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Research Article

Comparative Study on the Digestive Enzyme Activities, Hematology and Histopathology in Wild and Farmed *Labeo rohita*

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Abstract

This study compared the amylase and lipase activity, hematology, and histopathology of wild and farmed *Labeo rohita*. A total of 18 fish (9 wild and 9 farmed) in triplicate were used in the present study. Blood samples were taken, and organs were dissected for analysis. The results showed that wild fish had significantly higher amylase activity in the liver (8.00±0.27 U/L) compared to farmed fish (1.00±0.65 U/L), while farmed fish had higher amylase activity in the intestine (13.60±0.44 U/L) than wild fish (9.30±0.37 U/L). Lipase activity in both the liver and the intestine was similar between the two groups. Hematologically, farmed fish had higher levels of hemoglobin (8.90 g/dL), hematocrit (26.7%), and plasma protein (2.86 g/dL), while wild fish showed elevated WBC counts ($11.10\times10^3/\mu L$) and MCHC (41.20 g/dL). Histopathological examination revealed that farmed fish had healthy hearts, kidneys, and gill structures, while wild fish showed elongated cardiac vessels, hydropic degeneration in the kidneys, and gill damage, including epithelial rupture. Overall, farmed *Labeo rohita* appeared to be healthier than its wild counterparts. The findings of this study have significant future implications for improving aquaculture practices, as they can inform the development of optimized diets, enhance fish health management strategies, and guide selective breeding programs to bridge the physiological gap between wild and farmed populations.

Keywords: Labeo rohita, Enzymes, Hematology, Histopathology, Wild, Farmed.

1. Introduction

Aquaculture, which involves farming fish and other aquatic organisms, has become a vital industry in meeting the increasing global demand for fish, a crucial source of animal protein. As the world's population continues to grow and eating habits evolve, the need for fish as a fundamental part of human nutrition has reached unprecedented levels. In many developing countries, fish represents a major part of daily protein consumption, supplying around 26% of the necessary animal protein for millions. With wild fish populations under threat from overfishing [1] and dwindling catches, aquaculture presents a sustainable way to satisfy this rising demand.

There is a notable difference in nutritional value, growth, and overall health between wild fish and farmed fish [2]. Wild fish, caught in their natural environments, typically have superior nutritional profiles, boasting higher levels of protein, omega-3 fatty acids, and other essential nutrients. This advantage comes from their varied diet, which includes smaller fish, plankton, and other nutrient-rich organisms. Consequently, wild fish often exhibit a richer flavor and greater nutritional benefits [3].

In contrast, fish raised in aquaculture systems typically have lower nutritional value. This is likely due to differences in their diet and living conditions. Farmed fish are usually fed commercial feed designed for rapid growth, but this may not provide the same nutrient levels as a natural diet. Additionally, farmed fish are often kept in overcrowded and stressful environments [4], which can weaken their immune systems and make them more susceptible to diseases.

Farmed fish often face a higher risk of disease and parasites due to the overcrowded and stressful environments in which they are raised [5]. This can negatively influence their overall health and well-being, potentially affecting the nutritional quality of the fish as well. For instance, farmed fish might have elevated levels of contaminants like PCBs and dioxins, which can pose risks to human health [6].

Even with these differences, farmed fish are easier to find and generally cheaper, making it simpler for people to access protein compared to wild fish. Additionally, some farmed fish producers are making efforts to enhance the nutritional value and sustainability of their offerings, so it's a good idea to investigate where your farmed fish is coming.

In Pakistan, the consumption of fish has been increasing steadily, driven by population growth and a trend towards more fish-based diets. Consequently, the aquaculture sector has seen significant expansion [7], especially in the farming of species like *Labeo rohita.* Rohu plays a vital role in Pakistan's aquaculture industry due to its high nutritional value, fast growth rate, and economic significance [8].

As the demand for fish continues to rise, it presents a variety of challenges that must be tackled to ensure the industry's sustainability. A major challenge is understanding and optimizing the digestive physiology of farmed fish [9], especially in species like *Labeo rohita.* Their growth, health, and overall productivity are closely tied to how effectively they convert feed into body mass. The efficiency of fish digesting and absorbing nutrients from their diet is affected by several factors, including the species' digestive anatomy, the quality and composition of the feed, and the surrounding environmental conditions.

Digestive enzymes play an important role in breaking down food into simpler [10], absorbable nutrients. Among the key enzymes involved in fish digestion are amylase and lipase. Amylase breaks down carbohydrates into simpler sugars, while lipase aids in fat digestion by converting fats into fatty acids and glycerol. The effectiveness of these enzymes is vital for transforming feed into energy and supporting growth. In farmed fish, particularly in intensive aquaculture systems with diverse feed formulations [11]. Recent studies indicate that the activity of digestive enzymes in fish is affected by various factors, such as diet composition [2], environmental conditions, and the unique digestive adaptations of different species. Additionally, the efficiency of enzyme activity can offer valuable information about the overall health and growth potential of farmed fish [13]. The role of digestive enzymes is important, but the histology of fish organs like the liver and intestines is also crucial for determining the overall health and productivity of farmed fish [14]. The liver serves as a vital organ for metabolism and detoxification; any damage or dysfunction can hinder nutrient utilization and overall growth. Histological changes in the liver, such as fatty degeneration or necrosis, may indicate stress from a poor diet, disease, or environmental pollutants [15]. Likewise, the intestine is the main site for nutrient absorption, and its health is closely linked to the fish's ability to derive energy from food. Therefore, histopathological evaluations of the liver and intestines are essential for assessing the impact of diet and environmental factors on fish health and productivity [16].

When it comes to nutritional value, growth, and overall health, there is a significant difference between wild fish and farmed fish. Wild fish, which are caught in their natural habitats, have better nutritional profiles with higher levels of protein, omega-3 fatty acids, and other essential nutrients [17]. This is because they feed on a diverse diet of smaller fish, plankton, and other organisms that are rich in nutrients. Despite these differences, farmed fish are more accessible and affordable [18], which helps to overcome the challenges of obtaining wild fish. But the fish farms, especially those situated near urban or industrial zones, often face exposure to pollutants like pesticides, heavy

metals, and agricultural runoff [19]. Hematological parameters are valuable indicators of fish health, offering insights into how well fish are managing environmental stressors [20]. For instance, increased white blood cell counts may suggest an active immune response to infections or environmental toxins, while fluctuations in hemoglobin levels could indicate oxygen deprivation or anemia.

This study aims to compare the different parameters of wild and farmed *Labeo rohita*. Like the comparison between the activities of amylase and lipase in the liver and the intestine. Secondly, determine the difference among the blood parameters of the two groups. Also, to compare the histopathological parameters in the heart, kidney, and gills of both groups. Mainly to find out which fish are healthier and which environment is more suitable, from the wild and farmed environments.

2. Materials and Methods

2.1. Sampling Sites:

Wild fish were trapped by netting in the lower Chanab canal & Farmed fish were collected by random sampling from aquaculture ponds from Chehnawan fish hatchery. The water quality parameters recorded at the Chehnawa Fish Hatchery (farmed environment) were: temperature 28–30°C, pH 7.4–8.0, and dissolved oxygen (DO) 5.5–7.0 mg/L. In contrast, the Lower Chenab Canal (wild environment) showed a temperature range of 25–29°C, pH 7.0–7.8, and DO levels of 4.0–5.5 mg/L.

2.2. Sample Size:

A total of 18 fish (9 wild and 9 farmed) in triplicate were used in the present study. The total lengths and weights of the wild specimens, W_1, W_2, and W_3, ranged from 27.5–29.8 cm and 950–977 g, as shown in Table 1. As we can see in Table 2, the total length of the three farmed Rohu samples (F1, F2, and F3) ranged from 25.5 cm to 30.4 cm, with a weight variation from 944 g to 980 g. The age of the fish was about 1.7 to 2 years. Fish were fed a commercial diet having 34% protein in the ponds before sampling. Fish were brought to the Zoology lab, the blood was taken from the samples and transferred to EDTA vials, and the dissected organs were removed.

2.3. Enzymes Essay:

Stored tissue samples were ground in cold 50 mM Tris-HCl buffer and centrifuged at 6000 rpm for 15 minutes at 4°C. The supernatant was collected and kept cold for later enzyme tests. All tests were done three times [21].

2.4. Amylase Essay

Amylase activity was measured using starch as the substrate, following Bernfeld's method (1955). One millilitre of diluted enzyme extract was mixed with one millilitre of 1% starch solution and incubated at 37°C for 3 minutes.

Table 1: Total length (cm) and weight (g) of three sample groups of Wild Rohu collected at Chehnawan Fish Hatchery Canal.

Sr. No.	Common Name	Scientific Name	Total Length (cm)	Total Weight (g)
W_1	Rohu	*Labeo rohita*	27.5±05	950±20
W_2	Rohu	*Labeo rohita*	29.8±05	977±20
W_3	Rohu	*Labeo rohita*	28.3±05	965±20

Table 2: Total length (cm) and weight (g) of three sample groups of Farmed Rohu collected at the Chehnwan Hatchery Ponds

Sr. No.	Common Name	Scientific Name	Total Length (cm)	Total Weight (g)
F_1	Rohu	*Labeo rohita*	25.5±05	944±20
F_2	Rohu	*Labeo rohita*	30.4±05	980±20
F_3	Rohu	*Labeo rohita*	28.0±05	959±20

The reaction was stopped by adding 3,5-dinitrosalicylic acid reagent and then heating the mixture in boiling water for 5 minutes. After cooling, distilled water was added, and the absorbance was measured at 540 nm using a spectrophotometer. A standard curve was prepared using maltose (10–100 µg/mL) to estimate the amount of starch hydrolyzed [21] (Equation 1).

Lipase Assay

Lipase activity was measured by mixing 3.5 ml of phosphate buffer (0.2 M, pH 6.9), 1 ml of enzyme extract, and 0.5 ml of olive oil substrate. This mixture was incubated at 37°C for 30 minutes with shaking. After incubation, 1 ml of acetic acid was added, and the mixture was titrated with 10 mM NaOH until the pH reached 10 (Equation 2) [22].

2.5. Hematological Analysis:

Blood samples were diluted using a solution containing sodium citrate (31.3 g), 10 ml of 37% formalin, Cresyl brilliant blue (1 g), and 1000 ml of purified water. Total erythrocyte and leukocyte counts were performed using a Neubauer hemocytometer. Differential leukocyte count was carried out by the Giemsa staining technique [23].

2.6. Hemoglobin Estimation and Hematocrit Measurement Hemoglobin concentration was determined using the cyanmethemoglobin method with Drabkin's reagent. Packed Cell Volume (PCV) was measured through the capillary tube microhematocrit method [24].

2.7. Calculation of Erythrocyte Indices

Erythrocyte indices provide important information about the size and hemoglobin content of red blood cells and are valuable in diagnosing different types of anemia. The following erythrocyte indices were calculated:

2.7.1. Mean Corpuscular Hemoglobin Concentration (MCHC)

The MCHC reflects the average hemoglobin concentration in a given volume of packed red cells. It is expressed as a percenta**ge (%**) (Equation 3).

2.7.2 Mean Corpuscular Hemoglobin (MCH)

The MCH indicates the average amount of hemoglobin per red blood cell. It is expressed in picograms (pg) (Equation 4).

2.7.3 Mean Corpuscular Volume (MCV)

The MCV represents the average volume of a single red blood cell. It is expressed in femtoliters (fL) (Equation 5).

2.8. Protein Determination

Total protein in the serum was estimated using the Biuret method, which is based on forming a colored complex between copper ions and peptide bonds under alkaline conditions. Absorbance was measured at 546 nm using an Erba Diagnostic Kit. The concentration was calculated by following the procedure as documented by Kumar et al. [25]. The concentration of serum protein was calculated following equation (6).

Amylase Activity (U/ml) = (Delta A enzyme - Delta A blank) * Standard Factor/Incubation Time* Dilution Factor Equation (1).

Lipase Activity (U/ml) = Volume of NaOH used* Molarity of NaOH*1000 * 2 *Dilution Factor/Volume of Sample Used. Equation (2).

$$MCHC(\%) = \left(\frac{Hemoglobin\ (g/dl)}{Packed\ cell\ volume\ (\%)}\right) \times 100$$

Equation (3)

$$MCH(pg) = \left(\frac{Hemoglobin\ (g/dl)}{Erythrocyte\ count\ (million/mm^3}\right) \times 10$$

Equation (4)

$$MCV(fL) = \left(\frac{Packed\ cell\ volume\ (\%)}{Erythrocyte\ count\ (million/mm^3}\right) \times 10$$

Equation (5)

$$\text{Total serum Protien (g/dL)} = \left(\frac{\text{Absorbance of test}}{\text{Abdorbance of standard}}\right) \times \text{Concentration of standard(g/dL)}$$

Equation (6)

$$\text{Serum albumin (g/dL)} = \left(\frac{\text{Absorbance of test}}{\text{Absorbance of standard}}\right) \times \text{Concentration of standard (g/dL)}$$

Equation (7)

$$\text{Serum Globulin(g/dL)} = \text{Total serum protein} - \text{Serum Albumin}$$

Equation (8)

Where MCHC (%) indicates mean corpuscular hemoglobin concentration in percent, MCH (pg) in picograms, and MCV (fl) in femtoliters.

Serum Albumin

Serum albumin levels were determined by the Bromocresol Green (BCG) assay, in which albumin binds to the dye to form a green complex. The absorbance was recorded at 630 nm using the same diagnostic kit. Albumin concentration was calculated following equation 7.

Serum Globulin

Serum globulin was calculated indirectly by subtracting serum albumin from total protein (Equation 8):

2.9. Histopathological Analysis:

Extracted organs were immediately fixed in Bouin's for 24 hours to ensure proper storage of cell structures. After fixation, the samples were thoroughly washed with distilled water to remove any remaining fixatives. Clearing was performed using xylol to promote tissue transparency, and the sample was created for embedding. The removed fabric was embedded in a paraffin wax block to provide mechanical support for cutting [26]. These sections were carefully attached to clean glass slides and coated with adhesive to ensure proper adherence. The object rack was then unfolded with xylol and rehydrated by rinsing with distilled water by reducing the ethanol concentration.

Staining was performed using standard hematoxylin and eosin (H&E) protocols to allow visualization of cell and tissue structures. After staining, sections of elevated ethanol quality were dehydrated and cleared again with xylol. Finally, I installed a colored section with deck glass as mount medium using DPX. Microshoppers were recorded to document the results and to facilitate other comparative analyses.

Fish handling and sampling procedures were approved by the Departmental Ethical Review Committee, University of Central Punjab. Wild fish were collected with proper permissions, and all specimens were humanely euthanized using an overdose of MS-222 before sampling.

2.10. Statistical analysis

All data were presented as the mean value's standard deviation. The data was analyzed by SPSS software (14 versions). The data was analyzed by independent T test. The mean values were compared, and the values less than 0.05 were considered to be significant.

3. Results

In this study, we took a closer look at the enzymatic activities in both the liver and intestine of wild and farmed freshwater *Labeo rohita*. We also measured various biochemical and hematological parameters, and our histopathological comparisons revealed significant differences between the farmed and wild fish, particularly in the heart, kidney, and gills.

3.1. Enzymes assay:

Amylase assay: When it comes to enzymatic activity, we observed a striking difference in amylase levels between wild and freshwater Rohu, as highlighted in Table 3. Figure 1 illustrates that the liver amylase activity in wild Rohu was notably higher at 8.0 U/L, compared to just 1.0 U/L in farmed Rohu. On the flip side, lipase activity was significantly greater in the farmed Rohu at 13.6 U/L, while wild Rohu had a lower level of 9.3 U/L.

This suggests that wild Rohu have adapted to make use of a broader range of dietary carbohydrates available in their natural habitats, unlike the more limited options found in aquaculture settings.

3.2. Lipase assay

The activity of hepatic lipase in the intestines was found to be identical in both farmed and wild Rohu, sitting at 9.3 U/L. Similarly, amylase activity showed no significant differences between the two groups. However, when it came to intestinal lipase and amylase, there were no notable variations as seen in the liver, which is illustrated in Figure 2. The increased liver lipase levels in Rohu raised in aquaculture might be an adaptation to help them make the most of the lipid-rich feed that's typically used in these settings. The fact that both groups exhibited similar intestinal lipase activity suggests that their digestive functions were quite alike, regardless of their different rearing environments.

3.3. Hematological parameters:

As illustrated in Table 4, farmed Rohu had a bit more plasma proteins, clocking in at 2.86 g/dL, compared to their wild

counterparts, which measured 2.552 g/dL. The concentrations of albumin and globulin followed a similar pattern, with farmed Rohu showing significantly higher levels of 1.617 g/dL for albumin and 1.243 g/dL for globulin, while wild Rohu had lower values of 1.53 g/dL for albumin and 1.017 g/dL for globulin. These differences suggest that the controlled diet in farming conditions may lead to an increase in circulating proteins, which can help support growth and boost immunity.

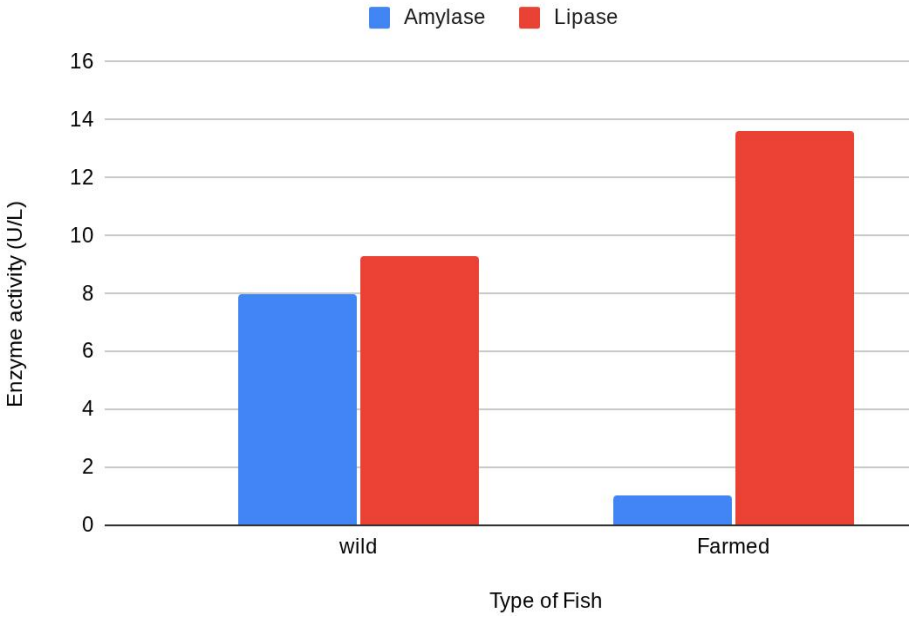

Figure 1. Comparative amylase and lipase activities in the liver of wild and farm-raised *Labeo rohita*.

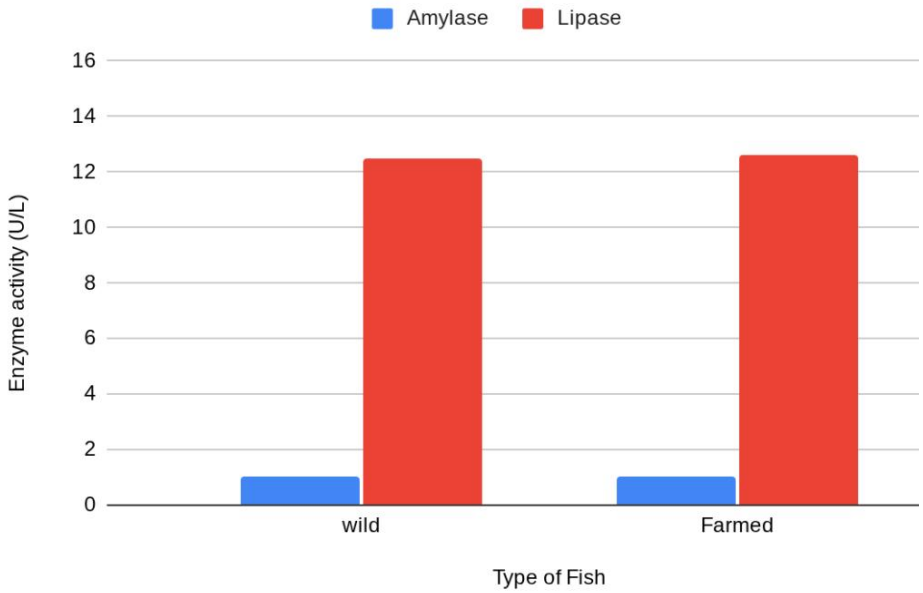

Figure 2. Comparative amylase and lipase activities in the intestine of wild and farm-raised *Labeo rohita.*

Table 3. Comparison of Amylase and Lipase activities in the Liver and Intestine of wild and farmed fish

Type of fish	Amylase in Liver (U/l)	Amylase in Intestine (U/l)	Lipase in Liver (U/l)	Lipase in Intestine (U/l)
Wild	08.00±0.27	01.00±0.33	09.30±0.37	12.50±1.09
Farmed	01.00±0.65	01.00±0.43	13.60±0.44	12.60±0.73

Table 4. Comparative biochemical and hematological parameters of farmed and wild *Labeo rohita*

Parameters	Farmed	Wild
Hemoglobin (g/dl)	08.90±0.27	06.70±0.35
WBC (TLC) (x10 /uL)	07.10±0.33	11.10±0.46
Total RBC (x10 /uL)	01.60±0.36	01.80±0.12
HCT (PVC) %	26.70±1.02	21.60±0.87
MCV (fL)	166.8±2.15	166.1±2.69
MCH (pg)	52.30±1.57	48.00±1.43
MCHC (g/dL)	33.30±0.99	41.20±1.05
Plasm protein (g/dL)	02.86±0.08	2.552±0.12
Albumin (g/dL)	1.617±0.53	01.53±0.78
Globulin (g/dL)	1.243±0.47	1.017±0.32

Wild Rohu fish show a higher mean corpuscular hemoglobin concentration (MCHC) of 41.2 g/dL, compared to just 33.3 g/dL in farmed varieties. This natural adaptation could explain why wild Rohu are better equipped for oxygen transport in their cells, especially since their natural habitats often have fluctuating oxygen levels.

Farmed Rohu fish showed significantly higher hemoglobin levels compared to their wild counterparts, with readings of 8.9 g/dL versus 6.7 g/dL. This notable difference suggests that the diet and living conditions of farmed Rohu contribute to a better ability to transport oxygen. Similarly, the hematocrit values (HCT) were also elevated in the farmed Rohu, measuring 26.7% compared to 21.6% in the wild ones. This indicates that the farmed group has an overall improved capacity for oxygen transport.

We discovered that wild Rohu kala had notably higher white blood cell (WBC) counts, measuring 11.1 x10³/uL, compared to their farmed counterparts, which had 7.1 x10³/uL. This elevated WBC level in wild Rohu could suggest that they are experiencing an adaptive immune response to the various environmental challenges and pathogens they encounter in their natural habitat.

While the total red blood cell (RBC) counts in both wild and farmed Rohu were comparable, wild fish showed a slight edge with RBC levels at 1.8 x10³/uL, compared to 1.6 x10³/uL in farmed ones, as highlighted in Table 4. This suggests that wild fish might be experiencing a bit more hypoxic or anoxic stress, prompting a physiological response that demands more efficient oxygen delivery to cope with the ever-changing conditions of their environment.

3.4. Histopathological parameters:

When we take a closer look at the histopathology, we can see some significant differences between farmed and wild fish, especially in their heart, kidneys, and gills.

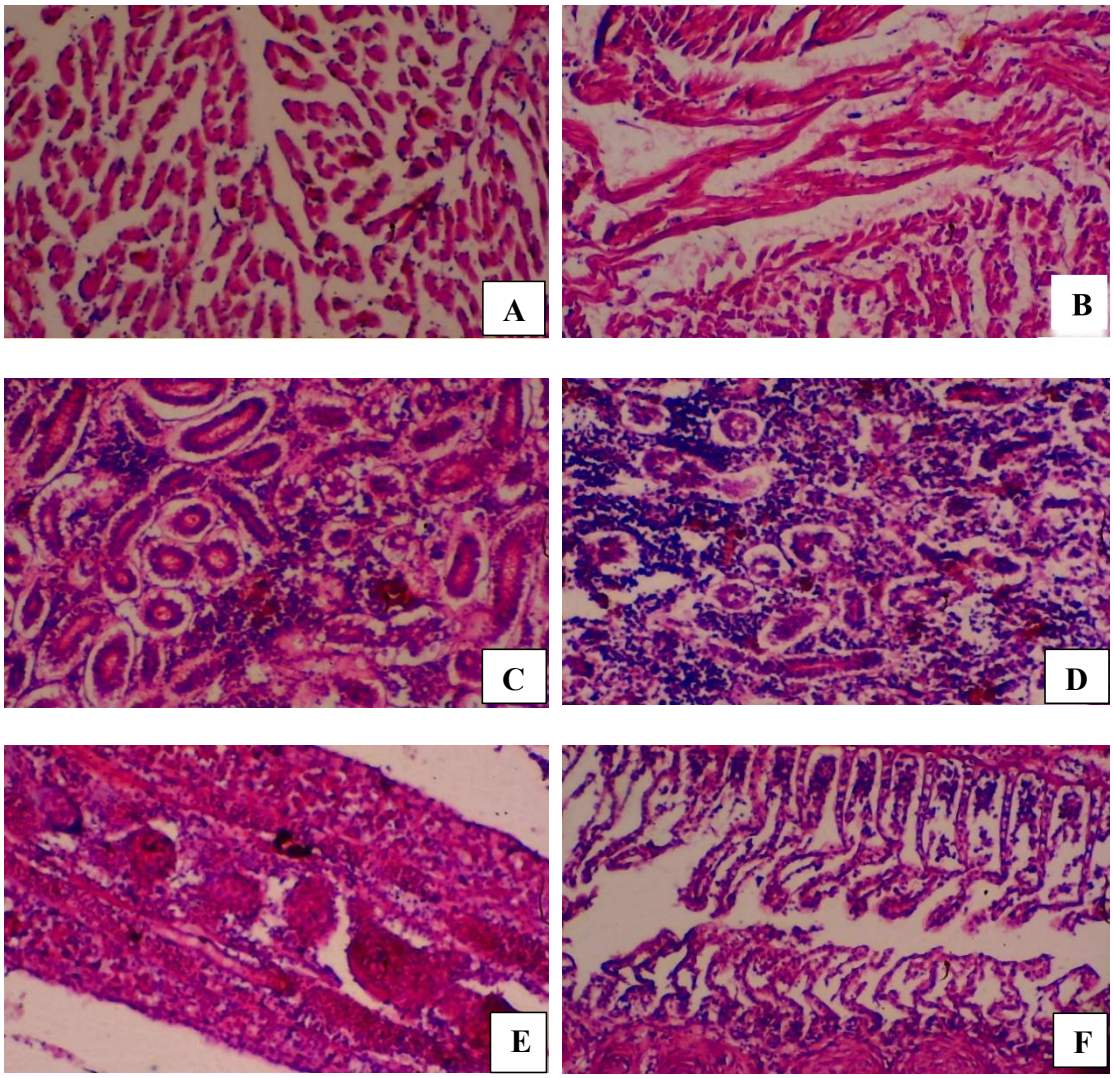

Figure 3. Histopathological results of farmed and wild fish. Histopathology of the heart (A), kidney (C), and gills (E) of farmed fish. Histopathology of the heart (B), kidney (D), and gills (F) of wild fish.

The farmed fish have hearts that look structurally normal, complete with a well-defined vascular supply, intact nerve bundles, and healthy muscle fibers. There's no sign of inflammatory diseases, granulomas, or cancer as shown in Figure 3 (A). In contrast, the wild fish exhibit elongated blood vessels in their hearts and softer muscle fibers, which suggests they might be under a lot of stress or have a weaker structure, which is illustrated in Figure 3 (B). However, just like the farmed fish, they also show no signs of granulomas or malignancy. As illustrated in Figure 3 (C), the kidneys of farmed fish show normal renal tissue, complete with nephrons that feature healthy tubules and ducts, a steady vasculature ratio, and no signs of inflammatory disease, granuloma, or cancer. However, there are some notable differences compared to wild fish, such as an increase in the interstitial space between glomeruli, congestion in the blood vessels, focal hemorrhages, and hydronic degeneration of the renal tubular epithelium, which you can observe in Figure 3 (D). Additionally, the glomerular Bowman spaces displayed edema, which may indicate a reduced GFR, suggesting renal dysfunction even in the absence of granuloma or malignancy.

As illustrated in Figure 3 (E), farmed fish look healthy and

have intact gill filaments when examined under a microscope, showing no signs of disease, granulomas, or cancer. In contrast, wild fish display significant gill damage, including hemorrhages and ruptured lamellar epithelium. We can also see micro-aneurysms and congested blood vessels on their gills, as shown in Figure 3 (F). This kind of morphological damage in wild-caught fish can seriously hinder their ability to exchange gases. Various environmental factors, like pollutants or poor water conditions, can affect how well the gill's function. In summary, while farmed fish exhibit healthy organ structures, wild fish reveal multiple signs of stress and possible physiological issues in their vital organs.

4. Discussion:

In this study, we investigated the effects of rearing environments wild versus farmed on the digestive enzyme activities, hematological parameters, biochemical profiles, and histopathological characteristics of *Labeo rohita.* By integrating our findings with prior research, we aim to elucidate how environmental and dietary factors shape the physiology, health, and adaptability of these species in contrasting habitats. The merged analysis highlights consistent patterns and provides insights into the implications for aquaculture management and environmental monitoring.

Our results revealed significant differences in digestive enzyme activities between wild and farmed *Labeo rohita* reflecting dietary adaptations to their respective environments. Wild fish exhibited higher liver amylase activity compared to farmed counterparts, likely due to a varied, natural diet rich in complex carbohydrates that necessitate enhanced carbohydrate digestion. In contrast, farmed fish displayed greater liver lipase activity, which can be attributed to the higher fat content in formulated aquaculture feeds. These findings align with Iqbal et al. (2018), who reported elevated lipase levels in fish fed plant-based diets [22], and Murtaza et al. (2016), who noted increased intestinal lipase activity in fish fed corn gluten, mirroring the higher lipase activity in our farmed *Labeo rohita* [21]. Interestingly, intestinal amylase and lipase activities were comparable between wild and farmed groups, supporting Lundstedt et al. (2004), who found no significant differences in intestinal enzyme activities between cultured and wild fish, indicating that intestinal digestion adapts to functional demands regardless of environment [37]. These similarities suggest that while liver enzyme profiles are sensitive to dietary composition, intestinal digestion maintains a stable capacity across environments, ensuring efficient nutrient breakdown.

Hematological data revealed distinct differences between wild and farmed *Labeo rohita* reflecting physiological adaptations to their environments. Farmed fish exhibited higher hemoglobin (8.9 g/dL vs. 6.7 g/dL) and hematocrit (26.7% vs. 21.6%) compared to wild fish, suggesting enhanced oxygen transport capacity likely due to nutrient-rich diets and stable oxygen levels in aquaculture settings. These findings are consistent with Prasad and Charles (2010), who reported elevated hemoglobin and hematocrit in cultured *Catla catla* under controlled conditions [42]. Similarly, Habib et al. (2021) observed higher hemoglobin and hematocrit in farmed fish, supporting the notion that controlled diets enhance circulatory protein levels, potentially improving immune function and growth in farmed *Labeo rohita* [31].

Conversely, wild *Labeo rohita* showed higher white blood cell (WBC) counts (11.1 $x10^3/\mu L$ vs. 7.1 $x10^3/\mu L$) and mean corpuscular hemoglobin concentration (MCHC, 41.2 g/dL vs. 33.3 g/dL), indicating an adaptive immune response and enhanced oxygen-carrying efficiency in response to environmental stressors. These results align with Nwani et al. (2013), who reported elevated WBC counts in wild *Channa punctatus* exposed to environmental pollutants, suggesting that wild *Labeo rohita* face greater immune challenges in natural [40]. The slightly higher red blood cell (RBC) count in wild fish (1.8 $x10^3/\mu L$ vs. 1.6 $x10^3/\mu L$) corroborates Kumar et al. (2017), who noted increased RBC counts in wild fish under hypoxic conditions, reflecting physiological adaptations to fluctuating oxygen levels [36]. These hematological differences highlight how controlled aquaculture conditions enhance oxygen transport and metabolic efficiency, while wild fish develop robust immune and adaptive responses to environmental variability.

Biochemical profiles further underscored the impact of rearing environments on *Labeo rohita* health. Farmed fish exhibited higher total plasma protein levels, with slightly elevated albumin and higher globulin levels, indicating better nutritional status and immune function due to consistent feeding and reduced pathogen exposure in controlled settings. In contrast, wild fish showed lower albumin levels, likely reflecting unpredictable food availability in natural habitats. These findings are supported by Asghar et al. (2023), who reported higher protein content in farmed *Labeo rohita* emphasizing the nutritional advantages of aquaculture [29]. Additionally, Pradhan et al. (2013) noted that environmental factors and sex influence biochemical parameters, with males showing higher total protein, globulin, and albumin levels, aligning with our observations of environment-driven differences in protein profiles [32]. The elevated plasma proteins in farmed fish suggest improved physiological regulation and nutrient transport, contributing to their overall health and growth potential.

Histopathological analysis revealed stark contrasts between farmed and wild *Labeo rohita* highlighting the impact of environmental stressors. Farmed fish displayed normal heart, kidney, and gill structures, with no signs of inflammation, granulomas, or malignancy, consistent with Rašković et al. (2011), who reported normal histology in cultured carp under controlled conditions with clean water and balanced diets. In contrast, wild fish exhibited significant abnormalities, including cardiac stress (elongated blood vessels, softer muscle fibers), renal and gill damage (hemorrhages, ruptured lamellar epithelium, micro-aneurysms) [16]. These findings align with Poleksic et al. (2010), who reported gill damage (e.g., lamellar rupture, hemorrhages) in wild fish exposed to environmental pollutants, and Abalaka et al. (2015), who noted renal abnormalities like glomerular edema and tubular degeneration in wild fish exposed to heavy metals [41,43]. Bhanot et al. (2021) similarly reported severe histopathological damage in muscular tissue of *Labeo rohita* exposed to untreated sewage water [27], while another study observed necrosis and significant gill damage in fish exposed to high arsenic concentrations (30 mg/L) [33]. Mallik et al. (2020) also reported gill pathologies in wild *Labeo rohita* from polluted rivers, linking them to heavy metals and organic pollutants [34]. The absence of granulomas or malignancy in both groups aligns with Fernandes et al. (2007), who found no neoplastic changes in fish unless exposed to specific carcinogens [38].

The integrated findings underscore the profound influence of rearing environments on *Labeo rohita* physiology. Higher liver amylase in wild fish and lipase in farmed fish reflect dietary adaptations, consistent with studies on cyprinids [21, 22]. Hematological differences, such as elevated hemoglobin and plasma proteins in farmed fish and higher WBC and MCHC in wild fish, align with research on nutritional and environmental influences. Histopathological damage in wild fish, particularly in gills and kidneys, mirrors the report of environmental stress in wild populations, while healthy tissues in farmed fish highlight the benefits of controlled conditions. These patterns emphasize the role of diet, water quality, and environmental stressors in shaping fish health.

For aquaculture management, the lower amylase activity in farmed *Labeo rohita* suggests that carbohydrate-rich feeds could be optimized to enhance digestion, potentially improving feed efficiency and growth. The elevated WBC counts and histopathological damage in wild fish highlight the need for monitoring environmental pollutants in natural habitats, as suggested by Poleksic et al. (2010),[41]. The nutritional advantages of farmed fish, as evidenced by higher plasma proteins, underscore the importance of maintaining high-quality feed and water in aquaculture to ensure fish health and safety for consumption which is aligned with the Asghar et al, (2023) [29]. Future research could explore proteomic or transcriptomic profiles, as in Nissa et al. (2022), to uncover molecular mechanisms underlying these differences, further informing sustainable aquaculture practices and environmental conservation strategies [39].

These findings contribute to a comprehensive understanding of how farming practices and environmental conditions affect *Labeo rohita* quality and health, emphasizing the need for

controlled management strategies in aquaculture to ensure the sustainability and safety of fish as a vital protein source.

.Conclusion

This study aimed to evaluate the comparative health status of wild and farmed *Labeo rohita* by analyzing their hematological, biochemical, enzymatic, and histological profiles. The findings suggest that farmed fish generally exhibited better hematological and biochemical parameters, including higher hemoglobin, albumin, and plasma protein levels, as well as structurally healthier organs with no visible signs of inflammation or degeneration. In contrast, wild fish showed elevated WBC counts and signs of immune activation, which may reflect a response to environmental stressors. Despite their compromised organ structures, these immune-related adaptations in wild fish suggest potential resilience under natural conditions. Farmed fish also demonstrated higher lipase activity, especially in the liver, indicating efficient lipid metabolism. In summary, farmed fish show better overall health and organ condition due to controlled feeding and environment. Wild fish, however, seem to have stronger immune responses but also show more physical damage, likely because of the challenges in their natural habitat. This means there is a trade-off between health and adaptability, and neither environment is better, both have advantages depending on what is most important for the fish's survival and well-being.

Data availability statement

The data supporting the results of this study can be obtained from the corresponding author upon request.

Conflicts of interest

All authors declare that they have no conflicts of interest.

Ethical approval

This work is done and approved by the DERC of the university.

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Authors Contribution

Umar Tayyab, Tehseen Hajra Iqbal, Jawairia Zahid, Noor Fatima, Muhammad Zain Ullah, Manahil, and Jamil Ahmad conducted the research, collected and analyzed the data, and wrote the manuscript. Dilawar Hussain and Muhammad Mudassar Shahzad supervised, edited, and reviewed the article

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