

# BIOACCUMULATION OF HEAVY METALS BY catfish (*Clarias gariepinus*) IN E-WASTE SOIL POLLUTED AQUARIA AND ASSOCIATED FUNGI

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**Abstract** - Soil samples from Alaba International Market, Lagos, were analyzed for physiochemical and microbiological parameters. Alaba International Market, Lagos is a market known for sales of fairly used electrical electronic appliances and computers. This market also has a site where electrical electronic appliances that are condemned and have reached their life cycle end are dumped. The soils were used to pollute a species of catfish's aquaria in the ratio of 1:1, 1:2 and 1:3 of water to soil. Differences were observed in pH and the BOD<sub>5</sub> of the fish water which were monitored weekly for five weeks. The soil from e-waste dumpsite differ from the control in most of the parameters (pH, moisture content, organic contents, minerals and heavy metals) measured, specifically, higher organic contents (17.60%), moisture content (3.86%), organic carbon (10.17%) and higher value of all the heavy metals analyzed (Pb, Cd, Zn, Co, Cr, Mn and Ni) and the numbers of fungi isolated from the e-waste soil is more than the soil without e-waste. There were decreases in fungal population with increase in the e-waste soil pollution while there were increases in the fungal population with increase in the soil without e-waste pollution. Fungi were isolated from the soils, polluted fish water and parts of the harvested fish. The fungi isolated include; *Penicillium italicum*, *Aspergillus paraciticus*, *Aspergillus niger*, *Aspergillus flavus*, *Varicosporium elodeae*, *Saccharomyces* sp, *Mucor mucedo*, *Articulospora inflata*, *Candida* sp, *Rhizopus stolonifer*, *Zoopage nitospora*, *Rhodotorula rubra* and *Aureobasidium pullulans*. Bioaccumulations of the heavy metals after 5 weeks were significantly difference at  $P \leq 0.05$  between treatments. The sequence of the heavy metals concentrations in the fish sample was Zn>Pb>Mn>Cr>Cd>Ni>Co. Manganese exceeded it recommended limits of 0.01 – 0.05ppm. The results of the research showed that the dumping of e-waste on land leads to increase in heavy metals released to the land, e-waste soil pollution of fish water affect the pH, BOD<sub>5</sub>, fungal loads, fungal types, leads to bioaccumulation of heavy metals in catfish.

**Keywords** – Bioaccumulation, e-waste, fungi, heavy metals.

## INTRODUCTION

Burning of e-waste may generate dioxins, furans, polycyclic aromatic hydrocarbons (PAHs), polyhalogenated aromatic hydrocarbons (PHAHs), and hydrogen chloride [14]. The chemical composition of e-waste changes with the development of new technologies and pressure from environmental organisations on electronics companies to find alternatives to environmentally damaging materials [17]. Most e-waste is disposed in landfills, effective reprocessing technology, which recovers the valuable materials with minimal environmental impact, is expensive. Consequently, although illegal under the Basel Convention, rich countries export an unknown quantity of e-waste to poor countries, where recycling techniques include burning and dissolution in strong acids with few measures to protect human health and the environment [14]. Such reprocessing initially results in extreme localized contamination followed by migration of the contaminants into receiving waters and food chains [14], [42]. E-waste workers suffer negative health effects through skin contact and inhalation, while the wider community is exposed to the contaminants through smoke, dust, drinking water and food. There is evidence that e-waste associated contaminants may be present in some agricultural or manufactured products for export [42]. E-waste contains valuable metals (copper, platinum group) as well as potential environmental contaminants, especially lead, mercury, nickel, selenium, cadmium, polybrominateddiphenyl ethers (PBDEs), and polychlorinated biphenyls (PCBs) [14]. Fish are always at the top of aquatic food chain and when pollutants build up in the food chain, fish are widely used to evaluate the health of aquatic ecosystems [2], [21]. Bioaccumulation of heavy metals in fish has been reported by many researchers. The uptake of heavy metals in fish was found to occur through absorption across the gills surface or through the gut wall trait. Diffusion facilitates the transportation in gills and surface mucus and mechanisms of uptake through the gut from food and the rate of excretion. Gills generally have the

highest metal concentration due to their intimate contact with the environment and their importance as an effect or of ionic and osmotic regulation [2], [15]. Liver in its role as a storage and detoxification organ can also accumulate much less. The aspect of human health linked to the consumption of contaminated fish as a result of the presence of heavy metals is of great concern [15], [21]. Bioaccumulation of heavy metals by fish in its true sense is not only a danger to the fish accumulating the heavy metals but more importantly great danger is posed to man consuming the fish. There is need to increase the public awareness of the danger of improper disposal of e-waste which is a source of heavy metals. Therefore, this work focused on isolation and identification of fungi from e-waste dumpsite soil, e-waste soil polluted aquaria and bioaccumulation of heavy metals from e-waste with a view to create more awareness of bioaccumulation of heavy metals from e-waste.

## MATERIALS AND METHODS

### Collection of samples

Soil samples were collected from Alaba International Market, Lagos, Lagos State, Nigeria, in sterile container using soil auger and were taken to the Department of Microbiology, Department of Fisheries and Aquaculture Technology, Department of Chemistry, all in The Federal University of Technology, Akure, Ondo State, for analyses.

### Set up and pollution of aquaria

A set of aquaria made up of seven aquaria each containing six juvenile catfish (set A), were polluted with three different quantities of the e-waste soil sample and soil without e-waste (25g, 50g and 75g for both soil samples) in the ratio of 1:1, 1:2, 1:3 of water to soil samples after acclimatization of the fishes for six weeks and the seventh aquarium in each set is the control. The aquaria were monitored weekly for five weeks for physiochemical parameters; pH, dissolved oxygen, biochemical oxygen demand, total, titratable acidity while the fungi analyses are; weekly monitoring of fungi loads, isolation and identification of fungi and fungal succession in the polluted fish waters.

### Identification of fungi

This was done based on the cultural, morphological and microscopic examination of the colonies;

### Cultural characteristics of fungi

Using visible observation and microscope at low power magnification (x40), the parameters such as colony colour, characteristics of the submerged hyphae rhizoid, spiral or regular and characteristic shape of mature fruiting bodies were all observed.

### Microscopic examination of fungi

This involved transferring a small piece of mycelium free of medium using a sterile inoculating loop unto a clean glass slide containing a drop of cotton blue-in-lactophenol and the mycelium was spread properly. The

preparation was covered with a clean grease free cover slip and observed under medium power (x100). The observations made were used in identifying the fungi organism [30].

### Total plate count

Plates in triplicates from e-waste soil, soil without e-waste, unpolluted fish water, e-waste soil polluted fish water, soil without e-waste polluted catfish water and harvested catfish samples were observed for their fungi loads. The unpolluted fish water, e-waste soil polluted catfish waters and the soil without e-waste polluted catfish water samples were plated weekly for five weeks and the fungi loads of each interval were noted.

### Enumeration of fungi counts

Spore counting was carried out by counting the number of visible spores that appears on the plates. Calculation of spore forming unit (sfu) per ml for fungi was based on the volume of the sample used.

### Physiochemical parameters

The physiochemical parameters measured are; temperature, pH [16], total titratable acidity [38], Biochemical oxygen demand (APHA, 1995), Organic carbon determination and Organic matter [35], total phosphate determination and nitrogen determination [1] and heavy metals determination in soil samples [23].

### Heavy metals bioaccumulation analysis

This was done by determining the heavy metal in the catfish (*Clarias gariepinus*) tissues from the different treatments [37]. The data obtained were subjected to statistical analysis.

## Results

**Table 1: Soil physiochemical parameters and heavy metal profile**

PARAMETER	A	B
pH	7.90	8.70
Moisture content (%)	3.86	2.24
Organic matter (%)	17.60	5.00
Organic carbon (%)	10.17	2.89
Organic nitrogen (%)	0.35	0.21
Organic phosphorus (mg/kg)	146.65	160.00
Lead (mg/kg)	64.90	3.06
Cadmium (mg/kg)	0.32	0.02
Zinc (mg/kg)	35.50	3.34
Cobalt (mg/kg)	0.83	0.05
Chromium (mg/kg)	0.54	0.26
Manganese (mg/kg)	18.60	2.99
Nickel (mg/kg)	2.82	0.08
Sodium (mg/kg)	24.40	31.40
Potassium (mg/kg)	33.30	32.90
Calcium (mg/kg)	182.00	245.00
Magnesium (mg/kg)	34.00	29.70

**Key: A- Soil from e-waste dumpsite  
B- Soil without e-waste**

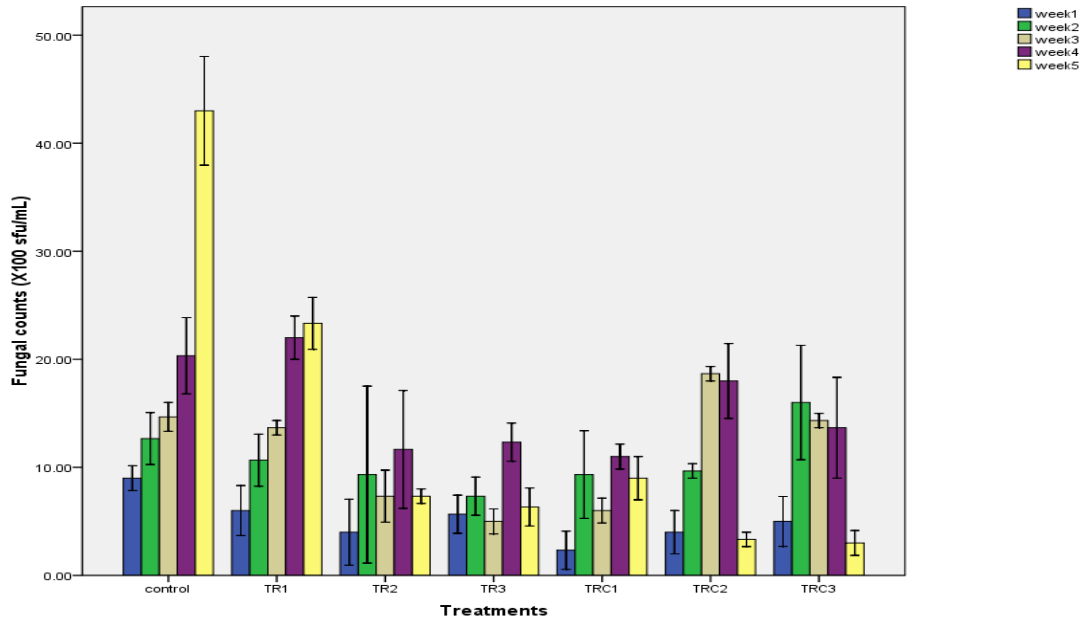


Figure 1: Weekly fungal count of catfish aquaria

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste.

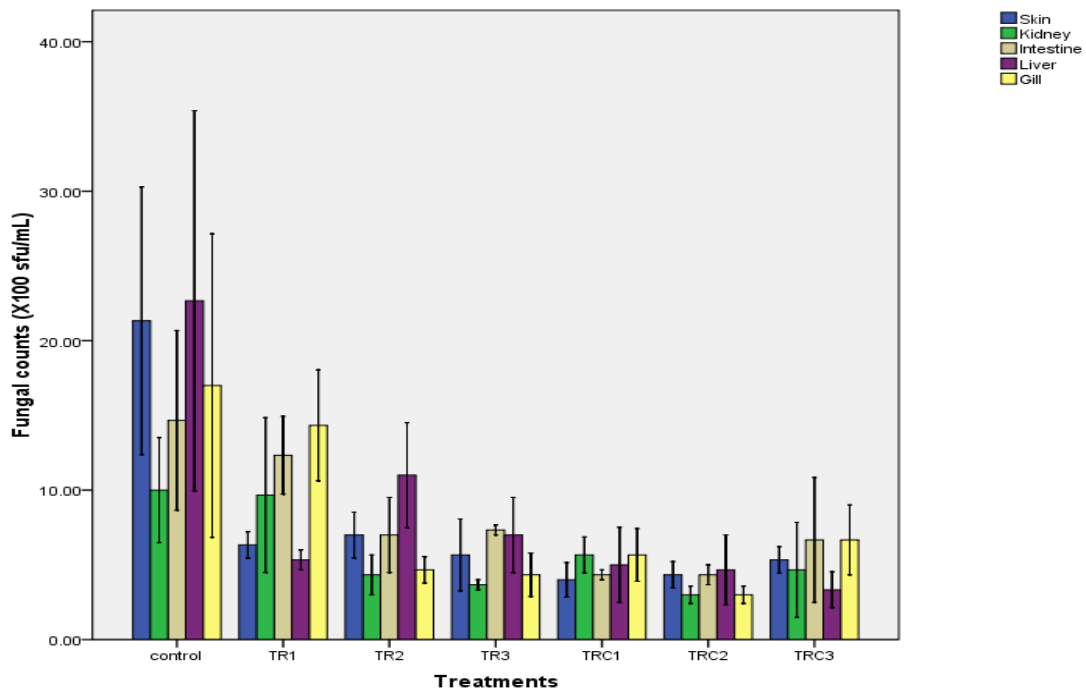


Figure 2: Fungal count of harvested catfish body parts after 5 weeks

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste.

**Table 2: Isolated fungi from the e-waste soil and the soil without e-waste**

Isolates	E-waste soil	Soil without e-waste
<i>Candida</i> sp	+	+
<i>Zoopage nitospora</i>	+	-
<i>Articulospora inflata</i>	+	+
<i>Varicosporium elodeae</i>	+	-

**Table 3: Isolated fungi in the control, soil without e-waste and e-waste catfish aquaria**

Isolate	Control	Soil without e-waste	E-waste soil
<i>Penicillium italicum</i>	+	+	+
<i>Candida</i> sp	+	+	+
<i>Articulospora inflata</i>	+	+	+
<i>Aspergillus niger</i>	-	+	+
<i>Rhizopus stolonifer</i>	-	+	-
<i>Mucor mucedo</i>	-	-	+
<i>Zoopage nitospora</i>	-	-	+
<i>Varicosporium elodeae</i>	-	-	+
<i>Rhodotorula rubra</i>	-	-	+
<i>Aureobasidium pullulans</i>	-	+	-
<i>Aspergillus paraciticus</i>	-	+	+

**Key: + = Present, - = Absent**

**Table 4: Numbers of isolated fungi from fish aquaria**

Treatments	Catfish
Control	301
TR1	207
TR2	109
TR3	110
TRC1	117
TRC2	160
TRC3	157
Total	1161

**Key: Control – Not polluted, TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste.**

**Table 5: Fungi isolated from body parts of harvested catfish**

Isolate	Control	Soil without e-waste	E-waste soil
<i>Penicillium italicum</i>	+	+	+
<i>Candida</i> sp	+	+	+
<i>Articulospora inflata</i>	+	+	+
<i>Aspergillus niger</i>	+	+	+
<i>Rhizopus stolonifer</i>	+	+	+
<i>Aspergillus flavus</i>	+	+	+
<i>Mucor mucedo</i>	-	+	+
<i>Zoopage nitospora</i>	-	+	+
<i>Varicosporium elodeae</i>	-	+	+
<i>Rhodotorela rubra</i>	-	+	+
<i>Aureobasidium pullulans</i>	-	+	+

**Key: + = Present, - = Absent**

**Table 6: Numbers of isolated fungi in the body parts of harvested catfish**

Treatments	Gills	Skin	Liver	Intestine	Total
Control	21	34	30	34	119
TR1	16	20	21	42	99
TR2	14	21	33	21	89
TR3	18	17	21	22	78
TRC1	17	12	15	13	57
TRC2	9	13	15	13	50
TRC3	20	16	10	20	66
Total	110	128	140	160	538

**Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste**

**Table 7: Heavy metal present in the harvested catfish tissue (mg/kg)**

Treatment	Pb	Cd	Zn	Co	Ni	Mn	Cr
Control	0.12 <sup>a</sup> ±0.02	0.01 <sup>a</sup> ±0.00	2.63 <sup>a</sup> ±0.11	ND	0.02 <sup>b</sup> ±0.01	0.20 <sup>ab</sup> ±0.01	0.04 <sup>a</sup> ±0.01
TR1	0.45 <sup>c</sup> ±0.01	0.02 <sup>a</sup> ±0.00	3.33 <sup>cd</sup> ±0.02	ND	ND	0.24 <sup>bc</sup> ±0.02	0.04 <sup>ab</sup> ±0.01
TR2	0.83 <sup>e</sup> ±0.03	0.07 <sup>b</sup> ±0.02	9.03 <sup>e</sup> ±0.02	ND	ND	0.32 <sup>d</sup> ±0.01	0.06 <sup>bcd</sup> ±0.00
TR3	0.48 <sup>c</sup> ±0.03	0.04 <sup>a</sup> ±0.02	3.42 <sup>d</sup> ±0.09	ND	ND	0.36 <sup>d</sup> ±0.02	0.07 <sup>cd</sup> ±0.01
TRC1	0.10 <sup>a</sup> ±0.01	0.03 <sup>a</sup> ±0.01	2.71 <sup>a</sup> ±0.07	ND	0.03 <sup>b</sup> ±0.01	0.17 <sup>a</sup> ±0.01	0.06 <sup>abc</sup> ±0.01
TRC2	0.24 <sup>b</sup> ±0.02	0.03 <sup>a</sup> ±0.01	3.15 <sup>bc</sup> ±0.05	ND	ND	0.22 <sup>bc</sup> ±0.01	0.11 <sup>e</sup> ±0.01
TRC3	0.67 <sup>d</sup> ±0.01	0.02 <sup>a</sup> ±0.01	2.98 <sup>b</sup> ±0.03	ND	0.03 <sup>b</sup> ±0.01	0.25 <sup>c</sup> ±0.01	0.08 <sup>d</sup> ±0.01

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste, ND – Not detected

**Table 8: Weekly pH of water in the catfish aquaria**

Sample	WK1	WK2	WK3	WK4	WK5
Control	6.70 <sup>b</sup> ±0.00	6.20 <sup>a</sup> ±0.25	7.20 <sup>b</sup> ±0.05	7.30 <sup>a</sup> ±0.05	7.20 <sup>c</sup> ±0.00
TR1	6.98 <sup>cd</sup> ±0.03	6.60 <sup>b</sup> ±0.00	7.10 <sup>b</sup> ±0.00	7.00 <sup>c</sup> ±0.05	5.30 <sup>a</sup> ±0.15
TR2	7.20 <sup>f</sup> ±0.00	6.63 <sup>b</sup> ±0.03	6.70 <sup>a</sup> ±0.05	6.50 <sup>a</sup> ±0.05	6.50 <sup>d</sup> ±0.05
TR3	7.00 <sup>d</sup> ±0.00	6.60 <sup>b</sup> ±0.00	6.80 <sup>a</sup> ±0.00	6.70 <sup>b</sup> ±0.00	5.40 <sup>a</sup> ±0.00
TRC1	6.93 <sup>c</sup> ±0.03	6.70 <sup>bc</sup> ±0.00	7.20 <sup>b</sup> ±0.00	7.10 <sup>c</sup> ±0.00	6.20 <sup>bc</sup> ±0.05
TRC2	7.13 <sup>c</sup> ±0.03	6.90 <sup>bc</sup> ±0.00	7.15 <sup>b</sup> ±0.00	7.00 <sup>c</sup> ±0.00	6.10 <sup>b</sup> ±0.05
TRC3	6.40 <sup>a</sup> ±0.00	7.00 <sup>c</sup> ±0.05	7.20 <sup>b</sup> ±0.05	7.10 <sup>c</sup> ±0.05	6.40 <sup>d</sup> ±0.00

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste and WK - Week

**Table 9: Weekly biochemical Oxygen demand of water in catfish aquaria (mg/l)**

Sample	WK1	WK2	WK3	WK4	WK5
Control	7.31 <sup>b</sup> ±0.21	7.44 <sup>a</sup> ±0.04	0.00 <sup>a</sup> ±0.00	2.79 <sup>b</sup> ±0.15	2.59 <sup>a</sup> ±0.02
TR1	13.17 <sup>c</sup> ±0.03	3.05 <sup>bc</sup> ±0.10	5.75 <sup>e</sup> ±0.10	5.69 <sup>d</sup> ±0.20	4.44 <sup>a</sup> ±0.11
TR2	1.83 <sup>a</sup> ±0.10	2.76 <sup>b</sup> ±0.10	9.28 <sup>g</sup> ±0.04	10.55 <sup>f</sup> ±0.12	9.71 <sup>c</sup> ±1.58
TR3	10.57 <sup>d</sup> ±0.30	8.52 <sup>e</sup> ±0.12	8.56 <sup>f</sup> ±0.03	8.06 <sup>e</sup> ±0.32	6.66 <sup>b</sup> ±0.16
TRC1	16.46 <sup>f</sup> ±0.20	10.20 <sup>f</sup> ±0.08	5.16 <sup>d</sup> ±0.02	3.82 <sup>c</sup> ±0.11	4.51 <sup>a</sup> ±0.10
TRC2	8.12 <sup>c</sup> ±0.21	3.15 <sup>c</sup> ±0.05	3.13 <sup>c</sup> ±0.08	3.37 <sup>bc</sup> ±0.12	4.15 <sup>a</sup> ±0.03
TRC3	10.58 <sup>d</sup> ±0.37	1.02 <sup>a</sup> ±0.10	1.87 <sup>b</sup> ±0.04	1.92 <sup>a</sup> ±0.08	2.52 <sup>a</sup> ±0.26

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste and WK - Week

**Table 10: Weekly temperature of water in catfish aquaria (°C)**

Sample	WK1	WK2	WK3	WK4	WK5
Control	23.35 <sup>b</sup> ±0.25	23.80 <sup>c</sup> ±0.00	23.80 <sup>abc</sup> ±0.00	23.85 <sup>a</sup> ±0.05	22.70 <sup>d</sup> ±0.10
TR1	22.95 <sup>ab</sup> ±0.05	23.70 <sup>ab</sup> ±0.00	23.75 <sup>ab</sup> ±0.05	23.80 <sup>a</sup> ±0.00	22.45 <sup>c</sup> ±0.05
TR2	22.90 <sup>a</sup> ±0.10	23.65 <sup>a</sup> ±0.05	23.85 <sup>bc</sup> ±0.05	23.80 <sup>a</sup> ±0.00	22.00 <sup>a</sup> ±0.00
TR3	23.00 <sup>ab</sup> ±0.10	23.75 <sup>bc</sup> ±0.05	23.80 <sup>abc</sup> ±0.00	23.40 <sup>a</sup> ±0.40	22.00 <sup>a</sup> ±0.00
TRC1	22.95 <sup>ab</sup> ±0.05	23.80 <sup>c</sup> ±0.00	23.90 <sup>c</sup> ±0.00	23.80 <sup>a</sup> ±0.00	22.20 <sup>b</sup> ±0.00
TRC2	22.85 <sup>a</sup> ±0.05	23.80 <sup>c</sup> ±0.00	23.70 <sup>a</sup> ±0.00	23.80 <sup>a</sup> ±0.00	22.20 <sup>b</sup> ±0.00
TRC3	22.90 <sup>a</sup> ±0.00	23.80 <sup>c</sup> ±0.00	23.75 <sup>ab</sup> ±0.05	23.70 <sup>a</sup> ±0.00	22.20 <sup>b</sup> ±0.00

Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste and WK - Week

**Table 11: Weekly titratable acidity of water in catfish aquaria**

Sample	WK1	WK2	WK3	WK4	WK5
Control	0.06 <sup>a</sup> ±0.00	0.08 <sup>a</sup> ±0.02	0.16 <sup>bc</sup> ±0.02	0.27 <sup>c</sup> ±0.03	0.27 <sup>b</sup> ±0.03
TR1	0.08 <sup>a</sup> ±0.02	0.08 <sup>a</sup> ±0.02	0.18 <sup>c</sup> ±0.01	0.23 <sup>bc</sup> ±0.02	0.12 <sup>a</sup> ±0.00
TR2	0.06 <sup>a</sup> ±0.00	0.08 <sup>a</sup> ±0.01	0.07 <sup>a</sup> ±0.01	0.15 <sup>a</sup> ±0.03	0.09 <sup>a</sup> ±0.03
TR3	0.05 <sup>a</sup> ±0.02	0.09 <sup>a</sup> ±0.02	0.15 <sup>bc</sup> ±0.03	0.18 <sup>ab</sup> ±0.00	0.09 <sup>a</sup> ±0.03
TRC1	0.03 <sup>a</sup> ±0.00	0.08 <sup>a</sup> ±0.022	0.12 <sup>ab</sup> ±0.01	0.23 <sup>bc</sup> ±0.02	0.12 <sup>a</sup> ±0.00
TRC2	0.06 <sup>a</sup> ±0.03	0.08 <sup>a</sup> ±0.01	0.12 <sup>b</sup> ±0.00	0.18 <sup>ab</sup> ±0.00	0.12 <sup>a</sup> ±0.00
TRC3	0.06 <sup>a</sup> ±0.00	0.08 <sup>a</sup> ±0.00	0.14 <sup>bc</sup> ±0.02	0.20 <sup>ab</sup> ±0.02	0.09 <sup>a</sup> ±0.03

**Key: TR1 – Polluted with 25g of e-waste soil, TR2 – Polluted with 50g of e-waste soil, TR3 - Polluted with 75g of e-waste soil; TRC1 – Polluted with 25g of soil without e-waste, TRC2 – Polluted with 50g of soil without e-waste, TRC3 - Polluted with 75g of soil without e-waste and WK – Week**

### DISCUSSION

Soil from e-waste dumpsite where e-waste is disposed by burning and soil from without e-waste dumpsite were analyzed for the presence of heavy metals. The result revealed that soil from burnt e-waste dumpsite has the highest quantity of heavy metals in part per million (Table 1), this could be as a result of burning of e-waste on open land leads to increased in the release of heavy metals in the soil. A total of thirteen fungi were isolated from the e-waste soil sample, soil without e-waste sample, polluted catfish waters and harvested catfish parts. *Aspergillus flavus*, *Mucor mucedo* has been associated with fish spoilage [12], [22]. *Articulospora inflata*, *Zoopage nitospora*, *Varicosporium elodeae*, *Penicillium* sp, *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus stolonifer*, and *Mucor mucedo* had been isolated from many environments (such as crude oil polluted environment, gastrointestinal tract, agricultural soil) and their presence also in these environments (e-waste soil, soil without e-waste, polluted catfish waters and harvested catfish parts) could have been as a result of their ability to adapt to different environmental conditions and using of wide range of food substances as nutrient source [6], [22]. Some fungi which were not isolated at the beginning of the research were later isolated at the final stage of the research this is in line with the succession of a substrate, an area or polluted area by microorganisms [9], [28]. The fungi counts monitored (Figure 1 and 2) showed fungi counts decreases as the quantity of e-waste soil pollution increases and increases as the quantity of soil without e-waste pollution increases. These showed the effect of the pollution on the fungi in this environment which correlates the findings of [24] on the effect of oil pollution on an environment. E-waste soil and e-waste soil polluted catfish aquaria had higher number of isolates compared to soil without e-waste and soil without e-waste polluted catfish water. These can be attributed to high percentage of organic contents (carbon, organic matter, nitrogen) and moisture content of the e-waste soil, which might have encouraged and supported

the growth of those microbes. This is in conformity with the findings of [24] about microbial needs for growth. Fungi isolation and identification were determined weekly and a total of 1161 fungi from *Clarias gariepinus* (catfish) aquaria were isolated after 5 weeks (Table 4). The number of fungi isolated from each pollution treatments showed that the number of fungi isolated decreases with increases in e-waste soil pollution and increases with increase in soil without e-waste pollution. *Articulospora inflata*, *Zoopage nitospora*, *Candida* sp, *Penicillium italicum*, *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus stolonifer*, *Mucor mucedo*, *Varicosporium elodeae* and *Aureobasidium pullulans* were isolated from the e-waste soil, soil without e-waste, soil without e-waste polluted aquaria and e-waste soil polluted catfish aquaria for the period of the research (5 weeks) (Tables 2, 3 and 5), these fungi might have developed tolerance to this polluted environment or has the ability to use substances from the pollutant in the polluted environment [4], [8], [33], [36] and probably they can be used or manipulated to bioremediate or in biosorption of pollutants from e-waste. This is in line with the biosorption of heavy metals ions using *Aspergillus niger* by [20], removal (bioremediation) of heavy metals using *Aspergillus niger* and *Aspergillus flavus* by [36], which also agrees with the findings of [19], that fungi of metal contaminated soil have high level of metal tolerance and biosorption properties. The total numbers of fungi isolated from the body parts (skin, liver, gills and intestine) of catfish were 538 (Table 6). The number isolated from the fish parts in each aquarium reduces with increase in e-waste soil pollution and increases with increase soil without e-waste pollution, the same trend of fungi isolation from the fish water followed. This correlates the findings of [24]. The highest fungi isolates were from intestine (160) while the lowest fungi isolates were from the gills (110), this could be an indication of the type of feeding habit of the fish and part of the fish in which food interact more. High numbers of fungi (140) (Table 6) were also isolated from the liver this could be an indication of high detoxification processes undertaken by the liver which

probably supported the high fungi growth. This is in conformity with reports of [10], [25], [32] that the exposure of fish (fishes are known to possess the metallothioneine proteins) to elevated levels of heavy metals induces the synthesis of metallothioneine proteins (MT), which are metal binding proteins. Metallothioneine proteins have high affinities for heavy metals and in doing so, concentrate and regulate these metals in the liver and metallothioneine proteins bind and detoxify the metal ion. Fungi of considerable number were also found on the skin part of these fishes this is probably because the skin of fish is usually in direct contact with water.

It was observed on the mixed culture of many of the fungi plates that the growth of some fungi (*Aspergillus flavus*, *Aspergillus paraciticus*, *Aspergillus niger*, *Aspergillus repens*, *Penicillium italicum*, *Mucor mucedo*, *Zoopage nitospora* and *Rhizopus stolonifer*) on many of the fungi plates lead to reduction in the population and poor growth of other fungi on the same plate. This could be due to the production of some inhibitory substances such as toxin or toxic metabolites (aflatoxins, cyclopiazonic acid, ochratoxins, kojic acid) which might have affected or inhibited the growth of the other fungi on the plate. It correlates the report of [11] that reported that some of these fungi secrete aflatoxins and other substances that are capable of inhibiting the growth of some microorganisms and can lead to declination and eventual death of some of these microorganisms that were found present at the early stage of fermentation. The high numbers of *Aspergillus* species in exhibiting this probable inhibitory property in this research attest to the known ability of many of the species in the general of *Aspergillus* to release harmful or inhibitory substances especially aflatoxin [22], [39].

Fish has been reported to accumulate metals from water by diffusion via skin and gills as well as oral consumption [26], [27]. 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. Bioaccumulation of the heavy metals in the fish's tissue in this research was proportional to the concentration of e-waste soil pollution and the quantity of the heavy metals present in the soil samples (Table 7). The research also showed that some of the heavy metals were poorly bioaccumulated while cobalt was not bioaccumulated, these could be due to the inability of these fishes to absorb the heavy metals in their tissues and probably the presence of enzymes that can detoxified the heavy metals or have the ability to metabolize them. This result correlated the reports of [40] who worked on cyanide bioaccumulation, that cyanide bioaccumulation and biomagnification in food web has not been established, possibly due to rapid detoxification of sub-lethal doses by most species, and death at higher doses. The quantity of lead, cadmium and zinc absorbed by catfish is more than chromium, manganese and nickel, these could be due to the presence of the types of

enzymes for their metabolic activities, this is in agreement with the work of [5], [18] that accumulation of metal in different species is the function of their respective membrane permeability and enzyme system, which is highly species specific and because of this fact metals accumulated differently in the tissues of fishes. Enzymes have a great potentiality to effectively transform and detoxify polluting substances because they are able to transform pollutants to less toxic forms [3], [29], [34], [41]. The bioaccumulations of the heavy metals by *Clarias gariepinus* were within the acceptable limits for most of the heavy metals observed in the period of the research (5 weeks) and may be exceeded if these fishes are exposed for a longer time and the resultant increased in the metal concentrations can be toxic to fishes and render the water unsuitable for other uses [2]. The maximum recommended limit for manganese was exceeded by *Clarias gariepinus* (Table 7) (Mn accumulates from 0.16ppm in TRC1 to 0.38ppm in TR3, recommended limit is between 0.01 – 0.05ppm [13].

The e-waste soil pollution affects the pH and the BOD<sub>5</sub> of the culturing water. The pH (Initial pH of water used was 6.40 ± 0.00, pH of e-waste and off e-waste soils are 7.9 and 8.70 respectively) was shifting between neutrality (ranges from 6.55 ± 0.05 – 7.20 ± 0.00) in week 1, 3, 4 and acidic (5.15 ± 0.15 – 6.95 ± 0.05) in week 2 and 5 with e-waste soil polluted catfish water having lesser values for pH compared to the off e-waste soil polluted fish water. This variation in pH (Table 8) could have resulted from the fermentative activities of the microorganisms present [31]. It could also have resulted from interaction between the materials and the hydrogen ions from the soil types (e-waste and off e-waste soil), water used and catfish waste products or metabolites. Catfish waste, especially unionized ammonia (NH<sub>3</sub>) and the ionized ammonium (NH<sub>4</sub><sup>+</sup>) released into the fish water is likely to be low probably catfish conversion rate (that is the conversion of protein in food materials to body tissue and nitrogenous compounds) is low, therefore the influence on the pH of the fish water should be more from the soils types and temperature. Feeding rate, metabolism, decomposition of metabolite and organic matters are temperature dependant, at low temperature these activities will be slower compared to the rate at optimum temperature 26°C – 32°C and catfish water temperature in this research ranges from 22°C – 23.9°C which might have influence the pH of the fish water. The lower pH values in the e-waste polluted fish water could be due to higher dissolved CO<sub>2</sub> from the decomposition of organic matters which was higher in the e-waste soil.

Day 5 analysis of the BOD (Table 9) shows that differences between the two soil types polluted fish water was only evident at week 3 – 5, with e-waste soil polluted catfish water having higher BOD compared to soil without e-waste polluted catfish these could be due to the organic matter presence, types of metabolites and the rate of release of these metabolites.

The significant different at  $P \leq 0.05$  between the titratable acidity (Table 11) of polluted aquaria and the control of the fish (*Clarias gariepinus*) showed that the e-waste soil pollution has effect on the titratable acidity of the fish water, since the results of the polluted aquaria were lower than the unpolluted (control). The control aquarium titratable acidity values ranges from 0.06 – 0.30 and the polluted aquaria ranges from 0.06 – 0.24 while 0.12 – 0.36 in control aquaria. These differences are probably due to the interference in the fermentative processes of the microorganisms present by the pollutants in the polluted aquaria giving rise to lower titratable acidity in these aquaria, since titratable acidity and fermentation has a relationship. This is in agreement with the report of [31] that variation in titratable acidity and pH is a function of the fermentative activities of microorganisms especially lactic acid bacteria present in a product and the length of fermentation.

## CONCLUSION

The study of heavy metals from e-waste has contributed to understanding of bioaccumulation in catfish (*Clarias gariepinus*) and associated fungi. Since e-waste production has increased in recent years, the need to create more public awareness of the potential harm of some e-waste constituents (heavy metals) is necessary, many products that end up as e-waste such as electrical materials, electrical appliances, both household and office equipments are widely used and commonly discarded or incinerated. Substantial dangers therefore continue to be presented by release of heavy metals through e-waste disposal into the environment if e-waste is not properly disposed.

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