al. (2004) concluded that the observed effects were likely due to aromatase induction by atrazine. Aromatase converts testosterone to E<sub>2</sub> in vertebrate species. Atrazine induces aromatase in humans (Sanderson et al., 2001), reptiles (Crain et al., 1997; Keller and McClellan-Green, 2004), and frogs (Hayes, 2004; Miyahara et al., 2003). Aromatase induction by atrazine may result in a parabolic dose response (Hayes



et al., 2003) in which lower concentrations of atrazine can have a greater effect than higher concentrations. This is because peaks of high estrogen concentrations within an organism are countered by negative feedback pathways within the endocrine system which inhibit estrogen production. Leopard frogs (*Rana pipiens*) exposed to 0.1 µg/L atrazine had a higher incidence (36 percent) of gonadal dysgenesis compared to those exposed to 25 µg/L atrazine (12 percent; Hayes et al., 2003).

#### Hepatic EROD Activity

The EROD activities measured in 53 shovelnose sturgeon liver samples were generally low (Appendix Table A.7). The average EROD activity for the 19 samples that were above the level of quantification was  $3 \pm 0.4$  pmol resorufin/min/mg protein and ranged from 1.6 to 9.1 pmol/min/mg. Differences between genders and sites were not analyzed statistically due to the large number of non-detects. However, EROD activity appeared to be lower in females compared to males and was above detection limits more frequently at some sites (e.g., 15LR and 7550) compared to others (Table 2). There were no significant differences between EROD induction and carcass concentrations of Se or PCBs; however, sample size for the PCB analysis was small ( $n = 8$ ) and the comparison with Se was limited by overall low EROD activity.

Carp collected in 1995 from the lower Platte River at Louisville as part of the BEST program (Schmitt, 2002) also had a depressed mean EROD activity ( $1.6 \pm 0.48$  pmol/min/mg,  $n = 9$ ). Female carp and largemouth bass collected by the BEST program in 1995 generally tended to have lower EROD activity than males. EROD activity in carp greater than 6 and 4 pmol/min/mg for males and females, respectively, was considered induced (Schmitt, 2002).

In this study, shovelnose sturgeon from the Platter River had lower EROD activity than previously reported for sturgeon. EROD activity in shovelnose sturgeon ( $n = 57$ ) from the Missouri River were generally above the level of detection (LOD), and those above detection averaged  $4.01 \pm 0.23$  pmol/min/mg (D. Papoulias, unpublished data). Lake sturgeon from the Ottawa River in Canada had a mean EROD activity of  $3.39 \pm 0.57$  pmol/min/mg (standard deviation) (Rousseaux et al., 1995). Mean hepatic

EROD activity in white sturgeon from the Columbia River was as high as  $138 \pm 23$  pmol/min/mg ( $n = 15$ ), whereas white sturgeon ( $n = 10$ ) from a commercial fish hatchery at University of California Davis had a mean hepatic EROD activity of  $11 \pm 2$  pmol/min/mg (Foster et al., 2001). Exposure to chlorinated dioxin, chlorinated furans, and/or PAHs could be responsible for the elevated EROD activity in white sturgeon (Foster et al., 2001). Interspecies differences in basal EROD activity levels and hatchery fish food contamination with EROD inducing pollutants (Wilson et al., 2001) are potential reasons for the difference between EROD activity in hatchery-reared white sturgeon and in shovelnose sturgeon from the lower Platte River.

Table 2. Summary statistics for Ethoxyresorufin-*O*-deethylase (EROD) activity (pg/min/mg) in shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

Site(s)	Gender	$N_A$	$N_{BDL}$	$N_{BLQ}$	$N_Q$	Mean $\pm$ SE	Min	Max
All	F	20	10	7	3	NC	< 0.2	2.8
All	M	33	9	8	16	NC	< 0.2	9.1
6477	M	6	4	1	1	NC	< 0.2	4.5
6477	F	4	2	1	1	NC	< 0.2	1.9
7550	M	7	0	1	6	$2.21 \pm 0.38$	< 0.7	3.2
7550	F	4	1	2	0	NC	< 0.35	1
15LR	M	10	0	3	7	$2.90 \pm 0.78$	< 0.45	9.1
15LR	F	2	2	0	0	NC	< 0.2	0.3
50SC	F	6	1	3	2	NC	< 0.35	2.8
50SC	M	4	0	3	1	NC	< 0.7	2.4
SCER	M	6	5	0	1	NC	< 0.2	2.3
SCER	F	4	4	0	0	NC	< 0.3	0.4

Note: M = male, F = female,  $N_A$  = number of samples analyzed,  $N_{BDL}$  = number below detection limit,  $N_{BLQ}$  = number below level of quantification,  $N_Q$  = the number of samples quantified, SE = standard error, Max = maximum. NC = mean not calculated because most samples were below the level of quantification.

#### H4IIE Bioassay

Dioxin equivalents (TCDD-EQs) were above the level of quantification in four of eight shovelnose sturgeon carcass extract samples (Table 3). The amount of dioxin-like potency in the 4 samples above the LOQ ranged from 2.2 to 3.9 pg/g and indicate potential toxicity (Nicks et al., 2005). The shovelnose sturgeon with the highest concentration of total PCBs (SP-14) also had the highest concentration of TCDD-EQs; however, small sample size and the number of samples with TCDD-EQs below quantification did not allow for statistical comparisons between PCBs and dioxin equivalents. There also was no correlation between EROD activity and TCDD-EQs.

Table 3. H4IIE results as TCDD equivalents (pg/g wet weight) for shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

Fish ID	Site	Gender	Age	TCDD-EQ	SD
SP14	7550	Male	13	3.9	0.8
SP25	SCER	Female	NA	2.7	0.6
SP53	15LR	Male	14	2.2	0.5
SP10	7550	Male	8	2.2	0.5
SP40	6477	Male	8	<LOQ	NA
SP11	7550	Male	9	<LOQ	NA
SP47	15LR	Male	9	<LOD	NA
SP45	15LR	Male	9	<LOD	NA

Note: TCDD-EQ = dioxin equivalents, NA = not applicable, < = less than, SD = standard deviation. The level of detection and level of quantification for these samples were 0.7 pg/g and 1.8 pg/g, respectively.

## Elemental Contaminants

Concentrations of Se were measured in 22 shovelnose sturgeon carcasses, and eight carcasses were analyzed for a suite of elemental contaminants (Appendix Tables A.8 and A.9). Concentrations of elemental contaminants were also determined in 11 samples of shovelnose sturgeon digesta (seven from individual fish and four composite samples of 5 - 11 individuals) and 11 composite samples of cyprinid minnows (Appendix Tables A.10 and A.11).

Shovelnose Sturgeon Carcasses. Concentrations of elemental contaminants in shovelnose sturgeon carcasses from the lower Platte River were compared to those previously reported in shovelnose sturgeon from the Missouri River (Ruelle and Henry, 1994a) and Atchafalaya River (Conzelmann, 1997). Shovelnose sturgeon from the lower Platte River had significantly ( $p < 0.05$ ) greater concentrations of Ba, Se, Sr, and Zn than those from the Atchafalaya River, whereas concentrations of Al, Cr, Cu, Fe, Ni, and Pb were significantly lower (Table 4). Shovelnose sturgeon from the lower Platte River also had greater concentrations of Ba, Hg, Mg, and Se than those collected previously from the Missouri River (Ruelle and Henry, 1994a); however, the only significant difference was for Ba (Table 4). Mean wet weight concentrations of Se ( $1.25 \mu\text{g/g}$ ) and Ba ( $11.3 \mu\text{g/g}$ ) in shovelnose sturgeon from this study also were greater than those in carp (Se =  $0.79 \mu\text{g/g}$ ; Ba =  $3.1 \mu\text{g/g}$ ) collected from the lower Platter River at Louisville, Nebraska in 1995 (Schmitt, 2002; BEST, 2004).

Mean concentrations of Pb in shovelnose sturgeon from this study were lower than those reported for shovelnose sturgeon from the Missouri River and Atchafalaya River (Table 4). This may be a result of reduced environmental emissions of Pb from smelters (Schmitt et al., 2002) and automobile fuels since it was banned as an additive in 1990 (ATSDR, 1999). A similar reduction in Pb concentrations in whole-body carp was found by the BEST program in 1995 (Schmitt, 2002) when compared to samples collected in 1986 by the National Contaminant Biomonitoring Program. Concentrations of Pb in shovelnose sturgeon from this study averaged  $0.08 \pm 0.01 \mu\text{g/g}$  wet weight (ww)

and were similar to those measured in carp from the lower Platte River in 1995 ( $n = 9$ ; mean =  $0.07 \mu\text{g/g ww}$ ; Schmitt, 2002; BEST, 2004).

Possible explanations for differences in elemental contaminant uptake among shovelnose sturgeon from the Platte River, Missouri River, and Atchafalaya River include temporal variation, potential differences in dietary food items, water quality, local/regional sources of contamination, and sediment quality. In addition, carcass samples from the lower Platte River had several tissues removed for other analyses, unlike the whole-body samples analyzed by Conzelmann (1997) and Ruelle and Henry (1994a). Nevertheless, basic comparisons between these data sets may help identify local contaminant issues.

In this study, concentrations of Se in shovelnose sturgeon carcasses averaged  $4.8 \pm 0.25$  milligrams per kilogram (mg/kg) dw and ranged from 3.2 - 9.3 mg/kg. Compared to concentrations of elemental contaminants in shovelnose sturgeon from other areas and carp from the lower Platte River, shovelnose sturgeon in the lower Platte River have elevated concentrations of Se. Concentrations of Se in all shovelnose sturgeon from this study exceeded the normal background range of 1 to  $4 \mu\text{g/g dw}$  for whole body fish and 24 of 30 were within the 4 to  $6 \mu\text{g/g Se}$  range where reproductive impairment may begin to occur in sensitive species such as perch, bluegill, and salmon (USDOI, 1998).

Principal anthropogenic sources of Se to aquatic ecosystems include coal-fired power plants and irrigation return flows (Schmitt, 2002). There are naturally high Se concentrations in Upper Cretaceous marine and sedimentary bedrock which underlies the lower Platte River (USDOI, 1998) and watersheds that drain into the lower Platte River are dominated by irrigated land. However, a low evaporation index for central and eastern Nebraska indicates that a Se problem due to irrigated lands is unlikely (USDOI, 1997). Runoff from cattle feedlots also may contribute Se releases into the lower Platte River as Se is often used as a feed additive by large livestock operations (Sims, 1995) and feedlot runoff is known to enter the Elkhorn River.

Table 4. Differences in mean  $\pm$  standard error (SE) concentrations of elemental contaminants in shovelnose sturgeon carcasses collected from the lower Platte River, Missouri River, and Atchafalaya River.

Trace Element	Site	N	Mean $\pm$ SE	Site	N	Mean $\pm$ SE	Site	N	Mean $\pm$ SE
Al	Atchafalaya River <sup>A</sup>	10	234.10 $\pm$ 36.79	lower Platte River <sup>B</sup>	8	38.86 $\pm$ 7.88	Missouri River <sup>B</sup>	10	29.50 $\pm$ 8.75
Ba	lower Platte River <sup>A</sup>	8	45.95 $\pm$ 7.37	Atchafalaya River <sup>B</sup>	10	26.10 $\pm$ 3.65	Missouri River <sup>B</sup>	10	25.34 $\pm$ 3.71
Cd	Missouri River <sup>A</sup>	9	0.47 $\pm$ 0.13	lower Platte River <sup>A</sup>	8	0.37 $\pm$ 0.04	Atchafalaya River <sup>A</sup>	10	0.35 $\pm$ 0.08
Cr	Atchafalaya River <sup>A</sup>	10	5.78 $\pm$ 1.51	Missouri River <sup>AB</sup>	10	2.14 $\pm$ 0.17	lower Platte River <sup>B</sup>	8	1.64 $\pm$ 0.33
Cu	Atchafalaya River <sup>A</sup>	10	5.09 $\pm$ 0.72	Missouri River <sup>B</sup>	10	2.14 $\pm$ 0.17	lower Platte River <sup>B</sup>	8	1.75 $\pm$ 0.13
Pb	Missouri River	3	1.63 $\pm$ 0.05	Atchafalaya River <sup>A</sup>	8	1.07 $\pm$ 0.21	lower Platte River <sup>B</sup>	7	0.33 $\pm$ 0.04
Fe	Atchafalaya River <sup>A</sup>	10	573.50 $\pm$ 90.89	Missouri River <sup>B</sup>	10	229.97 $\pm$ 52.48	lower Platte River <sup>C</sup>	8	73.75 $\pm$ 6.42
Hg	lower Platte River <sup>A</sup>	8	0.34 $\pm$ 0.06	Atchafalaya River <sup>A</sup>	10	0.27 $\pm$ 0.09	Missouri River <sup>A</sup>	9	0.20 $\pm$ 0.06
Mg	lower Platte River <sup>A</sup>	8	1740.00 $\pm$ 69.80	Atchafalaya River <sup>A</sup>	10	1549.40 $\pm$ 132.02	Missouri River <sup>A</sup>	10	1333.40 $\pm$ 126.42
Ni	Atchafalaya River <sup>A</sup>	10	14.05 $\pm$ 2.72	Missouri River <sup>B</sup>	10	0.89 $\pm$ 0.23	lower Platte River <sup>B</sup>	8	0.68 $\pm$ 0.08
Se	lower Platte River <sup>A</sup>	8	4.80 $\pm$ 0.25	Missouri River <sup>A</sup>	10	4.72 $\pm$ 0.29	Atchafalaya River <sup>B</sup>	10	3.58 $\pm$ 0.57
Sr	Missouri River <sup>A</sup>	10	104.09 $\pm$ 21.86	lower Platte River <sup>A</sup>	8	85.58 $\pm$ 6.04	Atchafalaya River <sup>B</sup>	10	46.61 $\pm$ 7.45
V	Atchafalaya River <sup>A</sup>	10	0.69 $\pm$ 0.14	Missouri River <sup>A</sup>	10	0.64 $\pm$ 0.10	lower Platte River	2	0.50 $\pm$ 0.00
Zn	Missouri River <sup>A</sup>	10	123.05 $\pm$ 39.73	lower Platte River <sup>A</sup>	8	116.01 $\pm$ 5.27	Atchafalaya River <sup>B</sup>	10	76.68 $\pm$ 7.85

Note: All concentrations are in milligrams per kilogram (mg/kg) dry weight. Missouri River data are from Ruelle and Henry (1994a), Atchafalaya River data are from Conzelmann (1997), lower Platte River data are from this study. Superscript letters indicate significance ( $p < 0.05$ ) as determined by a Kruskal-Wallis test followed by pairwise Wilcoxon rank sums tests. Sites with a sample size (N) less than 5 were not analyzed statistically.

Barium concentrations in shovelnose sturgeon from the lower Platte River also appeared to be elevated; however, the effects (if any) of elevated Ba in sturgeon are unknown due to very limited information on this subject. Anthropogenic releases of Ba are primarily from industry and can result from its use in cement, glass industries, electronics, cosmetics, pharmaceuticals, and paints (WHO, 2001). Barium also is emitted from the burning of coal, fossil fuels, and waste (WHO, 2001).

Sturgeon Food Items. The diet of shovelnose sturgeon in the lower Platte River consists mainly of aquatic invertebrates but larval fish and terrestrial invertebrates also have been found in stomach contents (Shuman, 2003). Adult pallid sturgeon feed primarily on fish of the family Cyprinidae, including carps and minnows (Dryer and Sandvol, 1993). The diet of pallid sturgeon in the lower Platte River is unknown; however, fish comprised 38 percent (by volume) of the stomach contents in nine pallid sturgeons from the Missouri River (Carlson, 1985).

Cyprinid samples used to evaluate contaminants in potential pallid sturgeon food items had significantly ( $p < 0.05$ ) greater concentrations of Hg, Mg, Se, and Zn than shovelnose sturgeon digesta, whereas shovelnose sturgeon digesta had greater concentrations of Al, B, Ba, Be, Cd, Co, Cu, Fe, Mn, Pb, and V (Table 5). Selenium, Zn, and Hg bioaccumulate in fish (Wiener et al., 2003; USDOJ, 1998). In addition, Se accumulation in fish tends to be greater from dietary sources than from water (Sandholm et al 1973). Therefore, the results of this study indicate that the more piscivorous pallid sturgeon may be exposed to greater concentrations of these elements through ingestion than shovelnose sturgeon. Although dietary uptake can contribute more than 90 percent of Hg accumulated in fish (Wiener and Spry, 1996), the results of this study indicate Hg in pallid sturgeon food items were at or below normal background concentrations (Eisler, 1987). However, concentrations of Se as low as 3 mg/kg dw in the diet of warm-water fish can lead to reproductive impairment (Lemly, 1996), and this threshold concentration was exceeded in all 11 cyprinid samples and in 4 of 11 shovelnose sturgeon digesta samples (Appendix Tables 10 and 11).

Table 5. Mean concentrations of elemental contaminants in shovelnose sturgeon digesta compared to those in potential pallid sturgeon food items from the Platte River, NE 2002.

Trace Element	Sample Matrix	N <sub>D</sub> /N <sub>A</sub>	Concentration (µg/g dw) Mean ± Standard Error	Range	Significance*
Al	Digesta	11/11	1,483 ± 296	611 - 3,750	A
Al	Cyprinid	11/11	233.16 ± 56.00	54.00 - 675.00	B
As	Cyprinid	11/11	1.94 ± 0.07	1.63 - 2.48	A
As	Digesta	11/11	1.60 ± 0.18	0.80 - 2.47	A
B	Digesta	6/11	1.08 ± 0.23	0.46 - 2.94	NA
B	Cyprinid	0/11	NA	NA	NA
Ba	Digesta	11/11	104.15 ± 38.75	24.50 - 364.00	A
Ba	Cyprinid	11/11	27.26 ± 1.81	21.90 - 41.00	B
Be	Digesta	9/11	0.06 ± 0.01	0.02 - 0.11	NA
Be	Cyprinid	0/11	NA	NA	NA
Cd	Digesta	11/11	1.58 ± 0.41	0.05 - 4.28	A
Cd	Cyprinid	11/11	0.07 ± 0.00	0.05 - 0.08	B
Cr	Digesta	11/11	1.34 ± 0.22	0.62 - 3.17	NA
Cr	Cyprinid	1/11	0.79 ± NA	NA	NA
Cu	Digesta	11/11	9.71 ± 1.78	1.18 - 19.00	A
Cu	Cyprinid	11/11	2.90 ± 0.09	2.44 - 3.27	B
Fe	Digesta	11/11	2,478 ± 542.54	969 - 7,390	A
Fe	Cyprinid	11/11	275.66 ± 59.12	88.30 - 735.00	B
Hg	Cyprinid	11/11	0.137 ± 0.007	0.11 - 0.16	A
Hg	Digesta	4/11	0.03 ± 0.00	0.02 - 0.04	B
K	Cyprinid	11/11	11,909 ± 120	11,300 - 12,600	A
K	Digesta	11/11	1711.00 ± 353.00	332.00 - 3620.00	B
Mg	Cyprinid	11/11	1,574 ± 63	1,220 - 1,960	A
Mg	Digesta	11/11	586.09 ± 114.12	168.00 - 1480.00	B
Mn	Digesta	11/11	71.69 ± 10.24	29.20 - 140.00	A
Mn	Cyprinid	11/11	38.22 ± 5.10	22.00 - 79.30	B
Ni	Digesta	11/11	2.07 ± 0.35	0.72 - 3.93	NA
Ni	Cyprinid	2/11	0.75 ± 0.13	0.62 - 0.88	NA
Pb	Digesta	11/11	2.27 ± 0.20	1.05 - 3.17	A
Pb	Cyprinid	11/11	0.27 ± 0.05	0.11 - 0.66	B
Se	Cyprinid	11/11	4.50 ± 0.27	3.15 - 5.51	A
Se	Digesta	11/11	2.49 ± 0.62	0.19 - 6.25	B
V	Digesta	11/11	4.39 ± 1.24	1.46 - 16.00	NA
V	Cyprinid	2/11	1.27 ± 0.20	1.07 - 1.47	NA
Zn	Cyprinid	11/11	207.36 ± 3.37	182.00 - 223.00	A
Zn	Digesta	11/11	91.85 ± 19.62	8.97 - 216.00	B

Note: \*Different letters indicate significant ( $p < 0.05$ ) differences as determined by a Wilcoxon test. SE = standard error, NA = not applicable.



## Organochlorine Chemical Residues

Organochlorines detected in less than 50 percent of ovary and carcass tissues included DDT, hexachlorobenzene, endrin, mirex, lindane, toxaphene, and cis-nonachlor. Organochlorines frequently detected in either shovelnose sturgeon carcass (n = 8) or ovary (n = 9) samples included dieldrin, DDE, DDD, alpha, and gamma chlordane, oxychlordane, heptachlor epoxide, trans-nonachlor, and PCBs (Appendix Tables A.12 and A.13). For these OCs, there are no known published toxicity thresholds for sturgeon based on tissue residues; therefore, concentrations in shovelnose sturgeon were compared to guidelines for the protection of fish-eating wildlife or published toxicity thresholds for other fish species (Table 6). It is noteworthy that two ovary samples exceeded U.S. Food and Drug Administration (USFDA) human health action levels for concentrations of total PCBs and/or total chlordane (i.e., the sum of alpha and gamma chlordane, oxychlordane, and cis and trans nonachlor) in edible fish tissue ( 2.0 and 0.3 mg/kg ww, respectively) (USFDA, 2002).

Dieldrin, total DDT (i.e., the sum of p,p' and o,p' isomers of DDT, DDE and DDD), and total chlordane exceeded protective guidelines for fish-eating wildlife but did not exceed any known fish toxicity thresholds. Dieldrin was detected in 5 of 8 carcass samples and all ovary samples, but concentrations did not exceed a 1.2 - 1.4 µg/g ww toxicity threshold range for freshwater fish (Jarvinen and Ankley, 1999 as cited by Schmitt, 2002). Total DDT consisted mainly of p,p'-DDE (the most stable and toxic DDT metabolite) and was detected in 7 of 8 carcasses sampled and all ovary samples. Concentrations of total DDT in carcass tissues did not exceed a 0.2 µg/g ww total DDT guideline for the protection of fish-eating wildlife (Newell et al., 1987); but this guideline was exceeded in three ovary samples. Total chlordane residues exceeded a 0.1 mg/kg ww guideline for the protection of predatory fish (Eisler, 1990) in one carcass sample and four ovary samples.

Concentrations of total PCBs in shovelnose sturgeon tissues exceeded a number of toxicity thresholds for fish, and potential adverse effects to sturgeon in the lower Platte River warrant concern. PCBs in six carcasses were above the 0.11 µg/g ww guideline for fish-eating wildlife (Newell et al., 1987) and two ovary samples exceeded a 0.3 mg/kg ww egg guideline for the protection of aquatic life (Eisler, 1986). Mean percent lipids

Table 6. Concentrations of organochlorine contaminants in shovelnose sturgeon ovary and carcass samples from the lower Platte River, 2002, compared to fish tissue guidelines or effects thresholds.

Organochlorine	Ovary				Carcass				Tissue Guideline or Effects Threshold and References
	N <sup>D</sup> /N <sup>A</sup>	Mean ± SE	Min	Max	N <sup>D</sup> /N <sup>A</sup>	Mean ± SE	Min	Max	
chlordane (alpha)	9/9	0.038 ± 0.013	0.012	0.110	3/8	NA	< 0.002	0.057	NA
chlordane (gamma)	9/9	0.020 ± 0.007	0.004	0.061	5/8	0.008 ± 0.004	< 0.002	0.037	NA
oxychlordane	9/9	0.014 ± 0.004	0.004	0.037	2/8	NA	< 0.002	0.009	NA
trans-nonachlor	9/9	0.063 ± 0.023	0.017	0.220	7/8	0.024 ± 0.008	< 0.002	0.068	NA
Total Chlordane	9/9	0.158 ± 0.058	0.039	0.515	7/8	0.047 ± 0.020	< 0.002	0.193	0.1 <sup>A</sup> , 0.5 <sup>B</sup>
dieldrin	9/9	0.039 ± 0.013	0.005	0.110	5/8	0.020 ± 0.010	< 0.002	0.060	0.12 <sup>B</sup> , 1.2 <sup>C</sup>
heptachlor epoxide	9/9	0.014 ± 0.005	0.003	0.045	1/8	NA	< 0.002	0.013	0.2 <sup>B</sup>
PCB-1254	8/9	0.246 ± 0.116	< 0.01	1.090	2/8	NA	< 0.01	0.31	0.33 <sup>D</sup> , 0.66 <sup>E</sup>
PCB-1260	6/9	0.162 ± 0.113	< 0.01	1.060	8/8	0.251 ± 0.071	< 0.01	0.54	2.1 <sup>F</sup>
Total PCBs	8/9	0.405 ± 0.227	< 0.01	2.150	6/8	0.320 ± 0.130	< 0.01	1.100	0.02 <sup>G</sup> , 0.11 <sup>B</sup> , 0.12 <sup>H</sup> , 0.3 <sup>I</sup> , 0.4 <sup>J</sup>
p'p'-DDD	9/9	0.073 ± 0.027	0.022	0.240	5/8	0.018 ± 0.007	< 0.002	0.053	NA
p'p'-DDE	9/9	0.265 ± 0.114	0.051	0.900	7/8	0.058 ± 0.013	< 0.002	0.110	NA
Total DDT	9/9	0.354 ± 0.149	0.074	1.211	7/8	0.075 ± 0.02	< 0.002	0.160	0.014 <sup>K</sup> , 0.2 <sup>B</sup>

Note: All concentrations are in mg/kg wet weight. N<sup>D</sup> = number of detects; N<sup>A</sup> = number of samples analyzed; SE = standard error; Min = minimum; Max = maximum; NA = not applicable; Total Chlordane = the sum of alpha and gamma chlordane, oxychlordane, and cis and trans nonachlor; Total PCBs = the sum of Aroclors 1242, 1248, 1254, and 1260; and Total DDT = the sum of p,p' and o,p' isomers of DDT, DDE and DDD.

References:

- <sup>A</sup> Eisler, 1990 (wholebody concentration guideline to protect predatory fish).
- <sup>B</sup> Newell et al., 1987 (fish flesh guideline to protect fish-eating wildlife).
- <sup>C</sup> Jarvinen and Ankley, 1999 (whole body concentration for survival in freshwater fish).
- <sup>D</sup> Eisler, 1986 (egg concentration for prehatch mortality in trout eggs).
- <sup>E</sup> McCarthy et al., 2003 (egg concentration resulting in decreased growth and startling response).
- <sup>F</sup> Matta et al., 1998 (concentration in larvae with abnormal oocyte development).
- <sup>G</sup> Nakayama et al., 2005 (egg concentration resulting in delayed time to hatch).
- <sup>H</sup> Von Westernhagen 1981 (egg concentration resulting in decreased hatchability).
- <sup>I</sup> Eisler and Belisle, 1996 (egg derived criteria for the protection of aquatic life).
- <sup>J</sup> Eisler and Belisle, 1996 (whole body derived criteria for the protection of aquatic life).
- <sup>K</sup> Environment Canada, 1998 (tissue residue guidelines for protection of aquatic life).

were greater for ovary (average =  $52 \pm 5$  percent) than carcass (average =  $5 \pm 1$  percent), and likely accounted for the greater concentrations of PCBs in ovaries (Niimi, 1983).

Although the shovelnose sturgeon (SP-14) with the greatest concentration of PCBs was older (13 years) than the average age of sturgeon collected for this study (9 years), small sample size ( $n = 7$ ) precluded any significance in the correlation between age and PCB carcass residues. Older fish typically have greater tissue concentrations of PCBs (Ion et al., 1997; Lafontaine et al., 2002).

Concentrations of PCBs in shovelnose sturgeon carcass samples were generally below published toxicity thresholds in the Environmental Residue-Effects Database (ERED, 2004). Toxicity concentrations in ERED for whole body adult fish (six different species) ranged from a 0.14 mg/kg ww lowest observed effects concentration (LOEC) for liver effects (increased EROD and mass) to a 170 mg/kg ww toxicity threshold for reduced egg hatchability by 83 percent (ERED, 2004).

The transfer of PCBs from female fish to eggs is a concern because it can result in developmental toxicity to offspring including decreased egg and larval viability (Willford et al., 1981; Black et al., 1988; Ankley et al., 1991). Toxic effect concentrations in fish based on PCB residues in eggs, as reported by publications in ERED (2004), ranged from 0.02 mg/kg ww (decreased time to hatch) to 170 mg/kg ww (72 percent decrease in hatchability of eggs). Although most of the PCB toxicity thresholds for fish in ERED were not exceeded in shovelnose sturgeon ovaries from this study, some published thresholds were exceeded including delayed time of hatch for Japanese Medaka (Nakayama et al., 2005), and decreased hatchability of flounder (Von Westernhagen et al., 1981; as cited by Ray et al., 1984). Two ovary samples and two carcass samples (all from different fish) also exceeded total PCB criteria for the protection of aquatic life (0.3 and 0.4 mg/kg ww, respectively)(Eisler and Belisle, 1996).

Comparisons between field studies of health effects in fish are typically confounded by exposure to chemical mixtures that vary between sites. However, health assessments of wild fish populations can include more realistic scenarios than controlled lab experiments. Health effects reported in wild fish from areas predominately contaminated with PCBs included reduced fertility, hormone imbalance, delayed ovarian

development, ovarian atresia, decreased egg and larval viability, and reduced larval growth (Casillas et al., 1991; Niimi, 1996).

PCB contaminated waterbodies are a concern in Nebraska. There are 17 impaired waterbodies in Nebraska where efforts are required by the U.S. Environmental Protection Agency (USEPA) to reduce PCB contamination (USEPA, 2004). One of these sites includes the lower Platte River from the confluence of the Loup River diversion canal to the confluence of the Elk Horn River (segment LP1-20000; NDEQ, 2004). Sources for the PCB contamination in this river segment have not been identified (Patrick O'Brien, Nebraska Department of Environmental Quality, pers. comm., 2004).

Concentrations of most OCs in shovelnose sturgeon from the lower Platte River were generally less than those reported from other sites known to be contaminated by OCs (Table 7). The shovelnose sturgeon from the OC-affected site on the Mississippi River near Chester, Illinois had health anomalies and increased liver mass relative to shovelnose sturgeon from a reference site near Davenport, Iowa, where no health anomalies were observed (Table 7) (Coffey et al., 2003). These health anomalies included ova-testis in two male fish as well as increased plasma estrogen and VTG in blood plasma of all males from the OC contaminated site (Coffey et al., 2003).

Concentrations of total OCs were similar between shovelnose sturgeon from the Platte River and those collected previously in the Missouri River by Ruelle and Henry (1994a); however, limited sample sizes in both studies preclude any statistical comparisons.

Shovelnose sturgeon from the lower Platte River had lower concentrations of total chlordane (mean =  $0.047 \pm 0.02$ ) than those previously reported for the Missouri River below Gavins Point Dam, Nebraska (Allen and Wilson, 1991). Chlordane residues in shovelnose sturgeon collected in 1988 increased along a downstream gradient from 0.14  $\mu\text{g/g}$  near Blair, Nebraska, to 0.22  $\mu\text{g/g}$  at Omaha, Nebraska, to 0.33  $\mu\text{g/g}$  at Atchison, Kansas (Allen and Wilson, 1991). These results, in conjunction with the results from this study, indicate that chlordane residues in shovelnose sturgeon have decreased since 1988. The BEST program reported a general decline in chlordane residues from fish sampled from the Mississippi River basin in 1986 and 1995 (Schmitt, 2002).

Table 7. Mean  $\pm$  standard error (SE) concentrations of organochlorine contaminants (OCs) in shovelnose sturgeon carcass or whole body samples from the Mississippi River, Atchafalaya River, and Platte River.

Organochlorine	River	N <sup>D</sup>	N <sup>A</sup>	Concentration $\mu\text{g/g}$ wet weight		Reference
				Mean $\pm$ SE	Range	
Dieldrin	*Mississippi	9	10	0.06 $\pm$ 0.01	< 0.01 - 0.11	Coffey et al., 2000
	Mississippi Reference	8	10	0.05 $\pm$ 0.01	< 0.01 - 0.11	Coffey et al., 2000
	*Atchafalaya	10	10	0.05 $\pm$ 0.01	0.01 - 0.08	Conzelman et al., 1997
	Lower Platte	5	8	0.02 $\pm$ 0.01	< 0.002 - 0.06	Current study
	Missouri	1	4	NA	<0.01 0.01	Ruelle and Henry, 1994a
Total PCBs	*Mississippi	10	10	0.81 $\pm$ 0.12	0.31 - 1.50	Coffey et al., 2000
	*Atchafalaya	10	10	0.45 $\pm$ 0.11	0.13 - 1.40	Conzelman et al., 1997
	Lower Platte	6	8	0.32 $\pm$ 0.13	<0.01 - 1.10	Current study
	Missouri	4	4	0.23 $\pm$ 0.04	0.16 0.32	Ruelle and Henry, 1994a
	Mississippi Reference	10	10	0.23 $\pm$ 0.03	0.09 - 0.49	Coffey et al., 2000
Total DDT	*Atchafalaya	10	10	0.53 $\pm$ 0.08	0.25 - 1.10	Conzelman et al., 1997
	*Mississippi	10	10	0.19 $\pm$ 0.03	0.07 - 0.34	Coffey et al., 2000
	Missouri	4	4	0.17 $\pm$ 0.07	0.04 - 0.34	Ruelle and Henry, 1994a
	Lower Platte	7	8	0.07 $\pm$ 0.02	< 0.002 - 0.16	Current study
	Mississippi Reference	10	10	0.04 $\pm$ 0.01	0.02 - 0.07	Coffey et al., 2000
Total OCs	*Atchafalaya	10	10	2.10 $\pm$ 0.37	1.09 - 5.28	Conzelman et al., 1997
	*Mississippi	10	10	1.24 $\pm$ 0.16	0.53 - 2.34	Coffey et al., 2000
	Lower Platte	7	8	0.46 $\pm$ 0.17	< 0.01 - 1.53	Current study
	Missouri River	4	4	0.44 $\pm$ 0.13	0.21 0.74	Ruelle and Henry, 1994a
	Mississippi Reference	10	10	0.34 $\pm$ 0.05	0.12 - 0.64	Coffey et al., 2000

\* = site with known OC contamination. N<sup>D</sup> = number of detects, N<sup>A</sup> = number analyzed, NA = not applicable. Half the detection limit was substituted for values below detection to calculate the mean. Total PCBs include the sum of Aroclors 1242, 1248, 1254, and 1260. Total DDT = p,p' and o,p' isomers of DDT, DDE, and DDD. Total OCs equals the sum of all OCs that were above the detection limit. For the Atchafalaya River and Missouri River, total OCs included contaminants (toxaphene, mirex, endrin, hexachlorobenzene, and lindane) that were below detection limits in shovelnose sturgeon from the lower Platte River.

Many of the organochlorines detected in shovelnose sturgeon tissues can disrupt hormone systems including dieldrin, DDE, PCBs, and chlordane. Dieldrin has a low affinity for the estrogen receptor in fish and can result in decreased E<sub>2</sub> plasma concentrations in fish (Petit et al., 1997; Muller et al., 2002). Anti-androgenic effects in fish exposed to p,p'-DDE can include suppressed courtship behavior, delayed maturation, decreased plasma E<sub>2</sub>, and reduced sperm count and testes size (Baatrup and Junge, 2001; Bayley et al., 2002; Muller et al., 2002). PCBs can produce either estrogenic or anti-estrogenic effects in fish depending on their chemical structure (Westerlund et al., 2000; Olsson et al., 2000; Letcher et al., 2002).

Concentrations of organochlorines in cyprinid samples (n = 10) and shovelnose sturgeon stomach contents (n = 10) were all below detection limits (Appendix Tables 14 and 15). The apparent low OC dietary exposure to shovelnose sturgeon supports the finding of generally low residues in sturgeon tissues. However, tissue residues indicate that some organochlorines, such as PCBs, are bioaccumulating in shovelnose sturgeon and exposure to PCBs in sediments may be an important exposure pathway.

#### Herbicides in Water

Concentrations of total herbicides in water grab samples (n = 5) ranged from 2 to 48 µg/L, with atrazine being the predominant herbicide detected (Appendix Table A.16). Equal-width and depth-integrated water-column samples collected by the USGS-WRD in Lincoln, Nebraska also contained detectable concentrations of atrazine on all sampling occasions (n = 8) with mean concentrations ranging from 0.6 µg/L at Duncan Bridge to 14.3 µg/L at the highway 50 bridge near Louisville, NE (Appendix Table A.17). Overall, waterborne atrazine exceeded the Nebraska chronic water quality criterion for aquatic life (12 µg/L; NDEQ, 2002) in three of 13 samples analyzed.

Previous monitoring of atrazine concentrations in the lower Platte River by NAWQA in May of 1992 found that it often exceeded 20µg/L (Frenzel et al., 1998). Atrazine is the most heavily applied pesticide for corn production in Nebraska (NASS, 2003). In 2002, Nebraska farmers applied 0.91 lbs of atrazine per acre to 64 percent of

the total corn crop for a total usage of 5,356,000 lbs (NASS, 2003). Atrazine degradation products (desethyl atrazine and deisopropyl atrazine) were detected at much lower concentrations than the parent compound, indicating that most of the atrazine detected was from applications made during the spring of 2002. However, sources of atrazine input into the lower Platte River are not necessarily local as degradation by photolysis and hydrolysis can occur slowly (half-life of 335 days; Solomon et al., 1996) and result in long-distance transport.

#### Herbicides in Shovelnose Sturgeon

Concentrations of herbicides were detected in all shovelnose sturgeon blood plasma samples (n = 50), but not in any liver samples (n = 19; all less than 0.05 µg/g or parts per million ww). Although atrazine was not detected in liver, concentrations in the liver were likely greater than those measured in blood plasma because the bioconcentration factor of atrazine in fish is greater in liver tissue than blood (Gunkel, 1981). Whitefish exposed to atrazine in water at a concentration of 50 µg/L resulted in bioconcentration factors as high as 8.2 and 2.2 for liver and blood, respectively (detection limit = 0.05 µg/g, Gunkel, 1981). The detection of atrazine in shovelnose sturgeon blood but not liver is likely due to differences in specificity and sensitivity between the ELISA and analytical methods used. Furthermore, liver samples from our study had an average mass of  $3.8 \pm 0.3$  grams, whereas the recommended tissue sample mass for analytical atrazine analysis is 5 grams (Dr. Christina Lusk, Analytical Chemist, Mississippi State Chemical Lab, pers. comm., 2005).

Concentrations of atrazine in shovelnose sturgeon blood plasma ranged from greater than 30 µg/L (actual concentration not determined by FCSC) to 0.24 µg/L (Appendix Table A.6). Atrazine concentrations differed significantly among sites (Figure 10); however, variation in atrazine concentrations both within sites and among sites was apparently influenced more by peaks in daily river flow velocity than by location. The highest atrazine concentrations in shovelnose sturgeon blood plasma (i.e., concentrations > 12 µg/L) occurred during peaks in daily mean flow rate, whereas lower concentrations of atrazine were detected in fish collected between peaks (Figure 11). These peak flows

were attributed to rainfall events and increased runoff from agricultural fields, resulting in increased atrazine exposures to shovelnose sturgeon. There were no significant correlations between concentrations of atrazine in shovelnose sturgeon blood samples and biomarker measurements (E2, 11KT, E/KT, Vt, HSI, SSI, GSI, CF). Neither were there differences in atrazine concentrations between genders.

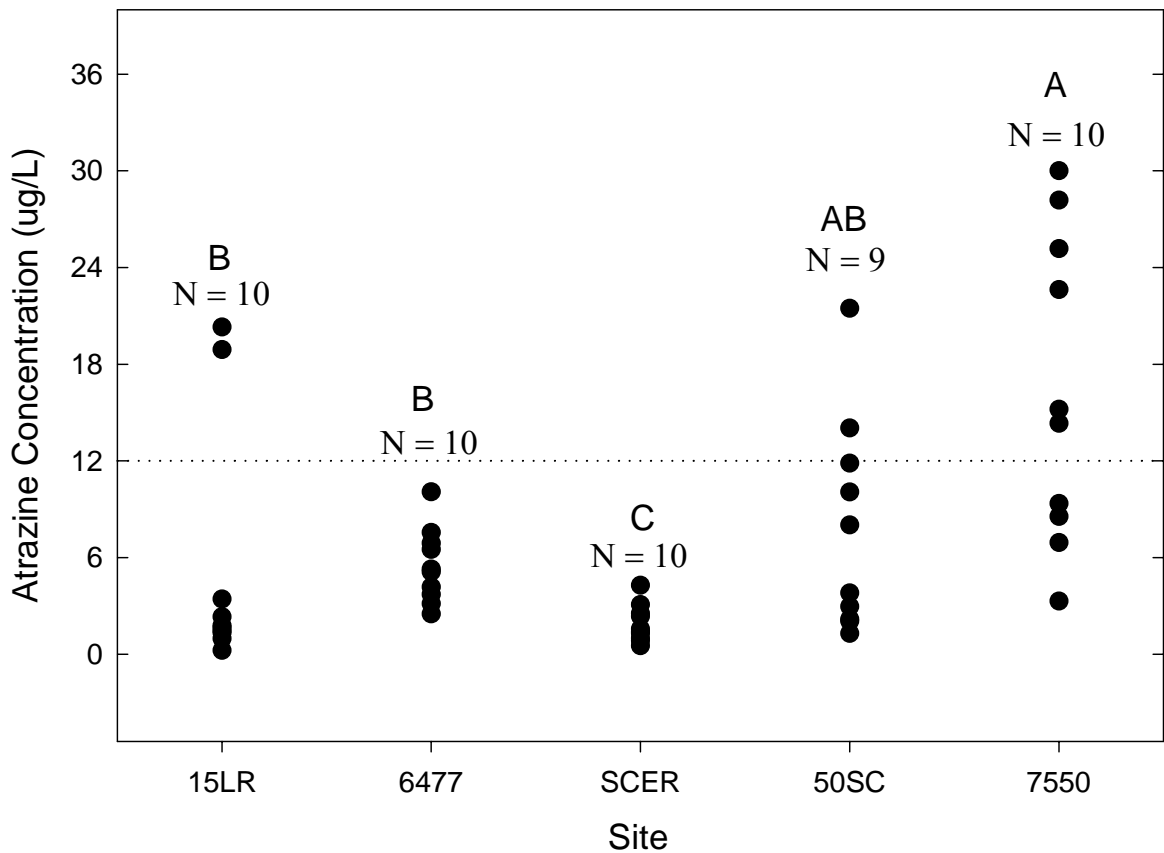


Figure 10. Concentrations of atrazine in blood plasma from shovelnose sturgeon collected at five sites in the lower Platte River, Nebraska. Note: N = sample size, letters indicate significant differences ( $p < 0.05$ ) as determined by a Kruskal-Wallis test followed by pairwise Wilcoxon rank sums tests. Sites progress downstream from 15LR to 7550 (see Figure 2).



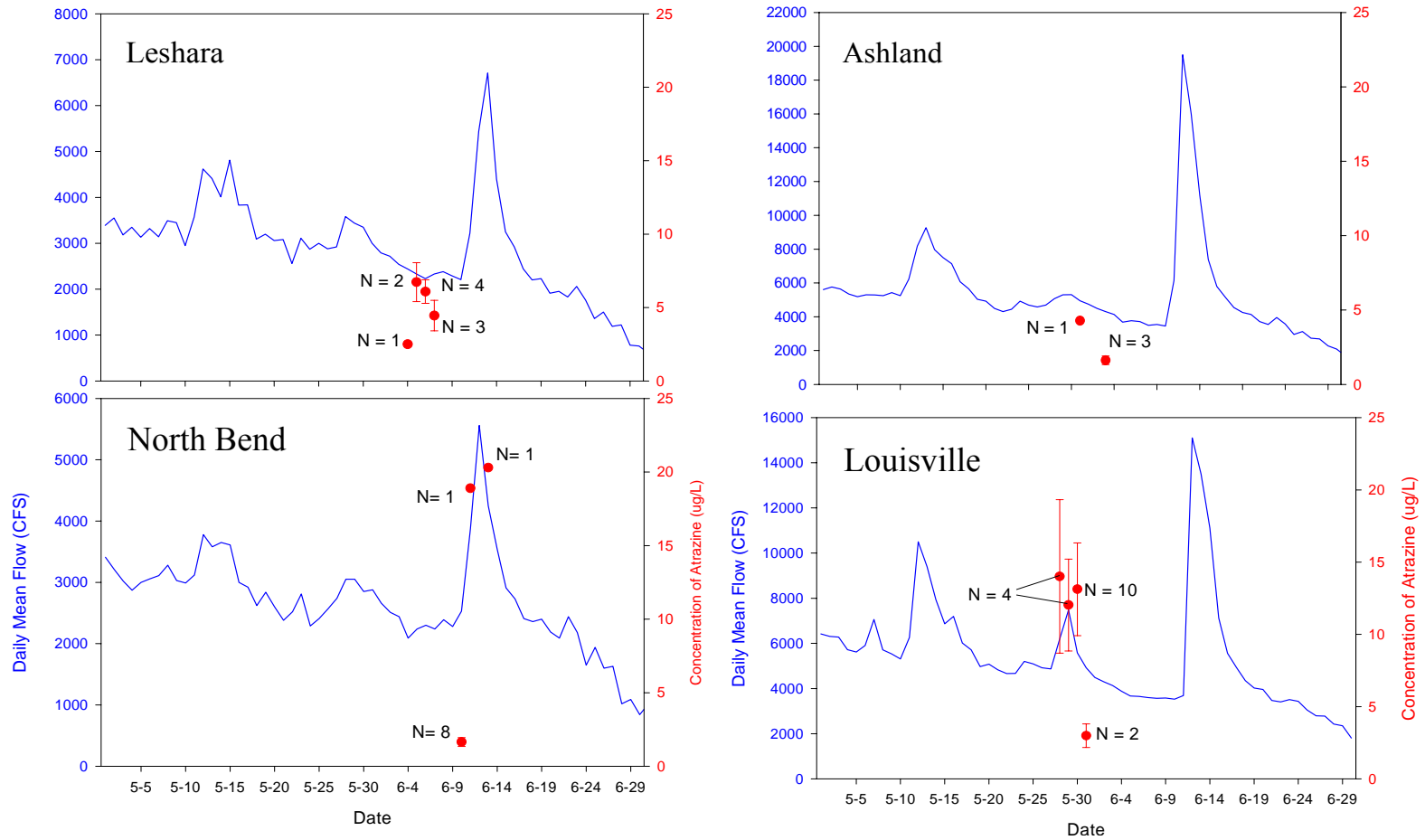


Figure 11. Concentrations of atrazine in blood plasma from shovelnose sturgeon compared to daily flow velocity in the lower Platte River measured at Leshara, Ashland, North Bend, and Louisville, NE, 2002. N = sample size and error bars indicate the standard error for the mean. Dates are in month-day format. Daily flow velocity is from NWIS (2004).

Waterborne atrazine is rapidly taken up by fish through the gills where it enters the bloodstream and is systemically transported to the other organs (Gunkel and Streit, 1980). Although the effects of atrazine on shovelnose sturgeon and pallid sturgeon are unknown, the ability of atrazine to adversely affect other fish species is well documented and includes endocrine disruption (Moore and Waring, 1998; Moore and Lower, 2001; (Spanó et al., 2004), altered kidney morphology (Fisher-Scheri et al., 1991; Oulmi et al., 1995), reduced larval growth (Alvarez and Fuiman, 2005) and altered behavior (Saglio and Trijasse, 1998).

#### Condition and Organo-somatic Indices

Condition and organo-somatic indices were measured for 53 shovelnose sturgeon from the lower Platte River (Appendix A.18). There were no significant differences in CF,  $W_R$  or HSI between genders for any of the sites; however, male shovelnose sturgeon had a significantly ( $p < 0.05$ ) greater mean SSI than females (Table 8).

Condition factor,  $W_R$ , and  $HSI_{GI}$  in shovelnose sturgeon from the lower Platte River were comparable to those measured from a reference site on the Mississippi River by Coffey et al. (2003), whereas shovelnose sturgeon from the contaminated site on the Mississippi River had a significantly ( $p < 0.05$ ) greater  $HSI_{GI}$  and lower CF and  $W_R$  (Figure 12). The greater HSI and decreased CF and  $W_R$  in shovelnose sturgeon from the contaminated site on the Mississippi River may be in response to increased exposure to OC compounds. Decreased CF and increased HSI have been reported in fish from sites contaminated with PCBs (Buckley et al., 1985; Jaworska et al., 1997; Khan, 1999).

Interpretation of the SSI and GSI measurements for shovelnose sturgeon from this study was limited by a lack of other SSI and GSI data for the species. However, the BEST program also reported a significant difference in SSI between male and female carp and found that male carp from the Platte River (BEST station 89) had the greatest mean SSI compared to 47 other sites in the central U.S. (Schmitt, 2002). It is unclear why male shovelnose sturgeon and carp from the lower Platte River have a higher SSI compared to their female counterparts. However, increased SSI can be indicative of

disease or immune dysfunction (Goede and Barton, 1990 as cited by Schmitt, 2002) and decreased SSI has been associated with exposure to PCBs, PAHs, and metals (Schmitt and Dethloff, 2000).

Table 8. Condition indices for shovelnose sturgeon collected from the lower Platte River, Nebraska, in May and July of 2002.

Condition Index	Gender	N	Mean $\pm$ SE	*Significance	Range
CF	F	20	0.38 $\pm$ 0.01	A	0.31 - 0.47
	M	33	0.36 $\pm$ 0.01	A	0.29 - 0.44
W <sub>R</sub>	F	20	89.78 $\pm$ 2.23	A	74.72 - 112.60
	M	33	84.73 $\pm$ 2.05	A	66.19 - 106.85
SSI	M	33	98.15 $\pm$ 0.18	A	77.08 - 99.40
	F	20	93.51 $\pm$ 1.54	B	94.49 - 99.50
HSI	F	20	1.05 $\pm$ 0.07	A	0.70 - 1.75
	M	33	0.95 $\pm$ 0.06	A	0.38 - 1.58
GSI	F	20	6.50 $\pm$ 1.54	A	0.60 - 22.92
	M	33	1.85 $\pm$ 0.18	B	0.50 - 5.51

Note: \* Different letters indicates significant ( $p < 0.05$ ) differences between genders as determined by a Wilcoxon rank sums test. N = sample size, CF = condition factor, W<sub>R</sub> = relative weight, SSI = spleno-somatic index, HSI = hepato-somatic index, GSI = gonado-somatic index, F = female, M = male.

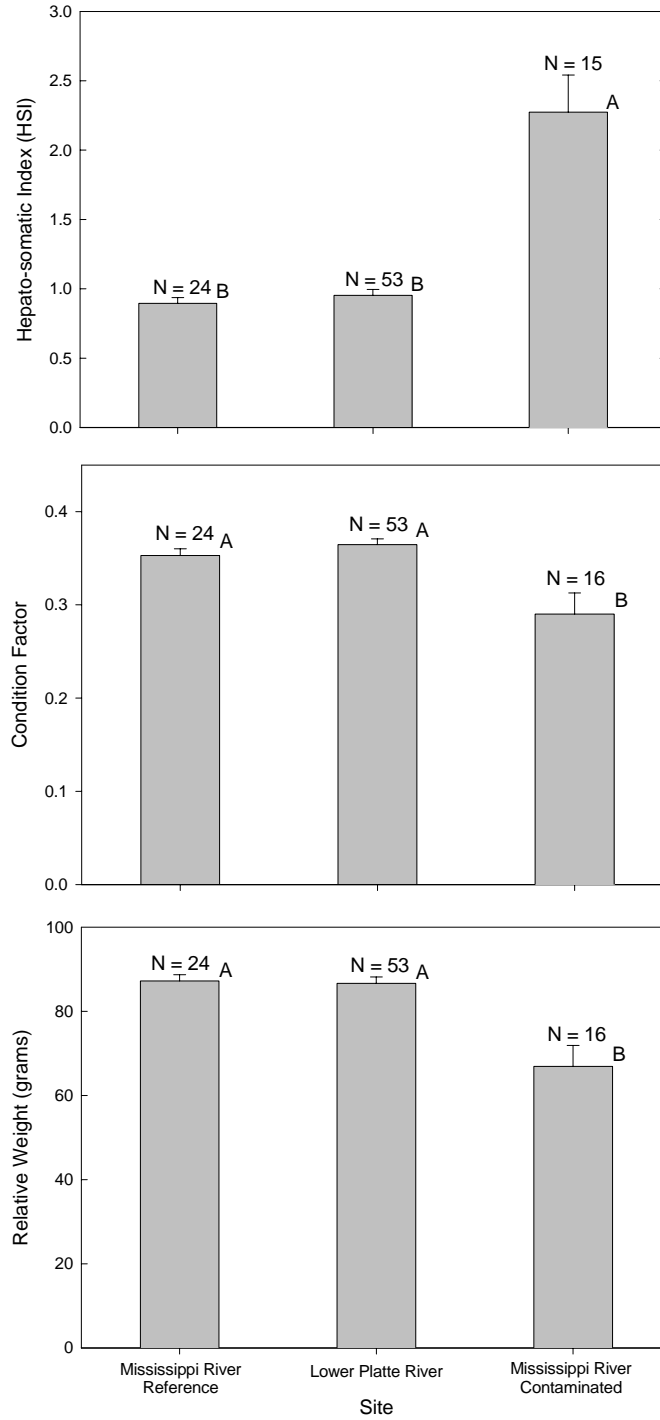


Figure 12. Mean ( $\pm$ SE) relative weight ( $W_r$ ), condition factor (CF), and hepato-somatic index for shovelnose sturgeon from the lower Platte River and two sites on the Mississippi River sampled by Coffey et al. (2003). N = sample size. Letters indicate significance ( $p < 0.05$ ) as determined by a Kruskal-Wallis test followed by pairwise Wilcoxon rank sums tests.

### Risk to Pallid Sturgeon

A principal objective of this research was to use data from shovelnose sturgeon to evaluate contaminant exposure and potential adverse effects to pallid sturgeon. This can be accomplished by considering the likely differences between these species in contaminant exposure and effects based upon their different life history characteristics. Pallid sturgeon have a more piscivorous diet, greater maximum life-span, and longer reproductive cycle than shovelnose sturgeon (Moos, 1978; Carlson, 1985; Ruelle and Keenlyne, 1994). These differences may make pallid sturgeon more susceptible than shovelnose sturgeon to environmental contaminants that bioaccumulate and/or cause reproductive effects.

A limited comparison of OC and elemental contaminant uptake between pallid sturgeon and shovelnose sturgeon can be made by comparing tissue residues for these species. A number of studies have reported concentrations of elemental contaminants and OCs in shovelnose sturgeon (Allen and Wilson, 1991; Welsh and Olson, 1992; Welsh, 1992; Ruelle and Henry, 1994a; Palawski and Olsen, 1996; Conzelmann, 1997; Coffey et al., 2003). Reports of elemental contaminant concentrations in pallid sturgeon tissues (i.e., gonad, liver, and muscle) are much more limited and were first reported only in three fish from the Missouri River by Ruelle and Keenlyne (1993). This was followed by a report that included data for 13 other pallid sturgeon, mainly from the Mississippi River (Ruelle and Henry, 1994b). Differences in the tissues analyzed, collection locations, and time of collections limit the usefulness of these data for comparing contaminant accumulation between the species. Probably the best comparisons that can be made are for elemental contaminants and OC concentrations in liver and muscle samples collected from sturgeon in the Missouri River between 1983 and 1992 (Tables 9 and 10).

Table 9. Mean concentrations of organochlorine contaminants in pallid sturgeon and shovelnose sturgeon tissues collected from the Missouri River, 1983 to 1991.

Tissue	Organochlorine	Pallid Sturgeon <sup>1</sup>			Shovelnose Sturgeon <sup>2</sup>		
		N	Concentration ( $\mu\text{g/g}$ wet weight)		N	Concentration ( $\mu\text{g/g}$ wet weight)	
			Mean $\pm$ SE	Range		Mean	Range
Liver	Dieldrin	2	0.07 $\pm$ 0.02	0.05 - 0.08	5	NC	<0.01 - <0.01
	o,p'- DDD	2	0.09 $\pm$ 0.07	0.02 - 0.16	5	NC	<0.01 - <0.01
	o,p'-DDE	2	NC	<0.010 - 0.10	5	NC	<0.01 - <0.01
	o,p'-DDT	2	0.12 $\pm$ 0.02	0.10 - 0.13	5	NC	<0.01 - <0.01
	p,p'.DDD	2	0.98 $\pm$ 0.73	0.25 - 1.71	5	NC	<0.01 - 0.08
	p,p'.DDE	2	3.41 $\pm$ 0.59	2.82 - 4.00	5	0.40	0.11 - 0.86
	p,p'.DDT	2	0.11 $\pm$ 0.03	0.08 - 0.14	5	NC	<0.01 - <0.01
	total PCB	2	11.12 $\pm$ 9.40	1.72 - 20.51	5	1.50	1.20 - 2.00
Muscle	Dieldrin	3	0.07 $\pm$ 0.04	0.01 - 0.14	5	NC	<0.01 - 0.01
	o,p'- DDD	3	0.04 $\pm$ 0.02	0.01 - 0.08	5	NC	<0.01 - <0.01
	o,p'-DDE	3	NC	<0.010 - 0.12	5	NC	<0.01 - <0.01
	o,p'-DDT	3	0.18 $\pm$ 0.13	0.01 - 0.44	5	NC	<0.01 - <0.01
	p,p'.DDD	3	0.52 $\pm$ 0.37	0.01 - 1.24	5	0.01	<0.01 - 0.02
	p,p'.DDE	3	2.42 $\pm$ 1.20	0.02 - 3.67	5	0.05	0.02 - 0.09
	p,p'.DDT	3	0.14 $\pm$ 0.07	0.01 - 0.26	5	NC	<0.01 - <0.01
	total PCB	3	9.29 $\pm$ 8.08	0.07 - 25.38	5	0.17	<.005 - 0.23

Note: <sup>1</sup> = data from Ruelle and Henry (1994b), <sup>2</sup> = data from Welsh and Olson (1992).

A standard error (SE) was not reported for the shovelnose sturgeon data. < indicates a sample was below the detection limit (value = detection limit). NC = a mean and SE was not calculated because 50 percent or more of the samples analyzed were below the detection limit.

Although the data in Tables 8 and 9 should be interpreted cautiously due to small sample sizes and differences in collection sites and date, it is noteworthy that pallid sturgeon consistently had higher concentrations of OCs than shovelnose sturgeon (Table 9). Differences between pallid sturgeon and shovelnose sturgeon metal accumulation are less obvious; however, pallid sturgeon appear to accumulate more Hg and less Se and Zn than shovelnose sturgeon (Table 10).

Table 10. Mean ( $\pm$ SE) concentrations of elemental contaminants in shovelnose sturgeon and pallid sturgeon tissues collected from the Missouri River between 1983 and 1992.

Species	Tissue	Sample Sizes and Concentration in mg/kg dry weight								Reference								
		Ba		Hg		Se		Zn										
		N <sub>A</sub>	N <sub>D</sub>	Mean	$\pm$ SE	N <sub>A</sub>	N <sub>D</sub>	Mean	$\pm$ SE		N <sub>A</sub>	N <sub>D</sub>	Mean	$\pm$ SE				
Shovelnose Sturgeon	Gonad	3	6	NC		3	6	NC		6	6	2.22	$\pm$ 0.30	6	6	20.42	$\pm$ 2.62	1
	Liver	6	6	21.60	$\pm$ 4.29	6	6	0.83	$\pm$ 0.13	6	6	9.34	$\pm$ 2.22	6	6	69.73	$\pm$ 7.44	
	Muscle	6	6	6.60	$\pm$ 1.72	6	6	0.47	$\pm$ 0.05	6	6	3.29	$\pm$ 0.14	6	6	33.35	$\pm$ 8.37	
	Gonad	9	9	6.77	$\pm$ 1.80	1	9	NC		9	9	6.63	$\pm$ 1.07	9	9	99.09	$\pm$ 37.65	
	Liver	9	9	35.81	$\pm$ 6.23	9	9	1.10	$\pm$ 0.21	9	9	16.59	$\pm$ 1.61	9	9	99.21	$\pm$ 5.63	
Pallid Sturgeon	Gonad	2	2	1.17	$\pm$ 0.05	2	2	0.72	$\pm$ 0.45	2	2	1.49	$\pm$ 0.91	2	2	30.33	$\pm$ 26.17	3
	Liver	3	3	7.63	$\pm$ 2.65	3	3	6.45	$\pm$ 4.79	3	3	5.63	$\pm$ 1.08	3	3	38.80	$\pm$ 11.54	
	Muscle	4	4	1.23	$\pm$ 0.27	4	4	2.03	$\pm$ 0.18	4	4	1.88	$\pm$ 0.89	4	4	15.25	$\pm$ 4.22	

Note: N<sub>A</sub> = the number of samples analyzed and N<sub>D</sub> = the number of detects. NC = not calculated because 50% or more of the samples analyzed were below the detection limit. For the Reference column, 1 = Welsh and Olson, 1992; 2 = Fannin and Esmoil, 1992; 3 = Ruelle and Henry, 1994b.

The results from this study indicate that the diet of pallid sturgeon could result in greater exposure to Se, Hg, and Zn relative to shovelnose sturgeon in the lower Platte River. Of these elements, Se appears to be of most concern regarding pallid sturgeon. Concentrations of Se in pallid sturgeon are not necessarily expected to be greater than those measured for shovelnose sturgeon because Se does not tend to biomagnify in fish when considering whole-body residues (Ohlendorf, 2003). However, concentrations of Se in potential pallid sturgeon food items and shovelnose sturgeon carcasses from the lower Platte River exceeded levels where reproductive impairment may begin to occur in fish. More work is needed to better evaluate whether these concentrations are detrimental to pallid sturgeon recovery.

With the exception of total PCBs, organochlorine contamination in shovelnose sturgeon does not appear to be a concern. However, pallid sturgeon are more likely to accumulate greater concentrations of OCs based on their piscivorous diet, potentially longer life-span, and higher percentage of lipids in muscle tissue (Ruelle and Keenlyne, 1994).

The results of this study clearly indicate that shovelnose sturgeon in the lower Platte River are exposed to atrazine. Atrazine concentrations in shovelnose sturgeon are apparently a function of waterborne atrazine concentrations and not diet; therefore, pallid sturgeon exposure to atrazine is probably similar to that of shovelnose sturgeon.

Atrazine concentrations in lower Platte River water may adversely affect pallid sturgeon either directly by endocrine disruption as explained above, or indirectly by decreasing their prey base. Previous studies indicate that atrazine concentrations detected in the lower Platte River can adversely effect aquatic invertebrates (Kettle et al., 1987) and cyprinids (Messadd et al., 2000). Experimental ponds containing 20 µg/L atrazine for 136 days resulted in significantly ( $p < 0.001$ ) fewer invertebrates in bluegill (*Lepomis macrochirus*) stomach contents when compared to control ponds (Kettle et al., 1987). The decreased prey base in the experimental ponds was linked to significantly ( $p < 0.01$ ) lower bluegill reproduction (Kettle et al., 1987). Red shiners (*Cyprinella lutrensis*), a



potential prey item for pallid sturgeon, can also be adversely affected by atrazine exposure, especially during the summer. Red shiners (*Cyprinella lutrensis*) collected from the lower Platte River and exposed to atrazine concentrations of 10 µg/L at 23 and 30 °C had a significantly lower Critical Thermal Maximum (CTM) compared to controls (Messadd et al., 2000). Temperatures above the CTM disrupt locomotory activity to the point where fish lose their ability to escape from conditions that promptly lead to death (Cowles and Bogert, 1944 as cited by Messadd et al., 2000).

In general, pallid sturgeon may be more susceptible than shovelnose sturgeon to the main contaminants of concern identified by this study (i.e., Se, PCBs, and atrazine). These contaminants have all been linked to potential reproductive effects in fish. The reproductive cycle of the pallid sturgeon is longer than that of the shovelnose sturgeon. Male pallid sturgeon reach sexual maturity at 5 to 7 years, and females begin egg development at 9 to 12 years and first spawn at around 15 years (Keenlyne and Jenkins, 1993). Shovelnose sturgeon reach sexual maturity at 5 to 7 years after which they typically undergo a 2 to 3 year interval between spawns (Moos, 1978).

#### Future Research Needs

High VTG and abnormal E/KT ratios in shovelnose sturgeon from the lower Platte River need to be further evaluated. These VTG concentrations were measured using the same carp antibody that was used for carp by Goodbred et al. (1997) and for shovelnose sturgeon by Coffey et al. (2003). Although VTG is thought to be well conserved across fish species by some, the use of a shovelnose sturgeon-specific VTG antibody will provide a more accurate evaluation as the cross reactivity of the carp VTG antibody with non-VTG proteins in shovelnose sturgeon blood is unknown (Kevin Kroll, Interdisciplinary Center for Biotechnology Research, pers. comm., 2004). In addition, future monitoring of hormones in blood plasma should try to account for seasonal variation.

Further research on contaminants that could contribute to pallid sturgeon reproductive failure in the lower Platte River and Missouri River is needed and should

include a screening for HACs. Disruption of the hormonal functions in fish can adversely affect sexual maturation, gamete transport, sexual behaviour, fertility, and embryo development (Arukwe, 2001). Hormonally active compounds not tested for in this study include natural and synthetic hormones, detergents and their breakdown products (i.e., alkylphenols, nonylphenols, and octylphenols), and select pesticides such as chlorpyrifos and diazinon (Arcand-Hoy and Benson, 1998; Kolpin et al., 2002). Research by the USGS indicates that many of these contaminants are present in rivers throughout the U.S. (Kolpin et al., 2002; Barber et al., 2003) including the lower Platte River (Frenzel et al., 1998; Dr. Jason Vogel, USGS Hydrologist, pers. comm., 2004). Furthermore, androgenic and estrogenic substances were detected in the Elkhorn River (Soto et al., 2004), which is a major tributary to the lower Platte River. Fathead minnows (*Pimephales promelas*) collected from the Elkhorn River immediately downstream from cattle feedlots exhibited decreased testosterone synthesis, altered head morphometrics, smaller testis size in males, and a decreased estrogen to androgen ratio in females (Orlando et al., 2004).

Many of the fish health assessment biomarkers used in this study (e.g., EROD, liver MAs, organo-somatic indices, and H4IIE dioxin equivalents) have not been previously reported for shovelnose sturgeon and their interpretation is limited. Further research is needed to establish baseline and threshold values for these parameters and improve their usefulness in evaluating sturgeon health.

It is important to further evaluate whether atrazine exposure to Platte River sturgeon is adversely affecting their reproduction, development, and survival. The results of this study, in combination with research in the laboratory by others, indicates that atrazine exposure to shovelnose sturgeon may be causing endocrine disruption as evidenced by abnormal E/T ratios, follicular atresia, and perhaps induced VTG. In addition, laboratory studies have linked atrazine exposure to adverse effects for many fish species including Atlantic salmon (*Salmo salar*) (Moore and Lower, 2001), goldfish (*Carassius auratus*) (Saglio and Trijasse, 1998; Spanó et al., 2004), red drum larvae (*Sciaenops ocellatus*) (Alvarez and Fuiman, 2005), and rainbow trout (*Oncorhynchus*

*mykiss*) (Fisher-Scheri et al., 1991). Laboratory evaluations are needed to determine the effects of atrazine exposure to adult sturgeon and their larvae.

Studies are needed to estimate the relative number of female shovelnose sturgeon that spawn each year. We found that only 30 percent (6 of 20) adult shovelnose sturgeon females were in spawning condition. Previous studies have reported higher percentages in May and June (78 percent by Moos, 1978). Sturgeon population recruitment is highly sensitive to the percentage of females that spawn annually (Pine et al., 2001).

### Recommendations

In this study, atrazine was detected in all water samples and all shovelnose sturgeon blood plasma samples. Based on this data it is reasonable to assume that the endangered pallid sturgeon is exposed to atrazine at concentrations similar to those found in this study. The current Nebraska chronic aquatic life water quality standard for atrazine is 12 µg/L. Based on the results of this study, pallid sturgeon are likely exposed to concentrations of atrazine that exceed this chronic standard and atrazine loading into the lower Platte River should be reduced. Non-point sources of atrazine can be reduced by implementing Best Management Practices (BMPs) to reduce run-off from cornfields. Such BMPs include pre-plant application and the use of alternative herbicides (Franti et al., 1997). In addition to these voluntary measures, it is recommended that the current atrazine standard of 12 µg/L be changed to a more protective level of 3 µg/L or less. Peer-reviewed scientific literature reports indicate adverse effects in fish when exposed to atrazine concentrations between 3 and 12 µg/L including altered behavior at 5 µg/L (Saglio and Trijasse; 1998), kidney damage at 5 and 10 µg/L (Fisher-Scheri et al., 1991; Oulmi et al., 1995), decreased temperature tolerance at 10 µg/L (Messaad et al., 2000), and DNA strand breaks at 7 µg/L (Chang et al., 2005). In addition, atrazine toxicity to aquatic plants, both algae and macrophytes, commonly occurs at concentrations of 10 µg/L and above (USEPA, 2003). A few studies have documented endocrine effects in fish exposed to concentrations of atrazine below 3 µg/L (Moore and Waring, 1998; Moore and Lower, 2001). Atlantic salmon (*Salmo salar*) males exposed to 0.5 µg/L

atrazine had significantly greater concentrations of plasma testosterone and a reduced reproductive priming ability of prostaglandin which resulted in significantly less expressible milt (Moore and Lower, 2001). Exposure to 0.5 µg/L simazine also resulted in these effects as well as significantly greater concentrations of 11-ketotestosterone in blood plasma. The authors concluded that simazine and atrazine toxicity is additive and that their effects are not restricted to the reproductive system, but may also affect olfactory imprinting to the natal river and subsequent homing of the adults. In the future, it may be determined that exposure to concentrations of atrazine less than 3 µg/L are harmful to pallid sturgeon. If this occurs, then more protective site specific standards may be needed or atrazine use in watersheds that drain into pallid sturgeon habitat may need to be prohibited.

Although PCBs have been banned since 1977, the results of this study indicate that fish are still exposed to these compounds at concentrations that are potentially toxic to fish and piscivorous wildlife. It is recommended that sources for PCB contamination be identified for possible remediation, especially in areas used by pallid sturgeon.

The lower Platte River segment downstream from the Elkhorn River is currently listed as impaired by selenium contamination and results of this study found concentrations of Se in sturgeon tissues that exceeded levels where reproductive impairment may begin to occur in some fish species. Selenium exposure to pallid sturgeon may be decreased by identifying and reducing anthropogenic sources in the area. This can be accomplished by screening for Se in National Pollutant Discharge and Elimination System (NPDES) permits and reducing run-off from cattle feedlots by implementing BMPs. Such BMPs include streambank fencing, relocating feedlots away from streams, constructing roofs over concentrated feeding areas, and establishing filter strips. Further studies are needed to evaluate how much irrigation drainage in the Platte River Basin is contributing to Se contamination in the lower Platte River.

Many of the adverse conditions found in shovelnose sturgeon from this research (e.g., ovicular atresia, increased macrophage aggregates, and hermaphroditic gonads), have been observed in other fish species exposed to high water temperatures (Blazer et

al., 1987; Webb et al., 1999). The higher than average water temperatures recorded in this study indicate that efforts may be needed to reduce high water temperatures in the lower Platte River during the spawning period. Magnitude and frequency of high water temperatures in the lower Platte River can be reduced by increasing river flows in the spring and by establishing temperature restrictions in NPDES permits.

### Conclusions

Shovelnose sturgeon in the lower Platte River are exposed to a mixture of elemental contaminants, organochlorines, and triazine herbicides. Although there were no anomalies detected in shovelnose sturgeon from the lower Platte River based on gross observations and condition indices, histological examination of gonads, and reproductive biomarkers indicate that potential adverse reproductive effects are occurring. Adverse reproductive conditions observed in shovelnose sturgeon include ovarian atresia and abnormal VTG concentrations and E/T ratios in blood plasma. These conditions may not be conducive to population growth and could be detrimental to pallid sturgeon recovery efforts. Factors that could be responsible for the observed adverse reproductive conditions include high water temperatures and exposure to environmental contaminants including Se, PCBs, atrazine, and other hormonally active compounds. Concentrations of elemental contaminants in shovelnose sturgeon carcasses were generally at or below background with the exception of Se and Ba. Concentrations of Se in shovelnose sturgeon were within the 4 to 6 µg/g threshold range for reproductive impairment in sensitive fish species. The effects (if any) of the elevated Ba tissue concentrations measured in this study are unknown.

The results of this study clearly indicate that shovelnose sturgeon are exposed to atrazine. Concentrations of atrazine in blood plasma were related to storm related peak flows in the lower Platte River and increased agricultural runoff. In addition, research by others (Moore and Lower; 2001; Spanó et al., 2004) suggests that the follicular atresia and abnormal E/T ratios seen in shovelnose sturgeon from this study may be due to atrazine exposure. Although the effects of atrazine exposure to shovelnose sturgeon are

unknown, research on other fish species indicate that atrazine may be adversely affecting shovelnose sturgeon health and reproduction by mechanisms that are both direct (endocrine disruption) and indirect (decreased prey base).

Many of the fish health assessment biomarkers used in this study (e.g., liver MAs, organo-somatic indices, and H4IIE dioxin equivalents) have not been previously reported for shovelnose sturgeon and their interpretation is therefore limited. However, extrapolation from studies on other fish species indicate that the high number of liver MAs and increased SSI may be in response to environmental contaminant exposure. In addition, four shovelnose sturgeon had TCDD-EQs within the possibly toxic range.

A piscivorous diet and longer life-span and reproductive cycle likely make pallid sturgeon more susceptible than shovelnose sturgeon to toxins that bioaccumulate and/or cause adverse reproductive effects. To protect pallid sturgeon in the lower Platte River, this research indicates that efforts are needed to limit exposure to Se, PCBs, atrazine, other potentially hormonally active compounds, and water quality conditions (e.g., increased temperature) that can be detrimental to pallid sturgeon reproduction.

Recommended strategies to reduce shovelnose sturgeon and pallid sturgeon exposure to environmental contaminants and adverse water quality conditions include strengthening water quality standards, implementing BMPs, further limiting pollutant discharges in NPDES permits, and increasing river flows to avoid high water temperatures during the spawning period. The unknown cumulative effects of the multiple contaminants and stressors identified by this research emphasize the need for a precautionary approach in evaluating potential adverse effects, especially to pallid sturgeon.

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APPENDIX: ADDITIONAL TABLES

Table A.1. Shovelnose sturgeon and cyprinid samples analyzed for contaminant residues through the U.S. Fish and Wildlife Service's Analytical Control Facility.

Analysis	Sample ID	Sample Mass (grams)	Percent moisture	Percent Lipid
Catalog 6050106	SP-07-C-R	100	72.7	NA
Elemental Contaminants	SP-19-C-R	100	72.8	NA
Shovelnose Carcass	SP-22-C-R	100	76.5	NA
	SP-27-C-R	100	76.7	NA
	SP-33-C-R	100	78.3	NA
	SP-37-C-R	100	73.3	NA
	SP-49-C-R	100	77.4	NA
	SP-50-C-R	100	74.1	NA
Catalog 6050103	SP-10-C-R	468	72.8	7.3
Organochlorines	SP-11-C-R	636	69.1	8.6
Shovelnose Carcass	SP-14-C-R	770	67.1	11.9
	SP-25-C-R	873	76.0	3.9
	SP-40-C-R	557	69.8	5.6
	SP-45-C-R	621	77.7	1.0
	SP-47-C-R	457	77.8	0.3
	SP-53-C-R	663	71.0	3.4
Catalog 6050105	SP-01-L-R	2	76.5	3.5
Atrazine	SP-03-L-R	6	63.3	14.9
Shovelnose Liver	SP-07-L-R	4	71.8	4.4
	SP-08-L-R	5	70.0	8.1
	SP-09-L-R	2	66.0	14.5
	SP-10-L-R	2	66.7	15.0
	SP-11-L-R	4	64.9	11.0
	SP-14-L-R	6	54.1	25.4
	SP-15-L-R	3	72.0	5.1
	SP-16-L-R	3	76.2	3.4
	SP-18-L-R	4	72.7	3.2
	SP-20-L-R	2	69.7	6.4
	SP-22-L-R	2	83.8	1.0
	SP-23-L-R	2	73.1	5.9
	SP-37-L-R	3	70.8	1.8
	SP-40-L-R	2	69.1	11.3
	SP-43-L-R	7	67.6	5.0
	SP-50-L-R	4	73.3	5.3
	SP-51-L-R	2	71.8	4.5
Catalog 6050094	SP-03-Sc-R	7	41.5	0.8
Organochlorines	SP-06-Sc-R	2	51.9	1.1
Shovelnose Digesta	SP-08-Sc-R	5	73.9	0.7
	SP-127511-Sc-R	5	91.9	0.9
	SP-13-Sc-R	5	72.0	1.0
	SP-15-Sc-R	5	65.0	1.1
	SP-17-Sc-R	5	75.5	1.0
	SP-18-Sc-R	9	71.5	1.3
	SP-21-31-Sc-R	20	92.2	1.5
	SP-32-41-Sc-R	100	53.5	0.4
	SP-42-53-Sc-R	5	74.8	0.5
	CY-01-T-R	321	75.5	NA
	CY-02-T-R	366	74.8	NA
	CY-03-T-R	366	76.2	NA
	CY-04-T-R	338	73.9	NA
	CY-05-T-R	206	74.2	NA
	CY-06-T-R	202	75.4	NA

Table A.1. Continued.

Analysis	Sample ID	Sample Mass (grams)	Percent moisture	Percent Lipid
Catalog 6050097	CY-07-T-R	364	76.3	NA
(Continued)	CY-08-T-R	347	74.1	NA
	CY-09-T-R	333	74.3	NA
	CY-10A-T-R	87	73.9	NA
	CY-10B-T-R	94	73.5	NA
Catalog 6050097	CY-01-T-R	357	80.3	6.2
Organochlorines	CY-02-T-R	402	72.1	6.0
Pallid Food Items	CY-03-T-R	405	81.5	5.9
	CY-04-T-R	375	79.1	6.5
	CY-05-T-R	234	77.8	8.4
	CY-06-T-R	223	78.5	10.8
	CY-07-T-R	396	79.1	8.0
	CY-08-T-R	381	75.7	7.2
	CY-09-T-R	379	71.4	5.8
	CY-10A	105	76.9	6.6
Catalog 6050093	50SCA	1005	NA	NA
Atrazine	6477A	968	NA	NA
water grab samples	7915A	1010	NA	NA
	HWY81A	968	NA	NA
	SCERA	1040	NA	NA
Catalog 6050114	SP-02-C-R	100	72	NA
Selenium	SP-03-C-R	100	69	NA
Shovelnose Carcass	SP-04-C-R	100	70	NA
	SP-05-C-R	100	72	NA
	SP-08-C-R	100	75	NA
	SP-13-C-R	100	73	NA
	SP-15-C-R	100	70	NA
	SP-21-C-R	100	71	NA
	SP-26-C-R	100	71	NA
	SP-28-C-R	100	76	NA
	SP-30-C-R	100	72	NA
	SP-31-C-R	100	75	NA
	SP-32-C-R	100	73	NA
	SP-35-C-R	100	73	NA
	SP-38-C-R	100	76	NA
	SP-39-C-R	100	72	NA
	SP-41-C-R	100	76	NA
	SP-43-C-R	100	72	NA
	SP-44-C-R	100	71	NA
	SP-48-C-R	100	78	NA
	SP-51-C-R	100	78	NA
	SP-52-C-R	100	77.3	NA
Catalog 6050116	SP-03-G-R	7.26	59.9	32.6
Organochlorines	SP-15-G-R	7.91	68.5	24.4
Shovelnose Ovary	SP-19-G-R	50	12.4	56.9
	SP-22-G-R	64.5	7.35	59.6
	SP-25-G-R	72.8	8.5	56.5
	SP-30-G-R	48.7	9.86	58.3
	SP-33-G-R	64.3	6.79	59.6
	SP-39-G-R	82.4	11.5	54.9
	SP-49-G-R	43.9	11.9	61.4

Table A.2. Demographics and morphometrics for shovelnose sturgeon collected from the lower Platte River in NE, 2002.

Fish ID	Site	Date	Gender	Gonad Stage	Age	Body mass (g)	Fork Length	Morphological Measurements (mm)								mCI	
								IBL				OBL					
								R	L	R	L	MIB	IL	HL	PP		NHL
SP-01	7550	020528	M	V	7	600	593	33	33	40	40	32	62	152	67	2.55	0.52
SP-02	7550	020528	M	V	7	400	510	23	24	35	34	25	39	125	60	2.69	1.11
SP-03	7550	020528	F	II	8	750	582	36	35	50	50	35	58	157	67	2.24	0.75
SP-04	7550	020528	M	V	10	500	531	36	36	51	52	32	55	144	63	2.44	0.64
SP-05	50SC	020529	F	II	6	675	583	31	32	42	40	34	63	150	71	2.41	0.44
SP-06	50SC	020529	M	II	6	600	564	31	30	41	41	29	56	144	65	2.53	0.55
SP-07	7550	020529	M	V	12	900	663	35	37	50	46	33	67	172	77	2.11	0.51
SP-08	50SC	020529	M(TO)	III(late)	11	900	605	36	36	48	49	35	55	155	74	2.18	0.94
SP-09	7550	020530	F	II	8	600	544	35	35	47	47	30	59	146	63	2.26	0.42
SP-10	7550	020530	M	III(late)	8	520	513	29	29	38	39	31	53	137	61	2.59	0.68
SP-11	7550	020530	M	III(late)	9	730	579	33	34	47	45	34	56	153	69	2.38	0.80
SP-12	7550	020530	F	II	7	490	519	33	33	43	42	30	54	144	63	2.60	0.77
SP-13	7550	020530	F	II	5	460	506	30	30	41	41	29	47	130	60	2.91	0.86
SP-14	7550	020530	M	III	13	900	602	35	36	47	50	37	56	157	70	2.38	0.91
SP-15	50SC	020530	F	II	10	730	602	33	33	44	44	33	60	160	69	2.45	0.70
SP-16	50SC	020530	F	III	9	720	592	35	35	48	48	33	64	161	70	2.21	0.46
SP-17	50SC	020530	M	II	6	450	510	31	25	42	43	30	50	133	66	2.34	0.60
SP-18	50SC	020530	M	V	10	900	643	38	40	50	na	37	66	175	81	2.77	0.76
SP-19	50SC	020530	F	III	12	1100	624	39	39	52	51	37	51	165	76	2.45	1.38
SP-20	SCER	020531	M	II	6	520	555	29	29	38	38	33	56	141	62	2.84	0.64
SP-21	50SC	020531	F	II	7	500	503	28	28	36	36	31	51	137	57	2.49	0.82
SP-22	50SC	020531	F	V	13	950	625	37	38	50	50	35	68	172	71	2.39	0.51
SP-23	SCER	020603	F	II	ND	595	535	33	34	42	40	33	56	144	66	2.63	0.77
SP-24	SCER	020603	M	III	7	610	538	29	27	39	38	37	52	134	67	2.63	0.76
SP-25	SCER	020603	F	V	ND	1050	630	39	36	50	49	40	61	167	76	2.43	0.89
SP-26	SCER	020603	M	III(early)	8	600	534	35	31	47	45	37	50	143	68	2.69	0.96
SP-27	SCER	020603	M	V	9	800	581	37	38	41	47	34	35	150	71	2.41	2.23
SP-28	SCER	020603	M	V	6	500	531	27	28	44	40	27	53	135	65	3.24	0.35
SP-29	SCER	020603	F	III	10	800	598	39	38	51	52	31	61	152	79	2.80	0.44
SP-30	SCER	020603	F	V	8	800	568	39	37	49	48	30	60	148	63	2.50	0.44
SP-31	SCER	020603	M	V	8	600	521	30	29	37	36	31	53	130	59	2.51	0.64
SP-32	6477	020604	M	V	ND	600	562	30	30	43	45	38	59	147	64	2.33	0.57
SP-33	6477	020605	F	V	ND	1000	662	31	31	45	45	36	76	117	79	2.28	-0.58
SP-34	6477	020605	M	V	ND	600	560	32	31	42	41	31	55	134	62	2.44	0.51
SP-35	6477	020605	M	III	ND	650	605	34	36	41	45	33	55	139	68	2.62	0.74
SP-36	6477	020605	M	V	ND	700	609	35	33	47	46	33	62	149	69	2.59	0.37
SP-37	6477	020606	M	V	ND	1200	677	39	37	57	58	40	76	180	75	3.33	0.22
SP-38	6477	020606	F	II	ND	800	594	37	34	48	45	40	57	141	75	2.55	0.76
SP-39	6477	020607	F	V	7	900	575	34	34	46	46	33	57	148	69	2.45	0.67
SP-40	6477	020607	M	III	8	650	561	33	33	47	48	30	56	145	65	2.92	0.53
SP-41	6477	020607	F	II	8	560	564	34	34	45	45	32	69	161	71	2.93	0.15
SP-42	15LR	020610	M	V	7	560	560	31	32	43	44	31	62	144	62	2.67	0.20
SP-43	15LR	020610	M	V	10	1050	660	41	41	54	54	36	65	164	77	2.82	0.59
SP-44	15LR	020610	M	V	7	750	553	25	25	32	34	32	55	142	66	2.62	0.65
SP-45	15LR	020610	M	V	9	850	663	32	31	40	40	31	60	144	67	2.38	0.42
SP-46	15LR	020610	M	V	9	825	648	31	31	43	43	35	65	152	70	2.51	0.34
SP-47	15LR	020610	M	V	8	700	544	31	32	39	38	31	61	146	69	2.30	0.44
SP-48	15LR	020610	M	ND	8	750	570	27	28	37	37	32	61	144	67	2.50	0.37
SP-49	15LR	020610	F	V	10	1000	634	36	38	43	43	33	59	154	66	IE	0.82
SP-50	15LR	020610	M	V	12	900	597	30	30	44	44	32	61	157	71	2.38	0.45
SP-51	15LR	020611	F	II	13	650	582	32	32	43	44	34	63	149	72	2.57	0.37
SP-52	15LR	020613	M	V	13	500	538	28	28	35	35	26	56	136	60	3.34	0.34
SP-53	15LR	020613	M	V	14	700	598	32	32	41	46	30	55	142	70	2.14	0.60

Note: Site descriptions are provided in the Results and Discussion section, Date = date captured (yymmdd), M = male, F = female, SP = Scaphirhynchus platyrhynchus, TO = teste-ova (male fish with ova in the testes), mCI = morphometric character index as determined by Sheehan et al. (1999). Gonad stage is according to Moose (1978). R = right, L = left, IBL = inner barbell lengths, OB = outer barbell lengths, MIB = Mouth-to-Inner barbell distance, IL = inter-rostrum length, HL = head length, PP = protrusion to protrusion, NHL = New Head Length.

Table A.3. Measurements of water temperature, specific conductivity, and dissolved oxygen from the lower Platte River, NE, 2002.

Measurement ID	Collection Date			Latitude	Longitude	Temperature (°C)	Disolved Oxygen (mg/L)	Specific Conductivity (µs/cm)
	Month	Day	Site					
COL 061002A	June	10	1	47.3931	97.2756	25	7.25	309.2
COL 061002B	June	10	1	41.3857	97.2445	25.1	7.78	340.5
COL 061002C	June	10	1	41.3849	97.2418	25.1	7.78	340.5
COL 061002D	June	10	1	41.3843	97.2253	25.5	8.58	341.2
COL 061002E	June	10	1	41.3858	97.2146	26.1	8.75	334.9
COL 061002F	June	10	1	41.3988	97.2806	26	6.88	305.1
COL 061002G	June	10	1	41.3993	97.2833	27.1	10.3	553
COL 061002H	June	10	1	41.3988	97.2806	27.1	10.3	553
COL 061002I	June	10	1	41.3987	97.2813	26.1	8.3	360.2
COL 061002J	June	10	1	41.3968	97.2791	26.1	8.3	360.2
COL 061002K	June	10	1	41.3953	97.2779	25.9	8.28	336.2
COL 061102A	June	10	1	41.3957	97.2758	23.6	7.73	271.4
SCH 061102A	June	11	1	41.4152	97.0463	26.2	10.27	286.1
SCH 061102B	June	11	1	41.4154	97.0286	26.8	10.86	299.1
SCH 061102C	June	11	1	41.4131	97.0267	27.1	11.2	361.8
SCH 061102D	June	11	1	41.4154	97.0219	27.7	11.25	277.9
SCH 061102E	June	11	1	41.4562	96.9281	28	10.3	295
SCH 061102F	June	11	1	41.4562	96.9281	28	10.3	295
B052102A	May	21	2	41.05344	96.32252	15.7	9.55	544
B052102B	May	21	2	41.05581	96.32530	16.7	9.81	548
B052102C	May	21	2	41.06086	96.32423	17.4	9.81	569
B052102D	May	21	2	41.05996	96.32249	17.9	10.36	612
B052102E	May	21	2	41.05842	96.32050	17.9	10.36	612
B052102F	May	21	2	41.05670	96.32016	18	10.45	578
B052102G	May	21	2	41.05415	96.31796	18.2	10.45	578
B053102A	May	31	2	41.08342	96.33809	26.3	9.35	446.7
B053102B	May	31	2	41.08342	96.33809	26.3	NA	NA
B053102C	May	31	2	41.09214	96.33942	27.2	10.35	427.9
B053102D	May	31	2	41.08956	96.34011	27.2	10.35	427.9
B060302A	June	3	2	41.12245	96.31210	24.6	7.93	404.3
B060302B	June	3	2	41.12324	96.31544	25	8.79	415.1
B060302C	June	3	2	41.07314	96.33382	26.5	7.43	490
B060302E	June	3	2	41.06924	96.33201	26.6	7.03	422.5
B060402A	June	4	2	41.06533	96.32640	21	7.34	519
B060402B	June	4	2	41.06536	96.32640	20.8	8.32	518
B060402C	June	4	2	41.07130	96.33450	20.5	8.5	458.1
B060402D	June	4	2	41.06836	96.33176	20.4	7.88	434.1
B060402E	June	4	2	41.06653	96.32836	20.4	8.6	411.4
B060402F	June	4	2	41.06132	96.32529	20.6	8.91	426.2
B060402G	June	4	2	41.05674	96.32579	21.8	8.48	456.4
B060402H	June	4	2	41.05352	96.32226	21.8	8.48	456.4
B062002A	June	20	2	41.06118	96.32482	23.4	7.84	478
B062002B	June	20	2	41.05622	96.32579	24.65	8.21	503
B062002C	June	20	2	41.07125	96.33437	24.5	8.6	454
B062002D	June	20	2	41.06865	96.33167	24.5	8.6	454
B062002E	June	20	2	41.06037	96.32276	26.09	8.88	514
B062002F	June	20	2	NA	NA	26.09	8.8	514

Table A.3 Continued.

Measurement ID	Collection Date		Site	Latitude	Longitude	Temperature (°C)	Dissolved Oxygen (mg/L)	Specific Conductivity (µs/cm)
	Month	Day						
C052302A	May	23	3	NA	NA	15.9	9.94	964
C052302B	May	23	3	41.01400	96.15406	15.8	9.45	921
C052302C	May	23	3	41.01466	96.15406	15.8	9.78	956
C052302D	May	23	3	41.01746	96.15234	15.7	10.18	613
C052302E	May	23	3	41.00998	96.16894	16.6	11.54	598
C052302F	May	23	3	41.01027	96.16770	16.6	11.93	617
C052802A	May	28	3	41.01231	96.15705	21.7	7.78	850
C052802B	May	28	3	41.01308	96.15721	21.7	7.78	850
C052802C	May	28	3	41.01443	96.15484	21.7	7.78	850
C052802D	May	28	3	41.01316	96.15179	21.7	7.78	850
C052802E	May	28	3	41.00332	96.18919	23.3	9.23	NA
C052802F	May	28	3	41.00404	96.18156	24	7.19	NA
C052802G	May	28	3	41.00694	96.17581	24	7.19	NA
C052802H	May	28	3	41.00740	96.17331	24	10.9	NA
C052902A1	May	29	3	41.00445	96.18771	22.3	7.33	459
C052902A	May	29	3	40.99595	96.20306	22.8	8.7	454.3
C052902B	May	29	3	40.99715	96.20039	23.5	9.66	554.1
C052902B1	May	29	3	41.00445	96.18771	22.3	7.33	459
C052902C	May	29	3	40.99775	96.20405	25	11.88	467.1
C052902C1	May	29	3	41.00505	96.18182	22	6.78	489.7
C052902D	May	29	3	40.99918	96.20141	25	11.88	467.1
C052902D1	May	29	3	40.99576	96.21445	25.5	14.06	448.5
C052902E1	May	29	3	41.00696	96.17602	25.5	7.24	456.5
C052902F1	May	29	3	41.00696	96.17602	25.5	7.24	456.5
C052902G1	May	29	3	40.99733	96.21135	26.7	14.56	451.2
C052902H1	May	29	3	40.99782	96.21142	26.7	14.56	451.2
C053002A	May	30	3	41.05782	96.01351	24.6	7.51	485.9
C053002A1	May	30	3	41.01915	96.26058	24.9	10.4	433.2
C053002B	May	30	3	41.01511	96.24327	29.3	12.12	416.4
C053002C	May	30	3	40.99558	96.21465	29.2	15.02	440.1
C053002C1	May	30	3	41.01781	96.25852	25.6	9.89	394.5
C053002D	May	30	3	40.91553	96.20953	29.7	15.7	439.8
C053002D1	May	30	3	41.01845	96.25540	25.6	9.89	394.5
C053002E	May	30	3	41.01392	96.24273	27.5	9.29	464.7
C053002E1	May	30	3	41.01790	96.25217	27.5	12.3	443.7
C053002F	May	30	3	41.00567	96.23480	27.2	14.55	467
C053102A	May	31	3	41.01912	96.25941	26.3	9.1	452.2
C053102B	May	31	3	41.01883	96.25713	26.8	9.06	445.7
C053102D	May	31	3	41.01725	96.24699	28	10.43	467.7
C053102E	May	31	3	41.01471	96.24309	28.7	9.25	576

Note: Data obtained from the University of Nebraska at Lincoln (UNL). Occasion ID = unique identifier used by UNL. The sample collection date is included within this ID in MMDDYY format. NA = data not available.

Table A.4. Incidence and severity of histological lesions in spleen, kidney, liver, and gonad tissues from shovelnose sturgeon collected in the lower Platte River, Nebraska, 2002.

Fish ID	Gender	Age	Spleen			Kidney			Liver		Stage	Condition
			RBC reduction	Inflammation	other	debris	present	glomeruli	fat/glycogen	leuckocytes		
SP-20	M	6	1	2	some fat	2	no	normal	2	1	II	Fat
SP-06	M	6	1	0	none	1	no	normal	2	0	II	Fat
SP-17	M	6	3	1	none	2	no	normal	2	1	II	Fat
SP-28	M	6	2	1	none	3	yes	normal	3	1	V	Spawning
SP-01	M	7	3	3	none	nd	nd	normal	1	1	V	Spawning
SP-02	M	7	1	0	none	4	yes	normal	2	2	V	Spawning
SP-24	M	7	1	0	none	3	yes	normal	2	1	III	Inactive
SP-42	M	7	2	2	none	1	no	normal	2	1	V	Spawning
SP-44	M	7	0	0	necrosis 1	nd	nd	normal	3	0	V	Spawning
SP-26	M	8	1	0	none	2	no	normal	2	1	III (early)	Inactive
SP-31	M	8	1	3	necrosis 2	2	no	normal	2	2	V	Spawning
SP-47	M	8	0	0	none	2	no	hypotrophy 1	2	1	V	Spawning
SP-40	M	8	1	1	none	1	no	normal	1	1	III	Inactive
SP-48	M	8	0	0	none	2	no	hypotrophy 1	3	1		Inactive
SP-10	M	8	3	0	none	2	yes	normal	2	1	III(late)	Inactive
SP-45-	M	9	1	0	none	2	yes	hypotrophy 1	2	1	V	Spawning
SP-46	M	9	1	0	none	2	yes	hypotrophy 1	2	1	V	Spawning
SP-11	M	9	2	1	none	3	yes	normal	1	0	III(late)	Inactive
SP-27	M	9	1	2	none	2	no	normal	2	2	V	Spawning
SP-43	M	10	0	0	none	2	yes	normal	2	1	V	Spawning
SP-04	M	10	1	0	none	3	no	normal	2	1	V	Spawning
SP-18	M	10	1	1	none	2	no	normal	2	1	V	Inactive
SP-08	TO	11	2	2	none	3	no	fatty	2	1	III(late)	Inactive
SP-07	M	12	1	0	fatty	3	yes	hypotrophy 1	2	1	V	Spawning
SP-50	M	12	1	0	none	2	yes	normal	1	1	V	Spawning
SP-52	M	13	3	2	none	3	yes	normal	3	1	V	Spawning
SP-14	M	13	2	1	none	1	no	normal	0	2	III	Inactive
SP-53	M	14	1	0	none	2	no	normal	2	1	V	Spawning
SP-32	M	NA	1	0	none	2	yes	normal	2	1	V	Spawning
SP-34	M	NA	1	0	none	1	no	normal	2	2	V	Inactive
SP-35	M	NA	1	2	none	2	no	normal	2	1	III	Inactive
SP-36	M	NA	1	0	none	2	yes	normal	3	1	V	Spawning
SP-37	M	NA	1	0	none	2	yes	normal	2	1	V	Spawning

Table A.4. Continued.

Fish ID	Gender	Age	Spleen			Kidney			Liver		Stage	Condition
			RBC reduction	Inflammation	other	debris	present	glomeruli	fat/glycogen	leukocytes		
SP-13	F	5	4	3	none	NA	NA	NA	2	0	II	Fat; could be virgin; may spawn in 2 yrs
SP-05	F	6	1	0	none	2	no	hypotrophy 1	2	1	II	Fat; could be virgin; may spawn in 2 yrs
SP-12	F	7	2	2	none	2	no	normal	1	0	II	Fat; could be virgin; may spawn in 2 yrs
SP-39	F	7	1	2	none	1	no	hypotrophy 1	3	0	V	Atresia (slight); empty follicles
SP-21	F	7	1	0	none	2	no	normal	2	1	II	Fat; could be virgin; may spawn in 2 yrs
SP-30	F	8	1	2	edema 1	2	no	normal	2	2	V	Spawning
SP-41	F	8	0	1	none	2	no	normal	2	1	II	Fat; could be virgin; may spawn in 2 yrs
SP-09	F	8	1	0	none	3	no	normal	3	1	II	Fat; could be virgin; may spawn in 2 yrs
SP-03	F	8	1	0	none	3	no	normal	1	1	II	Fat; could be virgin; may spawn in 2 yrs
SP-16	F	9	1	0	none	2	no	normal	2	1	III	Atresia (high); pigment (heavy); spawn last year; may spawn next year
SP-49	F	10	2	1	none	2	no	normal	3	0	V	Atresia (heavy); empty follicles; pigment (slight); looks like resorbing without spawning
SP-29	F	10	1	1	none	1	no	normal	2	1	III	Spawned last year or year before
SP-15	F	10	2	1	none	1	no	normal	2	2	II	Fat; pigment (slight); spawned last year or year before;
SP-19	F	12	1	2	none	2	no	normal	2	1	III	Atresia (moderate); pigment (slight); spawn last year; may spawn next year
SP-22	F	13	1	0	none	NA	NA	NA	4	1	V	Empty follicles; atresia (slight); Spawning
SP-51	F	13	1	0	none	4	no	hypertrophy 1	2	1	II	Atresia (heavy); pigment (heavy); looks like didn't spawn last year.
SP-23	F	NA	1	0	none	3	no	normal	2	1	II	No fat; empty follicles; spawned last year;
SP-25	F	NA	1	2	none	2	no	normal	2	1	V	Spawning
SP-33	F	NA	1	0	none	1	no	normal	3	2	V	Atresia (slight); empty follicles
SP-38	F	NA	1	1	none	2	no	hypotrophy 1	2	1	II	Empty follicles; atresia (slight-moderate); pigment (slight); little fat; spawned last year

Note: 1=minimal, 2=mild, 3=moderate, and 4=moderately-severe. SP = *Scaphirhynchus platyrhynchus*, M=male, F=female, NA= data not available.



Table A.5. Macrophage aggregate parameters measured in liver tissue from shovelnose sturgeon collected in the lower Platte River, Nebraska, 2002.

Fish ID	Gender	Age	Mean MA density (# per mm <sup>2</sup> ± Std)	Mean MA area (mm <sup>2</sup> ) ± Std	Mean Percent Area MA per mm <sup>2</sup> ± Std	Pigment
SP-01	M	7	25 ± 3	0.0023 ± 0.0006	3.112 ± 0.0016	none
SP-02	M	7	12 ± 1	0.0014 ± 0.0006	1.812 ± 0.011	none
SP-04	M	10	38 ± 0	0.0007 ± 0.0003	1.41 ± 0.005	BL,BR
SP-06	M	6	17 ± 7	0.0013 ± 0.0001	2.344 ± 0.008	BL,BR
SP-07	M	12	20 ± 5	0.004 ± 0.001	9.561 ± 0.032	BL,BR
SP-10	M	8	14 ± 3	0.0006 ± 0.0002	0.833 ± 0.002	none
SP-11	M	9	14 ± 6	0.0006 ± 0.0002	0.108 ± 0.133	BL,BR
SP-14	M	13	11 ± 3	0.0018 ± 0.001	2.881 ± 0.015	BL,BR
SP-17	M	6	15 ± 0	0.0012 ± 0	1.873 ± 0	BL,BR
SP-18	M	10	26 ± 1	0.0025 ± 0.0012	5.602 ± 0.029	BL,BR
SP-20	M	6	27 ± 6	0.0006 ± 0.0003	1.79 ± 0.012	none
SP-24	M	7	13 ± 4	0.0021 ± 0.0009	2.563 ± 0.01	none
SP-26	M	8	18 ± 2	0.0011 ± 0.0002	1.949 ± 0.002	none
SP-27	M	9	26 ± 0	0.0092 ± 0	28.081 ± 0	BL,BR
SP-28	M	6	16 ± 3	0.002 ± 0.003	5.104 ± 0.017	none
SP-31	M	8	14 ± 2	0.0014 ± 0.0003	2.225 ± 0.002	BL,BR
SP-32	M	NA	24 ± 12	0.0033 ± 0.0007	9.586 ± 0.056	none
SP-34	M	NA	19 ± 0	0.0046 ± 0	5.758 ± 0.039	none
SP-35	M	NA	73 ± 15	0.0012 ± 0.0002	4.167 ± 0.001	none
SP-36	M	NA	27 ± 5	0.0022 ± 0.0016	5.758 ± 0.039	none
SP-37	M	NA	35 ± 4	0.0029 ± 0.0007	9.963 ± 0.017	BL,BR
SP-40	M	8	17 ± 4	0.0016 ± 0.0005	2.607 ± 0.008	none
SP-42	M	7	27 ± 10	0.0018 ± 0.0011	5.612 ± 0.052	BL,BR
SP-43	M	10	25 ± 5	0.0022 ± 0.0003	5.495 ± 0.004	none
SP-44	M	7	23 ± 5	0.0061 ± 0.0031	13.599 ± 0.042	none
SP-45	M	9	46 ± 7	0.0083 ± 0.008	19.662 ± 0.006	none
SP-46	M	9	26 ± 4	0.0033 ± 0.0011	8.918 ± 0.028	none
SP-47	M	8	35 ± 2	0.0046 ± 0.0001	17.074 ± 0.006	none
SP-48	M	8	47 ± 7	0.005 ± 0.0009	23.813 ± 0.007	none
SP-50	M	12	26 ± 3	0.0011 ± 0.0006	4.535 ± 0.022	BL,BR
SP-52	M	13	38 ± 1	0.0011 ± 0.0002	7.617 ± 0.009	none
SP-53	M	14	42 ± 4	0.0038 ± 0.0015	10.536 ± 0.021	BL,BR
SP-08	M(TO)	11	39 ± 6	0.0024 ± 0.0022	4.237 ± 0.006	BL,BR

Table A.5. Continued.

Fish ID	Gender	Age	Mean MA density (# per mm <sup>2</sup> ± Std)	Mean MA area (mm <sup>2</sup> ) ± Std	Mean Percent Area MA per mm <sup>2</sup> ± Std	Pigment
SP-03	F	8	17 ± 1	0.0013 ± 0.0006	2.37 ± 0.013	BL,BR
SP-05	F	6	19 ± 2	0.001 ± 0.0005	1.738 ± 0.007	none
SP-09	F	8	29 ± 13	0.0006 ± 0.0001	1.652 ± 0.005	none
SP-12	F	7	13 ± 5	0.0008 ± 0.0004	0.905 ± 0.004	none
SP-13	F	5	9 ± 0	0.0003 ± 0	0.312 ± 0	none
SP-15	F	10	42 ± 5	0.0008 ± 0.0002	1.6394 ± 0.0006	none
SP-16	F	9	35 ± 4	0.0018 ± 0.0002	6.496 ± 0.002	none
SP-19	F	12	39 ± 7	0.002 ± 0.0003	8.562 ± 0.016	BL,BR
SP-21	F	7	23 ± 8	0.0007 ± 0.0003	1.007 ± 0.007	none
SP-22	F	13	27 ± 3	0.0015 ± 0.0006	4.517 ± 0.001	none
SP-23	F	NA	22 ± 5	0.0013 ± 0.0003	2.663 ± 0.001	BL,BR
SP-25	F	NA	22 ± 7	0.003 ± 0.0009	9.002 ± 0.043	none
SP-29	F	10	24 ± 4	0.0022 ± 0.0012	5.912 ± 0.032	none
SP-30	F	8	13 ± 0	0.002 ± 0	2.627 ± 0	BL,BR
SP-33	F	NA	15 ± 0	0.0002 ± 0	0.377 ± 0	none
SP-38	F	NA	21 ± 4	0.0026 ± 0.0007	5.472 ± 0.014	BL,BR
SP-39	F	7	33 ± 8	0.0014 ± 0.0002	4.713 ± 0.011	none
SP-41	F	8	12 ± 1	0.0016 ± 0.0004	1.872 ± 0.004	none
SP-49	F	10	123 ± 18	0.0037 ± 0.0008	21.424 ± 0.005	none
SP-51	F	13	29 ± 4	0.0021 ± 0.0003	7.111 ± 0.011	BL,BR

Note: SP = *Scaphirhynchus platyrhynchus*, NA= data not available, M= male, F = female, TO = testis-ova, MA = macrophage aggregates, Std = standard deviation, BL = blue, BR = brown. Age = estimated age in years. Mean values are for 3 subsamples of tissue.

Table A.6. Concentrations of 17-beta estradiol (E2), 11-ketotestosterone (11KT), vitellogenin (VTG), and atrazine in blood plasma from shovelnose sturgeons collected from the lower Platter River, 2002.

Fish ID	Gender	Plasma Hormones and Atrazine			
		plasma E2 (pg/ml)	plasma 11KT (pg/ml)	plasma VTG (mg/ml)	plasma atrazine (ppb)
SP-01	M	567	1039	0.132	28.173
SP-02	M	303	1816	0.075	3.293
SP-03	F	713	458	1.108	9.358
SP-04	M	501	241	2.152	15.2
SP-05	F	530	949	0.024	21.472
SP-06	M	667	548	1.396	10.067
SP-07	M	706	1450	0.163	8.551
SP-08	M(TO)	606	1363	0.092	8.025
SP-09	F	457	205	0.562	> 30
SP-10	M	365	1254	0.025	6.935
SP-11	M	511	1472	0.132	14.325
SP-12	F	NA	NA	NA	NA
SP-13	F	707	311	0.4	22.63
SP-14	M	691	305	0.136	25.165
SP-15	F	1034	788	0.723	14.032
SP-16	F	792	295	0.074	11.854
SP-17	M	489	105	0.234	2.962
SP-18	M	850	520	0.562	1.288
SP-19	F	412	129	0.004	2.047
SP-20	M	603	883	0.007	4.285
SP-21	F	664	1327	0.149	2.171
SP-22	F	690	105	0.054	3.801
SP-23	F	759	1043	0.042	2.533
SP-24	M	867	1405	0.142	1.568
SP-25	F	688	835	0.846	0.99
SP-26	M	570	778	0.703	1.408
SP-27	M	412	601	0.541	0.518
SP-28	M	665	446	0.572	1.299
SP-29	F	742	107	1.91	0.842
SP-30	F	500	979	0.077	3.081
SP-31	M	829	1146	0.074	2.35
SP-32	M	653	1218	0.075	2.506
SP-33	F	1750	206	1.748	4.17
SP-34	M	589	898	0.43	5.112
SP-35	M	719	661	0.056	10.085
SP-36	M	637	350	0.167	7.55
SP-37	M	579	656	0.168	5.28
SP-38	F	599	635	0.141	6.89
SP-39	F	441	515	0.154	3.143
SP-40	M	571	122	0.128	3.709
SP-41	F	564	761	0.168	6.5
SP-42	M	729	1040	0.065	1.784
SP-43	M	374	246	0.094	3.429
SP-44	M	874	254	0.934	NA
SP-45	M	559	1116	0.137	1.644
SP-46	M	723	732	0.065	0.975
SP-47	M	643	542	0.303	1.461
SP-48	M	540	629	0.303	1.34
SP-49	F	818	101	1.969	0.241
SP-50	M	690	246	1.623	2.327
SP-51	F	399	478	0.095	18.896
SP-52	M	712	265	0.848	NA
SP-53	M	507	181	1.377	20.304

Note: M = male, F = female, TO = testis-ova (intersex); SP = *Scaphirhynchus platyrhynchus*, NA= data not available.

Table A.7. Ethoxyresorufin-*O*-deethylase (EROD) activity in liver tissue from *Scaphirhynchus platyrhynchus* (SP) collected in the lower Platter River, 2002.

Fish ID	Site	Gender	Age	Mean EROD Rate (pmol/min*mg) ± SE	CV %	LOD	LOQ
SP-01	7550	Male	7	<LOQ	0.1	58.6	0.4 1.4
SP-02	7550	Male	7	1.7	0	3.7	0.4 1.4
SP-03	7550	Female	8	<LOQ	0.1	21.5	0.4 1.4
SP-04	7550	Male	10	1.9	0.1	7	0.4 1.4
SP-05	50SC	Female	6	2.2	0.1	4.5	0.4 1.4
SP-06	50SC	Male	6	<LOQ	0	4.3	0.4 1.4
SP-07	7550	Male	12	1.6	0.1	9.1	0.4 1.4
SP-08	50SC	Male	11	<LOQ	0.1	16.9	0.4 1.4
SP-09	7550	Female	8	<LOQ	0.1	9	0.4 1.4
SP-10	7550	Male	8	3.2	0	2.6	0.4 1.4
SP-11	7550	Male	9	3.2	0.2	10.7	0.4 1.4
SP-12	7550	Female	7	<LOQ	0.1	14.7	0.7 1.9
SP-13	7550	Female	5	<LOD	NA	NA	0.7 1.9
SP-14	7550	Male	13	3.2	0.2	12.1	0.7 1.9
SP-15	50SC	Female	10	<LOQ	0.1	5.3	0.7 1.9
SP-16	50SC	Female	9	<LOQ	0.1	20.1	0.7 1.9
SP-17	50SC	Male	6	<LOQ	0.3	90.1	0.7 1.9
SP-18	50SC	Male	10	2.4	0.1	7	0.7 1.9
SP-19	50SC	Female	12	<LOD	0	21.3	0.7 1.9
SP-20	SCER	Male	6	2.3	0.1	5.5	0.7 1.9
SP-21	50SC	Female	7	2.8	0.2	26.8	0.7 1.9
SP-22	50SC	Female	13	<LOQ	0	6.8	0.7 1.9
SP-23	SCER	Female	ND	<LOD	0.1	32	0.7 1.9
SP-24	SCER	Male	7	<LOD	0.1	78.6	0.7 1.9
SP-25	SCER	Female	ND	<LOD	0.1	85.1	0.7 1.9
SP-26	SCER	Male	8	<LOD	0	0.9	0.7 1.9
SP-27	SCER	Male	9	<LOD	0.1	44.4	0.7 1.9
SP-28	SCER	Male	6	<LOD	0	52.1	0.7 1.9
SP-29	SCER	Female	10	<LOD	0	19.9	0.6 0.9
SP-30	SCER	Female	8	<LOD	0	30.1	0.7 1.9
SP-31	SCER	Male	8	<LOD	0	0.3	0.4 1.8
SP-32	6477	Male	ND	<LOD	0.1	28.9	0.4 1.8
SP-33	6477	Female	ND	<LOD	0.1	NA	0.4 1.8
SP-34	6477	Male	ND	<LOD	0.1	24.1	0.4 1.8
SP-35	6477	Male	ND	<LOD	0	18.2	0.4 1.8
SP-36	6477	Male	ND	<LOD	0.1	181.7	0.4 1.8
SP-37	6477	Male	ND	<LOQ	0.1	23.4	0.4 1.8
SP-38	6477	Female	ND	<LOD	NA	NA	0.4 1.8
SP-39	6477	Female	7	<LOQ	0.1	16.8	0.4 1.8
SP-40	6477	Male	8	4.5	0.2	6.5	0.4 1.8
SP-41	6477	Female	8	1.9	0.1	11.8	0.4 1.8
SP-42	15LR	Male	7	<LOQ	0	1.2	0.4 1.8
SP-43	15LR	Male	10	2.8	0.4	24.8	0.4 1.8
SP-44	15LR	Male	7	2.1	0.1	10.8	0.4 1.8
SP-45	15LR	Male	9	4	0.5	21.3	0.4 1.8
SP-46	15LR	Male	9	2.6	0.1	9.6	0.4 1.8
SP-47	15LR	Male	8	3.6	0.1	9.1	0.4 1.8
SP-48	15LR	Male	8	<LOQ	0.1	25.7	0.4 1.8
SP-49	15LR	Female	10	<LOD	0.1	24.2	0.4 1.8
SP-50	15LR	Male	12	<LOQ	0.2	76.4	0.6 0.9
SP-51	15LR	Female	13	<LOD	0.5	277	0.6 0.9
SP-52	15LR	Male	13	2.5	0.3	40.2	0.6 0.9
SP-53	15LR	Male	14	9.1	0.4	8.4	0.6 0.9

Note: < = less than, LOQ = level of quantification, LOD = level of detection, CV% = coefficient of variation, SE = standard error, ND = not done, NA = not applicable (i.e., Induction in 1 well only).

Table A.8. Concentrations of selenium in carcass samples of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF Catalog ID	Sample ID	Concentration mg/kg	
		Dry Weight	Wet Weight
6050114	SP-02	4.2	1.2
6050114	SP-03	3.2	1.0
6050114	SP-04	3.6	1.1
6050114	SP-05	4.1	1.2
6050114	SP-08	5.0	1.3
6050114	SP-13	3.8	1.0
6050114	SP-15	4.1	1.2
6050114	SP-21	4.3	1.2
6050114	SP-26	3.8	1.1
6050114	SP-28	4.5	1.1
6050114	SP-30	4.5	1.3
6050114	SP-31	4.7	1.2
6050114	SP-32	4.5	1.2
6050114	SP-35	3.6	1.0
6050114	SP-38	7.8	1.8
6050114	SP-39	4.6	1.3
6050114	SP-41	4.0	1.0
6050114	SP-43	5.6	1.6
6050114	SP-44	4.3	1.2
6050114	SP-48	4.4	1.0
6050114	SP-51	7.6	1.7
6050114	SP-52	6.2	1.4
6050106	SP-07	3.6	1.0
6050106	SP-19	4.2	1.1
6050106	SP-22	5.2	1.2
6050106	SP-27	4.4	1.0
6050106	SP-33	5.2	1.1
6050106	SP-37	5.8	1.5
6050106	SP-49	9.3	2.1
6050106	SP-50	4.0	1.0

Note: SP = *Scaphirhynchus platyrhynchus*, PACF = Patuxent Analytical Control Facility, Laurel MD.

Table A.9. Concentrations of elemental contaminants in carcass samples of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF Catalog ID	Fish ID	Trace Element Concentration in mg/kg dry weight (D.W.) and wet weight (W.W.)											
		Al		As		B		Ba		Be		Cd	
		D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.
6050106	SP-07	20	5.5	< 0.20	< 0.05	< 2.00	< .500	48.6	13.3	< 0.10	< 0.03	0.34	0.093
6050106	SP-19	50	14	< 0.20	< 0.05	< 2.00	< .500	41.1	11.2	< 0.10	< 0.03	0.31	0.08
6050106	SP-22	57	13	0.2	0.05	< 2.00	< .500	39.8	9.37	< 0.10	< 0.02	0.49	0.12
6050106	SP-27	30	6.9	< 0.20	< 0.05	< 2.00	< .500	93.6	21.7	< 0.10	< 0.02	0.35	0.081
6050106	SP-33	58	13	0.2	0.05	< 2.00	< .400	50.6	11	< 0.10	< 0.02	0.59	0.13
6050106	SP-37	17	4.6	< 0.20	< 0.05	< 2.00	< .500	36.3	9.7	< 0.10	< 0.03	0.39	0.1
6050106	SP-49	69	16	0.3	0.07	4	0.9	29.5	6.66	< 0.10	< 0.02	0.2	0.04
6050106	SP-50	9.9	2.6	< 0.20	< 0.05	< 2.00	< .500	28.1	7.3	< 0.10	< 0.03	0.3	0.07
		Cr		Cu		Fe		Hg		Mg		Mn	
		D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.
6050106	SP-07	2	0.56	1.9	0.53	87	24	0.32	0.09	1610	440	13	3.5
6050106	SP-19	3.3	0.89	1.6	0.43	81	22	0.2	0.07	1480	403	14	3.7
6050106	SP-22	1	0.31	2.1	0.49	79	19	0.2	0.05	1830	430	19	4.5
6050106	SP-27	1	0.2	1.7	0.39	67	15	0.51	0.12	1910	443	20	4.6
6050106	SP-33	1	0.3	1.8	0.39	86	19	0.59	0.13	1910	414	24	5.3
6050106	SP-37	1.6	0.43	1.3	0.35	51	14	0.52	0.14	1660	443	14	3.8
6050106	SP-49	2.6	0.58	2.3	0.51	95	21	0.2	0.05	2000	451	15	3.4
6050106	SP-50	0.6	0.2	1.3	0.33	44	11	0.2	0.05	1520	394	11	2.9
		Mo		Ni		Pb		Sr		V		Zn	
		D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.	D.W.	W.W.
6050106	SP-07	< 2.00	< 0.50	0.7	0.2	0.5	0.1	78.5	21.4	< 0.50	< 0.10	110	30
6050106	SP-19	< 2.00	< 0.50	1	0.39	0.3	0.08	81.5	22.2	< 0.50	< 0.10	102	27.9
6050106	SP-22	< 2.00	< 0.50	0.5	0.1	0.3	0.07	87.6	20.6	< 0.50	< 0.10	115	27.1
6050106	SP-27	< 2.00	< 0.50	0.6	0.1	0.4	0.1	101	23.4	< 0.50	< 0.10	141	32.7
6050106	SP-33	< 2.00	< 0.50	0.7	0.2	0.4	0.08	100	21.7	0.50	0.10	132	28.5
6050106	SP-37	< 2.00	< 0.50	0.9	0.2	0.3	0.08	64.5	17.2	< 0.50	< 0.10	109	29.1
6050106	SP-49	< 2.00	< 0.50	0.8	0.2	0.3	0.07	109	24.5	0.50	0.10	122	27.5
6050106	SP-50	< 2.00	< 0.50	<0.50	< 0.10	< 0.20	< 0.05	62.5	16.2	< 0.50	< 0.10	97.1	25.2

Note: SP = *Scaphirhynchus platyrhynchus*, < = less than the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD.

Table A.10. Concentrations of elemental contaminants in digesta (stomach contents) of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF		Trace Element Concentration in mg/kg dry weight													
Catalog ID	Fish ID(s)	Al	As	B	Ba	Be	Cd	Co	Cr	Cu	Fe	Hg	K	Mg	
6050094	SP-04	662	1.79	< 0.97	34.9	< .0484	0.0619	1.27	0.704	1.37	1700	< 0.0194	337	206	
6050094	SP-09	744	1.51	< 0.93	24.5	0.09	0.0518	1.08	0.62	1.18	2020	<0.0186	332	168	
6050094	SP-12	1550	2.35	1.58	354	0.088	1.46	4.73	1.74	9.88	2800	<0.0205	1430	476	
6050094	SP-14	2890	2.47	2.94	364	0.113	3.1	4.4	3.17	19	3750	< 0.0397	3460	1000	
6050094	SP-16	1310	2.11	1.2	111	0.057	1.34	2.3	1.61	10.6	2220	0.0216	1410	483	
6050094	SP-19	1020	1.05	< 0.98	31.4	< 0.05	1.02	0.927	1	6.19	1430	< 0.0197	802	302	
6050094	SP-20	611	0.82	1.06	63.2	< 0.05	4.28	1.01	0.719	16.8	969	0.0357	3620	634	
6050094	*SP-21-31	981	1.28	1.06	67.1	0.059	3.12	1.56	1.28	13.2	1450	0.0263	3030	622	
6050094	*SP-32-41	1250	0.803	1.59	28	0.058	0.97	0.77	1.49	6.06	1450	< 0.0202	1250	413	
6050094	*SP-42-53	1550	1.28	< 1.03	37.2	0.079	0.846	1.63	1.34	14.9	2080	0.0263	1910	663	
6050094	*SP-1,2,7,5,11	3750	2.19	< 1.00	30.4	0.098	1.08	2.52	1.06	7.63	7390	< 0.0201	1240	1480	
		Mn	Mo	Na	Ni	P	Pb	S	Se	Si	Sr	Ti	V	Zn	
6050094	SP-04	102	< 0.97	392	2.02	890	3.17	654	0.629	1430	4.3	18.6	2.65	29.4	
6050094	SP-09	45.9	< 0.93	418	0.831	325	2.16	298	0.185	1300	3.03	27.8	1.81	8.97	
6050094	SP-12	105	< 1.02	906	2.99	2920	2.94	2170	1.14	2450	12.6	47.9	4.83	76.9	
6050094	SP-14	140	< 1.99	4270	3.79	5840	3.05	4370	6.25	4130	31.8	64.4	6.14	216	
6050094	SP-16	80.3	< 1.00	878	3.93	2840	2.41	2160	2.29	2350	10.3	36.6	4.41	79.7	
6050094	SP-19	42.5	< 0.98	695	1.87	1930	2.04	1180	0.973	1770	6.02	37.5	2.47	64.5	
6050094	SP-20	39.4	< 0.98	2400	0.723	6250	1.05	4700	3.45	1300	12.9	16.7	1.46	198	
6050094	*SP-21-31	54.9	< 1.03	2360	1.23	5770	1.91	3850	3.36	1930	14.5	27.3	2.38	135	
6050094	*SP-32-41	29.2	1.15	1080	0.9	2330	1.54	1670	1.67	2040	12.2	32.8	2.71	68.3	
6050094	*SP-42-53	82	< 1.03	3840	1.61	4630	2.13	3910	6.02	2440	27.8	35.9	3.41	73.6	
6050094	*SP-1,2,7,5,11	67.4	< 1.00	1230	2.83	2180	2.6	1800	1.47	1540	6.93	22.5	16	60	

Note: \* = composite sample, SP = *Scaphirhynchus platyrhynchus*, < = less than the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD.

Table A.11. Concentrations of elemental contaminants in potential pallid sturgeon food items (cyprinids less than 5 inches in length) collected from the Platte River, Nebraska, 2002.

PACF		Trace Element Concentration in mg/kg dry weight													
Catalog ID	Sample ID	Ag	Al	As	B	Ba	Be	Cd	Co	Cr	Cu	Fe	Hg	K	Li
6050097	CY-01	< 1.00	193	1.87	<0.99	22.7	< 0.05	0.07	< 0.50	< 0.50	2.82	205	0.16	12600	0.324
6050097	CY-02	< 1.02	110	1.76	< 1.02	21.9	< 0.05	0.07	< 0.51	< 0.51	2.98	156	0.158	12300	0.309
6050097	CY-03	< 1.03	121	1.85	< 1.03	23.7	< 0.05	0.07	< 0.52	< 0.52	3.27	151	0.154	12100	0.304
6050097	CY-04	< 1.01	262	1.99	< 1.01	25.3	< 0.05	0.08	< 0.50	< 0.50	2.8	342	0.125	12000	0.353
6050097	CY-05	< 1.01	185	1.99	< 1.01	27.8	< 0.05	0.08	< 0.51	< 0.51	2.97	243	0.109	11300	0.337
6050097	CY-06	< 1.04	346	2.48	< 1.04	36.2	< 0.05	0.08	< 0.52	< 0.52	3.2	410	0.119	11800	0.408
6050097	CY-07	< 1.08	675	2.09	< 1.08	41.0	< 0.05	0.08	0.59	0.79	3.18	735	0.109	11900	0.736
6050097	CY-08	< 0.98	422	1.92	< 0.98	27.4	< 0.05	0.08	< 0.49	< 0.49	3.15	457	0.113	11700	0.471
6050097	CY-09	< 1.00	54	1.63	< 1.00	23.6	< 0.05	0.07	< 0.50	< 0.50	2.61	88.3	0.157	12300	0.259
6050097	CY-10A	< 1.04	84.8	1.87	< 1.04	25.9	< 0.05	0.06	< 0.52	< 0.52	2.44	111	0.161	11500	0.265
6050097	CY-10B	< 1.01	112	1.88	< 1.01	24.4	< 0.05	0.05	< 0.50	< 0.50	2.52	134	0.143	11500	0.285
		Mg	Mn	Mo	Na	Ni	P	Pb	S	Se	Si	Sr	Ti	V	Zn
6050097	CY-01	1810	25.8	< 0.99	3910	< 0.50	25000	0.194	8870	3.44	378	70.5	5.54	< 1.00	223
6050097	CY-02	1610	23.7	< 1.02	3580	< 0.51	25700	0.165	8430	3.27	264	74.7	4.1	< 1.02	219
6050097	CY-03	1380	22.0	< 1.03	3400	< 0.52	24400	0.157	8120	3.15	291	71.5	4.04	< 1.03	210
6050097	CY-04	1690	43.8	< 1.01	3250	< 0.50	23200	0.314	8080	4.76	503	92.7	7.83	< 1.01	182
6050097	CY-05	1540	39.5	< 1.01	3090	< 0.51	23400	0.215	7580	5.51	398	92.3	6.1	< 1.01	199
6050097	CY-06	1570	48.7	< 1.04	3400	0.62	26300	0.393	8020	5.23	643	112	10.1	1.07	203
6050097	CY-07	1960	79.3	< 1.08	3120	0.88	23600	0.659	7600	5.49	1060	89.6	17.7	1.47	206
6050097	CY-08	1600	49.0	< 0.98	3190	< 0.49	23600	0.473	7960	5.3	706	93.4	11.5	< 0.98	205
6050097	CY-09	1220	23.5	< 1.00	3430	< 0.50	22800	0.109	8380	4.7	144	67.7	2.1	< 1.00	218
6050097	CY-10A	1570	33.3	< 1.04	3360	< 0.52	25800	0.143	7530	4.67	171	89.5	2.77	< 1.04	206
6050097	CY-10B	1360	31.8	< 1.01	3220	< 0.50	24700	0.143	7690	3.94	252	77.5	3.39	< 1.01	210

Note: CY = cyprinid, < indicates sample was below the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD.



Table A.12. Concentrations of organochlorine residues in carcasses of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF Catalog No.	Fish ID	Organochlorine Concentration mg/kg dry weight (DW) and wet weight (WW)									
		PCB-TOTAL		Hexachlorobenzene		Mirex		Toxaphene		Heptachlor Epoxide	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050103	SP-10	1.69	0.46	< 0.00735	< 0.002	< 0.00735	< 0.002	< 0.184	< 0.05	< 0.00735	< .00200
6050103	SP-11	0.712	0.22	< 0.00647	< 0.002	< 0.00647	< 0.002	< 0.162	< 0.05	< 0.00647	< .00200
6050103	SP-14	3.34	1.1	< 0.00608	< 0.002	< 0.00608	< 0.002	< 0.152	< 0.05	0.0395	0.013
6050103	SP-25	<0.0417	< .0100	< 0.00833	< 0.002	< 0.00833	< 0.002	< 0.208	< 0.05	< 0.00833	< .00200
6050103	SP-40	1.26	0.38	< 0.00662	< 0.002	< 0.00662	< 0.002	< 0.166	< 0.05	< 0.00662	< .00200
6050103	SP-45	0.583	0.13	< 0.00897	< 0.002	< 0.00897	< 0.002	< 0.224	< 0.05	< 0.00897	< .00200
6050103	SP-47	<0.045	< .0100	< 0.00901	< 0.002	< 0.00901	< 0.002	< 0.225	< 0.05	< 0.00901	< .00200
6050103	SP-53	0.966	0.28	< 0.0069	< 0.002	< 0.0069	< 0.002	< 0.172	< 0.05	< 0.0069	< .00200
		Alpha Chlordane		Gama Chlordane		Oxychlordane		Cis-nonachlor		Trans-nonachlor	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050103	SP-10	0.0809	0.022	0.0331	0.009	< 0.00735	< .00200	< 0.00735	< 0.002	0.129	0.035
6050103	SP-11	<0.00647	< .00200	0.0129	0.004	< 0.00647	< .00200	< 0.00647	< .00200	0.055	0.017
6050103	SP-14	0.173	0.057	0.112	0.037	0.0334	0.011	0.0608	0.02	0.207	0.068
6050103	SP-25	0.025	0.006	0.0333	0.008	< 0.00833	< .00200	< 0.00833	< .00200	0.112	0.027
6050103	SP-40	<0.00662	< .00200	0.0166	0.005	< 0.00662	< .00200	< 0.00662	< .00200	0.0894	0.027
6050103	SP-45	< 0.00897	< .00200	< 0.00897	< .00200	< 0.00897	< .00200	< 0.00897	< .00200	0.0135	0.003
6050103	SP-47	< 0.00901	< .00200	< 0.00901	< .00200	< 0.00901	< .00200	< 0.00901	< .00200	< 0.00901	< .00200
6050103	SP-53	< 0.0069	< .00200	< 0.0069	< .00200	0.031	0.009	< 0.0069	< .00200	0.0414	0.012
		Dieldrin		Endrin		p,p'-DDD		p,p'-DDE		p,p'-DDT	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050103	SP-10	0.00735	0.002	< 0.00735	< 0.002	0.125	0.034	0.32	0.087	< 0.00735	< 0.002
6050103	SP-11	0.0583	0.018	< 0.00647	< 0.002	0.0615	0.019	0.136	0.042	< 0.00647	< 0.002
6050103	SP-14	0.185	0.061	< 0.00608	< 0.002	0.161	0.053	0.334	0.11	< 0.00608	< 0.002
6050103	SP-25	0.0875	0.021	< 0.00833	< 0.002	0.0542	0.013	0.267	0.064	< 0.00833	< 0.002
6050103	SP-40	< 0.00662	< .00200	< 0.00662	< 0.002	0.0629	0.019	0.291	0.088	< 0.00662	< 0.002
6050103	SP-45	< 0.00897	< .00200	< 0.00897	< 0.002	< 0.00897	< .00200	0.117	0.026	< 0.00897	< 0.002
6050103	SP-47	< 0.00901	< .00200	< 0.00901	< 0.002	< 0.00901	< .00200	< 0.00901	< .00200	< 0.00901	< 0.002
6050103	SP-53	0.0241	0.007	< 0.0069	< 0.002	< 0.0069	< .00200	0.148	0.043	< 0.0069	< 0.002

Note: SP = *Scaphirhynchus platyrhynchus*, < indicates sample was below the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD. Lindane and o,p' isomers of DDT, DDD, and DDE (data not shown) were all below the detection limit of 0.002 mg/kg wet weight.

Table A.13. Concentrations of organochlorine residues in ovary of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF		Organochlorine Concentration mg/kg dry weight (DW) and wet weight (WW)									
Catalog No.	Fish ID	PCB-TOTAL		PCB-1254		PCB-1260		Hexachlorobenzene		Heptachlor Epoxide	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050116	SP-03-G-R	0.935	0.630	0.757	0.510	0.178	0.120	0.012	0.012	0.052	0.035
6050116	SP-15-G-R	2.840	2.150	1.440	1.090	1.400	1.060	0.008	0.008	0.060	0.045
6050116	SP-19-G-R	0.418	0.180	0.255	0.110	0.162	0.070	< 0.005	< 0.002	0.009	0.004
6050116	SP-22-G-R	0.126	0.051	0.126	0.051	< 0.025	< 0.01	< 0.005	< 0.002	0.007	0.003
6050116	SP-25-G-R	< 0.023	< 0.01	< 0.023	< 0.01	< 0.023	< 0.01	< 0.005	< 0.002	0.014	0.006
6050116	SP-30-G-R	0.456	0.190	0.336	0.140	0.129	0.054	0.005	0.002	0.022	0.009
6050116	SP-33-G-R	0.228	0.092	0.186	0.075	0.042	0.017	< 0.005	< 0.002	0.012	0.005
6050116	SP-39-G-R	0.157	0.071	0.157	0.071	< 0.022	< 0.01	< 0.004	< 0.002	0.022	0.010
6050116	SP-49-G-R	0.725	0.280	0.415	0.160	0.311	0.120	< 0.005	< 0.002	0.016	0.006
		Alpha Chlordane		Gama Chlordane		Oxychlordane		Cis-nonachlor		Trans-nonachlor	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050116	SP-03-G-R	0.141	0.095	0.080	0.054	0.055	0.037	0.107	0.072	0.208	0.140
6050116	SP-15-G-R	0.146	0.110	0.081	0.061	0.036	0.027	0.128	0.097	0.291	0.220
6050116	SP-19-G-R	0.053	0.023	0.016	0.007	0.021	0.009	0.044	0.019	0.104	0.045
6050116	SP-22-G-R	0.030	0.012	0.012	0.005	0.010	0.004	< 0.005	< 0.002	0.042	0.017
6050116	SP-25-G-R	0.041	0.018	0.023	0.010	0.016	0.007	< 0.005	< 0.002	0.067	0.029
6050116	SP-30-G-R	0.103	0.043	0.058	0.024	0.036	0.015	< 0.005	< 0.002	0.113	0.047
6050116	SP-33-G-R	0.030	0.012	0.020	0.008	0.015	0.006	0.040	0.016	0.057	0.023
6050116	SP-39-G-R	0.042	0.019	0.013	0.006	0.024	0.011	< 0.004	< 0.002	0.040	0.018
6050116	SP-49-G-R	0.034	0.013	0.010	0.004	0.016	0.006	< 0.005	< 0.002	0.075	0.029
		Dieldrin		Gama BHC		p,p'-DDD		p,p'-DDE		p,p'-DDT	
		DW	WW	DW	WW	DW	WW	DW	WW	DW	WW
6050116	SP-03-G-R	0.138	0.093	0.009	0.006	0.356	0.240	1.340	0.900	0.105	0.071
6050116	SP-15-G-R	0.146	0.110	0.004	0.003	0.238	0.180	1.080	0.820	0.081	0.061
6050116	SP-19-G-R	0.044	0.019	< 0.005	< 0.002	0.100	0.043	0.209	0.090	< 0.005	< 0.002
6050116	SP-22-G-R	0.025	0.010	< 0.005	< 0.002	0.055	0.022	0.126	0.051	< 0.005	< 0.002
6050116	SP-25-G-R	0.115	0.050	< 0.005	< 0.002	0.062	0.027	0.161	0.070	0.016	0.007
6050116	SP-30-G-R	0.072	0.030	< 0.005	< 0.002	0.118	0.049	0.288	0.120	< 0.005	< 0.002
6050116	SP-33-G-R	0.020	0.008	< 0.005	< 0.002	0.064	0.026	0.176	0.071	< 0.005	< 0.002
6050116	SP-39-G-R	0.053	0.024	< 0.004	< 0.002	0.055	0.025	0.146	0.066	< 0.004	< 0.002
6050116	SP-49-G-R	0.013	0.005	< 0.005	< 0.002	0.111	0.043	0.518	0.200	< 0.005	< 0.002

Note: SP = *Scaphirhynchus platyrhynchus*, < indicates sample was below the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD. All samples were below detection (data not shown) for toxaphene (detection limit = 0.05 mg/kg ww), PCB-1242, PCB-1248 (detection limit = 0.01mg/kg ww), mirex, endrin, o,p'-DDE, alpha-BHC, beta-BHC, and delta-BHC (detection limit = 0.002 mg/kg ww).

Table A.14. Concentrations of organochlorine residues in digesta (stomach contents) of shovelnose sturgeon collected from the Platte River, Nebraska, 2002.

PACF Catalog No.	Sample ID	Organochlorine Concentration mg/kg dry weight			
		PCB-TOTAL	HCB	BHC*	Heptachlor Epoxide
6050094	SP-03	< 00.0855	< 0.0171	< 0.0171	< 0.0171
6050094	SP-06	< 0.104	< 0.0208	< 0.0208	< 0.0208
6050094	SP-08	< 0.192	< 0.0384	< 0.0384	< 0.0384
6050094	SP-13	< 0.179	< 0.0358	< 0.0358	< 0.0358
6050094	SP-15	< 0.143	< 0.0286	< 0.0286	< 0.0286
6050094	SP-17	< 0.204	< 0.0407	< 0.0407	< 0.0407
6050094	SP-18	< 0.175	< 0.0351	< 0.0351	< 0.0351
6050094	*SP-21-31	< 0.639	< 0.128	< 0.128	< 0.128
6050094	*SP-32-41	< 0.108	< 0.0215	< 0.0215	< 0.0215
6050094	*SP-42-53	< 0.199	< 0.0397	< 0.0397	< 0.0397
6050094	*SP1,2,7,5,11	< 0.618	< 0.124	< 0.124	< 0.124
		Chlordane	oxychlordane	cis-nonachlor	trans-nonachlor
6050094	SP-03	< 0.0171	< 0.0171	< 0.0171	< 0.0171
6050094	SP-06	< 0.0208	< 0.0208	< 0.0208	< 0.0208
6050094	SP-08	< 0.0384	< 0.0384	< 0.0384	< 0.0384
6050094	SP-13	< 0.0358	< 0.0358	< 0.0358	< 0.0358
6050094	SP-15	< 0.0286	< 0.0286	< 0.0286	< 0.0286
6050094	SP-17	< 0.0407	< 0.0407	< 0.0407	< 0.0407
6050094	SP-18	< 0.0351	< 0.0351	< 0.0351	< 0.0351
6050094	*SP-21-31	< 0.128	< 0.128	< 0.128	< 0.128
6050094	*SP-32-41	< 0.0215	< 0.0215	< 0.0215	< 0.0215
6050094	*SP-42-53	< 0.0397	< 0.0397	< 0.0397	< 0.0397
6050094	*SP1,2,7,5,11	< 0.124	< 0.124	< 0.124	< 0.124
		Dieldrin	Endrin	Mirex	toxaphene
6050094	SP-03	< 0.0171	< 0.0171	< 0.0171	< 0.0855
6050094	SP-06	< 0.0208	< 0.0208	< 0.0208	< 0.104
6050094	SP-08	< 0.0384	< 0.0384	< 0.0384	< 0.192
6050094	SP-13	< 0.0358	< 0.0358	< 0.0358	< 0.179
6050094	SP-15	< 0.0286	< 0.0286	< 0.0286	< 0.143
6050094	SP-17	< 0.0407	< 0.0407	< 0.0407	< 0.204
6050094	SP-18	< 0.0351	< 0.0351	< 0.0351	< 0.175
6050094	*SP-21-31	< 0.128	< 0.128	< 0.128	< 0.639
6050094	*SP-32-41	< 0.0215	< 0.0215	< 0.0215	< 0.108
6050094	*SP-42-53	< 0.0397	< 0.0397	< 0.0397	< 0.199
6050094	*SP1,2,7,5,11	< 0.124	< 0.124	< 0.124	< 0.618
		DDT	DDD	DDE	
6050094	SP-03	< 0.0171	< 0.0171	< 0.0171	
6050094	SP-06	< 0.0208	< 0.0208	< 0.0208	
6050094	SP-08	< 0.0384	< 0.0384	< 0.0384	
6050094	SP-13	< 0.0358	< 0.0358	< 0.0358	
6050094	SP-15	< 0.0286	< 0.0286	< 0.0286	
6050094	SP-17	< 0.0407	< 0.0407	< 0.0407	
6050094	SP-18	< 0.0351	< 0.0351	< 0.0351	
6050094	*SP-21-31	< 0.128	< 0.128	< 0.128	
6050094	*SP-32-41	< 0.0215	< 0.0215	< 0.0215	
6050094	*SP-42-53	< 0.0397	< 0.0397	< 0.0397	
6050094	*SP1,2,7,5,11	< 0.124	< 0.124	< 0.124	

Note: \* = composite sample, SP = *Scaphirhynchus platyrhynchus*, < indicates sample was below the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD. BHC = alpha, beta, delta and gama isomers. chlordane = alpha and gama isomers. DDD, DDE and DDT include p,p' and o,p' isomers.

Table A.15. Concentrations of organochlorine residues in potential pallid sturgeon food items (cyprinids less than 5 inches in length) collected from the Platte River, Nebraska, 2002.

PACF		Organochlorine Concentration mg/kg dry weight			
Catalog No.	Sample ID	PCB-TOTAL	HCB	BHC	Heptachlor Epoxide
6050097	CY-01	< 0.254	< 0.0507	< 0.0507	< 0.0507
6050097	CY-02	< 0.179	< 0.0358	< 0.0358	< 0.0358
6050097	CY-03	< 0.270	< 0.0540	< 0.0540	< 0.0540
6050097	CY-04	< 0.240	< 0.0480	< 0.0480	< 0.0480
6050097	CY-05	< 0.225	< 0.0450	< 0.0450	< 0.0450
6050097	CY-06	< 0.233	< 0.0465	< 0.0465	< 0.0465
6050097	CY-07	< 0.240	< 0.0479	< 0.0479	< 0.0479
6050097	CY-08	< 0.206	< 0.0412	< 0.0412	< 0.0412
6050097	CY-09	< 0.175	< 0.0350	< 0.0350	< 0.0350
6050097	CY-10A	< 0.217	< 0.0433	< 0.0433	< 0.0433
		Chlordane	oxychlordane	cis-nonachlor	trans-nonachlor
6050097	CY-01	< 0.0507	< 0.0507	< 0.0507	< 0.0507
6050097	CY-02	< 0.0358	< 0.0358	< 0.0358	< 0.0358
6050097	CY-03	< 0.0540	< 0.0540	< 0.0540	< 0.0540
6050097	CY-04	< 0.0480	< 0.0480	< 0.0480	< 0.0480
6050097	CY-05	< 0.0450	< 0.0450	< 0.0450	< 0.0450
6050097	CY-06	< 0.0465	< 0.0465	< 0.0465	< 0.0465
6050097	CY-07	< 0.0479	< 0.0479	< 0.0479	< 0.0479
6050097	CY-08	< 0.0412	< 0.0412	< 0.0412	< 0.0412
6050097	CY-09	< 0.0350	< 0.0350	< 0.0350	< 0.0350
6050097	CY-10A	< 0.0433	< 0.0433	< 0.0433	< 0.0433
		Dieldrin	Endrin	Mirex	toxaphene
6050097	CY-01	< 0.0507	< 0.0507	< 0.0507	< 0.254
6050097	CY-02	< 0.0358	< 0.0358	< 0.0358	< 0.179
6050097	CY-03	< 0.0540	< 0.0540	< 0.0540	< 0.270
6050097	CY-04	< 0.0480	< 0.0480	< 0.0480	< 0.240
6050097	CY-05	< 0.0450	< 0.0450	< 0.0450	< 0.225
6050097	CY-06	< 0.0465	< 0.0465	< 0.0465	< 0.233
6050097	CY-07	< 0.0479	< 0.0479	< 0.0479	< 0.240
6050097	CY-08	< 0.0412	< 0.0412	< 0.0412	< 0.206
6050097	CY-09	< 0.0350	< 0.0350	< 0.0350	< 0.175
6050097	CY-10A	< 0.0433	< 0.0433	< 0.0433	< 0.217
		DDD	DDE	DDT	
6050097	CY-01	< 0.0507	< 0.0507	< 0.0507	
6050097	CY-02	< 0.0358	< 0.0358	< 0.0358	
6050097	CY-03	< 0.0540	< 0.0540	< 0.0540	
6050097	CY-04	< 0.0480	< 0.0480	< 0.0480	
6050097	CY-05	< 0.0450	< 0.0450	< 0.0450	
6050097	CY-06	< 0.0465	< 0.0465	< 0.0465	
6050097	CY-07	< 0.0479	< 0.0479	< 0.0479	
6050097	CY-08	< 0.0412	< 0.0412	< 0.0412	
6050097	CY-09	< 0.0350	< 0.0350	< 0.0350	
6050097	CY-10A	< 0.0433	< 0.0433	< 0.0433	

Note: \* = composite sample, SP = *Scaphirhynchus platyrhynchus*, < indicates sample was below the detection limit (value is the detection limit), PACF = Patuxent Analytical Control Facility, Laurel MD. BHC = alpha, beta, delta and gama isomers. chlordane = alpha and gama isomers. DDD, DDE and DDT include p,p' and o,p' isomers.

Table A.16. Concentrations of herbicides and herbicide metabolites in water grab samples from the lower Platte River, 2002.

Herbicide Classification	Analyte	Sample ID	Concentration $\mu\text{g/L}$	Analyte	Sample ID	Concentration $\mu\text{g/L}$
Triazine	Atrazine	50SCA	23.00	Propazine	50SCA	0.29
		SCERA	13.00		SCERA	0.20
		6477A	0.99		6477A	< 0.1
		7915A	8.60		7915A	0.20
		HWY81A	2.00		HWY81A	0.20
	Desethyl atrazine	50SCA	3.50	Metribuzin	50SCA	2.70
		SCERA	1.90		SCERA	2.90
		6477A	0.19		6477A	0.16
		7915A	0.95		7915A	0.62
		HWY81A	0.69		HWY81A	< 0.1
	Desisopropyl atrazine	50SCA	0.60	Cyanazine	50SCA	0.19
		SCERA	0.59		SCERA	0.16
		6477A	< 0.1		6477A	< 0.1
		7915A	0.33		7915A	< 0.1
		HWY81A	0.14		HWY81A	< 0.1
	Simazine	50SCA	0.28	TOTAL TRIAZINES	50SCA	30.56
		SCERA	0.26		SCERA	19.01
		6477A	< 0.1		6477A	1.34
		7915A	< 0.1		7915A	10.08
		HWY81A	< 0.1		HWY81A	3.03
Acetanilide	Metolachlor	50SCA	4.20	TOTAL HERBICIDES	50SCA	47.59
		SCERA	4.10		SCERA	24.78
		6477A	0.33		6477A	11.94
		7915A	4.00		7915A	10.95
		HWY81A	1.9		HWY81A	2.35
Pyridazone	Norflurazone	50SCA	0.1			
		SCERA	< 0.1			
		6477A	0.11			
		7915A	< 0.1			
		HWY81A	0.10			

Note: Herbicides classifications according to Ware (1994), < indicates sample was below the detection limit (value is the detection limit),

Table A.17. Concentrations of herbicides and herbicide metabolites in integrated equal width and depth water grab samples collected by the U.S. Geological Survey from the lower Platte River, 2002.

Station Location	Date (YYMMDD)	Concentration µg/L								
		Atrazine	deethyl- atrazine	Deisopropyl- Atrazine	Cyanazine	Simazine	Propazine	TOTAL TRIAZINES	Metolchlor	Metribuzin
Duncan Bridge near Duncan, NE	020528	3.38	0.62	0.32	< 0.05	< 0.05	< 0.05	4.32	0.97	< 0.05
Duncan Bridge near Duncan, NE	020610	0.60	0.23	0.06	< 0.05	< 0.05	< 0.05	0.89	0.05	< 0.05
Hwy 64 bridge near Leshara, NE	020530	1.51	0.18	< 0.05	< 0.05	< 0.05	< 0.05	1.69	0.22	< 0.05
Hwy 64 bridge near Leshara, NE	020610	0.72	0.11	0.05	< 0.05	< 0.05	< 0.05	0.88	0.06	< 0.05
Hwy 50 bridge near Louisville, NE	020510	2.00	E 0.05	NA	0.02	0.01	NA	2.07	0.24	0.01
Hwy 50 bridge near Louisville, NE	020523	0.94	E 0.07	NA	< 0.018	0.01	NA	1.02	0.18	E 0.01
Hwy 50 bridge near Louisville, NE	020613	14.30	E 0.54	NA	< 0.05	0.05	NA	14.89	2.37	0.13
Hwy 50 bridge near Louisville, NE	020627	1.14	E 0.13	NA	< 0.018	0.01	NA	1.28	0.18	< 0.006

Note: < indicates sample was below the detection limit (value is the detection limit),

Table A.18. Condition and organo-somatic indices and body and organ weights for shovelnose sturgeon collected from the lower Platte River, NE, 2002.

Fish ID	Gender	CF	Liver mass	Gonad Mass	HIS	HSI <sub>GI</sub>	SSI	GSI	Standard Weight (Ws)	Relative Weight (Wr)
SP-01	M	0.29	8.30	14.30	1.42	1.38	97.62	2.38	885.67	67.75
SP-02	M	0.30	4.50	8.90	1.15	1.13	97.78	2.23	536.05	74.62
SP-03	F	0.38	12.30	20.00	1.68	1.64	97.33	2.67	832.13	90.13
SP-04	M	0.33	6.60	7.80	1.34	1.32	98.44	1.56	613.15	81.55
SP-05	F	0.34	6.50	12.40	0.98	0.96	98.16	1.84	836.91	80.65
SP-06	M	0.33	4.80	9.70	0.81	0.80	98.38	1.62	749.48	80.06
SP-07	M	0.31	9.20	11.00	1.03	1.02	98.78	1.22	1284.22	70.08
SP-08	M	0.41	10.40	13.20	1.17	1.16	98.53	1.47	946.77	95.06
SP-09	F	0.37	6.90	16.40	1.18	1.15	97.27	2.73	664.58	90.28
SP-10	M	0.39	7.80	25.10	1.58	1.50	95.17	4.83	546.63	95.13
SP-11	M	0.38	9.60	40.20	1.39	1.32	94.49	5.51	817.94	89.25
SP-12	F	0.35	6.90	10.80	1.44	1.41	97.80	2.20	568.21	86.24
SP-13	F	0.36	4.50	8.50	1.00	0.98	98.15	1.85	522.18	88.09
SP-14	M	0.41	11.20	27.30	1.28	1.24	96.97	3.03	931.23	96.65
SP-15	F	0.33	8.00	19.70	1.13	1.10	97.30	2.70	931.23	78.39
SP-16	F	0.35	8.90	15.10	1.26	1.24	97.90	2.10	880.71	81.75
SP-17	M	0.34	5.90	2.60	1.32	1.31	99.42	0.58	536.05	83.95
SP-18	M	0.34	10.50	16.60	1.19	1.17	98.16	1.84	1159.69	77.61
SP-19	F	0.45	17.40	108.20	1.75	1.58	90.16	9.84	1049.46	104.82
SP-20	M	0.30	5.60	7.40	1.09	1.08	98.58	1.42	710.39	73.20
SP-21	F	0.39	5.30	10.60	1.08	1.06	97.88	2.12	511.94	97.67
SP-22	F	0.39	5.90	152.80	0.74	0.62	83.92	16.08	1055.07	90.04
SP-23	F	0.39	6.30	9.50	1.08	1.06	98.40	1.60	628.66	94.65
SP-24	M	0.39	7.10	17.00	1.20	1.16	97.21	2.79	640.48	95.24
SP-25	F	0.42	6.20	172.20	0.71	0.59	83.60	16.40	1083.44	96.91
SP-26	M	0.39	7.10	13.40	1.21	1.18	97.77	2.23	624.76	96.04
SP-27	M	0.41	6.40	7.20	0.81	0.80	99.10	0.90	827.38	96.69
SP-28	M	0.33	3.70	8.10	0.75	0.74	98.38	1.62	613.15	81.55
SP-29	F	0.37	9.20	14.70	1.17	1.15	98.16	1.84	910.78	87.84
SP-30	F	0.44	6.00	117.20	0.88	0.75	85.35	14.65	767.33	104.26
SP-31	M	0.42	6.60	11.30	1.12	1.10	98.12	1.88	575.53	104.25
SP-32	M	0.34	5.80	11.90	0.99	0.97	98.02	1.98	740.67	81.01
SP-33	F	0.34	6.90	120.90	0.78	0.69	87.91	12.09	1277.78	78.26
SP-34	M	0.34	2.64	5.40	0.44	0.44	99.10	0.90	731.92	81.98
SP-35	M	0.29	4.50	10.90	0.70	0.69	98.32	1.68	946.77	68.65
SP-36	M	0.31	5.90	9.60	0.85	0.84	98.63	1.37	967.78	72.33
SP-37	M	0.39	8.79	18.50	0.74	0.73	98.46	1.54	1376.77	87.16
SP-38	F	0.38	5.60	4.80	0.70	0.70	99.40	0.60	890.65	89.82
SP-39	F	0.47	5.20	206.30	0.75	0.58	77.08	22.92	799.27	112.60
SP-40	M	0.37	6.60	7.40	1.03	1.02	98.86	1.14	736.29	88.28
SP-41	F	0.31	5.50	3.90	0.99	0.98	99.30	0.70	749.48	74.72
SP-42	M	0.32	4.40	11.60	0.80	0.79	97.93	2.07	731.92	76.51
SP-43	M	0.37	12.80	18.50	1.24	1.22	98.24	1.76	1264.97	83.01
SP-44	M	0.44	4.20	15.10	0.57	0.56	97.99	2.01	701.90	106.85
SP-45	M	0.29	4.70	11.10	0.56	0.55	98.69	1.31	1284.22	66.19
SP-46	M	0.30	4.50	11.60	0.55	0.55	98.59	1.41	1189.99	69.33
SP-47	M	0.43	3.00	3.50	0.43	0.43	99.50	0.50	664.58	105.33
SP-48	M	0.40	4.50	9.50	0.61	0.60	98.73	1.27	776.36	96.60
SP-49	F	0.39	7.40	135.90	0.86	0.74	86.41	13.59	1106.51	90.37
SP-50	M	0.42	8.00	16.30	0.91	0.89	98.19	1.81	905.72	99.37
SP-51	F	0.33	5.00	9.00	0.78	0.77	98.62	1.38	832.13	78.11
SP-52	M	0.32	1.90	5.90	0.38	0.38	98.82	1.18	640.48	78.07
SP-53	M	0.33	5.10	13.40	0.74	0.73	98.09	1.91429	910.78	76.86

Note: SP = *Scaphirhynchus platyrhynchus*, M = male, F = female, CF = condition factor, HIS = hepato-somatic index, HSI<sub>GI</sub> = same as HIS except the calculation included gonad weight as part of the body mass, SSI = spleno-somatic index, GSI = gonado-somatic index.