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Bioaccumulation of heavy metals (Cadmium and Lead) in liver, gills and muscle of sea-water fish *Mugil cephalus* from Benghazi lake and Qaminus beach, Libya.

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Highlights

- **This study aims to determine the possibility of using fish as an indicator of marine pollution.**
- **Liver, gills and muscles were used to determine the bioaccumulation of heavy metals.**
- **From this study, it is possible to use fish organs to determine the presence of marine pollution especially the organs that are sensitive to any environmental changes.**

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ABSTRACT

The concentrations of heavy metals (Pb and Cd) were measured in the liver, gills and muscles of one hundred and sixty seawater fish of *Mugil cephalus* collected from two different areas (80 fish from Benghazi Lake and 80 fish from Qaminus beach) in the east of Libya. The levels of heavy metals diverse significantly between fish groups and organs (liver, gills and muscle). Gills always possessed the highest concentrations of two metals followed by liver and muscle. In all studied fish, the gills and livers were the target organs for lead and cadmium accumulation and the high accumulation of lead and cadmium in the gills and liver is likely linked to the roles of organs. Different groups of *Mugil cephalus* showed variations in metal concentrations. These concentrations of lead and cadmium in tissues were higher in groups collected from Benghazi Lake than in groups from Qaminus Beach. These differences among the two groups could be recognized as untreated pollutants that drifted directly to Benghazi Lake.

1. Introduction

Toxic waste from the aquatic ecosystem has advanced into severe health suffering in recent years. Furthermore, marine ecosystems are exposed to several kinds of toxins that are usually released from sewage, pesticide plants, and drainage from agricultural areas and wastes of domestic. These pollutants resulted in severe damage to freshwater and seawater organisms (Karbassi *et al.,* 2008). Pollution of seawater by chemical toxins has befallen one of the most vital seawater complications in recent years. The unlimited increase in the use of heavy metals over the past few decades has certainly resulted in an increased metallic substances change in the water environment (Yang *et al.,* 2021). The heavy metals are of actual concern due to their extended result and the range of concentration stirred toxic poorly motive to the sea life types. Industrial litters form the main basis of heavy metal pollution in natural water (Kadirvelu *et al.,* 2001). Increased discharge of heavy metals into water locations might expose water animals to damaging ranks of metals (Sorogy and Attiah, 2015; Shetaia *et al.,* 2023). In addition, metal bioaccumulation in marine animals depends on the total amount of heavy metals, the bioavailability of each metal in the seawater environment and the way of uptake, as well as their storage and excretion by the sea water animals (Eldurssi *et al.,* 2022). Fishes are regularly exposed to highly contaminated water, which leads to different changes ranging from biochemical alterations in single cells to changes in the whole body of fish. Furthermore, heavy metal toxicity can result in damaged or reduced mental, lower energy levels, and damage to the gills, liver, muscles and other vital organs of the fish. Heavy metals enter fish through water, skin, gills and food then to the blood and storage in the liver (Klavins *et al.,* 2009; Al-Ghanim *et al.,* 2016). Fish participate in a key function in the ecosystems since they are at the peak of the food chain. Furthermore, there are several advantages to using fish as an indicator of heavy metal in seawater, for example, fish are ideal models for addressing questions about marine pollution because they are totally depending on their surrounding environment for survival and are very sensitive to the pollution of the aquatic ecosystem (Goldstien and Dewoese, 1999; Klavins *et al.,* 2009). Moreover, fish are easily bred and raised in a laboratory, inexpensive to purchase and most fishes are economically important (Padmini *et al.,* 2004; Mohboob *et al.,* 2009).

This study aimed to detect the rate of heavy metals (lead and cadmium) in the liver, gills and muscles of *Mugil cephalus*, and to compare the lead and cadmium in the liver, gills and muscles of *Mugil cephalus* collected from Benghazi Lake and *Mugil cephalus* collected from Qaminis beach (unpolluted area).

2. Materials and methods

Approximately 80 specimens of *Mugil cephalus* were collected from Benghazi Lake (the 23rd July Lake) and 80 specimens were collected from Qaminis Beach by commercial fishing boat, using gill nets with a 40 mm stretched mesh. The specimens were directly kept in ice on board and transferred to the laboratory Department of Histopathology, Benghazi University. The total length (TL, in

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cm), and body weight (BW, in g), were determined and the condition factor (CF) was calculated according to the following equation:

$$CF = \left(\frac{BW}{L^3}\right) \times 100$$

where BW is the body weight, L is the total length.

Specimens were dissected and the gills, muscle and liver were quickly removed and placed in 70% nitric acid (5 ml). The solution was gently heated for 2 h at (70–80 C°). Five ml of hydrogen peroxide 30% H2O2 was added until the digestion was complete. The solution was transferred to a 25 ml measuring flask and completed with distilled water. For each series of 10 samples, two analytical blanks were prepared similarly without samples to check the possible contamination. The concentration of Cadmium (Cd) and Lead (Pb) was determined by an Atomic Absorption spectrophotometer-GTA120 Graphite Tube Atomizer.

Statistical Analysis

All statistical analysis was carried out by MINITABE statistical package. The data were carried out using ranges and mean ± standard deviations. Relationships between measurements were tested by correlation analysis. Finally, a T-test was used to compare the measurements of Benghazi Lake and Qaminus beach fish groups.

3. Results

Overall range of measured variables: The results of heavy metals concentration (µg/g per wet body weight) of the liver, muscles and gills of *Mugil cephalus* were collected from Benghazi Lake and Qaminus Beach are shown in Tables 1 A and B. In both areas, the heavy metals were distributed in all tissues (gills, liver and muscles). Furthermore, the comparison between the different organs regarding the lead and cadmium concentrations in fish (Figs. 1 and 2) found that the levels of both metals were highly significant (P=0.001) in gills as compared to that of the liver and muscles, moreover, the concentrations of the lead and cadmium in the liver were found higher (P=0.05) than the concentrations of the lead and cadmium in muscles.

Table 1

Range mean ± SD for total body length, body weight, condition factor and metals accumulation in liver, muscles and gills of *Mugil cephalus* collected from (A) Benghazi like and (B) Qaminus Beach.

A Variable	Range
Total body length (cm)	23.00 – 37.00
Body weight (g)	138.00 – 455.00
Condition factor	0.678 – 1.298
Lead liver (µg/g)	0.121 - 0.441
Lead muscle (µg/g)	0.013 – 0.047
Lead gills (µg/g)	0.212 – 0.934
Cadmium liver (µg/g)	0.125 – 0.971
Cadmium muscle (µg/g)	0.008 – 0.969
Cadmium gills (µg/g)	0.403 – 0.766

B Variable	Range
Total body length (cm)	20.00 – 34.00
Body weight (g)	145.00- 470.00
Condition factor	1.069 – 2.780
Lead liver (µg/g)	0.073 - 0.622
Lead muscle (µg/g)	0.007 – 0.097
Lead gills (µg/g)	0.013 – 0.712
Cadmium liver (µg/g)	0.081 – 0.771
Cadmium muscle (µg/g)	0.005 – 0.051
Cadmium gills (µg/g)	0.196 – 0.531

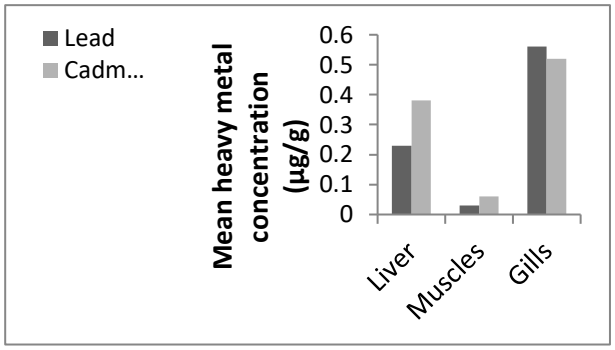

Fig. 1. Distributions of cadmium and lead in tissues of fish at Benghazi Lake.

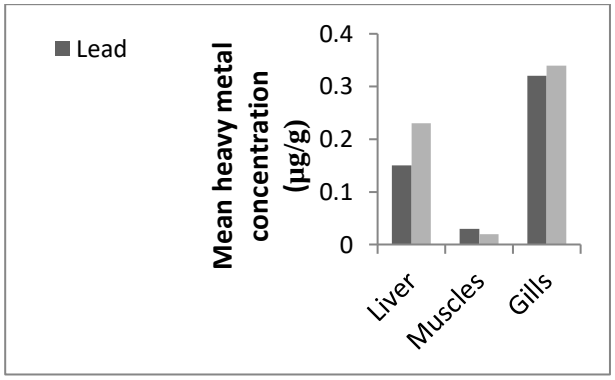

Fig. 2. Distributions of cadmium and lead in tissues of fish at Qaminus Beach.

Correlations among measured variables: Tables 2 and 3 show the matrix of correlations for total body length and body weight across metal concentrations in all tissues of *Mucil cephalus* from Benghazi Lake and Qaminus Beach. For fish from Benghazi Lake, many of the variables were correlated with each other. For total body length, a significant positive correlation was found between total body length with body weight and lead concentration in gills. However, a significant negative correlation was found between total body length with lead concentration in muscles. Body weight is positively related to lead concentration in gills and negatively correlated to the concentration of lead in muscles, but body weight is unrelated to other measures. The lead concentration in the liver was unconnected to all measures. The lead concentration in the muscles was unrelated to cadmium concentration in the liver, but significantly negatively related to lead concentration in gills, cadmium concentration in muscles and in gills. The lead concentration in gills was positively correlated to cadmium concentration in gills, but no significant relationship was found between lead gills with cadmium in the liver and muscles. Cadmium in the liver was not significantly related to cadmium in muscles and cadmium in gills, furthermore, cadmium in muscles was not significantly correlated to cadmium in gills.

For fish from Qaminus Beach, the total body length was positively related to the body weight, lead in the liver and in the gills, also, cadmium in the liver, muscles and gills. Nevertheless, total body length is negatively significantly related to lead in muscle. Body weight is also positively correlated to lead in the liver, lead in gills, and cadmium in both the liver and gills, but negatively correlated with lead in muscle. Lead in the liver was not correlated with lead in muscle. However, positive relationships were found between lead in the liver with lead in the gills and cadmium in the liver, although, negative relationships were found between lead in the liver with cadmium in muscle and gills. Lead in muscle was negatively correlated to lead in gills, cadmium in the liver, muscle and in gills. However, lead in gills was positively correlated with cadmium in the liver and gills. For cadmium in the liver was no significant correlation to cadmium in muscles and gills. However, Cadmium concentration in muscles was significantly positively correlated to cadmium concentration in gills.

Table 2

The correlation matrix the total body length (cm), body weight (g), lead in liver, lead in muscle, lead in gills, cadmium in liver, cadmium in muscle and cadmium gills for fish at Benghazi Lake, the top figure is the correlation coefficient and the bottom figure is the P- value.

	Total length	Body weight	Lead liver	Lead muscle	Lead gills	cadmium liver	cadmium muscle
Body weight	0.924						
	0.000						
Lead liver	0.065	0.051					
	0.564	0.650					
Lead muscle	-0.306	-0.252	0.125				
	0.005	0.022	0.263				
Lead gills	0.549	0.536	0.002	-0.523			
	0.000	0.000	0.983	0.000			
cadmium liver	0.034	-0.032	-0.013	0.011	0.059		
	0.764	0.776	0.906	0.925	0.597		
Cadmium muscle	0.100	0.086	-0.029	-0.263	0.172	-0.040	
	0.372	0.442	0.799	0.017	.123	0.724	
Cadmium gills	0.168	0.129	-0.161	-0.221	0.469	0.116	0.163
	0.130	0.244	0.149	0.045	0.000	0.300	0.144

Table 3

The correlation matrix the total body length (cm), body weight (g), lead in liver, lead in muscle, lead in gills, cadmium in liver, cadmium in muscle and cadmium gills for fish at Qaminus Beach, the top figure is the correlation coefficient and the bottom figure is the P- value**.**

	Total length	Body weight	Lead liver	Lead muscle	Lead gills	cadmium liver	Cadmium muscle
Body weight	0.902						
	0.000						
Lead liver	0.431	0.523					
	0.002	0.000					
Lead muscle	-0.362	-0.441	-0.107				
	0.012	0.002	0.470				
Lead gills	0.879	0.828	0.391	-0.392			
	0.000	0.000	0.006	0.006			
cadmium liver	0.933	0.903	0.625	-0.304	0.850		
	0.000	0.000	0.000	0.036	0.000		
Cadmium muscle	0.264	0.120	-0.387	-0.393	0.181	0.099	
	0.070	0.416	0.007	0.006	0.219	0.061	
Cadmium gills	0.467	0.257	-0.288	-0.333	0.365	0.273	0.634
	0.001	0.078	0.047	0.021	0.011	0.061	0.000

Comparison of Mugil cephalus with different areas: Fig. 3 and 4 show means ± standard deviation values for total body length, body weight and condition factor in the fish group from Benghazi lake and fish group from Qaminus Beach. The results showed that the fish group from Benghazi Lake had significantly larger total body length (P=0.001) and lower condition factor (P=0.001) than the group from Qaminus Beach, although, there was no significant difference between two groups regarding body weight (P=0.117). Furthermore, in terms of heavy metal contents, Table 4 shows means ± standard deviation values for all variables in the fish group from Benghazi lake and a fish group from Qaminus Beach., the comparison between the two groups of *Mugil cephalus* regarding the lead levels in different organs, there was a significant increase in the level of lead in the liver (P= 0.021) and gills (P=0.002) in a fish group of Benghazi Lake more than fish group collected from Qaminus Beach, but the level of lead in muscles in the two

groups were matched without significant difference (P=0.462). Regarding the cadmium concentrations in different organs, the fish group collected from Benghazi Lake had significantly higher levels of cadmium in the liver (P=0.001), muscles (P=0.016) and gills (p=0.001) than in organs of the fish group collected from Qaminus Beach.

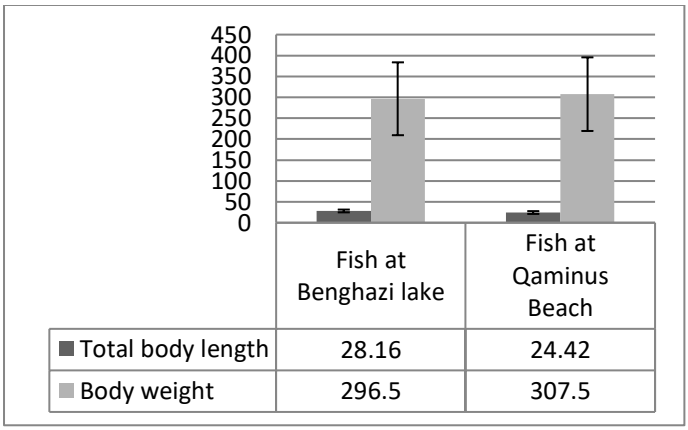

Fig. 3. Comparison of fish total body length (cm) and body weight (gm) between fish at Benghazi Lake and fish at Qaminus Beach.

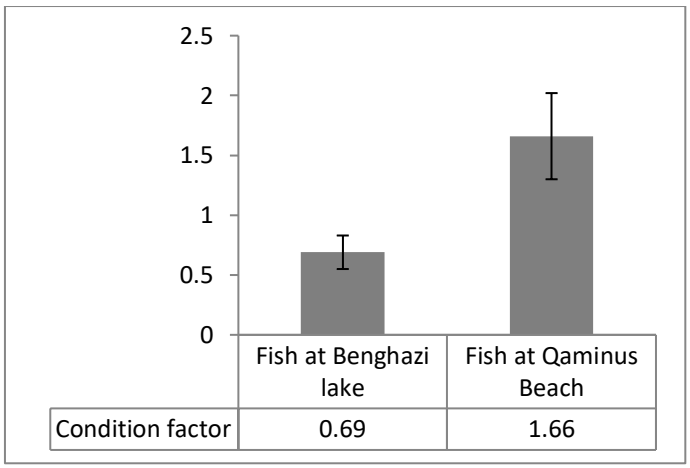

Fig. 4. Comparison of fish condition factor between fish at Benghazi Lake and fish at Qaminus Beach.

Table 4

A comparison of the heavy metal concentrations (Mean ± SD) among fish collected from the area.

Variable	Mean ± SD		P-value
	Fish at Benghazi lake	Fish at Qaminus Beach	
Total body length	28.16 ± 3.31	24.42 ± 3.36	0.001
Body weight	296.50± 87.14	307.50 ± 88.04	0.117
Condition factor	0.69 ± 0.14	1.66 ± 0.36	0.001
Lead liver	0.23 ± 0.09	0.15 ± 0.08	0.021
Lead muscle	0.03 ± 0.01	0.03 ± 0.01	0.462
Lead gills	0.52 ± 0.16	0.32 ± 0.16	0.001
Cadmium liver	0.38 ± 0.12	0.23 ± 0.18	0.001
Cadmium muscle	0.06 ± 0.12	0.02 ± 0.01	0.016
Cadmium gills	0.56 ± 0.10	0.34 ± 0.09	0.001

4. Discussion

Heavy metals are considered to be the most important form of water pollution due to their toxicity, long perseverance and accrual by marine organisms. In addition, the heavy metals (Pb and Cd) are poisonous to all seawater-living organisms when subjected to elevated concentrations (Oktariani *et al.,* 2023). Heavy metals in the sea even in small amounts, might accrue and reach to toxic level and cause organic pollution (Obasohan and Eguayoen, 2008). Furthermore, heavy metals are extremely toxic as ions or in complex shape, because these may become soluble in water and readily absorbed in living organisms and these metals preserve connections to very important cellular apparatus for instance structural proteins and nucleic acids and obstruct their functioning (Landis and Yu, 2000; Yia and Zhang, 2012).

In the present study, the levels of heavy metals were determined in the liver and gills because the liver is significant in metabolism and the accumulation site for elements whereas the gills have a large surface area, which is the primary site for elements uptake from water and gills are very sensitive, (Zaynab *et al.,* 2022). However, the muscles are not important tissue in metals agglomeration except mercury (Oktariani *et al.,* 2023), but the muscles are consumed as a source of a good protein and important in supporting the good health of humans (Hashim *et al.,* 2014).

In the current study, the levels of lead and cadmium have high concentrations in the gills followed by the liver and the lowest concentration was in muscles in the two fish groups. The physiological variations between organs control the bioaccumulation of a particular metal. The high concentration of metal in gills could be related to the character of metals with the gill mucus to be removed totally from the gill lamellae before analysis (Wasilah *et al.* 2021). Normally, the metal accumulation in different tissues and organs could be related to the method of exposure as diet or their elevated level in the closed water environment (El-Moselhy *et al.,* 2014). The study by Yilmaz *et at.* (2010) showed that the highest levels of Pb were found in the gills and the lowest concentration was found in the muscles of *Mugil cephalus* and *Oreochromis niloticus*. However, Dural *et al.* (2006) stated that the liver accumulated more metals than the gills and the muscles.

In the present study, there were positive and negative correlations for all measurements in the two groups of fish, (i.e. body weight, body length as well as lead and cadmium in liver, gills and muscles). Furthermore, the positive correlations between measurements and body weight and length could be due to loss of homeostasis capacity under stress and metal exposure leading to bioaccumulation. Kamaruzzaman *et al* (2010) indicated that there was a correlation between metal concentrations and fish body size. According to Yia and Zhang, (2012), the rise in fish length is correlated with the increase of heavy metal concentrations in some tissues.

The present study found that *Mugil cephalus* from Qaminus Beach has a higher condition factor than does *Mugil cephalus* from Benghazi Lake. Condition factor is an index for health and obviously responds in different ways to environmental conditions. Furthermore, condition factors in fish generally respond to some factors, for example, the status of diet, contact with toxic and the effects of pathogens (Jenjan, 2011)

Lead and cadmium in different livers and gills had higher concentrations in *Mugil cephalus* from Benghazi Lake than those in *Mugil cephalus* from Qaminus Beach. This high level of heavy metals in fish from Benghazi Lake could be attributed to local sources (untreated pollutants that drifted directly to the Benghazi Lake), for example, wastes of metals, domestic garbage deposits, medical wastes and household liquids. Nisbet *et al.* (2010) stated that the main sources of pollution in seawater are disposal of domestic and wastes of industrial. Furthermore, metal concentrations strongly depend on environmental conditions (Gaber, 2007; Djedjibegovic *et al.,* 2020; Eldurssi *et al.,* 2022) and can disturb wholly water ecosystems (Palaniappan and Muthulingam, 2016). In general, the rise of lead and cadmium levels in organs of *Mugil cephalus* from Benghazi Lake could also be attributed to the increase of the total dissolved heavy metal consequently leading to the increase of free cadmium and lead levels and then an increase in the metals uptake by fish. This pollution can disturb wholly water ecosystems directly or through the food chain.

5. Conclusions

The result showed that metal accumulation varied depending on location and different organs. Metal concentrations were higher in the Benghazi fish group and lower in the Qaminus beach fish group. Variations in metal concentrations were recorded in the body organs of the fish groups. Lead and cadmium bioaccumulations were higher in the gills followed by liver and muscles. The great bioaccumulation of lead and cadmium in the liver and gills is possibly connected to characters of organs.

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