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Thermal tolerance and oxygen consumption rates of the catfish *Horabagrus brachysoma* (Günther) acclimated to different temperatures

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ABSTRACT

Thermal tolerance using critical thermal methodology and oxygen consumption rates of *Horabagrus brachysoma* (23.01 ± 2.04 g) were determined after acclimating the fish to 15, 20, 26, 31, 33 and 36 °C temperature for 30 days. Critical thermal maxima, CT_{Max} , (34.86 ± 0.09, 38.05 ± 0.06, 40.23 ± 0.12, 41.79 ± 0.09, 42.44 ± 0.05, and 42.79 ± 0.02) and critical thermal minima, CT_{Min} , (13.17 ± 0.09, 13.56 ± 0.05, 14.15 ± 0.10, 15.37 ± 0.10, 16.39 ± 0.12, and 18.37 ± 0.04) increased significantly ($P < 0.05$) with increasing acclimation temperatures. A thermal tolerance polygon over the range of 15 to 36 °C had a calculated area of 526.6 °C². Oxygen consumption rate increased significantly ($P < 0.05$) with increasing acclimation temperature between 15 to 31 °C and 33 to 36 °C. Maximum and minimum temperature quotients (Q_{10}) were observed between 15–20 and 31–33 °C as 2.59 and 1.53, respectively. Final preferred temperature estimated from the Q_{10} value was between 31 and 33 °C. Results indicate that *H. brachysoma* has better capability for adapting to higher temperatures and suggest their culture potential in tropical regions.

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1. Introduction

Among the various physical factors affecting the aquatic environment, temperature is of paramount importance and is considered as the 'abiotic master factor' for fishes (Brett, 1971). Any rise in the atmospheric temperature due to natural variations or pollution-induced greenhouse effect will influence the water temperature. Global climate change is suggested to potentially affect freshwater fisheries by lowering productivity in wild fish populations and in intensive aquaculture systems worldwide (Ficke et al., 2007). As fishes are poikilotherms, drastic change in their surrounding water temperature will influence their metabolic processes, behavior, migration, growth, reproduction, and survival (Fry, 1971; Pörtner, 2001). Therefore, researchers are making continuous efforts to define thermal tolerance of various fish species of aquaculture importance. Long-term changes in the environmental temperature induce ectothermic animals to display compensatory responses (which include changes in the metabolic enzymes and tissue chemistry) that are suggested to mitigate the effect of temperature on metabolism (Hazel and Prosser, 1974; Hochachka and Somero, 1971). Temperature beyond the optimum limits of a particular species, however, adversely affects fish health by increasing metabolic rate and subsequent oxygen demand, invasiveness and virulence of bacteria

and other pathogens which in turn may cause a variety of pathophysiological disturbances in the host (Wedemeyer et al., 1999).

Freshwater aquaculture constitutes one-third of the total fish production in India, where Indian major carps (IMCs; Family: Cyprinidae) contribute a major share (ICAR, 2005). Nevertheless, diversification of aquaculture, by introducing new fish species is gaining impetus due to the wide agro-climatic conditions of India and to keep pace with the mounting demand for fish protein. Catfishes are the preferred candidate species for aquaculture in India owing to their consumer preference, commercial and medicinal value (Auddy et al., 1994). Culture practices of *Clarias batrachus* and *Heteropneustes fossilis* have been popularized widely. Studies on thermal tolerance of catfishes native to India are reported for *H. fossilis* (Vasal and Sundararaj, 1978) and *Pangasius pangasius* (Debnath et al., 2006). However, the yellow catfish, *Horabagrus brachysoma* (Günther) has not received much research attention.

H. brachysoma is a rare and endemic species belonging to the family Bagridae. The distribution of this catfish is restricted to the river systems of Kerala (Karuvannoor, Chalakkudy, Pampa, Achankovil and Vembanad Lake) (Anvar Ali et al., 2007) and Karnataka (Kali and Aghanashini) (Bhat, 2001) states of Southern India. The catfish has great aquaculture potential because it is relished as a food fish and also valued in the ornamental trade (Anvar Ali et al., 2007). However, during the last few years, the population of *H. brachysoma* in Kerala has sharply declined due to overexploitation, habitat loss, pollution and anthropogenic pressures (Anvar Ali et al., 2007; Sreeraj et al., 2006) and has been listed as an endangered species (Molur and Walker, 2001). Conservation measures such as, captive breeding, milt

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cryopreservation, and culturing in small-scale rural farming systems have been considered (Sreeraj et al., 2006). Studies pertaining to the length–weight relationship (Kumar et al., 1999), characterization using cytogenetic markers (Nagpure et al., 2003), food and feeding habits (Sreeraj et al., 2006), and population genetics (Abdul Muneer et al., 2007) are reported for *H. brachysoma*. However, there are currently no published data on adaptive thermal responses of this endemic catfish. Both for successful cultivation as well as conservation of this species, it is important to understand its tolerance and adaptive responses to various environmental conditions. Therefore, the present study was conducted to investigate the thermal tolerance and oxygen consumption rates of *H. brachysoma* acclimated to different acclimation temperatures. This study will provide a better understanding of the fish's culture potential across a wide agro-climatic region, and will also provide insights into potential impacts of global warming and climatic change on *H. brachysoma*.

2. Materials and methods

2.1. Fish handling and acclimation

H. brachysoma (23.01 ± 2.04 g) procured from the local vendor (Aquatic world, Mumbai, India) were transported with proper oxygenation to the laboratory of the Central Institute of Fisheries Education, Mumbai, India. They were first given a prophylactic dip in salt solution (2%) and were then maintained in the laboratory conditions (26 ± 1 °C) for 15 days. Fish were fed *ad libitum* daily with live tubifex worms during the experimental period. Acclimation of fish (12 per test temperature; 6 per aquarium) was carried out in thermostatic aquaria (water capacity 52 L, sensitivity ± 0.2 °C) separately at test temperatures of 15, 20, 26, 31, 33, and 36 °C. The water temperature in the aquaria was increased or decreased at a rate of 1 °C per day from the ambient temperature (26 ± 2 °C) to reach the treatment temperatures. After achieving the desired test temperatures, fish were maintained in their respective temperatures for a period of 30 days. The acclimation procedure followed in our present study was based on our earlier investigations on various fishes (Chatterjee et al., 2004; Das et al., 2004; Debnath et al., 2006; Sarma et al., in press), and from other studies on sheepshead minnow, *Cyprinodon variegatus* (Bennett and Beitinger, 1997) and channel catfish, *Ictalurus punctatus* (Bennett et al., 1998). During the acclimation period, dissolved oxygen (DO) concentrations of 5.8 ± 0.5 mg/L were maintained by continuous aeration using a 2 HP centralized air blower. Water exchange was carried out on alternate days to maintain the water quality. The fish were not fed for 24 h before being subjected to the temperature tolerance and oxygen consumption experiments.

2.2. Determination of temperature tolerance

Thermal tolerance of *H. brachysoma* was evaluated after 30 days of acclimation using the critical thermal methodology (CTM). Because the CTM does not involve death as the experimental end point, it is a useful method for estimating the thermal tolerances of endangered or threatened fish species (Bennett et al., 1997; Gelbach et al., 1978). A total of 72 fish were used for determination of thermal tolerance. The CTM tests were conducted in their respective thermistatic aquarium to avoid handling stress in the fish. Fish (12 per treatment; 6 per aquarium for CT_{Max} and CT_{Min} tests, respectively) acclimated to a particular temperature were subjected to constant rate (0.3 °C per min) of increase or decrease in the water temperature until loss of equilibrium (LOE) was reached, which were designated as the critical thermal maxima (CT_{Max}) and critical thermal minima (CT_{Min}), respectively (Becker and Genoway, 1979; Beitinger et al., 2000). Continuous aeration was provided during each test to maintain adequate DO levels, and the temperature at which LOE occurred was recorded for each fish. The fish were then subsequently transferred to

their respective acclimation temperatures and monitored for the next 24 h, during which all fish were recovered. The thermal tolerance polygon was generated by plotting the acclimation temperatures on the X-axis and the mean CT_{Max} and CT_{Min} values on the Y-axis. The thermal tolerance zone area was calculated from the polygon and expressed as °C².

2.3. Determination of oxygen consumption rates

Oxygen consumption rates were measured in a static respirometer chamber, using a separate group of fish acclimated to different temperatures (15, 20, 26, 31, 33, and 36 °C), following the method adopted in our earlier investigations (Debnath et al., 2006). Briefly, fish (6 per treatment), acclimated to a particular temperature (30 days) were placed individually into a sealed glass chamber (5 L) with 6.4 mm thick glass lid, cut to cover the top portion completely. An opening in the lid fitted with a gasket to ensure an air-tight seal permitted the insertion of a DO probe. The chamber was placed inside the thermostatic aquarium set at the respective test temperatures. All four sides of the aquarium were covered with opaque screens to minimize visual disturbances of the experimental fish. The oxygen consumption experiment was carried out for an hour. The initial and final oxygen contents in the static respirometer were measured using a digital oxy-meter 330 (sensitivity 0.01 mg O₂/L, E-Merck, Germany), and the oxygen consumption rates for individual fish were expressed as mg O₂/kg/h. The temperature quotients (Q₁₀) were calculated to assess the effect of acclimation on oxygen consumption rate by using the formula:

$$Q_{10} = (\text{Rate}_2 / \text{Rate}_1)^{(10 / \text{Temp}_2 - \text{Temp}_1)}$$

2.4. Statistical analysis

Statistical analyses for the acclimation temperature dependent relationships of CT_{Max}, CT_{Min}, and oxygen consumption rate were performed using one-way analysis of variance (ANOVA) via SPSS 14.0 (SPSS, Chicago, IL, USA) for Windows. Oxygen consumption values were mass-adjusted considering mass exponent of 0.80 and reported at standard temperature and pressure (Cech, 1990). Duncan's multiple range tests were used to compare the differences among treatment means at *P* < 0.05. Data presented in the text, figures, and table are means ± standard error. Simple regression analysis was performed using Microsoft Office Excel spreadsheet.

3. Results and discussion

No mortality was recorded during the 24 h observation period subsequent to CTM trials. The CT_{Max} and CT_{Min} values increased significantly (*P* < 0.05) with increasing acclimation temperature (Table 1). The strong relationship between the acclimation temperatures and the thermal tolerance level (CTM) of *H. brachysoma* supports evidence that temperature adaptation is an essential physiological phenomenon in

Table 1
Critical thermal maxima (CT_{Max}) and critical thermal minima (CT_{Min}) of *Horabagrus brachysoma* acclimated to 15, 20, 26, 31, 33, and 36 °C.

Acclimation temperature (°C)	CT _{Max} (°C)	CT _{Min} (°C)
15	34.86 ± 0.09 ^a	13.17 ± 0.09 ^a
20	38.05 ± 0.06 ^b	13.56 ± 0.05 ^b
26	40.23 ± 0.12 ^c	14.15 ± 0.10 ^c
31	41.79 ± 0.09 ^d	15.37 ± 0.10 ^d
33	42.44 ± 0.05 ^e	16.39 ± 0.12 ^e
36	42.79 ± 0.02 ^f	18.37 ± 0.04 ^f

Values are expressed as mean ± SE (*n* = 6). Different superscript letters (a, b, c, d, e and f) in the same column indicate significant difference (ANOVA, *P* < 0.05).

fishes and is dependent on the acclimation temperature (Beitinger and Beninett, 2000). These results are in agreement with the findings of our earlier investigations on IMCs (*Labeo rohita*, *Catla catla* and *Cirrhinus mrigala*) advance fingerlings (Das et al., 2004), common carp (*Cyprinus carpio*) early fingerlings (Chatterjee et al., 2004), yellowtail catfish (*P. pangasius*) (Debnath et al., 2006), and climbing perch (*Anabas testudineus*) (Sarma et al., in press). Similar observations were also reported for CT_{Min} in red-bellied piranha (*Pygocentrus natterii*) (Bennett et al., 1997), largemouth bass (*Micropterus salmoides*) (Currie et al., 1998), and for CT_{Max} in *M. salmoides* (Currie et al., 1998), tigerperches (*Therapon jarbua*), and pearl spot (*Etroplus suratensis*) (Rajaguru, 2002). The thermal tolerance polygon area for *H. brachysoma* acclimated between 15 and 36 °C was calculated as 526.6 °C² (Fig. 1). There was no parallel report available for *H. brachysoma* or its closely related species to compare our present finding. Data extracted from our earlier investigation on the IMCs, *L. rohita*, *C. catla*, and *C. mrigala* (Das et al., 2004) over the same acclimation temperature range (15–36 °C) revealed thermal tolerance zones of 562.1, 543.4, and 603.8 °C², respectively. Results indicated that the zone of thermal tolerance of *H. brachysoma* is smaller than IMCs. The regression slope for CT_{Max} and CT_{Min} of *H. brachysoma* revealed that for every 1 °C increase in the acclimation temperature the CT_{Max} ($y = 0.37x + 30$, $R^2 = 0.9652$) and CT_{Min} ($y = 0.22x + 9.15$, $R^2 = 0.8447$) increased by 0.37 and 0.22 °C, respectively. These results showed that, in *H. brachysoma* changes in the acclimation temperatures have a greater effect on their tolerance to high temperatures than to cold temperatures; therefore, it could be suggested that *H. brachysoma* has better adaptive capability to warm temperatures than to cold temperatures.

The oxygen consumption rates of *H. brachysoma* increased significantly ($P < 0.05$) with increasing acclimation temperatures. However, no significant change in the oxygen consumption rates were observed between 31 and 33 °C (Fig. 2). In poikilothermic animals, the metabolic responses that are quantified in terms of oxygen consumption show a linear correlation to temperature due to its direct effect on the kinetics of the enzyme reactions involved (Hazel and Prosser, 1974; Hochachka and Somero, 1971). Similar observations were made for IMCs (*L. rohita*, *C. catla*, and *C. mrigala*) (Das et al., 2004), *P. pangasius* (Debnath et al., 2006), early fingerlings of *L. rohita* and *C. carpio* (Chatterjee et al., 2004), crucian carp *Carassius carassius* and *C. auratus* (Sollid et al., 2005), and *M. salmoides* juveniles (Díaz et al., 2007).

The Q₁₀ values calculated from the oxygen consumption rates decreased with increasing acclimation temperature, however, the

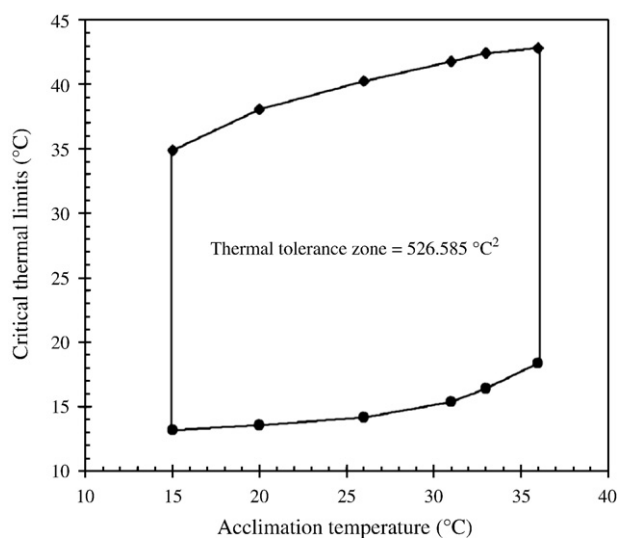


Fig. 1. Thermal tolerance polygon of *Horabagrus brachysoma* generated from the critical thermal limits (CT_{Max} and CT_{Min}) at six acclimation temperatures.

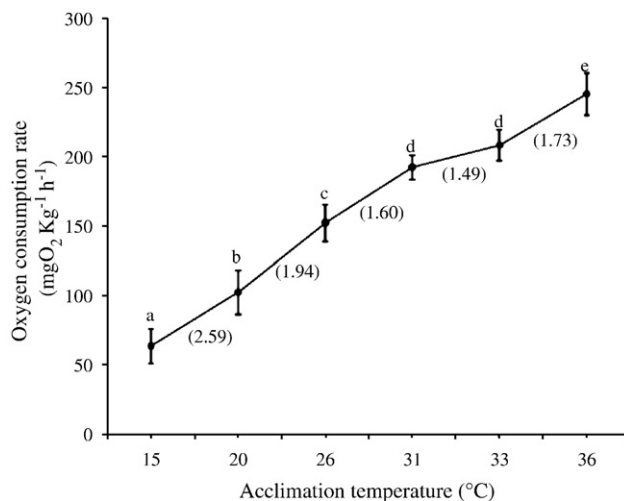


Fig. 2. Oxygen consumption rates of *Horabagrus brachysoma* acclimated to 15, 20, 26, 31, 33, and 36 °C. Values are expressed as mean \pm SE ($n = 6$). Different superscript letters (a, b, c, d, and e) indicate significant difference (ANOVA, $P < 0.05$). Numbers in parenthesis are Q₁₀s between acclimation temperatures (15–20, 20–26, 26–31, 31–33 and 33–36 °C).

values increased between 33 and 36 °C. Highest and lowest Q₁₀ values of 2.59 and 1.49 were observed between 15–20 and 31–33 °C acclimation temperatures, respectively. The Q₁₀ increased to 1.72 between 33 and 36 °C acclimation temperatures (Fig. 2). Generally, Q₁₀ values in ectotherms fall with increasing temperature (Prosser, 1986; Sollid et al., 2005; Withers, 1992). The results obtained in our present investigation are in agreement with earlier reports in *C. carassius* (Sollid et al., 2005) and *M. salmoides* juveniles (Díaz et al., 2007). However, the metabolic responses depend on the temperature, acclimation period, and the species under investigation (Dent and Lutterschmidt, 2003; Díaz et al., 2007). Dent and Lutterschmidt (2003) suggested that freshwater fish demonstrate physiological plasticity when they are able to regain or approach their metabolic set-point (viz., quantitative or qualitative changes in enzyme expression) within the context of their thermally fluctuating environments. This relationship predicts that fish with greater plasticity in the routine metabolic rate will have a smaller Q₁₀. In the present study, lowest Q₁₀ value was observed between 31 and 33 °C (1.53), which indicates that within this range *H. brachysoma* has a better capacity for maintaining homeostasis. Brett (1971) and Kellog and Gift (1983) have pointed out that the final preferred temperature coincides with optimum temperature for various physiological processes, particularly for growth (Tsuchida, 1995). Kita et al. (1996) reported that the final preferred temperature corresponds to the temperature at which increase in oxygen consumption rate with temperature is gradually lessened. The decrease in Q₁₀ indicates that the metabolic rate of the fish has decreased and that more energy is potentially available for growth (Díaz et al., 2007). Thus, the final preferred temperature may be estimated indirectly based on the relationship between Q₁₀ for oxygen consumption rates and the acclimation temperatures (Das et al., 2004). Therefore, it could be suggested that the final preferred temperature of *H. brachysoma* is in the range of 31–33 °C. Interestingly, the preferred temperature of the IMCs (*L. rohita*, *C. catla*, and *C. mrigala*) is also reported to be in the range of 31–33 °C (Das et al., 2004). Similarly, the catfish *H. fossilis* acclimatized to 28 °C (summer) or 16 °C (winter) temperatures finally selected temperatures ranging from 31.3 to 32 °C, when placed in a temperature gradient of 15 to 35 °C (Vasal and Sundararaj, 1978). These observations indicated a common trend in the final preferred temperature in these tropical water fishes, which suggests that *H. brachysoma* may also exhibit a similar pattern of energy utilization during thermal acclimation.

In conclusion, the present study showed that *H. brachysoma* has better adaptive capabilities at higher temperatures. In addition, the

final preferred temperature of *H. brachysoma* is comparable to that of the IMCs and the catfish *H. fossilis* indicating high culture potential for *H. brachysoma* in tropical freshwater regions. Further investigations on various biochemical changes including the induction of stress proteins and metabolic enzymes may help to understand the adaptive physiological responses of this catfish to high temperatures. Studies on the nutrient requirements at different life stages are also required before adopting this species for aquaculture.

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