



Temperature Of Growth And Biochemical Reaction In Freshwater Fish Tissue: Applications To Indoor And Outdoor Culture Systems (Labeo Rohita)

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Abstract :

Temperature has a major impact on freshwater fish raised for growth and metabolism's physiological characteristics. Ten fish were used, kept in two tank aquaculture systems, and exposed to different temperatures, for example, in order to compare the temperatures inside and outside simultaneously. The impact of four weeks of aquarium-raised *Labeo rohita* fingerlings on their growth response to different indoor and outdoor conditions. The growth rates varied greatly amongst the indoor treatment groups, but they first grew and then declined in the 28°C to 29°C temperature range. Within the 29–32°C range, this straight forward growth model yielded a dependable growth (SGR%) estimate with unit rise of outdoor temperature. The purpose of this work is to estimate the amounts of lipids, proteins, and carbohydrates in these two kinds of temperature-variation culture systems. Muscle (104.73 ± 0.00089) and intestinal (103 ± 0.00081) had a high carbohydrate content, in contrast to the low amounts reported in indoor fish muscle (100.33 ± 0.00047) and gut (106 ± 0.0008). Fish raised outdoors have a high protein content in their muscles (205 ± 0.216) and guts (176.36 ± 0.262). The external muscles and intestines of fish exhibit significant lipid contents (74.54 ± 0.270 and 30.36 ± 0.207 , respectively). fish with low intestine (24.38 ± 0.192) and muscular (62.40 ± 0.274) levels indoors. The results demonstrated that the outdoor culture system had a higher concentration of protein, lipids, and carbohydrate than the other fish species that were assessed for temperature.

Aquaculture ,Climate change , Water temperature , *Labeo rohita*,

Biochemical Analysis.

The past 20 years have seen the aquaculture sector emerge as a key player in the food production landscape, providing a significant portion of the animal protein needed by all people, independent of lifestyle globally, and its capacity to feed the expanding global population may be estimated by looking at its yearly growth rate of more than 7% (Jayasankar, 2018). Aquaculture has been producing fish for a long time; now, 82.1 million tons (46%) of the 179 million tons of fish produced globally come from this source. Furthermore, it is anticipated that by 2030, aquaculture output would account for 53% of all fish produced worldwide, surpassing 46% of fish production worldwide (FAO 2020). According to Collins et al. 2020 and Khalid 2022, Rising temperatures have the potential to accelerate the emergence of exotic diseases and increase the

susceptibility of domesticated animals to heat-related ailments. While fish diseases and water temperature can be directly impacted by one another, multivariate environmental change may have unpredictable effects on both. The combined effects of these three interrelated factors may lead to variations in the frequency and intensity of the condition, as well as its growth and decline over time, according to (Chiaramonte et al.2016). Temperature,precipitation,photoperiod duration, and other climatic factors are among the many that physically affect water bodies. Freshwater bodies' primary production is influenced by temperature and other physical conditions. Optimal fish production requires an understanding of the local environmental factors and how management tactics may be combined with them to enhance fish yield.T13

One of the primary environmental factors influencing fish development and metabolism is temperature. Aquaculture is concerned about the rising water temperatures brought on by global warming.Fish populations may decrease as a result of physiological processes disrupted by high temperatures, and certain species may even become extinct (Ashawaf-Ud-Doulah et al., 2019).Temperature of the aquatic environment has been found to affect fish survival, distribution, reproduction, and proper metabolism (Shahjahan et al., 2013; 2017). Fish are cold-blooded animals, therefore the temperature of the water they live in affects their body temperature, growth rate, ability to consume food, ability to convert feed, and other bodily processes (Azevedo et al., 1998; Britz et al., 1997). It feeds on organic stuff that has decomposed and vegetable waste, making it a bottom feeder. Physiological research can be used to anticipate how temperature changes would affect different fish species (Somero, 2010). As fish are aquatic poikilothermic animals, increased water temperatures generate stress and change blood chemistry standards, which impact almost all biochemical and physiological activities. High temperatures, when they above the threshold of tolerance, accelerate the chemical reactions in fish bodies and have a significant impact on physiological processes, according to Chatterjee et al. (2004).

High-quality protein and other organic compounds may be found in abundance in fish. They are the primary source of animal protein and are generally regarded as a nutritious food item that also contains other nutrients. Andrew.A.E(2001) .Due to its low cholesterol, great palatability, and soft meat, a significant portion of the population consumes fish, which is high in protein. It is the most affordable way to obtain animal protein and other vital elements that are needed in a human diet, especially for those with poor and moderate incomes. The sort of food an animal eats has a significant impact on the type and quality of nutrients it contains. Furthermore, the nutritional makeup of a fish's flesh is significantly influenced by its eating habits. Fish flesh contains all of the necessary amino acids for diet and around 85–90% of the digestible protein. A food's proximate composition, such as its protein and fat level, must frequently be measured to make sure it complies with regulations. b21

Fish nutritional value has been the primary focus of biochemical component analysis in India. Due to its comparatively high digestion, fish protein is regarded to have a high biological and growth-promoting effect (Shekhar et al., 2004).It also contains all 10 necessary amino acids in optimal quantities for human intake (Bhilave et al., 2013).Understanding the biochemical makeup of various species, including fish, is very beneficial for determining their nutritional worth. It also aids in assessing the quality of these natural resources and maximizing their use (Rodriguez-Gonzalez et al., 2006). Fisheries biochemical studies are useful in assessing environmental effects. Due to their specificity about the dietary values of fish and their ability to assess the physiological requirements of fish at various stages of life, biochemical investigations of fish tissues are of great interest. The manufacturing of dried fish, canning, and fish meal preparation are among the major fish processing sectors that benefit from an understanding of the nutritional composition of freshwater fish.Species-specific responses to heat acclimation in tropical freshwater fishes, however, are comparatively little understood, particularly with regard to stress responses and the use and mobilization of energy stores. This viewpoint led to the current study's attempt to ascertain the effects of various acclimation

temperatures (26, 31, 33, and 36° C) on biochemical variables, such as protein, carbohydrates in the gut and muscle, and total lipid in *L. rohita* tissue.

2. Materials and Method

2.1 Experimental of fish collection:

Fresh water fish *Labeo rohita* weighing (5.15.-5.25)g were collected from the suriya fish farm, Kallidaikuruchi, Tirunelveli. The fish were thereafter brought to the lab in polythene bags with low-temperature, oxygenated water that was disturbed as little as possible. Then they were acclimatized to the ambient laboratory room temperature range (27 to 29°C) in FRD tank. During the period of acclimatization, which lasted for two weeks.

2.2 Experimental setup:

The trial was carried out in nine thermostatic aquariums at various temperature levels, including Indoor and Outdoor culture system. The fish were kept in controlled laboratory tanks, each of which had filtration and aeration systems to keep the water clean. We acquired *L. rohita* fingerlings from nearby fish farms. For a duration of two weeks, the fish were housed in a flowing water fibre tank at a room temperature range of (28.13-29.50) and a stocking density and outdoor fluctuation temperature range of (29.63-32.25). The pH range of the water was (7.-8.6) alkalinity (110 -130 mg l-1), DO (5-6 mg l-1), ammonia (0.25-0.55) with 24 hours.

2.3 Experimental diet In the second experiment, nine (n = 10) identically sized *L. rohu* fingerlings were divided evenly among 500 L of (FRD) water tanks that had sufficient aeration. The meal was provided in the morning (07.00 a.m.) and at nightfall (06.00 p.m.), with 16.22% protein, 58.87% carbohydrate, 3.78% fat, 10.62% ash, 2% crude fiber, and 8.45% moisture content. For four weeks, the experimental fish were exposed to two distinct temperature regimes: inside temperature and outdoor temperature.

Table 1 Water quality parameters (Mean ± SD) during the 30 days periods

Water Parameter	Indoor culture	Outdoor culture
Temperature	(28.70±0.32)	
Dissolved oxygen (mg/L)	4.95 ± 0.16	5.33 ± 0.16
pH	7.35 ± 0.09	7.00 ± 0.11
Total alkalinity (mg/L)	118.0 ± 6.2	132.0 ± 7.3
Ammonia	0.35 ± 0.004	0.52 ± 0.00

Table 2 Growth Responses of in Two variations temperature treatments for 30 days

Growth parameter	Indoor temperature	Outdoor temperature
Initial BW (g)	5.17±0.07	5.25±0.01
Final BW (g)	6.19±0.09	7.24±0.02
Weight gain (g)	1.01±0.09	1.99±0.03
% weight gain	19.57±1.43	37.90±0.84
SGR (% / day)	0.64±0.05	1.14±0.02
FCR	1.15±0.02	1.44±0.01
Survival (%)	90± 0.00	90± 0.00

Salinities vary considerably ($p < 0.05$) across values in a row with various alphabetical superscripts. Every value is given as mean \pm SD.

The following formulas were used to get the feed intake (FI), feed conversion ratio (FCR), weight gain (WG), and specific growth rate (SGR).

$$\text{SGR}(\%) = \frac{(\text{Final Body Weight} - \text{Initial Body Weight}) \times 100}{\text{Expressent Duration}}$$
 The formula for WG (%) is $[(\text{Final body weight} - \text{Initial body weight}) / \text{Initial body weight}]$.
 FCR is equal to the wet weight growth of each fish and the dry weight of feed that each fish ingested throughout the trial.

Estimation of Total carbohydrate

The technique developed by Carroll et al. (1956) was used to assess the total carbohydrate content. After adding 10% Trichloroacetic acid (TCA) to the test sample, the mixture was centrifuged for 15 minutes at 3000 rpm. A 15-minute boil was achieved by adding 5 milliliters of anthrone reagent to 0.5 milliliters of supernatant. Following cooling of the tubes, the absorbance was measured at 620 nm using a spectrophotometer and a blank solution consisting of the same ratio of 10% TCA and anthrone. The measured quantities were given in units of glucose per milliliter.

Estimation of Total Protein

The approach of Lowry et al. (1951) was used to determine the total protein content. 1 ml of 1 N NaOH was used to dissolve 1 ml of the sample. This resulted in the addition of 5 ml of alkaline copper solution (50 ml of 2% Na₂CO₃ and 1 ml of 0.5% CuSO₄. 5H₂O in 1% sodium potassium tartrate) to 0.2 ml of the extract. After thoroughly mixing, the mixture was let to stand for ten minutes. This was mixed with 0.5 ml of a 50% Folin-Ciocalteau reagent that had been diluted 1:1 with distilled water. In a spectrophotometer, the optical density was measured at 620 nm after 30 minutes in comparison to a blank. The Lowry et al. (1951) technique was used to plot the standard graph using bovine serum albumin (Sigma Chemical Company, U.S.A.). The measurements were given in $\mu\text{g/ml}$. Three copies of each sample were collected.

Estimation of Total Lipids

The Barnes et al. (1976) approach was utilized to estimate lipids. 50 mg of tissue was homogenized in a 2:1 chloroform:methanol mixture using 10 ml of water in a mixing blender. Whatmann No. 1 filter paper was used to filter the homogenates after the residue had been previously homogenized and filtered. 0.88% KCl, which was added as one-fourth of the volume, was shaken vigorously to eliminate the non-lipid materials from the pooled filtrate. In a test tube, 1 ml of the filtrate was extracted, evaporated under nitrogen, and then 1 ml of concentrated H₂SO₄ was added, boiling for 10 minutes. Two milliliters of the vanillin reagent were added to 0.2 milliliters of the solution to estimate the total amount of lipid. The produced color was measured at 520 nm using a spectrophotometer against a blank for the reagent. Using the aforesaid approach and

powdered cholesterol, the standard graph was drawn. The results were given in units of $\mu\text{g/ml}$ moist weight of the tissue.

Table 3

Biochemical Analysis	Organs	Indoor Temperature	Outdoor Temperature
Total Carbohydrate($\mu\text{g /ml}$)	Muscle	104.73 \pm 0.0089	100.33 \pm 0.00047
	Intestine	103. \pm 0.00081	106 \pm 0.008
Protein($\mu\text{g /ml}$)	Muscle	150.23 \pm 0.169	205 \pm 0.205
	Intestine	170.3 \pm 0.216	176.366 \pm 0.262
Total Lipids($\mu\text{g /ml}$)	Muscle	62.40 \pm 0.274	74.54 \pm 0.270
	Intestine	24.38 \pm 0.192	30.36 \pm 0.207

The means differ by a considerable amount ($p < 0.5$), with the data shown as Mean \pm SD.

Result and Discussion

Water parameter:

During the research period, the pH varied seasonally between 6.8 (rainy) and 7.8 (spring), and the water temperature ranged from a low of 20.5°C in winter to a high of 30.5°C in summer. A temperature range of 28–32°C in tropical seas is favorable for fish development, according to Jinghran (1968). The content of dissolved oxygen ranged from 5. to 5. mg/l . Both indoor and outdoor temperatures were found to have the greatest value of the dissolved oxygen content. The dissolved oxygen value that was recorded was within the boundaries that Boyd (1979) and Almabasta et al. (1980) had established for excellent water quality in fish production. The pH data that was observed fell within the range that Swingle (1961) and Boyd et al. (1985) had identified as being most appropriate for fish production to achieve optimal productivity. There are some grounds for thinking that, under some situations, ammonium ions may play a major role in ammonia toxicity. Rising water temperatures led to a substantial ($P < 0.05$) rise in pH and a drop in dissolved oxygen content, according to studies by Das et al. (2005) and Brahmane et al. (2014). While the pH and total alkalinity of the water remained rather constant, there was a drop in dissolved oxygen in the outdoor culture ($P < 0.05$) when the inside temperature increased (Shahjahan et al., 2018; Islam et al., 2019).

Growth characteristics of *L. rohita* at various temperature variations:

(Table 2) *L. rohita*'s growth performance at two distinct temperature regimes. In comparison to fish acclimated at (control) indoor temperature, fish acclimatized in the outdoor temperature range (29.63-32.25) had substantially higher weight gain (WG), specific growth rate (SGR), and FCR over the 30-day study period. When compared to fish kept in a room temperature range (29.63-32.25), an increase in WG, SGR, and

rise was noted in fish raised outdoors in the 29.63-32.25) temperature range. Different temperature settings had no effect on the fish's survival rate. The study's findings aligned with those of Das et al. (2005) and Ashaf-Ud-Douhah et al. (2020). The results of this study showed that growth performance (weight gain, SGR, and FCR) was much greater at indoor and outdoor temperature acclimation than it was at more fluctuating temperatures. The observed reduced growth performance of *L. rohita* at outdoor temperatures may have resulted from the organism's heightened hunger at higher temperatures, as evidenced by greater feed conversion rates (FCR) during the course of the 30-day feeding period.

Biochemical Analysis:

Analyses protein, carbohydrate, and lipid contents of the fish were conducted at two distinct temperature ranges (Table 3). 150.23±0.169 and 170.3±0.216, respectively, were the measurements made of the total protein content in the muscles (205±0.205) and the intestines (176.366±0.262). The overall amount of carbohydrates in the stomach (106±0.008), outdoor muscle (100.33±0.00047), and indoor muscle (104.73±0.0089), respectively. The total lipid content of the intestine (30.36±0.207), outdoor muscle (74.54±0.270), and indoor muscle (62.40±0.274) differs from the total lipid content of these tissues. The results demonstrated that the two stress variation fishes had significantly different amounts of protein, fat, and cars in the ambient temperature. Stress may be applied to the modifications in carbohydrate metabolism that would be necessary to fulfill the shifting energy requirements (Lacerda et al. 1986), (Santos et al. 1987) In most vertebrates, including fish and humans, the normal metabolic rate and blood glucose level match. (Umminger et al. 1987). The severe stress on the metabolic process and the impairment of the internal organs' ability to synthesis proteins might be the cause of the loss in muscle protein. (Jha et al. 2002). The function of biological components including lipids, proteins, and carbohydrates varies with temperature, and these changes are crucial in indicating how susceptible organ systems are to temperature fluctuations.

Conclusion:

The findings of the study showed that water temperature significantly impacted both water quality indicators and the growth performance of *Labeo rohita*. In *Labeo rohita*, we investigated the impacts of temperature, biochemical analysis, and fish tissue efficiency. The findings suggest that there are differences in the ideal water temperature for rohu development between indoor and outdoor settings.

Because of its minerals and biological components, fish is a diet high in nutrients. This study found that when all variables are taken into account, temperature stresses Indian main carp because it contains more amounts of fat, protein, and carbohydrate than when development is under stress.

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