

Research Article

Thermal tolerance and aerobic scope of tetra-hybrid tilapia Pargo-UNAM

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ABSTRACT. Fish actively select an adequate environment that fits their optimum culture/preferred temperature, this mechanism is called thermoregulatory behavior. However, what exactly is this mechanism, how does it work and what can we learn from it? Helped by their thermal limits, fish avoid temperature variations not favorable for their maximum growth. They find a thermal window where optimal temperature culture is located and use it in the best way possible for all activities. The present study is based on thermal biology studies, and its purpose is to learn the aerobic scope functions on Pargo-UNAM juveniles. The importance of this study is related to the fact that Pargo-UNAM fish, being a hybrid, has five different genetic backgrounds. We found that acclimation temperature influenced the preferred temperature of Pargo-UNAM, having a metabolic adjustment in the 20-32°C range; the Final preferendum obtained was 29.5°C. The maximum and minimum range of critical thermal limits was between 39.2-43.5°C and 8-14.9°C, respectively. The thermal window had an area of 355.2°C². The acclimation response ratio had a 0.40-0.35 interval for CTMax, and 0.52-0.69 for CTmin. Chase method used in Pargo-UNAM caused a maximum aerobic scope at 29°C. Blood lactate concentration was the highest in fish acclimated at 20°C; these values decreased while acclimation temperature increased. Results from Pargo-UNAM juveniles showed that these can be grown successfully in a 26-32°C temperature range, with their greater performance at 29°C, where the aerobic scope was at its maximum capacity.

Keywords: critical thermal, preferred temperature, thermal window, metabolism, lactate, aquaculture.

INTRODUCTION

Tilapia is the most cultivated fish group around the world, ranking second in production after carp (FAO, 2014). It would be important to have a thorough knowledge of the basics about its biological cycle, specifically concerning the thermotolerance aspects to produce tilapia commercially. Optimum temperature from tilapia ranges from 20°C to 30°C, and the reproduction occurs successfully at 26-29°C (El-Sayed,

2006; Saavedra-Martínez, 2006). Pargo-UNAM (PU) is a red tilapia obtained via hybridization through inter-specific crosses (Ramírez-Paredes *et al.*, 2012). It was developed in the Centro de Enseñanza, Investigación y Extensión en Ganadería Tropical (CEIEGT) by Muñoz-Cordova & Garduño-Lugo (2003), in an attempt to improve the production of red tilapia strains in Mexico. Learning about the potential of hybrid tilapia through thermal biology would give us an advantage on aquaculture research.

The temperature is the main abiotic factor that influences numerous aspects of fish biology, distribution, and behavior (Fry, 1947). Thermal preference is an aspect of each species genetically inherent in its behavior. Aquatic organisms have developed a thermoregulatory behavior that enables them to actively select a thermal habitat that matches with their optimum temperature. In this habitat, it avoids temperature variations and reaches its best performance (Kelsch & Neill, 1990; Golovanov, 2013). The influence of endogenous factors such as age, weight, food availability, density, and pathogens, as well as abiotic factors such as season, water quality, and light intensity have a profound impact in the thermal behavior (Wedemeyer *et al.*, 1976; Reynolds & Casterlin, 1978; Díaz *et al.*, 2004).

Thermal tolerance of many aquatic ectotherms has been calculated through the critical thermal limits, consisting of exposing the fish to a constant increase or decrease rate of water temperature until a non-lethal endpoint is reached. At this point, fish may show a loss in the righting response or onset muscle spasms (Bennet & Beitinger, 1997). Thermal windows, minimum and maximum temperatures an organism can successfully tolerate, provide general insights into the niches of taxa (Fry, 1971; Rezende *et al.*, 2014). Thermal thresholds represent a combination of physiological and biochemical features (Somero, 2004).

The oxygen consumption rate increases as the temperature rises, until the critical temperature threshold, is reached. At this point, there is a cellular transition from aerobic to anaerobic metabolism; therefore, the critical thermal limit becomes a match when the aerobic scope is close to zero. Above this point, the aerobic metabolism is suppressed due to a failure in the functions of ventilation and/or blood circulation, even if there is enough oxygen concentration available in the environment (Jost *et al.*, 2012). Survival beyond the critical thermal maximum (CTMax) point is limited by time, due to the insufficient ATP yield from anaerobiosis. Temperature intervals between the critical thermal limits and optimal temperature are called Pejus, a transitory temperature where protection mechanisms against radical oxygen molecules are activated (Cumillaf *et al.*, 2016; Rodríguez-Fuentes *et al.*, 2017).

The fish aerobic scope is determined by the difference between the oxygen consumption maximum or maximum metabolic rate (MMR) and the standard metabolic rate (SMR) (Clark *et al.*, 2013; Farrell, 2013, 2016; Roche *et al.*, 2013; Chabot *et al.*, 2016). Pörtner (2010) and Sokolova *et al.* (2012) propose that the maximum aerobic scope (AS) can be obtained in the

optimal temperature because in this condition the organism reaches its maximum performance. Nonetheless, when the relationship between the MMR and SMR curves and acclimation temperature is extreme, the aerobic scope is almost zero; therefore, the metabolic condition has a minimum performance (Fry, 1947; Ferreira *et al.*, 2014).

The response of blood lactate to exercise has been used to evaluate the aerobic capacity of sedentary, active or slow swimmers. After 5 min of a fish being chased, lactate builds up in its blood. For this reason, blood levels have been included as an indicator of the onset of anaerobic metabolism (Peak *et al.*, 1997; Brooks *et al.*, 1999).

The present study was designed to evaluate thermal tolerance responses after different acclimation temperatures in Pargo-UNAM juveniles and determine the aerobic scope with the chase method. The main goal is to comprehend the thermal physiology of this hybrid, to improve and extend its aquacultural practices. Another goal is to understand how hybrid tilapia could deal with climate changes, which may affect its distribution.

MATERIALS AND METHODS

Obtaining experimental organisms

Pargo-UNAM fingerlings were obtained from the aquaculture facility at Centro de Enseñanza, Investigación y Extensión de Ganadería Tropical (CIEGT-UNAM) at Tlapacoyan, Veracruz, and then transferred to Departamento de Biotecnología Marina del Centro de Investigación Científica y de Educación Superior de Ensenada (CICESE). A group of 250 fish ranging from 0.5 to 1.5 g wet weight was maintained in two 2000 L reservoirs with a continuous freshwater flow and constant aeration (water temperature was maintained at 28°C). Fish were fed with the commercial diet Nutripec^{MR} Purina^{MR} pellets (2.4 mm, 44% protein, 15% lipids, Agribands Purina México S. de R.L, México) three times per day to satiation, and feces were removed daily. Water quality parameters were as follows: pH 8.0 ± 0.1 , 6.6 ± 0.8 mg O₂ L⁻¹ and alkalinity was 115.5 ± 9.5 mg CaCO₃ L⁻¹.

Thermal acclimation

Two hundred and fifty Pargo-UNAM juveniles were acclimated to determine the preferred temperature (PT), critical thermal maximum (CTMax), critical thermal minimum (CTMin), and aerobic scope (AS), in five 500 L tanks with 50 individuals for each acclimation temperature (AT), regulated with a 1000 W immersion heater or cold finger cooler. The temperature was increased or reduced from 28°C (1°C per day) until

reaching temperatures of 20, 23, 26, 29, and 32°C with a daily variation of $\pm 1.0^\circ\text{C}$; acclimation time lasted for 30 days in controlled conditions (Saavedra-Martínez, 2006). The system allows an open water flow. Water quality was maintained at pH 7.8 ± 0.1 , dissolved oxygen at $6.6 \pm 0.3 \text{ mg O}_2 \text{ L}^{-1}$, and alkalinity by $115.5 \text{ mg CaCO}_3 \text{ L}^{-1}$. Organisms were fed at satiation every day three times a day. Feces were removed daily. During acclimation, no signs of mortality were detected.

Preferred temperature

An acute method was used to determine the preferred temperature with a horizontal temperature gradient, as described by Díaz *et al.* (2007). The temperature gradient ranged from 8 to 40°C ($y = 3.542 + 1.426 x$; $R^2 = 0.95$, where y = segment temperature, and x = gradient segment). The intersection graphically calculated the final preferendum (FP) between preferred temperatures and the equality line (González *et al.*, 2010).

To learn the preferred temperature (PT) of the organisms, we followed the acute method described by Reynolds & Casterlin (1979). 12 h before introducing the Pargo-UNAM in the horizontal thermal gradient, we labeled two groups of five organisms each with a plastic mark sewn to the caudal fin. Each group was placed in the virtual chamber of the corresponding experimental acclimation temperature. All temperatures are measured with an infrared digital thermometer (Steren model Her-425, Mexico). To avoid interference from feeding, and as recommended by Beamish & Trippel (1990), fish were not fed in a 24 h period before the trials. The experiment lasted for 2 h, and during its course, every 10 min we were recording the chamber (among the 20 chambers making up the gradient) at which organisms were located and the temperature.

CTMax, CTmin, ARR, and thermal window

CTMax data were obtained using 10 individuals, from each experimental condition, 40 L aquarium provided with 1000 W immersion heater and permanent aeration, to maintain a uniform temperature (González *et al.*, 2010). Fish were introduced to the aquarium 30 min before the temperature started to increase, to reduce the direct effects of handling stress during measurements (Pérez *et al.*, 2003). The temperature was increased at a rate of 1°C min^{-1} from each acclimation temperature. This rate should be slow enough to allow deep body temperature and to follow the test temperature without a significant time delay until they showed a loss of righting response (LRR) symptoms (Lutterschmidt & Hutchinson, 1997). After reaching this point, organisms were returned to their acclimation temperature.

CTmin data were obtained from 10 Pargo-UNAM juveniles from each experimental condition. Then, they were introduced into the thermal horizontal gradient; a minimum temperature of 6.8°C was set in the first segment. A fish was transferred 30 min before the experiment into the plastic cage, reducing the stress caused by handling (Pérez *et al.*, 2003). Each cage was placed in sequential order in the gradient segment with AT and gradually was moved into the colder side at the rate of 1°C min^{-1} . LRR symptoms were recorded when the organism showed them, and they were returned immediately to acclimation temperature.

A thermal window was built with CTMax and CTmin values at each AT using a modified version of the method of Bennet & Beitinger (1997). Preferred temperature, $\text{Pejus}_{\text{max}}$, and $\text{Pejus}_{\text{min}}$ were included in the thermal window (Cumillaf *et al.*, 2016).

The acclimation response ratio (ARR) was calculated as an index to learn the magnitude of thermal acclimation using equation 1, proposed by Claussen (1977).

$$ARR = \frac{\Delta CT_{\text{Max}} \text{ or } \Delta CT_{\text{Min}}}{\frac{\Delta T}{CT_{\text{Max}2} \text{ or } CT_{\text{Min}2} - CT_{\text{Max}1} \text{ or } CT_{\text{Min}1}}} \quad (1)$$

where: $CT_{\text{Max}2} - CT_{\text{Max}1}$ or $CT_{\text{Min}2} - CT_{\text{Min}1}$ (1) represent the difference between the values of critical temperatures (max or min) within the whole acclimation temperature range examined ($TA_2 - TA_1$).

Chase method

Twelve juveniles were randomly selected from each AT tank to calculate the maximum metabolic rate (MMR). First, fish were placed into a circular exercise tank with fresh water; one by one (47 cm diameter, 15 cm water depth); temperature and aeration in the tank were controlled. After 15 min of recovery time by handling, fish were continuously chased with nets and lightly pushed when they stopped swimming or if they slowed down. The chasing time given was 5 min, according to Roche *et al.* (2013) and Norin *et al.* (2014); after that time, they were immediately introduced in a hermetic respirometric chamber and submerged in a freshwater bath, having the temperature controlled at the same AT as in the last environment they were at. We used six respirometric chambers (1 L) for each repetition provided with an optic fiber oxygen sensor (precision $\pm 0.005\% \text{ O}_2$, detection limit $0.03\% \text{ O}_2$) and connected them to an OXY-10 mini-amplifier (PreSens GmbH, PreSens[®], Germany). A respirometric chamber without fish was used as a control to measure the oxygen consumed by microorganisms, by subtracting it in the final data. The initial oxygen concentration was recorded with an opened water flow; the water flow was immediately interrupted for 5 min,

and the final oxygen concentration was recorded. The following equation was used to calculate the respiration rate:

$$MO_2 = (O_{2(i)} - O_{2(f)}) \times \frac{V/t}{M} \quad (2)$$

where MO_2 is the respiration rate ($\text{mg O}_2 \text{ h}^{-1} \text{ kg}^{-1} \text{ ww}$), $O_{2(i)}$ is the initial oxygen concentration in the chamber ($\text{mg O}_2 \text{ L}^{-1}$), $O_{2(f)}$ is the final oxygen concentration in the chamber ($\text{mg O}_2 \text{ L}^{-1}$), V is the water volume in the chamber, minus the water volume displaced by the animal (L), t is the time (h), and M is the body mass of the experimental animal (kg^{-1} wet weight). Fish were weighed, and blood samples were taken from the caudal vein and finally returned to their respective AT tank.

Twelve fish were maintained for 24 h in the respirometric chamber and submerged in a bath of controlled temperature with open flow water to calculate SMR, at the same AT as in the last environment they were at. During this evaluation, fish were not fed. After that, flowing water was interrupted for 5 min, and the oxygen concentration was measured. The lower oxygen consumption was used to calculate the SMR. Both respiration rates were calculated from the oxygen consumption as $\text{mg O}_2 \text{ h}^{-1} \text{ kg}^{-1} \text{ ww}$. Induced AS can be obtained as MMR-SMR , where AS reflects the muscle activity of the chased fish. The factorial aerobic scope was obtained by MMR/SMR and $Q_{10} = (\text{MMR/SMR})^{10/(T_2-T_1)}$ where T_2 and T_1 represent AT.

Blood lactate

Blood samples were obtained with disposable 2 mL (pediatric) syringes from the caudal vein from each experimental fish, exposed to the chase method; then, a drop was immediately placed on a disposable lactate test strip of the Accutrend® Plus (Roche Diagnostics GmbH). For all the trials of metabolic rates, the measure of lactate was expressed as mmol L^{-1} .

Data and statistical analysis

First, data were pooled for each acclimation temperature, to find differences among medians obtained from Acute method results. An exploratory data analysis (Tukey, 1977) and a one-way ANOVA Kruskal-Wallis analysis were performed. Linear regression was calculated from median values of Preferred Temperatures to determine the kind of response shown by organisms, in accordance with Johnson & Kelsch (1998). A descriptive statistical analysis (Tukey, 1977) and the one-way ANOVA test (Shapiro-Wilk) by Holm-Sidak were applied to detect the differences, with an All Pairwise comparison between mean values from each CTMax and CTmin data. MMR and SMR metabolic rates and lactate mea-

Table 1. ARR of tilapia Pargo-UNAM juveniles at different ARR acclimation temperatures, acclimation response ratio, obtained from mean values of CTMax and CTmin and temperature intervals.

	Acclimation temperature (°C)			
ARR	20-26	23-29	26-32	Average ARR
CTmin	0.59	0.69	0.52	0.60
CTMax	0.40	0.37	0.35	0.37

surements were plotted, and the differences were statistically compared from chase results. All statistical tests and graphics were performed using Sigma Plot v.12.

RESULTS

The temperature selection of Pargo-UNAM increased with the rise of AT. The slope obtained from the linear regression equation was 0.29 ($\text{PT} = 21.420 + 0.290 \times \text{AT}$), $R^2 = 0.831$. The final preferendum was 29.50°C . The critical thermal limits for Pargo-UNAM juveniles increased as the acclimation temperature rose ($P < 0.05$). CTMax and CTmin ranges were 39.2 to 43.5°C and 8 to 14.9°C , respectively. The acclimation response ratio (ARR) range was 0.35 - 0.40 for CTMax and 0.52 - 0.69 for CTmin (Table 1).

The thermal window obtained for Pargo-UNAM showed a total area of 359.1°C^2 . The optimal zone outlined by the preferred temperatures showed an area of 63.93°C^2 (Fig. 1). The zone representing $\text{Pejus}_{\text{max}}$ and $\text{Pejus}_{\text{min}}$ showed areas of 122.5°C^2 and 169.3°C^2 , respectively.

The maximum metabolic rate increased with acclimation temperature from 20 to 29°C , showing its peak in organisms acclimated to 29°C and $261.0 \pm 11.6 \text{ mg O}_2 \text{ h}^{-1} \text{ kg}^{-1}$ wet weight, and oxygen consumption is significantly reduced to 32°C . SMR values maintained a similar trend in organisms acclimated from 20 through 32°C (Fig. 2a). The highest AS was in the fish acclimated to 29°C , with $187 \text{ mg O}_2 \text{ h}^{-1} \text{ kg}^{-1}$ wet weight value, AS decreased in organisms acclimated to 20 and 32°C (Fig. 2b). Blood lactate concentrations showed that the value decreased significantly from 1.8 to 1.15 mmol L^{-1} in 20°C and 32°C , respectively ($P < 0.05$) from fish exposed to chase. Controls ranged from 0.5 to 0.7 mmol L^{-1} (Fig. 2c).

DISCUSSION

According to Kelsch & Neill (1990) and Johnson & Kelsch (1998), the relationship between preferred temperature (PT) and the acclimation temperature can be divided into three classes: positive, independent, and

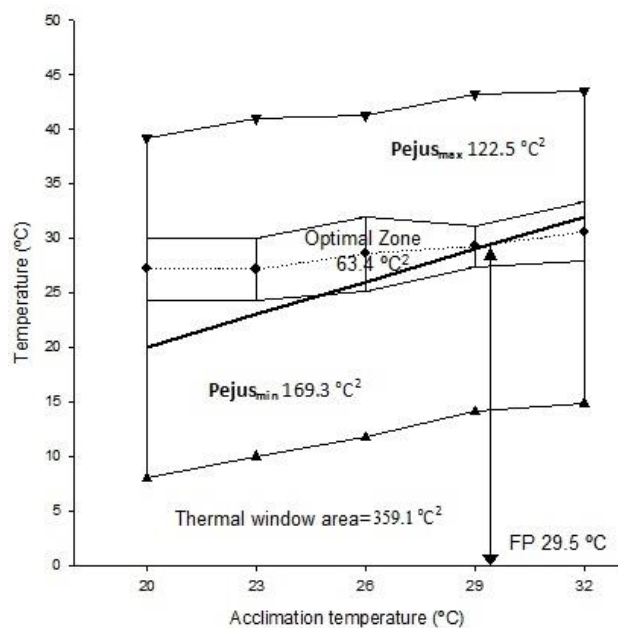


Figure 1. A thermal window for Pargo-UNAM juveniles acclimated to different temperatures. The black dots with lines represent mean values from preferred temperatures \pm SD. The 45° line represents the point where preferred and acclimation temperature are equal. The triangles represent the CTMax and CTmin, and the area outlined by these points includes $Pejus_{max}$ and $Pejus_{min}$.

negative. In the case of Pargo-UNAM juveniles, the 0.29 slope was positive, which represents a partial physiological adjustment to improve the metabolic efficiency at the new temperature. Later, this will reflect on a preferred temperature increase (Johnson & Kelsch, 1996). A positive response has been found in other fish such as Mozambique tilapia (*Oreochromis mossambicus*), common carp (*Cyprinus carpio*), bluegill (*Lepomis macrochirus*), and largemouth bass (*Micropterus salmoides*) (Badenhuizen 1967; Cherry *et al.*, 1975; Cincotta & Stauffer, 1984). Species experiencing annual cycles of relatively high amplitude were expected to show temperature-preference relationships that are positive functions. Therefore, Pargo-UNAM juveniles can be classified in the eurytherms group.

Pargo-UNAM (PU) juveniles final preferendum (FP) was very close to other species of tilapia: Badenhuizen (1967) and Stauffer (1986) obtained a preferred temperature for Mozambique tilapia between 28 and 32.2°C, respectively; for Nile tilapia (*Oreochromis niloticus*), Beamish & Trippel (1990) reported a 28-29.5°C interval. Some studies in *O. niloticus* (Azaza *et al.*, 2008; Abdel-Tawwab & Wafeek, 2014) have shown preferred temperature in the 28-32°C interval. Watanabe *et al.* (1993) reported that

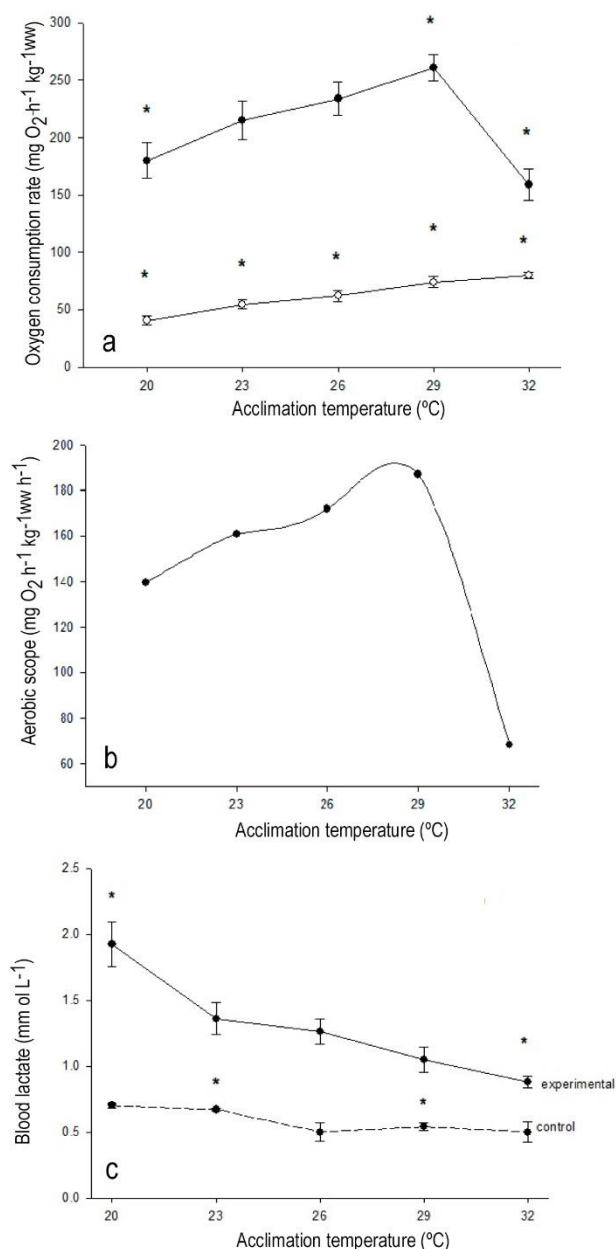


Figure 2. a) Maximum metabolic rate (MMR) and standard metabolic rate (SMR) of fish exposed to chase, b) aerobic scope (AS) calculated as $MMR - SMR$, black dots represent the mean \pm standard error, c) blood lactate of fish exposed to chase, the continuous line is experimental, and the dashed line represents control values.

Florida red tilapia reached a maximum growth at 27°C, which is very close to its FP. The results obtained in this study are important to improve Pargo-UNAM culture, so it can be developed successfully in regions where the water temperature is near 29.5°C.

Given the above, it can be assumed that in the FP organisms will optimize their physiological process in

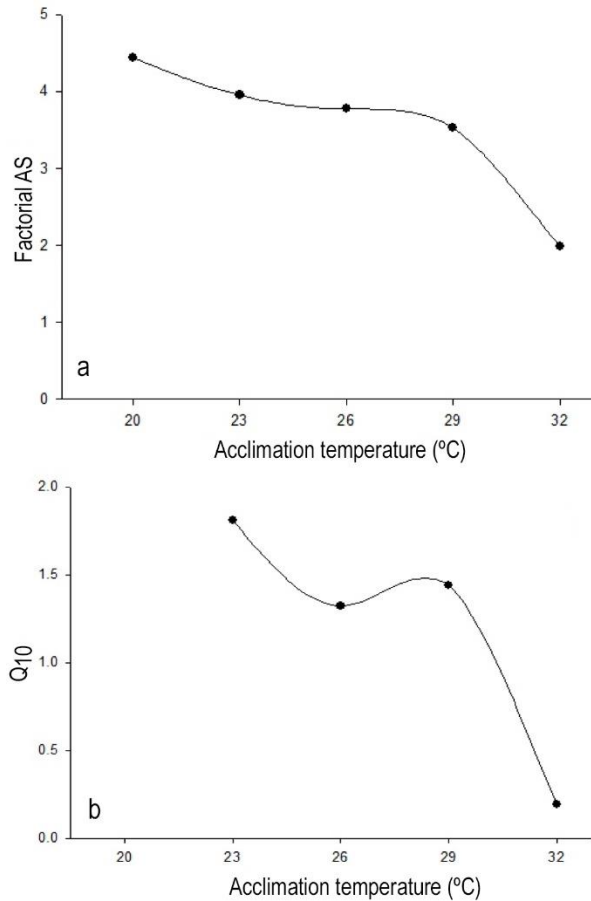


Figure 3. a) Factorial AS obtained from MMR/SMR from the chase, b) metabolic cost as Q_{10} obtained from the MMR. Black dots represent the mean values.

such manner that energy expenditure is reduced, which may translate in energy savings aimed directly to potential growth.

Therefore, the FP can be used as a temperature measurement, selected by PU and as an index of the magnitude of temperatures to which the species is adapted (Johnson & Kelsch, 1998). This result demonstrates that PU was able to choose within the horizontal thermal gradient the most advantageous temperatures available on the virtual chambers, like those conditions in its environment. We conclude that PU was able to regulate its body temperature by using behavioral thermoregulation.

According to Pörtner (2010) and Sokolova *et al.* (2012), CTMax and CTmin represent the upper and lower limits corresponding to the critical thermal threshold. PU juveniles reached the CTMax at 43.5°C. Thermal thresholds obtained from the thermal window are found in the geographical zones where tilapia is grown in Mexico. These areas are affected by temperatures that have increased or decreased, origi-

nated by climate changes that have occurred in recent years (Noyola *et al.*, 2015). That is why is crucial to learn the thermotolerance of aquatic species on a planet that is changing due to global warming. In the present study, we propose that values reported of CTMax in PU correspond to the critical threshold temperature at which the scope of metabolic activity is zero. The above explains why organisms in this threshold temperature can stay alive for a short period, where animals enter into physiological repair (Pörtner, 2010), and demonstrates that intervals between 26 to 29°C correspond to an acclimation temperature where the maximum performance is observed (Pörtner, 2010).

The CTmin reached by PU juveniles had an 8-14°C interval, as compared to the 2.5-11-3°C range of *Cyprinodon variegatus* (Bennet & Beitinger, 1997) and to the interval obtained by Currie *et al.* (1998) for *Ictalurus punctatus* (2.7-9.8°C) and *M. salmoides salmoides* (3.2-10.7°C); these ranges indicate that when comparing PU to these species, PU is more susceptible to cold water; except for *Argyrosomus regius* (11.7-13.24°C) (Kir *et al.*, 2017) which is intolerant to cold. Concerning other fish, tilapia is more susceptible to low temperatures; therefore, its growth is reduced.

Acclimation response ratio is usually considered a reliable measurement to indicate the physiological response of fish to a given temperature change (Claussen, 1977; Chatterjee *et al.*, 2004), although it depends on previously experienced temperatures (Díaz *et al.*, 1998). ARR can be interpreted as the heat-tolerance degrees gained for each centigrade grade increased in acclimation temperature (He *et al.*, 2014; Kir *et al.*, 2017). Aquatic organisms inhabiting in cold or temperate regions show lower ARR values because they experience longer and more gradual temperature fluctuations; as a result, they have a reduced tolerance to abrupt temperature changes (Díaz *et al.*, 1998, 2004). Cold-water species such as *Schizothorax kozlovi* (He *et al.*, 2014), *Oncorhynchus mykiss* (Brett, 1952), and *Salvelinus fontinalis* (Currie *et al.*, 1998) have shown ARR values of 0.21, 0.18, 0.14 and 0.1-0.30, respectively. In contrast, high ARR values such as PU values are characteristic of organisms experiencing wide temperature fluctuations in a short period, since they have no time to use adjustment mechanisms. For this reason, PU can be characterized as a subtropical fish because its ARR values are in the 0.34-0.67 range. Many commercially important fish are geographically located in subtropical and tropical zones, such as *Tilapia* spp., *Ctenopharyngodon*, *Cyprinus*, and *Ictalurus* and share similar ARR values, but the most important characteristic is that they are eurythermic; allowing them to tolerate changes and deal with global warming (Gunderson & Stillman, 2017).

The thermal window provides important insights regarding fish ecology, distribution, and survival tactics (Bennet & Beiting, 1997) and determines optimal culture conditions (Noyola *et al.*, 2013), providing a comparative index of eurythermicity among species (Eme & Bennet, 2009). The usefulness of thermal windows (reported as °C²) relies on its ability to offer a visual comparison among species, instead of only a thermal preference point. Also, thermal windows define intrinsic thermal tolerance zones, *i.e.*, tolerance-independent from previous thermal acclimation history. A complete thermal window for PU juveniles with different zones such as the optimal, transition (Pejus) and critical limits threshold was included. According to this, the best performance of PU juveniles occurred in the optimal zone with a 63.4°C² area. The total thermal window area of PU juveniles (355.2°C²) is higher than *Labeo rohita*, (273.5°C²), *Anabas testudineus* (278.3°C²), *Cyprinus carpio* (311.6°C²), *Lutjanus guttatus* (344.25°C²), *Ocyurus chrysurus* (282.0°C²) (Chatterjee *et al.*, 2004; Larios, 2014; Noyola *et al.*, 2015) and lower than *Horabagrus brachisoma* (526°C²), and *Argyrosomus regius* (460°C²) (Dalvi *et al.*, 2009).

Due to the last characteristic, PU could be cultivated in many areas of Mexico where the minimum and maximum temperatures are between the 20–32°C range (INEGI, 2015). The relationship between the preferred temperature and metabolic optimal of fish is linked with the concept of aerobic scope (Fry, 1947). The maximum aerobic scope for activity generally occurs at the preferred temperature. The highest aerobic scope obtained for PU was 29°C, where this temperature corresponds to the preferred temperature. Thus, the highest amount of available energy could be channeled to adaptive functions in PU juveniles, such as activity, growth, reproduction, and survival, in a similar manner as other species (Beamish, 1981; Jobling, 1981; Kelsch & Neill, 1990; Kelsch, 1996; Alsop *et al.*, 1999; Lee *et al.*, 2003).

In PU, AS values are like the aerobic scope of *Ocyurus chrysurus*, a tropical fish with similar thermal tolerance. Chase increased swimming activity, which led the organism to increase its muscular energy demand and, as a result, a rise in oxygen consumption. Factorial aerobic scope shows that PU and *O. chrysurus* could be cataloged as semi-sedentary fish (Noyola *et al.*, 2015)

Lactate and amino acids are the preferred substrates for gluconeogenesis when there is an oxygen deficit (Moon & Foster, 1995; Suarez & Mommsen, 1997). It has been demonstrated that the lactate concentration in the blood increases after some stress; this is due to muscle glycolysis (Wood *et al.*, 1985). Blood lactate values obtained from fish exposed to chase were very

low. Thus, they used the aerobic metabolism even at temperatures from 20 to up to 29°C. Blood lactate values found were in the 1.2–1.8 range; this could be associated with constant values in blood, liver, and muscle (Frederich & Pörtner, 2000, 2001; Sokolova & Pörtner, 2002).

Under prolonged stress conditions such as a 20°C temperature, in which an organism spends more energy than the one available on its active metabolism, PU is forced to activate anaerobic pathways (Priede, 1977). When *Ocyurus chrysurus* was exposed to chase, it had lower blood lactate in the 20–26°C interval temperature and a higher lower blood lactate when they were exposed to 30 and 32°C, indicating that ATs affected its metabolism, probably because they were close to their upper threshold limit (Noyola *et al.*, 2015).

Knowledge regarding the thermoregulatory behavior of PU provides important information for aquaculture and in selecting the places where the temperature is optimal for its growth. PU juveniles showed a high degree of eurythermicity and a positive response to thermal changes; and this leads us to conclude that PU will be grown successfully in a 26–32°C temperature range, having its greater performance at 29°C, where the aerobic scope was at its maximum performance.

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