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Temporal Variation in the Physiological Responses in Largemouth Bass following Small Club Angling Tournaments

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Abstract

Sublethal physiological disturbances and mortality were quantified in Largemouth Bass *Micropterus salmoides* subjected to small, club-style angling tournaments (<30 teams) held at two central Illinois lakes. Between April and October, physiological disturbances were assessed in four tournaments at Lake Bloomington, and mortality was assessed in four tournaments at Evergreen Lake. Indicators of physiological disturbances were evident in Largemouth Bass following club angling tournaments, with some temporal variation in responses. Plasma glucose concentrations increased in tournament-caught fish relative to reference fish in all months, except during October, when glucose concentrations did not change; plasma cortisol values among tournament fish also were lowest during October. Plasma potassium levels decreased only in April, whereas chloride levels were unaffected by tournaments. Sodium concentrations varied across months, but the magnitude of tournament-induced decreases were similar across all months. Whole-blood hemoglobin was lowest in May, and although hematocrit significantly decreased in tournament-caught fish in May, it remained unchanged in other months. Lactate increases occurred during all tournaments and were of similar magnitudes even though water temperatures ranged from 15.7°C to 27.6°C. Small, yet significant, temporal differences were observed in plasma sodium and in whole-blood hemoglobin concentrations from reference fish collected in each month via electrofishing, which indicated temporal changes in baseline values for some parameters. Mortality at tournaments was low (<5%) and did not appear to vary across months. Our results suggest that physiological responses of Largemouth Bass to small, club-style tournaments can vary temporally and are similar to those sustained during professional tournaments, even if mortality rates are generally low.

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The frequency of competitive angling tournaments has risen dramatically during the last 30 years; a recent survey estimated that over 32,000 events were held in the United States in 2005 (Schramm and Hunt 2007). Black bass *Micropterus* spp. are the most common species targeted by tournament anglers, although some tournaments focus on other species (Schramm and Hunt 2007). Concerns regarding the impacts of tournaments on populations of Largemouth Bass *M. salmoides* have prompted several studies to examine issues related to the mortality of tournament-caught fish (reviewed in Siepker et al. 2007). Several factors, such as high water temperatures, the presence of Largemouth Bass virus, and poor organizational procedures, have all been shown to influence tournament mortality (Schramm et al. 2006; Siepker et al. 2007).

Impacts during tournaments, however, are not limited to mortality; sublethal physiological disturbances of the fish (Suski et al. 2003; Killen et al. 2006) may reduce growth rates (Wendelaar Bonga 1997), affect fitness (Schreck et al. 2001), and compromise the effectiveness of immune systems (Pickering and Pottinger 1989). Moreover, the return to physiological homeostasis is fueled by the utilization of stored energy resources, which can be depleted by stress (Richards et al. 2002). Changes in physiological parameters are also more sensitive than direct estimates of mortality and can provide insight into the nature and severity of stressors associated with angling tournaments. More specifically, changes in hematocrit and hemoglobin levels can indicate a reduced capacity to deliver oxygen to tissues (Houston 1997), and elevated lactate levels are indicative of increased anaerobic respiration (Furimsky et al. 2003). Further, the concentration of cortisol increases during exposure to a stressor and prompts several physiological changes designed to withstand and overcome the stressor (Mommsen et al. 1999). Cortisol promotes the release of glucose to provide fuel for aerobic tissues, such as the heart and gills, and increases perfusion of the gill lamellae to increase the surface area for oxygen uptake (Wendelaar Bonga 1997). Perfusion of the gill lamellae can also result in loss of ions to the environment (Gonzalez and McDonald 1992), which can be problematic for fish because ionic balance is costly to maintain (Febry and Lutz 1987). Clearly, identifying the factors that influence lethal and sublethal responses of Largemouth Bass in angling tournaments can provide information useful to anglers, tournament organizers, and fishery managers to minimize the negative impacts on individual fish and fish populations.

Angling tournaments for black bass can vary widely with respect to their timing, magnitude, and organizational procedures. Currently, bass angling tournaments can be scheduled throughout the year; fish have been shown to exhibit temporal changes in behavior (e.g., spawning), swimming ability (Kolok 1992), metabolic rate (Evans 1984), and partitioning of energy stores (Adams et al. 1982; Brown and Murphy 2004). Variations in these behavioral and physiological characteristics can combine to significantly affect physiological disturbances in Largemouth Bass (Gingerich et al. 2010) and may

produce temporal variation in the lethal and sublethal responses of fish in angling tournaments. For example, higher mortality of Largemouth Bass has been noted during spring tournaments, presumably due to the synergistic interactions of tournament-induced stressors and spawning behavior (Kwak and Henry 1995). Tournament-induced mortality has also been shown to correlate positively with water temperature (e.g., Wilde 1998), suggesting a temperature-mediated impairment of physiological systems. Temporal changes in physiological parameters and environmental conditions may therefore strongly influence the sublethal effects of competitive angling tournaments on black bass.

In addition to variation in the timing of angling tournaments, the great deal of variation in the size of tournaments. For example, local club tournaments with a small number of competitors (typically <30; Edwards et al. 2004a) are held more frequently than are larger, high-profile tournaments that have many competitors (50–500 participants). The greater negative effects of club-style tournaments on fisheries than those of larger tournaments have been ascribed to lower levels of organization, concentration of fishing effort on a limited number of water bodies, and the higher frequency of club-style tournaments (Wilde 1998). To date, research has focused only on quantifying the physiological (sublethal) effects of large, high-profile angling events (Suski et al. 2003), and the conclusions from those studies were based only on single visits to several tournaments at the same time of the year (summer). Currently, little is known regarding the sublethal effects of small, club-style tournaments despite the fact that they are more numerous and organizationally distinct from large, professional events.

The primary objective of this study was to quantify physiological disturbances of Largemouth Bass caught during small, club-style tournaments throughout the year. We tested the hypothesis that physiological disturbances incurred by Largemouth Bass during these events vary temporally due to changes in water temperature or inherent physiological properties of the fish. In an effort to minimize intertournament variability, our approach was to examine a single tournament series in a single lake several times in 1 year. A secondary goal was to quantify initial and delayed mortality of tournaments over a similar time frame to provide a basis for comparison with previous studies.

METHODS

Tournament locations and procedures.—Eight live-release bass tournaments were visited in 2008, four at each of two central Illinois lakes: Lake Bloomington (257 hectares; 40.6617°N, 89.6501°W) and Evergreen Lake (364 hectares; 40.6503°N, 89.0542°W). Nonlethal blood sampling was performed only at the tournaments at Lake Bloomington, and delayed mortality was estimated only at the tournaments at Evergreen Lake. In effect, we performed two studies to prevent potential confounding effects of one sampling procedure on the other. At

each tournament, surface water temperatures were measured immediately before the weigh-in. All events lasted for 8 h, beginning at 0600 hours and ending at 1400 hours, except for tournaments in October, which began at 0700 hours and ended at 1500 hours. Surface water temperatures and dates of tournaments at Lake Bloomington were: 15.7°C on 26 April, 17.0°C on 24 May, 27.6°C on 26 July, and 18.7°C on 5 October. Surface water temperatures and dates of tournaments at Evergreen Lake were: 14.0°C on 10 May, 25.2°C on 28 June, 27.6°C on 16 August, and 19.6°C on 4 October. All tournaments were part of the Ever-Bloom Tournament Trail, in which a common group of anglers participated and weigh-ins were officiated by the same individual at each event. Each event followed procedures similar to those in larger tournaments (e.g., Suski et al. 2003), except for the number of anglers. About 20 boats were used, each boat containing a team of two anglers. Each team was allowed to weigh in a maximum of five largemouth bass of at least 38.1 cm TL each (equal to the creel and size limits for a single angler on tournament lakes). Anglers were allowed to use artificial baits only, captured fish were held in aerated live wells aboard boats, anglers convened at a designated weigh-in location at the end of the tournament, and anglers incurred penalties for deceased fish brought to the scales (e.g., Schramm et al. 1987; Kwak and Henry 1995; Neal and Lopez-Clayton 2001; Suski et al. 2003; Edwards et al. 2004a). The dead-fish penalty was 453 g (1 lb.), which is between two- and ninefold greater than penalties previously reported (53 g, Schramm et al. 1987; 227 g, Weathers and Newman 1997). Culling, or the replacement of a smaller fish with a larger fish once the bag-limit has been reached, was permitted except that culling dead fish was prohibited. The weigh-in procedure was similar to that in previous studies where, at the conclusion of the tournament, anglers used plastic bags containing about 20 L of lake water to transport fish from their boat to a scale, where the fish were weighed in air in aggregate. After a total weight was determined, fish were placed back into water-filled bags and carried by the anglers to the lake for release. At Lake Bloomington, prior to final release of the fish into the lake, a subset of Largemouth Bass were intercepted directly from anglers for nonlethal blood sampling; all fish caught at Evergreen Lake were used for the estimation of delayed mortality (both described below).

Physiological disturbances.—To study physiological disturbances, we collected blood samples nonlethally by randomly taking a fish from an angler (just before the fish was to be released into the lake) and placing it into a 378-L aerated holding tub half-filled with lake water. Up to two fish at a time were held in the holding tub for up to 3 min before being placed into an aerated 45-L cooler half-filled with lake water. Fish that had been held longer than 3 min were released back into the lake without being sampled for blood, to ensure that sampling-related disturbances remained at a minimum; indicators of physiological disturbances can begin to be displayed after several minutes of confinement (Wedemeyer et al. 1990). Once placed into the

cooler, Largemouth Bass were held in a supine position on a foam-lined tray, ensuring that gills were submerged in water while the posterior portion of the body was exposed. Approximately 1 mL of blood was then drawn by caudal puncture (generally taking <1 min) using an 18-gauge hypodermic needle and a 1-mL syringe rinsed with lithium heparin (Sigma-Aldrich, St. Louis, Missouri; Houston 1990). Immediately following blood withdrawal, we placed a small portion (about 100 μL) of whole blood into a 1.5-mL centrifuge tube, and flash-froze it in liquid nitrogen for later determination of hemoglobin. Hematocrit values for whole blood (% packed cell volume, PV) were determined on-site by inducing a small amount of whole blood into two heparinized microcapillary tubes (about 20 μL each), which were then sealed and centrifuged at 15,800 rpm (13,700 × g) for 3 min in a hematocrit centrifuge, after which the hematocrit was read using a digital reader (CritSpin Models CS22 and CSD2; Iris International, Inc., Chatsworth, California). The remaining whole blood (about 800 μL) was placed into a 1.5-mL microcentrifuge tube and centrifuged for 2 min at 2,000 × g for 2 min. Plasma was immediately separated from erythrocytes using a transfer pipette, divided into three aliquots of approximately 100 μL each, placed into 1.5-mL microcentrifuge tubes, and flash-frozen in liquid nitrogen before being transferred to an ultracold freezer (<−75°C) until processing (Suski et al. 2003). Following blood collection, the TL to the nearest millimeter and weight (to the nearest gram) of the fish were measured, after which they were released into the lake. Blood was collected at random from at least six fish per tournament (April: $n = 8$, May: $n = 6$, July: $n = 8$, October: $n = 7$), approximately one-quarter to one-third of the total number of fish weighed in during each tournament, and similar to the number of fish previously examined for physiological parameters during professional tournaments (e.g., Suski et al. 2003).

To distinguish tournament-induced physiological disturbances from natural temporal changes in physiological parameters, we collected a group of Largemouth Bass from Lake Bloomington immediately after the completion of each tournament (subsequently termed the reference group) to determine resting values for blood parameters. For this, six Largemouth Bass were collected using DC electrofishing gear but no samples were taken until later, to avoid any physiological changes due to electrofishing. After collection, fish were transported in aerated hauling tanks to the Kaskaskia Biological Station, Illinois Natural History Survey, Sullivan, Illinois. Once they arrived at the laboratory, the reference fish were placed into individual opaque, aerated containers continuously supplied with water from a central basin and held for 60 h to allow them to recover from capture and transport-induced disturbances. Water in the central basin was preheated or prechilled to maintain temperatures in the individual containers ± 1°C of the surface temperature of the lake at the time the fish were collected.

At the end of 60 h of acclimation, resting Largemouth Bass were overdosed with anesthetic [3-aminobenzoic acid ethyl ester methanesulfonate (MS-222), 250 mg/L, buffered with 500 mg/L

sodium bicarbonate] added directly to each aerated chamber. After the fish ceased to ventilate, approximately 1 mL of blood was collected and processed identically to the tournament sampling procedures described above. Total length and weight of reference fish were recorded and found to be not significantly different across months or significantly different from tournament fish during any month, as determined by two-way analysis of variance (ANOVA); main effects: month (length: $P = 0.73$, weight: $P = 0.24$); tournament/reference (length; $P = 0.18$, weight: $P = 0.11$); month × tournament/reference (length: $P = 0.22$, weight: $P = 0.39$).

Laboratory analyses.—A detailed description of the analyses of plasma parameters are described in Suski et al. (2003). Briefly, plasma cortisol, whole-blood hemoglobin, and plasma hemoglobin concentrations were determined with commercially available kits (cortisol: kit 900-071; Assay Designs, Ann Arbor, Michigan, and hemoglobin: QuantiChrom Hemoglobin Assay Kit, DIHB-250; BioAssay Systems, Hayward, California). Plasma sodium and potassium concentrations were determined using a flame photometer (Model 2655-00; Cole-Parmer Instrument Co., Chicago, Illinois), and plasma chloride concentrations were determined using a digital chloride titrator (Model 4435000; Labconco Corp., Kansas City, Missouri). Plasma glucose and lactate concentrations were determined enzymatically following the methods of Lowry and Passonneau (1972), using a 96-well microplate with a commercially available spectrophotometer (Spectra Max Plus 384, Model 05362; Molecular Devices, Union City, California).

Mortality estimates.—Delayed mortality of Largemouth Bass caught during club angling tournaments was estimated using fish in three submerged holding pens as well as a fourth holding pen of reference Largemouth Bass collected using electrofishing gear. At the conclusion of the weigh-in at Evergreen Lake, all Largemouth Bass weighed during the tournament were placed into holding pens consisting of a 2.7-m-deep soft, nylon-mesh box secured to a floating frame made of schedule-40 polyvinyl chloride pipe (3 cm inside diameter) 1.2 m wide × 1.8 m long (5.8 m^3 total volume). Fish were randomly placed into one of three holding pens and no more than 10 fish were placed into an individual holding pen. The maximum density of fish in holding pens was 10 fish/5.8m^3, which is less than densities in some previous studies using holding pens (e.g., Hartley and Moring 1995; Kwak and Henry 1995; Edwards et al. 2004a). Holding pens were placed into water more than 3 m deep and secured to a dock structure. Weights were attached to the bottom of the netting to sink the bottom of the cage while the frame remained on the surface, providing the fish inside with a range of available depths. Temperature and oxygen profiles were taken both inside and outside of the cages (YSI 55; Yellow Springs Instruments, Yellow Springs, Ohio) and did not differ by more than 0.1°C or 0.2 mg/L. Holding-pen—induced mortality and angling–related mortality are commonly addressed by

two methods, both of which utilize control fish captured outside of the tournament setting (e.g., electrofishing). The first method places angled fish and control fish into separate pens, while the second method places angled and control fish into the same pen, after individuals from each group have received an identifying mark (e.g., fin clip, tag; Pollock and Pine 2007). We chose to utilize the first method by collecting Largemouth Bass using DC electrofishing gear immediately after the conclusion of the tournament and placing 10 of these fish into a separate holding pen, identical to the ones described above. Although this method cannot account for events affecting the fish in a single pen (e.g., tampering), it eliminates the additional handling needed to mark individuals. All fish were retained for a period of 72 h and all pens were checked for mortality 24 and 72 h after the conclusion of the tournament. These lengths of time have been previously used to assess posttournament mortality of small tournaments (Edwards et al. 2004a). In addition, the probability of additional tournament-induced mortality after 4 d has been reported to be negligible (Weathers and Newman 1997; Neal and Lopez-Clayton 2001) and, as Schramm et al. (1987) showed, the greatest portion of delayed mortality occurred within 24 h of the conclusion of an angling tournament. At the end of 72 h, we counted all fish in each pen and then released all living Largemouth Bass. Initial mortality (dead Largemouth Bass brought by anglers to the weigh-in) was also recorded. Fish were judged as dead if they displayed no opercular movement (e.g., Schramm et al. 1987; Meals and Miranda 1994; Kwak and Henry 1995).

Statistical analyses.—For physiological parameters, a two-way ANOVA was used to test for an interaction of month and treatment (tournament-caught or reference). Main effects of ANOVAs were not interpreted when the interaction term in the two-way ANOVA was significant; they were interpreted, however, when the interaction was not significant. A Tukey–Kramer honestly significant difference test was used for all post hoc comparisons when appropriate. Statistical analyses were performed using SAS Version 8.2 (SAS Institute, Cary, North Carolina) and the level of significance (α) for all tests was 0.05. For mortality determination, each pen served as a replicate estimate of mortality, which was calculated by dividing the number of deceased fish at the end of 72 h by the number of fish originally put into the pen (Pollock and Pine 2007). The overall percent of delayed mortality for the entire tournament and the variance of these estimates were calculated following the methods of Pollock and Pine (2007). Initial mortality was calculated as the number of deceased fish the anglers brought to the weigh-in divided by the total number of fish weighed.

RESULTS

Physiological Disturbances

Significant changes in physiological parameters of Largemouth Bass were observed during the four small, club-style angling tournaments examined. In April, plasma cortisol values

of tournament-angled fish increased by sevenfold over those of the April reference fish (Figure 1A; Table 1). Although the concentration of cortisol in tournament-angled Largemouth Bass was greater relative to reference fish in May, July, and October, the changes in these months were not statistically significant (Figure 1A). Additionally, although concentrations of cortisol in the plasma of tournament-caught Largemouth Bass did not vary across months, the concentrations were significantly higher in reference fish in July than in April (Figure 1A). Plasma glucose concentrations of tournament-angled Largemouth Bass were 60% greater than for reference fish in April, and double that for reference fish in May (Figure 1B; Table 1). Plasma glucose concentrations of tournament-caught fish did not differ significantly from values obtained from reference fish during the July or October sampling periods (Figure 1B). Concentrations of plasma glucose were similar for April, May, and July tournaments; the concentrations observed during October tournaments were approximately 25% lower than April and May values (Figure 1B). Plasma lactate values were significantly higher for tournament-caught Largemouth Bass than for reference fish in every month (Figure 1C; Table 1). Plasma lactate concentrations were similar for April, May, and July tournaments but were about 50% lower for the October tournament than for the earlier tournaments (Figure 1C).

Compared with the changes in plasma cortisol, glucose, and lactate, the changes observed in plasma ions of tournament-caught Largemouth Bass were relatively minor. Plasma sodium concentrations changed significantly across months; the April and October tournaments had the highest sodium concentrations, July the lowest, and May values were intermediate (Figure 2A; Table 1). Plasma sodium values were significantly greater in tournament-caught fish than in reference fish (indicated by the significant tournament/reference ANOVA; Table 1); however, this may be driven by a large difference between groups in October (Figure 2A). No significant changes in plasma chloride concentrations were observed within or across months for either tournament-caught or reference Largemouth Bass (Figure 2B; Table 1). During April, plasma potassium concentrations were significantly lower by about 30% in tournament-caught Largemouth Bass than in reference fish (Figure 2C; Table 1). No significant differences were observed between reference fish and tournament-caught fish for the other months (Figure 2C).

Both month and the stressors associated with tournaments had a significant effect on the indicators of oxygen transport ability for Largemouth Bass. The concentration of hemoglobin in whole blood of Largemouth Bass caught during angling tournaments was lower than for reference fish in all months (Figure 3A; Table 1). Whole-blood hemoglobin concentrations differed significantly across months, the highest concentrations being observed in October, followed by those in April and July (which were not significantly different from each other), and the lowest in May (Figure 3A). Whole-blood hematocrit values
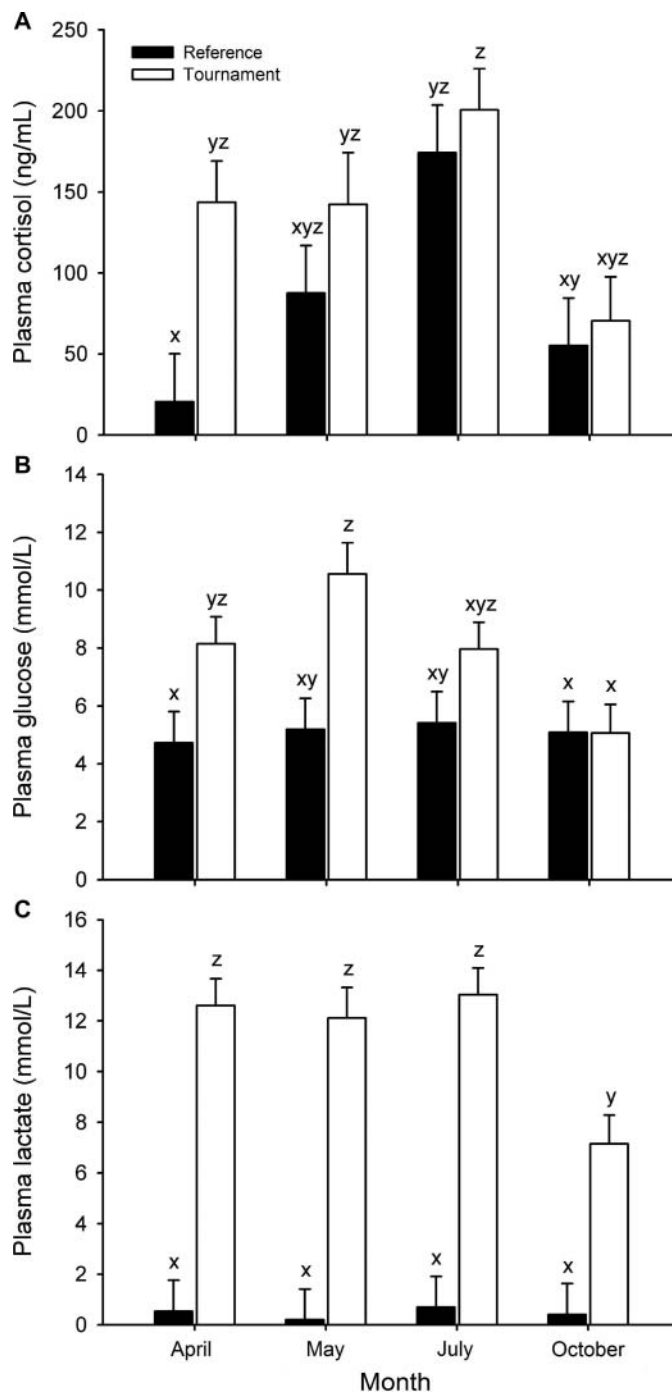

FIGURE 1. Concentrations of cortisol (**A**), glucose (**B**), and lactate (**C**) in plasma of Largemouth Bass from Lake Bloomington, Illinois, in April, May, July, and October. Largemouth Bass either were sampled immediately following a club-style angling tournament (Tournament, open bars) or were collected using DC electrofishing gear and sampled after a 60-h recovery period in the laboratory (Reference, filled bars). Mean separation is indicated by lowercase letters; statistically different means do not share letters (reference groups: $n = 6$ for all months; tournament-caught groups: April, $n = 8$; May, $n = 6$; July, $n = 8$; October, $n = 7$).

TABLE 1. Two-way ANOVA for plasma parameters measured in Largemouth Bass in April, May, July, and October at Lake Bloomington, Illinois. Largemouth Bass either were sampled immediately following a club-style angling tournament or were collected using DC electrofishing gear and sampled after a 60-h recovery period in the laboratory (reference groups: $n = 6$ for all months; tournament-caught groups: April, $n = 8$; May, $n = 6$; July, $n = 8$; October, $n = 7$).

Response Variable	Source	SS	df	F	P
Plasma cortisol (ng/mL)	Tournament/reference	15.9	1	14.6	0.0004
	Month	16.6	3	5.1	0.0042
	Tournament/reference × month	12.5	3	3.8	0.016
	Error	48.5	44		
Plasma glucose (mmol/L)	Tournament/reference	3.3	1	19.4	<0.0001
	Month	1.4	3	2.6	0.068
	Tournament/reference × month	1.4	3	2.8	0.049
	Error	7.8	45		
Plasma lactate (mmol/L)	Tournament/reference	124.7	1	388.0	<0.0001
	Month	3.8	3	3.9	0.015
	Tournament/reference × month	2.8	3	2.9	0.048
	Error	14.7	45		
Plasma sodium (mEq/L)	Tournament/reference	0.03	1	4.5	0.040
	Month	0.14	3	7.2	0.0005
	Tournament/reference × month	0.03	3	1.5	0.23
	Error	0.29	45		
Plasma chloride (mEq/L)	Tournament/reference	155.4	1	0.5	0.50
	Month	2,280.0	3	2.3	0.092
	Tournament/reference × month	1,057.8	3	1.1	0.38
	Error	15,037.0	45		
Plasma potassium (mEq/L)	Tournament/reference	0.3	1	0.8	0.36
	Month	0.1	3	0.1	0.98
	Tournament/reference × month	2.7	3	2.9	0.047
	Error	13.8	43		
Whole blood hemoglobin (g/dL)	Tournament/reference	1.4	1	14.6	0.0004
	Month	4.1	3	14.1	<0.0001
	Tournament/reference × month	0.3	3	1.2	0.33
	Error	4.3	44		
Whole blood hematocrit (% PV)	Tournament/reference	0.03	1	0.0	0.84
	Month	787.46	3	7.8	0.0004
	Tournament/reference × month	220.76	3	2.9	0.049
	Error	1,294.36	38		
Plasma hemoglobin (mg/dL)	Tournament/reference	3.1	1	3.3	0.078
	Month	9.2	3	3.2	0.033
	Tournament/reference × month	2.7	3	0.9	0.43
	Error	43.6	45		

for tournament-caught fish significantly differed from reference fish only during May, when hematocrit values for tournament-caught fish were approximately 20% lower than values from reference fish (Figure 3B; Table 1). Hematocrit was significantly greater in April reference fish than in October reference fish, but no other significant differences were observed among reference fish in the other months (Figure 3B). Plasma hemoglobin concentrations varied across months, being the highest during July, lowest during October, and intermediate during April and May (Figure 3C; Table 1).

Mortality Estimates

No delayed mortality of reference Largemouth Bass collected by electrofishing was observed at any tournament during the observation period. A total of 96 Largemouth Bass were weighed in during the four tournaments surveyed at Evergreen Lake (Table 2). Initial mortality was 4.0% for the June tournament, 3.8% for the August tournament, and 0% in October (Table 2). Some fish escaped from the holding pens during the first 24 h after the May tournament; their delayed mortality was estimated from the remaining fish (10 of the original 25).
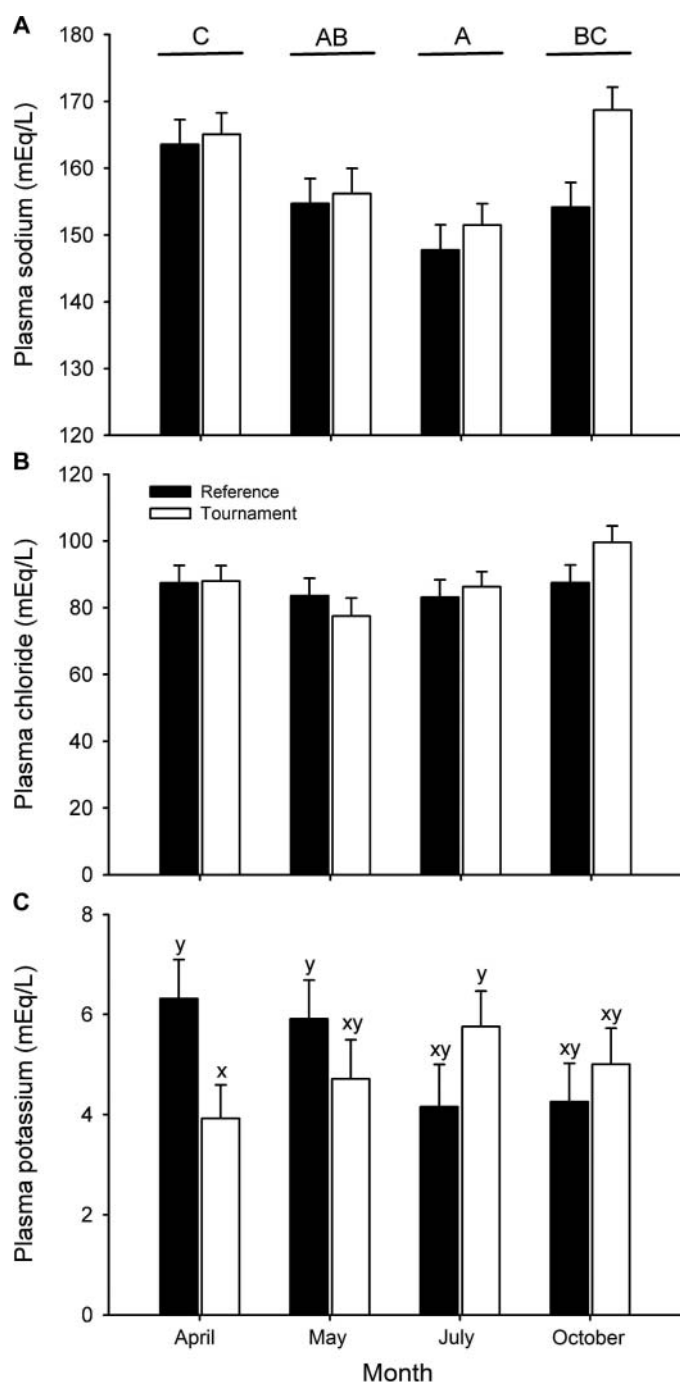
FIGURE 2. Concentrations of sodium **(A)**, chloride **(B)**, and potassium **(C)** in plasma of Largemouth Bass from Lake Bloomington, Illinois, in April, May, July, and October. Largemouth Bass either were sampled immediately following a club-style angling tournament (Tournament, open bars) or were collected using DC electrofishing gear and sampled after a 60-h recovery period in the laboratory (Reference, filled bars). Mean separation is indicated by lowercase letters; statistically different means do not share letters. Uppercase letters are used to separate monthly means when the main effect of month was significant but the main effect of tournament/reference and the interaction of the main effects (tournament/reference × month) were not significant (reference groups: $n = 6$ for all months; tournament-caught groups: April, $n = 8$; May, $n = 6$; July, $n = 8$; October, $n = 7$).
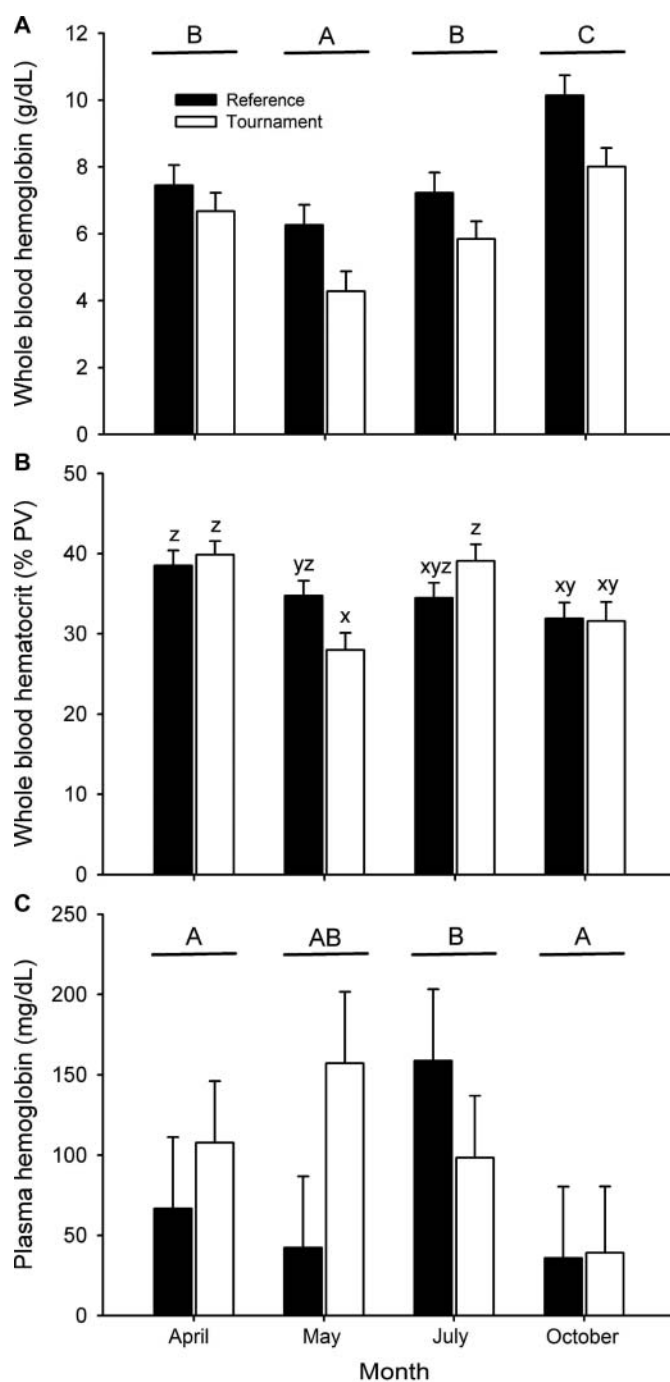
FIGURE 3. Concentrations of whole blood hemoglobin **(A)**, whole blood hematocrit (% PV is percent packed volume of red blood cells) **(B)** , and plasma hemoglobin **(C)** of Largemouth Bass from Lake Bloomington, Illinois, in April, May, July, and October. Largemouth Bass either were sampled immediately after a club-style angling tournament (Tournament, open bars) or were collected using DC electrofishing gear and sampled after a 60-h recovery period in the laboratory (Reference, filled bars). Mean separation is indicated by lowercase letters; statistically different means do not share letters. Uppercase letters are used to separate months when the main effect of month is significant but the main effect of tournament/reference and the interaction of the main effects (tournament/reference × month) are not significant (reference groups: $n = 6$ for all months; tournament caught groups: April, $n = 8$; May, $n = 6$; July, $n = 8$, October, $n = 7$).

TABLE 2. Summary of the total number of fish weighed-in, and the initial, delayed, and total mortality of largemouth bass caught during small, club-style tournaments at Evergreen Lake, Illinois, across four months. Mortality is expressed as a percentage of the total number of fish weighed-in, with the actual number of fish in parentheses. Initial mortality refers to the number of fish that were deceased before being weighed-in at the tournament. Delayed mortality refers to the number of fish that died within 72 h of the conclusion of the tournament in a submerged holding pen, but had been alive immediately after the weigh-in. Total mortality is the sum of initial and delayed mortality. No delayed mortality was observed during any month for reference Largemouth Bass collected using electrofishing gear and held in holding pens for 72 h.

	Month			
	May	June	August	October
Water temperature (°C)	14.0	25.2	27.6	19.6
Number of teams	21	22	21	18
Total number fish weighed-in	25	25	26	20
Average length of fish (mm)	418.3	422.5	419.1	422.3
Initial mortality	0%	4.0% (1)	3.8% (1)	0%
72 h delayed mortality	0–60%[a]	0%	0%	5% (1)
Standard error of delayed mortality	[a]	0%	0%	5%
Total number reference Largemouth Bass	15	14	15	10
Total mortality	0–60%[a]	4.0% (1)	3.8% (1)	5% (1)

[a]Estimates of mortality for the May tournament account for largemouth bass that escaped from holding pens during the first 24 h following the tournament. Mortality is estimated to be 0% if it is assumed all escaped fish survived; whereas mortality is estimated to be 60% if it is assumed all escaped fish died.

No delayed mortality of tournament-caught Largemouth Bass was observed after 72 h following the May, June, and August tournaments. Because of the escape of Largemouth Bass after the May tournament, the estimate of delayed mortality ranged from 0%, if all escaped fish were presumed to have survived, to 60%, if all escaped fish were presumed to have died. Due to the low number of fish caught during the October tournament, we used only two holding pens to assess delayed mortality. One tournament-caught Largemouth Bass died within 72 h after the tournament, resulting in 5% delayed mortality for the October tournament (Table 2).

DISCUSSION

To determine temporal variability in the effects of tournaments on Largemouth Bass physiology, we examined a single angling organization to control for variation in fish-handling practices among tournament groups and interlake variability. The physiological responses of Largemouth Bass to tournaments have been shown to exhibit little variation among lakes when conducted in the same season (summer: June–August; Suski et al. 2003). Nevertheless, a key next step would be to evaluate many club angling tournaments in which fish would be subjected to a wide range of conditions, including organizations that adhere to fish care guidelines of larger tournaments and those that disregard the guidelines. The inference space of our results is somewhat limited as we examined only one lake for physiological parameters and only one lake for mortality, but it does establish the temporal variation in physiological parameters that could be expected in future studies with more lakes. Furthermore, our study serves as a starting point for future studies aimed at examining club-style events in more detail. Specifically, quantification of exposure to air and the total handling times would help interpretation of physiological variation.

Largemouth Bass displayed a suite of physiological disturbances in plasma and whole-blood parameters after participation in small, club-style angling tournaments. As part of the stress response in fishes, stress hormones such as corticosteroids (e.g., cortisol) and catecholamines are released into the blood to trigger physiological adjustments that allow fish to withstand and overcome stressors (Mommsen et al. 1999). Cortisol is commonly used in studies to examine stressors in teleosts; it is readily measured and its concentrations in plasma increase during exposure to stressors (Mommsen et al. 1999). Plasma cortisol concentrations observed following weigh-ins throughout the current study ranged from 70 to 200 ng/mL, slightly greater than those previously reported (80–120 ng/mL) during large tour-style tournaments held across a number of lakes in Ontario, Canada (Suski et al. 2003). The peak in cortisol concentrations in club-style tournaments (July) was also greater than the cortisol levels observed in Largemouth Bass after exhaustive exercise (60–110 ng/mL; Suski et al. 2006, 2007), thus indicating cortisol levels in Largemouth Bass may take longer to recover from tournament stressors than from exercise alone. Plasma glucose, a common stress indicator released during the secondary stress response (Wendelaar Bonga 1997), increased more in tournament-caught fish than in reference fish in all tournaments except the October tournament; however, the difference was significant only with regard to the values in laboratory reference fish following the April and May tournaments. Plasma glucose concentrations observed in Largemouth Bass after club-style tournaments were similar in magnitude to those observed in Largemouth Bass after large, tour-style events (Suski et al. 2003).

Metabolic disturbances resulting from anaerobic respiration were seen in Largemouth Bass caught during all tournaments, as indicated by greater plasma lactate values in tournament-caught fish than in reference fish. The magnitude of these disturbances was about equal to that of the disturbances during angling and simulated weigh-in (Suski et al. 2004), exhaustive exercise (Suski et al. 2006), and professional tournaments with many (>75) anglers (Suski et al. 2003). This disturbance most likely resulted from some combination of exposure to air during the weigh-in (Suski et al. 2004) or culling (Cooke et al. 2002), hypoxia in live wells or in weigh-in bags, or angling stress (Suski et al. 2004, 2006).

Changes in ionic concentrations, whether increasing or decreasing, are indicative of disruptions to the ionic balance of a fish and can provide insight into the nature of the disturbance.

More specifically, potassium increases can be indicative of a hematological disturbance, given that potassium is released by the rupturing of red blood cells, whereas potassium decreases may be the result of gill lamellae perfusion, which allows greater oxygen uptake but also increases the surface area over which ions can be lost to the environment (Gonzalez and McDonald 1992; Nielsen and Lykkeboe 1992). A decrease in plasma potassium similar to the one observed following the April club-style tournament did not occur during professional tournaments (Suski et al. 2003) or in any of the other tournaments in the current study. Stressors causing disruptions to the ionic balance of fish can result in reduced growth rates (Bury et al. 1995), thus highlighting the importance of this disturbance even if it does not result in mortality.

Hematological parameters, such as hemoglobin and hematocrit, are used to assess the oxygen-carrying capacity of the blood (Houston 1997). Decreases in either hematocrit or hemoglobin indicate a reduced ability for the blood to transport oxygen, whether because of reduced binding capacity (hemoglobin) or fewer red blood cells (hematocrit; Houston 1997). Hemoglobin concentrations can begin to change within 5 min following exposure to a stressor (Gustaveson et al. 1991), and sharp decreases in concentrations to below 4 g/dL have often been thought to indicate anemia (Barton et al. 2002). Blood hemoglobin concentrations of reference and tournament-caught Largemouth Bass in May were only slightly above 4 g/dL, indicating Largemouth Bass may have taken longer to recover from exposure to air following this tournament due to impaired ability to deliver oxygen to tissues and thus to fuel recovery (Richards et al. 2002). In the current study, variation in hemoglobin concentrations was unlikely to be related to changes in the quantity of red blood cells. Typically, hemoglobin concentrations will correlate positively with the quantity of red blood cells (Houston 1997). In the current study, however, high hemoglobin and low hematocrit in October indicated that other factors such as release and absorption by the spleen are likely involved (Pearson and Stevens 1991).

Overall, small, club-style tournaments induced significant changes in several physiological parameters, the responses varying somewhat over the tournaments examined. This variation could be due to a number of biotic and abiotic factors. Temporal changes in Largemouth Bass physiology are one possible explanation for these differences. For example, Largemouth Bass exhibit seasonal changes in several indices of physiological condition (e.g., hepatosomatic index), which can be influenced by several factors, including spawning behavior during spring, high water temperatures during summer, and accumulation and use of energy stores prior to winter (Adams et al. 1982, Brown and Murphy 2004). Illustrating temporal changes in Largemouth Bass physiology throughout the current study, we noted that cortisol levels of both tournament-caught and laboratory-reference Largemouth Bass reached peak levels during July but, on average (but not significantly), were lower during October, suggesting a positive relationship between cortisol and temperature.

The July peak in cortisol levels may have also influenced concentrations of sodium ions, which were lowest after the July tournament, presumably a result of increased gill permeability, concomitant with high cortisol levels (Gonzalez and McDonald 1992).

Factors other than temporal changes in Largemouth Bass physiology and water temperature could also be responsible for the variation in the observed physiological responses. Exposure to air during tournament weigh-ins has been shown to induce metabolic disturbances in Largemouth Bass (Suski et al. 2004); therefore, differences in duration of air exposure could explain variation in plasma lactate levels across tournaments. Although plasma lactate concentrations observed during the October tournament were about half that in the other three tournaments, we do not suspect shorter air exposure in October than in the other months, as the procedural guidelines and the official performing the weigh-in were the same for all tournaments; these effects cannot, however, be ruled out. Physiological differences among tournaments could also be due to variations in fish care practices. For example, the recommendation to add ice to live wells might be followed only during summer tournaments (Gilliland et al. 2002). For Largemouth Bass, this practice has been shown to reduce mortality of tournament-caught fish (Gilliland 2002); however, large reductions in water temperature can also induce physiological disturbances (VanLandeghem et al. 2010) and increase physiological recovery times following angling (Suski et al. 2006).

Electrofished Largemouth Bass likely provide a reasonable source for baseline physiological values. Although electrofishing can have an immediate effect on some physiological parameters, recovery to precapture physiological levels occurs within 48 h for wild Striped Bass *Morone saxatilis* (Harrell and Moline 1992) and within 24 h for European Chub *Leuciscus cephalus* (Bracewell et al. 2004). Furthermore, Suski et al. (2006) demonstrated that several physiological parameters of Largemouth Bass, including many examined in this study, return to predisturbance levels after 4 h of recovery following exhaustive exercise. Thus, 60 h probably provided ample time for electrofished Largemouth Bass to recover from capture- and transport-induced physiological disturbances and therefore measurement of resting values was most likely accurate.

Mortality of Largemouth Bass caught during club events was generally low and similar to mortality rates previously reported for small club tournaments. Specifically, Edwards et al. (2004b) reported mortality rates between 1.3% and 4.6% for small club tournaments in Connecticut; we too did not observe initial or delayed mortality rates above 5% for the June or August tournaments, and our estimate of delayed mortality during the October tournaments was between 0% and 10%. Several factors may have contributed to the overall low mortality in the current study despite temperatures as high as 27.6°C. Tournament mortality has been shown to correlate positively with the numbers of fish weighed per angler (Wilde et al. 2002); during tournaments at Evergreen Lake, however, most teams weighed

in fewer fish than the 5 fish limit. We would expect that fewer Largemouth Bass than the limit could result in lower mortality, as more oxygen is available to each individual fish in live wells and/or transport bags, reducing the metabolic demand for oxygen during confinement (Killen et al. 2006). Overall, mortality at the club tournaments monitored in the current study was low relative to that seen in previous studies and did not appear to follow any trends with water temperature.

In conclusion, physiological results exhibited temporal patterns, whereas mortality did not. Although tournaments at both lakes followed the same organizational procedures, involved a common group of anglers, and were conducted at relatively similar temperatures, the physiology and mortality results for Largemouth Bass caught during club tournaments at Lake Bloomington and Evergreen Lake do not appear to be related. These comparisons highlight the importance of minimizing the sublethal physiological disturbances incurred by Largemouth Bass during tournaments, even if such disturbances are not associated with mortality. Physiological disturbances and associated stress responses have previously been linked to negative outcomes, including disease susceptibility (Wendelaar Bonga 1997), an impaired ability to escape predators (Schreck et al. 1997), altered movement and activity patterns (Pankhurst and Dedualj 1994; Pankhurst and van der Kraak 1997; Gurshin and Szedlmayer 2004; Thorstad et al. 2004), and reduced fitness (Ostrand et al. 2004). Physiological parameters examined in this study suggest that small, club-style tournaments can have physiological effects on Largemouth Bass similar to those sustained during professional tournaments (Suski et al. 2003), despite earlier work suggesting that a reduced degree of organization might actually result in greater negative impacts relative to those of larger, tour-style events (Ostrand et al. 1999). Because not all tournament groups follow the same procedural guidelines as the one in this study, incorporating tournaments with different levels of organization should be a goal for future studies. The results of this study strongly suggest that small and large angling tournaments can have similar effects on Largemouth Bass, and tournament organizers and anglers should implement previously suggested procedures (e.g., proper aeration and minimized exposure to air), regardless of season and temperature.

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