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Evaluation of Some Heavy Metal Levels in Tissue of Fish Collected from Coasts of Susa Region, Libya

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ABSTRACT
This study aims to estimate the levels of various heavy metals (Zn, Cu, Pb, Co, Cd, and Cr) in different tissues of fish samples collected from various locations along the Libyan coast. The fish species included in the study were Siganus rivulatus, Mugil spp, and Mullus barbatus. The metal concentrations in the studied fish samples were found to follow this order of abundance: Zn> Pb> Cu>Co. The detected metal concentrations varied as follows: (56.74 - 141.52 ppm), (29.74 -101.84 ppm), (51.33-111 ppm), (3.19-5.03 ppm), (1.60-1.93 ppm), and (1.26-33.60 ppm). These variations in metal concentrations are attributed to differences in the activities and metal sources in the various sampling locations. The accumulation of metals in the fish samples suggests that these species could potentially serve as bioindicators of heavy metal pollution.

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INTRODUCTION

Heavy metal pollution primarily originates from various sources along beaches worldwide, often without treatment [1]. It has been noted that the concentration of heavy metals in the water column of most marine samples depends on numerous factors, and the Water Quality Index (WQI) is commonly used to assess the distribution and presence of pollutants in water samples. Studies have reported that aquatic environments are primarily polluted by heavy metals such as Cd, Fe, Pb, Hg, Cu, Cr, and Zn [2]. Additionally, other research has indicated that the presence of heavy metals in marine samples is largely attributed to human activities [3].

In many studies, the concentrations of heavy metals like Pb, Zn, Cu, and Cd were found to be lower in sediment samples compared to fish. The contamination of aquatic ecosystems with heavy metals, above natural background levels, has become a significant concern among researchers. These metals can accumulate in aquatic organisms and enter the food chain, potentially affecting various tissues in the human body and posing risks to human health when contamination levels are toxic. The primary objective of this study is to assess the metal concentrations in fish samples collected from the coastal region of Susa Beach in Libya.

METHODS

Area of study

Susa, located on the southern coast of the eastern Mediterranean in northern Libya, is one of the most significant fishing centers in the country. It supplies approximately 25% of the total seafood for the Green Mountain region. The city, with a population of around 40,000, is bordered to the south by a chain of rocky hills and has expanded along the coastline. Susa's fisheries are a primary source of fish for nearby large cities such as El-Bayda, Shahhat, and other local markets in Libya. However, the quality of its coastal waters has deteriorated over time, largely due to the inflow of untreated domestic, industrial, and agricultural waste, which has led to a decline in fishing. In this study, fish are considered bio-indicators of contamination.

Samples

Three different fish species were selected for this study: Mugil cephalus, Siganus rivulatus, and Mullus. These fish were collected from the coast of Susa city and stored frozen in the laboratory until further analysis. Different tissues, including muscles, gills, and liver, were separated from the samples to assess the bioaccumulation of heavy metals in each part. The heavy metals were measured following the Association of Official Analytical Chemists (AOAC) method. Each tissue was digested using concentrated HNO_3, filtered, and then diluted to a final volume of 50 mL. The

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heavy metals were determined using atomic absorption spectroscopy at the central laboratory of the institute of aquatic research in Alexandria, Egypt [4-15]

RESULTS

The concentrations of selected metals in the studied samples were illustrated in Tables 1-3.

The concentrations of zinc in the three organs (gills, liver, and muscles) of the selected fish species from Susa city are presented in Tables 1-3. For Siganus rivulatus, the zinc concentrations were 46.18 µg/g in the gills, 60.55 µg/g in the liver, and 20.44 µg/g in the muscles. In Mugi1 sp., fish samples, the zinc concentrations were 50.83 µg/g in the gills, 76.19 µg/g in the liver, and 14.50 ug/g in the muscles, with a total concentration of 141.50 µg/g. In Mullus barbatus, the zinc concentrations were 15.87 µg/g in the gills, 32.54 µg/g in the liver, and 8.33 µg/g in the muscles. The results clearly show that the highest concentrations of zinc were recorded in the liver for all the studied fish species, with the following order of concentration: liver>gills> muscles. The highest zinc levels were found in Mugil sp., relative to the other species. This herbivorous species primarily feeds on green, red, and brown algae, as well as detitus.

The concentration of copper (Cu) in different fish species collected from Susa is presented in Tables 1-3. The results reveal that the liver accumulates the highest amount of copper among the studied organs, regardless of species. This finding aligns with previous studies, which reported that copper residues in the liver of fish, such as eel, whiting, and flounder from the Medway Estuary (UK), were 5-60 times greater than in muscle. The elevated liver content observed in Mugil sp. can be attributed to its omnivorous diet, whereas other species, like Siganus rivulatus, recorded high concentrations in both the liver and gills due to their herbivorous feeding habits. Among all species, the gills rank as the second-highest organ for copper accumulation. However, unlike soft tissues (liver and muscles), gills appear to be less influenced by species type. Notably, the copper levels in gills and muscles are relatively similar across species. Hard tissues, such as gills, accumulate less copper compared to soft tissues, indicating that diet is the primary pathway for copper uptake. This conclusion is supported by the significantly higher copper concentrations recorded in the liver compared to the gills.

The distribution pattern of lead in the selected organs follows the order: liver > gills > muscle for Mugil sp., Siganus rivulatus, and Mullus barbatus. Although gills, considered hard tissues, show high lead content, it remains lower than the levels observed in the liver. The elevated lead levels in gills may be attributed to particulate or organic lead adsorbed onto the gill surface, as suggested by the NRCC. Additionally, the lower pH on the gill surface, caused by respired CO2, can dissolve lead into a soluble form that diffuses into gill tissues. This suggests that lead tends to accumulate more in hard tissues (like gills) than in soft tissues. Among the studied species, the lead concentration in gills decreases in the order: Mugilsp. > Siganus rivulatus > Mullus barbatus. Lead is also found in the brain and liver due to its affinity for thiol and phosphate-containing ligands.

The concentration of cadmium (Cd) in the fish species collected from the investigated area is presented in Tables 1-3. The distribution pattern of cadmium in the studied fish follows the order: muscles <gills < liver. This indicates that the liver accumulates the highest levels of cadmium in most species, except for Mugil sp., where the muscle recorded a higher concentration than the gills.

The concentrations of chromium in the three organs of selected fish species from Susa city are presented in Tables 1-3. In Siganutus rivulatus, the chromium levels were 11.33 ug/g in the gills, 19.50 µg/g in the liver, and 2.77 µg/g in the muscles. For Mugil sp., the chromium concentrations measured were 1.60 µg/g in the gills, 2.91 µg/g in the liver, and 0.33 µg/g in the muscles, with a total concentration of 4.48 ug/g. In Mullus barbatus, chromium levels were recorded as 0.06 µg/g in the gills, 0.76 µg/g in the liver, and 0.44 µg/g in the muscles. Correlation coefficients between individual metals showed a strong positive relationship(r>0.9) across all metals and tissue types (Tables 4-6).

Table 1. Concentrations (ppm) of heavy metals in different tissues of Siganutus rivulatus.

Tissue Metal	Gills	Liver	Muscles	Total
Zn	46.18	60.55	20.40	127.130
Cu	9.92	15.77	4.05	29.740
Pb	18.44	36.92	5.82	61.180
Co	0.30	1.11	0.52	1.930
Cd	1.60	2.48	0.95	5.030
Cr	11.33	19.50	2.77	33.600

Table 2. The concentrations (ppm) of heavy metals in Mugil sp. tissues.

Tissue Metal	Gills	Liver	Muscles	Total
Zn	50.83	76.19	14.50	141.520
Cu	12.44	80.70	8.70	101.840
Pb	31.19	60.48	19.33	111.000
Co	0.95	0.66	0.21	1.820
Cd	0.82	1.61	1.50	3.930
Cr	1.60	2.91	0.33	4.840

Table 3. The concentrations of heavy metals in Mullus barbatus tissues (ppm)

Tissue Metal	Gills	Liver	Muscles	Total
Zn	15.87	32.54	8.33	56.740
Cu	11.90	18.54	6.55	36.990
Pb	12.23	30.44	8.66	51.330
Co	0.11	1.45	0.09	1.650
Cd	0.86	1.56	0.77	3.190
Cr	0.06	0.76	0.44	1.260

Table 4. Correlation coefficients between metals in the Siganuts rivulatus Fishes

Tissue Metal	Zn	Cu	Pb	Co	Cd	Cr
Zn	1					
Cu	0.995	1				
Pb	0.976	0.993	1			
Co	0.921	0.939	0.942	1		
Cd	0.996	0.995	0.980	0.953	1	
Cr	0.983	0.995	0.998	0.924	0.981	1

Table 5. Correlation coefficients between tissues in the Siganuts rivulatus fishes

Tissues	Gills	Liver	Muscles	Total
Gills	1			
Liver	0.977	1		
Muscles	0.986	0.933	1	
Total	0.997	0.991	0.972	1

Table 6. Correlation coefficients between metals in the Mugil sp fishes

Tissues	Zn	Cu	Pb	Co	Cd	Cr
Zn	1					
Cu	0.907	1				
Pb	0.987	0.936	1			
Co	0.932	0.693	0.885	1		
Cd	0.855	0.794	0.915	0.781	1	
Cr	0.995	0.940	0.984	0.895	0.828	1

Table 7. Comparison of the ppm of metals in different fish

Metals	Zn	Cu	Pb	Co	Cd	Cr
Siganus rivulatus Fishes	56.74	36.99	51.33	1.65	3.19	1.26
Mugil sp Fishes	141.52	101.84	111	1.82	3.93	4.84
Mullus barbatus Fishes	127.13	29.74	61.18	1.93	5.03	33.6

DISCUSSION

It is well known that algae accumulate higher levels of heavy metals compared to other organisms, such as crustaceans, mollusks, and smaller fish, which are the primary food source for other fish species. This finding aligns with a previous study [16], which reported that feeding habits and environmental conditions influence the levels of heavy metals in different species. According to a study stated that the lower zinc concentration in muscle tissue [17], compared to the liver and gills, is attributed to a decrease in zinc levels in the muscles rather than an increase in the liver and gills. The zinc concentrations in the gills exhibit a different pattern from the liver across species, suggesting that zinc levels in the gills are influenced more by species-specific factors and surrounding water conditions (such as particulate and dissolved metal forms, pH, salinity, temperature, and dissolved oxygen) than by the fish's feeding habits. From the discussion, it is evident that the zinc levels in soft tissues (liver, gills, and muscles) are primarily influenced by the feeding habits of the species, rather than environmental parameters.

In contrast, hard tissues like bones appear to be dependent solely on the food source, while the zinc levels in gills are influenced by both food habits and environmental factors. When comparing the current findings with those reported by a previous study [18], where zinc levels in fish tissues ranged from 205 to 400 µg/g, a notable difference is observed. However, the present study aligns more closely with the results from fish samples collected from Derna city, where zinc concentrations ranged from 56.74 to 141.52 µg/g. The World Health Organization [19] recommends an allowable zinc concentration of 40 µg/g in fish. In this study, the total zinc concentrations in the fish samples exceed this allowable limit, except during non-feeding periods.

Fish typically obtain the majority of zinc from their diet rather than from water. When contaminated food is absent, the rate of zinc uptake depends on the zinc concentration in water and the exposure duration. Furthermore, the presence of organic and inorganic chelates in the water may significantly reduce zinc absorption by fish. Temperature stress and low oxygen levels likely increase the susceptibility of many fish species to zinc. Residue levels in freshwater and marine fish are generally much lower than those found in algae and invertebrates, indicating that zinc accumulation in many fish species is minimal.

Previous studies [20] reported similar findings, noting that digestive tract copper concentrations exceeded those in gills. The distribution of copper in the studied organs follows the order: liver/>gill>muscle. The low copper concentration in muscle tissue has been widely documented, although Mugil sp. recorded the highest muscle copper levels among the studied species. According to WHO guidelines, the allowable copper concentration for human consumption is 30 µg/g wet weight (ww).

While most studied tissues fall within acceptable safety ranges, the liver of Mugil sp. exceeded this limit significantly, with a recorded concentration of 80.70 µg/g. Contaminated food is likely a more critical source of copper in fish than water. Consequently, copper burdens in fish cannot be consistently correlated with water pollution levels. Experimental studies have shown that copper uptake is inversely related to the presence of chelators and inorganic ions in water and directly related to exposure duration and concentration. Marine fish generally exhibit higher copper residues than freshwater species due to the increased bioavailability of chloro-Cu(II) species. Although plankton, fish, and shellfish from contaminated areas contain higher copper levels than those from uncontaminated regions, copper does not typically bioaccumulate in food chains.

The present study indicates that the liver accumulates the highest levels of lead (Pb)among the studied fish organs. This observation is consistent with findings by a previous study [21], which noted that lead and calcium share similarities in deposition and mobilization in the liver. Lead can remain immobilized for years, often causing metabolic disturbances to go undetected. Under normal conditions, over 90% of the retained lead in the body is stored in the skeleton. This interaction inhibits hem biosynthesis and affects the membrane permeability of kidney, liver, and brain cells, potentially leading to reduced function or total breakdown of these tissues. As a cumulative poison, lead poses significant risks. Conversely, muscle tissue showed the lowest lead concentrations in all species, likely due to the reduced rate of binding to SH groups and the restricted movement of lead salts across cell membranes. The WHO recommends a maximum lead intake of 3 mg per person per week, with children and infants (more vulnerable to lead poisoning) advised to limit intake to less than 1 mg per week. In urban areas with high lead inputs from motor vehicles, children's intelligence levels may be negatively impacted.

Australia's National and Medical Research Council (NHMRC) specifies that lead concentrations in edible fish should not exceed 2 ug/g. However, the study's results reveal levels exceeding these limits, raising concerns for human consumption. Although lead accumulation in marine and freshwater species is typically minimal, extreme pollution may pose risks. Feeding habits generally show no correlation with lead residues. Bernhard and Zattera noted that lower trophic levels like phytoplankton, zooplankton, and macrophytes can enrich lead concentrations considerably compared to seawater. However, further enrichment at higher trophic levels is negligible.

Lead concentrations in crustaceans, mollusks, and fish from uncontaminated areas are comparable to those at the first trophic level. Compared to other metals, lead in marine environments is less toxic; for instance, concentrations up to 0.8 ppm of lead nitrate may even enhance diatom growth due to nitrate's nutrient effects. Nevertheless, lead nitrate inhibits several major enzymes, with the decline in activity proportionate to the lead concentration and exposure duration, this finding underscores the toxicological implications of lead exposure on aquatic species and the broader ecosystem

These findings align with the WHO and IPCS Environmental Health Criteria for cadmium, which state that Cd is stored in various tissues, with the liver and kidney being the primary sites of accumulation in aquatic organisms. Additionally, significant amounts of Cd can accumulate in gills and exoskeleton. Similar results have been reported by Heghazi [22] for fish species collected from the Red Sea. In Mugil sp., the concentration of cadmium in the gills was comparable to that in the liver, suggesting that Cd uptake occurs through both the gills and the food chain. The higher cadmium levels observed in Siganus rivulatus compared to other studied species may be attributed to its dietary preference for algae, which are known to contain elevated levels of cadmium. There was minimal variation in cadmium concentrations between the brain and muscle across all collected fish. Although cadmium is not an essential element for any organism, it has been observed to enhance phytoplankton photosynthesis and growth at concentrations up to 100 ppm. This is likely due to its association with phosphates, which are absorbed by phytoplankton.

It was noted that [23], despite being non-essential, cadmium is accumulated by marine animals through pathways similar to those used for essential metals. Davies et al. found that the dominant route of cadmium accumulation in crustaceans is through food sources. In aquatic organisms, cadmium uptake occurs via two primary pathways: dietary intake and absorption through body surfaces such as cell membranes, epithelia, and skin.

The results indicate that chromium accumulates most significantly in the liver across all studied species, following the pattern liver >gills>muscles. According to the Western Australian Food and Drug Regulation, the permissible chromium concentration is 5.5 ppm. All the fish species studied exhibited chromium levels in their edible tissues well below this threshold. Heavy metals, particularly lead, were found to accumulate more in the liver than in other tissues, and were highest in Mugil sp. among the studied species

CONCLUSION

This study identified the presence of various heavy metals in different fish tissues, including the liver, gills, and muscles. The most prominent metals detected were zinc (Zn), lead (Pb), and copper (Cu), along with trace amounts of cadmium (Cd), cobalt (Co), and chromium (Cr). A strong correlation among the measured metals suggests they originate primarily from common sources.

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