

Available online at www.sciencedirect.com



Aquaculture

Aquaculture 258 (2006) 606-610

www.elsevier.com/locate/aqua-online

Thermal tolerance and metabolic activity of yellowtail catfish *Pangasius pangasius* (Hamilton) advanced fingerlings with emphasis on their culture potential

D. Debnath, A.K. Pal*, N.P. Sahu, K. Baruah, S. Yengkokpam, T. Das, S.M. Manush

Fish Biochemistry Laboratory, Central Institute of Fisheries Education, Fisheries University Road, Versova, Mumbai-400 061, India

Received 15 March 2005; received in revised form 17 April 2006; accepted 24 April 2006

Abstract

Thermal tolerance and metabolic activity of *Pangasius pangasius* fingerlings (22.7±1.5 g) was evaluated in terms of critical thermal maximum (CTmax), critical thermal minimum (CTmin), lethal thermal maximum (LTmax), lethal thermal minimum (LTmin) and rate of oxygen consumption after acclimating the fingerlings at 30, 34 and 38 °C for 30 days. CTmax (mean±S.E.) (42.68±0.03, 43.67±0.05, 44.05±0.12), CTmin (12.37±0.03, 14.48±0.03, 17.22±0.09), LTmax (42.95±0.02, 44.35±0.02, 44.53±0.03) and LTmin (11.75±0.02, 12.52±0.03, 14.35±0.02) increased significantly (p < 0.05) with increasing acclimation temperatures. Oxygen consumption rate increased significantly (p < 0.05) with increasing acclimation temperature from 30 to 34 °C, but the change was not significant from 34 to 38 °C. Temperature quotient (Q_{10}) of acclimated fish (2.47) was lesser than the non-acclimated fish (3.27). The area of thermal polygon over the range of acclimation temperatures (30–38 °C) of *P. pangasius* was calculated as 231 °C². Overall results indicate that thermal tolerance and metabolic activity in *P. pangasius* is dependent on acclimation temperature and is comparable to Indian major carps, suggesting their culture potential in tropical region. © 2006 Elsevier B.V. All rights reserved.

Keywords: Thermal tolerance; Oxygen consumption; Pangasius pangasius; Partial tolerance polygon

1. Introduction

As poikilotherms, fish cannot maintain body temperatures different from the surrounding water. Temperature affects biochemical and physiological activities of fish. Any rise in the atmospheric temperature due to pollution-induced greenhouse effect will influence the water temperature. Therefore, fisheries researchers have been making continuous attempts to define thermal tolerance of various species of aquaculture importance. Long-term changes in temperature lead ectothermic animals to display acclimatory responses, which may include enzymatic changes thought to mitigate the effect of temperature on metabolism (Hazel and Prosser, 1974). Prior acclimation before releasing to open water bodies has been reported to increase the post-stocking survival of walleye, *Sander vitreus* fry (Clapp et al., 1997). Rising temperature up to a certain limit, favors aquaculture production by increasing the growth rate and reducing the maturation period of fish. However, temperature beyond optimum limits of a particular species adversely affects the health of aquatic animal due to metabolic stress and hence increases susceptibility to diseases, proliferation of pathogens and oxygen demand.

^{*} Corresponding author. Tel.: +91 22 26361446; fax: +91 22 26361573.

E-mail address: akpal_53@rediffmail.com (A.K. Pal).

Fish production from freshwater aquaculture constitutes more than half of the total fish production in India, where Indian major carps contribute a major share. Nevertheless, diversification of aquaculture is very important to keep pace with the mounting demand for fish protein. Diversification by introducing new candidate species of fish/shellfish is getting impetus considering the wide agro-climatic conditions of India. Species selection for culture is primarily based on the level of input required, growth rate and tolerance to a wide range of environmental conditions. Catfishes are the preferred candidate species for aquaculture in India owing to their consumer preference, commercial and medicinal value. Culture practices of Clarias batrachus (magur) and Heteropneustes fossilis (singhi) have been popularized widely. However, catfishes of the family Pangasiidae have not received much research attention in India. Pangasius sutchi (striped catfish) and Pangasius lamaudii (black-ear catfish) are cultured in Southeast Asian countries (Paripatananont, 2002). They are naturally found in the rivers of India, Burma, Indonesia, Malaysia, Cambodia and Thailand (Ling, 1977). Pangasius pangasius (yellowtail catfish) is a fast growing omnivorous fish, which tolerate poor water quality and is a natural inhabitant of Indian waters.

Considering the future prospects and needs of culturing this catfish, we investigated temperature tolerance and oxygen consumption of P. pangasius fingerlings at three different acclimation temperatures (30, 34 and 38 °C). Lethal temperature was determined to know the thermal limit at which mortality occurs. Rate of oxygen consumption was used to estimate the metabolic activity of the fish. Critical thermal methodology (CTM) is used to estimate thermal tolerance as the mean temperature at which fish exposed to slow, constant changes in water temperature reach a predefined nonlethal (but near lethal) end point (Cox, 1974). We recommend this method as reliable to envisage the suitability of any species for aquaculture in different agro-climatic regions. This preliminary investigation highlights the impact of global warming and climatic change on the tolerance capability of P. pangasius.

2. Materials and methods

2.1. Experimental animal

Fingerlings of *P. pangasius* were brought from the Ghosh Spawn Culture Enterprise, Kolkata, West Bengal to Fish Biochemistry Laboratory, Central Institute of Fisheries Education, Mumbai, India, and were fed with a

practical diet (35% crude protein) for four weeks at ambient water temperature (28 °C).

2.2. Acclimation of experimental animals

As a pilot study, twelve fish $(22.7\pm1.5 \text{ g})$ were acclimated per treatment, separately for critical thermal methodology (CTM) and lethal thermal methodology (LTM) at 30, 34 and 38 °C in 75-L plastic tubs. Temperature in each treatment was maintained using temperature controller fitted with sensors (Selectron Process Controls Pvt. Ltd., Mumbai, India). Fish were fed twice daily with practical diet and water exchange was carried out on alternate days to maintain water quality. Acclimation of fish was carried out at 1 °C per day over ambient water temperature (28 °C) to reach the test temperatures (30, 34 and 38 °C) and maintained for 30 days. Acclimation procedure followed in this experiment was based on our earlier investigations (Manush et al., 2004; Das et al., 2004; Chatterjee et al., 2004) and from other reports on Sheepshead minnow, Cyprinodon variegates (Bennett and Beitinger, 1997). Therefore, we assume that the test fish were completely acclimated prior to CTM tests. Fish were fasted for 24 h before being subjected to CTM tests and oxygen consumption experiments.

2.3. Determination of thermal tolerance

Six fish from each acclimation temperature were transferred one by one to thermostatic aquarium (52-L water capacity, sensitivity ±0.2 °C) maintained at particular acclimation temperature before conducting thermal tolerance studies. Dissolved oxygen concentration was maintained at $5.5\pm0.5 \text{ mg l}^{-1}$ by continuous aeration. The temperature was increased or decreased at a constant rate of 0.3 °C/min until loss of equilibrium (LOE) was reached, to determine critical thermal maximum (CTmax) and critical thermal minimum (CTmin), respectively. Subsequently, fish were transferred to their respective acclimation temperatures after CTM tests and all the fish were recovered. This method has been established as a powerful tool to study stress physiology and thermal adaptation in fish (Paladino et al., 1980; Beitinger and McCauley, 1990). Similarly, lethal thermal maximum (LTmax) and lethal thermal minimum (LTmin) were determined using another lot of 12 fishes to know the lethal temperature limits of P. pangasius fingerlings in relation to acclimation temperatures. Cessation of opercular movement was considered as the end point of LTmax and LTmin (Tsuchida, 1995).

A partial thermal tolerance polygon was generated from the CTmax and CTmin data by plotting acclimation temperatures (°C) on the *X*-axis and thermal tolerance limit (°C) on the *Y*-axis. The area of thermal tolerance was calculated from the polygon (Fig. 1).

2.4. Determination of oxygen consumption

Oxygen consumption was measured at the end of acclimation period (30 days) in different acclimation temperatures (30, 34 and 38 °C) following the method adopted in our earlier investigation (Chatterjee et al., 2004) using a separate lot of acclimated fish. Briefly, six acclimated fish per treatment were placed individually in a sealed glass chamber (5 L capacity) 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 dissolved oxygen probe. An inlet and outlet using large diameter tubes were used to flush water (from thermostatic aquarium maintained at test temperatures) and continuous aeration was provided during the acclimation of fish (for 1 h), before performing oxygen consumption experiment, as per the requirement of a static respirometer (Cech, 1990). The chamber was placed inside the thermostatic aquarium set at the respective test temperatures. The oxygen consumption experiment is carried out for 1 h. All four sides of the aquarium were covered with opaque screen to minimize visual disturbances of the experimental fish. The initial and final oxygen concentration was measured using a digital oxy-meter 330 (sensitivity

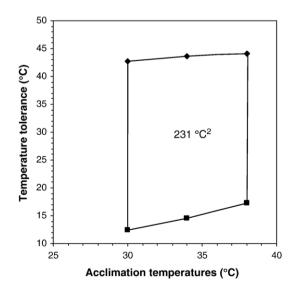


Fig. 1. Partial thermal tolerance polygon of yellowtail catfish, *P. pangasius*, fingerlings generated from critical thermal maxima and minima data collected at three acclimation temperatures.

0.01 mg O₂ l⁻¹, E-Merck, Germany) and the difference was expressed as mg O₂ kg⁻¹ h⁻¹. Similarly another set of 18 fish, which were maintained at ambient water temperature (28 °C) for 30 days, were subjected to an increase in water temperature to reach the test temperatures (30, 34 and 38 °C) in the thermostatic aquarium separately, so as to delineate the effect of an acute increase of temperature vs. acclimation procedure on oxygen consumption rate. Temperature quotient (Q_{10}) was calculated to assess the effect of acclimation on oxygen consumption using the formula:

$$Q_{10} = (\text{Rate } 2/\text{Rate } 1)^{(10/\text{Temp } 2-\text{Temp } 1)}$$

2.5. Statistical analysis

Statistical analysis of CTmax, CTmin, LTmax, LTmin and the rate of oxygen consumption were carried out using one-way analysis of variance (ANOVA via SPSS 11.0 for Windows). Oxygen consumption values were mass-adjusted considering mass exponent b=0.80 and reported at STP (Cech, 1990). Duncan's multiple range test (DMRT) was carried out for comparison of means (p<0.05), if they were significant.

3. Results and discussion

Mean values of CTmax (42.68±0.03, 43.67±0.05, 44.05 ± 0.12) and CTmin (12.37 ± 0.03 , 14.48 ± 0.03 , 17.22 ± 0.09) of *P. pangasius* fingerlings increased significantly (p < 0.05) with increasing acclimation temperatures (30, 34 and 38 °C). Similarly, LTmax $(42.95\pm0.02, 44.35\pm0.02, 44.53\pm0.03)$ and LTmin $(11.75\pm0.02, 12.52\pm0.03, 14.35\pm0.02)$ of *P. pangasius* fingerlings increased significantly (p < 0.05) with increasing acclimation temperatures. These results are in agreement with recent investigations in our laboratory on finfish (Das et al., 2004; Chatterjee et al., 2004) and shellfish (Manush et al., 2004). It is obvious from the results that the thermal tolerance of aquatic animals is dependent on the thermal exposure history before the experiment, which is regarded as acclimation. Fish have specific behavior in response to thermal acclimation, beyond which breakdown occurs, leading to stress and production losses due to anorexia, disease outbreak and ultimately death.

Data pertaining to oxygen consumption of *P. pangasius* fingerlings is presented in Table 1. Rate of oxygen consumption increased significantly (p<0.05) with increasing acclimation temperatures. Oxygen consumption is an index of metabolism in freshwater fish (Kutty and Peer Mohamed, 1975) and is dependent

609

on acclimation temperatures (Kita et al., 1996). Absolute values of oxygen consumption rate were similar to our earlier investigation in Indian Major Carps (Das et al., 2004). Temperature quotient (Q_{10}) was found to be 2.47 for acclimated fish and 3.27 for non-acclimated fish over the range of acclimation temperatures (30–38 °C), which indicates that acclimation episode has played vital role to maintain the homeostasis of *P. pangasius* over the test temperatures.

The zone of thermal tolerance polygon (Fig. 1) over the range of acclimation temperatures (30, 34 and 38 °C) was calculated as 231 °C². Data extracted from our earlier investigation in Indian major carps (*Labeo rohita*, *Catla catla*, *Cirrhinus mrigala*) and the common carp (*Cyprinus carpio*) over the same acclimation range (30– 38 °C) reveal similar zone of thermal tolerance polygon (219.44, 216.56, 231.4 and 260.48 °C²) as indicated in Table 2. Results indicate that zone of thermal tolerance of *P. pangasius* over the acclimation range (30–38 °C) is higher than Indian major carps. However, their tolerance limit was lower than *C. carpio*.

Behavior of the fingerlings subjected to critical thermal methodology was recorded. Fish acclimated to 30 °C showed restlessness and escape attempts at 39 °C, but settled to the bottom of the aquarium at 41 °C. Unorganized swimming with intermittent swift movements was noticed and fish tried to jump out of the aquarium at 42 °C during CTmax tests. Fish subjected to CTmin tests appeared motionless as CTmin endpoint was reached. Stiffening of pectoral fins was observed during LTmin tests. Fish acclimated to 34 and 38 °C showed common behavioral changes such as hanging of fish in the column with head up and tail down (vertically), swirling movement, cloudiness of eyes, gasping, frequent opercular movements, jerky motions, sluggish movements, open-mouth, bent body, etc.

Our investigation clearly indicates that thermal tolerance and metabolic activity of *P. pangasius* is dependent on acclimation temperature. There are no parallel reports available on the thermal tolerance limit of *Pangasius* spp. although they are being considered as

Table 1

Mass-adjusted rate of oxygen consumption (mg $O_2 kg^{-1} h^{-1}$) in acclimated and non-acclimated condition of *P. pangasius* fingerlings acclimated to three temperatures

Temperatures (°C)	30	34	38
Acclimated Non-acclimated		$\begin{array}{c} 231.72{\pm}3.10^{b} \\ 258.29{\pm}0.80^{b} \end{array}$	$\begin{array}{c} 236.25 {\pm} 1.50^{b} \\ 265.01 {\pm} 1.15^{b} \end{array}$

Different superscripts in the same row indicate significant (p<0.05) difference. Values are reported as mean±S.E. (n=6) at standard temperature and pressure.

Table	2
-------	---

Thermal tolerance data of Indian major carps and common carp (extracted from earlier investigations)

Acclimation temperatures (°C)	Species and stage	CTmax	CTmin	Reference
30	Labeo rohita advanced fingerling	41.52	14.01	Das et al., 2004
30	Catla catla advanced fingerling	41.16	14.23	Das et al., 2004
30	Cirrhinus mrigala advanced fingerling	42.46	13.52	Das et al., 2004
30	Cyprinus carpio early fingerling	40.60	8.60	Chatterjee et al., 2004
34	L. rohita advanced fingerling	42.87	15.19	Das et al., 2004
34	C. catla advanced fingerling	42.85	15.37	Das et al., 2004
34	<i>C. mrigala</i> advanced fingerling	42.84	13.99	Das et al., 2004
34	<i>C. carpio</i> early fingerling	42.58	10.02	Chatterjee et al., 2004
38	L. rohita advanced fingerling	43.30	15.95	Das et al., 2004
38	C. catla advanced fingerling	43.18	15.97	Das et al., 2004
38	<i>C. mrigala</i> advanced fingerling	43.23	14.32	Das et al., 2004
38	<i>C. carpio</i> early fingerling	43.86	10.74	Chatterjee et al., 2004

a candidate species for aquaculture in Southeast Asian countries. Therefore, our report on the thermal tolerance limits may help to assess the culture potential of this species in various agro-climatic regions of the world. The thermal tolerance study is considered as a useful approach in assessing potential survival, the impact of local and global climatic change, assessing colonization and potential culture of any candidate fish species. From this pilot study, it is evident that thermal tolerance limit of P. pangasius is comparable to Indian major carps, indicating their potential for diversifying freshwater aquaculture, owing to the better flesh quality and lesser intermuscular bones of former over latter. However, more research in future is required to be carried out at wide acclimation temperatures to know the complete zone of thermal tolerance, which may strengthen our hypothesis. Over all results highlight the potential of P. pangasius as a candidate species for aquaculture in tropical region.

Acknowledgements

Authors are thankful to the Director, Central Institute of Fisheries Education, Mumbai, India for providing all the facilities required for the present work. Thanks are due to Board of Research in Nuclear Sciences (BRNS), Department of Atomic Energy (DAE), Government of India for providing the thermostatic aquaria for our study (BRNS Sanction No. 99/36/22/BRNS. Grant No. 089).

References

- Beitinger, T.L., McCauley, R.W., 1990. Whole animal physiological processes of the assessment of stress in fishes. J. Great Lakes Res. 16, 542–575.
- Bennett, W.A., Beitinger, T.L., 1997. Temperature tolerance of the sheepshead minnow, *Cyprinodon variegates*. Copeia 77–87.
- Cech, J.J., 1990. Respirometry. In: Schreck, C.B., Moyle, P.B. (Eds.), Methods for Fish Biology. American Fisheries Society, Bethesda, MD, pp. 335–356.
- Chatterjee, N., Pal, A.K., Manush, S.M., Das, T., Mukherjee, S.C., 2004. Thermal tolerance and oxygen consumption of *Labeo rohita* and *Cyprinus carpio* early fingerlings acclimated to three different temperatures. J. Therm. Biol. 29, 265–270.
- Clapp, D.F., Bhagwat, Y., Wahl, D.H., 1997. The effect of thermal stress on walleye fry and fingerling mortality. North Am. J. Fish. Manage. 17, 429–437.
- Cox, D.K., 1974. Effects of three heating rates on the critical thermal maximum of bluegill. In: Gibbons, J.W., Sharitz, R.R. (Eds.), Thermal Ecology. National Technical Information Service, CONF-730505, Springfield, Virginia, pp. 158–163.

- Das, T., Pal, A.K., Chakraborty, S.K., Manush, S.M., Chatterjee, N., Mukherjee, S.C., 2004. Thermal tolerance and oxygen consumption of Indian major carps acclimated to four temperatures. J. Therm. Biol. 29, 157–163.
- Hazel, J.R., Prosser, C.L., 1974. Molecular mechanisms of temperature compensation in poikilotherms. Physiol. Rev. 54, 19–42.
- Kita, J., Tsuchida, S., Setoguma, T., 1996. Temperature preference and tolerance, and oxygen consumption of the marbled rock-fish, *Sebastiscus marmoratus*. Mar. Biol. 125, 467–471.
- Kutty, M.N., Peer Mohamed, M., 1975. Metabolic adaptations of mullet, *Rhinomugil cersula* (Hamilton) with special reference to energy utilization. Aquaculture 5, 253–270.
- Ling, S.W., 1977. Aquaculture in Southeast Asia: A Historical Overview. University of Washington Press, Seattle, WA, USA, p. 108.
- Manush, S.M., Pal, A.K., Chatterjee, N., Das, T., Mukherjee, S.C., 2004. Thermal tolerance and oxygen consumption of *Macro-brachium rosenbergii* acclimated to three temperatures. J. Therm. Biol. 29, 15–19.
- Paladino, F.V., Spotila, J.R., Schubauer, J.P., Kowalski, K.T., 1980. The critical thermal maximum: a technique used to elucidate physiological stress and adaptation in fish. Rev. Can. Biol. 39, 115–122.
- Paripatananont, T., 2002. Snakehead and *Pangasius* catfish. In: Webster, C.D., Lim, C. (Eds.), Nutrient Requirements and Feeding of Finfish for Aquaculture. CAB International, Wallingford, UK, pp. 396–401.
- Tsuchida, S., 1995. The relationship between upper temperature tolerance and final preferendum of Japanese marine fish. J. Therm. Biol. 20, 35–41.