



Research Article

Examination of the Physiological, Health Status and Heavy Metals in Cultured Carp Fish (*Cyprinus carpio*)

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Abstract | Carp belongs to the Cyprinidae, which is considered one of the largest family of freshwater fish. in rivers, lakes and ponds. Aquaculture in Iraq depends on the availability of purified water, this study aimed to assessment of metals concentration (Cd, Pb, Zn, and Fe) existing in different organs, including the gills, muscles, reproductive gonads, and liver of the common carp fish as well as to evaluate the physiological and health status (Total protein, Albumin, Cholesterol, Glucose, ALT and AST) of fish. Two places were selected for the study at the aquacultures affiliated with the University of Basrah. Heavy metals were analyzed by using atomic absorption spectrometer as well as biochemical properties measured via A Humalalyzer Primus spectrophotometer. This study recorded that all values were within the recommended normal range for healthy fish. Therefore, they were considered a reference value for heavy metal levels. Therefore, these values may be useful as a tool for monitoring heavy metal pollution indicators and monitoring the health status of this fish and other related fish species.

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Introduction

Fish diversity plays a vital role in maintaining aquatic ecosystem stability and providing a significant source of high-quality nutrition for humans. Diverse fish species, including both indigenous and exotic forms, serve as important bioresources contributing to food security and livelihood sustainability (Pervaiz *et al.*, 2012; Naeem *et al.*, 2012a). Fish are widely recognized for their rich nutritional composition,

being excellent sources of high biological value proteins, essential fatty acids, and vital micronutrients that support human health (Naeem *et al.*, 2016). The proximate composition of various freshwater species such as *Oncorhynchus mykiss*, *Mystus bleekeri*, and *Notopterus notopterus* indicates that fish flesh is low in saturated fats and rich in polyunsaturated fatty acids, particularly omega-3 fatty acids, which are essential for cardiovascular and neurological health (Naeem *et al.*, 2011b; Naeem *et al.*, 2016). Moreover, fish contain

essential macro- and micro-elements including calcium, phosphorus, potassium, magnesium, zinc, and iron, which are necessary for maintaining metabolic balance and preventing nutritional deficiencies (Naeem *et al.*, 2010). Studies on the morphometric and reproductive biology of species such as *Catla catla*, *Ctenopharyngodon idella*, and *Tor macrolepis* highlight their ecological and economic importance in both aquaculture and wild ecosystems of Pakistan (Naeem *et al.*, 2005; Naeem *et al.*, 2011a; Khan *et al.*, 1992). The relationship between fish body composition, size, and condition factor further reflects their adaptive responses to environmental conditions, emphasizing the role of fish diversity in sustaining healthy aquatic food webs (Naeem *et al.*, 1992; Naeem *et al.*, 2012b).

Fish are an important food resource and play a key role in many food chains because they contain high levels of important fats, proteins, omega-3 fatty acids, amino acids, vitamins, and vital minerals such as copper, zinc, calcium, and iron (Naeem *et al.*, 2011b; Naeem *et al.*, 2016). They are known for their benefits in supporting general health, especially their cardiovascular benefits, as it helps in preventing and treating cardiovascular complications, in addition to reducing neurological, immune and reproductive symptoms. (Rodrigues *et al.*, 2024). Many Asian and European countries have been interested in fish farming, especially common carp (*Cyprinus carpio*), because it is a significant aquatic species that has been able to benefit from the available nutrients from aerobically decomposed organic matter found in bottom sediments, and it has the ability to tolerate the low density of dissolved oxygen and high density of carbon dioxide in the water compared to other carp species (Rahman, 2015). The culture has been widely successful in subtropical and tropical regions, with carp has a daily growth rate of 2-4% of body weight, consequently the fish weighs 0.6 to 1.0 kg in one season. (FAO 2004).

Carp belong to the Cyprinidae, a freshwater fish found in rivers, lakes, and ponds. Aquaculture in Iraq has relied heavily on abundant freshwater, suitable locations, and good soil leading to its widespread distribution in central and southern Iraq. There are public and private aquaculture farms. Carp has become more popular for aquaculture in Iraq (FAO 2009; FAO 2017).

In general, the environment was affected by climatic

conditions, especially the environment of the Arabian Gulf and the Shatt al-Arab River (Yaseen *et al.*, 2024). Negative changes in the surrounding environment have a profound impact on physiological activities, this can be due to blood imbalances, which reduces resistance to disease, and also affects metabolism and reproductive performance, thus altering the health of the entire body. and thus, biochemical studies of the blood and serum of farmed fish are important to monitor their health during farming (Batool *et al.*, 2024a). These limits are thoroughly linked to the organism's response as they affect the survival, reproduction and growth of fish (Casanovas *et al.*, 2021). Therefore, this study aimed to assessment of metals concentration (Cd, Pb, Zn, and Fe) examined in four important organs of these fish, which are the gills, muscles, reproductive gonads, and liver. as well as to evaluate the physiological and health status (Total protein, Albumin, Cholesterol, Glucose, ALT and AST) of common carp fish at the aquacultures affiliated with the University of Basra.

Materials and Methods

Two stations were chosen, Figure 1, the first of which was represented by the aquaculture ponds of the Marine Sciences Center and the aquaculture basins of the Al-Hartha Research Station. These locations were selected for the reason that they are considered field stations for scientific research, in addition to their economic importance for fish marketing and environmental sustainability.

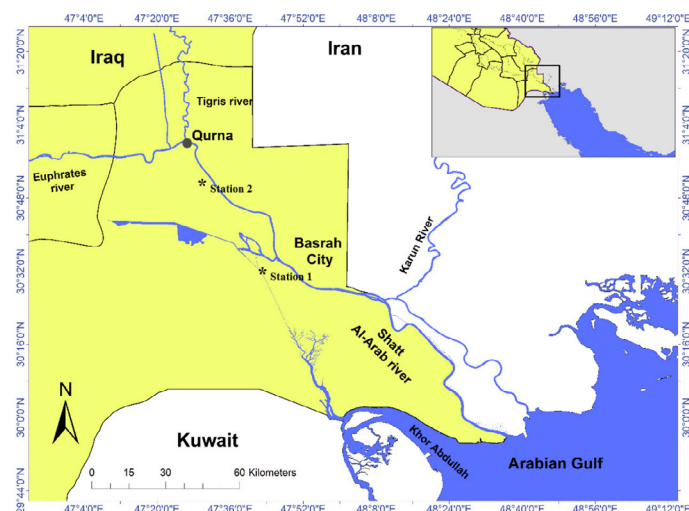


Figure 1: Map of the stations used in the study area, the first station is the Marine Sciences Center station and the second is the Al-Hartha Research Station / University of Basra.

Measurement of heavy metals

Fish samples were collected from the culture tanks and transported to the laboratory for freezing. They were then thawed at room temperature. The fish were dissected to isolate the organs to be examined and then washed with deionized water., dried at 105°C overnight, and crushed. 1 g of muscle, gills, and gonads were taken, while the dry weight of the liver was 0.1 g. Samples were digested in an acidic medium of nitric acid (65%) and hydrogen peroxide (35%) (6-1 ml) (Aldoghachi, and Alabdul Aziz, 2023). The solution was made up to 50 ml with deionized water. For water samples, 100 ml were filtered using Millipore filter paper, digested by adding 6 ml of concentrated nitric acid, dried by heating to 10 ml and completed with 50 ml of deionized water. Samples were analyzed using a GBC Savant AA Ver3.02 atomic absorption spectrometer (mg/kg dry weight).

Biochemical investigations

Fish feeding was discontinued within 24 hours until blood samples were taken. Five replicate fish samples, averaging 225±4 g, were taken and immediately anesthetized in ice water. A 3 ml syringe was used to withdraw blood from the caudal vein as described by Zang *et al.* (2015) and collected in 5 ml glass test tubes and allowed to reach room temperature (21°C). Serum was separated from the clot using a centrifuge at 3,000 rpm for 10 minutes. The serum was kept

frozen at -20°C until further investigations. Glucose Measurement via Enzymatic Colorimetric Method mentioned in Afiyah *et al.*, (2023) and total proteins were determined by Biuret Method according to Shaalan and Sayed, (2025). ALT and AST activities in fish serum were measured with the ultraviolet assay technique as described in Rahimikia (2017). A Humalyzer Primus spectrophotometer was used to measure absorbance at a wavelength of 546 nm, measured in U/L.

Data analysis

The collected data were analyzed by SPSS (version 22) using one-way analysis of variance (ANOVA) at a 0.05 level of accuracy.

Results and Discussion

The study results showed that iron (Fe) recorded the highest concentration (3.18 and 0.59 mg/l) in the water used at Al-Hartha Research Station (Station 2) and the Marine Science Center Station (Station 1), respectively. Zinc (Zn) followed, with the highest concentration recorded at Station 2, 0.22 mg/l, while it was 0.031 mg/l at Station 1. Lead (Pb) was recorded in the water used at Station 2 only, at 0.202 mg/l. Cadmium was not recorded at either of the stations studied (Figure 2).

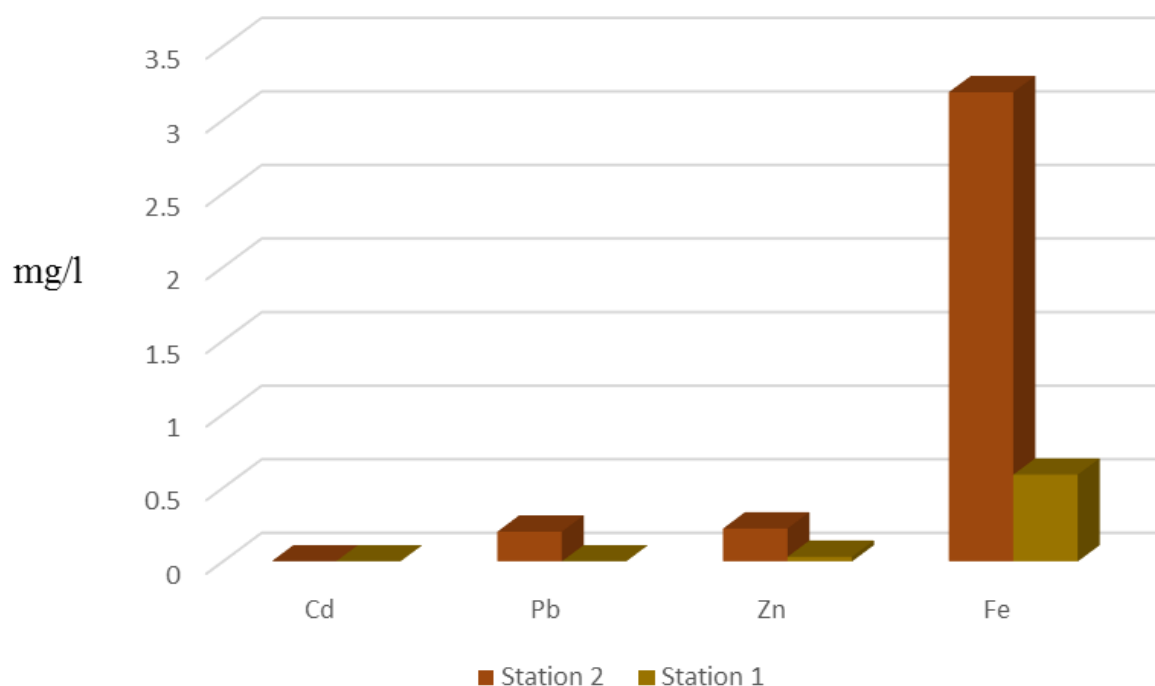


Figure 2: Measuring heavy elements in the water used in the aquaculture of the Marine Sciences Center (Station 1) and Al-Hartha Research Station (Station 2) / affiliated with the University of Basra.

Canadian water quality guidelines (mg/l) CCME (2025): Cd < 0.005, Pb < 0.05, Zn < 2.00 and Fe < 2.00

Data of the current study (Tables 1 and 2) displayed that all studied tissues, iron had the highest concentration among the tested elements. (Muscles, gills, liver, and female gonads). The gill tissues showed accumulation rates of 15.204 and 54.01 mg/kg for the first and second stations, while cadmium showed a very low concentration and was not detected at the second station, where the order of accumulation was (Fe > Zn > Pb > Cd). In general, many previous studies have demonstrated that fish organ tissues have the ability to concentrate and accumulate metals. (Aldoghachi and Altamimi, 2021 and Batool *et al.*, 2024b).

The liver recorded the highest accumulation of non-essential elements (cadmium 0.0047 mg/kg and lead 0.186 mg/kg) at the first station, these concentrations are less than permissible limits by EC 2006 while in a study FAO (2009) on carp fish in the artificial lake in Baghdad, which proved that heavy elements have the aptitude to concentrate in the gills and liver, their results showed that lead values in whole fish reached a concentration of (6.834 mg/kg), while cadmium values reached (0.567 mg/kg) in places near to factory waste. Their data designated that lead and cadmium levels were higher than permitted concentrations. The current study referred to the second station had the highest levels for zinc 5.73 mg/kg and iron 53.81 mg/kg. Zinc is an essential element recorded at normal concentrations within the permissible limits. The highest concentration was in the gill tissues, which was 5.80 mg/l in the second station, while the highest concentration in the liver was 3.1798 mg/l in the first station (Tables 1 and 2). This is mainly due to its presence in water used for aquaculture, which amounted to (0.031 mg/l) (Figure 2). As for the muscles, concentrations were also recorded within the permissible limits, which were (0.433 and 0.5) for the first and second stations. This is agreement with the data of FAO (2017) which reported that the value of zinc ranged between (0.21-100 mg/kg) recorded in the muscles of many carps living in Iraqi rivers in different geographical areas. Zinc is a cofactor for many biological activities, but increasing its concentration converts extra dangerous and causes undesirable impacts on fish in relations of accumulation leading to changes in enzymatic activity (Gashkina, 2024). All element concentrations were within the permissible levels (Tables 1 and 2).

Table 1: Means of metals concentrations (mg/kg) in tissues of *Cyprinus carpio* at the aquaculture ponds of Marine Science Center

Sample	Cd	Zn	Pb	Fe
Gills	0.000567	4.195	0.093	15.20433
Muscles	ND	0.433	0.080	15.811
Female gonads	ND	0.756	ND	8.228
Liver	0.0047	3.179	0.1865	11.5585
Acceptable limit ¹	0.05 for muscle 0.5 for liver	< 5.00	0.3 for fish	--

¹Permissible limit by European Communities (2006)

ND: none detectable

Table 2: Means of metals concentrations (mg/kg) in tissues of *Cyprinus carpio* at the aquaculture ponds of Al-Harthia station.

Sample	Cd	Zn	Pb	Fe
Gills	ND	5.80	0.08	54.01
Muscles	ND	0.50	0.016	9.01
Female gonads	ND	0.55	0.014	4.41
Liver	ND	1.24	0.19	6.02
Acceptable limit ¹	0.05 for muscle 0.5 for liver	< 5.00	0.3 for fish	--

¹Acceptable limit by european communities (2006)

ND: none detectable

Serum biochemical composition

The serum biochemical parameters of carp reared in earthen ponds (T. protein, albumin, cholesterol, glucose, ALT, and AST) were recorded in Table 3. The data showed no significant differences ($P > 0.05$) in the levels of albumin, protein, cholesterol, glucose and ALT in carp for both rearing ponds. However, the biochemical level of AST obtained exhibited a significant difference ($P < 0.05$). The data detected that the first station had the highest AST content of 75.54 ± 3.50 , while the second station had the lowest content of 60.74 ± 3.02 .

Table 3: Serum biochemical parameters of *Cyprinus carpio* from the two fish enclosures Earthen Ponds

Parameters	Earthen Pond 1	Earthen Pond 2
Total Protein (g/dl)	6.14 ± 0.09^a	4.70 ± 0.34^a
Albumin (g/dl)	2.12 ± 0.9^b	1.49 ± 0.04^b
Glucose (mg/dl)	129.82 ± 5.32^c	118.32 ± 1.15^c
Cholesterol	100.85 ± 2.3^d	95.50 ± 0.70^d
ALT (U/L)	70.30 ± 2.20^f	61.0 ± 0.15^f
AST (U/L)	75.54 ± 3.50^g	60.74 ± 3.02^h

Values with the same letter have no significant differences ($P > 0.05$)

In general, fish proteins play an important role in maintaining blood glucose levels and are considered a major energy source. Serum biochemical parameters vary from one species to another and can be affected by numerous abiotic and biotic components. For example, a high-protein diet increases total protein, which in turn reduces immune indicators. Similarly, the sex and age of fish, as well as the seasonal pattern, affect liver enzymes, proteins, and glucose (Fazio, 2019). Changing environmental conditions, such as temperature, salinity, oxygen deficiency, and increased pollution, cause stress that leads to significant changes in biochemical parameters (AlKatrani *et al.*, 2018; Owen *et al.*, 2025).

Total serum protein rises with each stress, especially starvation. In this study, the total protein concentration varied slightly between the two culture stations. The total serum protein of carp was recorded as (6.14-4.70 g/dl) for the two stations, respectively. The level at station 2 was consistent with the normal range of healthy fish, while station 1 was slightly above the normal limits, which was higher than the value recorded by Syawal *et al.* (2023) of 4.5 g/dl in grass-fed carp. This is attributed to the salinity changes in the culture water, which was 2.2 g/L compared to the second station, which was 0.5 g/L which measured in preliminary test. This is consistent with what was indicated by Alfonso *et al.* (2024) that the reason for the increase in total protein in fish blood increases with a noticeable change in water temperature or salinity concentration, which leads to an increase in protein as part of the response to osmoregulatory stress.

Albumin contributes significantly to osmotic regulation through water balance between blood and tissues and helps the general metabolism of fish by transporting vital substances such as free fatty acids and ions. It also contributes to acid-base balance. Albumin is strongly affected by diet and environmental conditions in addition to environmental or physiological stress. The albumin content recorded in the samples of the studied stations did not differ significantly and was within the normal limits recorded by Owen *et al.* (2025) between 1.3 - 2.6 g / deciliter.

Glucose is an important energy source for organisms. It is preserved in the complex system of endogenous production through the breakdown of dietary carbohydrates. However, the glucose level was within

the normal range of 50-150 mg/dl (Afiah *et al.*, 2023). Temperature affects blood sugar levels, and a very slight difference in glucose concentration between the two stations was observed, attributed to differences in activity, environmental habitats, and culture conditions. This was confirmed by Xi *et al.* (2023) in their study on zebrafish (*Danio rerio*). Excessive dietary carbohydrate intake (30%-50%) leads to metabolic disturbances in fish, particularly increased glucose levels, which then leads to the accumulation of lipids in the blood and subsequent liver damage. This is in addition to processes associated with glycolipid metabolism and inflammation in fish, which they attributed to increased expression of genes associated with glycolysis in the liver and process of inhibited genes associated with gluconeogenesis in the fish liver.

The values of the current research presented that the cholesterol level in carp fish was (95-100 mg/dl) (Table 3). This is within the average normal level of cholesterol in fish blood of 100-140 mg/dl. Cholesterol level is affected by the temperature of the water used in fish farming as well as the level of fat nutrition. In *Ctenopharyngodon idella* fish, studied by Kovacic *et al.* (2023) recorded 145 mg/dl during the summer at moderate fat nutrition levels, while cholesterol level doubled during the autumn. Exposure to heavy metals disrupts liver and kidney tissue, leading to increased cholesterol levels as part of the environmental stress response. A study by Guo *et al.* (2022) on carp exposed to environmental cadmium showed increased blood cholesterol levels, which leads to the occurrence of non-alcoholic fatty liver disease, while Anjum *et al.* (2024) found that grass carp (*Ctenopharyngodon idella*) were exposed to high concentration (38 mg l⁻¹) of Zinc ions contaminants, resulting in a decrease in their cholesterol levels from 378 to 262 mg/dl. This decrease was attributed to the consumption of fat as an energy source in response to stress, which reduced their cholesterol levels.

As for the activity of the ALT enzyme, no significant differences were recorded between the two stations, and the highest value was (70.30±2.20 U/L), while the AST enzyme recorded few significant differences between the two studied stations, and the highest value was (75.54±3.50 U/L), which was less than what was recorded by Aldoghachi (2022) after exposing fish to the pollutant cadmium for 48 hours (ALT 223.6±3.03 U/L) and (AST 240.5±2.15 U/L).

Increased AST and ALT activity is due to cell membrane disruption, which leads to these enzymes leak from the cells into the interstitial fluid and then into the bloodstream. These enzymes are called “leakage enzymes.” They have been used primarily as biomarkers of liver damage. ALT is more specific and is considered the gold standard for liver dysfunction than elevated AST activity (Shahsavani *et al.*, 2010). Elevated AST activity indicates increased amino acid metabolism, stress, and dysfunction of liver or kidney (Matsche, and Gibbons, 2012).

To predict and diagnose the health of fish reared in aquaculture and research, biochemical tests are accepted as a biomarker of exposure to toxic pollutants. These limits assistance determine the degree of poisonousness and the mechanisms by which certain toxic factors affect living fish. In a review by Bojarski *et al.* (2025), they indicated that total protein, albumin, bilirubin, triglycerides, and cholesterol, in addition to ALT and AST, are biomarkers of general physiological stress and liver dysfunction, which are significantly affected by toxic pollutants, including exposure to heavy metals, depending on the concentration and exposure duration, these indicators have the potential to increase or decrease in their levels in fish blood.

Conclusions and Recommendations

All values recorded in this research were within the recommended standard extent for healthy fish. Therefore, they were considered a reference value for heavy metal levels in various fish tissues, as well as for designated biochemical limits in the blood serum of fish carp reared in earthen ponds. Therefore, these values may be useful as a tool for monitoring heavy metal pollution indicators and monitoring the health status of this fish and other related fish species.

Ethics approval

Live fish were handled under the supervision of the Ethical Committee of the Marine Science Center, University of Basra.

Consent for publication

I am (Mohammed A. Jasim Aldoghachi) give my consent for information about ourself to be published in Journal.

Availability of data and material

Yes, all data and materials are available on request.

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Novelty Statement

The research provides health indicators for farmed carp and monitors heavy metal pollution levels and their physiological impact to assess the aquaculture environment and the fish used for human consumption.

Authors' Contribution

Mohammed A. Jasim Aldoghachi: Fish work and inspection as well as wrote the text of the manuscript and analyzed the data. Format the manuscript and the figures as well as, Collected and examined the raw data of the manuscript.

Dunya A. Hussein: Collecting samples, analyzing and digesting fish water and tissue samples, water quality tests, in addition to contributing to statistical analysis.

Generative AI or AI assisted technology statement

The authors declare that no generative AI or AI-assisted technology was used in the writing or editing of this manuscript.

Conflict of interest

The authors declare that there is no conflict of interest among the author of the manuscript.

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