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Blood lactate levels as a biomarker for angling-induced stress in tigerfish *Hydrocynus vittatus* from the Okavango Delta, Botswana

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Although critical in catch-and-release angling, no data are available on angling-induced stress in African gamefish. Blood lactate levels were used as a biomarker for angling-induced metabolic stress in tigerfish caught by angling in the Okavango Delta, Botswana. Blood was drawn and analysed for blood lactate from 66 anaesthetised fish. The landing time, handling time, body mass and total length were recorded prior to reviving and keeping the fish in aerated water for recovery before release. A strong positive relationship (r^2 = 0.607) was found between landing time and body mass, as well as significant elevations in blood lactate concentrations following rod-and-line angling, regardless of angling time. These data suggest that longer angling time significantly increases physiological stress, which may have an impact on breading success and mortality in tigerfish.

Keywords: catch-and-release angling, fish, physiology, recreational fisheries management, stress

Introduction

The tigerfish *Hydrocynus vittatus* is not only the most sought-after freshwater angling fish in Africa, but is one of the most important predatory fishes in African waters (Winemiller and Kelso-Winemiller 1994, Skelton 2001, Økland et al. 2005). Although abundant in certain areas throughout Africa, its numbers have declined in many rivers due to water extraction, pollution, obstructions such as dams and weirs, and fishing pressure (Steyn et al. 1996, Skelton 2001). This has been recognised specifically in South Africa by the Department of Environmental Affairs and Tourism who recently included tigerfish on the protected species list, together with such marine icons as the great white shark *Carcharodon carcharius* and the once thought to be extinct coelacanth *Latimeria chalumnae* (DEAT 2007). Despite the high profile of the tigerfish as a protected ecologically and economically important species, only limited published information on its biology in specific populations is available. These include data on growth, feeding and population dynamics in Lake Kariba (Matthes 1968, Kenmuir 1973); movement patterns and ecology in the upper Zambezi River and Zambezi floodplain (Winemiller and Kelso-Winemiller 1994, Økland et al. 2005); allozyme variation in and between upper Zambezi River (Namibia) and Olifants River (South Africa) populations (Kotzé et al. 1998); and accumulation of selected metals in the tissue and organs of the Olifants River tigerfish (du Preez and Steyn 1992) and of mercury in the Lake Kariba tigerfish (Mhlanga 2000). To date, the majority of research on tigerfish has been on populations from the Zambezi and Olifants River systems with no biological information available on the populations found in the Okavango Delta, Botswana.

The Okavango Delta, specifically the panhandle region with deeper, faster flowing water, is one of the most popular areas in Africa for anglers to catch tigerfish (for the specific region, see van der Bank and Smit 2007, Figure 1). Many tourist lodges and camps in this area specifically cater for recreational anglers and are the source for valuable income for this region. While some anglers keep a portion of the fish captured (up to five per day are permitted), many fish are captured and immediately released. The game fishing industry further encourages anglers voluntarily to release fish as a way to expand recreational fishing and therefore catch-and-release angling is growing, as a proportion of total fishing, in this region. The ultimate success of catch-and-release angling, however, depends on ensuring high release survival rates by minimising injury and mortality (Bartholomew and Bohnsack 2005). Despite the importance of this principle, research examining the physiological response to angling and possible sublethal impacts thereof on freshwater game fish is only available for a few popular North American and European species, i.e. Atlantic salmon, rainbow trout and largemouth bass (Brobbel et al. 1996, Cooke et al. 2002, DuBois and Dubielzig 2004), and is completely lacking for any of Africa's freshwater game

fishes. Meka and McCormick (2005) state that although the specific cause of mortality from exhaustive exercise is unknown, it might be due to intracellular acid-base disturbance produced, in part by the generation of muscle lactic acid (also see Wood et al. 1983). Although this is not true for all fish species, various studies have found elevated levels of blood lactate following capture, which may be associated with delayed mortality following high intensity anaerobic exercise (Ferguson and Tufts 1992, van Raaij et al. 1996). In view of the paucity of data concerning Africa's game fish, the aim of this study was to test the use of blood lactate as a biomarker for angling-induced metabolic stress in tigerfish and to examine the relationship between angling time and blood lactate levels.

Materials and methods

Sixty-six *Hydrocynus vittatus* were caught during the months of August and September 2007 and 2008 along 10 km of the main channel in the Okavango panhandle south of Shakawe (see van der Bank and Smit 2007, Figure 1). All fish were caught from a boat using standard recreational fishing gear, as recommended by local guides. During the capture of the fish, the time to land fish and remove hooks was recorded. Landing time was determined as the time from hooking a fish to landing it at the boat in a landing net. The handling time was determined as the time interval from when a fish was netted until the hook was successfully removed. Since plasma lactate levels in rainbow trout are significantly related to water temperature (Meka and McCormick 2005), the water temperature was measured daily throughout the collection area.

Anaesthesia and blood sampling

After unhooking, each fish was anaesthetised for two minutes in an insulated rectangle container (100 l) containing a solution of 32 mg l^{-1} clove oil and fresh river water. Once anaesthetised, the fish was placed on a wet surface and a 25 gauge needle and 1 ml syringe were used to aspirate approximately 500 µl of whole blood from the caudal vein. Blood was then immediately used to determine blood lactate with the aid of a portable blood lactate analyser (Lactate Pro Test Meter, Arkray Inc., Kyoto, Japan); the reliability of this instrument (coefficient of variation) was <3%. Anaesthetised fish were also measured (total length [TL] and fork length [FL]) and weighed before being revived in the river and subsequently released (following Meka and McCormick 2005).

Control group

Fifteen randomly chosen fish were used as controls. Following the initial blood collection, these fish were revived and kept in an aerated 100 l vessel containing fresh river water, transported to the field laboratory within 60 minutes and released into a 12 000 l aquarium containing aerated fresh river water. Twenty-five percent of the water was replaced daily with fresh river water. Following a 72-hour period (Gustaveson et al. 1991), to allow physiological stress from capture to subside, clove oil was added to the water to anaesthetise the fish and a further blood sample was taken and analysed as described previously. This sample was used as a control value (per individual fish) to examine the differences with values attained from the same fish at capture.

Statistical analysis

All data were analysed using SPSS for Windows, version 14. All descriptive data are reported as means ± SD (range). To elucidate the influence of fish mass on landing time, the relationship between these variables was examined using a Pearson's correlation coefficient. Differences between blood lactate concentrations of the control fish after capture and following 72 hours in the aquarium were analysed using a paired-samples *t*-test. Landing time and total angling time (landing time + handling time) were divided into minute intervals (<1 min, 1–2 min, 2–3 min, etc.) and a one-way ANOVA was used to examine the lactate response resulting from the different intervals; an LSD *post hoc* test was used to identify differences between groups. Finally, a stepwise multiple regression analysis was used to examine the variance that dependent variables (handling time, landing time, total angling time, mass, fork length, total length) contributed to blood lactate concentrations and hence metabolic angling stress.

Results

One of the 66 fish was foul-hooked (hooked on the body, rather than in or around the mouth) and as a result the data from this fish were considered anomalous and therefore removed from any further analysis. The water temperature range was 22–24 °C. The mean body mass and total length of the remaining 65 specimens was 1.71 ± 0.87 kg (0.45–4.02 kg) and 561 ± 79 mm (385–733 mm), respectively. Mean total angling time was 1 min 43 s ± 58 s (30 s–5 min 5 s) of which the landing was 1 min 2 s ± 44 s (15 s–3 min 5 s). The relationship between fish body mass and landing time (Figure 1) was significantly positively correlated ($r^2 = 0.607$, $p < 0.001$).

The mean body mass from the 15 control fish was 1.31 ± 0.16 kg (1.02–1.58 kg) and the mean blood lactate was 3.8 ± 1.5 mMol l^{-1} (1.6–7.4 mMol l^{-1}) at the time of capture. Following 72 hours in an aquarium, the mean blood lactate concentration was 1.6 ± 0.7 mMol l^{-1} (0.9–3.3 mMol l^{-1}). There was a poor relationship between the body mass and the control blood lactate ($r^2 = 0.114$). A paired-samples *t*-test revealed that blood lactate concentrations in the control fish were significantly higher following capture than following 72 hours in an aquarium ($t_{(14)} = 6.130$, $p < 0.001$).

A one-way ANOVA revealed that blood lactate concentrations were significantly different between groups when calculated using landing time ($F = 23.454$, $p < 0.001$) (Figure 2) and total angling time ($F = 17.538$, $p < 0.001$) (Figure 3). *Post hoc* analyses revealed that at every time point there was a significant elevation in blood lactate concentration when compared to the control group ($p < 0.001$). A stepwise multiple regression analysis demonstrated that only one variable contributed significantly to the variance in blood lactate concentration. Landing time was shown to account for 32% of the variance in blood lactate.

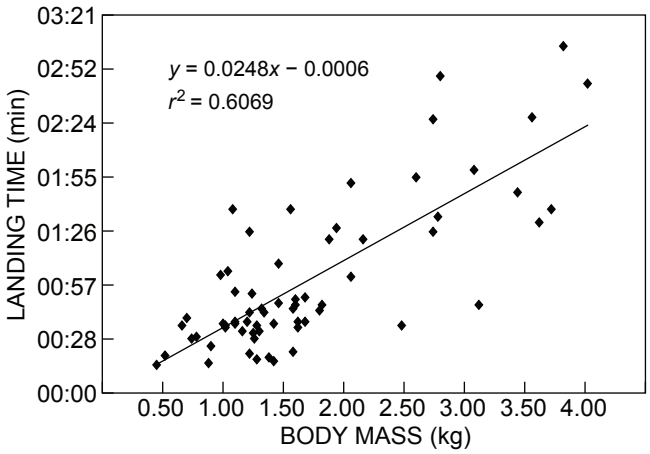

Figure 1: Relationship between tigerfish body mass and landing time (n = 65)

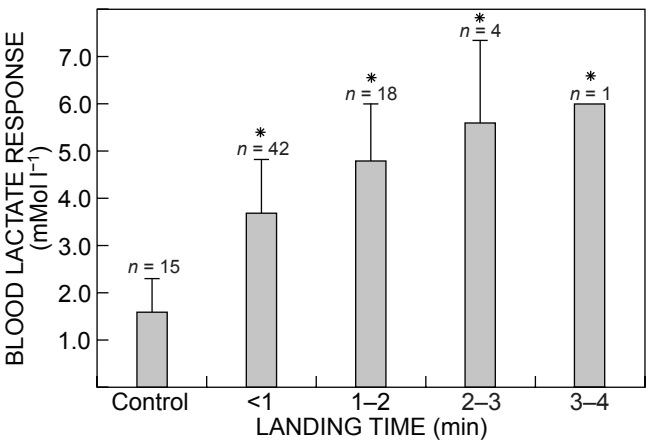

Figure 2: Blood lactate responses of control fish and angled tigerfish that were landed over various lengths of time. Values are presented as mean + SD; n = 65; * denotes that blood lactate concentrations were significantly greater than the control group (p < 0.001)

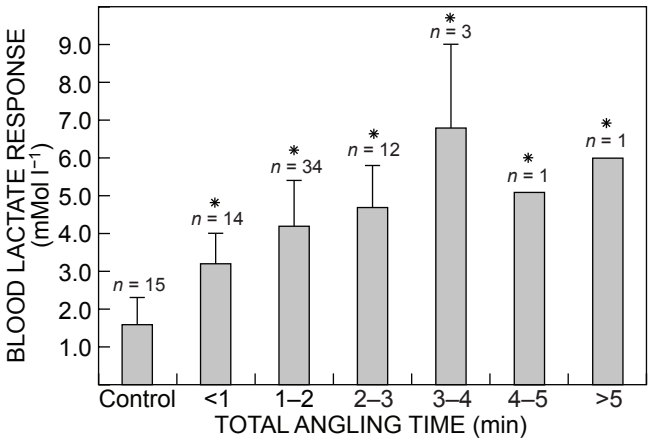

Figure 3: Blood lactate responses of control fish and tigerfish that were angled for various lengths of time. Values are presented as mean ± SD; n = 65; * denotes that the blood lactate concentrations were significantly greater than the control group (p < 0.001)

Discussion

Water temperature

Water temperature plays an important role in physiological responses to angling. Higher temperatures result in significantly greater responses to physiological variables such as lactate and cortisol in trout (Meka and McCormick 2005), which may have a detrimental effect on the prognosis of the fish. In addition, Thorstad et al. (2003) found that Atlantic salmon mortality after hook and release can increase with warmer water temperatures. During this investigation (August–September, Southern Hemisphere spring), the water temperature in the Okavango Delta ranged between 22 °C and 24 °C, but may vary from 17 °C in winter to 35 °C in summer (Mbongwe et al. 2003). If the response in tigerfish is similar to trout and salmon, then one might expect far greater levels of blood lactate during the summer months.

Angling duration versus fish size

Angling time increased with heavier fish (r^2 = 0.607). This is not surprising, given that similar results have been demonstrated in Atlantic salmon (Thorstad et al. 2003) and rainbow trout (Meka and McCormick 2005). In addition, Skelton (2001) has shown that southern African female tigerfish are generally heavier than males. This is certainly true for the Okavango, where in a sample of 208 tigerfish (~1:1 male:female ratio), all fish larger than 2 kg were female (see Gerber et al. [2009] for data on fish length–weight relationship and size at sexual maturity). Our data demonstrate that blood lactate increased with angling duration, up to approximately 6 mMol l^{-1} at 3–4 minutes. Since larger fish take longer to land, the female tigerfish, which grow the largest (>2 kg), are likely to be subjected to greater physiological stress from rod-and-line angling than the males.

In addition, the largest angling intensity in this system tends to occur during the annual catfish migrations in spring and early summer. As most large males (>1 kg) caught during this period were ripe-running, it seems that the catfish migrations coincide with the sexual ripening of male and female tigerfish (NS pers. obs.). In largemouth bass and salmon, angling stress may affect spawning behaviour and spawning success (Cooke et al. 2002, Thorstad et al. 2003). The same may also be true of tigerfish, however this requires further investigation.

Control data

This study demonstrated that there was a poor relationship between blood lactate and body mass in the control group, which suggests that blood lactate is independent of body mass and that the metabolic angling stress is the probable principal cause of elevated lactate levels (Meka and McCormick 2005).

During this study, 15 fish were successfully kept for 72 hours in an aquarium and showed significantly lower

blood lactate levels than after capture (1.6 mMol l^{-1} versus 3.6 mMol l^{-1} respectively). Results also show tigerfish blood lactate levels are significantly elevated less than one minute (3.2 mMol l^{-1}) after hooking when compared to the control (1.6 mMol l^{-1}). This indicates that there is an acute metabolic response that is likely a result of increased muscular work following hooking. These control values are lower than those reported for largemouth bass (Gustaveson et al. 1991) and rainbow trout (Meka and McCormick 2005) who reported values ranging from 1.8 mMol l^{-1} to 3.5 mMol l^{-1}. In addition, data from this study are similar (1.0 mMol l^{-1}) to what Milligan and McDonald (1988) found in Coho salmon 48 hours after capture. Based on these data we are confident that lactate levels in control fish returned to concentrations that are akin to resting levels.

Blood lactate and angling duration

The blood lactate response of the angled tigerfish was significantly greater than that of the control group at every time point, indicating that a large degree of metabolic stress is evident even following angling durations lower than one minute. Gustaveson et al. (1991) showed a comparable response in largemouth bass at similar water temperatures (28–30 °C) whereby fish angled for one minute or more had a significantly higher blood lactate level than controls. Conversely, Meka and McCormick (2005) found a significant increase in blood lactate levels of angled rainbow trout only after three minutes of total angling time; it is, however, important to note that they used rapid angled fish as controls and fished at lower water temperatures (~10–13 °C).

The multiple regression analysis showed that landing time was the only variable that significantly contributed to blood lactate elevations. This confirms that metabolic work done by the fish whilst hooked is a major contributor to the physiological stress of the fish. In addition, previous research in bluegill (*Lepomis macrochirus*) has shown that the longer the duration of air exposure (handling time) the longer the recovery period and the greater risk of mortality (Gingerich et al. 2007). Future work should consider the effect of extended air exposure time on tigerfish.

Conclusion

This study shows that blood lactate can be a useful biomarker for physiological stress in tigerfish following hook-and-line capture. Although blood lactate does provide clear evidence of significant metabolic stress, it cannot be used to determine global physiological stress and possible delayed mortality. Further research is warranted to elucidate all possible factors (e.g. cortisol, blood glucose) that may contribute to sublethal consequences (i.e. behaviour alterations, fitness impairment) of catch-and-release angling of tigerfish (also see Arlinghaus et al. 2007, Gingerich et al. 2007, White et al. 2008). Data from this investigation, plus proposed future research, should be used to make scientifically sound, evidence-based, recreational fisheries management decisions in order to provide sustainable utilisation of this popular angling species.

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