Risks assessments of heavy metals bioaccumulation in water and *Tilapia nilotica* fish from Maguite Island of Fitri Lake.

ABSTRACT

The purpose of this study is to assess concentration of selected metals (Zn, Cu, Cr, Pb, Cd and Fe) in the muscle, flesh and bone of *Tilapia nilotica* fish and in the water of Fitri Lake. It was carried out in Maguite Island of Fitri Lake, Batha region in Chad. Samples were collected from April to December 2016. A hundred of fish samples and three (3) sites of water collection were concerned. The content in heavy metals is determined using a spectrophotometer. The averages of heavy metals (mg/L) in water are 0.04 (Fe), 0.01 (Zn), 3.01 (Cu), 5.78 (Cr), and 0.07 (Cd). All the fish tissues have an important level of heavy metals content. Muscles have the low content. Heavy metals content (mg/kg) vary from 55.72 to 88.45 for Fe, 36.45 to 50.24 for Zn, 20.26 to 32.19 for Cu, 0.34 to 17.82 for Cr and 0.02 to 0.07 for Cd. All the analyzed parameters exceed the acceptable limit according to international standards. The high level of heavy metal content in fishes and water of the lake is an indicator of an important pollution activity around the lake or drained by the lake.

Keywords: Heavy metals, Bioaccumulations, *Tilapia nilotica*, Water, Fitri Lake

1. INTRODUCTION

Since many years ago, more attention is paid to the dispersion of polluting agents in the environment. Heavy metals are listed among the main environment polluting agents. There
are widely dispersed in the environment with a high level of potential bioaccumulation in many animal species and toxicity [1]. They may lead to a destructive effect on the ecological balance of the aquatic environment [1, 2] and probably on human health. Human activities have increased the concentration of metals in many of the natural water systems, which have raised concerns. Heavy metal pollution has been associated with anthropogenic activities, such as effluents and emissions from mainly mines, which often contain high concentrations of heavy metals [3]. Many reports underline an intensification of mining activities in the region of Batha (Chad) department of Fitri where is located the concerned lake. Some heavy metals are drained by the stream from the neighbor countries until the lake where they are accumulated. In the aquatic environment, larger animals such as fish have been exposed to heavy metals as a direct significance of biomagnifications [4-7]. The danger is that heavy metals even at low concentrations in fish and water have a particular importance in ecotoxicology and their toxic effects have been widely published for a number of water bodies [4, 8-14]. Among aquatic species, fishes are the inhabitants that cannot escape from the harmful effects of heavy metal pollution. \textit{Tilapia nilotica} is an import fish in natural lake and basin in Africa (Nil, Chad, Niger, Volta, Senegal, Jou) [15]. This is because of their very close contact with water that carries the heavy metals and also because fish have to extract oxygen from water by passing water over their gills. Muscles provide relatively low content of heavy metal content because of their low metabolic activity, but they may serve as a circumstantial biological indicator because of their relative mass compared to other organs such as bone. This study, therefore, aims to determine the concentration of trace elements such as Cd, Cr, Cu, Fe, Pb, Zn in bone, flesh and muscle in \textit{Tilapia nilotica} fish in Fitri Lake, Maguite Island.

\section*{2. MATERIAL AND METHODS}

\subsection*{2.1. Site location and description}
The study focuses on Fitri Lake. Fitri Lake is located in the central part of Chad Republic, in the region of Batha, Fitri department. It connected to Batha River. Fitri Lake is a great biosphere reserve and one of the important ecosystems of the Country. It is located about 300 km from N'djamena, the capital. Its area is estimated to about 500,000 hectares. And *T. nilotica* is the most fished fish.

![Figure 1: Sampling sites in Fitri Lake](image)

### 2.1. Sampling

Water is collected at three different sites of the lake during different seasons. A total of nine samples of water were collected. A polypropylene bottle is used for water sampling. The bottle is previously washed with demineralized water. All bottles are transported under 4°C and then homogenized with 0.5 mL (for 125 mL of sample) of HNO₃ 50% [16].

One hundred of *T. nilotica* fish were collected for heavy metal analysis. The collected fish samples were sent directly to the food control center lab (CECOQDA) by land in an ice-filled...
All samples were conditioned under 4 °C until laboratory where it stored in the freeze for analyzing.

### 2.3. Sample preparation and analyze

In the laboratory, we immediately weighed, measured the size of the fish which were then chilled at 4ºC. Subsequently, the fish were dissected and the tissue was dried and digested with HCl / HNO3 following the method of the American Society for Testing and Materials. Flesh, muscles and bone were removed and dried in an oven at 105 ºC. Dry sample was incinerated at 550 ºC in the oven and the ashes obtained were finally kept in plastic bags in the refrigerator. Total ash was quantified by standard method.

To each flask of 50 ml content, 0.5 ml of concentrated nitric acid is added. The flask is shaken and filtered throughout cellulose membrane. Fish are immediately weighted, size measured and then chilled at 4ºC. Subsequently, the fish were dissected and the tissue was dried and digested with HCl / HNO3 according to the method of the American Society for Testing Materials [17]. Flesh, muscles and bone were removed and dried in an oven at 105 ºC until constant weight and then incinerated at 550° C in the oven and the ashes obtained were finally kept in plastic bags in the refrigerator in the meantime [16, 17].

Heavy metals are determined using atomic absorption spectrometry (VARIAN spectra AA 50B) powered by an air-acetylene flame at the air pressure of 2.5-4 bar and that of the acetylene: 0.5-0.6 bar. The supply current was 4 mA.

This colorimetric method is based on Lambert-Beer law according to which the optical density of the element is directly proportional to its concentration in the solution. Thus, by adding the dithiver pre dissolved reagent in a capsule to the solution, coloration develops and its intensity is directly proportional to the concentration of the metal. The reading is at 515 nm for Pb and at 525 nm for Cd, Fe, Cu, Zn, Cr. For each analysis, 1 g of dry ash from
the sample is digested with fuming nitric acid (65%) before dilute with deionized distilled water.

2.4. Standard preparation

A solution containing the concerned metal is injected into a flame in which metals tend to remain in the ground state. The principle is based on the decrease in the intensity of radiation due to energy absorption is measured by a spectrophotometer, and it is a function of the number of atoms present on the path of radiation at any time [16, 17]. Measurements of optical densities were made wavelengths $\lambda$ (228.8, 324.7, 283.3, 213.9, 248.3 and 357.9 nm) respective for Ca, Cu, Pb, Zn, Fe and Cr. The concentration of the metal in the sample is determined according to the formula:

$$C = \frac{(C_1 \cdot V)}{P}$$

C: Concentration in the metal of the sample (mg/kg)
C1: Concentration in the metal of the sample solution (mg/l)
V: Final volume of the mineralization solution in ml
P: Mass of the sample in g

2.5. The bioaccumulation factor

Bioaccumulation: Ratio of the steady-state chemical concentrations in an aquatic water-respiring organism and the water determined from field data in which sampled organisms are exposed to a chemical in the water and in their diet. It is a combination of chemical bioconcentration and biomagnification.

The bioaccumulation factor was calculated according [18], using the following formula

$$BF = \frac{\text{Concentration of metals in fish tissues}}{\text{Concentration of metals in abiotic media}}$$

Where the abiotic media represents the water of the lake.
The intensity of contamination of the carp fish Tilapia nilotica of Lake Fitr is estimated from the comparison between the determined trace element values and the reference values (conventional) of the maximum limit concentrations of these elements in fish organs [36].

2.6. Statistical analysis

The results are expressed in milligrams per kilogram (mg/kg or ppm) of fresh weight. Means and analyze of variance (ANOVA) were determine using Sphinx plus2 software.

3. RESULTS AND DISCUSSION

3.1. Results

3.1.1. Size of the Fish

Collected fish was range according to their size. The fish sizes are ranged between 23 and 39 cm with an average of 29 cm as described in Table 1.

Table 1: Size of fish

<table>
<thead>
<tr>
<th>Size (cm)</th>
<th>Effective</th>
<th>Frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>[22- 24]</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>[24 - 26]</td>
<td>16</td>
<td>16</td>
</tr>
<tr>
<td>[26 -28]</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>[28 - 30]</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>[30 - 32]</td>
<td>7</td>
<td>7</td>
</tr>
<tr>
<td>[32 - 34]</td>
<td>9</td>
<td>9</td>
</tr>
<tr>
<td>[34 – 40]</td>
<td>19</td>
<td>19</td>
</tr>
</tbody>
</table>
3.1.2. Weight of the fish

The weight of the collected fish is ranged according to table 2. The maximum of weight is 408 g, with an average of 247.26 g.

Table 2: Weight of fish

<table>
<thead>
<tr>
<th>Weight (g)</th>
<th>Effective</th>
<th>Frequency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt; 60</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>60 – 120</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>120 – 180</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>180 – 240</td>
<td>28</td>
<td>28</td>
</tr>
<tr>
<td>240 – 300</td>
<td>6</td>
<td>6</td>
</tr>
<tr>
<td>300 – 360</td>
<td>18</td>
<td>18</td>
</tr>
<tr>
<td>360 – 408</td>
<td>21</td>
<td>21</td>
</tr>
</tbody>
</table>

3.1.3. Heavy metal content in the water of the lake

The water of the lake is alkaline with a pH varying between 7.2 and 8.5. The average content of the heavy metal is 0.4 mg/l, 0.07 mg/l, 0.17 mg/l, 5.78 mg/l, 0.01 mg/l respectively for Fe, Cd, Pb, Cr and Zn as showed in the following Table 3.

Table 3: Heavy metal content of Friti lake water (mg/l)
The evolution of heavy metal content in the lake from April to December is as indicated in the following Figure 2.

![Figure 2: Evolution of heavy metal content in the lake from April to December](image)

<table>
<thead>
<tr>
<th></th>
<th>pH</th>
<th>Fe</th>
<th>Zn</th>
<th>Cu</th>
<th>Cr</th>
<th>Pb</th>
<th>Cd</th>
</tr>
</thead>
<tbody>
<tr>
<td>April</td>
<td>7.8</td>
<td>0.04±0.02</td>
<td>0.01±0.00</td>
<td>3.01±0.09</td>
<td>5.78±0.85</td>
<td>0.17±0.04</td>
<td>0.07±0.05</td>
</tr>
<tr>
<td>May</td>
<td>8.2</td>
<td>0.04±0.02</td>
<td>0.01±0.01</td>
<td>2.66±0.29</td>
<td>5.22±0.11</td>
<td>0.14±0.03</td>
<td>0.07±0.03</td>
</tr>
<tr>
<td>June</td>
<td>7.6</td>
<td>0.16±0.19</td>
<td>0.1±0.02</td>
<td>2.9±0.98</td>
<td>5.11±0.32</td>
<td>0.15±0.05</td>
<td>0.07±0.04</td>
</tr>
<tr>
<td>July</td>
<td>7.9</td>
<td>0.12±0.04</td>
<td>0.12±0.04</td>
<td>2.9±0.11</td>
<td>5.22±0.54</td>
<td>0.18±0.06</td>
<td>0.07±0.03</td>
</tr>
<tr>
<td>August</td>
<td>8.5</td>
<td>0.11±0.10</td>
<td>0.11±0.04</td>
<td>2.61±0.11</td>
<td>4.67±0.01</td>
<td>0.17±0.05</td>
<td>0.08±0.03</td>
</tr>
<tr>
<td>September</td>
<td>7.2</td>
<td>0.14±0.11</td>
<td>0.1±0.07</td>
<td>2.71±0.16</td>
<td>5.11±0.96</td>
<td>0.16±0.05</td>
<td>0.09±0.01</td>
</tr>
<tr>
<td>October</td>
<td>7.5</td>
<td>1.14±0.18</td>
<td>0.13±0.06</td>
<td>2.61±0.08</td>
<td>5±0.12</td>
<td>0.17±0.06</td>
<td>0.11±0.01</td>
</tr>
<tr>
<td>November</td>
<td>7.8</td>
<td>0.11±0.1</td>
<td>0.14±0.05</td>
<td>3.01±0.09</td>
<td>5.22±0.59</td>
<td>0.18±0.06</td>
<td>0.07±0.02</td>
</tr>
<tr>
<td>December</td>
<td>8.3</td>
<td>0.12±0.04</td>
<td>0.15±0.07</td>
<td>2.97±0.17</td>
<td>4.44±0.19</td>
<td>0.19±0.08</td>
<td>0.1±0.06</td>
</tr>
</tbody>
</table>
3.1.4. Heavy metal content in fish tissues

The content in heavy metal of the fish bone is 0.22 mg/Kg, 36.45 mg/Kg, 65.97 mg/Kg, 20.26 mg/Kg, and 17.82 mg/Kg respectively for cadmium, zinc, iron, copper and chrome. For fish flesh, the content in heavy metal is 88.45 mg/kg, 22.87 mg/Kg, 0.34 mg/Kg, 0.43 mg/Kg, 0.07 mg/kg respectively for Fe, Cu, Cr, Pb and Cd. In muscle it about 55.72 mg/kg, 32.19 mg/kg, 0.36 mg/kg, 0.58 mg/kg, 88.45 mg/kg for Fe, Cu, Cr, Pb and Cd respectively (Table 4).

Table 4: Heavy metal content of fish part (mg/kg)

<table>
<thead>
<tr>
<th>Fish parts</th>
<th>Fe</th>
<th>Zn</th>
<th>Cu</th>
<th>Cr</th>
<th>Pb</th>
<th>Cd</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flesh</td>
<td>88.45±11.48</td>
<td>42.83±22.55</td>
<td>22.87±9.05</td>
<td>0.34±0.034</td>
<td>0.43±0.36</td>
<td>0.07</td>
</tr>
<tr>
<td>Bone</td>
<td>65.97±25.66</td>
<td>36.45±21.7</td>
<td>20.26±9.59</td>
<td>17.82±1.13</td>
<td>0.25±0.20</td>
<td>0.22±0.019</td>
</tr>
<tr>
<td>Muscle</td>
<td>55.72±29.45</td>
<td>50.24±16.67</td>
<td>32.19±9.83</td>
<td>0.36±0.033</td>
<td>0.58±0.48</td>
<td>0.02</td>
</tr>
</tbody>
</table>

3.1.5. Bioaccumulation of heavy metals in fish part

The bioaccumulation factor of each heavy metal in fish fleshes, bones and muscles is as presented in the following Table 5. It varies from 253.27 to 402.05 for Fe, 379.69 to 523.33 for Zn, 6.99 to 11.11 for Cu, 0.7 for Cr, from 1.50 to 3.47 for Pb and from 0.88 to 2.72 for Cd.

Table 5: Bioaccumulation factor of heavy metals in fish parts

<table>
<thead>
<tr>
<th>Fish parts</th>
<th>Fe</th>
<th>Zn</th>
<th>Cu</th>
<th>Cr</th>
<th>Pb</th>
<th>Cd</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flesh</td>
<td>402.05</td>
<td>446.15</td>
<td>7.89</td>
<td>0.07</td>
<td>2.69</td>
<td>0.88</td>
</tr>
<tr>
<td>Bone</td>
<td>299.86</td>
<td>379.69</td>
<td>6.99</td>
<td>0.07</td>
<td>1.50</td>
<td>2.72</td>
</tr>
<tr>
<td>Muscle</td>
<td>253.27</td>
<td>523.33</td>
<td>11.11</td>
<td>0.07</td>
<td>3.47</td>
<td>0.25</td>
</tr>
</tbody>
</table>

Concerning the bioaccumulation factor, the general decreasing order of heavy metals in fish respectively analyzed part is as shown in the following Table.

Table 6: General decreasing order of bioaccumulation of heavy metals in fish parts
<table>
<thead>
<tr>
<th>Variable</th>
<th>General order</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zn</td>
<td>Muscle &gt; Flesh &gt; Bone</td>
</tr>
<tr>
<td>Fe</td>
<td>Flesh &gt; Muscle &gt; Bone</td>
</tr>
<tr>
<td>Cu</td>
<td>Muscle &gt; Flesh &gt; Bone</td>
</tr>
<tr>
<td>Pb</td>
<td>Muscle &gt; Flesh &gt; Bone</td>
</tr>
<tr>
<td>Cd</td>
<td>Bone &gt; Flesh &gt; Muscle</td>
</tr>
<tr>
<td>Cr</td>
<td>Muscle &gt; Flesh &gt; Bone</td>
</tr>
</tbody>
</table>

The general decreasing order of the heavy metals in each part of the fish is as followed:

- **Flesh:** $\text{Zn} > \text{Fe} > \text{Cu} > \text{Pb} > \text{Cd} > \text{Cr}$
- **Bone:** $\text{Zn} > \text{Fe} > \text{Cu} > \text{Cd} > \text{Pb} > \text{Cr}$
- **Muscle:** $\text{Zn} > \text{Fe} > \text{Cu} > \text{Pb} > \text{Cd} > \text{Cr}$

### 3.2. Discussion

The fish samples have size ranged between 23 and 39 cm with an average of 29 cm as showed in table 1. There has an average weight of 247.26 g. This result is similar to those of Lake Chad with the same fish species [15]. Collected fish can then be classified in little fish, middle fish and great fish according to their size and weight.
The content in heavy metals of Fitri Lake as showed in Table 3 put in evidence the fact that Cr, Cu, Pb, Zn, Fe and Cd are respectively the more concentrate in the water of the Lake. The concentration level of Cd, Cr and Cu is similar to those find in the river of Ravi, Pakistan [19]. In Lake Chad, only Cr was detected as heavy metals with an average content of 2.3 ppm [15]. In Lufira Lake of Katanga, Congo Republic, the concentration in heavy metals is less than this result [1]. A high level of heavy metal concentration was also observe in Cameroun [20]. There is not a significant variation in the content of heavy metals in the lake except Cr and Fe. A high level of Fe content is observed in October. The concentration in the content of Cr decrease from April to December. Heavy metals concentration is then evidence in Fitri Lake. The concentration of heavy metal is Fitri Lake water is several time more high than international standard recommendation [21, 22]. It is a general environmental and health preoccupation considering its incidence in several aquatic environment particularly in Africa [1, 2, 9, 15, 20, 23-28]. Unfortunately, there is not yet standard neither law nor political engagement in African countries to contain heavy metals bioaccumulation in natural aquatic environment. Traditional mining exploitation and manure use contribute a lot to heavy metals bioaccumulation. In Fitri department as in some others places, aquatics resources are the only proteins resources for communities. With this high level of concentration, the risk is then evident. A hazard analyze would have considered the Lake as a critical point for any operation cannot be done to reduce resources from the Lake. This indicate why fish have an important concentration level of heavy metals.

The content in heavy metal of the fish vary from one tissue to another one. It is about 0.25, 36.45, 65.97, 20.26, and 17.82 mg/Kg respectively for cadmium, zinc, iron, copper and chrome in bone. For flesh, it is 88.45, 22.87, 0.34, 0.43, 0.07 mg/Kg respectively for Fe, Cu, Cr, Pb and Cd. In muscle it about 55.72, 32.19, 0.36, 0.58, 88.45 mg/Kg for Fe, Cu, Cr, Pb and Cd respectively (Table 4). In general, flesh is the tissue that concentrates the more heavy metals, followed by muscle and then bone. In the same T. nilotica in lake Chad, Cd...
was only detected in big fish liver and Pb was absent in fish liver. Only Cr was detected in all tissues of fish. Fish bone was the more concentrated in heavy metals [15]. The content in heavy metals is more higher than those of some fish tissues of Red Volta (Ghana), in Nigeria, in Pakistan, Morocco, in Cotonou, RD Congo, in the Mediterranean Sea, the Red Sea and North Sea [1, 18, 19, 28-31]. Fish from Fitri lake are then among the more concentrate in heavy metals in the world. All the fish tissues are concerned by the bioaccumulation. The importance concentration of heavy metals in bone may be an indicator of a long time pollution of the lake. For more study, only flesh, muscles, gills or liver are concerned [18, 26, 30, 31]. The acceptable limit is about 50 μg/L for Pb and 0.05 mg/L for Cr in water. For fishes, acceptable limit are about 0.5 and 0.1 (mg/kg of fresh weight) for Pb and Cd respectively according to CE standard [32]. The acceptable limit of Cu content in fish and fishery products is 30 mg/g [33]. All the analyzed heavy metals exceed largely the acceptable limit for both water and fish. And, the bioaccumulation of heavy metals concerns all the fish tissues. Which mean that population who consume the fish from Fitri Lake is at risk, for the exposure level is high. The population around the lake are in majority fishers men and then fish have an important place in their diet [34]. For the thousands who live in the department there is an emergency to contain the pollution, for is a long time public health problem. The danger is that heavy metals even at low concentrations in fish and water have a particular importance in ecotoxicology and their toxic effects have been widely published. Unlike many organic pollutants, heavy metals are not eliminated biologically, which promotes their cumulative effect in various compartments of the ecosystem. The accumulation of toxic metals in the human food chain is recognized globally as a risk to public health [35]. These results also put in evidence the importance of industries, manure use, mining activities around the lake. The water of the lake must immediately be avoided by population in food preparation without adequate treatment.
4. CONCLUSION

The study showed a permanent concentration of heavy metals in Fitri Lake. The content in heavy metal in the lake is higher than the acceptable limit, according to standard. All the analyzed tissues (flesh, muscles, bone) of *T. nilotica* are concentrated in heavy metal. The content in heavy metals is higher. All the suspected metals were detected. Corrective action is needed to protect the health of consumers.

Ethical Approval:

As per international standard or university standard written ethical approval has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES


28. Moslen M. Risk Assessment and Bioconcentration of Heavy Metals in Mugil Cephalus (Mullet) obtained from Azuabie Creek in Port Harcourt, Nigeria. 2017.


