EFFECTS OF PECTORAL FIN RAY REMOVAL ON WHITE STURGEON (ACIPENSER TRANSMONTANUS) AND SIBERIAN STURGEON (ACIPENSER BAERII) SWIMMING PERFORMANCE

By,

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(Under the Direction of Douglas L. Peterson)

ABSTRACT

The effects of two pectoral fin ray sampling methods on swimming performance were evaluated for hatchery-reared white sturgeon and Siberian sturgeon. Fish were subjected to either a notch removal or a full removal of the pectoral fin ray whereas control fish were subjected to a sham operation. Mean relative growth in fork length (F = 1.30; df = 2, 27; P = 0.29) and weight (F = 0.38; df = 2, 27; P = 0.69) were not significantly different among treatments white sturgeon. Mean 10-min critical-station holding speeds (CSHS) was not significantly different among treatments for white (F = 1.58; df = 2, 44; P = 0.22) or Siberian sturgeon (F = 0.55; df = 2, 42; P = 0.58). Analysis of variance indicates that there were no treatment effects on the CSHS and survival and growth of white sturgeon. Survival and growth was not evaluated for Siberian sturgeon.

INDEX WORDS:Fin ray removal, Swimming performance, Survival and growth, Whitesturgeon, Siberian sturgeon, Critical swimming speed

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DEDICATION

I dedicate this thesis to my family, especially my parents Ky and Hoa Nguyen, who have given me a tremendous amount of support throughout my graduate study. I would also like to dedicate this to my girlfriend Lara Church and her family: for their support, love, and patience.

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CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

The order Acipenseriformes is comprised of 27 extant species of sturgeons and paddlefishes distributed exclusively within the Northern Hemisphere. To varying degrees all members of the order are characterized by a protracted life cycle, slow growth, and late maturation (Bemis and Kynard 1997; Birstein 1993; Billard and Lecointre 2001). Sturgeons (family Acipenseridae) are prized for their high-quality flesh and roe and have been commercially exploited throughout the 19th and 20th centuries, resulting in severe declines of most populations (Brennan and Caillet 1989; Birstein 1993; Collins et al. 2000; Billard and Lecointre 2001; Schueller and Peterson 2010). Environmental degradation, such as pollution and dam construction, has exacerbated population declines for many sturgeon species worldwide (Rieman and Beamesderfer 1990; Birstein 1993; Luk'yanenko et al. 1999; Collins et al. 2000; Billard and Lecointre 2001). Presently, 24 species of sturgeons are currently listed on the IUCN Red List for Threatened Species 2012. Within the U.S., six out of nine species of sturgeon are currently protected under the Endangered Species Act (Williams et al. 1989).

Two species of sturgeon, the white (*Acipenser transmontanus*) and Siberian sturgeon (*Acipenser baerii*), are the most commonly farmed sturgeon species in commercial aquaculture (Williot et al. 2001). Global aquaculture of these species has increased dramatically over the past few decades, which has mitigated population declines of species associated with overfishing (Bronzi et al. 2011). Consequently, successful commercial production of these two species also

allow them to be readily available, hence, researchers can utilize them as surrogate species in many different types of laboratory studies that evaluate their physiology, biology, and behavior.

General Life History and Contemporary Status of White Sturgeon

White sturgeon are endemic to the Pacific coast of the United States, where they are distributed from the southern parts of Alaska south to California and Mexico. The most abundant populations are found in the Sacramento, Columbia, and Fraser river systems (Scott and Crossman 1973). The white sturgeon is the largest species of freshwater fish in North America but only the third largest of all sturgeon species. The largest white sturgeon on record was a 630 kg individual captured from the Fraser River in 1897 (Scott and Crossman 1973). Like many other sturgeon species, white sturgeon are anadramous – the adults spending a majority of their lives in brackish or marine environments and migrating up natal rivers to spawn at various times of year depending on latitude. Many natural and anthropogenic factors, however, have led to the isolation of several populations and as a result, some populations have become landlocked, completing their life cycle entirely within freshwater (Duke et al. 1999; Paragamian and Kruse 2001; Paragamian and Hansen 2008).

White sturgeon populations supported several vigorous commercial and recreational fisheries from the mid-1870s until the late 1880s throughout the Columbia River and the San Francisco Bay (Galbreath 1985; Brennan and Cailliet 1991). By the early 1890s, however, commercial harvest had peaked at approximately 2,000 tones, and soon thereafter many of these fisheries collapsed (Galbreath 1985). A 35-year fishing ban started for the species in 1917, which allowed some stocks to rebound, and by 1954 sport fishing of some populations, such as the San Francisco Bay, was resumed (Brennan and Cailliet 1991). In the Columbia River, multi-state agencies cooperatively imposed strict regulations on white sturgeon bag limits, size limits,

and fishing techniques. This allowed the Columbia River populations to flourish below Bonneville Dam, and soon thereafter the commercial harvest also rebounded (Galbreath 1985). Although commercial harvest of white sturgeon in the Columbia River in 1985 rivaled numbers of fish harvested in the mid-1880s, overall fish size has dramatically decreased from an average weight of 68 kg to an average of 14-16 kg (Galbreath 1985). Reduced mean fish sizes landed in that fishery suggest that over-exploitation is still occurring (Kohlhorst et al. 1980). Continued chronic overfishing coupled with habitat loss and population fragmentation (i.e. dam construction) has led to the recent listing of several white sturgeon populations. The most notable of these are found in the upstream reaches of the Columbia, Nechako, and Fraser Rivers (UCWSRI 2002; Paragamian and Hansen 2008). A genetically distinct population also occurs in the Kootenai River, Idaho, and has been listed as a federally endangered since 1994 (USFWS 1994; Duke et al. 1999). This population was isolated from the Columbia River at the end of the last glacial period (Duke 1999; Paragamian et al. 2001).

General Life History and Contemporary Status of Siberian Sturgeon

In contrast to the white sturgeon, the Siberian sturgeon has been poorly studied, probably because the species is rare and populations are dispersed throughout large remote rivers where access is difficult (Ruban 1997). Siberian sturgeon are widely distributed throughout all major rivers in Siberia, with the most notable populations occurring in Lake Baikal, and the Ob, Aldan, and Lena River systems (Billard and Lecointre 2001). There currently are three recognized subspecies of Siberian sturgeon: *A. b. baerii*, which occurs mainly in the Ob River Basin, and *A. b. baicalensis* and *A. b. stenorhynchus*, which occur in the Eastern Siberian River basins (Ruban 1997). Unlike most other sturgeon species, Siberian sturgeon are predominantly a freshwater

species; however, certain populations may inhabit estuarine environments on a seasonal basis (Birstein 1993; Billard and Lecointre 2001).

All major populations of Siberian sturgeon have suffered dramatic declines (50–80%) during the last 60 years as a result of overfishing, dam construction, and water pollution (Ruban and Zhu 2010). Up to 40% of the spawning grounds of *A. b. baerii* are now inaccessible to migrating adults because of dam construction on the Ob River and both *A. b. baerii* and *A. b. stenorhynchus* have been severely overfished (Ruban 1997). The remaining sub-species, *A. b. baicalensis* is currently listed in the Red Data Book of the Russian Federation (Kolosov 1983; Ruban 1997). All populations of Siberian sturgeon, especially those in the Ob and Kolyma rivers, are also hindered by reproductive abnormalities attributed to pollution. A recent study by Ruban and Zhu (2010) documented reproductive abnormalities in 80–100% of females in these rivers, including several instances of complete sterility. Currently, published stock assessments of Siberian sturgeon are lacking.

Assessing Age and Growth in Sturgeon

Given the history of commercial sturgeon fisheries, the need for current age data from extant stocks is crucial to understanding their population dynamics and abundance trends because they provide the quantitative basis for calculations of growth, mortality, and recruitment rates (Campana 2001). Understanding these basic population parameters allows researchers to estimate both current and future sizes from quantitative population models. Thus, accurate age information is a critical "lynchpin" in the effective management of sturgeon stocks, especially given their protracted life cycle and slow reproductive rates (Beamish and McFarlane 1983; Paragamian and Beamesderfer 2003). Historical age estimates of sturgeon were obtained from internal calcified structures, such as the clavicles, cleithrums, and otoliths (Cuerrier 1951).

However, traditional methods of estimating fish age from these structures are impractical for imperiled populations of sturgeons because removing these boney structures requires that the fish be sacrificed. Recent studies also show that age estimates derived from these structures is frequently inaccurate for sturgeons (Cuerrier 1951; Brennan and Caillet 1989). Consequently, the analysis of annular growth rings present on cross-sections of the calcified pectoral fin ray is currently the most widely accepted method for estimating age in sturgeons because they can be obtained from non-lethal biopsy and because annuli are more easily interpreted (Cuerrier 1951; Rien and Beamesderfer 1994; Rien et al. 1994). The two widely accepted methods for sampling pectoral fin rays in sturgeon are referred to as the "notch removal" method and the "full removal" method. In the notch removal method, only a small section (2-4 cm) of the marginal pectoral ray is removed (Figure 1-1; Brennan and Cailliet 1989; Peterson et al. 2002) whereas in the full removal method, the entire marginal pectoral ray is removed (Cuerrier 1951; Brennan and Caillet 1989; Collins and Smith 1996;; Paragamian and Beamesderfer 2003; Hurley et al. 2004; Koch et al. 2008).

Although obtaining age and growth information from pectoral fin rays provides critical data for stock assessment and stock recovery programs, some researchers have expressed concerns regarding the sub-lethal effects of fin ray removal on sturgeons (Kohlhorst 1979; Collins and Smith 1996; Parsons et al. 2003). Because sturgeons use their pectoral fins for maneuvering, swimming, station-holding, and benthic "shuffling" (Findeis 1997; Wilga and Lauder 1999); any mutilation of pectoral fin ray could affect their swimming performance. Consequently, evaluating the sub-lethal effects of pectoral fin ray sampling on the swimming performance of sturgeons is critical in determining the utility and practicality of the practice.

Sturgeon Swimming Behaviors

Sturgeons typically employ different swimming behaviors in various water velocities, allowing them to effectively move or maintain position within the lotic environment (Adams et al. 1997; Adams et al. 1999; Adams et al. 2003; Hoover et al. 2011). Observed swimming behaviors in sturgeons are characterized as (1) station-holding, in which the sturgeon appresses itself to the substrate, thereby maintaining station; (2) substrate skimming, in which the ventral side of the sturgeon is in contact with the substrate while swimming; and (3) free swimming, in which the fish propels itself through the water column without contacting the substrate (Adams et al. 1997, 1999, 2003; Hoover et al. 2011). In previous swim studies, researchers have found that prolonged periods of free swimming are rare in sturgeon and that the fish seem to prefer substrate skimming especially at higher current velocities (Adams et al. 1997; 2003; Boysen and Hoover 2009; Parsons et al. 2003).

Evaluating Swimming Performance

One method of evaluating a fish's swimming performance is to evaluate its critical swimming speed (U_{crit}), which is defined as the speed at which a fish can maintain station for a prescribed period of time (Brett 1964). By quantitatively evaluating the U_{crit} of fishes, biologists can determine how different environmental factors affect the fish's swimming performance (Plaut 2001). To evaluate U_{crit} values in fish, Brett (1964) designed a method in which fish are placed into an enclosed swim chamber or "flume" in which water velocity is increased incrementally at specific time intervals. For benthic fishes like sturgeons, however, Parsons et al. (2003) developed the term "critical station-holding speed" (CSHS) which accounts for benthic station-holding behaviors. Although similar to U_{crit} , critical station-holding speed is defined as the speed at which a fish can maintain station, either by swimming *or adherence to*

the bottom, for a prescribed period of time. The equation for CSHS is defined as (Parsons et al. 2003):

$$CSHS = U_1 + [U_2 (T_1/T_2)]$$
 Equation 1

Where:

CSHS = speed at which a fish can maintain station, either by swimming or adherence to the bottom, for a prescribed period of time

 U_1 = highest velocity maintained for the entire interval (cm/s)

 U_2 = velocity increment (cm/s)

 T_1 = time elapsed at fatigue velocity (min)

 T_2 = the prescribed interval time (min)

Current methods for reliable estimating age and growth of wild sturgeons require the sampling of the pectoral fin ray; however, studies evaluating the effects of sub-lethal effects of this practice are lacking. Although several authors have evaluated the general swimming performance of shovelnose (*Scaphirhynchus platorynchus*), pallid (*Scaphirhynchus albus*), and lake (*Acipenser fulvescens*) (Adams et al. 1997; Adams et al. 2003; Peake et al. 1995; Hoover at al. 2011), few have specifically evaluated the effects of fin ray removal on sturgeon swimming performance. Only Parsons et al. (2003) has conducted such a study, concluding that full ray removal has no significant effect on the 10-min CSHS of adult shovelnose sturgeon. Studies evaluating the effects of pectoral ray sampling on the survival and growth of different sturgeon species have produced contradictory results. Collins and Smith (1996) conclude that full ray removal had no significant effect on the survival and growth of Atlantic (*Acipenser oxyrinchus oxyrinchus*) or shortnose (*Acipenser brevirostrum*); however, Kolhourst (1979) suggests that full ray removal caused substantial mortality in wild white sturgeon. To date, no studies have

evaluated the effects of pectoral fin ray sampling on the health and swimming performance of white or Siberian sturgeon in a controlled setting.

The need for sampling fin rays to obtain age information in sturgeons coupled with contradicting results from previous fin ray removal studies warrants a controlled laboratory experiment to more accurately quantify the effects of fin ray removal on sturgeons. In this thesis, I present the results of a controlled experiment which specifically evaluated the effects of two fin ray sampling methods on two species of sturgeons. Chapter 2 describes the acute and long-term effects of two fin ray sampling methods on the survival, growth, and swimming performance of sub-adult white sturgeon. Chapter 3 describes the acute effects of these same two fin ray sampling methods on the swimming performance of Siberian sturgeon. The concluding chapter provides a summary and synthesis of key findings from both studies with implications for future management decisions regarding sampling of pectoral fin rays in wild sturgeon.

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Figure 1-1. View demonstrating the two common methods of obtaining pectoral fin rays from sturgeon. The notch removal method (A) and full removal method (B) are both non-lethal methods.

CHAPER 2

EFFECTS OF FIN RAY REMOVAL ON THE HEALTH AND SWIMMING PERFORMANCE

OF WHITE STURGEON (ACIPENSER TRANSMONTANUS)¹

¹ Nguyen, P. L., Z. Jackson, and D.L. Peterson. To be submitted to *Transactions of the American Fisheries Society*.

Abstract

Age information is necessary to successfully manage threatened or endangered populations of white sturgeon. Sturgeon are typically aged by analyzing cross-sections of their pectoral fin ray, however, removal of the fin ray may have lasting deleterious effects. The effects of two fin ray sampling methods on growth, survival, and swimming performance were evaluated for hatchery-reared sub-adult white sturgeon. Fish were subjected to either the notch removal treatment in which a small notch was removed from the marginal pectoral fin ray, or the full removal treatment in which the entire marginal pectoral fin ray was removed. Control fish did not have their fin ray removed, but they were placed through a sham operation. Survival and growth were evaluated on a group of fish housed in a 42,000-L outdoor raceway over a 6-month period. A modified 3,230-L Brett-type swim tunnel was used to evaluate the 10-min critical station-holding speeds (CSHS) on a separate group of fish. Water quality and fish size were comparable among treatments. Analysis of variance indicated that there were no significant differences in relative growth among treatment groups, and mortality of fish was not observed in any treatment. Mean 10-min CSHS (mean \pm SE) were 108 \pm 2.3 cm/s, 110 \pm 2.6 cm/s, and 115 \pm 3.5 cm/s for the notch removal treatment, full removal treatment, and control treatment, respectively, and were not significantly different among treatments. A temporary loss of horizontal swimming orientation was observed in some individuals from the full removal treatment only. Results indicate that both fin ray sampling methods have a negligible effect on the swimming performance of sub-adult white sturgeon. However, because of some behavioral anomalies observed only in the full removal treatment, the notch removal method is recommended.

Introduction

White sturgeon are endemic to the Pacific coast of the North America, where they are distributed from the southern parts of Alaska south to California and Mexico, with the most abundant populations found in the Sacramento, Columbia, and Fraser river systems (Scott and Crossman 1973; Perrin et al. 2003). They are the largest freshwater fish in North America and are a popular target for both commercial and recreational anglers because of their large size and valuable meat and roe. In the late 1890s, annual commercial harvest of white sturgeon peaked at approximately 2,500 tons, and many targeted white sturgeon fisheries collapsed (Galbreath 1985). Since then, chronic overfishing, coupled with habitat degradation and population fragmentation resulting from dam construction has led to the federal listing of several critically imperiled populations (Brennan and Caillet 1989; Rieman and Beamesderfer 1990; Birstein 1993; Collins et al. 2000; Billard and Lecointre 2001). The most notable of these, occur in the Columbia, Nechako, Fraser, and Kootenai Rivers (Setter and Brannon 1990; USFWS 1994; Duke et al. 1999; UCWSRI 2002; Paragamian and Hansen 2008).

To better understand the population dynamics of critically imperiled white sturgeon populations, biologists must obtain quantitative age data from each population (Campana 2001). Because of their protracted life cycle, white sturgeon populations are typically comprised of dozens of different age classes. Consequently, accurate determinations of the population agestructure are critical to understand population status and hence the most appropriate management strategy (Beamish and McFarlane 1983; Paragamian and Beamesderfer 2003). Unfortunately, traditional methods of estimating ages from internal calcified structures, such as otoliths, are impractical for threatened or endangered species and inaccurate for sturgeon (Cuerrier 1951; Brennan and Caillet 1989). Alternatively, analysis of cross-sections of the calcified pectoral fin

ray is the most widely accepted method for estimating age in sturgeons because fin rays can be easily sampled and annuli are typically easier to interpret (Cuerrier 1951; Rien and Beamesderfer 1994; Rien et al. 1994). The most widely accepted methods for sampling sturgeon fin rays are referred to as the "notch removal" method and the "full removal" method. In the notch removal method, only a 2-4 cm section of the marginal pectoral ray is removed (Brennan and Cailliet 1989; Peterson et al. 2002). In the full removal method, the entire marginal pectoral ray is removed (Cuerrier 1951; Brennan and Caillet 1989; Collins and Smith 1996; Paragamian and Beamesderfer 2003; Hurley et al. 2004; Koch et al. 2008). Although several previous studies have suggested that both methods are non-lethal, the pectoral fins of sturgeon are critically important for the fish in maneuvering, swimming, station-holding, and benthic shuffling (Findeis 1997; Wilga and Lauder 1999). Consequently, mutilation of the pectoral fin ray may alter the structural integrity of the pectoral fin, possibly affecting swimming performance and health of sturgeons subjected to fin ray sampling.

One way to assess how fin ray removal might affect a sturgeon's overall health is to quantify sturgeon swimming performance in an environmentally controlled swim chamber (Plaut 2001; Parsons et al. 2003). Although many studies have quantified swimming performance by determining critical swimming speeds (U_{crit}) and swimming endurance of shovelnose s (*Scaphirhynchus platorynchus;* Adams et al. 1997; Adams et al. 2003; Hoover et al. 2008), pallid (*Scaphirhynchus albus*; Adams et al. 1999) and lake (*Acipenser fulvescens*; Peake et al. 1995), only Parsons et al. (2003) specifically evaluated the effects of fin ray removal on the swimming performance of sturgeon. Parsons et al. (2003) concluded that full fin ray removal has no significant effect on the swimming abilities of wild adult shovelnose sturgeon.

Previous studies evaluating the effects of fin ray removal on sturgeon survival or growth have produced contradictory results. In a controlled laboratory experiment, Collins and Smith (1996) concluded that full ray removal has no significant effect on the survival or growth in Atlantic sturgeon (*Acipenser oxyrinchus oxyrinchus*) or shortnose sturgeon (*Acipenser brevirostrum*). In contrast, Kohlhorst (1979) found that wild white sturgeon subjected to the full removal method had lower return rates than those that had no fin ray alteration over a 3-yr period. However, many environmental factors could have affected his results, and the author acknowledged that his results may be imprecise because of small sample sizes. Consequently, controlled studies are needed to ensure that sampling of pectoral fin rays does not reduce survival of wild sturgeon. The objectives of this study were to assess effects of two fin ray sampling methods on the swimming performance, survival, and growth of white sturgeon.

Methods

Experimental Fish & Fish Care

Hatchery-reared sub-adult white sturgeon (n = 100) from the Sacramento – San Joaquin River brood stock were obtained from the Lazy Q Fish Ranch in Dixon, CA. Fish were age-4 with mean fork length (FL) and weight of 97.4 cm (range: 84-116 cm) and 7.3 kg (4.3 - 12.0 kg), respectively. All fish were thoroughly screened for bacterial and viral diseases at the University of California, Davis, Fish Pathology Laboratory, after which they were transported to the University of Georgia's Cohutta Fisheries Center, in Cohutta, GA. Upon arrival, individual fish were implanted with passive-integrated-transponders (PIT) tags to facilitate individual identification during trials. Fish were acclimated to local conditions for eight months in a 42,500 L-outdoor-raceway with spring-fed water supplied at a constant rate of 95 L/min. Temperature and dissolved oxygen in the raceway were monitored daily with a portable multi-meter

(YSI® 55; Yellow Springs Instruments, Inc., Yellow Springs, Ohio). Fish were fed a diet of size 6-mm Skretting sinking trout pellets (44% protein content, 28% oil content) at a rate of approximately 1% of total fish biomass per day.

Swimming Performance Study

Swimming performance trials were conducted from Nov 9, 2012 to Dec 6, 2012 using a specially-designed, 3,230-L (total water volume) Brett-type (1964) swim flume constructed by the U.S. Corps of Engineers in Vicksburg, MS (Hoover et al. 2011). The flume measured 244 cm x 91 cm x 91 cm (L x H x W) and was capable of maintaining boundary-layer or rectilinear flow with the use of different inserts. For this study, a cylindrical tube insert, measuring 46 cm in diameter and 150 cm in length was placed in the swim tunnel to induce and maintain rectilinear flow throughout the experiment. Acrylic grids were placed at both ends of the tunnel to entrain the fish within the swim chamber of tunnel. The cathode of a pulsed DC electro-fisher (Advanced Backpack AbP-3) was attached to a third grid which was positioned 61-cm behind the posterior end of the tube insert. During the trials, a localized electric current was used ad libitum to prevent fish from resting against the rear grid.

Current velocities within the tube insert were confirmed by placing the probes from two electronic flow meters (Marsh-McBirney Flo-Mate 2000) into the center of the tube insert. Water within the flume was changed daily. Throughout the trials temperature and dissolved oxygen within the flume were constantly monitored by using a portable multi-meter (YSI 55 multi-meter). Each swim trial was video-recorded (Sony DCR-SX45) to facilitate subsequent evaluation of behavioral responses.

Immediately before each trial, a single fish was randomly selected from the raceway and assigned to one of the three previously described treatments. Fish were introduced into the swim

tunnel where they were allowed to acclimate for 10-min at a base flow velocity of 10 cm/s. Acclimation velocities were then incrementally increased to 20, 40, 60, and 80 cm/s at 10-min intervals with a 1-min rest between each increment. Velocity increment values for the acclimation period were selected based on pre-experimental trials in which we determined that white sturgeon of comparable sizes required little effort to maintain station at velocities between 10-80 cm/s. Upon completion of the acclimation period, fish were allowed to rest for 10-min after which water velocity was increased to 90 cm/s. Water velocity was then increased incrementally by 10 cm/s every 10-min, with a 10-min rest between each successive increment. The 10-min critical station-holding speed (CSHS), which is the maximum speed at which a fish can maintain station, was determined from the maximum velocity at which the fish reached fatigue (the point at which the fish could no longer maintain position). Critical station-holding speed was then calculated as described by Parsons et al. (2003) by using the equation:

$$CSHS = U_1 + [U_2 (T_1/T_2)]$$
Equation 1

Where:

CSHS = speed at which a fish can maintain station, either by swimming or adherence to the bottom, for a prescribed period of time

 U_1 = highest velocity maintained for the entire interval (cm/s)

 U_2 = velocity increment (cm/s)

 T_1 = time elapsed at fatigue velocity (min)

 T_2 = the prescribed interval time (min)

The weight (kg), fork length (cm), cross-section (cm), and PIT tag number of each fish was recorded after each swim trial. All fish were swum only once to minimize any potential bias associated with training or habituation.

The CSHS of fish with cross-sectional areas >10% of the cross-sectional area of the tube insert were corrected for "solid blocking" as described by Bell and Terhune (1970) by using the equation:

$$UF = UT (1 + \varepsilon s)$$
 Equation 2

Where:

UF = corrected velocity

UT = velocity in the tunnel without the fish

 $\varepsilon s = a$ fractional error resulting from solid blocking

For each fish, as is defined as:

$$\epsilon s = \tau \lambda (A_o/A_T)^{1.5}$$
 Equation 3

Where:

 τ = a dimensionless value depending on the swim chamber cross section (0.8 in this study)

 λ = shape factor for the fish (λ = 0.5 x body length/body width)

 $A_o = cross$ sectional area of the fish

 $A_T = cross \ sectional \ area \ for \ swim \ tunnel$

In addition to measuring CSHS, I also quantified three behavioral parameters: time spent hunkering (TSH), tail beat frequency (TBF), and gill beat frequency (GBF) from video recordings. A fish was determined to be hunkering if it was able to maintain station by adhering to the bottom of the tunnel without body or caudal fin undulation. The amount of time a fish spent hunkering was recorded with a stopwatch for each velocity increment. The total TSH at each velocity for all fish within a treatment was then averaged to obtain a mean TSH for that treatment. The amount of TSH for each treatment group was expressed as a percent of total time swam at each velocity. Tail beat frequency was determined by using the method described by Parsons et al. (2003) whereby tail beats was estimated by counting the number of beats over a 5-s time period only when the fish is actively swimming. Estimations of TBF were repeated five times for an individual fish at each velocity, and those values were averaged to obtain an average TBF for that fish at each specific velocity. TBF values for all fish within a treatment were then averaged to obtain the average TBF at each velocity, for each treatment. The average values were then multiplied by 12 to obtain an average beats/min value. Gill beat frequencies were recorded for 30-sec during the 1-min rest between velocity increments during the acclimation period. Gill beat frequency was recorded for 30-sec immediately before and after each velocity increment. Any unusual behaviors were also noted.

Survival and Growth Study

Using a separate group of fish, a 6-month evaluation of survival and growth was conducted from Nov 2012 – May 2013. Sub-adult white sturgeon (n = 10 per treatment) were randomly assigned to one of three groups representing a control and two independent treatment groups. In the first treatment group (notch removal), a 2-4 cm section of the pectoral fin ray was removed near the point of articulation of the joint using a mini-hacksaw and knife as described in Schueller and Peterson (2010). In the second treatment group (full removal), the fin ray was similarly sampled, except that the entire marginal fin ray was removed from the point of articulation down to the terminal end of the ray as described by Koch et al. (2008). In the third group (control), fish were handled in exactly the same manner as the treatment groups, except that no fin ray samples were collected. Fish were tagged with one of three different colored floy tags to facilitate visual identification of treatment groups. Fish handling and sampling of pectoral fin rays were conducted on the same day. During this process, each individual fish was
dip-netted from the raceway and placed into lateral recumbency within a V-shaped restraining board. Water was then pumped through a plastic tube (3/4 cm diameter) into the fish's mouth to supply a continuous flow of water over the gills during the procedure. Regardless of method, fin ray samples were always obtained from right pectoral fin. Control fish were put through a sham operation to minimize potential bias associated with fish handling. The initial weight, fork length (FL), and PIT tag number of each fish were recorded immediately after fin ray sampling, and just prior to releasing the fish back into the raceway. Final weight and length data were recorded 6-month after the initial sampling date to facilitate calculations of relative growth of individuals by using the equation described by Busaker et al. (1990):

$$RG = [(D_1-D_2)/D_2] \times 100$$
 Equation 4

Where:

RG = relative growth D_1 = final length or weight D_2 = initial length or weight

Data Analysis

Analysis of variance (ANOVA) was used to identify significant effects of fin ray sampling on the survival, growth, and 10-min CSHS of sub-adult white sturgeon. Average relative growth in FL and weight was compared among fin ray removal treatments by ANOVA to detect significant differences. Survival was assessed daily and mean survival among treatments was compared at the end of the study using ANOVA. The effect of different fin ray removal treatments on swimming performance was analyzed by comparing 10-min CSHS among treatment groups by using ANOVA. A one-way repeated measures ANOVA was used to identify treatment and velocity effects on behavioral parameters (i.e., TSH, TBF, and GBF) with velocity as the within-subjects factor. Linear regression analyses were used to quantify correlations between CSHS and predictor variables (i.e., water temperature, DO %, and fork length). Significant differences were further analyzed using Tukey's test. All statistical analyses were performed in SAS 9.1 (SAS Institute, Cary, NC) and all test of significance were conducted at $\alpha = 0.05$. All data met the assumptions of normality and homoscedasticity (Shapiro-Wilk test).

Results

Swimming Performance Study

Swimming trials were completed on 45 sub-adult white sturgeon (n = 15 per treatment group). There were no significant differences in water quality or fish size among treatment groups (Table 2-1) and CSHS was not significantly related to water quality or fish size (Table 2-2). Nine fish had cross-sectional areas that were >10% of the cross-sectional area of the swim were corrected for solid blocking. Of these nine fish, one was a control, three were from the notch removal group,, and five were from the full removal group. Corrected 10-min CSHS values from these fish averaged 11.0 ± 0.2 % greater than the original values. These fish were not included in further analysis of TSH, TBF, or GBF because velocities they experienced were different from velocities experienced by smaller fish in the experiment. The control group had the highest mean 10-min CSHS (115.0 ± 3.5 cm/s) while the notch removal group was 110.0 ± 2.6 cm/s. Mean 10-min CSHS did not differ significantly among treatments (F_{2,44} = 1.58; P = 0.22; Figure 2-1).

Hunkering, skimming, and free swimming behaviors were observed in all three treatment groups. At low velocities (<60 cm/s), sturgeon in all treatments spent a majority (> 50%) of time

hunkering. However, as velocity increased, their ability to hunker on the curved tunnel bottom was reduced and sturgeon steadily increased their active swimming behaviors (i.e. skimming or free swimming; Figure 2-2). There was no significant interaction between velocity and treatment effect ($F_{12,198} = 1.43$; P = 0.15) on TSH, and there was no significant treatment effect on TSH for fish swimming at the same velocity ($F_{2,33} = 0.17$; P = 0.84). Tail beats, which were observed only when fish were actively swimming, ranged from approximately 6 – 8 beats/min at 10 cm/s to 127 – 133 beats/min at 110 cm/s (Figure 2-3). Tail beat frequency was closely correlated with water velocity ($R^2 = 0.96$; df = 1, 17; P < 0.0001). Linear regression between TBF and water velocity was analyzed only for velocities > 60 cm/s in which TBF counts were measurable;

TBF = 2.15(velocity) - 114.2

There was no significant interaction between velocity and treatment on the TBF of fish $(F_{12, 198} = 0.84; P = 0.61)$, and there was no significant treatment effect on TBF of fish swimming at the same velocity $(F_{2,33} = 0.84; P = 0.44)$. Tail beat frequency and TSH were not compared among treatment groups at velocities > 100 cm/s because of reduced sample sizes (missing data cells resulting from individual fish not able to swim at higher velocities).

Sturgeon typically switched to ram ventilation when water flow was engaged, thus gill beat frequencies were determined during the resting period immediately before and after each velocity increment. There was no significant interaction between current velocity and treatment on GBF prior-to ($F_{12, 198} = 0.42$; P = 0.96) and post-swimming ($F_{12, 198} = 0.78$; P = 0.67). Gill beat frequencies were not significantly different among treatment groups swimming at the same velocities (Figure 2-4). Mean difference in GBF before and after swim bouts for the control group was 1 beat/30-sec (range: -1 - 2), and 0 beat/30-sec for the notch removal group (range: -1 - 2) and full removal group (range: 0 - 1). Gill beat frequencies were not compared among

treatment groups at velocities > 100 cm/s because of reduced sample sizes (missing data cells resulting from individual fish not able to swim at higher velocities).

Anomalous swimming behaviors were observed in three of the full removal treatment fish. This behavior appeared to be caused by a temporary loss of horizontal swimming orientation during free swimming and was observed only at velocities > 80 cm/s. A sturgeon was considered to have lost its horizontal orientation if the entire body of the fish rolled left or right at an approximately 90° angle. Sturgeon that displayed this behavior would repeatedly swim on their sides before recovering. The mean (range) duration of this loss of horizontal axis was 12-sec (4 – 26 sec); however one fish exhibited this behavior for up to 101-sec at 90 cm/s (<17% of total time swam). The mean 10-min CSHS of these three fish was 110 cm/s (107 – 113 cm/s). These fish also exhibited typical swimming behaviors including hunkering and substrate skimming.

Survival and Growth Study

There was an overall increase in relative growth of sturgeon for each treatment group (Figure2-5) although mean relative increases in FL ($F_{2,29} = 1.30$; P = 0.29) and weight ($F_{2,29} = 0.38$; P = 0.69) did not differ significantly among treatments. Mean (± SE) relative growth in FL was 2.4 ± 0.6 % for the control group, 2.8 ± 0.6 % for notch removal group, and 3.5 ± 0.4 % for full removal group. Mean (± SE) relative weight increased by 7.6 ± 3.6 % for the control group, 8.7 ± 3.3 % for the notch removal group, and 12.3 ± 5.1% for the full removal group. Pooled relative increases in FL among treatments ranged from 0.6% to 6.7% of initial length. Pooled relative growth in weight for all treatments ranged from -10.26% to 37.61% of initial weight.

Within a week after initial fin ray sampling, fish in the notch and full removal treatment groups developed fungal growth where the fin ray was sampled. However, fish were left un-

treated to simulate natural conditions as best as possible. Fungal growth subsided after approximately two weeks and no mortality was observed in any treatment. Sites of fin ray biopsies 6-month after initial sampling appeared to be completely healed in all fish, regardless of sampling method. In the notch removal group a close inspection was sometimes necessary to determine where the ray had actually been biopsied. Fish subjected to the notch treatment suffered little bleeding during the sampling procedure. In contrast, all fish from the full removal group exhibited red scar tissue at the biopsy site. During sampling of the full fin ray, bleeding was observed in all fish, although the amount varied among individuals.

Discussion

My results showed that pectoral fin ray removal had no significant effect on swimming performance, survival or growth of sub-adult white sturgeon. These finding corroborate those of previous studies by Collins and Smith (1996) and Parsons et al. (2003) for other sturgeon species. Although my conclusions contradict those of Kohlhorst (1979), this author acknowledged that his survival estimates were based on a small number of recaptured white sturgeon (3.6%). Furthermore, because his study was conducted on wild fish in their natural habitat, many other environmental factors may have influenced the results. Regardless, data from the controlled experiments presented in this study clearly showed that fin ray biopsies have no detectable deleterious effects on white sturgeon, at least within a captive environment.

Although I could detect no significant differences in swimming performance among treatment groups and controls in my study, anomalous swimming behaviors in the full removal treatment group suggest that notch removal method should be adopted as the standard practice for obtaining fin ray biopsies of wild sturgeon. Although the proximate mechanisms that resulted in the temporary loss of horizontal swimming orientation observed in the full removal

treatment group are unclear, I suspect that complete removal of the pectoral fin ray may alter the hydrodynamics of the leading edge of the pectoral fin – particularly at higher current velocities. Furthermore, the handling time required for the notch removal biopsy was typically less than half that required for the full removal biopsy (approximately 15-sec versus 45 sec). Finally, the notch removal biopsy appeared to cause less damage to the pectoral fin (based on the subsequent formation of scar tissue) and appeared to heal more quickly and completely than biopsies obtained using the full ray removal method.

Results from this study provide new information on the swimming speeds of sub-adult white sturgeon. Previous studies have only provided data on swimming performance of small (80-100 mm) juvenile (Boysen and Hoover 2011) and large (143–164 cm TL) adult white sturgeon (Cheong et al. 2006). The use of CSHS in this study is analogous to critical swimming speeds (U_{crit}), which has been used in previous swim trial studies of sturgeon. The 10-min CSHS speeds reported in this study are comparable to 10-min U_{crit} speeds observed by similarly sized lake sturgeon. Pooled data for all fish in this study showed that at a mean FL of 97.4 ± 1.4 cm, the mean 10-min CSHS of white sturgeon was 111 ± 2.8 cm/s, at water temperatures of $13.4 \pm$ 0.1 °C. These data are consistent with swimming performance of lake sturgeon (117 - 122 cm)TL) which, under similar conditions, are expected to be able to swim for 10-min at a current velocity of 130 cm/s (Peake et al. 1995, 1997). Findings from this study are consistent with data collected by Parsley et al. (1993) who observed wild adult white sturgeon spawning on the lower Columbia River in current velocities of 210 cm/s. Considering that fish tested in this study were sub-adults, the results of my experiment appear biologically realistic compared to the swimming speeds typical of wild fish.

Previous studies have shown that white sturgeon are more susceptible to handling stress than other sturgeon species (Barton et al. 2000). Belanger et al. (2001) found physiological stress responses of white sturgeon exposed to handling were more extensive and more easily induced than those of other sturgeon species. Quantifying the primary responses (i.e. cortisol and lactate levels) of handling stress is an effective method of evaluating the physiological response of white sturgeon subjected to fin ray sampling; however, in this study I was only interested in the tertiary responses (e.g. growth, survival, and behavioral effects) as an indicator of how sub-lethal effects of fin ray biopsy could affect long-term survival in the wild sturgeons. As such, I quantified survival and growth, swimming performance, and behavioral parameters as a means of comparing a sturgeon's overall condition. The experiment also provided new insights into the subtle changes in swimming behavior that may result from fin ray sampling.

Although previous studies have shown that white sturgeon are more responsive to acute stressors than other sturgeon species, the results of my study corroborates with previous studies, which suggest that their responses are still relatively minor compared to those typically reported for teleostean fishes (Barton et al. 2000; Belanger et al. 2001; Barton 2002). Previous studies have shown that sturgeons typically employ different swimming behaviors at different water velocities, allowing them to effectively maneuver or maintain position at high water velocities (Adams et al. 1997; Adams et al. 1999; Adams et al. 2003; Hoover et al. 2011). Undoubtedly, these abilities are especially important to white sturgeon, which spend much of their life cycle in large swift flowing rivers. Regardless of species, all sturgeons have an inherent ability to maintain station when exposed to current by either hunkering or skimming along the substrate - behaviors that allow them make them highly efficient benthic foragers (Hoover et al. 2011).

These behaviors, presumably, would also allow the fish to rest after the physical exertion of handling by maintaining station along the substrate.

Fish tested in this study were hatchery-reared and have never been exposed to high flow conditions. Consequently, swimming performance of fish determined in this study may underestimate the swimming performance of similar sized wild white sturgeon, and therefore my ability to detect significant differences in treatments could have been slightly diminished. Likewise, growth rates of sturgeon observed in this study are also not typical of those typically reported for wild fish which typically endure a much harsher environment than captive fish. For these reasons, I suggest that future studies of wild fish be conducted *in vivo* to corroborate the conclusions of this study.

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Table 2-1. Means (±SE) of key experimental variables and critical-station holding speeds from
swimming performance trials of sub-adult white sturgeon (n = 15 per treatment) subjected to
different fin ray removal methods. Water quality and fish size were not significantly different
among treatment groups.

	Treatments			
	Control	Notch	Full	P-value
Water Temperature, ^o C	13.2 (0.2)	13.5 (0.3)	13.5 (0.3)	0.69
DO, %	86.9 (1.2)	87.2 (0.6)	87.1 (0.7)	0.96
Fork length, cm	95.5 (1.6)	98.6 (1.4)	98.0 (1.2)	0.26
Weight, kg	7.1 (0.4)	7.8 (0.4)	7.2 (0.3)	0.38
10-min CSHS, cm s ⁻¹	115.0 (3.5)	108.0 (2.3)	110.0 (2.6)	0.22

Variable	R ²	F - value	df	P - value		
Water Temperature						
Control	0.003	0.04	1, 14	0.83		
Notch removal treatment	0.040	0.54	1, 14	0.48		
Full removal treatment	0.029	0.39	1, 14	0.54		
Water DO %						
Control	0.033	0.45	1, 14	0.51		
Notch removal treatment	0.133	2.00	1, 14	0.18		
Full removal treatment	0.007	0.09	1, 14	0.76		
Fork Length						
Control	0.123	1.83	1, 14	0.20		
Notch removal treatment	0.317	3.03	1, 14	0.28		
Full removal treatment	0.124	3.88	1, 14	0.07		

Table 2-2. Linear regression analyses of water temperature, dissolved oxygen (DO), and fork length on the 10-min CSHS of sub-adult white sturgeon subjected to different pectoral fin ray sampling methods.



Figure 2-1. Mean 10-min critical-station holding speeds (CSHS) of sub-adult white sturgeon subjected to different methods of fin ray sampling. Mean 10-min CSHS did not differ significantly among treatments ($F_{2,44} = 1.58$; P = 0.22; Figure 2-2). Numbers within each bar represent the mean 10-min CSHS for the treatment. Error bars represent ± 1 SE.



Figure 2-2. Mean percent time spent hunkering (TSH) across a range of velocities for sub-adult white sturgeon subjected to different fin ray sampling methods. There was no significant effect of treatment, or an interaction between treatment and velocity on TSH. Numbers above each bar represent sample sizes. Error bars represent ± 1 SE.



Figure 2-3. Mean tail beat frequency (TBF) regressed against swimming velocity of sub-adult white sturgeon subjected to different fin ray sampling methods. There was no significant effect of treatment, or an interaction between treatment and velocity on TBF. Linear regression was not analyzed at velocities ≤ 40 cm/s because of low TBF counts.



Figure 2-4. Mean gill beat frequency across a range of velocities for sub-adult white sturgeon subjected to different fin ray sampling methods. Gill beats were recorded prior to the fish swimming at each velocity and immediately after. There was no significant effect of treatment, or an interaction between treatment and velocity on GBF. Error bars represent ± 1 SE.



Figure 2-5. Mean relative growth in fork length (FL) and weight of hatchery-reared sub-adult white sturgeon over a 6-month period from different treatment groups. Mean relative increases in FL ($F_{2,29} = 1.30$; P = 0.29) and weight ($F_{2,29} = 0.38$; P = 0.69) did not differ significantly among treatments. Error bars represent ± 1 SE.

CHAPTER 3

EFFECTS OF FIN RAY REMOVAL ON THE SWIMMING PERFORMANCE OF SIBERIAN

STURGEON (ACIPENSER BAERRI)²

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Abstract

Sturgeon are most commonly aged by analysis of their pectoral fin rays, which requires the removal of the ray. However, there are concerns associated with the post-release survival of fish subjected to fin ray removal. The effects of two fin ray sampling methods on growth, survival, and swimming performance were evaluated for hatchery-reared Siberian sturgeon. Fish were subjected to either a notch removal treatment in which a small notch was removed from the marginal pectoral fin ray, or a full removal treatment in which the entire marginal pectoral fin ray was removed. Control fish were not subjected to either of the fin ray sampling methods, but they were handled in the same manner to ensure that there were no biases related to stress associated with fish handling. A modified 3,230-L Brett-type swim tunnel was used to evaluate the 10-min critical station-holding speeds (CSHS) and behavioral characteristics (i.e. time spent hunkering, tail beat frequency, and gill beat frequency) associated with swimming. Water quality and fish size were comparable among treatments. Analysis of variance indicated that fin ray sampling had no significant effect on the 10-min CSHS of Siberian sturgeon. The 10-min CSHS (mean \pm SE) were 113 \pm 3.4 cm/s, 109 \pm 2.5 cm/s, and 111 \pm 2.8 cm/s for the notch removal treatment, full removal treatment, and control treatment, respectively. Time spent hunkering, tail beat frequency, and gill beat frequency were also not significantly different among treatments. Results indicate that both fin ray sampling methods have a negligible effect on the swimming performance of Siberian sturgeon. However, the full removal treatment required greater handling time and was much more physically invasive, thus only the notch removal method is recommended for use in fin ray sampling.

Introduction

Siberian sturgeon (Acipenser baerii) are widely distributed throughout all the rivers in Siberia, with the most notable populations occurring in Lake Baikal, and the Ob, Aldan, and Lena River systems (Billard and Lecointre 2001). There are currently three recognized subspecies of Siberian sturgeon: A. b. baerii, which occurs mainly in the Ob River Basin, and A. b. baicalensis and A. b. stenorhynchus, which occur in the Eastern Siberian River basins (Ruban 1997). Populations of Siberian sturgeon throughout the species' range have suffered dramatic declines within the last half century as a result of overfishing and habitat loss (Ruban and Zhu 2010). In the Ob River, up to 40% of the spawning grounds are no longer accessible to migrating adults as a result of dam construction (Ruban 1997). Both A. b. baerii and A. b. stenorhynchus have declined because of overfishing. The remaining sub-species, A. b. baicalensis is currently listed as endangered on the Red Data Book of the Russian Federation (Kolosov 1983; Ruban 1997). All populations of Siberian sturgeon, especially those in the Ob and Kolyma rivers, have exhibited reproductive abnormalities, which have been attributed to excessive pollution (Ruban and Akimova 1991; Akimova and Ruban 1993; Ruban and Akimova 1993; Ruban and Zhu 2010). Unfortunately, stock assessment data for remaining populations are lacking.

Accurate age information is needed to better understand the population dynamics of threatened or endangered sturgeon populations and for developing stock assessment models, which are necessary for effective management of these stocks (Beamish and McFarlane 1983; Campana 2001; Paragamian and Beamesderfer 2003). To obtain age information without lethal sacrifice of wild sturgeon, researchers typically analyze cross-sections of the pectoral fin ray to quantify annual growth rings (Cuerrier 1951; Rien and Beamesderfer 1994; Rien et al. 1994).

The most commonly used fin ray sampling methods require either the complete removal of the marginal pectoral fin ray (Cuerrier 1951; Brennan and Caillet 1989; Collins and Smith 1996; Paragamian and Beamesderfer 2003; Hurley et al. 2004; Koch et al. 2008), or the collection of a fin ray biopsy (aka: the "notch removal" method; Brennan and Cailliet 1989; Peterson et al. 2002). Regardless of method, some researchers have expressed concerns about the potential effects on swimming performance of sturgeons subjected to fin ray sampling (Kohlhorst 1979; Collins and Smith 1996; Parsons et al. 2003). As a benthic cruiser, sturgeons use their pectoral fins for in maneuvering, swimming, station-holding, and benthic shuffling (Findeis 1997; Wilga and Lauder 1999). Consequently, any mutilation of the pectoral fin ray may potentially alter the structural integrity of the pectoral fin, possibly affecting swimming performance and long-term health of fish subjected to fin ray sampling.

Few studies have evaluated the effects of fin ray removal on sturgeon in a controlled study. Although many studies have evaluated and quantified critical swimming speeds (U_{crit}) and swimming endurance of shovelnose sturgeon (*Scaphirhynchus platorynchus;* Adams et al. 1997; Adams et al. 2003; Hoover et al. 2008), pallid sturgeon (*Scaphirhynchus albus;* Adams et al. 1999) and lake sturgeon (*Acipenser fulvescens*; Peake et al. 1995), only Parsons et al. (2003) specifically evaluated the effects of fin ray removal on the swimming performance of sturgeon. Parsons et al. (2003) concluded that full fin ray removal has no significant effect on the swimming performance of wild adult shovelnose sturgeon. In a previous mark-recapture study of white sturgeon in the Sacramento-San Joaquin Estuary, Kohlhorst (1979) suggested that fin ray removal caused mortality in white sturgeon (*Acipenser transmontanus*) subjected to full fin ray removal (Kohlhorst 1979). Unfortunately, controlled laboratory evaluations of the effects of fin ray sampling on Acipenserid swimming performance are rare. Although wild populations of

Siberian sturgeon are listed as endangered, the species is readily available through commercial propagation and, hence, is an effective surrogate for wild Acipenserid species (Williot et al. 2001). The objectives of this study were to assess the effects of two different fin ray sampling methods on the swimming performance of hatchery-reared Siberian sturgeon.

Methods

Experimental Fish & Fish Care

Hatchery-reared juvenile Siberian sturgeon were obtained from the UGA Cohutta Fish Hatchery in Cohutta, Georgia. Fish were kept in a 42,500-L raceway supplied with fresh spring water at a constant rate of 95 L/min. All fish used in the experiment were tagged with passiveintegrated-transponder (PIT) tags to facilitate individual identification. Temperature and dissolved oxygen in the raceway was monitored daily with a portable multi-meter (YSI® 55; Yellow Springs Instruments, Inc., Yellow Springs, Ohio). Fish were fed a diet of size 6-mm Skretting sinking trout pellets (44% protein content, 28% oil content) at a rate of 2% of total fish biomass per day.

Swimming Performance Study

A total of 45 age-3 Siberian sturgeon (n = 15 per treatment) were randomly assigned to one of three experimental groups consisting of a control and two independent treatment groups. In the first treatment group (notch removal), a 2-4 cm section of the pectoral fin ray was removed near the point of articulation of the joint using a mini-hacksaw and knife as described in Schueller and Peterson (2010). In the second treatment group (full removal), the fin ray was similarly sampled, except that the entire marginal fin ray was removed from the point of articulation down to the terminal end of the ray as described by Koch et al. (2008). In the third

group (control), fish were handled in the same manner as the treatment groups, except that no fin ray samples were collected.

Swim trials were conducted between Jan 6, 2013 – Feb 3, 2013 by using in a speciallydesigned, mobile 3,230-L (total water volume) Brett-type (Brett 1964) swim flume constructed by the U.S. Corps of Engineers in Vicksburg, MS. The flume measured 244 cm x 91 cm x 91 cm (L x H x W) and was capable of maintaining boundary-layer or rectilinear flow with the use of different inserts. For this study, a cylindrical tube insert, measuring 46 cm in diameter and 150 cm in length was placed in the swim tunnel to induce and maintain rectilinear flow throughout the experiment. Acrylic grids were placed at both ends of the tunnel to entrain the fish within the swim chamber of tunnel. The cathode of a pulsed DC electro-fisher (Advanced Backpack AbP-3) was attached to a third grid which was positioned 61-cm behind the posterior end of the tube insert. During the trials, a localized electric current was used ad libitum to prevent fish from resting against the rear grid.

Current velocities within the tube insert were confirmed by placing the probes from two electronic flow meters (Marsh-McBirney Flo-Mate 2000) into the center of the tube insert. Water within the flume was changed daily and temperature and dissolved oxygen within the flume were monitored with a portable multi-meter (YSI 55 multi-meter). Each swim trial was video-recorded (Sony DCR-SX45) to facilitate subsequent evaluation of behavioral effects.

Immediately before each trial, a single fish was randomly selected from the raceway and assigned to one of the three previously described treatments. All fish were handled identically (except for the specific fin ray sampling treatment) after which they were immediately introduced into the swim tunnel where they were allowed to acclimate for 10-min at a base flow velocity of 10 cm/s. Acclimation velocities were then incrementally increased to 20, 40, 60, and

80 cm/s at 10-min intervals with a 1-min rest between each increment. The range of velocity increments used for the acclimation period were selected based on pre-experimental trials that showed that the fish could maintain station at velocities between 10-80 cm/s with little effort. Upon completion of the acclimation period, fish were allowed to rest for 10-min after which water velocity was increased to 90 cm/s. Water velocity was then increased incrementally by 10 cm/s every 10-min, with a 10-min rest between each successive increment. The 10-min critical station-holding speed (CSHS), which is the maximum speed at which a fish can maintain station, was determined from the maximum velocity at which the fish reached fatigue (the point at which the fish could no longer maintain position). Critical station-holding speed was then calculated as described by Parsons et al. (2003) by using the equation:

$$CSHS = U_1 + [U_2 (T_1/T_2)]$$
 Equation 2

Where:

CSHS = speed at which a fish can maintain station, either by swimming or adherence to the bottom, for a prescribed period of time

 U_1 = highest velocity maintained for the entire interval (cm/s)

 U_2 = velocity increment (cm/s)

 T_1 = time elapsed at fatigue velocity (min)

 T_2 = the prescribed interval time (min)

The weight (kg), fork length (cm), and PIT tag number of each fish was recorded after each swim trial. All fish were swum only once to minimize any potential bias associated with training or habituation. Critical-station holding speeds were not corrected for solid blocking as recommended by Bell and Terhune (1970) because all fish had cross-sectional areas <10% of the cross-sectional area of the tunnel.

In addition to measuring CSHS, I also quantified three behavioral parameters: time spent hunkering (TSH), tail beat frequency (TBF), and gill beat frequency (GBF) from video recordings. A fish was determined to be hunkering if it was able to maintain station by adhering to the bottom of the tunnel without body or caudal fin undulation. The amount of time a fish spent hunkering was recorded with a stopwatch for each velocity increment. The total TSH at each velocity for all fish within a treatment was then averaged to obtain a mean TSH for that treatment. The TSH for each treatment group was expressed as a percent of total time swam at each velocity. Tail beat frequency (TBF) was determined by using the method described by Parsons et al. (2003) whereby rate of tail beating was estimated by counting the number of beats over a 5-s period when the fish is actively swimming. Estimates of TBF were repeated five times for each individual fish at each velocity. Those values were then averaged to obtain an average TBF for each fish at each specific velocity. TBF values for all fish within a treatment were then averaged to obtain the average TBF at each velocity, for each treatment. The mean values were then multiplied by 12 to obtain mean beats/min. Gill beat frequencies (GBF) were recorded for 30-sec before and immediately after each velocity increment. Any anomalous behaviors were also noted.

Data Analysis

Analysis of variance (ANOVA) was used to identify significant effects of fin ray sampling methods on the 10-min CSHS of Siberian sturgeon. One-way repeated measures ANOVA was used to identify treatment and velocity effects on behavioral parameters (i.e., TSH, TBF, and GBF) with velocity as the within-subjects factor. Linear regression analyses were used to quantify correlations between CSHS and predictor variables (i.e., water temperature, DO %, and fork length). Significant differences were further analyzed using Tukey's test. All statistical

analyses were performed in SAS 9.1 (SAS Institute, Cary, NC) and all tests of significance were conducted at $\alpha = 0.05$. All data met the assumptions of normality and homoscedasticity (Shapiro-Wilk test).

Results

ANOVA results showed that there were no significant differences in water quality or fish size among treatment groups (Table 3-1). Likewise, critical-station holding speeds were not significantly related to water quality or fish size for any treatment group (Table 3-2). The notch removal treatment group had the highest mean 10-min CSHS (113.0 ± 3.4 cm/s) while the full removal treatment group had the lowest mean 10-min CSHS (109.0 ± 2.5 cm/s). Mean 10-min CSHS for the control treatment was 111.0 ± 2.8 cm/s. Mean 10-min CSHS did not differ significantly among treatment groups ($F_{2, 42} = 0.55$; P = 0.58; Figure 3-1).

Typical swimming behaviors (i.e. hunkering, skimming, and free swimming) were routinely observed in all three treatment groups, which facilitated evaluations of TSH and TBF. No abnormal swimming behaviors were observed in any treatments, although most fish typically spent a majority (> 50%) of the time thrashing about in the tunnel at 10cm/s. At acclimation velocities of 20 and 40 cm/s however, sturgeon in all treatments began to exhibit a higher percentage of hunkering behavior (50 - 68 %). As velocity increased, their ability to adhere to the curved tunnel bottom was reduced, such that the fish in all three treatments spent a majority of time actively swimming at velocities > 60 cm/s (Figure 3-2). There was no significant treatment effect on TSH ($F_{2, 42} = 0.80$; P = 0.45), and the interaction between velocity and treatment on TSH was not significant ($F_{12, 252} = 0.40$; P = 0.96). Mean TBF ranged from approximately 8 – 17 beats/min at 10 cm/s to 198 beats/min at 140 cm/s (Figure 3-3). Tail beat

frequency had a strong positive relationship with increasing current velocity ($R^2 = 0.97$; df = 1, 31; P = 0.01):

$$TBF = 1.59$$
 (velocity) - 11.68

There was no significant effect of treatment ($F_{2, 42} = 0.24$; P = 0.78) on TBF and the interaction between velocity and treatment on the TBF was not significant for fish swimming at the same velocity ($F_{12, 252} = 1.46$; P = 0.14). Tail beat frequency and TSH was not compared among treatment groups at velocities > 100 cm/s because of reduced sample sizes (missing data cells resulting from individual fish not able to swim at higher velocities).

All sturgeon switched to ram ventilation when current was induced inside the tunnel. There was no significant interaction between velocity and treatment ($F_{12, 252} = 0.51$; P = 0.91) and there was no significant effects of treatment on GBF prior to ($F_{2, 42} = 0.06$; P = 0.94) or after swimming ($F_{12, 252} = 0.83$; P = 0.62) for fish swimming at the same velocities (Figure 3-4). Mean differences in GBF before and after swim bouts for all three treatments were 1 beat/30-sec (range: -1 - 5). Gill beat frequencies were not compared among treatment groups at velocities > 100 cm/s because of reduced sample sizes (missing data cells resulting from individual fish not able to swim at higher velocities).

Discussion

The results from my study that pectoral fin ray removal has no significant effect on the swimming performance, and hence the survival of Siberian sturgeon, corroborate the findings of Collins and Smith (1996) and Parsons et al. (2003) for other species of sturgeons. Findings from this study do contrast those of Kohlhorst (1979) who determined that pectoral fin ray removal caused significant mortality in white sturgeon. However, Kohlhorst (1979) acknowledged an important caveat of his study, which was that his survival estimates were based on a small return

sample size (3.6%) of tagged individuals. Furthermore, because the study was conducted on wild fish in their natural habitat, many other environmental factors may have influenced the results. Regardless, data from the controlled experiments presented in this study clearly show that fin ray biopsies have no detectable deleterious effects on Siberian sturgeon in a captive environment.

Although fin ray sampling methods evaluated in this study do not significantly affect the swimming performance of Siberian sturgeon, I do suggest that only the notch sampling method be used. Evidence from previous studies that sturgeon subjected to the notch removal method will always regenerate the missing section whereas sturgeon subjected to the full removal method may not regenerate the ray (Peterson, unpublished data). The handling time required for the notch removal method was typically less than half that required for the full removal method (approximately 15-sec versus 45 sec). Finally, the notch removal method appeared to cause less damage to the pectoral fin (based on the subsequent formation of scar tissue) and appeared to heal more quickly and completely than biopsies obtained using the full ray removal method.

Swimming speeds determined in this study were similar to those obtained by Qu et al. (2013) who reported a mean 10-min critical swimming speed of 106 ± 2.2 cm/s for Siberian sturgeon measuring 61.8 ± 3.1 cm TL. In comparison, the pooled 10-min CSHS for fish in this study was 111 ± 1.7 cm/s for fish measuring 74.5 ± 0.6 cm FL. Although fish used in my study were slightly larger than those used by Qu et al. (2013), water temperatures in my study were approximately 10° C colder that those used by Qu et al. (2013). Higher water temperatures may explain why swimming speeds of the smaller fish used by Qu et al. (2013) were comparable to those of the larger fish used in my trials.

Swimming performance of sturgeons is typically reported to be relative poor, compared to similarly size teleost species (Peake et al. 1995; Lee et al. 2003). Relatively poor swimming performance is generally attributed to their unique combination of physiological and morphological characteristics, which includes a slower metabolism (Singer et al. 1990), a heterocercal tail which sacrifices thrust for dynamic lift, and a high drag coefficient incurred by their protective bony scutes and rough skin (Webb 1986). Interestingly, studies have also shown that sturgeons are able to compensate for this disadvantage by employing a variety of specialized swimming behaviors which they use under specific conditions (Adams et al. 2003; Hoover et al. 2011). For example, most sturgeon species have an inherent ability to maintain station in flowing water by either by hunkering or skimming along the substrate bottom. Both of these energy conserving behaviors are aided by their flat ventral surface, large pectoral fins, and pointed and flattened rostrums (Adams et al. 2003). As such, these same behaviors should, presumably, allow sturgeon to efficiently maintain station in along a riverine substrate even after the extreme physical exertion caused by capture and handling.

A lack of a significant effect from fin ray sampling on the swimming performance of Siberian sturgeon is likely attributable to the fact that sturgeon are generally are more resistance to stress compared to other teleostean fishes. Sturgeons exhibit low physiological responses to management stressors such as handling and transportation (Barton et al. 2000; Belanger et al. 2001). Low production of stress-related chemicals (e.g. plasma cortisol) after acute disturbances suggests that sturgeon may be better adapted to handling stress than other species of fishes (Barton et al. 2000). Although evaluating levels of stress-related chemicals is an effective method to determine physiological responses of sturgeons to fin ray sampling, in this study, I

was only interested in the tertiary responses (i.e. swimming performance and behavioral characteristics) of sturgeon subjected to fin ray removal.

Fish tested in this study are hatchery-reared fish that have never been exposed to high flow conditions. Consequently, swimming performance of fish determined in this study may underestimate the swimming performance of similar sized wild sturgeon, and therefore the ability to detect a difference in treatments could have been diminished. For these reasons, I suggest that future studies of wild fish be conducted *in vivo* to corroborate my conclusions.

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	Treatments			
_	Control	Notch	Full	P-value
Water Temperature, °C	13.2 (1.4)	13.9 (1.4)	13.6 (0.3)	0.92
DO, %	86.3 (2.6)	86.1 (3.8)	85.7 (0.7)	0.88
Fork length, cm	74.4 (4.7)	74.5 (3.2)	74.7 (1.2)	0.97
Weight, kg	2.4 (0.4)	2.3 (0.3)	2.3 (0.3)	0.95
10-min CSHS, cm s ⁻¹	111.0 (2.8)	113.0 (3.4)	109.0 (2.6)	0.58

Table 3-1. Means (SE) of key experimental variables and critical-station holding speeds (CSHS) from swimming performance trials of Siberian sturgeon (n = 15 per treatment) subjected to different fin ray removal methods.

Variable	R ²	F - value	df	P - value
Water Temperature				
Control	0.175	2.76	1, 14	0.12
Notch removal treatment	0.183	2.91	1, 14	0.11
Full removal treatment	0.006	0.09	1, 14	0.77
Water DO %				
Control	0.084	1.19	1, 14	0.29
Notch removal treatment	0.004	0.06	1, 14	0.81
Full removal treatment	0.002	0.03	1, 14	0.87
Fork Length				
Control	0.309	5.81	1, 14	0.30
Notch removal treatment	0.031	0.42	1, 14	0.52
Full removal treatment	0.001	0.01	1, 14	0.91

Table 3-2. Linear regression analyses of water temperature, dissolved oxygen (DO), and fork length on the 10-min CSHS of Siberian sturgeon subjected to different pectoral fin ray sampling methods.



Figure 3-1. Mean 10-min critical-station holding speeds (CSHS) of Siberian sturgeon subjected to different methods of fin ray sampling. Mean 10-min CSHS did not differ significantly among treatments ($F_{2,44} = 0.55$; P = 0.58). Numbers within each bar represent the mean 10-min CSHS for the treatment. Error bars represent ± 1 SE.



Figure 3-2. Mean percent time spent hunkering (TSH) across a range of velocities for Siberian sturgeon subjected to different fin ray sampling methods. There was no significant effect of treatment, or an interaction between treatment and velocity on TSH. Numbers above each bar represent sample sizes. Error bars represent ± 1 SE.



Figure 3-3. Mean tail beat frequency (TBF) regressed against swimming velocity of Siberian sturgeon subjected to different fin ray sampling methods. There was no significant effect of treatment, or an interaction between treatment and velocity on TBF. Error bars represent ± 1 SE.



Figure 3-4. Mean gill beat frequency across a range of velocities for Siberian sturgeon subjected to different fin ray sampling methods. Gill beats were recorded prior to the fish swimming at each velocity and immediately after. There was no significant effect of treatment, or an interaction between treatment and velocity on GBF. Error bars represent ± 1 SE.

Chapter 4

CONCLUSIONS

Results from my study provide evidence that full fin ray removal (Cuerrier 1951) and notch fin ray removal (Brennan and Cailliet 1989) do not significantly affect the swimming performance of white or Siberian sturgeon. However, I do suggest that only the notch sampling method be used, based on numerous observations of wild sturgeon where fin rays sampled using the notch removal method had completely regenerated within 12 months (Peterson, unpublished data). In contrast, similar observations of wild sturgeon fin rays sampled using the full ray removal method have reportedly not regenerated - even after several years. Although anecdotal, these observations further support the conclusions of my laboratory experiment that evaluated the survival and growth white sturgeon subjected to the two fin ray sampling methods. At the end of the 6-month experiment, biopsies obtained using the notch removal had almost completely healed whereas the majority of biopsies obtained using the full removal method were either still inflamed or covered with scar tissue over the entire length of the fin margin. My experience in the lab trials also suggested that the notch removal method was faster and easier, which may help reduce handling time, and ultimately, the amount of stress to the fish. Furthermore, the notch biopsy appeared to be less invasive base on the relative absences of bleeding observed in fish subjected to the notch removal method. I also observed the loss of horizontal swimming orientation in several white sturgeon subjected to the full removal treatment only. Although these behaviors did not appear to persist after the swim trials were concluded, there were clearly important behavioral anomalies during the trials, which were only

observed at velocities > 80 cm/s. Consequently, I concluded that this anomalous swimming behavior was an artifact of an interaction between fish size, water velocity, and the degree of pectoral fin ray mutilation incurred during the full removal method.

My study also provides new information on the swimming performance of white sturgeon, as it is the first to quantify critical swimming performance of sub-adult white sturgeon. The pooled 10-min CSHS speed of $(111 \pm 2.8 \text{ cm/s} \text{ at temperatures of } 13.4 \pm 0.1 \text{ °C})$ fish measuring 97.4 ± 1.4 cm in FL is comparable to previously published endurance curves of lake sturgeon of a similar size. The swimming speeds of white sturgeon in this study also corroborate previously reported swimming speeds of spawning white sturgeon on the lower Columbia River. Parsley et al. (1993) observed adult white sturgeon spawning in the Columbia River at mean water velocities of 210 cm/s. Given the size of the experimental fish evaluated in this study, my results appear to be biologically realistic compared to those documented for wild fish.

Another interesting result of this study was that white sturgeon appear to be weaker swimmers than Siberian sturgeon. Although the pooled 10-min CSHS of white sturgeon (111 \pm 2.8 cm/s) and Siberian sturgeon (111 \pm 1.7 cm/s) were very similar, the white sturgeon were significantly larger which should have enabled them to obtain a significantly higher CSHS. Similar speeds for Siberian sturgeon of smaller sizes have also been reported in a previous study. Qu et al. (2013) reported the mean 10-min U_{crits} of 11 Siberian sturgeon to be 106 \pm 2.2 cm/s for fish measuring 61.8 \pm 3.1 cm in body length swimming at 24.03 \pm 0.32°C. Although Siberian sturgeon tested in Qu et al. (2013) were significantly smaller than those tested in my study, they had comparable 10-min U_{crit} values - probably on account of the higher water temperature used in that study. Better swimming performance observed in Siberian sturgeon may also have been

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caused by their slightly different morphological characteristics which include a more elongated rostrum and thinner more streamlined body.

Finally, the results of this study provide quantified evidence that removal of the pectoral fin ray for age estimation in captive white and Siberian sturgeon has no significant acute or long term effects with regard to survival, growth, or swimming performance. Regardless of fin ray sampling method, I observed no mortalities over the 6-month experiment and all treatment groups had an overall increase in both length and weight. In my swimming performance studies of white and Siberian sturgeon, I observed no significant differences in the swimming performance of fish subjected to two different methods of fin ray removal. Behavioral responses including tail beat frequency (TBF), gill beat frequency (GBF), and time spent hunkering (TSH) were also not significantly different among treatment groups for sturgeon of the same species. I noted that data of GBF were not informative, and hence, should not be used as a behavioral response in future evaluations of sturgeon swimming performance. My results corroborate findings of several previous studies that have evaluated the effects of fin ray removal in sturgeon (Collins and Smith 1996; Parsons et al. 2003). However, because fish tested in my study are from hatchery-reared stocks that may not be representative of the physical fitness of wild fish, I recommend that further experiments evaluating the effects of fin ray removal be conducted on wild sturgeon to ensure that the method does not negatively affect survival in vivo.

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