Investigating the link between stress, temperature, and metabolic inertia in largemouth bass *Micropterus salmoides*

Shelby C. Mathieu and Peter J. Allen

Department of Wildlife, Fisheries, and Aquaculture, College of Forest Resources, Mississippi State University, Mississippi State, MS 39762

Abstract

Temperature is an important factor in fish survival. Temperature can affect where a fish lives, how quickly it responds to physiological factors, and the rate of metabolic processes. Stress is a common physiological response when fish are exposed to handling practices in hatcheries and angling by the public and by professionals in tournaments. Temperature and stress response research is vital in order to support fish survival, especially in hatcheries and tournaments where fish survival is of the utmost importance. This study looks at the relationship between handling stress typical of hatcheries or anglers and a temperature drop similar to what could occur in livewells or acclimation tanks on the metabolic inertia of largemouth bass *Micropterus salmoides*. Largemouth bass are often raised in hatcheries or caught by anglers and so were ideal for this study. We will look at the effect of handling stress and a 4°C drop in temperature on largemouth bass metabolic rate in order to determine how long it takes the fish to recover. Metabolic rate will be measured using respirometry and stress will be measured using plasma osmolality and plasma cortisol levels.

Introduction

Temperature is an important factor of survival in fishes as it plays a key role in their physiological responses, metabolic rate, and distribution. Fish prefer temperatures that maximize the portion of their metabolism that is available for growth, physiological responses, biochemical reactions, reproduction, and normal activity (Diaz et al. 2007). This is no different in largemouth bass. Largemouth bass tend to prefer water temperatures between 27.1°C and 29.2°C with their optimal temperature for growth being 28.1°C-28.6°C (Diaz et al. 2007). The critical thermal minima of largemouth bass is approximately 3.2°C and the critical thermal maxima of largemouth bass is approximately 38.5°C. (Beitinger et al. 1998).

Stressors have a large effect on the metabolic rate in fishes. Stress induces changes in the metabolic rate that do not allow energy to be put forth in growth, reproduction, or normal activity of the fish and disrupt the ability of the fish to return to normal patterns after the stress has passed. There are several types of stress response that fish undergo. The primary stress response is a hormonal response where catecholamines are released to stimulate oxygen and energy delivery to muscles which is then followed by cortisol release. Cortisol release is the part of the stress response that affects the metabolic rate, osmoregulation, and immune function (Kieffer and Cooke 2009). The secondary immune response can be sustained over long periods of time and is caused by changes in the metabolic, hydromineral, hematological, immunological, cardiovascular, and respiratory functions of the fish. These changes are used to provide the energy and oxygen needs that the fish require in order to "escape or fight" or to reinstate homeostasis in the fish. Plasma glucose, lactate, tissue glycogen, plasma chloride, plasma sodium, and plasma osmolality are measured in order to determine if the fish is undergoing a secondary stress response (Kieffer and Cooke 2009).

The tertiary stress response is the last stress response and reflects whole animal performance or behavior changes, and, in contrast to the primary and secondary stress responses, it is maladaptive. Tertiary stress response changes include growth rate changes, swimming capacity, disease resistance, reproductive functions, and death (Kieffer and Cooke 2009). Stress in largemouth bass has been tested by chasing with nets, cortisol implants, handling stress, such as simulated catch-and-release, and catch-and-release fishing. These studies have shown that short term stress will impair short term growth rate, plasma ion levels, and lactate levels (Suski et al. 2006; O'Connor et al. 2011; Cline et al. 2012).

As both temperature and stress affect the metabolic rate separately, it is important to see how both factors will impact metabolic rate together as this is a common concern for hatcheries and tournament anglers where fish are stressed and then moved to new water. It is important to especially concentrate on metabolic inertia, or the time for metabolic rate to change after a temperature shock to a new homeostasis, as the time this change takes can impact a fish's ability to survive as well as the duration of a fish's recovery from stress and conditions that will promote a faster recovery. Kieffer et al. (2011) studied brook trout (*Salvelinus fontinalis*) in order to determine if swimming after stress will promote a faster recovery period, and found that trout preferred to recover in flowing water and that fish that recovered in flowing water fatigued after a longer time than fish that recovered in still water. Largemouth bass will be used for this study as the time for their recovery is well documented at four hours at 25°C (Suski et al. 2006), their temperature preferences are well-known (Diaz et al. 2007), and they are a common game fish that would be often encountered in tournaments.

Hypothesis

The hypothesis for this study is that largemouth bass will have a shorter metabolic inertia time, i.e. the metabolic rate will return to a new level of homeostasis more quickly, after a sudden temperature drop of 4°C occurring after induced stress.

Methods and Materials

Largemouth bass juveniles were obtained from the Mississippi Department of Wildlife, Fisheries, and Parks Turcotte Hatchery. Bass will be acclimated in 4000-L recirculating systems with automatic temperature controls. Bass will be acclimated to either 20°C or 28°C, suboptimal or preferred temperature ranges, for 2-3 weeks. For experiments, bass will be removed to a 500-L, 3.05 m x .61 m x .61 m fiberglass experimental tank where they will be placed into a respirometer that is at their acclimation temperature. Metabolic rate will be measured for 3 hours to derive a baseline level. After a baseline level is determined, fish will either be stressed by chasing for 60 seconds or not stressed, after which the temperature will be dropped by 4°C or will remain the same and their metabolic rate will be measured for a hour period.

This tank was set to 4°C lower than that of the acclimation tank that the individual fish came from in order to stimulate a sudden shock that fish might experience upon release after angling tournaments. Intermittent respirometry measurement conditions will likely be 720 seconds long with 240 seconds as a flushing period, 60 seconds as an interim period, and 420 seconds as the measuring period, following a similar design of Arnold and Allen (2014). During the measuring period, oxygen consumption will be recorded along with tailbeat frequency and gill ventilation frequency for one 30 second interval per variable.

For each acclimation temperature, 20 bass will be used, 10 stressed and 10 not stressed for a total of 40 fish per trial. Blood samples will also be collected in order to measure plasma osmolality and plasma cortisol, which are indicators of the stress response.

Statistical Analyses

Statistical analyses will be conducted using commercial software to analyze variance.

Expected Outcomes

This study has a broad significance in research of stress and temperature of largemouth bass. Stress and temperature research in largemouth bass is mainly studied with regards to upper temperature tolerance range levels making this study unique as it will focus on the optimal temperature range and the lower levels of the temperature tolerance range and how bass handle stress in these ranges. Outcomes of the study are expected to be new knowledge on the stress response in the lower temperature tolerance range of largemouth bass, knowledge of how the stress response is affected by a sudden decrease in temperature, and how these factors will affect the short-term survival of largemouth bass. This information will be beneficial to others as bass have a wide temperature tolerance range that could act as an example for other fishes with a narrower range and the effect of a sudden drop in temperature could be similar to that of fishes placed in livewells in angling tournaments or released from livewells back into the natural habitat.

References

- Beitinger, T. L., Currie, R.J., and Bennet, W.A. 1998. Critical thermal minima and maxima of three freshwater game-fish species acclimated to constant temperatures. Environmental Biology of Fishes 51(2):187-200.
- Cline, Timothy J., Weidel, Brian C., Kitchell, James F., Hodgson, James R., and Post, John. 2012. Growth response of largemouth bass (Micropterus salmoides) to catch-and-release angling: a 27-year mark-recapture study. Canadian Journal of Fisheries & Aquatic Sciences 69(2): 224-230.
- Diaz, Fernando, Re, Ana Denisse, Gonzalez, Ricardo A., Sanchez, L. Noemi, Leyva, Gustavo, and Valenzuela, Francisco. 2007. Temperature preference and oxygen consumption of the largemouth bass Micropterus salmoides (Lacepede) acclimated to different temperatures. Aquaculture research 38(13):1387-1394.
- Kieffer, J. D., Kassie, R. S., and Taylor, S. G. 2011. The effects of low-speed swimming following exhaustive exercise on metabolic recovery and swimming performance in brook trout (Salvelinus fontinalis). Physiological and Biochemical Zoology 84(4):385-393.
- Kieffer, J. D., and Cooke, S. J. 2009. Physiology and organismal performance of centrarchids.Pages 207-263 *in* S. J. Cooke and D. P. Philipp, editors. Centrarchid Fishes Diversity,Biology, and Conservation. Blackwell Publishing Ltd, Chichester, West Sussex.
- Suski, C. D., Killen, S. S., Kieffer, J. D., and Tufts, B. L. 2006. The influence of environmental temperature and oxygen concentration on the recovery of largemouth bass from exercise: implications for live-release angling tournaments. Journal of Fish Biology 68(1):120-136.