COMPARATIVE RESPONSES OF CLOSELY RELATED CICHLIDS TO GRADED HYPOXIA

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EXTENDED ABSTRACT ONLY - DO NOT CITE

Introduction

Fish of the Amazon developed several adaptive strategies to survive the regular episodes of low dissolved oxygen (Val and Almeida-Val, 1995). The Aquatic Surface Respiration (RSA) has appeared in many fish groups of the Amazon in response to hypoxic environments. This adaptation also occurs in cichlids. In addition to this behavioral response, other adjustments, at different biological levels, may also occur in these animals as a way to keep their metabolism in pace with environmental hypoxia. The efficiency of such responses is an indicative of hypoxia tolerance in different species of cichlids. The aim of this study is to compare the responses of two Amazon cichlids, *Astronotus crassipinis* and *Symphysodon aequifasciatus*, to graded hypoxia. Oxygen consumption, opercular movements and plasma levels of glucose and lactate were analyzed. Concentration of hepatic and muscle glycogen and glycolytic and oxidative enzymes in skeletal and cardiac muscles will be presented elsewhere (Chippari-Gomes, PhD dissertation).

Material and Methods

Astronotus crassipinis (N=70; 54.3 ± 17.4g) and *S. aequifasciatus* (N=50; 28.8 ± 7.9g) were acquired from fish farms near Manaus/Brazil and transferred to indoor tanks at the Laboratory of Ecophysiology and Molecular Evolution (LEEM - INPA) for an acclimation period of two weeks. Ten individuals of each species were exposed to different oxygen levels in single metabolic chambers. Oxygen levels in the aquaria were gradually lowered by bubbling N_2 (20% per hour) from the normoxic level (6.0mgO_2/L) down the levels 3.0; 1.5; 0.75; 0.6; 0.3; and 0.0 mgO_2/L. The specimens were kept in each of these oxygen levels for 8 hours, or until they lost the equilibrium. Oxygen consumption and opercular movements were estimated in 6.0, 0.75 and 0.3mgO_2/L. Blood was collected from the caudal vein into heparinized syringes immediately after the experiments, transferred to Eppendorf tubes and kept on ice. Plasma glucose and lactate were estimated by enzymatic method using commercial kits (Doles® and Sigma Chem. Co.). Results are expressed as means ± SEM. Statistical differences between groups were tested by one-way ANOVA (Sigma Stat). The significance of difference among means was accepted when $P<0.05$.

Results and Discussion

The species *A. crassipinis* tolerates 2.5 hours in anoxia, while *S. aequifasciatus* tolerates 4.5 hours at 0.6mgO_2/L, the latter being less tolerant than the former.

VO_2 and Opercular movements

Both *A. crassipinis* and *S. aequifasciatus* presented suppression of metabolic rates (oxygen consumption) when exposed to 0.3 and 0.75mgO_2/L, respectively (Figure 1). Their opercular movements increased in hypoxia compared to normoxia (6.0mgO_2/L) (Figure 1). These responses are very common in fishes. Oxygen uptake becomes dependent upon the ambient O_2 availability when animals are exposed to critical O_2 tension. Furthermore, the depletion in environmental oxygen induces fish to compensate for by increasing their ventilation rates (Schimidt-Nielsen, 1996). Reduction in metabolism is also an important strategy to cope with hypoxia (Hochachka and Guppy, 1987). Although respiratory responses have been qualitatively the same in both species, different metabolic rates and opercular movements, reflect higher metabolic activity of *S. aequifasciatus*, which is supposed to account for the differential response as regard as hypoxia/anoxia survival.

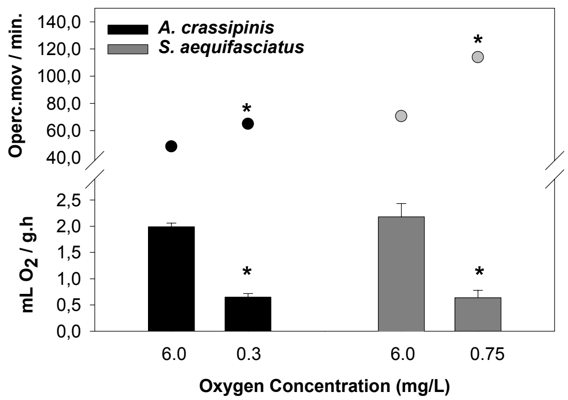

Figure 1. VO_2 and opercular movements of *A. crassipinis* and *S. aequifasciatus* exposed to normoxia and hypoxia. Symbol (*) represents significant difference compared to normoxia (6.0mgO_2/L), ($P<0.05$).

Plasma lactate and glucose

Glucose and lactate concentration in the plasma of both species increased when fishes were exposed to 0.75, 0.6, 0.3 and 0.0mgO_2/L (Figure 2) indicating an activation of anaerobic metabolism in both species. In general, levels of plasma metabolites, such as glucose, lactate, and fatty acids are altered when fishes are exposed to hypoxic stress (Val, 1993). Anaerobic metabolic activation has been observed in many fishes, including other species of the Amazon. Cichlids are particularly described as a hypoxia tolerant group, which respond to different oxygen concentration through their ability to regulate the enzyme concentration in their tissues. In fact, in another paper of this section we are describing the regulation of LDH-A gene in the same animals used in the present experiment (Oliveira et al. 2002, a companion paper). Other fish groups respond to hypoxia by depressing both aerobic and anaerobic metabolism as described in catfishes (N.P.Lopes, PhD thesis, in preparation). This does not seem to be the case of the species analyzed here.

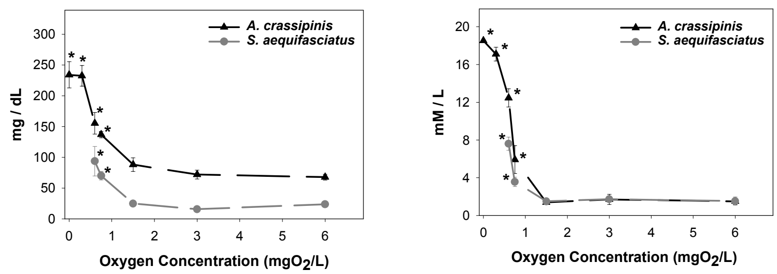

Figure 2. Concentration of plasma glucose (left panel) and lactate (right panel) of *A. crassipinis* and *S. aequifasciatus* exposed to normoxia and hypoxia. (*) represents significant difference compared to normoxia (6.0mgO_2/L), ($P<0.05$).

In the present work we observed that hypoxia tolerance ranges from moderate to high when fishes are exposed to graded hypoxia. However, this picture changes when fishes are exposed to acute hypoxia, varying from low to moderate tolerance, as observed by Chippari-Gomes et al. (2000) in four cichlid species of the Amazon . In fact, environmental oscillation in dissolved oxygen occurs gradually, both daily and seasonally (Val & Almeida-Val, 1995) allowing fish to improve physiological and biochemical adjustments to naturally survive hypoxia.

Acknowledgments

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