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# **Electroshock-Induced Injury in Juvenile White Sturgeon**

F. MICHAEL HOLLIMAN\* AND JAMES B. REYNOLDS

Alaska Cooperative Fish and Wildlife Research Unit<sup>1</sup> and School of Fisheries and Ocean Sciences, University of Alaska, Fairbanks, Post Office Box 757020, Fairbanks, Alaska 99775-7020, USA

Abstract.--Most sturgeon (Acipenseridae) populations are threatened or depleted. Assessment operations that could harm individuals in these populations, such as electrofishing, must be evaluated. The risk of electrofishing-induced injury in juvenile white sturgeon Acipenser transmontanus associated with electrical waveform (60-Hz pulsed DC [PDC] versus DC) and fish size (small [24-33 cm], age 1 versus large [37-54 cm] age 2) was evaluated in a tank experiment. Exposure to a homogeneous electric field of 1.2 V/cm immobilized all white sturgeons exposed to electrical treatments. No injury was found in the control groups. The risk for hemorrhage was significantly greater among white sturgeons exposed to PDC than among those exposed to DC (relative risk = 6.7; 95% CI = 3-29). Both size-groups were at significantly more risk for hemorrhage when exposed to PDC than to DC. All white sturgeons exposed to DC recovered (upright orientation and normal swimming) in less than 30 s; 95% recovered immediately. Most large sturgeons exposed to PDC required 1-2 min for recovery; most small sturgeons recovered immediately. Our results suggest that if electrofishing is conducted in waters where imperiled populations of white sturgeons are present, DC should be considered for use instead of 60-Hz PDC. If 60-Hz PDC must be used, electrofishers should be aware that a substantial portion of fish immobilized (complete cessation of movement) while electrofishing will probably be injured (hemorrhages). Results from other studies indicate that the incidence and severity of injury from PDC electrofishing can be reduced by using lower voltages and pulse frequencies (e.g., 60 Hz to 20-30 Hz) while maintaining capture responses less severe than immobilization (e.g., galvanotaxis).

Most extant sturgeon (Acipenseridae) populations are depleted or threatened (Beamesderder and Farr 1997), raising concern about sturgeon injury when conducting electrofishing in waters where they may be present. Although sturgeons may not be targeted for capture during electrofishing operations, they may nevertheless be exposed to the electric field. Because adult sturgeons occupy deeper waters where electrofishing fields have little or no effect, they probably are not at risk. Young sturgeons, however, may be at risk for exposure because they inhabit shallow water. Numerous deleterious effects (e.g., mortality, injury, stress, reduced growth, reduced fertility, and decreased swimming stamina) have been reported in fish captured by electrofishing or exposed to electrofishing fields (summarized by Snyder 1992; Nielsen 1998). Although these harmful effects in individual fish are negligible in large populations (Schill and Beland 1995; McMichael et al. 1998),

they could influence the survival of small populations (Nielsen 1998).

Electrofishing injury studies have primarily focused on hemorrhage and vertebral damage in bony fishes, particularly salmonids (Reynolds and Holliman 2000). However, the vertebral column in fish ranges from cartilaginous notochords to fully ossified vertebral columns. In sturgeons the axial skeleton includes a notochord, with a continuous internal lumen that lacks intervertebral joints. Other distinctive features of sturgeons include a cartilaginous skeleton and thick scutes covering the body (Long 1995; Moyle and Cech 2000). The information available regarding electrofishing injury in cartilaginous fishes is contradictory. Fredenburg (1992) found no hemorrhages in a small sample of shovelnose sturgeon Scaphirhynchus platorynchus collected with pulsed DC (PDC). Conversely, paddlefish Polyodon spathula, which have a cartilaginous skeleton and a notochord like that of sturgeons, were reported to have a high incidence of injury, some being severely injured with the notochord "blown apart" (Snyder 1992).

Electrical waveform and fish size influence the incidence and severity of electrofishing injury (Taylor et al. 1957; Spencer 1967; Sharber and Carothers 1988; Dalbey et al. 1996; Thompson et al. 1997; Ainslie et al. 1998; McMichael et al.

<sup>\*</sup> Corresponding author: ftfmh@uaf.edu

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FIGURE 1.—Voltage waveforms used in the electrofishing injury experiment on white sturgeons, Abernathy Fish Technology Center, Longview, Washington, on 3 and 4 August 1998; PDC = pulsed DC.

1998). On the other hand, Hollender and Carline (1994) found no waveform (AC versus 60-Hz PDC) effects. Fredenburg (1992) found no relationship between fish length and the incidence of fish injury.

Our objective was to evaluate the effect of fish size and electrical waveform on the risk and severity of electrofishing injury in juvenile white sturgeon *Acipenser transmontanus*. This work was part of a larger project addressing electrofishing injury in warm- and coolwater fishes.

#### Methods

Experimental procedure.—Hatchery-reared white sturgeons in two size-groups (corresponding to age 1 and age 2) were subjected to one of two waveforms (continuous DC or 60-Hz PDC; Figure 1) or used as controls. The experiment was conducted in a tank at the Abernathy Fish Technology Center, Longview, Washington, on 3 and 4 August 1998. Twenty white sturgeons were assigned to each of six experimental groups, the treatment groups being defined by four combinations of fish size and waveform. Sturgeons in the two control groups (20 age-1 fish, 20 age-2) were subjected to the same procedures as fish in the treatment groups, except that electrical power was applied to the test tank. All sturgeons used in the experiment were in excellent condition with no external signs of injury before treatment.

The experimental protocol consisted of selecting fish, one at a time, from the appropriate holding tank (fish were segregated by age-group); applying a randomly assigned treatment (including control designation); classifying the behavioral response; collecting fork length (mm) and weight (g) data; and evaluating the injury status of the treated fish. Each sturgeon was shocked with the head toward the cathode to determine whether galvanotaxis (forced swimming to the anode) occurred. Both DC and PDC treatments were applied at 150 V (mean = 150 V; SD = 5 V) for a period of 3 s, an electrical energy level determined during preliminary tests to be slightly above the immobilization response (complete cessation of movement) threshold in the age-2 sturgeons. Pulsed DC output was a square wave pulsed at 60 Hz with a 6-ms pulse width (36% duty cycle), a commonly used waveform (Figure 1). The time for recovery after exposure to the treatments, where recovery was defined as the resumption of an upright orientation and normal swimming motions, was used as an indicator of treatment severity. Videotape review was used to confirm fish response and to measure recovery time.

Immediately after removal from the test tank, the fish were killed with a lethal concentration of tricaine methanesulfonate (MS-222) for measurements of length and weight and evaluation of injury status. Despite the use of mammography film and exposure screens, which are commonly used for soft-tissue imaging, the sturgeon skeleton was not visible on radiographs. Therefore, injury evaluation consisted mainly of filleting both sides of each fish to expose the axial skeleton and lateral musculature. The notochord was visually inspected for damage. Before filleting, the fish were refrigerated for 24 h. Observed hemorrhages were rated by apparent severity, based on the worst hemorrhage observed (class 0 = none apparent; class 1 = one or more wounds in the muscle; class 2 =one or more small wounds  $\leq$  width of two notochordal segments] on the spine; class 3 =one or more large wounds  $\geq$  width of two notochordal segments] on the spine; Reynolds 1996).

Test equipment.—The tests were conducted in a commercially manufactured, rectangular (168-cm  $\times$  42-cm), fiberglass tank filled with water to a depth of 40 cm. Hatchery water was continuously supplied to the tank during the experiment (i.e., a flow-through system was used). Water conductivity ( $\mu$ S/cm) and temperature (°C) in the tank were measured at the outset and conclusion of the experiment. Identical steel plates, which served as

the tank electrodes, were placed parallel to each other 125 cm apart. Plastic screens prevented fish from making contact with the electrodes and reduced the effective length of the tank to 118 cm. The power supply for the exposure tank was a Smith-Root model 15 backpack electrofishing control unit, modified to allow fine adjustment of the output voltage and programmed to ensure that each treatment was applied for 3 s. A calibrated, digital oscilloscope was used to confirm the potential difference across the electrodes and the waveform applied during treatment of each fish.

The tank electrodes were flat, covered the crosssectional area of the tank, and were positioned parallel to each other. Thus, the electric field was expected to be homogeneous, that is, to have a linear change in voltage along the length of the tank. The expected voltage gradient was 150 V applied per 125 cm distance or 1.2 V/cm. In-water voltage measurements were used to evaluate the electric field (Kolz 1993). Voltage measurements were taken between the protective screens in water of 10, 100, and 1,000 µS/cm ambient conductivity, at applied voltages of 50 to 1,050 V, to create a series of percent-of-applied-voltage profiles. The measurements were taken at 10-cm intervals, at two depths, along three transects running the length of the tank. Linear regression analysis was performed on the profiles for voltage and water conductivity combinations and on a percent voltage profile pooled from all voltage and water conductivity values.

Data analysis.—Contingency tables were used to summarize the relationships between the explanatory (waveform and fish size) and response (injury) variables. The SAS FREQ (SAS Institute 1999) procedure was used in the analysis. Hemorrhage classifications were converted to a binary status (injured versus uninjured) for statistical analysis (Thompson et al. 1997). Because random allocation of the white sturgeons to the treatment groups fixed the row margin totals at the design stage, and the column margin totals were fixed by the null hypothesis of equal treatment effects, the data were distributed hypergeometrically (Stokes et al. 1995). Initially, associations between the explanatory variables in the experiment (i.e., waveform and fish size-group) and hemorrhage status were tested in pooled tables. Then, the modification of waveform effects by size-group was evaluated in a stratified analysis of the  $2 \times 2 \times 2$  table of fish size-by-waveform-by-hemorrhage status table, where white sturgeon size was the stratification variable and the large and small size-groups



FIGURE 2.—Observed and predicted applied voltage profiles for the electric fields used in the electrofishing-induced injury study, 3 and 4 August 1998.

were the two strata. Fisher's two-sided exact test was used to test the null hypothesis of no association between the explanatory variables and response variable in pooled tables and partial tables. The strength of associations in the tables was estimated with relative risk (RR), the risk of hemorrhage for one group compared with that for another group:  $RR = p_1/p_2$ , where  $p_1$  and  $p_2$  were the proportions of sturgeons with hemorrhages within the experimental groups compared. The Breslow– Day statistic was used to test for an interaction between waveform and fish size. Simple 95% confidence intervals (CI) for the measures of relative risk were estimated by bootstrapping (Efron 1979; Manly 1997).

#### Results

All voltage profiles were significant ( $P \le 0.01$ ) with  $r^2 \ge 0.99$  (Figure 2), providing evidence that the electric field between the protective screens was homogeneous across a wide range of water conductivity and applied voltages. The regression equation for the pooled profile was y = 1.84 +0.78x, where y is the percent voltage at the distance x cm from the anode. Multiplying the regression slope (0.78; 95% CI = 0.776-0.783) by our applied voltage (150 V) indicated that the voltage gradient (E) was 1.17 V/cm at all points between the electrodes during our experiment. The empirical slope (1.17 V/cm) was similar to the expected voltage gradient (1.2 V/cm). Water conditions within the test tank changed little during the experiment, being 13.2°C with an ambient conductivity of 296 µS/cm at the outset and 13.3°C with an ambient conductivity of 311 µS/cm at the conclusion.

A total of 120 white sturgeons were used in the

TABLE 1.—Number of white sturgeons injured, by hemorrhage class (see text), according to waveform and sizegroup. Control fish were not injured and are not included. The abbreviation PDC stands for pulsed DC; small fish were age 1, large fish age 2.

		Hemorrhage class					Number	
Waveform	Size-group	0	1	2	3	Total	injured (%)	
DC	Small	18	1	1	0	20	2 (10)	
	Large	18	0	2	0	20	2 (10)	
PDC	Small	5	1	14	0	20	15 (75)	
	Large	8	0	11	1	20	12 (60)	
Total	-	49	2	28	1	80	31 (39)	

study, where 20 age-1 and 20 age-2 sturgeons were assigned to each of the DC, PDC, and control groups. Age-1 (small) white sturgeons had a mean fork length (FL) of 277 mm (SD = 19 mm, range = 225-317 mm) and a mean weight 123 g (SD = 25 g). Age-2 (large) white sturgeons had a mean FL of 439 mm (SD = 33 mm, range = 355-523mm) and a mean weight of 520 g (SD = 131 g). No notochord damage was observed in any white sturgeons used in the experiment. No hemorrhages were found in the control groups. Overall, 31 (39%) of the 80 sturgeons in the treatment groups had at least one hemorrhage (Table 1). Hemorrhages were predominantly class 2 (90%); one sturgeon (3%) had a class 3 hemorrhage and two fish (7%) had class 1 hemorrhages. The class 3 hemorrhage occurred in a large white sturgeon exposed to 60-Hz PDC. The class 1 hemorrhages were found in two small sturgeons, one from each waveform group (Table 1). The class 2 and class 3 hemorrhages were found at multiple (2-6) sites, usually with a distinct spacing pattern, extending along the notochord from midlength into the anterior caudal region near the pelvic fins.

The probability of hemorrhage among all white sturgeons exposed to PDC (68%) was greater than for those exposed to DC (10%; RR = 6.75, 95% CI = 3-29, P = 0.00). The incidence of hemorrhage in the two size-groups was not significantly different (35% versus 43%; RR = 0.82, 95% CI = 0.5-1.4, P = 0.64). Large white sturgeons had a higher risk of injury (60% versus 10%) when exposed to PDC (RR = 6, 95% CI = 2.2–29, P = 0.00). Small white sturgeons were also more likely to be injured (75% versus 10%) when exposed to PDC (RR = 7.5, 95% CI = 3–33, P = 0.00). Sturgeon size did not modify the waveform effect, as indicated by the Breslow–Day statistic (P = 0.58). The average relative risk of hemorrhage for the two sturgeon size-groups was 6.7, indicating that, compared with DC, exposure to PDC greatly increased risk for hemorrhage regardless of fish size (Table 1).

Immobilization was evoked in all white sturgeons exposed to the PDC and DC treatments. Within the immobilization response, there was a dichotomy in the reactions to the waveforms. All sturgeons subjected to PDC straightened immediately and were completely immobile upon application of the treatment. Among sturgeons exposed to DC, anodic curvature and slight quivering were observed.

All white sturgeons exposed to DC recovered in less than 30 s, with 95% recovering immediately (Table 2). A net was needed to prevent many of these fish from leaping from the test tank upon cessation of the treatment. Recovery times were much more variable among fish exposed to PDC. Most small sturgeons (70%) recovered immediately; notable exceptions were two fish requiring 285 s and 600 s for recovery. Large sturgeons subjected to PDC took longer to recover than the other groups did: 68% took 60–120 s for recovery, and one took 285 s. Most of the sturgeons that did not

TABLE 2.—Recovery times for electroshocked white sturgeons, according to waveform and size group. PDC = pulsed DC.

		Recovery time (s)							
Waveform	Size-group	0	1-30	31-60	61–90	91-120	121-150	151-180	>180
DC	Small	19	1	0	0	0	0	0	0
	Large	19	1	0	0	0	0	0	0
PDC	Small	14	3	0	1	0	0	0	2
	Large	2	0	1	5	9	2	0	1

recover immediately after exposure to PDC displayed disoriented, periodically unbalanced swimming for as long as 30 s before sinking to the bottom of the tank, apparently unconscious. Upon regaining consciousness, they again experienced a period of disoriented swimming before recovering to an upright orientation with normal swimming.

## Discussion

White sturgeons exposed to 60-Hz PDC were nearly seven times more likely to be injured (hemorrhage) than those exposed to DC (68% versus 10%, a significant difference). There was no significant difference in injury risk between the two size-groups; the rates of injury after DC treatment were similar in large and small sturgeons, as they were after PDC. Most injured fish (90%) had moderately severe hemorrhages (class 2), based on the apparent severity index (Reynolds 1996). The increased severity of PDC, relative to DC, was also indicated by longer recovery times of PDC-treated fish.

The high risk of injury in PDC-exposed white sturgeons in our study was similar to that reported by others who studied teleosts (McMichael 1993; Dalbey et al. 1996; Thompson et al. 1997; Ainslie et al. 1998). The low incidence of injury induced by DC in our study appears to support Rayner's (1949) description of DC as relatively harmless to fish. Ainslie et al. (1998), however, reported that DC-exposed rainbow trout Oncorhynchus mykiss were more severely injured than those exposed to 30-Hz PDC. Most of the injured white sturgeons in our study had moderately severe hemorrhages, regardless of electrical waveform. We have no data on healing of hemorrhages from our study. Considering the results of hemorrhage healing reported for electroshocked rainbow trout (Schill and Elle 2000), we suspect that hemorrhages in juvenile white sturgeons would heal within one growth season-although the energy diverted to healing would retard growth (Dalbey et al. 1996; Ainslie et al. 1998).

A positive relation between fish size and the incidence of injury has been reported in previous work (Lamarque 1990; Hollender and Carline 1994; Dalbey et al. 1996; Thompson et al. 1997; Ainslie et al. 1998). In our study, however, both size-groups of white sturgeons were at similar risk for hemorrhage. The size range in our study may have been insufficient to detect a fish size-injury relationship.

All white sturgeons exposed to DC or PDC in this study were immobilized, a response usually

observed in fish near the anode during an electrofishing operation. Although all electroshocked sturgeons were immobilized, the physiological effects between DC and PDC are fundamentally different. Galvanonarcosis, induced by DC through central nervous system depression, has been likened to a chemical narcosis accompanied by muscle relaxation, whereas PDC induces immobilization though stimulation of the central nervous system and muscle tetany, or contraction (Halsband 1967). The action of PDC on the musculature has been described as cramp leading to constant irritation (Halsband 1959). Immobilization renders fish most vulnerable to capture, relative to other responses (e.g., taxis), but may also put a fish at greater risk of injury, especially if tetany is induced (Reynolds 1996).

We recommend that biologists seriously consider using DC to sample juvenile white sturgeons and other fishes, particularly salmonids. Uninterrupted DC is a power-demanding waveform and, in our experience, has a reputation among biologists as being ineffective. Nevertheless, DC needs to be evaluated fairly before it is discarded as an option; this waveform is effective in eliciting galvanotaxis with little likelihood of causing tetany (Lamarque 1990). In our study, the risk of injury among DC-shocked fish was 10%, a level likely to be accepted by biologists as realistic for livefish capture and handling (Reynolds and Holliman 2000).

When high risks of injury and assumed mortality in sampled salmonids were projected to the population level, only small fractions (1-3%) of populations seemed to be affected; compared to normal levels of natural mortality, such a sampling effect is quite small (McMichael et al. 1998). However, even a small sampling mortality may be unacceptable in threatened or endangered populations (Nielsen 1998). If biologists use 60-Hz PDC, our findings indicate that at least one-half of immobilized juvenile white sturgeons will be injured. If PDC must be used to maintain acceptable catch rates, we recommend that pulse rate be reduced from 60 Hz to 20-30 Hz and that voltage be decreased to elicit fish responses that favor capture (e.g., galvanotaxis) but are less injurious than immobilization resulting from tetany (Reynolds and Holliman 2000).

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