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Northern pike bycatch in an inland commercial hoop net fishery: Effects of water temperature and net tending frequency on injury, physiology, and survival

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ABSTRACT

In lakes and rivers of eastern Ontario (Canada) commercial fishers use hoop nets to target a variety of fishes, but incidentally capture non-target (i.e., bycatch) gamefish species such as northern pike (*Esox lucius*). Little is known about the consequences of bycatch in inland commercial fisheries, making it difficult to identify regulatory options. Regulations that limit fishing during warmer periods and that require frequent net tending have been proposed as possible strategies to reduce bycatch mortality. Using northern pike as a model, we conducted experiments during two thermal periods (mid-April: $14.45 \pm 0.32\,^{\circ}C$, and late May: $17.17 \pm 0.08\,^{\circ}C$) where fish were retained in nets for 2 d and 6 d. A '0 d' control group consisted of northern pike that were angled, immediately sampled and released. We evaluated injury, physiological status and mortality after the prescribed net retention period and for the surviving fish used radio telemetry with manual tracking to monitor delayed post-release mortality. Our experiments revealed that injury levels, in-net mortality, and post-release mortality tended to increase with net set duration and at higher temperatures. Pike exhibited signs of chronic stress and starvation following retention, particularly at higher temperatures. Total mortality rates were negligible for the 2 d holding period at 14 °C, 14% for 6 d holding at 14 °C, 21% for 2 d holding at 17 °C, and 58% for 6 d holding at 17 °C. No mortality was observed in control fish. Collectively, these data reveal that frequent net tending, particularly at warmer temperatures, may be useful for conserving gamefish populations captured as bycatch in inland hoop net fisheries.

1. Introduction

The conservation literature contains a vast number of papers that examine the magnitude and consequences of bycatch in marine waters (reviewed in Hall, 1996; Hall et al., 2000; Davis, 2002; Lewison et al., 2004). That knowledge base has been used to develop, test, and implement a variety of strategies that have reduced bycatch rates, as well as injury to and mortality of bycatch (Crowder and Murawski, 1998; Hall and Mainprize, 2005; Cox et al., 2007). Comparatively little is known about bycatch in inland (freshwater) fisheries. A recent review (Raby et al., 2011) revealed that bycatch does occur in inland fisheries around the globe but that there is a need to document its extent and develop mitigation strategies. Freshwater ecosystems are some of the most threatened on the planet (Ricciardi and Rasmussen, 1999; Jenkins, 2003) and freshwater fish are among the most imperilled taxa (Richter et al., 1997) so there is a need for research on the fate of discarded fish, some of which are economically important gamefish species.

Physiological tools and biotelemetry could be applied to the study of bycatch issues in inland waters with relative ease and could help to identify problems and solutions. Relatedly, there has been extensive research in freshwater recreational fisheries documenting the effects of catch-and-release on fish injury, physiology, and survival (Arlinghaus et al., 2007a). In particular, field physiology techniques (Cooke and Suski, 2005; Cooke et al., in press) and biotelemetry (Donaldson et al., 2008) have been used successfully to refine recreational fishing practices, more so than in commercial bycatch research in the marine environment (Davis, 2002). Most research on the post-release mortality of commercial bycatch has used inherently limited artificial holding environments (e.g., Neilson et al., 1989; Farrell et al., 2001; Davis, 2002; Marcalo et al.,

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2009) to monitor mortality, whereas biotelemetry permits monitoring of animals released into the wild (Donaldson et al., 2008). Likewise, the discipline of "conservation physiology" has emerged (Wikelski and Cooke, 2006; Seebacher and Franklin, 2012), and physiological tools can be used to reveal sub-lethal effects (Meka and McCormick, 2005; reviewed in Cooke et al., in press), to objectively quantify animal welfare (Arlinghaus et al., 2007b; Diggles et al., 2011), and to support mortality findings by illustrating mechanisms (e.g., Donaldson et al., 2011; reviewed in Cooke et al., in press). However, physiological tools have scarcely been used in freshwater bycatch. A recent literature survey identified only two freshwater bycatch studies that had incorporated physiological techniques (Raby et al., 2011).

In eastern Ontario, commercial fishers use hoop nets to target a variety of fishes, including sunfish (*Lepomis* spp.), crappie (*Pomoxis* spp.), and bullhead (*Ameiurus* spp.). A variety of gamefish species are captured incidentally, including black basses (*Micropterus* spp.) and northern pike (*Esox lucius*), all of which must be released as per the Ontario Ministry of Natural Resources Commercial Fishing Regulations. Regulators have a variety of options for managing bycatch. Gear modifications that reduce bycatch rates have been developed and tested extensively in the marine realm (e.g., Broadhurst, 2000; Cox et al., 2007) and in some instances in freshwater fisheries (Koed and Dieperink, 1999; Lowry et al., 2005; Gessner and Arndt, 2006; Fratto et al., 2008). Regulations in freshwater more commonly focus on seasonal restrictions (Bettoli and Scholten, 2006) or other policies related to fisher behaviour. For example, seasonal restrictions have been used to minimize bycatch during fish spawning periods or during warm periods when bycatch mortality may be higher (e.g., Johnson et al., 2004). Another regulatory strategy relates to the frequency at which nets are tended and emptied of bycatch. Hybrid regulations in which longer net tending periods are permissible at cooler temperatures and shorter net tending periods are required at higher temperatures may also be possible. However, there are no data in the peer-reviewed literature regarding such regulatory options and whether they would indeed reduce fish bycatch mortality and currently, there are no province-wide regulatory restrictions on either net set duration or fishing at high temperatures for inland hoop net fisheries in Ontario.

Using northern pike as a model, we examined the consequences of temperature and net tending frequency on the injury rate, physiological status, and mortality of fish held in hoop nets. In addition to monitoring in-net mortality, we used biotelemetry to observe post-release behaviour and estimate survival. We contrasted mid-April and late May (two thermal periods) and two net retention durations (2 d and 6 d) that have been identified as potential regulatory options in Ontario. Based on anecdotal evidence, commercial fishers in the area typically pull nets within 2–7 days. We predicted that injury, physiological stress, and mortality would each be greater at higher water temperatures (late May) and longer net set durations (6 d). Previous research has indicated that water temperature tends to be positively correlated with capture mortality (reviewed by Gale et al., in press). Similarly, it is well established that chronic confinement can be a stressor for wild fishes (reviewed by Portz et al., 2006), although the effects of confinement stress have not been well studied in the context of capture fisheries, particularly in freshwater. Northern pike were used because earlier pilot studies had indicated that they were commonly captured in hoop nets and were relatively susceptible to mortality (Larocque et al., 2012), and because they are an economically important sport fish in parts of North America and Europe. Unfortunately there are no bycatch monitoring programs for hoop net fisheries in Ontario inland from the Laurentian Great Lakes, so we have no information on the potential broader scope of this problem. Northern pike are widely distributed in temperate regions of North America and Europe and are likely encountered in the variety of passive commercial fishing gear types used; particularly traps such as hoop nets (also known as fyke nets), so our findings should be relevant beyond eastern Ontario. Moreover, given that this is one of the first studies to adopt an experimental approach to bycatch research in inland waters that incorporates sublethal and lethal endpoints, we hope that our study will serve as a model for future inland bycatch studies.

2. Materials and methods

2.1. Study site and fish capture

We conducted the study in Lake Opinicon in eastern Ontario, Canada (44°31′N, 76°20′W; mean depth = 4.5 m, surface area = 787 ha; Arlinghaus et al., 2009) in the vicinity of the Queen's University Biological Station during two thermally distinct segments of the spring fishing season: May 23–June 29 (in 2009) and April 15–May 12 (in 2010). Water temperature data were obtained from a temperature probe installed in the lake at a depth of 3.3 m from which readings were automatically recorded each day. We captured the 76 northern pike used for this study by angling from a boat using both trolling and active casting. We angled fish using a variety of artificial lures with one or more barbless treble hooks and we landed the fish as quickly as possible (≤60 s) to minimize capture stress. We landed all fish using smooth rubber landing nets to minimize dermal injury and we quickly de-hooked fish using pliers. Once landed, northern pike were held in large water-filled coolers (1 m × 0.40 m × 0.40 m) that were frequently replenished with lake water (every ~10 min). The reason for angling fish was to eliminate the variance associated with using the hoopnets to catch fish and the range of times fish would actually enter the net. All fish were angled in this study and angling constitutes an additional acute stressor that would not be present in a hoop net fishery. However, previous studies using northern pike in the same lake and across a similar range of water temperatures revealed that northern pike could be angled with minimal injury (Arlinghaus et al., 2008), physiological recovery was complete within 24 h (Arlinghaus et al., 2009), and that immediate and delayed mortality were negligible (Arlinghaus et al., 2008, 2009).

2.2. Hoop net treatment

We used two hoop net treatments whereby fish were captured by angling and placed in hoop nets that were set for either 2 d or 6 d. We used 10 identical double-ended hoop nets set within 20 m of shore and submerged at 2–3 m depths (see Larocque et al., 2012). All nets were set adjacent to one-another in the same part of the lake. Each hoop net comprised seven 0.91 m diameter steel or wooden hoops positioned 0.5 m apart with two throats per net at the second and fourth hoops. Nets were set by adjoining two hoop nets by their 10.97 m long and 0.91 m high leads with the wings (4.57 m long, 0.91 m high) sewn across the entrance of each net to prevent fish or other taxa from entering (and study animals from escaping). The hoop nets were made from 2.54 cm diamond nylon stretch mesh. Angled pike were placed in the hoop nets through the anchored end (i.e., directly into the last chamber) at a density of up to 4 fish per net. Once northern pike were inserted into nets, the nets were tied shut, re-set and left untouched for either 2 or 6 d. Fish were thus captured by angling for insertion into hoop nets on two dates in each year: April 15 and 19 in 2010 (water temperature = 15.82 °C and 13.91 °C, respectively), and May 23 and 27 in 2009 (16.81 °C and 17.46 °C). The nets were visually inspected daily by snorkelling to monitor mortality rates. Five nets were used each for the 2 and 6 d treatments, both in the warmer (2009) and cooler (2010) treatments.

Our hoop net treatment was modeled after commercial fishing methods used in the area by a fisher who we accompanied prior to this study while he operated his fishery. The two differences between our study and a true fishery were that (a) northern pike in our study were subjected to experimenter handling in the form of capture by angling, manual loading into nets, tagging and biopsy (see below) and (b) only northern pike (1–4 fish) were in the nets during retention. Based on a parallel study of freshwater bycatch (not focused on northern pike) using the same gear in an adjacent lake (Larocque et al., 2012), northern pike were commonly encountered, representing 2.6% of all fish captured (202 of 7702 fish) but accounted for ~20% of all fish bycatch. Pike were typically encountered in net sets of ≤6 days at densities between one and six fish per net. Although northern pike density was similar in our study, the total density of fish was far less because we sewed nets shut in order to prevent study animals from leaving the nets (to control retention duration). Lower overall density in the present study may have resulted in lower stress for northern pike than in a true fishery, although we are unaware of any studies on pike or other fish captured in fyke nets to support that supposition. Additionally, any bias towards more favourable conditions for the animals was likely offset somewhat by the added stress of angling and experimenter handling. In general, density was higher in the warmer treatment (mean 3.1 pike per net in 2009, 1.3 in 2010) because higher mortality rates meant more fish were needed in order to obtain sufficient sample sizes for radio telemetry tagging. However, as mentioned, relative to an operational fishery the density in both thermal periods was very low: 1–4 fish in a net rather than a crowded ~30–80 (Larocque et al., 2012).

2.3. Sampling and tagging

Following the experimental net treatment (2 or 6 d), the nets were pulled for processing, tagging, and release on one day in each of the two study periods: April 21 in 2010 (water temperature = 13.87 °C) and May 29 in 2009 (17.19 °C). Upon pulling nets, live northern pike were removed and held supine in a foam-padded v-shaped sampling trough filled with fresh lake water. A blood sample was quickly drawn from each fish using caudal puncture with a 3.8 cm, 21-gauge needle and a 2 ml heparinized vacutainer (lithium heparin, Becton-Dickson, NJ, USA). Blood samples were stored in ice slurry for up to 1 h until they could be analyzed. Following sampling, unique-frequency radio-telemetry transmitters (3.2 g in air, 25 x 13 x 6 mm, 120 mm antenna, battery life 6 months; Model PD-2, Holohil Systems Inc., Carp, ON, Canada) were externally affixed to surviving fish (following Arlinghaus et al., 2009). In 2009 (the warmer treatment) a subset of the surviving fish was tagged while in 2010 every surviving individual was radio tagged and released. To tag fish, two 22-gauge hypodermic needles mounted on 3 ml syringes were pushed through the dorsal musculature ventral and posterior to the origin of the dorsal fin. Wires attached to the transmitter (20-gauge surgical stainless steel) were threaded through the needles and pulled out on the opposite side of the fish through a small (10 mm × 5 mm × 1 mm) backing plate made from rubber gasket material. The wires were twisted carefully until the transmitter was snug to the fish and trimmed to minimize potential for snagging on macrophytes. No anaesthetic was used during the transmitter attachment. Following biopsy and tagging, each fish was measured for total length (mm), and injury was macroscopically evaluated with ratings for scale loss and fin fraying (both rated separately as 0 = none, 1 = minimal, 2 = severe). For each fish we also noted whether lip damage (i.e., from interaction with net mesh or fishing lures) was present and estimated percent body covered by fungal infection, if visible.

During both study periods, we also obtained baseline "control" physiological samples by angling and immediately blood sampling northern pike (within 2 min of hooking). We regard those field controls as the closest-as-possible to baseline value for free-swimming northern pike in the lake. The 'time 0' sample represents the culmination of angling capture and short term holding in coolers prior to tagging and release. For sampling fish from hoop nets we had large sampling teams (as many as seven people in the boat) such that we could pull the nets rapidly and blood sample multiple fish simultaneously (within 2 min of beginning to pull the net) to ensure that physiological measurements reflected the condition of the fish in the net prior to it being pulled (Barton, 2002).

Northern pike were blood sampled, telemetry tagged, and released on the same day, so that 6 d net treatment fish had been angled and placed in nets six days previously, with 2 d fish captured two days previously. Thus, there was one biopsy-tagging-release day on April 21 in 2010 (13.87 °C) and one on May 29 in 2009 (17.19 °C). Control fish were angled ($n = 9$ for each study period), held in coolers (2–3 h), sampled, tagged, and released throughout the tagging day concurrent with releases of tagged fish that were subject to hoop net treatments. Throughout the release day, radio-tagged northern pike were released from the same 25 m length of shoreline adjacent to the Queen's University Biological Station in series of three to five fish at a time (~5 m apart), always including at least one control fish.

Upon release of each radio tagged fish, we recorded the time required to move 100 m away from the release site using a combination of visual observation and manual radio tracking (see Thompson et al., 2008). This was done in order to evaluate whether there were any behavioural impairments for fish held in hoop nets relative to control fish simply captured by angling. Radio tracking was conducted with a hand-held radio receiver (R1000 Telemetry Receiver, Communications Specialists Inc., Orange, California, USA) and a three-element Yagi antenna. Beginning the day following release, we manually tracked fish by boat for seven days. Tracking occurred between 08:00 and 20:00 and we attempted to get one position per fish per day. We tracked individuals to within 10 m and recorded UTM coordinates using a handheld GPS (Garmin E-Trex). In 2009, following the initial 7-day tracking period, tracking was conducted every two days for an additional two weeks (three weeks in total). In 2010, following the 7-d tracking period, tracking occurred every four days for an additional two weeks (three weeks in total). If a tracked fish was a suspected to have died (i.e., did not move for consecutive days), we used an underwater antenna to gain a more precise location and a snorkeler tried to visualize the animal to assess whether it was alive.

2.4. Physiological analyses

Analyses were conducted on blood samples at the Queen's University Biological Station within 1 h of sampling. Handheld field meters were used to measure glucose (ACCU-CHEK glucose meter; Roche Diagnostics, Basel, Switzerland) and lactate using whole blood (Lactate Pro™ LT-1710 Blood Lactate Test Meter, Arkray Inc., Kyoto, Japan). Both instruments had previously been validated for use in fish (Venn Beecham et al., 2006; Cooke et al., 2008). Blood glucose was chosen as an indicator of stress given that, in fishes, cortisol acts to mobilize glucose stores (i.e. gluconeogenesis; Wendelaar-Bonga, 1997). Blood lactate was used as an indicator of metabolic exhaustion that increases following anaerobic exercise and can remain elevated following capture given a chronic confinement stressor (Farrell et al., 2001; Donaldson et al., 2011). We measured hematocrit with microhematocrit capillary tubes centrifuged for 5 min (using a CritSpin-Micro-Hematocrit Centrifuge) and calculated the percentage of red blood cells within the total volume of blood. The remaining blood was centrifuged at 10,000 × g for 5 min (Compact II Centrifuge, Clay Adams, NJ) and then ∼0.1 ml of plasma was used to measure plasma protein using a hand-held refractometer. Hematocrit was measured as an indicator for potential blood loss (Barton, 2002), while plasma protein was used to monitor potential starvation of fish retained in nets. The proteins measured by the refractometer used are known to decrease during starvation (Navarro and Gutiérrez, 1995).

2.5. Data analysis and statistics

In order to confirm that sizes of northern pike were consistent among treatment groups, we first tested for differences in fish size (total length) among treatments using a two-way analysis of variance (ANOVA) with temperature and net set duration as independent variables. Kruskal-Wallis ANOVA was used to test for the effects of both temperature and net set duration on ordinal injury scores (for fin fray and for scale loss). We observed that upper jaw injuries were occurring as a result of entanglement on the mesh of hoop nets (and not caused by angling). Therefore, we used contingency table analysis with Pearson's Chi-square test to determine if temperature and net set duration had significant effects on the occurrence of upper jaw injury. For physiological data, we tested for the effects of water temperature and net set duration on blood glucose, blood lactate, hematocrit, and plasma protein using two-way ANOVAs with temperature and net set duration as independent variables. When ANOVAs were significant, we used Tukey's HSD post hoc tests to identify differences. For dispersal data, we used temperature and net set duration as predicting factors in a two-way ANOVA with time to swim more than 100 m from the release site as the dependent variable. To confirm water temperature was significantly warmer during our "warm" thermal period, we performed a Student's *t*-test on daily mean temperatures for the capture-retention-release period for each of the two years (7 daily means for both years).

Finally, we used multiple logistic regression assess what factors affected survival. For overall survival (in-net and post-release combined) we explored whether there were effects of net tending frequency (0, 2, and 6 d) and temperature (14 and 17). For fish released using radio transmitters (both years pooled), we analyzed the effects of potential individual-level predictors of post-release mortality (total $n = 44$): scale loss score, fin fray score, presence of external fungus, blood glucose, blood lactate, plasma protein, and whether the fish exhibited negative orientation upon release, on post-release survival (fish that died during retention were not assessed for injury or physiology). Data were $\log_{10}$-transformed to meet the assumptions of normality as needed based on assessment using a Shapiro-Wilk's test. All statistical tests were conducted using JMP (V. 4.0, SAS Institute, Cary, NC) software and α was set at 0.05. We report all means ± SE.

3. Results

Overall, 76 northern pike were processed across the two thermal periods and three net set durations and there was no significant difference among the sizes of fish in each treatment group (Table 1). Water temperatures were significantly higher during the warmer late May study period (Student's *t*-test: $t = -8.16$, $P < 0.0001$): during capture, retention in nets, processing, and release, mean daily water temperatures (at 3.3 m depth) were 14.45 ± 0.32 °C in the earlier mid-April period (April 15–21, 2010) and 17.17 ± 0.08 °C in the warmer late May period (May 23–29, 2009). During the post-release radio tracking period, mean water temperatures for the cooler (April 22–May 12, 2010) and warmer periods (May 30–June 29, 2009) were 18.71 ± 0.64 °C and 19.93 ± 0.42 °C, respectively ($t = -1.67$, $P = 0.10$).

3.1. Injury

Rates of injury increased in both frequency and severity as both net set duration and water temperature increased. For fish held in both thermal periods, ≥80% of individuals exhibited some degree of scale loss and caudal fin fray regardless of the net set duration (2 vs. 6 day) (Figs. 1 and 2). Both fin fray and scale loss injury scores were significantly different among net set durations (Kruskal-Wallis ANOVA, $H_{2,60} = 33.08$, $P < 0.001$ and $H_{2,60} = 23.49$, $P < 0.001$, respectively). Fin fray score was significantly higher for

Table 1
Comparison of biological variables for northern pike held in two different thermal periods (mid-April and late May - 14 and 17 °C) for either 0, 2 or 6 days in hoop nets (mean ± S.E.). Baseline concentrations represent separate fish that were angled and immediately blood sampled. Analyses were conducted using two-way ANOVA with temperature and net set duration as the two independent variables. Italicized statistical output indicates significant models (at $\alpha = 0.05$). Dissimilar superscript (^{a,b,c}) indicates significant differences among durations (in the duration column). In the case of interactions, significant differences are denoted by dissimilar superscript only for the interaction terms.

Variables	Net set duration (days)	Mid-April 14 °C	*N*	Late May 17 °C	*N*	ANOVA output		
						Temperature	Duration	Interaction
Total length (mm)	Baseline	496.8 ± 1.7	4	475.7 ± 23.7	11	$F = 2.68$, $P = 0.106$	$F = 0.96$, $P = 0.417$	$F = 0.22$, $P = 0.884$
	0	511.0 ± 19.3	9	480.8 ± 30.7	9			
	2	493.0 ± 27.5	5	490.8 ± 16.6	19			
	6	543.3 ± 22.7	6	507.2 ± 20.1	12			
Blood glucose (mmol l^{-1})	Baseline	3.3 ± 0.3^{a,b}	4	3.00 ± 0.2^{b}	10	$F = 0.04$, $P = 0.85$	*$F = 21.46$, $P < 0.0001$*	*$F = 4.24$, $P = 0.010$*
	0	11.9 ± 0.9^{c,d}	9	9.8 ± 1.0^{c,d,e}	9			
	2	8.78 ± 4.5^{a,d,e}	5	12.9 ± 1.4^{c}	18			
	6	7.27 ± 0.7^{a,c,d,e}	6	5.8 ± 0.6^{a,e}	10			
Blood Lactate (mmol l^{-1})	Baseline ^{a}	4.6 ± 1.1	4	3.6 ± 0.7	10	$F = 0.96$, $P = 0.331$	*$F = 12.41$, $P < 0.0001$*	$F = 1.63$, $P = 0.192$
	0 ^{b}	11.2 ± 0.6	9	10.2 ± 1.3	9			
	2 ^{b}	5.3 ± 2.6	5	8.1 ± 0.7	17			
	6 ^{b}	4.9 ± 1.6	6	7.1 ± 0.8	11			
Hematocrit (% PCV)	Baseline	18.5 ± 1.7	4	22.9 ± 1.6	10	*$F = 4.18$, $P = 0.045$*	$F = 2.11$, $P = 0.107$	$F = 1.83$, $P = 0.151$
	0	18.1 ± 1.6	9	16.93 ± 0.1	9			
	2	19.0 ± 1.0	5	21.0 ± 0.8	18			
	6	17.8 ± 1.6	6	22.0 ± 1.8	10			
Plasma protein (mmol l^{-1})	Baseline	5.2 ± 0.4^{a}	4	3.5 ± 0.3^{b,c}	10	*$F = 42.45$, $P < 0.0001$*	$F = 1.20$, $P = 0.317$	*$F = 4.77$, $P = 0.005$*
	0	4.5 ± 0.2^{a,b}	9	3.6 ± 0.2^{b,c}	9			
	2	4.3 ± 0.3^{a,b}	5	4.1 ± 0.2^{a,b}	18			
	6	4.9 ± 0.1^{a}	6	2.8 ± 0.3^{c}	10			

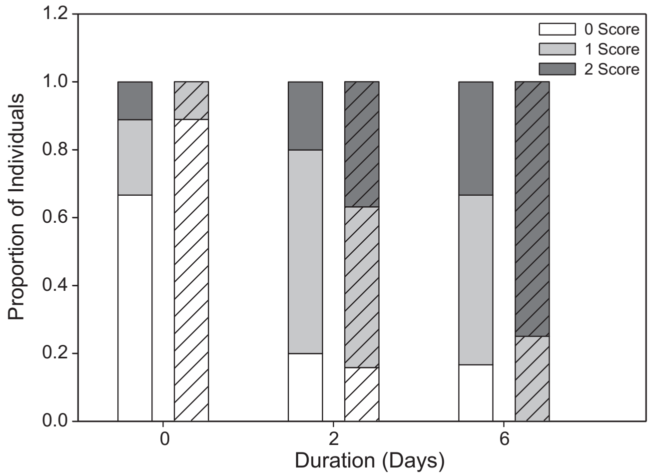

Fig. 1. The proportion of individuals in each treatment group exhibiting different levels of scale loss. Score 0 = None; Score 1 = Minimal; Score 2 = Severe. Non-hatched and hatched bars indicate northern pike captured and held at 14 °C (mid-April) and 17 °C (late May), respectively.

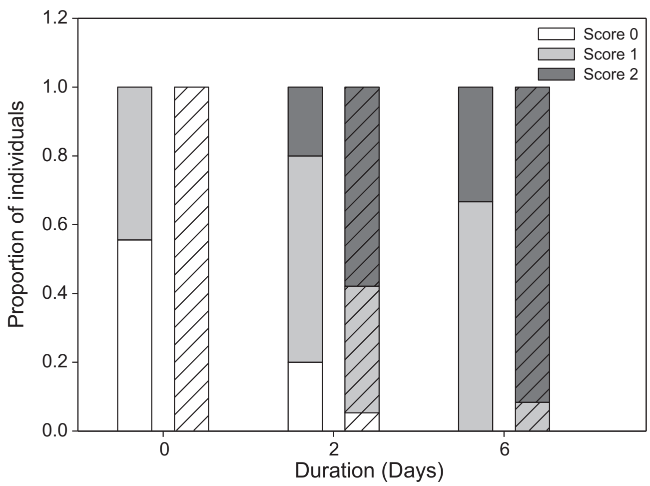

Fig. 2. The proportion of individuals in each treatment group exhibiting different levels of caudal fin fray. Score 0 = None; Score 1 = Minimal; Score 2 = Severe. Non-hatched and hatched bars indicate northern pike captured and held at 14 °C (mid-April) and 17 °C (late May), respectively.

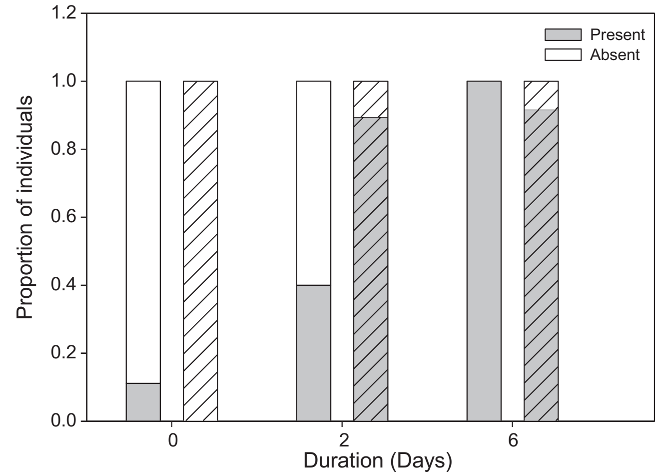

Fig. 3. The proportion of individuals in each treatment group in which damage to the upper jaw was present and absent. Non-hatched and hatched bars indicate northern pike captured and held at 14 °C (mid-April) and 17 °C (late May), respectively.

fish held in hoop nets in the warmer thermal period ($H_{1,60}$ = 4.67, P = 0.031), whereas scale loss was not significantly affected by temperature ($H_{1,60}$ = 2.14, P = 0.14). The frequency at which damage to the upper jaw was noted was significantly different among net set durations (Pearson's chi square, $\chi^2_{2,60} = 21.75$, $P < 0.001$) but not between temperatures ($\chi^2_{2,60} = 2.20$, $P > 0.10$) (Fig. 3). No fish were observed to have torn upper jaws at time of capture by rod and reel emphasizing that the large tears were associated with net retention. In general, hooking injuries resulting from angling were negligible such that there was no noticeable tearing of tissue or bleeding. Percent body affected by fungus was visually estimated and ranged from 2 to 10% for those fish where fungal infections were visible. The proportion of fish with fungal infections was 0.67 (8 of 12) for the 17 °C 6 d treatment, 0.29 (2 of 7) for the 14 °C 6 d treatment, and 0.21 for the 17 °C 2 d treatment with no fungal infections noted for other treatment groups (mean percent body coverage for individuals with fungal infections followed the same pattern among treatment groups: 7.5%, 2.5%, and 1.75%, respectively).

3.2. Mortality during retention

Only one northern pike in the study died during the cooler study period in mid-April (14 °C). This fish died while in the net and was part of the 6 d treatment (Table 2). Overall, 25% (N = 3/12) of northern pike held in the nets for 6 d at 17 °C (late May) died during the net retention portion of this study. Comparatively, there were no observed mortalities for fish that were simply angled and released (i.e., 0 d controls; N = 0/9) while 5.3% (N = 1/19) of fish died in the nets for the 2 d during the warmer late May period (17 °C).

3.3. Physiology

There was an interactive effect of temperature and net set duration on blood glucose and plasma protein (Table 1). Specifically, blood glucose levels were significantly lower for fish captured at 17 °C (late May) and held for 6 d in the nets, when compared to fish captured at 14 °C (mid-April) and held for 0 d in nets (controls) and fish captured at 17 °C and held for 2 d in the nets (Table 1). Plasma protein concentrations were significantly lower for fish captured at 17 °C and held in the nets for 6 d, when compared to all other treatments. Blood lactate values were significantly reduced with retention in the nets for both 2 d and 6 d, when compared with fish angled and held in coolers prior to tagging (i.e., 0 d; Table 1). There

Table 2
Pre-release, post-release and total mortality observed for northern pike held in hoops nets for 0, 2 and 6 days at two temperatures (in mid-April and late May).

Net retention duration (days)	Pre-release mortality % (N)		Post-release mortality % (N)		Total mortality % (N)	
	Mid-April 14 °C	Late May 17 °C	Mid-April 14 °C	Late May 17 °C	Mid-April 14 °C	Late May 17 °C
0	0.0% (N = 0/9)	0.0% (N = 0/9)	0.0% (N = 0/9)	0.0% (N = 0/9)	0.0% (N = 0/9)	0.0% (N = 0/9)
2	0.0% (N = 0/6)	5.3% (N = 1/19)	0.0% (N = 0/6)	33.3% (N = 3/9)	0.0% (N = 0/6)	21.1% (N = 4/19)
6	14.3% (N = 1/7)	25.0%(N = 3/12)	0.0% (N = 0/6)	57.1% (N = 4/7)	14.3% (N = 1/7)	58.3% (N = 7/12)

Table 3
Comparison of survivors with mortalities among northern pike released with radio transmitters, with all fish pooled across the three net set duration and two temperatures. For physiological measurements from blood and plasma, error terms represent standard error of the mean. For the remaining variables (presence of fungus, presence of upper lip rip injury, severe scale loss, severe fin fray, and loss of orientation) the values provided represent the proportion of fish exhibiting that state (e.g., the proportion of fish for which we noted external fungal growth). None of the variables listed were significant predictors of post-release mortality in a multiple logistic regression model ($P > 0.20$ for each variable).

Release measurement	Post-release survivors $N = 37$	Post-release mortalities $N = 7$
Blood glucose (mmol l^{-1})	9.36 ± 0.76	9.90 ± 2.29
Blood lactate (mmol l^{-1})	8.30 ± 0.67	6.13 ± 1.13
Plasma protein (mmol l^{-1})	4.08 ± 0.14	2.84 ± 0.36
Presence of fungus	0.09	0.43
Presence of upper lip rip	0.39	0.71
Severe scale loss (score of 2)	0.11	0.43
Severe fin fray (score of 2)	0.13	0.57
Loss of orientation	0	0.57

was a significant effect of temperature on hematocrit; however, there was no significant effect of net set duration or interaction between temperature and net set duration (Table 1).

3.4. Post-release behaviour and mortality

Most fish left the release site within 10 min, regardless of treatment group or water temperature. Overall, 94.0% of fish were >100 m from the release site within 8 h of release. There was an overall effect of holding time on time required for fish to swim more than 100 m from the release site (Two-way ANOVA, $F_{2,36} = 3.37$, $P = 0.046$; 6 d treatment fish dispersed the fastest), but not of temperature ($F_{1,36} = 3.04$, $P = 0.09$), and there was no interaction between the two ($F_{2,36} = 0.29$, $P = 0.75$). Post hoc comparisons of dispersal rate among the six duration/temperature combinations were not significant (all $P > 0.05$). No post-release mortality was observed in control fish or during the cooler thermal period and mortality rates were highest for fish held in the warmer treatment for 6 d (Table 2). Total mortality (representing combined initial and delayed mortality) differed by treatments and followed the same general pattern as observed for retention and post-release mortality (Table 2). The post-release mortality observed during the warmer thermal period (17 °C) occurred largely on the release day: two each for the 2 d and 6 d treatments. The third (and final) post-release mortality in the 2 d treatment occurred 12 days after release, while further delayed mortality for the warm 6 d treatment occurred two and seven days after release. Scale loss, fin fray, presence of external fungus, loss of orientation upon release, blood glucose, blood lactate, and plasma protein were not significant predictors of survival (multiple logistic regression, all $P > 0.20$) although there were only seven mortalities out of 44 tagged fish (over two years) from which to draw statistical conclusions (see Table 3). When combining the in-net period with the post-release period, net set duration had a significant negative effect on survival ($\beta = -0.539$, Wald statistic = 9.309, odds ratio = 0.583, $P = 0.002$), as did water temperature – although to a weaker extent ($\beta = -2.461$, Wald statistic = 4.454, odds ratio = 0.085, $P = 0.035$).

4. Discussion

The present study is one of the first to have examined the effects of commercial fisheries bycatch on gamefish in freshwater (Raby et al., 2011). Moreover, this study is one of the few to combine a variety of biological outcomes including injury, physiological status, behaviour, and mortality using an experimental approach for the study of bycatch in either marine or freshwater habitats. In accordance with our predictions, longer retention times of northern pike in hoop nets generally increased the level of injury (e.g., scale loss and fin fraying), physiological disturbances, and mortality. Furthermore, the outcomes were modulated by water temperature: fish held at 17 °C in late May tended to fare more poorly than fish held at 14 °C in mid-April. Herein we discuss the patterns of injury, physiology, and mortality observed in relation to experimental treatment groups before outlining potential relevance for management of freshwater fisheries.

4.1. Injury

Relative to control fish captured by angling, fish held in hoop nets exhibited more fin fraying, scale loss, and damage to their jaws. Fin fraying and scale loss are frequently used as macroscopic indicators of fish injury and condition in studies of bycatch and catch-and-release angling (Cooke and Hogle, 2000; Barthel et al., 2003). Previous studies have found that scale loss and fin fray in smallmouth bass (*Micropterus dolomieu*) and bluegill (*Lepomis macrochirus*), caused by net mesh similar to that used in the present study, was associated with post-release mortality (Cooke and Hogle, 2000; Barthel et al., 2003). Scales and fins are perfused tissues so blood loss is possible, while dermal abrasion and slime loss can serve as an entry route for opportunistic pathogens (Roberts and Bullock, 1980). In freshwater, water moulds are ubiquitous and have been implicated in the mortality of fish following seemingly benign injuries that are not directly lethal in the short term (Van West, 2006). The rate of growth of *Saprolegnia* spp. infections is temperature dependent (Van West, 2006). In our study, it was noted that by six days the majority of fish in the warmer treatment did exhibit some fungal development on their skin. Another injury that we observed was tearing of the upper jaw, an injury that was not the result of the initial angling. Rather, while snorkelling the nets and when pulling the nets we observed that the upper jaws of northern pike were frequently caught on the mesh. Fish would either thrash to free themselves, or were disentangled by the researchers: both of which resulted in some level of tearing. We have not observed similar jaw injuries in other species that we have encountered in hoop nets (e.g., lepomids, *Micropterus* spp., catostomids) suggesting that this phenomenon is specific to northern pike (and likely other species of the Esocidae) and may be a result of their anatomy and behaviour in the net. Use of smaller mesh may reduce the frequency of such injuries, particularly if used for the terminal compartment of the net, but that possibility remains to be tested. We are unsure what the long-term consequences of such injuries are on pike and their ability to obtain food and respire efficiently.

4.2. Blood physiology

Not surprisingly, the data for time-0 fish reflect what would be expected after exposing fish to an acute exercise and handling stressor (Kieffer, 2000) – lactate and glucose were elevated relative to the field controls. However, given that both glucose and lactate associated with a capture stressor should be recovered by 24 h (Kieffer, 2000; Arlinghaus et al., 2009), it was noteworthy that both lactate and glucose remained elevated relative to field control levels after 2 or 6 d of confinement in hoop nets. The elevated blood lactate concentrations may have been indicative of the fish actively attempting to escape, at times with their jaws being caught in the mesh, which could promote additional struggling that would constitute anaerobic exercise (Kieffer, 2000). Glucose levels in fish retained in hoop nets may have been elevated as a result of the stress associated with use of anaerobic pathways or could have resulted from crowding stress and mobilization of energy via stimulation by cortisol (Wendelaar-Bonga, 1997; Barton, 2002). Glucose can also be used to evaluate starvation, although its typical response is to decrease during starvation, not to increase (Barton, 2002). Plasma total protein levels indicate the amount of albumin and globulin molecules present in the blood; these molecules are known to decrease in concentration in plasma during starvation (Navarro and Gutiérrez, 1995; Congleton and Wagner, 2006; Hanson and Cooke, 2009). We noted that plasma protein levels were lowest for fish captured at 17 °C and retained for 6 d; possibly indicating those fish were beginning to experience starvation after six days of presumably little or no feeding. The reason for not observing the same trend for fish held for 6 d at the cooler temperature (in mid-April) likely relates to the increased energetic demands (Fry, 1971) as well as more rapid digestion of any pre-treatment gut contents at higher temperatures (Brett, 1995) which would lead to a more rapid depletion of plasma protein levels (Navarro and Gutiérrez, 1995). One key difference between our study and the fishery itself was the absence of other fishes in the hoop net given that we sewed nets closed to control retention. It is not known whether potential starvation effects would be offset by feeding opportunities while in a net crowded with live prey items or whether crowding in a true fishery would significantly elevate indices of stress. Our overall finding that stress resulted from confinement in hoop nets is not a controversial one – there is a wealth of literature demonstrating that confinement is a stressor for wild fishes (reviewed by Portz et al., 2006), though the present study is the first we are aware of to do so in the context of a freshwater commercial fishery.

The physiological measures used in this study were not predictive of post-release mortality (see Section 3) although low sample sizes likely precluded a rigorous assessment of mortality predictors (but see Table 3). Links between post-release fisheries mortality and blood constituents are often elusive (e.g., Davis et al., 2001; Davis and Schreck, 2005) and most such research has focused on acute capture stressors rather than the chronic stress associated with multiple days of confinement in a trap net. It is possible that other measures of stress not used here could have better predicted mortality (e.g., plasma cortisol, genomic indices, plasma ions and osmolality; Cooke et al., in press). Macroscopic indices of injury and vitality may be better predictors of delayed fisheries mortality (e.g., Campbell et al., 2010); although they were not statistically significant predictors in the present study (Table 3). There was a tendency for fish that suffered post-release mortality to more frequently exhibit injury and fungal infection upon release, in addition to having negative orientation. All four fish that exhibited negative orientation upon release subsequently died based on our radio tracking, lending some evidence to the idea that simple reflex measures can be effective predictors of delayed mortality (Davis, 2010; Raby et al., 2012; see Table 3).

4.3. Mortality

Some northern pike died during retention in hoop nets prior to release, with in-net mortality patterns suggesting duration was particularly influential. We used the state of fish post-angling that were held for 1 h as controls and observed no mortality among those fish at either temperature. Our net retention mortality rates were generally low for the 2 d period (0% at 14 °C and 5.3% at 17 °C). However, mortality rates were higher after 6 d of retention. At 14 °C (mid-April), mortality rates were 14.3% and at 17 °C (late May) they were 25%. Considering that mortality in control fish was 0% and that previous work on northern pike captured recreationally and held in tanks in Lake Opinicon was also 0% (Arlinghaus et al., 2009), these data suggest that some of the treatments that we tested have the potential to generate substantial pre-release bycatch mortality. No dataset exists on the tending frequency used by commercial fishers for this fishery, but anecdotally we can report that fishers do sometimes use net set durations longer than six days.

In addition to documenting mortality during retention, biotelemetry was used to evaluate post-release behaviour and mortality (Donaldson et al., 2008). Given that post-release behaviour was monitored for only 8 h and within 100 m of the release site, it is perhaps not surprising that clear among-treatment patterns in behavioural impairment were not identified. Subsequent tracking was done to simply determine if fish had moved from their last position as a means of detecting mortality. We observed no delayed mortality among fish captured and retained in nets at 14 °C during the cooler mid-April period. However, during the warmer late May period (17 °C) we observed a 33% delayed mortality rate for fish released after 2 d of retention and a 57% delayed mortality for fish released after 6 d of retention. Although it is not possible to properly account for tagging effects, we did not observe delayed mortality in our control tagged fish, suggesting that tagging is unlikely to induce mortality. In a previous study of post-release behaviour of northern pike that were angled and released in Lake Opinicon, no mortality was noted, similarly suggesting that tagging is not a significant source of mortality (Arlinghaus et al., 2009). Aside from potential tagging effects, it is possible that the cumulative stress of angling capture combined with subsequent net retention could exacerbate stress – normally in a hoop net fishery pike would enter the net on their own volition without an acute angling stress. In addition, hoop nets in the present study were closed (i.e., they were not fishing) whereas in a fishing hoop net the final chamber of the net can become crowded with hundreds of fish (mostly other species; Larocque et al., 2012). It is unknown whether the extra crowding in an operational fishery would appreciably exacerbate stress and increase mortality rates. Given some differences between the experiences of the northern pike in our study and those of an operational fishery, the mortality rates generated should be interpreted cautiously, with more attention paid to mortality patterns than to the absolute rates.

Study limitations aside, when the retention mortality (above) is combined with the delayed mortality, the total mortality rates are more than 50% for fish held at 17 °C for 6 d (Table 2). Given that northern pike are economically important gamefish that are required to be released so that they can be targeted by the sports fishery, 50% mortality is likely a level of mortality that would not be desirable for managers, particularly if there are localized areas with high fishing pressure and bycatch of northern pike. Although the difference in mean water temperature between the two treatment periods (i.e., capture, net confinement, tagging) was modest (∼3 °C), it was apparently wide enough to produce markedly higher mortality rates. Water temperatures used in this study are moderate for northern pike (McCauley and Casselman, 1980). The warmer late May netting treatment occurred at approximately 17 °C which is ∼5–6 °C lower than the peak summer water temperatures observed in lakes in this region where pike are plentiful (S. J. Cooke, unpublished data). Therefore, based on our findings, we would expect to observe substantially higher bycatch mortality if fishing were to occur during peak summer temperatures.

4.4. Management considerations

The present findings may have relevant implications for fisheries managers. Clearly, this study did not comprise exhaustive sample sizes across multiple years and in multiple systems. Therefore, the results should be interpreted with caution and further research on the roles of temperature and net set duration in freshwater commercial fisheries would be warranted. Nevertheless, the trends observed were in accordance with predictions based on established phenomena (the effects of temperature and confinement) and would likely apply to other inland fisheries around the globe, particularly for coolwater and coldwater fishes. It is well established that levels of physiological disturbance and mortality tend to be greater at warmer water temperatures (Wilkie et al., 1997; Davis, 2002; Gale et al., in press), recognizing that thermal thresholds and tolerances vary among species (Cooke and Suski, 2005). We are not suggesting that the exact temperatures or retention periods used here are directly transferable to other fisheries and systems. However, if efforts that result in frequent net tending are adopted, particularly when water temperatures are warm, fish captured as bycatch should experience less injury, stress and mortality during retention, and when released exhibit less delayed mortality. Seasonal closures are a common regulatory tool although they tend to be used to focus fisheries activities away from fish spawning periods. Thermal thresholds are more difficult to implement and are less common, but they have been used in the recreational sector to protect upriver migrating Atlantic salmon (Wilkie et al., 1997; Dempson et al., 2002). Another option is a hybrid approach in which, for a given range of temperatures (or seasonal periods), there is a specific regulation with respect to net tending frequency where durations vary relative to temperature. Such an approach adds regulatory complexity however; particularly with respect to enforcement. Our work revealed that even at cool temperatures, fish held in nets for extended periods of time (i.e., 6 d) exhibited significant levels of injury and stress at both temperatures even though little mortality was observed at the cooler temperature. Both injury and stress have the potential to influence immune function via injury creating pathways for opportunistic pathogens and through immunosuppression caused by the stress response. As such, from both a management and fish welfare perspective (Arlinghaus et al., 2007b; Diggles et al., 2011), the most risk-averse strategy would likely be to focus efforts on requiring frequent net tending irrespective of water temperature. Given that at warmer temperatures we did see mortality within 48 h, shorter periods might be considered as is common in fisheries research scenarios (e.g., 24 h net sets).

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