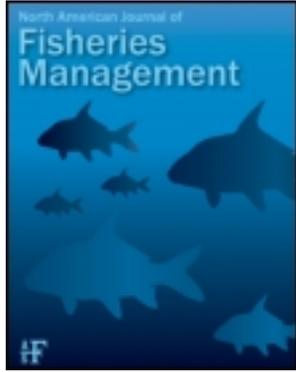


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ARTICLE

Evaluation of Two Forms of Electroanesthesia and Carbon Dioxide for Short-Term Anesthesia in Walleye

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Abstract

Anesthetics immobilize fish, reducing physical damage and stress during aquaculture practices, stock assessment, and experimental procedures. Currently, only tricaine methanesulfonate (MS-222) is approved for use as an anesthetic for food fish in Canada and the United States; however, MS-222 can only be used with certain fish species, and treated fish must be held for a specified period of time before release into the wild. Two forms of electroanesthesia and carbon dioxide (CO₂) were evaluated as anesthetics for adult walleye *Sander vitreus* to determine their suitability for use before intracoelomic implantation of telemetry transmitters. Walleyes were subjected to one of three treatment groups: constant direct current (CDC), pulsed direct current (PDC), and CO₂. Fish subjected to these treatments were monitored for induction (where appropriate) and recovery time and whether these forms of anesthesia were conducive to implanting telemetry transmitters, that is, whether they fit a surgery threshold range of 250–350 s. Additionally, all fish were monitored for posttrial survival, and radiographs were taken to determine whether any vertebral damage was associated with the electroanesthesia treatments. Although all anesthetic treatments successfully immobilized fish for enough time to implant a transmitter, PDC electroanesthesia is recommended because of its immediate induction time, quick recovery, high immediate and short-term survival, and lack of evidence of vertebral abnormalities.

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Anesthetics are valuable tools used to immobilize fish and reduce physical damage and stress during aquaculture practices (e.g., transport, sorting, spawning, and vaccination), stock assessment (e.g., enumeration of fish and collection of aging structures), and experimental procedures (e.g., telemetry transmitter implantation; Summerfelt and Smith 1990; AFS 2004). When such chemical or physical agents are applied to fish, the result is anesthesia, a loss of sensation through depression of the central and peripheral nervous systems (Iwama and Ackerman 1994). The fish undergo a series of physiological and behavioral changes in response to increasing concentration or exposure to the anesthetic. First the fish is calmed and then successively loses mobility, equilibrium, consciousness, and finally reflex action (Summerfelt and Smith 1990). The progression of these changes has been categorized into different stages of anesthesia by many researchers (e.g., McFarland 1959; Bell 1987; Yoshikawa et al. 1988; Iwama et al. 1989). Six stages of anesthesia described by Summerfelt and Smith (1990) are widely referenced. Stage-0 anesthesia refers to normal behavior, whereas stage-6 anesthesia refers to medullary collapse and asphyxia (i.e., death). Stage-4 anesthesia, characterized by total loss of equilibrium and no reaction to handling, is typically the stage required for surgical procedures on fish (Summerfelt and Smith 1990).

Although there are many factors to consider when selecting an appropriate anesthetic (e.g., cost, availability, induction and recovery times, ease of use, and human safety), local regulations and legislation may limit choices (Sattari et al. 2009). Because most chemical anesthetics are absorbed through the gills, a buildup of residue in tissues is likely to occur, making the fish unfit for human consumption until the compound is either metabolized or excreted (Marking and Meyer 1985). In Canada, tricaine methanesulfonate, commonly known as MS-222 or Aqualife TMS, is the only approved drug for use in food fish. The usage of MS-222 in food fish is limited to the Salmonidae family and is available only by veterinary prescription. Treated fish must be held for a minimum of 5 d at water temperatures of 10°C or higher (Health Canada 2010). MS-222 is also the only approved chemical anesthetic in the United States, but requires a 21 d withdrawal period (USOFR 1990). In the face of regulatory challenges, the use of carbon dioxide (CO₂) by aquaculturists and field researchers has become widespread, generally being recognized as safe when used with food items (Summerfelt and Smith 1990).

Carbon dioxide is considered a flexible anesthetic because the highest stage of anesthesia reached and the recovery time can be altered with slight changes in the CO₂ concentration or the duration of exposure (Post 1979); moreover, it can be introduced into the water directly via compressed gas or indirectly by the addition of sodium bicarbonate antacids (Peake 1998). While CO₂ gas in water effectively immobilizes fish, it does not effectively anesthetize fish to a stage that is deep enough to carry out surgery (Prince et al. 1995). This is may be a result of the disruption of the normal acid–base balance in fish caused by the hydration of CO₂; also gaseous CO₂ may not reach dissolved concentrations in the water appropriate for deep

anesthesia (Bell 1987). As such, it has been recommended that sodium bicarbonate be used in addition to CO₂ gas to act as a buffering agent (Bell 1987). Further studies by Prince et al. (1995) and Peake (1998) revealed that a sodium bicarbonate–acetic acid combination enhanced CO₂ liberation and stabilized the pH of the solution, resulting in stage-4 anesthesia of sockeye salmon *Oncorhynchus nerka* and walleye *Sander vitreus*.

Electric current can act as a nonchemical anesthetic by generating electrotetanus (muscle contraction) or electronarcosis (unconsciousness and muscle relaxation) in fish (Barham et al. 1987; Summerfelt and Smith 1990; Ross and Ross 2008). Electroanesthesia has primarily been investigated for use in hatcheries in which wild fish in spawning condition are used as broodstock to collect eggs and milt. Because most hatcheries have limited capacities to hold, feed, and maintain large numbers of fish while using the mandatory depuration time after chemical anesthesia, use of electroanesthesia is favorable (Walker et al. 1994; Redman et al. 1998). Electroanesthesia also has the benefit of rapid induction and recovery times, making it attractive for many aquaculture and laboratory applications (Chiba et al. 2006; Sattari et al. 2009). Additionally, the physiological effects of electroanesthesia have been shown to be similar to other forms of chemical anesthetics (Madden and Houston 1976; Barham et al. 1988; Henyey et al. 2002; Robb and Roth 2003). All of these characteristics make electroanesthesia appealing for such research applications as intracoelomic implantation of telemetry transmitters. Indeed, electroanesthesia effectively immobilized striped bass *Morone saxatilis* for surgery (Jennings and Looney 1998), and has since been used in telemetry studies on brown trout *Salmo trutta* (Gosset et al. 2006). Because a single combination of wave form, frequency, and shock duration may not induce suitable immobilization in all species, further investigation of electroanesthesia in other species is required (Kolz 1989; Gaikowski et al. 2001).

The walleye is an important commercial and sport fish in the Laurentian Great Lakes, particularly in Lakes Huron and Erie (Fielder et al. 2010; Vandergoot et al. 2010). The economic importance of walleyes has prompted interest in conducting large-scale, intra-lake, field telemetry studies on wild fish to answer questions about their spatial ecology and mortality. Given the high probability that some fish will be harvested and consumed shortly after implantation by recreational and commercial fishers (exploitation 12–22%; Vandergoot et al. 2009) and that most agencies cannot hold large numbers of fish for 21 d under field conditions, evaluation of nonchemical anesthesia for use on walleyes is essential. As such, the objective of this study was to evaluate two forms of electroanesthesia and CO₂ as potential alternative anesthetics to MS-222 for intracoelomic implantation of telemetry transmitters for adult walleyes.

METHODS

On 5 April 2010, adult walleyes ($n = 47$) were collected from the Maumee River, Ohio (a tributary of Lake Erie), with boat electrofishing gear (Smith-Root, Inc., 5.0 Generator

Powered Pulsator [GPP] Electrofisher; 60 pulses/s, 4–6 A). Walleyes were transported in aerated holding tanks to the Ohio Department of Natural Resources (ODNR), Sandusky Fisheries Research Station. They were then transferred to a 1.8 m × 2.4 m × 1.2 m floating net pen consisting of 13-mm bar mesh treated nylon at the station boat slip in Lake Erie, where they were held for 3 d at 13°C with 100% survival. On 8 April 2010, walleyes were collected from the net pen with the same electrofishing gear described above and transferred to a 1,514-L holding tank, where they were divided into three treatment groups (all to be described in detail): continuous direct current (CDC) to cause electrotetanus, pulsed direct current (PDC) to induce electronarcosis, and CO₂. A control group of walleyes ($n = 13$; 530 ± 19 mm total length [TL]; mean ± SE) was not subjected to anesthesia, to evaluate short-term survival. For each anesthesia treatment, fish were monitored for duration of induction to stage-4 anesthesia and duration of recovery to stage-0 anesthesia. The duration of recovery from anesthesia was compared with the time required to complete a surgery by surgeons with various degrees of experience (i.e., survey threshold range of 250–350 s; see Cooke et al. [2003]). At the end of each trial, walleyes subjected to an anesthesia treatment were tagged externally with a t-bar tag (Model FD-68B; Floy Manufacturing, Seattle, Washington), and returned to the floating net pen to assess short-term posttreatment survival for 5 d. Walleyes used as a control were also returned to the floating net pen after length was recorded. On 13 April 2010, all fish from the three treatment groups were killed with a lethal dose of MS-222 and immediately frozen in lateral aspect on a flat surface. To determine whether any vertebral damage occurred with the electrotetanus or electronarcosis treatments, vertebral radiographs for each fish (one per fish) were taken at Michigan State University, College of Veterinary Medicine, East Lansing, Michigan. Direct digital radiographic plates in the lateral view were taken for each walleye and examined by two veterinarian radiologists certified by the American College of Veterinary Radiology. Walleyes anesthetized with CO₂ served as a control for assessing vertebral abnormalities associated with the electroanesthesia treatments. The radiographs from each fish were scored according to the rating system proposed by Reynolds (1996): (0) no spinal damage apparent; (1) compression of vertebrae only; (2) misalignment of vertebrae, including compression; and (3) fracture of one or more vertebrae or complete separation of two or more vertebrae.

CDC (electrotetanus) treatment.—Immobilization (i.e., inducing a state in which fish were unable to exhibit voluntary movement) was achieved by subjecting walleyes ($n = 13$; 606 ± 21 mm TL [mean ± SE]) to a continuous electrical charge of 50 mA and 6–8 V for 300 s. CDC was applied to a 13-mm bar mesh nylon cradle with electrodes constructed of 5-mm-diameter stainless steel aircraft cable by way of a 12-V marine battery (Figure 1). The intensity and duration of the electrical current supplied to the electrodes was regulated with a control box. A detailed description of the control box and mesh cradle used is presented by Jennings and Looney (1998). Walleyes were

placed ventral side up in the mesh cradle, with a constant supply of freshwater irrigating the gills. To ensure that the fish were unresponsive to external stimuli, the individual administering the CDC treatment routinely exerted pressure on the abdomen with the aid of rubber gloves. When the 300 s had elapsed, each fish was immediately transferred back into the floating net pen as recovery was instantaneous. Posttreatment survival was monitored as previously described.

PDC (electronarcosis) treatment.—Narcosis was achieved by placing walleyes ($n = 12$; 580 ± 14 mm TL) into a confined electrical field for 3 s to induce stage-4 anesthesia. Each walleye was placed in a 379-L Rubbermaid tub filled with 284 L of Lake Erie water (temperature = 13.1°C; dissolved oxygen = 7.61 mg/L; conductivity = 328 µS; pH = 8.15); a 216-mm-diameter circular anode was placed at one end of the tub, and a square 305 mm × 305 mm cathode was placed at the other end, 125 cm apart. Both electrodes were constructed of 3.2-mm-thick stainless steel (Figure 1). The PDC was supplied to the electrodes by way of a 5.5-hp Honda generator and a Smith-Root, Inc., 2.5 GPP Electrofisher control box. Preliminary trials conducted during March 2010 with prespawning walleyes ranging in size from 550 to 721 mm TL, indicated that 45 V of PDC measured between the anode to cathode at 120 pulses/s for 3 s was sufficient for inducing narcosis for >600 s at a water temperature of 5.5°C; thus, these settings were used for the PDC treatment trials conducted on 8 April 2010. Power density (P_w), the amount of power applied to the water, was calculated as:

$$P_w = C_w \left(\frac{V}{D} \right)^2,$$

where C_w = is the conductivity of the water (µS/cm), V = the voltage, and D = the distance between the electrodes (cm; Kolz 1989). The P_w for the PDC treatments conducted on 8 April 2010 was 42.5 µW/cm³. After a fish was subjected to the PDC treatment, the fish was transferred to a 100-L cooler containing fresh lake water until it regained equilibrium and resumed regular opercular movements and swimming behavior. At this point, each fish was placed back into the floating net pen and its survival was monitored as previously described.

CO₂ treatment.—Walleyes ($n = 10$; 567 ± 16 mm TL) were anesthetized with CO₂ according to the method described by Peake (1998). Walleyes were placed in a 100-L cooler filled with 60 L of lake water, sodium bicarbonate 2.66 mg/L, and glacial acetic acid 1.0 mL/L until stage-4 anesthesia was achieved. Walleyes were immediately transferred to a different 100-L cooler containing fresh lake water and remained there until stage-0 anesthesia was regained. Survival to 5 d posttreatment was monitored as described above.

Effect of temperature on PDC recovery time.—Because water temperature has been shown to influence the duration of narcosis (Barham et al. 1989), additional electronarcosis trials were conducted on 19 April 2010 to determine whether warmer

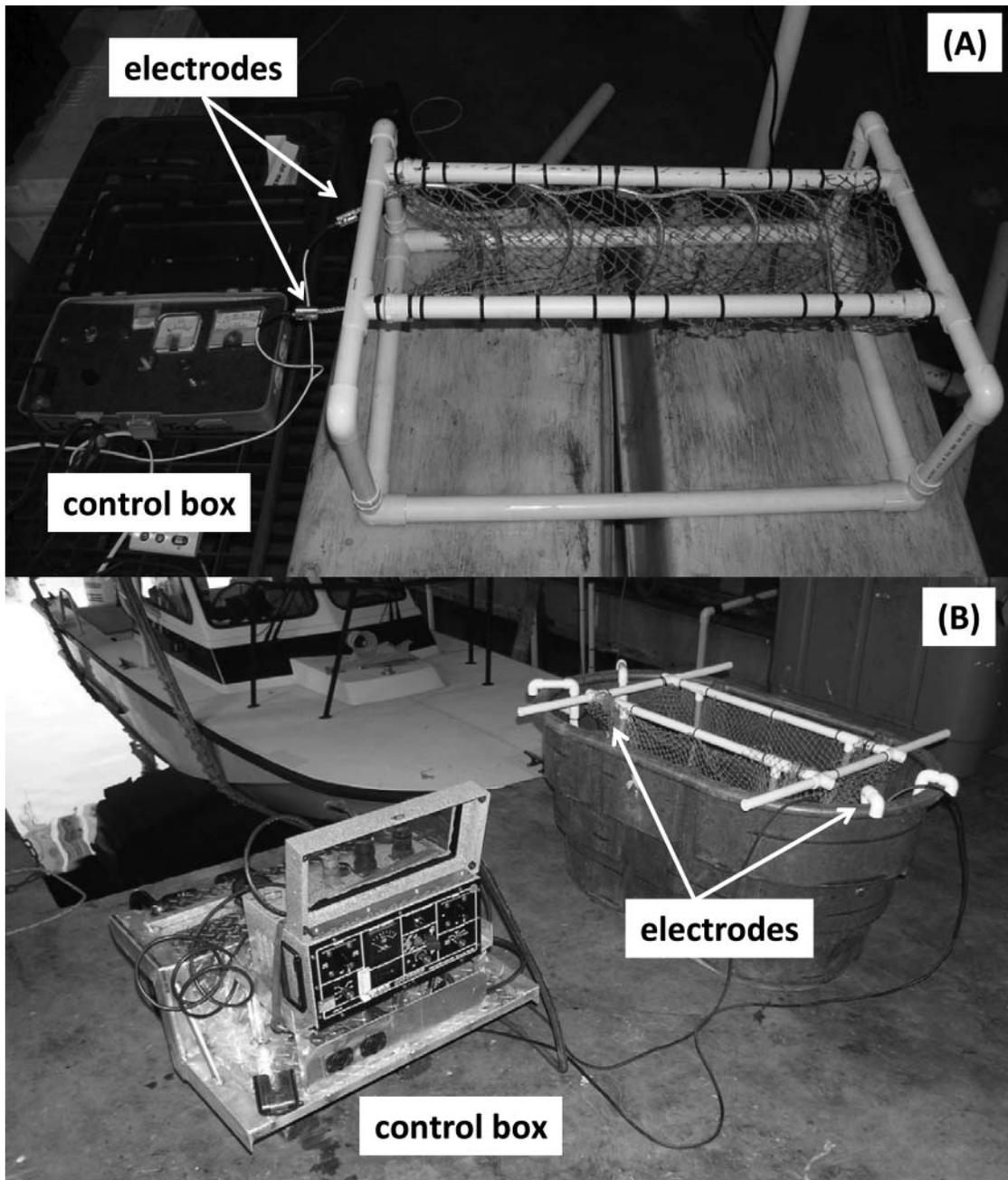


FIGURE 1. Photographs of (A) the continuous direct current (CDC) and (B) the pulsed direct current (PDC) units used for the electroanesthesia treatments. The control boxes and electrodes are labeled for each unit.

water temperatures had an effect on PDC recovery time. Spawning activity for walleyes in Ohio tributaries of Lake Erie typically peaks in April, when ambient water temperatures range between 8°C and 15°C (ODNR, unpublished data). Walleyes ($n = 11$; 500 ± 10 mm total length; mean \pm SE) were collected with electrofishing gear from the Sandusky River, Ohio (a tributary of Lake Erie), using the same collection protocol as described for the previous trials. Walleyes were transported to an adjacent

boat launch facility where the electroanesthesia trials were conducted as above with an ambient water temperature = 15°C, dissolved oxygen concentration = 14.8 mg/L, conductivity = 574 μ S/cm, and pH = 8.99. The P_w for the PDC treatments conducted on 19 April 2010 was 74.4 μ W/cm³. Postelectroanesthesia survival was monitored by holding the walleyes used in these treatments for 18 h in a floating net pen at the boat launch facility on the Sandusky River.

Data analysis.—Induction and recovery times were determined when appropriate for each of the three anesthesia treatments by videotaping the trials. Induction times were not recorded for the electroanesthesia treatments because immobilization and narcosis occurred instantaneously with the CDC and PDC treatments, respectively. For the CO₂ treatment, induction times were classified as the time required for individual fish to (1) reach stage-3 anesthesia (i.e., partial loss of equilibrium), (2) reach stage-4 anesthesia, and (3) be removed from the CO₂ treatment. Recovery times for the CO₂ and PDC treatments were characterized as the time elapsed from the time a fish was placed into the recovery cooler to (1) opercular movement, (2) fin movement, (3) stage-3 anesthesia, (4) stage-1 anesthesia (i.e., equilibrium normal, slight loss of tactile stimuli), and (5) stage-0 anesthesia. The stages of recovery used were similar to the methodology used in other electronarcosis studies (Madden and Houston 1976; Walker et al. 1994; Gaikowski et al. 2001).

The relationship between the different stages of recovery described above and fish length was examined for each treatment by correlation analysis. Because there was no evidence of a strong relationship between fish length and recovery times (Pearson's correlation coefficients <0.40), average recovery times for each treatment were estimated. Mean recovery times for the CO₂ and PDC trials conducted on 8 April 2010 and the PDC trials conducted on 8 April 2010 and 19 April 2010 were compared by *t*-tests ($\alpha = 0.05$; SAS Institute, Inc., 2010). If the *F*-statistic indicated that variance heterogeneity was significant ($P < 0.10$; SAS Institute, Inc., 2010) between treatment levels, approximate *t*-tests and Satterthwaite's approximation for degrees of freedom were used to compare mean recovery times. Use of Bonferroni adjusted *t*-tests (α/n tests performed) to compare treatment-level means, gave an adjusted α of 0.01 (0.05/5 recovery tests).

RESULTS

Physical Responses to Anesthesia Treatments

All three anesthesia treatments successfully immobilized or anesthetized walleyes in this study. Although walleyes typically remained motionless during the CDC procedure, occasional body movements were observed. If body movements became unsuitable for implanting a telemetry transmitter or suturing the incision, the intensity of the electrical current was either increased (maximum of 50 mA) or decreased until movement ceased. Once the electrical current was adjusted properly, body movements were minimal despite the repeated pressing on the abdomen in an attempt to elicit a response. At the end of the CDC treatment, walleyes were returned to the recovery tanks and normal swimming activity resumed immediately; immediate recovery meant recovery times were not recorded. The fish were then placed in the floating net pen for monitoring of posttreatment survival.

Once PDC was delivered to the electrodes, narcosis was instantaneous; fish immediately lost equilibrium, opercular move-

ment, and swimming ability. When first transferred to the recovery tanks, the fish were unresponsive to physical stimuli. Shortly after fish were placed in the tanks, however, opercular movement resumed, followed by stage-3 anesthesia and stage-1 anesthesia until stage-0 anesthesia occurred (Figure 2).

Walleyes anesthetized with CO₂ gradually exhibited stage-1 anesthesia, followed by stage-3 anesthesia as time passed (286 s; 95% confidence interval [CI], 225–346 s) until stage-4 anesthesia occurred. Although slower and more erratic than before their immersion, opercular movement was evident when walleyes were removed from the CO₂ and transferred to the recovery tanks. Although opercular movement was still visible during stage-4 anesthesia, recovery of walleyes anesthetized with CO₂ recovered from stage-4 to stage-0 anesthesia was similar to that of walleyes anesthetized with the PDC treatment.

PDC and CO₂ Solution Comparison

Walleyes anesthetized with CO₂ exhibited quicker recovery times than did walleyes anesthetized with PDC (Figure 2; 8 April 2010 treatments). Recovery times were significantly quicker with the CO₂ treatment than with the PDC treatment with respect to: opercular movement ($t = -7.10$, Satterthwaite $df = 11$, $P < 0.001$), fin movement ($t = -4.66$, Satterthwaite $df = 13.7$, $P < 0.001$), and stage-3 anesthesia ($t = -3.14$, $df = 18$, $P = 0.007$). The duration of time observed for walleyes to reach stage-1 anesthesia ($t = -2.14$, $df = 18$, $P = 0.046$) and total recovery times (stage-0 anesthesia) were similar ($t = -1.74$, $df = 19$, $P = 0.100$) between the treatments. Opercular movement was observed at the time that walleyes anesthetized with CO₂ were transferred to the recovery tanks and before the surgery threshold range had expired for the PDC treatment (Figure 2). Walleyes anesthetized with CO₂ exhibited fin movement within the surgery threshold range; whereas with PDC, fin movement occurred after the surgery threshold range had elapsed. Fin movement (522 s; 95% CI, 368–677 s) and transition to stage-3 anesthesia (543 s; 95% CI, 379–706) were observed almost simultaneously after the surgery threshold range elapsed with PDC; a similar phenomenon was observed with CO₂ but occurred before the onset of the surgery threshold. Recovery to stage-1 and stage-0 anesthesia occurred before the surgery threshold range had elapsed for CO₂ treatment, but after the threshold had elapsed with the PDC treatment (Figure 2).

Effect of Water Temperature on PDC

Time elapsed before fin movement ($t = 3.30$, $df = 20$, $P = 0.004$) was observed, and recovery to stage-3 anesthesia ($t = 3.33$, Satterthwaite $df = 15.6$, $P = 0.004$) was significantly quicker when the water temperature was 15°C (19 April 2010) rather than 13°C (8 April 2010; Figure 3). There was no statistical difference between average opercular movement ($t = 2.08$, $df = 21$, $P = 0.050$), stage-1 anesthesia ($t = 2.22$, Satterthwaite $df = 15.2$, $P = 0.042$), and stage-0 anesthesia ($t = 2.46$, Satterthwaite $df = 14.7$, $P = 0.027$) when the water was 15°C instead of 13°C (Figure 3). Similar to the trials conducted at 13°C, the

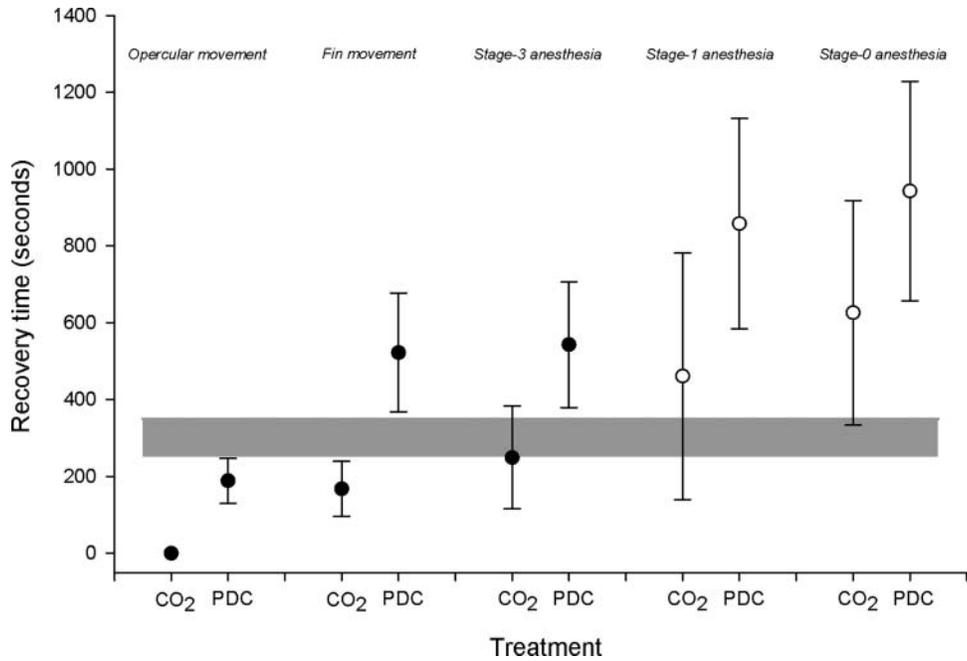


FIGURE 2. Average times required for walleyes to resume opercular and fin movements and achieve stage-3 anesthesia (partial loss of equilibrium), stage-1 anesthesia (normal equilibrium; slight loss of tactile stimuli), and stage-0 anesthesia (normal physical behavior) after CO₂ and pulsed-DC (PDC) anesthesia treatments on 8 April 2010. The shaded area denotes the surgery time threshold (300–350 s) used to compare the recovery times with the two treatments. The error bars represent the 95% confidence intervals; filled circles indicate significant differences ($P < 0.01$) in mean recovery time.

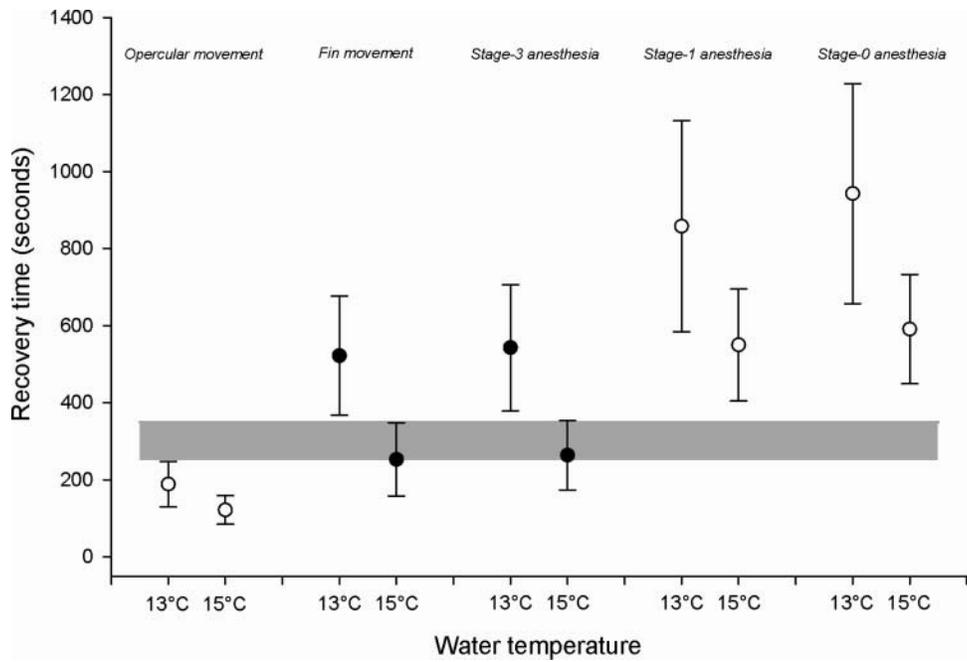


FIGURE 3. Average times required for walleyes to resume opercular and fin movements and achieve stage-3, stage-1, and stage-0 anesthesia after PDC treatments at 13°C and 15°C. The 13°C trials were conducted at the Sandusky Fisheries Research Station, Sandusky, Ohio, on 8 April 2010; the 15°C trials were conducted at the Sandusky River, Ohio, on 19 April 2010. See Figure 2 for other details.

TABLE 1. Percentage (actual numbers in parentheses) of walleyes anesthetized on 8 April 2010 with CO₂, continuous direct current (CDC), and pulsed direct current (PDC) exhibiting no vertebral abnormalities (0), compression of vertebrae only (1), misalignment of vertebrae, including compression (2), and fracture of one or more vertebrae or complete separation of two or more vertebrae (3), as determined by two veterinarian radiologists.

Reader	CO ₂ (n = 12)				CDC (n = 13)				PDC (n = 12)			
	0	1	2	3	0	1	2	3	0	1	2	3
1	100 (12)				84.6 (11)	15.4 (2)			100 (12)			
2	100 (12)				92.3 (12)		7.7 (1)					

average opercular movement recovery time (mean = 122 s; 95% CI, 85–159 s) occurred before the surgery threshold range expiration for the trials conducted at 15°C; at 13°C, however, fin movement (253 s; 95% CI, 158–348 s) and stage-3 anesthesia (264 s; 95% CI, 174–354 s) occurred during the surgery threshold range. Although stage-1 anesthesia and stage-0 anesthesia recovery times were 35.6% and 37.2% lower, respectively, for the PDC trials conducted at 15°C than at 13°C, these recovery times occurred after the surgery threshold range expired. The walleyes anesthetized with PDC at 13°C were significantly larger ($t = 4.14$, $df = 20$, $P = 0.001$) than the walleyes anesthetized at 15°C.

Survival and Vertebral Abnormalities

Walleye survival during and following the anesthesia treatments exceeded 90%. On 8 April 2010, survival was 100% during the CDC and CO₂ treatments. Similarly, survival was 100% during the PDC trials conducted on 19 April 2010. Only one mortality occurred during the anesthesia trials with the PDC treatment on 8 April 2010, for overall survival of 91.7%. Postanesthesia survival was 100% for the CDC, PDC, and CO₂ solution trials conducted on 8 April 2010 (i.e., 5 d) and for the PDC trials on 19 April 2010 (i.e., 18 h). Walleyes anesthetized with CO₂ (i.e., the control for the electroanesthesia treatments) and CDC exhibited signs of vertebral abnormalities (Table 1); however, these abnormalities were deemed to be congenital by both veterinarian radiologists. No vertebral abnormalities were observed among the walleyes anesthetized with PDC.

DISCUSSION

Our results demonstrate that CO₂ and electroanesthesia are suitable for immobilizing adult walleyes for surgical implantation of telemetry transmitters. Both electrotetanus and electroanesthesia had instantaneous induction times and met surgery thresholds. While vertebral abnormalities were noted for 23% of fish anesthetized with CDC, distinguishing between natural abnormalities in the vertebrae and electroanesthesia injuries can be subjective (Snyder 2003). One of the walleyes anesthetized with CDC was given a score of 2 (i.e., misalignment of vertebrae, including compression), which is generally indicative of an electroanesthesia injury (Sharber et al. 1994). However, the vertebral abnormalities observed in the walleyes examined in

this study were deemed congenital or the result of a previous injury by the veterinarian radiologists and not associated with the electroanesthesia treatments.

Fish anesthetized with electroanesthesia are susceptible to the same lethal and sublethal injuries as fish collected with electrofishing gear (Madden and Houston 1976; Gaikowski et al. 2001). Mortality associated with electroanesthesia can occur either immediately or shortly after the electrical current is administered (Gaikowski et al. 2001). In the current study, immediate mortality for walleyes anesthetized with electroanesthesia was 8.3%. The mechanism leading to the mortality observed in this study was not determined; however, mortalities in fish exposed to electrical shock are typically attributed to respiratory failure (Snyder 2003). If electroanesthesia is used in preparation for implanting transmitters, fish should be placed in well-oxygenated water during the surgery and postoperative period. Although vertebral abnormalities have been documented in electroanesthesia studies with lake trout *Salvelinus namaycush* (Gaikowski et al. 2001) and northern pike *Esox lucius* (Walker et al. 1994), none were observed with the walleyes used in this study. However, because vertebral injuries are difficult to detect, future vertebral assessments using fish anesthetized with electroanesthesia should include both lateral- and dorsal-view radiographs to facilitate determination of the nature and severity of spinal injuries if feasible (Thompson et al. 1997). Additionally, fish should be held at least 30 d after anesthetization to facilitate the detection of vertebral injuries such as compressed vertebrae which may not be readily evident; studies also should be conducted to further understand the latent effects of electroanesthesia associated with fish survival, behavior, and physiology (Snyder 2003).

In general, walleyes subjected to PDC exhibited behavior similar to that reported in studies of lake trout (Gaikowski et al. 2001), rainbow trout *Oncorhynchus mykiss* (Madden and Houston 1976), and Atlantic salmon *Salmo salar* (Robb and Roth 2003). Once the PDC ceased, walleyes momentarily floated to the surface of the electroanesthesia tank. When transferred to the recovery tank, fish often continued to float at the surface before sinking to the bottom of the cooler. Before opercular and fin movement were exhibited, uncoordinated twitching and flinching frequently occurred. These observations were consistent with fish in a state of narcosis as defined by Sharber and Black (1999) and Robb and Roth (2003). With the walleye electroanesthesia trials, the time to reach each recovery stage

evaluated decreased with an increase in ambient water temperature. Barham et al. (1989) observed an inverse relationship between water temperature and narcosis time for carp *Cyprinus carpio* anesthetized with electronarcosis; as water temperature increased, voltage, current, or shock duration also had to be increased to anesthetize fish. This phenomenon occurs because increasing water temperature increases is accompanied by a change in water conductivity (Reynolds 1996; Snyder 2003), a change that ultimately affects the ability of electrical power (voltage \times current) to be transferred from the water to the fish (Kolz 1989; Reynolds 1996). Although the behavioral response of fish to electrofishing gear is complex (Sharber and Black 1999) and influenced by a suite of variables, including gear configuration, fish physiology, and ambient water characteristics (Reynolds 1996); anecdotal observations suggest that fish collected with electrofishing gear at low water temperatures also require a longer period of time to recover than those collected at warmer temperatures (Reynolds 1996). However, the quicker recovery times observed in our electronarcosis trials with the elevated water temperature, despite more power being applied to the water, may also have been influenced by fish size. Barham et al. (1987) observed a positive relationship between narcosis time and fish length. Thus, the shorter recovery times observed during the 19 April 2010 PDC trials may have been influenced by using smaller adult walleyes than during the 8 April 2010 PDC trials. Unfortunately, this study was not designed to thoroughly investigate the influence of water temperature and fish size on narcosis duration and further investigation into this phenomenon is warranted. Regardless, narcosis durations for walleyes anesthetized with electronarcosis were still adequate for implanting transmitters, provided surgery time did not exceed 350 s.

As with the electroanesthesia treatments, walleyes anesthetized with CO₂ in this study exhibited high immediate and posttreatment survival; however, these fish reached stage-1 anesthesia before the surgery threshold elapsed. In the experiments conducted by Peake (1998), walleyes reached stage-1 anesthesia between 270 and 318 s at a water temperature of 10°C, compared with 140–782 s in the current study carried out when the water temperature was 13°C. Spawning activity for walleyes in Ohio tributaries of Lake Erie typically occurs at water temperatures between 10°C and 13°C; thus, anesthetizing walleyes with a solution of sodium bicarbonate before implanting intracoelomic transmitters is likely problematic because fish may recover before the surgeries are completed—unless a maintenance dose of sodium bicarbonate solution is administered during the implantation procedure (Summerfelt and Smith 1990). However, increasing the duration of time that fish are exposed to CO₂ may further increase recovery time.

Although both electroanesthesia treatments successfully immobilized fish for a sufficient time to perform intracoelomic implantation procedures, we recommend using electronarcosis. When anesthetized with electronarcosis, the body musculature became relaxed and fish were incapable of directed movement; conditions that are ideal for performing surgical implants. Con-

versely, the musculature of walleyes anesthetized with electrotetanus was rigid, and fish were capable of movement if the body ceased to remain in contact with the electrodes, a problem that could complicate the implantation procedure. The use of electronarcosis precludes the need to hold fish for a mandatory period of time when using MS-222 as an anesthetic as required by law in Canada and the USA. Furthermore, if standard electrofishing safety precautions are followed (see Reynolds 1996), the surgeon should have no human-health related concerns, which may exist with chemical anesthetics. Future electronarcosis studies should evaluate the affect of P_w , pulse frequency and duration as it pertains to fish physiology, recovery time, long-term (e.g., 30 d) behavior, and survival.

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