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Evaluating Growth, Survival and Swimming Performance to Determine the Feasibility of Telemetry for Age-0 Pallid Sturgeon (*Scaphirhynchus albus*)

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ABSTRACT.—Telemetry is valuable for understanding animal ecology and assessing conservation priorities. Sturgeon species worldwide are imperiled and telemetric methods have been applied to adults, but the feasibility of using this methodology on age-0 sturgeon remains unclear. The pallid sturgeon (*Scaphirhynchus albus*) is a federally endangered species in the central United States and little is known about its early life ecology. The use of telemetry on age-0 pallid sturgeon would greatly increase understanding of their early life ecology. We assessed growth, survival, and swimming performance of age-0 pallid sturgeon tagged with 0.2 g, nonfunctioning radio telemetry tags to determine whether radio telemetry can be used on such small sturgeon. Tags were surgically implanted internally and attached externally, with a control group that only experienced handling. Age-0 sturgeon with internally implanted tags grew slowly and had low survival, while counterparts within the control group and those with external tags grew faster and had almost 100% survival. No differences in critical swimming speed occurred among the internally tagged, externally tagged, and control fish. We suggest externally tagging age-0 sturgeon may be possible. However, a small telemetry transmitter that uses technology (*e.g.*, ultrasonic) which transmits well in deep rivers while maintaining a minimum tag weight and maximizing battery life is needed.

INTRODUCTION

Telemetry is one of the most widely used methods for examining ecological patterns of animals (Trefethen, 1956; Winter, 1996). Radio and ultrasonic transmitters are commonly used to monitor animal location, movement, behavior, and physiology (*e.g.*, Winter, 1996); but trade-offs exist for different tag types, particularly in aquatic ecosystems. For example radio telemetry transmitters are useful for searching large water bodies, can be detected in areas with high flows and abundant aquatic vegetation, and their receivers are highly mobile (Nielsen, 1992; Winter, 1996). However, these transmitters can encounter problems with excessive turbidity, high salinity, and deep water (Kenward, 2001). Ultrasonic tags have primarily been used for large fishes, but the size of these transmitters often limits their applicability to larval and juvenile fishes (Nielsen, 1992; Winter, 1996).

Transmitter placement (*i.e.*, internal or external) will influence efficacy and should be considered when using telemetry. External tags have an increased risk of abrasion and tag loss due to entanglement (Nielsen, 1992; Bridger and Booth, 2003). Internally implanted tags are protected from environmental entanglement and often placed near a fish's center

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of gravity, further minimizing balance concerns (Bridger and Booth, 2003). However, internal transmitters require longer surgical and recovery procedures and may also increase the risk of infection to the fish over external tags, as externally attached transmitters can be applied quickly with minimal fish handling.

Telemetry has been a valuable conservation tool for identifying adult sturgeon movement (McElroy *et al.*, 2012) and habitat use (Koch *et al.*, 2012), but its applicability to young-of-year (age-0) sturgeon remains unclear. Many fisheries scientists follow the '2% rule' and only apply transmitters weighing less than 2% of a fish's weight (Winter, 1996). This widely accepted recommendation and the current limitations of technology (*i.e.*, tag size) have restricted telemetry-based studies of larval and juvenile fish. Yet Brown *et al.* (1999), Jepsen *et al.* (2001, 2005), and Hall *et al.* (2009) have all challenged the 2% rule, suggesting telemetry could be applied to early life stages of many species. For endangered sturgeon, most of which occupy environments that are difficult to sample (*e.g.*, large rivers), using telemetry would greatly increase the resolution of ecological patterns of age-0 sturgeon.

In the central United States, the pallid sturgeon (*Scaphirhynchis albus*) was listed as federally endangered in 1990 (USFWS, 1990) due to habitat loss resulting from large-scale river modifications and overfishing, and they were often mistaken for shovelnose sturgeon *S. platorynchus* in roe fisheries (Bettoli *et al.*, 2009; USFWS, 2010). Pallid sturgeon are long lived, late maturing, and highly migratory (Tripp *et al.*, 2009). Using telemetry on age-0 pallid sturgeon would greatly increase the resolution of their early life ecology, but to date applying telemetric methods to these species has not been studied. Two assumptions of tagged fish are that the tags do not affect the behavior (*e.g.*, swimming performance) and physiology (*e.g.*, stress) of the fish (Nielsen, 1992). However, these assumptions have not been tested on age-0 pallid sturgeon.

The purpose of this study was to test the effects of internal and external 0.2 g nonfunctioning radio telemetry tags on age-0 pallid sturgeon growth, survival, swimming performance, and stress response to assess whether telemetry is feasible for age-0 pallid sturgeon. When compared to nontagged fish (*i.e.*, the control), we expected that internal tags would reduce growth, survival, and swimming performance of the age-0 sturgeon but an elevated stress response would be observed. Further, we expected externally tagged fish to exhibit similar growth and survival as the control fish, but swimming performance would be reduced and stress levels would be elevated.

MATERIALS AND METHODS

GROWTH AND SURVIVAL

Age-0 pallid sturgeon (total length [TL] range = 169–259 mm; mean = 210 mm TL; SE = 3.71; weight range = 9.71–32.02 g; mean = 19.21 g; SE = 0.809) were obtained from Milford Fish Hatchery (Junction City, Kansas, USA) and Gavins Point National Fish Hatchery (Yankton, South Dakota, USA) for the study. Fish were held and experiments were conducted in a recirculating aquaculture system equipped with 189 liter tanks (n = 2 sturgeon per tank, 1 if mortality occurred), mechanical and biological filtration units, and supplemental aeration at Southern Illinois University (SIU; 37°42′37″N, 89°14′17″W). Fish in each tank were fed, to apparent satiation, once daily on a diet of frozen chironomid larvae and artificial fish food (Silver Cup Fish Feed, Extruded Salmon 1.0 mm, Tooele, Utah, USA). Fish husbandry and surgical methods (described below) were approved by the Animal Care and Use Committee at SIU (Protocol # 10-004).

Sturgeon were tagged with 0.2 g, nonfunctioning radio telemetry tags from Advanced Telemetry Systems (ATS; Isanti, Minnesota). The tags did not exceed 2.5% (range = 0.9–

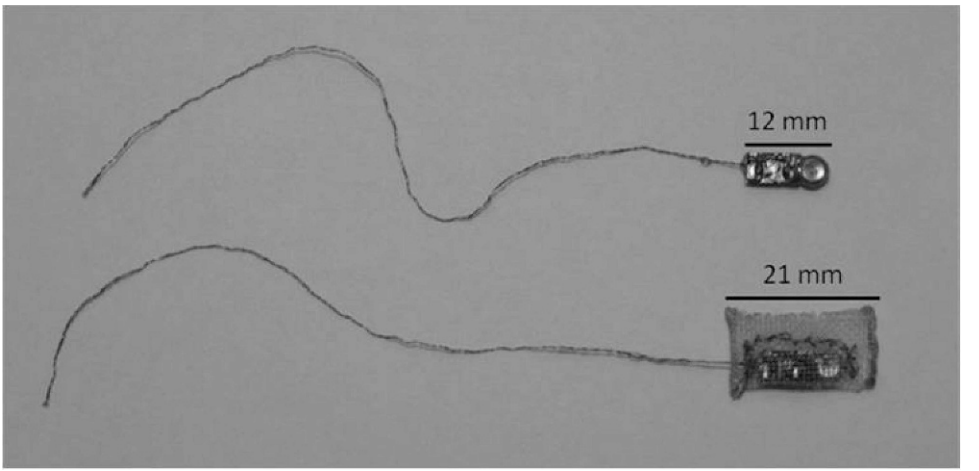

FIG. 1.—Nonfunctioning, 0.2 g telemetry tags from Advanced Telemetry Systems (ATS) used in the laboratory portion of the study to test growth, survival, and swimming performance of tagged age-0 pallid sturgeon; the bottom tag is enclosed in a mesh pouch for external attachment

2.5%; mean = 1.6%; SE = 0.068) of the sturgeon's body weight. Forty-five sturgeon were used to assess growth and survival; tags were internally implanted into 15 fish, externally attached to 15 fish, and 15 fish were used as controls (*i.e.*, handled, not tagged). Surgical instruments were disinfected with chlorhexidine before each use. Sturgeon were anesthetized individually using a carbon dioxide and oxygen mixture. Once equilibrium and swimming ability ceased, sturgeon were considered anesthetized. We kept enough water on the surgery platform to cover the sturgeon's gills but not the incision or attachment sites. Povidone iodine was swabbed over the attachment or incision site as a topical disinfectant before surgery. Internal tags were implanted by making a small 1 cm ventral incision, approximately at the fourth scute anterior to the pelvic fins. Tags were then inserted into the body cavity and pushed anteriorly away from the incision site to alleviate any added stress on the wound. The antenna exited the fish at the posterior end of the incision. Monofilament sutures, attached to a curved cutting needle, were used to close the incision with a simple interrupted suture pattern as described by Summerfelt and Smith (1990). For external attachment, tags were placed in small pouches (length ~21 mm) made of mesh netting to protect the tag and ease attachment (Fig. 1). Tags were attached dorsally, anterior to the dorsal fin. Sutures passed through the mesh pouch and under the scutes to secure the tag (Counihan and Frost, 1999). Control fish were handled similarly to the tagged fish (sedated, weighed, and measured), but they did not receive any incisions or tags.

The experiment ran for 8 w (double the expected battery life of this tag) to evaluate growth and survival. Each fish was weighed to the nearest 0.01 g and measured to the nearest mm (TL) at the beginning and conclusion of the experiment. Fish were visually assessed each day to monitor for any changes in behavior, such as feeding rate and loss of tags.

Kruskal-Wallis one-way analysis of variance (K-W ANOVA) test was used to test for differences in percent growth of sturgeon among control, externally tagged, and internally tagged fish because the data were nonnormal. If significant treatment effects were detected in the K-W ANOVA, Dunn's method was used to assess pairwise comparisons. Differences in

survival among the three treatment groups were evaluated with a chi-square test. Alpha was set at 0.05.

SWIMMING PERFORMANCE AND STRESS RESPONSE

Ten internally tagged, 10 externally tagged, and 10 control age-0 pallid sturgeon (TL range = 168–222 mm; mean = 196 mm TL; SE = 2.7; weight range = 11.73–27.10 g; mean = 17.56 g; SE = 0.81) were used to test whether tags affected swimming performance using a Brett-type swim tunnel (Brett, 1964); these fish were not the same individuals used to evaluate growth and survival, but they were from the same cohort. The tunnel had a working section, or length, of approximately 91 cm with screens placed at both the upstream and downstream ends of the chamber. Water velocities were calibrated with a Marsh-McBirney flow-meter (Flo-Mate 2000, Loveland, Colorado, USA). All swim trials were conducted 2–4 w after tagging.

Individual sturgeon were isolated and fasted 24 h before testing. Each sturgeon was acclimated in the swim tunnel for 1 h prior to testing. During that time, the velocity increased from 0 to 0.05 m/s after 30 min. Once the acclimation period was over, the water velocity was increased to 0.10 m/s and increased in a stepwise fashion by 0.05 m/s increments every 10 min to determine critical swimming speeds of the sturgeon. Trials ended when each sturgeon was fatigued, defined as a fish which was no longer able to leave the downstream end of the swim tunnel and would not respond to gentle stimulus (Adams *et al.*, 1999, 2003). Critical swimming speed (U_{crit}) was determined using the formula described by Brett (1964):

$$U_{crit} = u_1 + \left[u_2 \left(t_1 / t_2\right)\right],$$

where u_1 was the highest velocity maintained for the entire interval, u_2 was the velocity increment (0.05 m/s), t_1 was the time elapsed at fatigue velocity, and t_2 was the prescribed time period (10 min).

Critical swimming speed of pallid sturgeon was examined using analysis of covariance (ANCOVA) to test for differences among control, externally tagged, and internally tagged fish. Critical swimming speed was the response variable and tagging treatment was the predictor variable with three levels. Sturgeon total length and weight were the covariates in the ANCOVA model to determine whether they influenced U_{crit}. When significant treatment effects were detected, Tukey's HSD pairwise comparison tests were used to determine where differences occurred. Significance was set at $\alpha = 0.05$.

Hematology of sturgeon used in the swim trials was also examined to assess potential stress responses among internally and externally tagged, and control fish. After the sturgeon were considered fatigued, total lengths (mm) and weights (g) were measured. Heparinized, evacuated blood collection assemblies (Vacutainer®; Becton Dickinson and Co., Franklin Lakes, New Jersey, USA) were used to collect a blood sample from the caudal vasculature. To minimize the possibility of handling and venipuncture stressors, blood samples were collected within 5 min of fatigue. Hematocrit (Statspin® centrifuge; Fisher Scientific, Pittsburgh, Pennsylvania, USA) and glucose (Freestyle Freedom Lite® glucose meter; Abbott Laboratories, Abbott Park, Illinois, USA) were then immediately determined using the whole blood samples. Samples were centrifuged at 3000 × gravity at 4 C for 45 min, and the resulting plasma was stored at −80 C until further analysis. Osmolality (Vapro 5520; Wescor, Inc.; Logan, Utah, USA) and cortisol (EIA 1887; DRG International, Mountainside, New Jersey, USA) were determined using plasma samples.

One-way ANOVAs were used to analyze the hematocrit, glucose, and osmolality data for differences among control, externally tagged, and internally tagged sturgeon. Cortisol data

did not meet the assumption of constant variance after checking quantile plots. Thus, these data were transformed using $\log_{10}$ (x+1), after which a Welch's ANOVA was used to further control for heterogeneity in variances and potentially inaccurate P-values. Tukey's honest significant difference test was conducted to examine pairwise comparisons if the overall ANOVAs were significant. Hematology literature on sturgeon was referenced to determine if statistical differences in hematocrit, glucose, and osmolality were biologically relevant (Altinok *et al.*, 1998; Knowles *et al.*, 2006; Baker *et al.*, 2008; Bani *et al.*, 2009). Alpha was set at 0.05 for all tests.

RESULTS

GROWTH AND SURVIVAL

Overall, growth (% weight gain; $H = 6.672$; $df = 2$; $P = 0.035$) and survival ($x^2 = 13.07$; $df = 2$; $P < 0.001$) varied among treatment groups. Growth was not different between the control fish and the fish that were tagged externally (Fig. 2; $Q = 1.70$, $P > 0.05$). Internally tagged fish exhibited significantly less growth than the control fish ($Q = 2.48$, $P < 0.05$), but growth of internally tagged fish was not different from the externally tagged fish ($Q = 1.02$, $P > 0.05$). There was 100% survival in the externally tagged fish and 94% survival among the control fish. The internally tagged fish showed lowest survival (46%), which was substantially less than both the control and externally tagged fish (Fig. 2).

SWIMMING PERFORMANCE AND STRESS RESPONSE

The overall ANCOVA did not detect differences in swimming performance among the tag treatments of age-0 pallid sturgeon ($F = 1.80$; $df = 4,24$; $P = 0.16$; Fig. 2), and the covariates of total length ($t = 1.38$, $P = 0.18$) and weight ($t = -0.95$, $P = 0.35$) did not affect their swimming performance. Station-holding was observed periodically by fish in all treatment groups.

Plasma cortisol ($F = 3.06$; $df = 2,11$; $P = 0.09$) and blood glucose ($F = 0.01$; $df = 2,26$; $P = 0.99$) did not differ among externally tagged, internally tagged, and control fish. However, differences in hematocrit ($F = 33.43$; $df = 2,25$; $P < 0.001$) and osmolality ($F = 8.39$; $df = 2,21$; $P = 0.002$) occurred among treatments. Hematocrit levels were significantly different between all treatments (Table 1; for all pairwise comparisons, $P < 0.02$). On average hematocrit in the control fish was 12.5 and 30.6% greater than the internally and externally tagged fish, respectively, and 20.6% greater in the internally versus the externally tagged fish. Osmolality in the control fish was 6.0% higher than in the externally tagged fish ($P = 0.01$) and 7.5% in the internally tagged fish ($P = 0.002$). The osmolality between the internally and externally tagged fish was similar (Table 1; $P = 0.72$).

DISCUSSION

Age-0 pallid sturgeon can be tagged with 0.2 g nonfunctioning telemetry transmitters internally and externally, but internally implanted tags reduced growth and survival was less than 50%. Based on these findings, externally attached transmitters are a better option than internally inserted transmitters when tagging age-0 pallid sturgeon. Previous studies have also found tagged fish grow less than control fish (Paukert *et al.*, 2001 internal; Sutton and Benson, 2003 external; Zale *et al.*, 2005 internal; Weimer *et al.*, 2006 external). These authors argue as long as the appropriate transmitter (both internal and external) for the size of the fish and the length of the study are selected, successful telemetry studies can be completed. Here, we caution against the use of any internal transmitter, given the current sizes of transmitters available, for age-0 pallid sturgeon.

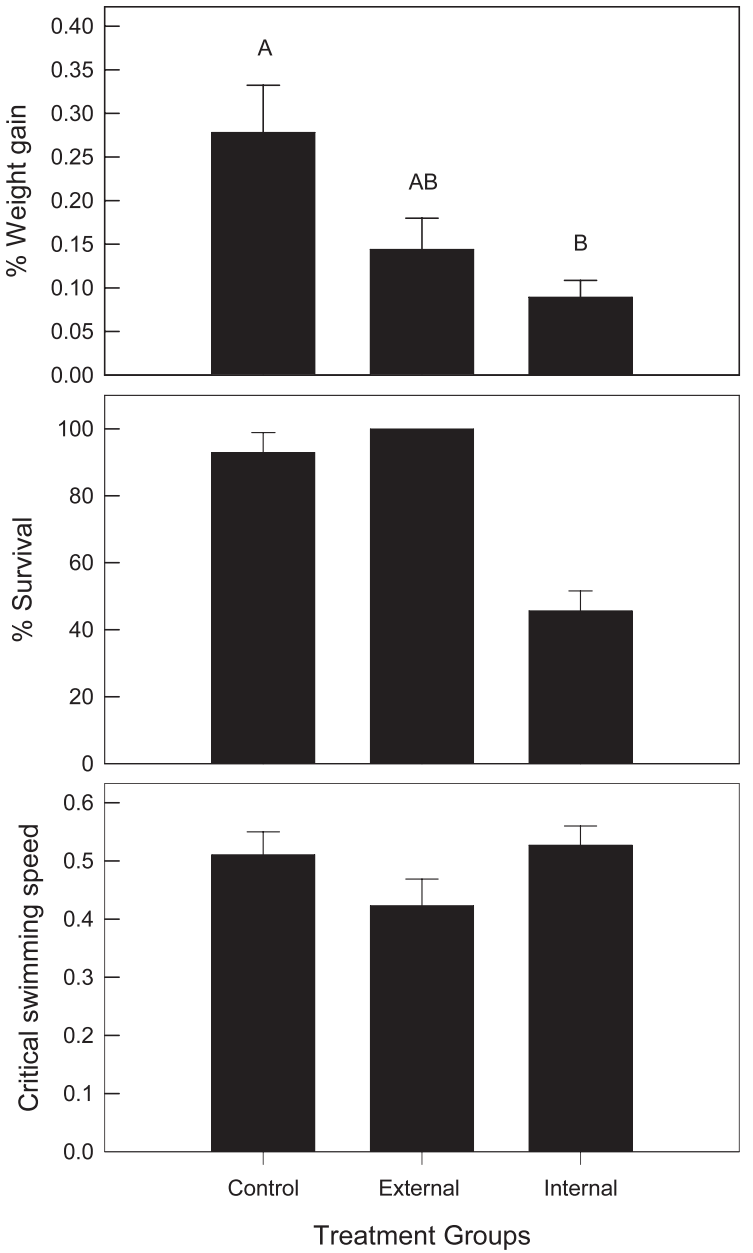

FIG. 2.—Average growth (±1 SE) as percent body weight gained (top), percent survival (middle), and mean critical swimming speed (m/s, ±1 SE) of age-0 pallid sturgeon tagged with 0.2 g, nonfunctioning telemetry tags. Different letters above the error bars for % weight gained indicate statistically different results between treatment groups based on Tukey's HSD pairwise comparisons

TABLE 1.—Hematological responses of age-0 pallid sturgeon following fatigue after measuring critical swimming speed

Parameter		Treatment groups			Overall mean (±1 SE)	P
		Internal	External	Control		
Cortisol (ng/mL)	Range	0–9.78	0–12.04	0–1.80	1.36 ± 0.21	0.09*
	Mean	2.99	4.83	0.58		
	SD	3.51	5.25	0.80		
Glucose (mg/dL)	Range	52–82	50–83	51–79	64.9 ± 1.85	0.99
	Mean	65.00	64.90	64.90		
	SD	9.14	12.09	9.45		
Hematocrit (%)	Range	14–18	12–16	17–23	16.25 ± 0.55	<0.001
	Mean	16.62	13.20	19.00		
	SD	1.51	1.32	1.89		
Osmolality (mOsm/kg)	Range	232–259	232–271	258–288	254.62 ± 2.68	0.002
	Mean	246.43	250.44	266.50		
	SD	8.96	11.52	9.65		

* Conducted using a Welch's ANOVA to control for heterogeneity in variances among treatments

The critical swimming speeds of sturgeon in this study were similar to those previously reported for juvenile *Scaphirhynchus* sturgeon. Unlike many other species (Koed and Thorstad, 2001; Robertson *et al.*, 2003; Zale *et al.*, 2005), sturgeon tend not to swim in the water column during swim trials. The majority of the time sturgeon swam along the bottom of the swim tunnel or used their pectoral fins to hold position (Adams *et al.*, 1997; Adams *et al.*, 1999; Adams *et al.*, 2003; Parsons *et al.*, 2003; Hoover *et al.*, 2011). This holding of position may account for the large range of critical swimming speeds observed in this study, by reducing swimming duration. Similarly though, 0.2 g transmitters do not affect the swimming ability of several nonsturgeon fishes (Koed and Thorstad, 2001; Robertson *et al.*, 2003; Zale *et al.*, 2005).

Our results suggest that cortisol and glucose levels do not vary or indicate a stress response in tagged (internally or externally) age-0 pallid sturgeon. The cortisol and glucose levels of pallid sturgeon in this study were slightly higher than those previously reported for other juvenile sturgeon species, but these values were well within the ranges reported for adult and juvenile *Scaphirhynchus* sturgeon. Adult pallid sturgeon have cortisol levels ranging from 0.67–15.5 ng/mL and glucose levels from 46.1 to 95.9 mg/dL (Webb *et al.*, 2007). Juveniles, however, have smaller ranges of cortisol reported from 1.16–5.42 ng/mL and glucose from 48.6–64.4 mg/dL (Barton *et al.*, 2000; Webb *et al.*, 2007; Haukenes *et al.*, 2008).

Even though hematocrit and osmolality of swim-trial sturgeon showed statistical differences among the tag treatments, it is likely they were not biologically relevant because they were well within the range of natural variability not associated with a stress response. Though hematocrit and osmolality have not been well reported in *Scaphirhynchus* sturgeon, the ranges and means of these two blood parameters were similar to those previously reported in other sturgeon species. For example juvenile Beluga sturgeon (*Huso huso*) have hematocrit levels of 14–32% (Falahatkar and Barton, 2007; Bani *et al.*, 2009), and juvenile shortnose sturgeon (*Acipenser brevirostrum*) have hematocrit levels of 22–28% and osmolality levels of 232–295 mOsm/kg (Baker *et al.*, 2005; Beyea *et al.*, 2005; Knowles *et al.*, 2006). More variable, adult lake sturgeon (*Acipenser fulvescens*) have hematocrit levels of 15–40% and osmolality levels of 207–318 mOsm/kg (Baker *et al.*, 2008). Furthermore, sturgeon have lower physiological responses to stressors than teleosts, with pallid sturgeon exhibiting even

lower responses than other sturgeon species (Barton *et al.*, 2000; Kieffer *et al.*, 2001; Baker *et al.*, 2005; Webb *et al.*, 2007).

It is worth noting that in natural systems external tags can lead to entanglement (*e.g.,* around submerged vegetation), and for small fish like age-0 sturgeon breaking away from entanglement may be difficult. This issue should be considered prior to the use of external tags on small fish. Further, due to environmental conditions pallid sturgeon occupy (*e.g.,* extreme depths and high conductivity), external radio telemetry tags may not be suitable for tracking sturgeon in large rivers (*e.g.,* Mississippi River), but this remains to be tested. As the depth of a radio telemetry transmitter increases, the surface range of the tag decreases, particularly in areas with high conductivity (Kenward, 2001). Freund and Hartman (2002) determined the maximum depth of detection for radio telemetry tags to be 10.3 m in the Ohio River making fish using deepwater habitats less likely to be detected. Additionally, Comben *et al.* (pers. comm., Grand Valley State University) found that age-0 and age-1 lake sturgeon tagged with radio telemetry tags at depths greater than 5 m were hard to detect. The Mississippi River, for example, can reach depths up to 20 m and has an average conductivity of 400 μS/cm (Jason Crites, pers. comm., Missouri Department of Conservation). Therefore, the range of the transmitters attached to the sturgeon in this study may not be optimal for tracking sturgeon in river systems similar to the Mississippi River. We suggest efforts be directed toward improving telemetry transmitter technology. Specifically, a tag which uses ultrasonic technology, a technique commonly used for tracking adult sturgeon, would be ideal (Koch *et al.*, 2012).

The smallest ultrasonic transmitter available at the time of this study was 0.6 g, which would reach 6% of an age-0 pallid sturgeons' weight; thus, the current limitation of this transmitter is its size. Juvenile sturgeon of other species (*i.e.*, lake sturgeon, gulf sturgeon, white sturgeon, and Atlantic sturgeon) were successfully tagged with ultrasonic telemetry transmitters (Counihan and Frost, 1999; Smith and King, 2005; Young and Scarnecchia, 2005; Sulak *et al.*, 2009; Fisher, 2011). However, the young of these species were slightly older and have a much greater mass [*e.g.,* Atlantic sturgeon: 80 to 300 g (Matthew Fisher, pers. comm., Delaware Division of Fish and Wildlife) and lake sturgeon: >35 g (Bradley Eggold, pers. comm., Wisconsin Department of Natural Resources)] than age-0 pallid sturgeon used in our experiment, and these other species are, therefore, able to be tagged with the ultrasonic transmitters which are currently available. Advancement in technology (*e.g.,* decrease in transmitter size), however, will be required to examine fine scale ecological relationships of age-0 pallid sturgeon and perhaps for other species as well.

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