Biochemical Study of Freshwater Fish Labeo rohita Infected with Helminthes Parasites from Nashik District, Maharashtra

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***Abstract*—Freshwater ecosystems include numerous aquatic resources, such as rivers and dams, which support diverse fish fauna and contribute significantly to inland fisheries. Among the commercially important species, Labeo rohita is widely distributed and is an important component of freshwater aquaculture. It is highly demanded and economically important fish in the aquaculture industries. As the Parasitic infections, particularly helminthes can influence the stress to the physiological and metabolic activities of the fish. Present study was to consider to evaluate the seasonal variations in selected biochemical parameters, such as glycogen, protein, and lipids, in helminthes-infected and non-infected L. rohita collected from freshwater bodies in the Nashik district of Maharashtra (India). Fish samples were collected and analyzed during three seasonal variation changes in water quality in summer, monsoon and winter of year 2023-2024 simultaneously. The results showed a consistent decline of nutrients, such as glycogen, protein, and lipids, in infected fish with helminthes parasites compared to non-infected fish.**

Keywords: Nutrients, Parasites, Infection, Metabolic activity, Aquaculture.

I. INTRODUCTION

Aquaculture refers to the scientific cultivation and management of aquatic organisms, such as fish, crustaceans, mollusks, and aquatic plants, under controlled or semi-controlled environmental conditions. It has emerged as one of the fastest-growing food production sectors worldwide and plays a vital role in ensuring food security, generating employment, and supporting economic development, particularly in developing countries such as India. With the rapid increase in the global population, the demand for high-quality protein sources has risen significantly. The freshwater fish serve as an important and affordable source of animal protein because of their high nutritional value, digestibility, and availability. Freshwater fish, particularly Indian major carps such as Labeo rohita (Hamilton, 1822), are widely cultured because of their fast growth rate, adaptability to diverse environmental conditions, and high consumer preference. The increasing demand for freshwater fish is mainly attributed to their rich composition of proteins, essential fatty acids, vitamins, and minerals, leading to the rapid expansion of aquaculture practices. Nashik district in Maharashtra is endowed with abundant freshwater resources, such as rivers, reservoirs, dams, and irrigation canals, which provide suitable habitats for fish production. These water bodies support diverse fish fauna and contribute significantly to regional fisheries. However, increasing anthropogenic activities, such as industrial discharge, agricultural runoff, and urbanization, have resulted in deterioration of water quality. Water pollution has emerged as a major threat to aquatic ecosystems, increasing the susceptibility of fish to various diseases, including parasitic infections. Among the disease-causing agents, helminth parasites, such as cestodes, trematodes, and nematodes, commonly infect freshwater fish. These parasites directly obtain nutrients from host tissues, thereby disturbing normal metabolic processes and physiological functions. Consequently, infected fish often exhibit reduced growth, stress, and compromised health. Biochemical constituents, such as glycogen, protein, and lipids, serve as key indicators of metabolic status in fish. Alterations in these constituents reflect disturbances in energy utilization and physiological balance. Parasitic infections are known to influence these biochemical parameters by increasing metabolic demand and diverting host nutrients toward parasite survival, ultimately reducing the nutritional quality and economic value of fish. Therefore, the present study aimed to evaluate the impact of helminth parasitic infection on the biochemical composition of glycogen, protein, and lipids in L. rohita from freshwater bodies in the Nashik district, along with seasonal variations in these parameters.

II.REVIEW OF LITERATURE

Freshwater fish are considered an important component of human nutrition because of their high protein content, essential fatty acids, vitamins, and minerals. Globally, fish contribute significantly to food security and provide an inexpensive source of animal protein for the growing population. The biochemical composition of fish tissues mainly includes proteins, lipids, carbohydrates, and mineral elements, which together determine the nutritional quality and physiological status of the organism (Jabeen & Chaudhry, 2016). Among freshwater fishes, Labeo rohita (Hamilton, 1822), commonly known as Rohu, is one of the most important major Indian carp species widely cultured in South Asian countries, including India, Bangladesh, and Pakistan. This species has high commercial value owing to its rapid growth rate, palatable flesh, and rich nutritional composition. Fish parasites represent a major threat to aquaculture and natural fisheries. Among them, helminth parasites, such as cestodes, trematodes, and nematodes, frequently infect the digestive tract and internal organs of fish. These parasites obtain nutrients directly from host tissues and intestinal contents, thereby altering the metabolic processes and biochemical composition of the host organism. Biochemical parameters, such as glycogen, protein, and lipids, serve as important indicators of metabolic health in fish. Glycogen acts as a primary energy reserve and is mainly stored in the liver and muscle tissues. Proteins are fundamental structural components involved in tissue repair and metabolic regulation. Lipids function as major energy sources and play an essential role in cellular membrane structure and hormonal regulation (Verma & Agarwal, 2005). Previous studies have demonstrated that parasitic infections significantly reduce the levels of biochemical constituents, such as glycogen, protein, and lipids, due to increased metabolic demand and nutrient utilization by parasites (Saraf & Katyayani, 2020; Jawale, 2023).

III.MATERIAL AND METHODOLOGY

The present investigation was conducted in freshwater bodies located in the Nashik district, Maharashtra, India. The Nashik district is characterized by several reservoirs, rivers, and irrigation canals, which provide favorable habitats for freshwater fish, including L. rohita. Fish samples were collected periodically from local fishing sites and fish markets during the study period from February 2023 to January 2024. Specimens of L. rohita were collected from local markets. The collected fish were transported to the laboratory. Each specimen was carefully examined for the presence of helminthes parasites by dissecting the gastrointestinal tract and internal organs. Helminthes parasites from infected fish were isolated, washed in physiological saline, and preserved in 70% ethanol. Identification was performed based on morphological characteristics using standard taxonomic keys. Glycogen content was estimated using the anthrone method (Kemp et al., 1954), protein content was estimated using the Lowry method (Lowry et al., 1951), and lipid content was estimated using the Folch extraction method (Folch et al., 1957). Biochemical constituents were estimated from fish muscle tissues using standard biochemical techniques. All biochemical estimations were performed in triplicate to ensure accuracy and reproducibility of the results. All measurements were carried out in triplicate ($n = 3$) and expressed as mean ± standard deviation

IV.RESULTS AND DISCUSSION

Glycogen, protein, and lipids are essential biochemical components that help fish maintain normal body functions and overall health. Glycogen acts as a quick source of energy, especially when the fish is under stress or requires immediate metabolic activity. Proteins are crucial Body growth, repair of tissues, and the execution of various biological processes through enzymes and hormones. Lipids, in contrast, serve as long-term energy reserves and play an important role in maintaining cell structure and supporting vital physiological functions. Together, these components provide a clear indication of the nutritional condition and metabolic balance of fish.

Table 1: Glycogen Content in non-infected and infected Labeo rohita

Seasonal period	Glycogen (mg/gm)			Source	Glycogen SS	Glycogen F	Glycogen p
	Non-Infected	Infected	Reduction (%)	Season (Factor A)	102.412	31.021	<0.0001
Feb – May (Summer)	14.84 ± 1.18	9.95 ± 0.80	32.98% (**)	Infection (Factor B)	154.038	93.319	<0.0001
June – Sept (Monsoon)	18.33 ± 1.88	12.86 ± 1.45	29.83% (*)	Season × Infection	4.267	1.293	0.3102
Oct – Jan (Winter)	21.82 ± 1.42	14.64 ± 0.48	32.94% (**)	Error (Within)	19.808	-	-

Table 2: Protein content in non-infected and infected Labeo rohita

Seasonal period	Protein (mg/gm)			Source	Protein SS	Protein F	Protein p
	Non-Infected	Infected	Reduction (%)	Season (Factor A)	2365.717	16.605	0.0003
Feb – May (Summer)	120.82 ± 7.48	83.97 ± 5.20	30.50% (**)	Infection (Factor B)	4653.902	65.333	<0.0001
June – Sept (Monsoon)	145.54 ± 6.68	115.40 ± 3.27	20.71% (**)	Season × Infection	49.964	0.351	0.7112
Oct – Jan (Winter)	131.66 ± 15.83	102.18 ± 6.20	22.39% (*)	Error (Within)	854.800	-	-

Table 3: lipid content in non-infected and infected Labeo rohita

Seasonal period	Lipid (%)			Source	Lipid SS	Lipid F	Lipid p
	Non-Infected	Infected	Reduction (%)	Season (Factor A)	1.853	1.748	0.2157
Feb – May (Summer)	12.96 ± 0.66	8.23 ± 0.64	36.45% (***)	Infection (Factor B)	72.000	135.786	<0.0001
June – Sep (Monsoon)	11.79 ± 0.85	8.02 ± 0.59	31.95% (**)	Season × Infection	1.223	1.153	0.3484
Oct – Jan (Winter)	12.33 ± 1.01	8.82 ± 0.49	28.47% (**)	Error (Within)	6.363	-	-

[Values: Mean ± SD (min – max). n = 3 specimens per group. Significance of pairwise t-tests is shown in the reduction column. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$, ns = not significant. Two-way ANOVA factors: Season (three levels) × infection status (2 levels). Sum of Squares (SS), F-value (or F-statistic), p-value (Probability)]

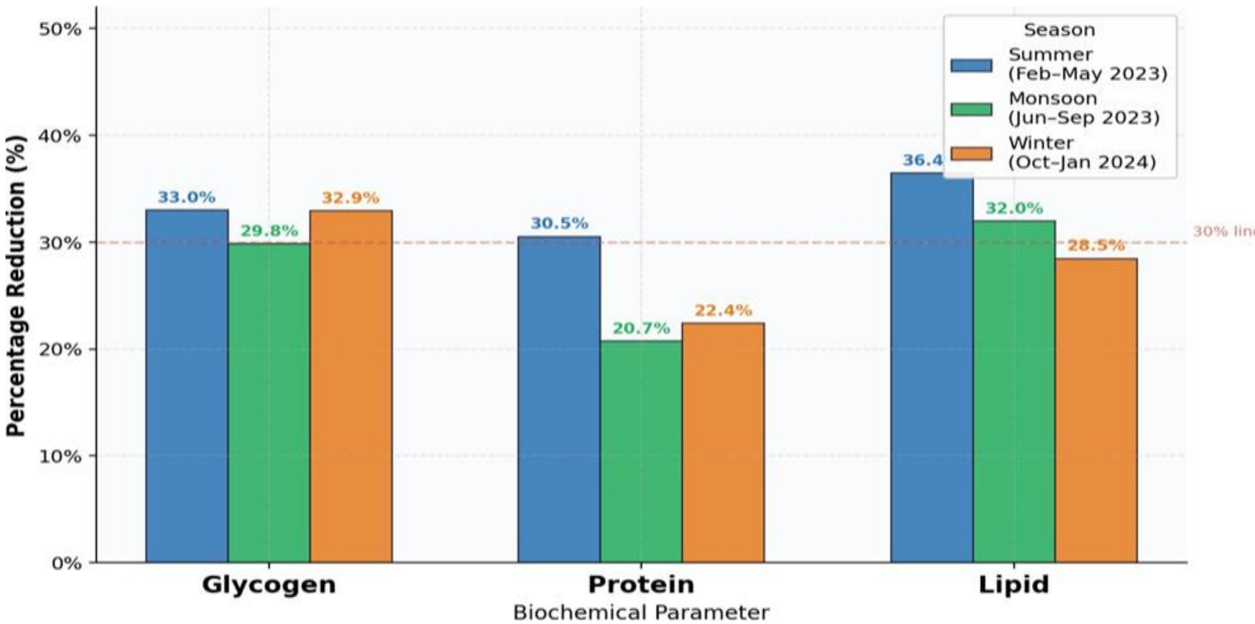

Figure 1 : Reduction (%) in Biochemical Parameters due to helminth infection

Biochemical analysis of L. rohita revealed a consistent and statistically significant depletion of glycogen, protein, and lipid reserves in helminth-infected fish compared to their healthy counterparts across all three seasons. While the baseline protein and glycogen levels of uninfected fish naturally fluctuated with changing seasons (ANOVA Season $p < 0.001$), the baseline lipid levels remained relatively stable year-round (ANOVA Season $p = 0.2157$).

Despite these natural seasonal baselines, the onset of parasitic infection triggered severe nutrient reductions regardless of the time of year. Protein concentrations declined by 20.71% to 30.50% ($p < 0.05$ to $p < 0.01$), and glycogen reserves dropped by 29.83% to 32.98% ($p < 0.05$ to $p < 0.01$). Lipids endured the most drastic degradation, decreasing by 28.47% to 36.45% ($p < 0.01$ to $p < 0.001$). A two-way analysis of variance (ANOVA) confirmed that infection status acted as a highly significant, independent driver of macronutrient loss ($p < 0.0001$). Furthermore, the statistical models revealed no significant interaction between season and infection status ($p > 0.05$ for all parameters), proving that the severity of parasitic nutrient extraction remained consistently uniform throughout the entire year. Present study demonstrates that helminthes infections induce profound metabolic stress in L. rohita, fundamentally disrupting the host's physiological homeostasis. Glycogen, the primary immediate energy source, is rapidly mobilized to meet the heightened energetic demands imposed by the parasite. This rapid utilization of carbohydrate reserves aligns with the findings of (Carroll et al.,1956) and (Jawale.,2023), who noted similar glycogen depletion driven by host energy requirements and direct nutrient diversion toward parasite metabolism. Similarly, a significant reduction in host protein content can be attributed to heightened proteolysis, in which amino acids are broken down for compensatory energy production. This is compounded by the parasites directly absorbing nutrients from the host's gastrointestinal tissues, a mechanism also observed in other parasitized freshwater fish (Roberts., 2012).

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