# Bioaccumulation of Heavy Metals in *Clarias Gariepinus* Exposed to Untreated Paint Effluent

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#### ABSTRACT

This study presents the heavy metal accumulation profile in Clarias gariepinus (catfish) exposed to untreated paint effluent. Sub-lethal concentrations of paint effluent were used to pollute catfish aquaria. This was followed by isolation and identification of microorganisms from the polluted aquaria after 96 h using conventional microbiological techniques. The tissue heavy metal composition and physicochemical parameters of the aquaria were also monitored using standard analytical methods. Microbial load in the range of 10.00a±2.00 to 189.00d±20.52 cfu/ml on tissues of catfish were observed. The heavy metals present in the paint effluent were mainly Lead  $(1.20\pm0.01$  ppm) and Zinc (0.34±0.01ppm). No significant difference in the proximate analysis between the tissue of catfish from the control and the polluted aquaria were observed. Catfish tissue accumulated Lead (0.16 ppm) from the polluted aquaria. Persistent exposure through pollution by effluent in aquatic environment can lead to increased metallic Lead bioaccumulation that could be unhealthy for both the fish and human consumption. Therefore, indiscriminate discharge of untreated effluent from industrial sites into water bodies should be discouraged.

KEYWORDS: Clarias gariepinus, bioaccumulation, heavy metal, paint effluent

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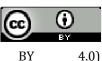
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#### 1. INTRODUCTION

Persistent contamination of water bodies through continuous discharge of industrial effluents is common and can produce varying degree of physiology and biochemical changes in fish species [1]. Rapid urbanization and industrialization has led to increased disposal of pollutants such as heavy metals, radio nuclides and various types of organic and inorganic into the aquatic environment. Extensive industrialization has measurably influences the quality of water of lakes, ponds and rivers all over the world. Industrial wastes constitute the major source of metal contamination of the aquatic environment [2]. Industrial effluents have been regarded as the main input source of various pollutants to the aquatic ecosystem and contain various organic and inorganic substances potentially toxic to aquatic biota. It is well established that pollution lowers the quality of life in various aspects and affects health and life span [3]. Besides the direct health effects, the subtle danger of pollutants lies in the fact that they may be mutagenic or toxic and lead to several human afflictions like cancer, atherosclerosis, cardiovascular diseases and premature ageing [4]. Discharging effluents without proper treatments may have an adverse effect on the receiving water bodies [5]. Heavy metals constitute the major contaminants. Metals are important pollutants, because they are not eliminated from the aquatic ecosystems by natural processes like organic pollutants and are enriched in mineral organic substances. Metallic contaminants are introduced into aquatic system through smelting process, effluents, sewage and leaching of garbage which cause serious damage to the aquatic fauna [6]. Heavy metals have long being recognized as serious pollutants of the aquatic ecosystem. Increase discharge of heavy metals into natural aquatic ecosystems can expose aquatic organisms to unnatural high levels of these metals [7]. The metals entering the aquatic ecosystem may not directly cause damage to organism but they can be deposited in aquatic organism through the effect of bio-concentration, bioaccumulation and other food chain processes thus gets to man in concentrations that poses threats to human health via consumption of sea foods [8]. Transport of metals in fish occurs through the blood where the ions are usually bound to proteins. The metals are brought into contact with the organs and tissue of the fish and consequently accumulated to a different extent in different organs or tissues of the fish. Once heavy metals are accumulated by an aquatic organism, they can be transported through the upper class of the food chain [9].

Fish is an important component of human nutrition, and those from contaminated sites present a potential risk to human health. Metals can accumulate in aquatic organisms including fish and persist in water and sediments. Fishes are the simple and reliable biomarkers of pollution of metal in a polluted aquatic ecosystem, they tend to take these metals up from their direct environment [10]. Therefore, this study was undertaken to assess the heavy metal accumulation and microbial safety of catfish grown in aquaria contaminated with paint effluent.

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#### 2. MATERIALS AND METHODS

#### 2.1. Collection of samples

The Paint effluent used in this study was obtained at point source of a Paint Factory in Lagos. The samples were collected in sterile containers and taken for physiochemical and microbiological analysis.

#### 2.2. Set up and Pollution of Aquaria

Six (6) aquaria in duplicates each contain 10 fingerlings. The fishes were then exposed to paint effluent at sub-lethal concentrations of 0.00 (control), 1.25mls/l, 2.50mls/l, 3.75mls/l, 5.0mls/l and 6.25mls/l for 96hours. Careful observations were then made to note the number of mortalities and effect of the paint on the test organism.

#### **Analytical Methods**

#### 2.3. Water Analysis for Heavy Metal

Water samples in the bottles were shaken sufficiently for thorough mixing, and then 50mls aliquots were filtered on Watman No. 41 filter papers and quantitatively transferred into 50mls volumetric flask before aspiring directly into the Inductively Coupled Plasms for analysis of heavy metals as describe by APHA [11].

#### 2.4. Analysis on fish

#### 2.4.1. Heavy Metals Analysis

The concentrations of the selected heavy metals (Pb and Zn) in fish flesh were determined using X-Ray Fluorescence analyzer.

#### 2.4.2. Nutritive Value Analysis of Fish

Nutritive value of the sample flesh was determined using Kjedahl Steam Distillation and Soxhlet methods respectively.

#### 2.5. Physiochemical Parameters

The physiochemical parameters such as temperature, total dissolved solids and total solids were determined using the methods of Aderinola et al. [12].

#### 2.6. Biochemical and Morphological Identification of Bacteria Isolates

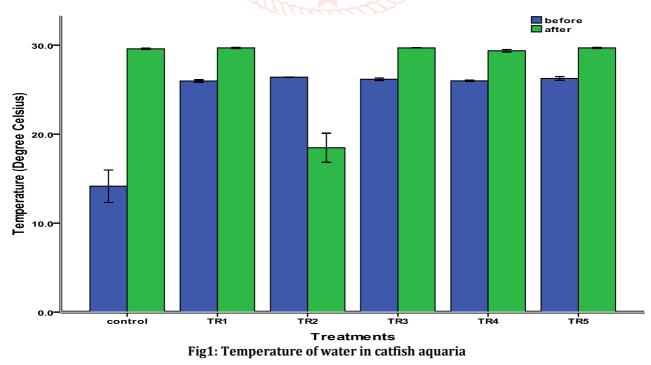
Bacteria were isolated using nutrient agar as well as selective media; and the individual colonies were identified by morphological and biochemical techniques using methods described by Osho and Fawole [13].

#### 2.7. Statistical Analysis

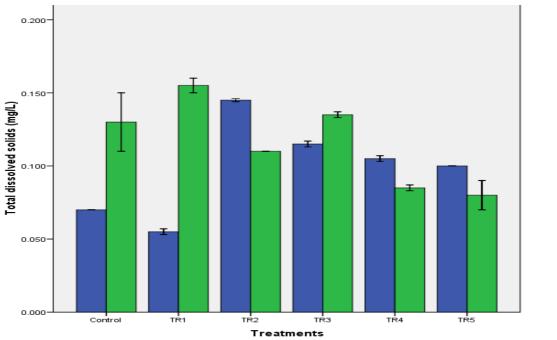
The data were analyzed for significant differences (P<0.05) by Analysis of Variance (ANOVA) using computer Statistical Package for Social Sciences (SPSS) for windows (v.25.0). Determined differences were partitioned by the Least Significant Difference (LSD) at P=0.05

#### 3. Results and discussion

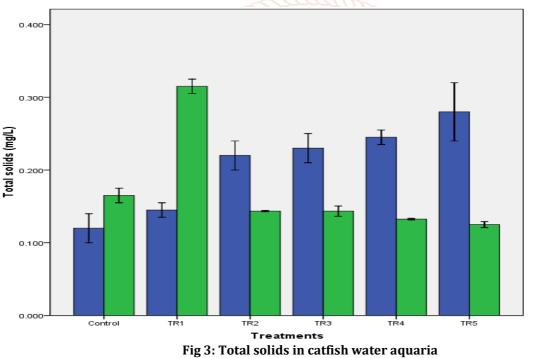
The temperature range of the aquaria contaminated with paint effluent is presented in figure 1. The figure shows no significant difference in the temperature of the control and other treatments except treatment 2 which had a lower temperature at the end of the experimental period. This suggest that the paint effluents had no effect on the temperature of the fish aquaria however, the lower temperature observed in the treatment 2 may need to be investigated more since there are no observation that may suggest possible explanation for the drop in the temperature.

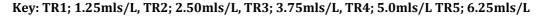


Key: TR1; 1.25mls/L, TR2; 2.50mls/L, TR3; 3.75mls/L, TR4; 5.0mls/L TR5; 6.25mls/L









Figures 2 and 3 presents the total dissolved solids and total solids in the aquaria water samples respectively. The total solids in the aquaria water increased with increase in the paint effluent contamination rising from 0.125mg/L in the control to 0.265mg/L in the treatment 5 (6.25ml/L of effluent) at the beginning of the experiment. Meanwhile, the total solids got reduced drastically in all the treated samples after four days trial except treatment 1. This may be due to assimilation of some of the solid particles by the fish [14]. Also, it may be due to settling of these solids at the bottom of the aquaria

Table1: Heavy metal	present in paint	effluent sample

Parameters	Composition		
Zinc	0.34±0.01		
Nickel	N.D		
Chromium	N.D		
Cadmium	N.D		
Lead	1.20±0.01		
Kev: ND- Not detected			

Day1

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Treatment	<b>Moisture content</b>	Crude protein	Fat contents	Ash content
Control	7.78c±0.12	32.71a±0.75	27.62a±0.38	11.73b±0.20
TR1	4.92a±0.21	47.86c±0.87	28.33a±0.23	13.77c±0.40
TR2	6.54b±0.48	38.88b±0.00	29.52a±0.13	10.50a±0.40
TR3	10.02d±0.07	48.5c±0.00	31.54ab±0.02	11.30ab±0.06
TR4	8.68c±0.45	31.71a±0.78	29.01a±0.05	10.46a±0.27
TR5	7.66c±0.29	37.17b±0.78	30.67ab±0.32	11.03ab±0.11

#### Table2: Proximate composition of catfish

Values are presented as Mean±S.E (n=3). Means with the same superscript letter(s) along the same column are significantly different (P>0.05).

#### Key: TR1; 1.25mls/L, TR2; 2.50mls/L, TR3; 3.75mls/L, TR4; 5.0mls/L TR5; 6.25mls/L

#### Table3: Heavy metal accumulation in harvested catfish

Treatments	Zinc	Lead	
Control	0.21	0.01	
TR1	0.22	0.06	
TR2	0.24	0.09	
TR3	0.15	0.12	
TR4	0.14	0.16	
TR5	0.22	0.15	

Key: TR1; 1.25mls/L, TR2; 2.50mls/L, TR3; 3.75mls/L, TR4; 5.0mls/L TR5; 6.25mls/L

Table4: Bacterial load of catfish tissue (x 10 <sup>2</sup> cfu/g)				
Treatment	Skin	Liver	Gill	Intestine
Control	10.00 <sup>a</sup> ±2.00	33.67 <sup>a</sup> ±3.67	35.33ª±7.57	100.67 <sup>b</sup> ±1.53
TR1	58.67°±13.32	75.67 <sup>ab</sup> ±4.30	80.67 <sup>b</sup> ±21.20	142.00°±5.05
TR2	70.67 <sup>b</sup> ±10.07	87.67 <sup>bc</sup> ±8.02	81.33 <sup>b</sup> ±6.11	180.00 <sup>e</sup> ±4.00
TR3	103.67°±4.73	126.33°±5.51	91.33 <sup>d</sup> ±3.22	104.67 <sup>b</sup> ±2.01
TR4	100.33°±20.50	90.00 <sup>bc</sup> ±15.10	89.00 <sup>d</sup> ±20.52	85.67 <sup>a</sup> ±6.03
TR5	73.00 <sup>b</sup> ±20.66	77.33 <sup>ab</sup> ±8.08	85.67°±16.01	168.00 <sup>d</sup> ±5.92
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## Table4: Bacterial load of catfish tissue (x 10<sup>2</sup> cfu/g)

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Values are presented as Mean±S.E (n=3). Means with the same superscript letter (s) along the same column are not significantly different (P>0.05).

#### Key: TR1; 1.25mls/L, TR2; 2.50mls/L, TR3; 3.75mls/L, TR4; 5.0mls/L TR5; 6.25mls/L

#### Table5: Frequency of occurrences of bacterial isolates in catfish grown in paint effluent contaminated aquaria

Isolates	Before	After	Total
Staphylococcus aureus	2	1	3
Salmonella sp		2	2
Bacillus substilis	3	4	7
Enterobacter aerogenes	2	_	2
Bacillus cereus	2	3	5
Micrococcus leteus	1	2	3
Klebsiella sp	1	1	2
Aeromonas hydrophila	2	1	3
Listeria monocytogenes	_	2	2
Shigella sp	1	1	2

The results of the heavy metal present in the paint effluent is shown in table 1 where it was revealed that Ni, Cr, and Cd were not detected in the effluent whereas, Pb and Zn were found to be 1.20 ppm and 0.34 ppm respectively. The non detection of the other heavy metals in the effluent samples may be due to pretreatment by the company before releasing their waste water into the environment, since the government require industries to treat their wastes to reduce toxicity to barest minimum before ejecting same into the environment [15]. The presence of lead in the effluent in high proportion suggests that the effluent may be dangerous to aquatic lives if released like that in to water bodies. Lead poisoning is one of the major problems associated with industrial effluent discharge with debilitating effects on animals. The heavy metals become toxic when they do not get metabolized by the body and end up accumulating in the soft tissues [16]. Ingestion is the most common route of exposure to heavy metals. Fish has been reported to accumulate metals from water by diffusion via skin and gills as well as oral consumption. Bioaccumulation of heavy metals in *Clarias gariepinus* probably might have resulted from inability of the fish to metabolize the heavy metals or the metabolism of the heavy metals in the fish tissues, were slow [17]. The results of the proximate composition and mineral content of the catfish used in this study are presented in table 2 and 3 respectively. There were no significant differences in the crude protein, fat and ash content of the fish. However, there was a gradual increase in lead concentration in the tissue of the cat fish with increase in the concentration of the paint effluent. The increase may be due to the availability of the metal in the water where the fish lives as a result of contamination by the effluent. Also, bioaccumulation of heavy metals especially lead in aquatic animals have been documented in the last decade. The high level of lead in the catfish tissue may be linked with bioaccumulation of same. Similar result was also documented by Igwegbe et al. [18].

The bacterial load on different parts of the cat fish is shown in table 4. The table revealed that all the catfish grown in paint effluent contaminated aquaria had higher bacterial count compared with the control. The bacterial load of the intestines were higher than those found in other parts of the body whereas the skin had the least count. The higher count in the intestine may be due to digestion activity going on in the intestine as well as the abundance of food available for microbial growth [19]. However, the high microbial count ranging from 75.67 to 126./33 x 10<sup>2</sup> cfu/g observed in the liver compared with  $33.67 \times 10^2$  cfu/g obtained in the control group suggests that the paint effluent may suppress the catfish immunity and allow infection of major internal organs. This may lead to death of the fish and general loss of aquatic diversity [20]. A total of 10 bacterial isolates were obtained based on their morphological and biochemical properties. The bacteria were: Micrococcus luteus, Bacillus subtilis, Staphylococcus aureus. Enterobacter aerogenes, Salmonella Sp, Bacillus cereus, Klebsiella spp, Aeromonas hydrophila, and Listeria monocytogenes. Tables 3 shows the frequency of occurrences of probable isolates from catfish, it was observed that Bacillus subtilis was the most frequent for catfish. Some of these bacteria are known pathogens of man and their presence in the fish suggests that the consumption of the fish pose serious health risk to the consumers [21].

#### 4. Conclusion

This study revealed the quality of fish in terms of heavy metal and microbial contamination, when exposed to an industrial paint effluent. The levels of the heavy metals detected in fish are not remarkable to cause acute health consequences, but chronic adverse health effects due to prolonged bioaccumulation and long term exposure through fish consumption can be inevitable. The public should be enlightened on the possibility of being susceptible to health problems when feeding on fish contaminated with heavy metals and harmful microorganisms

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