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Research article

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Comparative Study of Whole-Body Proximate Composition, Dressing Losses and Bioaccumulation of Heavy Metals in Muscle Tissues of Wild and Farm-Raised Labeo Rohita Imtisal Shahid¹, Remsha Bibi¹, Dilawar Hussain^{1*}, Esha Khan¹, Sidra Bibi¹, Muhammad Faheem¹, Fatima Gulzar¹ and Rimsha Munawar¹

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Abstract

The muscle tissues of wild and farm-raised rohu were analyzed for proximate composition (Crude protein, crude fat, ash, and dry matter) and uptake of heavy metals (Cd, Ni, Pb, and Cr) using the standard protocols. For this purpose, wild fish samples were collected from the Khanpur Canal, Sheikhupura, and farmed fish samples from a local fish market in Lahore. The results showed that the concentration (%) of protein (17.92 ± 0.36) , ash (06.54 ± 0.13) , and moisture (76.50 ± 1.53), was observed to be higher in the wild when compared to protein (16.90 ± 0.34) , ash (5.07 ± 0.10) and moisture (73.50 ± 1.47) of farm-raised rohu. In contrast, crude fat contents were observed to be higher (4.02 ± 0.08) in farm-raised rohu than wild (03.65 ± 0.07) . In terms of dressing losses, wild fish showed higher (26.37%) losses as compared to farmraised (25.83%). Results showed that in wild and farm fish's muscles, the trend of the heavy metals was Pb > Cd > Ni > Cr. The findings demonstrated that in comparison to Cd, Ni, and Cr, Pb concentration (ppb) was greater in both wild (128.32.82) and farm-raised (111.42.16) rohu. When compared to wild rohu, farmed rohu showed a higher bioaccumulation tendency for Cr and Ni, but Cd and Pb showed the reverse pattern. The findings of this study suggest that fish proximate composition, dressing losses, and bioaccumulation of heavy metals are certainly impacted by the aquatic life of freshwater fish like Labeo rohita both in wild and farmed conditions and these results will be helpful for the aquaculturists as well as the local consumers of the fish.

Keywords: Proximate composition, heavy metals, muscle tissues, dressing losses, wild, cultured.

1. Introduction

Fish is a worldwide recognized essential and prominent protein source, and it is playing an important role in aquaculture research. Fish provides a higher amount of easily digestible protein to over 4.5 billion individuals [1]. Fish is composed of 65-80% moisture, 15-20% protein, 5-20% fat, and 0.25% ash. 60% of individuals in developing nations ingest fish protein [1].

Pakistan is mostly an agricultural country with abundant natural water resources. Aquaculture's primary source is freshwater and seawater. Aquaculture accounts for 1% of the Gross Domestic Product (GDP). Because of the scarcity of culture, the majority of contributions come from catch fisheries [2]. The chemical composition of fish flesh is considered to be a good predictor of the quality, nutritional value, physiological state, and habitat of the fish. 66-81% water, 16-21% protein, 1.2-1.5% mineral, 0.2-25% fat, and 0-0.5% carbohydrate are found in fish. Fish's body composition is said to consist of 96-98% water, protein, fat, and ash. The term 'proximate composition' of fish refers to the evaluation of these elements. In order to interpret the nutritional value, physiological state, and overall health of any consumable part of the organism, a

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thorough examination of the species' proximate composition is necessary [3]. Increases in other parameters, such as feed ratio and fish size, also seemed to have an impact on fish body composition, as they led to increased adipose deposition and decreased water contents in the fish body [4].

All living things rely on river water for survival. Because of its environmental toxicity, persistence, bioaccumulation, and biomagnification in the food chain, heavy metal pollution has become a global issue [5,6,7,8]. Freshwater bodies with high levels of heavy metals are unfit for human consumption, livestock watering, or irrigation [9]. Natural processes like bacterial activity, spring water, erosion, and volcanic eruptions disperse heavy metals throughout the environment. However, a variety of human activities, including the discharge of industrial effluents, sewage sludge, home waste, agricultural runoff, and so forth, are also significant contributors to heavy metal pollution in water bodies [10]. Microbes can quickly transform heavy metals into more dangerous organic forms, some of which are deadly to people and aquatic species [11]. Human health issues such as liver damage, cardiovascular disease, and renal failure can be brought on by heavy metal pollution [12].

Fish consumption has expanded considerably in recent years, as has awareness of its nutritional and therapeutic benefits [13]. Fish is a complete food source that is extremely important in the diet because it has a high concentration of protein and fats with great biological value [14]. Fish tissue's heavy metal content indicates how much of these contaminants the fish have been exposed to in the past or are currently exposed to them [15]. In aquatic environments, fish classified as edible are a significant group of animals because fish tissue contains heavy metal ions that may function as potential food chain metal ion transporters. Ultimately, man is affected by metal ions in the aquatic medium, either directly or indirectly [16]. Concern over heavy metal bioaccumulation in fish tissue is growing since these metals are known to be toxic and to have detrimental effects on both human and animal health [17].

The current study is therefore designed to analyze and

compare the proximate composition (Crude protein, Crude fat, Ash, and Dry matter), dressing losses, and heavy metal load (Cd, Cr, Pb, Ni) in muscle tissues (meat) of wild and farmraised *Labeo rohita* (Rohu).

2. Materials and Methods

2.1. Selection of fish species

Labeo rohita, is one of the most significant, highly sought-after, consumable, and widely cultured fin fish species. It was chosen for the study because a greater proportion of the Punjab province's population consumes rohu than any other fin fish species, both for normal fish meals and for special occasions or festivals.

2.2. Site selection and sampling of wild Labeo rohita

Khanpur Canal is an important waterway located in Sheikhupura a district in the Punjab province of Pakistan (Figure 1). The canal serves as a significant source of drinking urban areas in the district, improving access to clean water for domestic use. The canal supports fish farming and serves as a habitat for various fish species. Fishermen in Sheikhupura rely on the canal for their livelihoods, engaging in fishing activities and selling fish in local markets. From a fish point of view, this canal is very significant due to its rich aquatic ecosystem on the availability of various fish species. This canal is known for its abundant fish population, making it an attractive location for anglers and fishing enthusiasts, and provides a favorable habitat for several fish species to thrive.

Healthy *Labeo rohita* (3 fish of Avg. wt. 950±50 g) were purchased from the local fisherman who *caught* that fish from the canal. The fish were washed and packed in polythene bags having ice cubes to prevent bacterial infections. The fish was then transported directly from the sampling site to the Quality Control Labs, Fisheries Research & Training Institute Manawan, Lahore for proximate composition, dressing losses, and heavy metal (cadmium, chromium, nickel, lead) analysis.

2.3. Market selection and sampling of farmed Rohu

The farmed fish samples of fresh *Labeo rohita* (3 fish of Avg. wt. 900±30 g) were purchased from the local fish market located at Lahori gate, old town Lahore (Figure 2). The fish were washed and packed in polythene bags having ice cubes to

Journal of Zoology and Systematics prevent bacterial infections.



Figure 1. Picture of Khanpur canal



Figure 2. Sampling site for market selection and sampling of farmed Rohu.

The fish was transported directly from the sampling site to the Quality Control Labs, Fisheries Research & Training Institute Manawa, Lahore for proximate composition, dressing losses, and heavy metal (cadmium, chromium, nickel, lead) analysis.

2.4. Proximate analysis

After measuring the weight and length, the fish samples were placed in a solution of water and clove oil. The sample was dried and pulverized in an oven at 70°C for 24 hours and stored in the refrigerator for further analysis (% Crude Protein, % Crude Fat, % Dry Matter, and ash %, according to AOAC [18].

2.5. Dressing losses

The fish were slaughtered and given a longitudinal cut from the ventral side after each sample was cleaned in tap dechlorinated water. After the fish was dissected, a few chosen visceral organs, the head, and the skin were taken out and weighed using an electric balance.

2.6. Digestion of the Samples for heavy metal analysis

Churn's (1975) dry-weight method was modified to prepare fish tissue samples for Cd measurement. Using mortars and pestles, the dried meat and offal of the fish sample were ground into a composite sample. In a 100 ml beaker, the dry weight of each composite sample (5.0 g for meat and offal) was stored in triplicate. Each sample received 10 millilitres of concentrated nitric acid (HNO3), which was then allowed to digest for the entire night. The very next day, after the samples had completely broken down, the metals were removed by heating the beakers containing the samples to 70 °C on a plate. The digestion was complete when the colour of the solution turned from pale yellow to translucent. To create a transparent solution, 1.0–2.0 ml of perchloric acid (HClO4) was added to the sample drop by drop. Once the solutions had fully broken down, they were allowed to cool to room temperature, diluted with ion-free double distilled water, filtered through Whatman filter paper No. 1 (110 mm), and finally placed into sample bottles (Tarson), each holding a final volume of 30 ml [19].

2.7. Detection of metals by atomic absorption spectrophotometer

Utilizing hollow cathode lamps made of Pb and Cd, the metal content of the sample was found using a Varian AA 240 Atomic Absorption Spectrophotometer. From stock solutions (1000 mg/1) were created three standard solutions (0.5 mg/1, 1.0 mg/1, and 1.5 mg/1) cd. Each sample's metal concentration was determined using a standard curve created by charting the standard solution's absorption values along the Y-axis and X-axis. Each sample's final concentration was given as micro g of metal/ (d wt) [19].

2.8. Statistical analyses

All the data were subjected to one-way ANOVA using Statistix 8.1 software. The comparison of means was calculated by Tuckey's test. Making of graphs and standard deviation were calculated by Microsoft Excel 2016 [19].

3. Results

3.1. Length and weight of fish

The length and weight of both wild and farm raised *Labeo rohita* are represented in Tables 1 and 2, respectively.

3.2. Proximate composition

In *Labeo rohita*, maximum crude protein (17.92 ± 0.36) was observed in the wild and minimum (16.90 ± 0.34) in farmraised rohu. Maximum ash content (06.54 ± 0.13) was observed in the wild and minimum (5.07 ± 0.10) in farm-raised rohu. A reverse trend was observed in terms of crude fat the maximum fat content was observed in the farm-raised (4.02 ± 0.08) and minimum (03.65 ± 0.07) in wild rohu. Analysis of Variance (ANOVA) showed that the differences remained statistically non-significant (P> 0.05) for dry matter, ash, and fat contents in wild and farmed rohu (Table 3). For moisture, maximum contents were observed in the wild (76.50) and minimum in farm-raised (73.50) rohu, and Analysis of Variance (ANOVA) showed that the differences remained statistically significant (P<0.05) for dry matter contents in wild and farmed rohu (Table 3; Figure 3).

3.3. Dressing Losses

Wild *Labeo rohita* showed more dressing losses (26.37%) when compared to farm-raised (25.83%) *Labeo rohita* (Table 4).

3.4. Bioaccumulation of Chromium (Cr) & Nickle (Ni)

In *Labeo rohita*, the maximum concentrations of both Cr (16.65 ± 0.65) and Ni (22.19 ± 1.80) were found in farm-raised rohu and minimum Cr (13.29 ± 0.72) and Ni (18.28 ± 0.28) in wild. Analysis of Variance (ANOVA) showed that the difference remained statistically significant (P<0.05) for Cr and Ni (Table 5).

3.5. Bioaccumulation of Cadmium (Cd) & Lead (Pb)

In terms of bioaccumulation of Cd and Pb, a reverse trend was observed. For both the metals, maximum concentrations of Cd (54.30 ± 0.66) and Pb (128.3 ± 2.82) were found in wild rohu, and minimum Cd (54.30 ± 0.66) and Pb (111.4 ± 2.16) in farmed raised rohu. Analysis of Variance (ANOVA) showed that the difference remained statistically significant (P<0.05) for Cd and Pb (Table 5; Figure 4).

4. Discussion

4.1. Proximate composition

Due to their delicate nature and wide tolerance range at the community level, fish are an important indicator in an ecosystem. The proximate composition of the same species of fish can change depending on the water's depth, quality, and environmental and feeding conditions. Our comparison of the proximate composition of farmed and wild rohu found that the wild fish has a greater crude protein content (17.92 ± 0.36) when compared to farmed fish (16.90 ± 0.34) . Our findings are in line with the studies of [1] and [20] who concluded that Wild Sea breams had significantly higher muscle protein than cultured conspecifics.

In the current study, moisture contents were found higher in wild fish (76.50±1.53) and lower in farm-raised fish (73.50 ± 1.47) . The present results contradict the findings but are in line with the results recorded by [21]. Lipids are generally regarded as the most important constituents, which determine the quality of fish meat. In the present experiment, lipid contents were observed at a maximum (4.02±0.08) in farmed fish and a minimum in (03.65±0.07) wild fish. The significant variation in the protein and fat composition of wild fish appears to be caused by the fish's size and weight as well as the lack of available food. Fish experienced a prolonged period of restricted food supply as a result of this state of scarcity, which led to a decrease in fish growth. Fish experienced a prolonged period of restricted food supply as a result of this state of scarcity, which led to a decrease in fish growth. Fish gradually reduced their fat stores, but before they reached a critical lowlevel, protein started to be used for energy. As a result, their protein contents decreased while their water content increased. According to [22], The fish initially feeds on lipids from the liver and only begins to mobilise muscle protein when its reserves of energy derived from fat are almost depleted. After then, water replaces the protein as it is used up. Increased water contents as a result of this change were negatively correlated with the amounts of protein and fat in their meats [23].

Fish gradually reduced their fat reserves, but before they reached a critical level, they started to be used for energetic –

Table 1: Total length (cm) and weight (g) of three samples of Wild (W) Labeo rohita collected at Khanpur canal					
Sr. No.	Common Name	Scientific Name	Total Length (cm)	Total Weight (g)	
W ₁	Rohu	Labeo rohita	27.5	950	
W_2	Rohu	Labeo rohita	29.8	977	
W3	Rohu	Labeo rohita	28.3	965	

Table 2: Total length (cm) and weight (g) of three samples of Farmed (F) Labeo rohita collected from the local market

Sr. No.	Common Name	Scientific Name	Total Length (cm)	Total Weight (g)
F1	Rohu	Labeo rohita	25.5	944
F_2	Rohu	Labeo rohita	30.4	980
F ₃	Rohu	Labeo rohita	28.0	959

Table 3: Proximate analysis (%) in muscle tissues of Wild and Farmed Rohu.

Type of fish	Crude protein	Ash	Crude Fat	Moisture
Wild	17.92±0.36 A	06.54±0.13 A	03.65±0.07 A	76.50±1.53 A
Farmed	16.90±0.34 A	5.07±0.10 A	4.02±0.08 A	73.50±1.47 B

Note: Values are presented as mean \pm standard deviation. Different uppercase letters denote significant differences (p < 0.05) between wild and farmed fish .

Table 4: Dressing losses in wild and farmed Rohu

Type of fish	Sample no	Avg live	Avg dressed	Avg fresh fish total	% Loss	Mean
		weight (g)	weight (g)	loss (g)		Loss
	\mathbf{W}_1	950	722	228	24	
Wild	W_2	977	721	256	26.2	26.37
	W ₃	965	686.15	278.88	28.9	
	F_1	944	722.44	221.55	23.5	
Farmed	F_2	980	730	250	25.5	25.83
	F ₃	959	685.61	273.31	28.5	

purposes and finally a decrease in their protein contents combined with an increase in water content meant that the fish initially consumed lipids from the liver and only began to mobilise muscle protein when the energy derived from fat had almost completely been consumed. After then, water replaces the protein as it is used up. Increased water contents as a result of this change were negatively correlated with the amounts of protein and fat in their meats. [24].

4.2. Dressing losses

When fish were dressed, the average total losses (Table 3) matched the live body weight of the fish exactly. The wild

Labeo rohita under W3 always had the highest dressing percentage, closely followed by the farmed Labeo rohita under F3. When compared to other weight categories of wild and farmed fish, the higher weights of external losses in wild fish were the cause of this. This might be caused by the fish's genetic function, according to those who believe that this is a physiological activity rather than a genetic function. These results are in line with the findings of [23] and [25].

4.3. Bioaccumulation of heavy metals

The results of this study are similar to previous research in Pakistan, which has identified heavy metal contamination in -

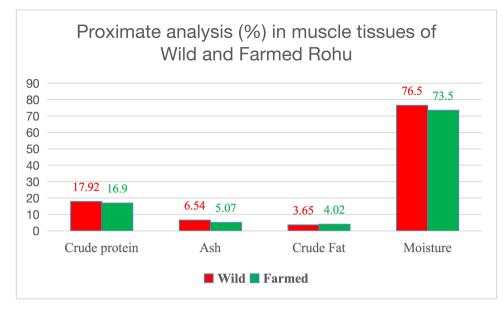


Figure 3. Proximate composition of wild and farm-raised Labeo rohita.

Table 5. Heavy metals concentrations (ppb) in muscle tissues of Wild and Farmed Rohu.

Type of fish	Cr	Ni	Cd	Pb
Wild	13.29±0.72 A	18.28±0.28 B	54.30±0.66 A	128.3±2.82 A
Farmed	16.65±0.65 B	22.19±1.80 A	36.4±2.41 B	111.4±2.16 B

Note: Cr: chromium, Ni: nickel, Cd: cadmium, Pb: lead, Values are presented as mean \pm standard deviation. Different uppercase letters denote significant differences (p < 0.05) between wild and farmed fish for each heavy metal.

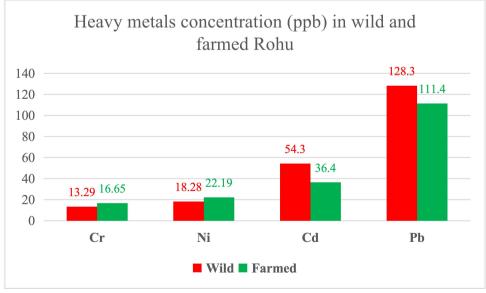


Figure 4. Heavy metals concentration (ppb) in wild and farmed Rohu.

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fish populations. However, the study is unique in that it specifically examines *Labeo rohita* and compares wild and farm-raised populations. The findings of the current study revealed a mixed trend of different metals both in the wild and farm-raised rohu. The concentration of Cr and Ni was found higher in the farmed rohu while Cd and Pb were in the wild. The authors attributed this disparity to the increased exposure of wild rohu to pollutants present in the water bodies.

The study, published in Environmental Monitoring and Assessment. provides valuable insights into the bioaccumulation of heavy metals in rohu fish and highlights the potential impact of water pollution on the contamination levels in aquatic organisms. The results revealed that both wild and farm-raised Rohu had detectable levels of heavy metals, with the wild group having significantly higher concentrations of Cd and Pb than the farmed group. [26] reported similar results in their study of the assessment and bioaccumulation of heavy metals in water, fish (wild and farmed), and associated human health risks.

The result of our findings is incompatible with [27, 28] and [29]. The findings revealed that the concentrations of two metals (Cr, Ni) were notably higher in farm-raised rohu in comparison to wild rohu. The authors attributed this disparity to the contamination of the feed given to farm-raised rohu, which was found to be contaminated with heavy metals.

By specifically examining *Labeo Rohita* and comparing wild and farm-raised populations, the study adds new information to the existing research and helps to build a more complete understanding of the extent and distribution of heavy metal contamination in fish populations in Pakistan. The results of this study can also be used to inform future research on heavy metal contamination in fish populations. For example, the findings suggest that there may be differences in heavy metal concentrations between wild and farm-raised populations, which could be further explored in future studies. Our findings are in line with the previous findings by [30,31] and [32] aimed to examine the bioaccumulation of heavy metals, namely chromium (Cr), nickel (Ni), cadmium (Cd), lead (Pb), and zinc (Zn), in wild and farmed rohu (*Labeo rohita*). The findings revealed that the concentrations of these heavy metals were significantly higher in wild rohu compared to farm-raised rohu. The fish was exposed to Cr, Pb, Ni, and Cd. The detailed results of heavy metals on fish *Labeo rohita* can be seen in Table 3. The highest mean deviation was seen in Pb of wild fish and Cd of farmed fish. The highest concentration was found in Cd and Pb and there was no significant difference between other metals Ni or Cr. Similar, studies on the determination of heavy metal load in river water so as to assess water quality have been conducted by several researchers [33,34,35,36,37].

5. Conclusion

Both in the wild and in captivity, *Labeo rohita* were examined to determine their overall proximate composition as well as the number of dressing losses and bioaccumulation of heavy metals. We found that there was a higher occurrence of dressing loss in wild Rohu (26.37%) in comparison to farmed fish. When the *Labeo rohita* is dressed, the fat contents doesn't get removed so the farm-raised Rohu could contain a higher percentage of fat than wild-caught Rohu. Since farm-raised Rohu are bred to be bigger and heavier, there is a possibility that they may generate more offal.

Data availability

The data that were analyzed in the present article are available upon justifiable request to the corresponding author.

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Author contributions

Imtisal Shahid, Remsha Bibi, Esha Khan, Sidra Bibi, Muhammad Faheem, Fatima Gulzar, and Rimsha Munawar conceived, collected, and analyzed data and wrote the manuscript. Dilawar Hussain, supervised the study design, and read, edited, and approved the final manuscript.

Conflicts of Interest

The authors declare no conflict of interest.

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