**SPECTROPHOTOMETRIC ANALYSIS OF HEAVY METALS OF SOME ORGANS OF LABEO DIPLOSTOMUS AND COERULEUS DAY COLLECTED FROM MARDAN KHEL DAM DISTRICT KARAK, KHYBER PAKHTUNKHWA, PAKISTAN**

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**Abstract**

The present study is focused on the analysis of some selected heavy metals like (Pb, Fe, Cu, Cd & Zn) in four body regions of a species *Labeo diplostomus, Labeo caeruleus day* collected from Ghol Mardan Khel dam of district Karak for their significance for human consumption. This study was conducted from April to August 2019 to find out accumulation of heavy metals in fishes of Ghol Mardan Khel Dam Karak, Khyber Pakhtunkhwa, Pakistan. Heavy metal tests were conducted by Atomic Absorption Spectrometry. The obtained results were expressed mg/kg dry weight. Results showed that (Pb, Fe & Cu) concentrations were the most abundant among all tissues according to WHO value whereas Cd and Zn levels were the lowest.

**Key Words;** Heavy Metal Concentration in *Labeo Diplostomus* and *Labeo caeruleus day*

**Introduction**

Fish perform an essential role in human health because they contain minerals and vitamins [1]. Fish are often used as bio-indicators of heavy metals pollution in marine ecosystem [2]. Metals are characterized as non-essential and essential groups based on their roles and needs for development and growth of human, animals and plants. Metals like Cadmium (Cd), Lead (Pb) and Chromium (Cr) are non-essential metals while Iron (Fe), Manganese (Mn) and Copper (Cu) are essential metals, but these must be with in their respective permissible limits. Cd is highly toxic environmental pollutant and it cause various disease like failure of kidney, heart problems, blood pressure cancer and bone damage [3]. However, heavy metals such as Pb and Cd are toxic even in trace amounts and to date, many studies have revealed the contamination of heavy metal, especially in ground water [4] . Pb in high concentrations can affect nerve system, cause headache, high blood pressure, lungs and stomach problems [5]. In the case of Cd, it is a non-essential, highly toxic metal [6].

The term “heavy metals” refers to any metallic element that has density greater than 4g/cm3. Heavy metals exhibit metallic properties such as ductility, malleability, conductivity, cation stability, and legend specificity. Heavy metals include; Lead (Pb), Cadmium (Cd), Copper (Cu), Iron (Fe), Manganese (Mn), Zinc (Zn), groups elements. Some heavy metals such as Co, Cu, Fe, Mn, Mo, Ni, V and Zn are required in minute quantities by organisms. However, excessive amounts of these elements can become harmful to organisms. Other heavy metals such as Pb, Cd and Hg as a metalloid but generally referred to as a heavy metal do not have any beneficial effect on organisms and are thus regarded as the “main threats” since they are very harmful to both plants and animals [7]. heavy metals eventually enter in food chains and their bioaccumulation and magnification can cause physiological and morphological alterations not only in aquatic animals but in human beings as well [8].

Heavy metals are essential for the regular metabolic activity in fish and are found in the aquatic food, in different sediments and water [9]. The rate of heavy metal accumulation in fish meat depends on the species of fish, age, gender and location where the fish are cultivated [10]. Heavy metals are released into aquatic environment from industrial waste discharge, mining activities, sewage sludge and discharge of municipal wastes [11].

The infants and younger population are more prone to the toxic effects of heavy metals, as the rapidly developing body systems in the fetus, infants and young children are far more sensitive [12]. Ghol Mardan Khel Dam site is in Urmar Algad, a left tributary of Teri Toi at about 20 km from Banda Daud shah on a road to Gurguri in district Karak .Cost of the Dam Project is Rs.439,974 million .The Dam has a crest length of 227 ft with height of 100 ft. The live storage capacity is 2383 AF having Cultivable Command area (CCA) of 1500 Acres . it is estimated that 3000 persons will be benefited from the project. Therefore, the present study was also conducted to examine some trace metals in the different parts of the Labeo species *( Labeo diplostomus and Labeo careallus day* ) collected from Ghol Mardan Khel Dam of district Karak to examine their impact on human health .

**Material and Method**

**Area of Sample Collection**

Fishes are collected randomly from GholMardan khel Dam Khyber Pakhtunkhwa at district Karak. During the month of April 2019 in order to analyse them for heavy metal detection.

**Sample Collection**

Fishes were collected from Ghol Mardan Khel Dam with the help of non-local special fisherman using different types of nets and hooks. Fishes were preserved in 5% formalin solution in separate bottles. The samples were collected in sterile polythene bags and kept in the laboratory deep freezer (-20°C) to prevent deterioration till further analysis. We worked on Dry method and a clean washed high quality corrosion resistant stainless knife was used to cut the fish into Head, Gills, Tail and Abdomen and was placed in china dishes.

**Preparation of Samples**

**Dissection of Fishes**

**Apparatus**

The tools that is used for dissection were (i) scissor, (ii) surgical blade, (iii) dissecting tray, (iv) dissection box, (v) forceps, (vi) gloves, (vi) clothes, (vii) disssection fluids, (viii) dissecting pans and pads .

**Procedure**

We take a fish samples then we put the said sample of fishes in the dissecting tray ,before the dissection we measure the organs of fish and also weighted the fish through digital balance and place the fish in dissecting try darsoventrly and then we start the dissection .

**Incision at anus**

At the beginning of dissection insert the surgical blade or cleaner blade at the anus of a fish.the anus is located in the front of anal fin. The incision is extended intrearly along the fish belly towards the head part. The incision passses interarly between the pelvic fins and at depends upon the types of the fish. Uses the scissor to cut anterioly through the bones attached to the pelvic fins cut forward along the narrow, fleshy space benaith the head and gills. Pull apart of the two walls of the body in order to expose the internal organ or viceral organ. We see the internal organs such as heart, lungs, kidney, pyloric ceaic, liver, gallblader, etc. We take only four body organs such as head, gills, abdomen & tail for heavy metal analysis.

**Refrigeration of fish sample**

These all fish said samples were refrigerate at 30 degree centigrade at the labortary of Bioinformatic deportment of Khushkhal Khan Khattak University Karak.

**Drying of samples**

All these samples were dried at 110 C for the next 24 hours and then these dried fish samples were grinded using pistol and mortar. Using Analytical balance one gram of each sample was taken.

**Digestion of Samples**

Each said samples we can put them in 16 ml of nitric acid and then we put the 8 ml of HCL. By putting these chemicals, we get a colorless solution. This solution we placed on a hot plate to become evaporated them by 100 centigrade

**Filtration of Samples**

After evaporating the remining sample we can filter them by using a filter paper at the chemistry lab KUST. We can add 15 ml distalled water to each samples then we get 25 ml of net solution of each sample.

**Analysis**

We can analyse the Heavy elements by using the Atomic Absorption Spectrometer (AAS) at the laboratory for the determination of each heavy element a specific lamp is used.

**Result and Desscustion.**

A survey was started from April to August 2019. According to our survey we captured the two species belonging to same order were selected from Ghol Mardan Khel Dam for heavy metal analysis. the consumption of highly contimenated food may prove to be lethal for human beings and can also produce some bad impact. therefore, this study is focused on evaluating the trace metal leavel in fish tissuses. The obtained results of the detection of heavy metal like Lead ( Pb), Iron (Fe), Cupper (Cu), Cadmium (Cd), & Zinc (Zn). The fish species *Labeo diplostomus* belong to phylum urochordata, class pieces, cohort Euteleostii, Order cypriniformes, Family cyprinidae, Genus labeo cuvier. According to table no1 the concentration of heavy metal in tissues of (Labeo diplostomus) the Lead (Pb) their concentration is in increasing order randamily according to WHO value (0.01) in Head (0.386), Gills (0.148), Abdomen (0.165) and Tail (5.057) . All the four value are high according to WHO value which is lethal for human beings. Iron (Fe) their concentration also increase randamily from WHO value (0.30) in Head, (11.77) Gills (4.350), Abdomen (7.583) and (6.915) Tail. The sample concentration is greater then that of the highest standard which is lethal for human beings. Copper (Cu) their concentration also increase randamily from WHO value (0.30) in Head (0.693), Gills (0.573), Abdomen (0.353) and Tail ( 0.337) there value is greater from the highest standard which is lethal for human health. Cadmium (Cd) their concentration is normally according to WHO value (0.003) compare to Head (0.013), Gills (0.002), Abdomen (0.005) & Tail (0.004) which have no bad impact for human health. Zinc (Zn) their sample concentration is normal which have no bad impact for human health according to WHO value (3.00), Head (0.02) Gills (0.453) Abdomen (0.003) & Tail (0.222). The fish species (*Labeo caeruleus day*) belong to Phylum urochordata, Class pices, Cohort Euteleostei,Order Cypriniformes, and Genus *labeo cuvier* .According to table no 2 the concentration of heavy metal in tissues of species (*Labeo caeruleus day*). The lead (Pb) their concentration is in increasing order randamily according to WHO value (0.01) in Head (0.105) Gills (0.136) Abdomen (0.054) Tail (0.035). All the four value are high according to WHO value which is lethal for human health. Iron (Fe) their concentration is also incressing randamily according to WHO value (0.30) in Head (2.035) Gills (1.734) Abdomen (3.669) Tail (3.109). All the four samples concentration is greater then that of the highest standered which become lethal for human health. Copper (Cu) their concentration is incressing randamly according to WHO value (0.30) in Head (0.162) Gills (0.064) Abdomen (0.120) Tail (0.0358). All Samples concentration is heigher according to WHO value which is lethal for human beings.Cadmium (Cd) their samples concentration is normal according to WHO value (0.003) in Head (0.008) Gills (0.003) Abdomen (0.006) Tail (0.001) . All the sample don’t show negative reaction in human health. Zinc (Zn) all the samples concentration is normal according to WHO value (3.00) in Head (0.652) Gills (0.218) Abdomen (0.163) Tail (0.122). All the four samples have no bad impact for human beings. It has been observed that different tissues of ﬁsh may have distinct afﬁnities for heavy metal accumulation due to their differing physiological functions [13]. To evaluate the health risk to Pakistan people through consumption of marine fish, daily intake of heavy metals was estimated on the basis of the concentrations of Mn, Zn, Cu, Cd and Pb in muscle of daily fish consumption. The average daily fish consumption in Pakistan is 33 g per person [14] . Trace metals such as Pb will interfere with essential nutrients of similar characteristics such as calcium (Ca) and Zn. Pb also causes renal failure and liver damage in humans [15] . The gills are in direct contact with the contaminated medium (water) and have the thinnest epithelium of all the organs and metals can penetrate through the thin epithelia cells [16].

**Table 1:** Metals concentration in *Labeo Diplostomus*organs sample.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Heavy Metals** | **Organs** | | | |
| **Head** | **Gills** | **Abdomen** | **Tail** |
| Lead (Pb) | 0.386 ± 0.077 | 0.148± 0.188 | 0.165± 0.023 | 5.057± 0.185 |
| Iron (Fe) | 11.77± 0.087 | 4.350±0.174 | 7.583±0.034 | 6.915± 0.024 |
| Copper (cu) | 0.693±0.008 | 0.573± 0.003 | 0.353± 0.008 | 0.337±0.005 |
| Cadmium (Cd) | 0.013±.009 | 0.002±0.010 | 0.005±0.008 | 0.004±0.007 |
| Zinc (Zn) | 0.002±0.010 | 0.453±0.007 | 0.003±0.010 | 0.222±0.010 |

Concentration of “Lead” Pb in fish *Labeo diplostomus*. Concentration of “Iron” Fe in fish *Labeo diplostomus.*

Concentration of “Copper” Cu in fish *Labeo diplostomus.* Concentration of “Cadmium” Cd in fish *Labeo diplostomus.*

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Concentration of “Zinc” Zn in fish *Labeo diplostomus .*

**Table 2:** Metals concentration in *Labeo caeruleus day* organs sample .

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Heavy Metals** | **Organs** | | | |
| **Head** | **Gills** | **Abdomen** | **Tail** |
| Lead (Pb) | 0.105±0.119 | 0.136± 0.127 | 0.054± 0.197 | 0.035± 0.106 |
| Iron (Fe) | 2.035± 0.011 | 1.734±0.023 | 3.669±0.026 | 3.109± 0.024 |
| Copper (cu) | 0.162±0.015 | 0.064± 0.013 | 0.120± 0.021 | 0.0358±0.009 |
| Cadmium (Cd) | 0.008±.010 | 0.003±0.004 | 0.006±0.007 | 0.001±0.005 |
| Zinc (Zn) | 0.652±0.007 | 0.218±0.001 | 0.163±0.001 | 0.122±0.001 |

Concentration of “Lead” Pb in fish *Labeo Caeruleus day*. Concentration of “Iron” Fe in fish *Labeo Caeruleus day*.

Concentration of “Copper” Cu in fish *Labeo Caeruleus day*. Concentration of “Cadmium” Cd in fish *Labeo Caeruleus day.*

. Concentration of “Zinc” Zn in fish *Labeo Caeruleus day.*

**Conclusions**

From the given result, it was concluded that the (LABEO) species *Labeo diplostomus*, *Labeo caeruleus day* found in from Ghol Mardan Khel Dam Karak contain metal concentration in high permissible limit, so hence, this Labeo species was not suitable for human consumption. Furthermore, as the environmental condition of this dam are not free from the accumulation of heavy metals pollution hence, it is more suitable to take some steps for the management is that to remove the heavy metals from this dam to provide such good environment for the good health of the fish’s community. but the local people of the district Karak used this fish in daily routine in medical point which is not good for their health

**References**

1. Al-Bader, N. (2008). Heavy metal levels in most common available fish species in Saudi market. J Food Technol, 6, 173-177.
2. Authman, M. M., Zaki, M. S., Khallaf, E. A., & Abbas, H. H. (2015). Use of fish as bio-indicator of the effects of heavy metals pollution. Journal of Aquaculture Research & Development, 6(4), 1-13.
3. Mijal, R. S., & Holzman, C. B. (2010). Blood cadmium levels in women of childbearing age vary by race/ethnicity. Environmental research, 110(5), 505-512.
4. Mahagamage, M., Chinthaka, S., & Manage, P. (2015). Assessment of water quality index for groundwater in the Kelani River basin, Sri Lanka.
5. Muhammad, S., Shah, M. T., & Khan, S. (2011). Health risk assessment of heavy metals and their source apportionment in drinking water of Kohistan region, northern Pakistan. Microchemical journal, 98(2), 334-343.
6. Shivakumar, C., Thippeswamy, B., Tejaswikumar, M., & Prashanthakumara, S. (2014). Bioaccumulation of heavy metals and its effect on organs of edible fishes located in Bhadra River, Karnataka. International Journal of Research in Fisheries and Aquaculture, 4(2), 90-98.
7. Offor, I., Ehiri, R., & Njoku, C. (2014). Proximate nutritional analysis and heavy metal composition of dried Moringa oleifera leaves from Oshiri Onicha LGA, Ebonyi State, Nigeria. Journal of Environmental Science, Toxicology and Food Technology, 8(1), 57-6 2.
8. Vinodhini, R., & Narayanan, M. (2008). Bioaccumulation of heavy metals in organs of freshwater fish Cyprinus carpio (Common carp). International Journal of Environmental Science & Technology, 5(2), 179-182.
9. Gupta, A., Rai, D. K., Pandey, R. S., & Sharma, B. (2009). Analysis of some heavy metals in the riverine water, sediments and fish from river Ganges at Allahabad. Environmental monitoring and assessment, 157(1-4), 449.
10. Thakur, J., & Mhatre, M. (2015). Assessment of potential dietary toxicity of heavy metals in Tilapia mossambica in the industrially polluted area of MIDC Taloja, India. International Journal of Engineering Technology, Management and Applied Sciences, 3, 85-90.
11. Gautam, S. K., Sharma, D., Tripathi, J. K., Ahirwar, S., & Singh, S. K. (2013). A study of the effectiveness of sewage treatment plants in Delhi region. Applied Water Science, 3(1), 57-65.
12. Johnson, D. B., & Hallberg, K. B. (2005). Acid mine drainage remediation options: a review. Science of the total environment, 338(1-2), 3-14.
13. Tapia, J., Vargas-Chacoff, L., Bertrán, C., Peña-Cortés, F., Hauenstein, E., Schlatter, R., . . . Tapia, C. (2012). Heavy metals in the liver and muscle of Micropogonias manni fish from Budi Lake, Araucania Region, Chile: potential risk for humans. Environmental monitoring and assessment, 184(5), 3141-3151.
14. Chughtai, M. I., & Mahmood, K. (2012). Semi-intensive carp culture in saline water-logged area: A multi-location study in Shorkot (District Jhang), Pakistan. Pakistan Journal of Zoology, 44(4).
15. Salem, H. M., Eweida, E. A., & Farag, A. (2000). Heavy metals in drinking water and their environmental impact on human health. ICEHM2000, Cairo University, Egypt, 542-556.
16. Bebianno, M., Geret, F., Hoarau, P., Serafim, M., Coelho, M., Gnassia-Barelli, M., & Romeo, M. (2004). Biomarkers in Ruditapes decussatus: a potential bioindicator species. Biomarkers, 9(4-5), 305-330.