



Journal of Fish Biology (2013) **83**, 1401–1406 doi:10.1111/jfb.12216, available online at wileyonlinelibrary.com

Blood lactate loads of redthroat emperor *Lethrinus miniatus* associated with angling stress and exhaustive exercise

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(Received 9 February 2013, Accepted 9 July 2013)

Baseline, post-angling and maximum attainable blood lactate concentrations were measured for the fishery species redthroat emperor *Lethrinus miniatus* to gain insight into the condition of fish released following c. 30 s angling and <45 s air exposure. Mean \pm s.D. baseline blood lactate was $1.5 \pm 0.6 \text{ mmol } l^{-1}$, which increased and plateaued around $6 \text{ mmol } l^{-1}$ at 15-30 min post-angling. These values were significantly lower than those obtained from fish maximally exhausted with a prolonged chase and air exposure protocol following capture ($10.9 \pm 1.8 \text{ mmol } l^{-1}$), suggesting that *L. miniatus* is not maximally exhausted during standard angling practices.

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Key words: catch-and-release; fish; fisheries; post-release; stress physiology.

Angling induces burst swimming in fishes, which is primarily reliant on fast-twitch, anaerobic white muscle fibres (Wood, 1991) and can only be sustained for short periods owing to physiological changes in blood and tissue chemistry (Milligan, 1996). The physiological response of fishes to capture is of interest because disrupted physiological homeostasis has been linked with decreased survival (Black, 1958). Knowledge of the range of blood lactate concentrations attainable following angling can help to understand responses to capture and ability to tolerate associated stressors, and ultimately can be used to refine capture and handling techniques to optimize welfare.

Responses to angling are species-specific (Barton, 2002) and variable increases in lactate have been documented in plasma, whole blood and muscle tissue. Maximum lactate concentrations range from low ($<5 \text{ mmol } l^{-1}$) in coralgrouper *Plectropomus leopardus* (Lacépède 1802) whole blood ($28-30^{\circ}$ C; Frisch & Anderson, 2000) to very high (*c*. 20 mmol l^{-1}) in plasma of exhaustively exercised white trevally *Pseudocaranx dentex* (Bloch & Schneider 1801) (24° C; Wells & Baldwin, 2006),

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rainbow trout *Oncorhynchus mykiss* (Walbaum 1792) (15° C; Milligan, 1996) and coho salmon *Oncorhynchus kisutch* (Walbaum 1792) (7° C; Clark *et al.*, 2012). The time to reach peak blood lactate after angling or handling also varies, as does recovery time. Peak plasma lactate for green sturgeon *Acipenser medirostris* Ayres 1854 occurs within 15 min (19° C; Lankford *et al.*, 2003), whereas in winter flounder *Pseudopleuronectes americanus* (Walbaum 1792) whole blood it may take 2–4h ($12-14^{\circ}$ C; Girard & Milligan, 1992). Recovery of plasma lactate and other physiological variables can take 12+ h for *O. kisutch* subjected to exhaustive exercise (7° C; Clark *et al.*, 2012).

Redthroat emperor *Lethrinus miniatus* (Forster 1801) is an important fishery species on the Great Barrier Reef, with individuals regularly released if undersize (<38 cm total length, $L_{\rm T}$) or if the catch limit has been reached. This study examined baseline, post-angling and maximum attainable lactate concentrations for *L. miniatus* to provide insight into the condition of fish released following brief angling and air exposure.

Research was conducted onboard a vessel at Heron $(23.4500^{\circ} \text{ S}; 151.9167^{\circ} \text{ E})$ and One Tree $(23.5083^{\circ} \text{ S}; 152.0917^{\circ} \text{ E})$ reefs during August 2011 and February 2012. Fish (mean 46 cm L_{T}) were caught using rod and reel (13.6 kg line and 13.6 kg leader) with barbless 8/0 hooks baited with pilchard *Sardinops sagax* (Jenyns 1842) or squid *Loligo* spp. Angling time was <30 s, and time between landing and when blood sampling was recorded. Standard capture involved <45 s of air exposure for hook removal and venting of the swim bladder, representing postcapture handling by commercial fishers (A. Tobin, pers. comm.).

Blood was taken within 2 min and at 30 min (n = 22 fish, August 2011) or at 5 and 15 min (n = 5, February 2012) postcapture to identify the time course of lactate accumulation in the blood. Blood was extracted from the caudal vasculature using heparinized 19 gauge hypodermic needles and syringes while the fish's gills were submerged. Blood taken immediately on landing (*i.e.* 2 min sample) was considered to be a baseline sample (lactate response from capture should not have been apparent yet; Clark *et al.*, 2011). Fish were allowed to recover between and after blood sampling in onboard tanks (521) filled with fresh seawater (21 and 27° C, August and February, respectively). Fish appeared calm and upright with gentle fin movement during recovery.

At 20 min postcapture (February 2012), 12 fish (n = 5 from above and n = 7 additional fish, all treated as above) were kept anaerobically active for 40 min in a 1951 tank by tapping the caudal peduncle to achieve maximal exhaustion, after which they were exposed to air for 1 min and blood was taken. All fish were sacrificed after sampling.

Lactate concentration in whole blood was measured immediately using a Lactate Pro (Akray Inc., www.arkray.co.jp) portable lactate analyser, previously validated for use with teleost blood (Brown *et al.*, 2008) and for tropical species (Cooke *et al.*, 2008; O'Toole *et al.*, 2010). Whole-blood lactate for each individual was plotted against time postcapture. For concentrations below limits of the Lactate Pro analyser (measurable range $0.8-23.3 \text{ mmol }1^{-1}$), $0.7 \text{ mmol }1^{-1}$ was substituted. Paired *t*-tests compared samples with repeated reads (2 and 30 min, 5 and 15 min) and independent samples *t*-tests compared readings from different individuals (15 and 30 min). A mixed effects model (restricted maximum likelihood estimation) with Tukey's *post hoc* test compared blood samples of all individuals in the R environment (R



FIG. 1. Blood lactate concentration of *Lethrinus miniatus* postcapture where boxes represent the 25th-75th percentiles, the median is represented by lines and whiskers indicate 10th and 90th percentiles. \Box , individuals caught and allowed to rest after capture (n = 22: 2 and 30 min; n = 5: 5 and 15 min); \Box , individuals subjected to additional exhaustive exercise postcapture (n = 12: 60 min); _____, mean lactate concentrations at time intervals between 2 and 30 min postcapture. Values with the same lower case letters are not significantly different (P > 0.05). Capture (i), recovery (ii) and continuous swimming (iii, for maximally exhausted fish only) periods are indicated.

Development Core Team: www.r-project.org). This model allowed for unbalanced design, repeated measurements and heterogeneity of variances, with lactate concentration, time postcapture (fixed effects) and individual (random effect) as variables. P values were significant at the <0.05 level.

Baseline lactate concentration at <2 min postcapture was $1.5 \pm 0.6 \text{ mmol } 1^{-1}$ (mean \pm s.D.; Fig. 1). Four samples were below the detectable limit of the analyser, but no difference in the overall mean was observed when $0.7 \text{ mmol } 1^{-1}$ was substituted with zero $(1.4 \pm 0.8 \text{ mmol } 1^{-1})$. Blood lactate concentrations at 2 min postcapture were similar to those reported for temperate *O. mykiss* (*c.* 2 mmol 1^{-1} ; Ferguson & Tufts, 1992) and plasma lactate concentrations of tropical blackeye thicklip *Hemigymnus melapterus* (Bloch 1791) (*c.* 1.5 mmol 1^{-1} ; Grutter & Pankhurst, 2000).

Lactate increased significantly from $3.0 \pm 0.5 \text{ mmol } 1^{-1}$ at 5 min to $6.1 \pm 1.8 \text{ mmol } 1^{-1}$ at 15 min post-angling (Fig. 1). Fish held for 30 min did not show further elevations in lactate $(4.6 \pm 1.6 \text{ mmol } 1^{-1}; t\text{-test}, t_{25} = -1.967, P > 0.05)$. There were significant increases in mean lactate between 2 and 30 min (*t*-test, $t_{21} = -8.675$, P < 0.001) and 5 and 15 min (*t*-test, $t_4 = -4.797$, P < 0.05). Post-angling lactate concentrations for *L. miniatus* were comparable to wild caught tropical bonefish *Albula vulpes* (L. 1758) (*c*. 3 and *c*. 6 mmol 1^{-1} following 1 and 4 min of exercise; Suski *et al.*, 2007).

Model results revealed that the mean maximum blood lactate concentration from the exhaustive exercise protocol for 40 min postcapture $(10.9 \pm 1.8 \text{ mmol } l^{-1})$ was significantly higher than all other lactate concentrations ($F_4 = 107.024$, P < 0.001; Fig. 1). *Post hoc* model results indicated significant differences between each time interval combination, except 2 and 5 min and 15 and 30 min (Fig. 1), in agreement with *t*-tests. Lactate significantly increased from baseline levels to a postcapture plateau that reflected only half of the maximum level attainable. This suggests that *L. miniatus* is not maximally exhausted following standard angling. Similarly, Roth & Tore Rotabakk (2012) reported that chasing further increased lactate in saithe *Pollachius virens* (L. 1758) after being captured by longline.

Given the plateau at 15-30 min, it is reasonable to assume, based on the shape of lactate response curves from other tropical species (Frisch & Anderson, 2000; Cooke *et al.*, 2008), that lactate would begin to clear from the blood with subsequent recovery (Milligan, 1996). These results are probably indicative of the lactate response of *L. miniatus* following a typical angling encounter. Post-angling lactate of *L. miniatus* plateaued faster than salmonids after simulated angling events (Ferguson & Tufts, 1992; Clark *et al.*, 2012) but over a similar period to tropical *P. leopardus* (30 min; Frisch & Anderson, 2000) and *A. vulpes* (seine caught, 20 min; Cooke *et al.*, 2008). In contrast, species subjected to exhaustive exercise have demonstrated elevations in blood lactate that may impede swimming ability and recovery (Ferguson & Tufts, 1992; Farrell *et al.*, 2000), such as 10-16 mmol 1^{-1} in salmonids (Stevens & Black, 1966; Farrell *et al.*, 1998). Low-speed swimming and increased water flows have been linked to enhanced recovery rates of blood lactate following exhaustive exercise in temperate (Milligan *et al.*, 2000) and tropical (Brownscombe *et al.*, 2013) fishes, which may improve the condition of fishes released.

No perfect techniques exist for measuring acute stress responses (Cooke *et al.*, 2013), however, it is important to understand interactive and synergistic effects on physiology. The influence of water temperature in the blood lactate time course should be the focus of future studies, as it probably plays some role (Lankford *et al.*, 2003; Clark *et al.*, 2012). Plasma lactate concentrations of *A. medirostris* acclimated to 11 and 19° C, however, were significantly different only at resting (Lankford *et al.*, 2003), and temperature variations (*c.* 4° C) exist within studies but have not been reported to have a significant influence (Farrell *et al.*, 2000; Cooke *et al.*, 2008). It was not possible to investigate temperature effects in this study (range $21-27^{\circ}$ C) because there was no overlap in blood sampling times across seasons. It is unlikely that temperature influenced the overall conclusions of this study, as clear differences were identified between the maximum, post-angling and baseline lactate values. Yet, future research examining seasonal differences in lactate responses would be beneficial, as would sampling over a longer time course.

In conclusion, a brief angling and air-exposure event elevated blood lactate concentrations of *L. miniatus* above baseline, but this value seemed to plateau at values well below the maximum values measured for fish exposed to continuous exercise for 40 min. This suggests that *L. miniatus* released after a typical angling encounter should not experience excessively elevated blood lactate.

This study was funded by an Australian Research Council Future Fellowship to M.R.H., AIMS@JCU stipend to L.M.C. and funding provided by James Cook University. The authors would like to thank the anonymous reviewers and everyone who helped with field collections, especially A. Tobin. Treatment of all animals was conducted under JCU animal ethics number A1566.

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