



# Physiological effects of ice-angling capture and handling on northern pike, *Esox lucius*

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**Abstract** Understanding how released fish recover following capture is vital information for researchers examining the effects of angling on exploited populations. This information is virtually non-existent for fish angled through the ice in winter, despite the popularity of ice-angling in many northern areas. To address this gap, 60 northern pike, *Esox lucius* L., were angled through the ice from an impoundment in eastern Wisconsin, USA, and subjected to one of ten combinations of handling and recovery duration. Plasma samples were collected and analysed for cortisol, lactate and glucose. The results showed a delayed response in the elevation of plasma variables, and a significant interaction between air exposure and recovery time for plasma lactate. No fish suffered mortality during the period of holding. Collectively, these data suggest that northern pike are physiologically resilient to ice-angling capture stress as long as air exposure times are kept at 4 min or less.

**KEY WORDS:** catch-and-release, fisheries conservation, stress response, sublethal effects, temperature, winter ecology.

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## Introduction

One of the goals of wildlife and fisheries managers is to create policies that ensure the sustainability of resources (Cox *et al.* 2010). The desire to conserve economically vital fisheries is well ingrained in managers and researchers as well as resource users (Cooke & Cowx

2006). This goal applies to the implementation of recreational angling regulations, where there is a major concern that overexploitation of recreational fisheries may lead to a loss in fishery quality or even complete collapse (Post *et al.* 2002; Post 2013). These management objectives can include the implementation of size limits that mandate anglers release their catch if it does not

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meet a certain minimum size (or exceeds a maximum size in the case of slot limits) (Noble & Jones 1993), the exclusion of anglers from certain geographical areas or restrictions limiting angling at certain times of the year (Kubacki *et al.* 2002).

The use of catch-and-release as a conservation tool has become widespread among freshwater anglers. Oftentimes, catch-and-release efforts go above and beyond what is mandated by angling regulations, particularly when these practices are seen by resource users as serving to maintain populations of large and/or highly prized gamefish species (Policansky 2008). In general, evidence suggests that catch-and-release angling is an effective tool in the conservation of inland fisheries as angled fish can be caught multiple times (Cooke & Schramm 2007). However, studies have demonstrated that, depending on the gear type used and the ambient conditions (e.g. temperature, water quality) at the time of capture, post-release mortality may counteract the otherwise positive effects of releasing captured fish whether release is mandated or voluntary (Ferguson & Tufts 1992; Cooke *et al.* 2002; Gingerich *et al.* 2007). Even if mortality does not occur, fitness consequences can still be incurred by captured fish (Lewin *et al.* 2006). These include numerous sublethal physiological and behavioural consequences such as blood acidosis (Donaldson *et al.* 2011), reduction of energy stores (Wilkie *et al.* 1997), reflex impairment (Raby *et al.* 2012), reduced movement post-capture (Klefoth *et al.* 2008) and in the case of brood-guarding individuals a greater chance of nest abandonment (Suski *et al.* 2003). Understanding how different contexts and conditions may influence these sublethal effects is of critical importance in understanding the biological consequences for captured-and-released individuals, because differences in abiotic factors at the time of capture (such as temperature) may play a major role in determining if fish suffer physiological disturbance or mortality (Gingerich *et al.* 2007).

Currently, knowledge of the ecology and physiology of fish in the winter lags behind knowledge of fish biology in warmer seasons (Lavery 2016). Indeed, nearly all studies of the sublethal consequences of capture and release on fish have been performed during warm weather periods. This exclusive focus on warm weather angling is surprising as ice-angling during the winter is quite popular, particularly in northern regions where lakes are ice-covered for several months of the year (Deroba *et al.* 2007). As an example of the popularity of ice-angling, a study of seasonal angler effort targeting northern pike *Esox lucius* L. in a set of lakes in Northern Wisconsin found that average ice-angling effort was only slightly lower than average open-water effort and that the average number of fish harvested in the winter actually exceeded that found in the

open-water season (Margenau *et al.* 2003). In the northern USA and in Canada, ice-angling typically involves suspending an artificial lure or live bait through a drilled hole in the ice, which is either left unattended by the angler (e.g. when an angler uses 'tip-ups', see methods below) or jigged on a small pole vertically down into the water column, although some fish are captured *via* spearing as well (Pierce & Cook 2000). During the winter, cold air and water temperatures present challenges to a captured fish that do not exist during the summer, including the risk of increased tissue damage (particularly to the gills) as a result of freezing in subzero temperatures, as well as reductions in the activity of enzyme-driven mechanisms to facilitate recovery from the capture stressor (Donaldson *et al.* 2008). Despite the potential of these factors to create major differences in the stress and recovery experienced in winter-captured fish, there exists no report to this point quantifying post-capture physiology in ice-angled freshwater fish.

The goal of this study therefore was to quantify the stress response and subsequent recovery of northern pike angled by common ice-angling techniques during winter. Because air exposure has been shown to be a major factor determining outcomes for captured fish (Cook *et al.* 2015), the study quantified how differing amounts of post-capture handling time with concurrent air exposure would impact the post-capture blood physiology of northern pike in the winter. Northern pike were chosen as the focal species to conduct this investigation because, in the USA and Canada, northern pike are one of the most sought after species during the winter months (Schroeder & Fulton 2014). Their popularity as a target species is partially due to their large size relative to other freshwater gamefish species and the fact that they continue to forage in the winter months (Sammons *et al.* 1994), thus making them easier to catch during this period compared with some other species. Furthermore, while a greater percentage of captured northern pike are harvested during the winter compared to the ice-free period, the release rate of captured fish in the winter has still been found to approach half of all captured fish (Margenau *et al.* 2003). While this study does not assess post-capture physiology in the summer, the existence of previous work on summer capture in pike allows for a cursory comparison of physiological disturbance between seasons (Arlinghaus *et al.* 2009).

## Methods

### Study site

All fieldwork took place at Grand Lake, a 0.98-km<sup>2</sup> impoundment of the Grand River in eastern Wisconsin,

USA (N 43.68938070, W 89.12186220), between 12 and 15 January 2016 (Fig. 1). Grand Lake is a shallow impoundment featuring a primarily sandy bottom covered with dense patches of aquatic macrophytes and a mean depth of approximately 1 m (Wisconsin Department of Natural Resources Website). In addition to northern pike, the sportfish community includes bluegill, *Lepomis macrochirus* Rafinesque, yellow perch, *Perca flavescens* Mitchill, largemouth bass *Micropterus salmoides* (Lacépède), and walleye, *Sander vitreus* (Mitchill). The minimum legal length for harvest of northern pike in this water body is 66 cm, with all fish captured in this study coming in below this length (i.e. all fish captured in this study legally would need to be released by anglers). Angling took place on the southwest end of the flowage where water depth was <1 m, as a result water temperatures as measured with a handheld thermometer did not vary by depth and were observed daily between 0 and 1 °C. Air temperature data were taken from a weather station in the town of Kingston, WI (which is on the shore of Grand Lake). Over the 4 days of angling daily high temperatures fluctuated between a low of -16 °C on 13 January and a high of 0 °C on 14 January, and temperatures within each day never fluctuated by more than 3 °C.

#### Ice-angling

All fish were captured using 'tip-ups', one of the most common gear types used for capturing northern pike in the winter (Fenske, personal observation). The tip-ups used were standard for pike anglers and consisted of a wooden or metal frame that suspended a spool of 4.5 kg

braided-line equipped with a steel leader and either a size 4 treble hook or a size 4/0 J-hook baited with a live golden shiner, *Notemigonus crysoleucas* (Mitchill). Lines were suspended through a drilled hole and into the water just below the ice-water interface. The spool of line was attached *via* a metal bar to a T-bar above the water that held a spring-loaded flag in place. When a fish became hooked by ingesting the bait, the spool and bar spun, releasing the flag to alert anglers. On each day of sampling, 18 tip-ups were spread evenly over a 100 m × 100 m area on the ice. Upon hooking, fish were retrieved hand-over-hand by the angler, with fight times never exceeding 15 s as is typical for the retrieval of ice-angled northern pike (Fenske, personal observation). Once the fish was landed, the hook was removed quickly (within 1 min) with pliers. At the outset, it was determined that only fish that were not 'deep-hooked' in the gills or gullet would be included to avoid the confounding influence of deep-hooking on post-release mortality (Stein *et al.* 2012; Lennox *et al.* 2015); however, all captured fish in the study were hooked in the mouth, which allowed for easy hook removal.

#### Handling and recovery

Following capture, each fish was processed according to one of four handling treatments. These included a control handling treatment in which fish were dehooked and immediately had blood drawn (recovery time = 0 min), 'low-stress' handling (fish was dehooked and immediately transferred to a holding container), 'medium-stress' handling (fish was dehooked, then subjected to an additional 2 min of air exposure in a nylon net before being

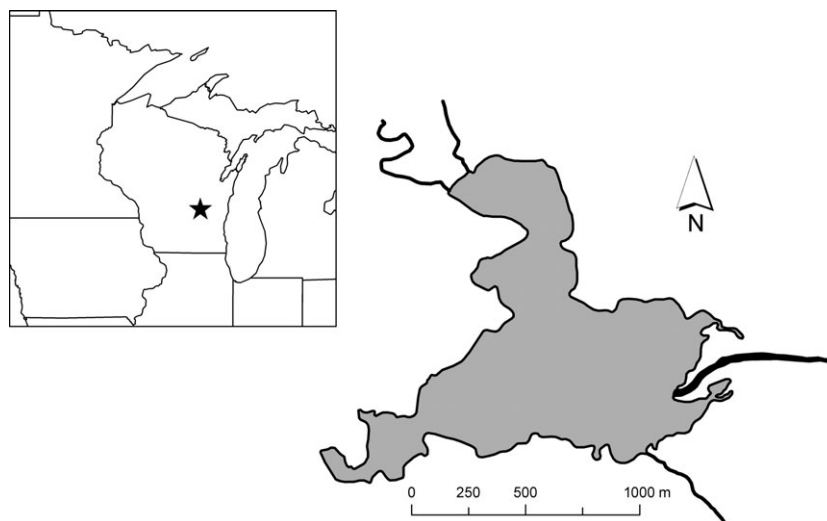


Figure 1. Map of Grand Lake in eastern Wisconsin, USA.

transferred to a holding container) and 'high-stress' handling (fish was exposed to an additional 4 min of air exposure, with the first 2 min consisting of the fish being laid directly on the ice). The high-stress ice-exposure treatment was designed as a result of communication with local anglers who described how northern pike are often laid on the ice following capture while the angler deals with their gear, retrieves a camera for a photograph, obtains their measurement device and/or decides whether or not a fish needs to be released. Fish holding took place in one of several 190-L plastic containers filled with fresh lake water. Containers all had a tight-fitting plastic lid to minimise light exposure and other sensory stimuli, making these containers similar to recovery chambers that have been used in past studies (Galloway & Kieffer 2003; Arlinghaus *et al.* 2009). While dissolved oxygen in the holding containers was not monitored during the study to avoid disturbing the fish, preliminary trials showed that oxygen levels remained above  $10 \text{ mg L}^{-1}$  even after holding a northern pike for 4 h, meaning hypoxia during holding was likely not a factor influencing the stress response of held fish. In addition, water temperatures in the holding tanks were unlikely to be a factor driving post-capture physiology, as variability in temperature (as assessed in a subset of tanks following fish processing) was very low (measured between  $1^\circ$  and  $2^\circ \text{C}$  over the course of the study). Each fish (except for those in the control treatment) was allowed to recover in the holding tanks for a period of 45 min, 2 h or 4 h prior to blood drawing, a time course common in studies of this type (Galloway & Kieffer 2003). Following recovery, each fish was observed for condition by simply noting whether it was maintaining equilibrium in the tank before having blood drawn as described below.

#### *Blood drawing and assessment of stress*

Following recovery (or capture in the case of control fish), 0.5–1.0 mL of whole blood was drawn from each fish *via* caudal puncture with a heparinised 21-gauge needle (40-mm long). Blood was immediately centrifuged for 2 min at 3000 *g* to separate plasma from other blood components. Plasma was then immediately frozen in liquid nitrogen and held prior to transport back to the laboratory where samples were stored at  $-80^\circ \text{C}$ . Each fish had a small clip taken from its caudal fin to avoid resampling a fish in the event it was recaptured, however no recaptures occurred during the study.

Cortisol concentration in the plasma was quantified using a commercially available ELISA Immunoassay kit (Enzo Life Sciences, Farmingdale, NY, USA), previously validated for use in fishes (Sink *et al.* 2008).

Plasma lactate concentrations were quantified calorimetrically from perchloric acid extracts on a 96-well spectrophotometry plate (Lowry & Passonneau 1972). Glucose was quantified calorimetrically on a 96-well plate (Bergmeyer 1974). All animals were processed according to protocols approved by the University of Illinois Care and Use Committee (IACUC), protocol #15169.

#### *Statistical analysis*

A one-way analysis of variance (ANOVA) was used to compare the mean total length of pike across all treatment blocks. Because mean total length was not significantly different across blocks (d.f. = 9,  $F = 0.251$ ,  $P = 0.984$ ), body length was ignored as a covariate (Engqvist 2005) and two-way ANOVAs were used to quantify the main effects of handling treatment (control, low/medium/high stress) and recovery time (0 min, 45 min, 2 h and 4 h) as well as their interaction, on concentrations of plasma cortisol, glucose and lactate. If a significant interaction was present, main effects were ignored. In the event of a significant main effect, or if the interaction term was significant, specific differences among treatment blocks were evaluated using Tukey's honest significance (HSD) test. All analyses were conducted in SPSS (version 23, Chicago, IL, USA) with significance values set at  $\alpha = 0.05$ . A total of  $n = 6$  fish were included in each treatment block (including controls), except for the control block for glucose, where a technical issue rendered one sample unusable leading to an  $n$  of 5 for that block.

#### **Results**

A total of 60 northern pike with a mean total length of 540 cm ( $\pm 8$  cm SE) were captured over the four angling days. All captured-and-held fish were able to swim away successfully when released. No impairment was observed in any captured fish in the holding tanks. Furthermore, no post-capture injury was observed in the fish resulting from holding (i.e. no bruises or abrasions from colliding with the walls of the tanks).

Plasma cortisol levels did not rise significantly above control values until 2 h post-capture (Table 1; Fig. 2a), at which point mean values across handling treatments were 15-fold higher than control values. Cortisol values at 4 h showed a further 36% increase over those at 2 h, which was statistically significant (Table 1; Fig. 2a). No significant effect of handling treatment, or its interaction with recovery time, was found for plasma cortisol (Table 1).

Plasma glucose levels did not increase relative to control values until 4 h post-capture, at which point they

**Table 1.** Statistical output for the effects of handling (control, quick, moderate, extended), recovery time (0 m, 45 m, 2 h, 4 h) and their interaction each concentrations of cortisol, lactate in plasma of northern pike derived from two-way analysis of variance (ANOVA). Significant results ( $\alpha < 0.05$ ) are given in bold

|                            | Plasma cortisol                               | Plasma lactate                                | Plasma glucose                                |
|----------------------------|---|---|---|
| Handling                   | d.f. = 2,45 $F < 0.001$ , $P = 1.000$         | d.f. = 2,45 $F = 18.200$ , $P = 0.095$        | d.f. = 2,45 $F = 0.171$ , $P = 0.844$         |
| Recovery time              | d.f. = <b>2,45</b> $F = 38.852$ , $P < 0.001$ | d.f. = <b>2,45</b> $F = 64.424$ , $P < 0.001$ | d.f. = <b>2,45</b> $F = 11.469$ , $P < 0.001$ |
| Handling $\times$ Recovery | d.f. = 4,45 $F = 0.970$ , $P = 0.433$         | d.f. = <b>4,45</b> $F = 2.788$ , $P = 0.038$  | d.f. = 4,45 $F = 2.189$ , $P = 0.085$         |

reached levels 2.3-fold higher than control values (Fig. 2b). Neither handling treatment, nor its interaction with recovery time, had any effect on plasma glucose levels (Table 1; Fig. 2b).

In contrast to cortisol and glucose, lactate concentrations for all handling treatments rose quickly and were above control levels at 45 min (Table 1; Fig. 2c). In addition, a significant interaction was found between handling and recovery time for levels of plasma lactate. Lactate concentrations for the low-stress and medium-stress recovery periods at 4 h were significantly lower than the medium-stress 45-min and high-stress 2-h treatment blocks (Fig. 2c). The only handling treatment where signs of recovery could be noted was the medium-stress treatment, which reached a peak at 45 min but had significantly declined from that peak by 4 h (Fig. 2c). Fish in both the high-stress and low-stress handling blocks reached their highest absolute mean lactate level 2 h post-capture. However, for each handling treatment, 2-h levels did not differ significantly from those at 45 min or 4 h.

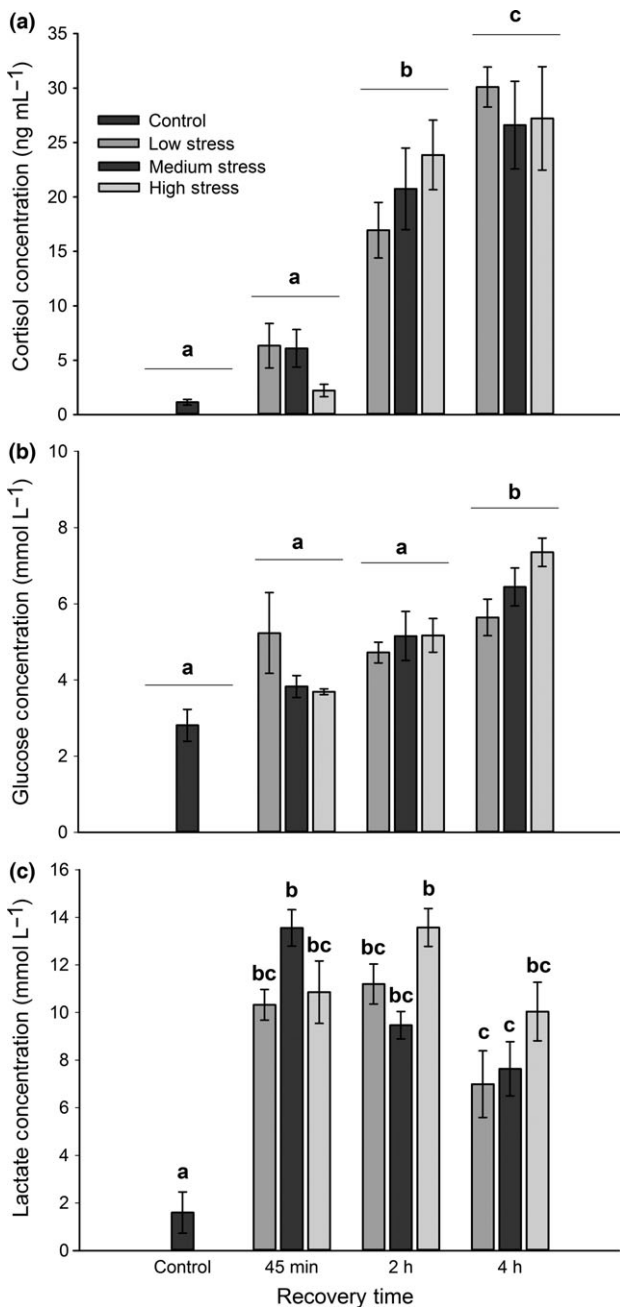
## Discussion

Northern pike angled through the ice and subsequently handled demonstrated elevated levels of cortisol, glucose and lactate. No studies have examined levels of plasma cortisol or lactate in northern pike following an angling event, although maximum mean plasma cortisol levels in the closely related muskellunge *Esox masquinongy* Mitchell following angling capture were found to be  $\sim 40$  ng mL<sup>-1</sup> (Landsman *et al.* 2011). In that study, however, blood samples were taken within 5 min of capture, meaning that cortisol levels likely had not had the chance to peak (Gesto *et al.* 2013). The maximum mean plasma cortisol concentration found in the present study (27.97 ng mL<sup>-1</sup>) is far lower than post-stress values (75–100 ng mL<sup>-1</sup>) found for adult sockeye salmon *Oncorhynchus nerka* (Walbaum) (Donaldson *et al.* 2011) and bluegill (Cousineau *et al.* 2014). Peak plasma glucose levels in the present study were also slightly lower than those found previously in northern pike (Arlinghaus

*et al.* 2009). Conversely, maximum plasma lactate concentrations were similar to those found in previous studies of peacock bass *Cichla ocellaris* Bloch & Schneider (Bower *et al.* 2016) and sockeye salmon *Oncorhynchus nerka* (Walbaum) (Gale *et al.* 2011). While cold temperatures may have served to mute the stress response, the low response could also be due to the low fight times for captured pike in this study (15 s or less) as physiological disturbance has been shown to correlate positively with fight time in angled fish (O'Toole *et al.* 2010, Meka & McCormick 2005). While direct comparisons to other studies should also be examined cautiously due to the different physiological responses to stress seen in different species (Pottinger 2010), the data presented here indicate that the magnitude of the stress response to ice-angling is less pronounced than that seen in conventionally angled fish.

In addition to a dampened stress response, rises in plasma cortisol and glucose levels were significantly delayed relative to that seen in previous studies. Several similar studies in which fish were held in static containers saw peak levels being reached within 1 h (Cook *et al.* 2011; Jeffrey *et al.* 2014). In the present study, plasma cortisol levels did not reach their highest levels until 4 h post-capture, and no indication of recovery was seen by 4 h for any of the metrics assessed. By comparison, prior work in warmer water has shown recovery to be complete by 4 h; for instance, a study of largemouth bass exercised at 10 °C showed complete recovery of plasma cortisol and glucose levels by 2 h (Suski *et al.* 2007) and work on northern pike has shown recovery of muscle lactate levels by 1 h (Arlinghaus *et al.* 2009). The course of recovery of plasma metrics may be directly dictated by temperature, possibly due to reduced enzymatic activity in cold conditions (Van Ham *et al.* 2003; Mills *et al.* 2015). For instance, a study of Atlantic salmon *Salmo salar* L., Wilkie *et al.* (1997) found recovery of plasma lactate levels by 4 h at relatively warmer temperatures (18° and 23 °C), but not at lower temperatures (12 °C). In contrast to plasma glucose and cortisol, plasma lactate levels in the present study did increase significantly above controls for all handling





**Figure 2.** Concentrations of (a) cortisol, (b) glucose and (c) lactate in the plasma of northern pike captured fish by handling treatment and recovery time. Letters indicate significant differences among the recovery times for cortisol and glucose. For lactate, owing to a significant handling × recovery interaction, differences between individual treatments are shown.

treatments by 45 min, but complete recovery did not occur by 4 h. While it is possible that holding fish in static tanks rather than in flowing water may have inhibited recovery (Milligan *et al.* 2000), prior work also suggests that flowing water recovery may induce

disturbances for sit-and-wait predators relative to static conditions (Suski *et al.* 2007), and the use of static containers for field studies such as this is not uncommon (Cook *et al.* 2011; Jeffrey *et al.* 2014). The general conclusion, therefore, is that the delay in recovery is the result of the angling and/or environmental conditions, meaning ice-angled fish will take longer to recover physiologically from capture than summer-angled fish.

The effect of handling time on the post-capture physiology of pike was rather muted; longer durations of air exposure did not have significant effects on cortisol or glucose levels. A significant interaction between handling treatment and recovery time indicates that longer periods of air exposure may have influenced plasma lactate concentrations, but the between-block comparisons yielded few significant differences. This lack of a handling effect on post-capture physiology was surprising, given that even 1 min of extra air exposure following either capture or capture simulation led to large physiological disturbances in many fish species (Cooke *et al.* 2002; Thompson *et al.* 2008). These disturbances led to alterations in behaviour and greater risk of mortality (Ferguson & Tufts 1992), but cooler conditions have been found to mitigate those effects (Gingerich *et al.* 2007). It is possible that, regardless of temperature, northern pike are simply resilient to the effects of extended air exposure relative to other fish species. This is evidenced by a previously described lack of air exposure (300 s) effect on the magnitude and recovery trajectory of plasma lactate and ions in angled northern pike, although it should be noted that behaviour post-capture was adversely impacted by additional air exposure (Arlinghaus *et al.* 2009). Further work will be necessary to determine whether the reduced sensitivity to additional handling time seen here is an across-the-board pattern for all ice-angled species, or whether this resilience is instead specific to northern pike.

Species- and context-specific knowledge of post-release physiology and behaviour in game fishes is critical to the informing of management strategies to maximise population health in exploited species (Cooke & Suski 2005). It should be noted that the overall goal of any of this work is to potentially quantify the likelihood of mortality occurring post-capture, which the present study also does not directly address. While no short-term (within 4 h) mortality was observed in any captured northern pike, the present study did not assess the possibility of delayed mortality, that is mortality taking place several hours or even days following capture (Davis 2002; Coggins *et al.* 2007). The low levels of physiological disturbance seen here combined with prior work indicating that colder water reduces the likelihood of mortality in both freshwater and marine species

(Bartholomew & Bohnsack 2005; Gingerich *et al.* 2007; Murphy *et al.* 2011) indicate that post-release mortality may be low in ice-angled and released northern pike. This conclusion, however, requires additional research to confirm, as physiological indicators have not always been found to be accurate predictors of mortality (Davis *et al.* 2001). It is also possible that the very low air and water temperatures as seen in the present study may have induced cold shock and/or damage to tissues (particularly the gills), which may have led to delayed mortality which was not predicted by the physiological variables measured (Donaldson *et al.* 2008). With this in mind, it is still suggested that managers advise anglers to minimise the time captured fish are exposed to air to minimise physiological disturbance (Cook *et al.* 2015). Given the current data, it appears likely that if efficient handling is practiced by anglers, then overall physiological disturbance in captured fish can be kept to a relatively low level.

### Acknowledgments

We would like to thank Chris Bruce for his guidance in the field during this project and his skilful use of ice-angling techniques and gears, which greatly increased our ability to conduct this study. We also thank Ryan Solomon for his assistance with the field work and sample processing. Funding for this research was provided in part by Federal Aid in Sport Fish Restoration Project F-69-R-29 to J.A.S.

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