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Acute Toxicity of House Boat Effluents on Paleamonetes africanus and Tilapia guineensis.

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Abstract: *Acute toxicity of house boat effluents (black water, grey water, ballast water and slop water) was tested on aquatic biota (Paleamonetes africanus and Tilapia guineensis). The toxicity assay based on lethal concentration (LC_{50}) able to kill 50% of the test organism showed at 48hrs, 72hrs and 96hrs exposure the LC_{50} for black water on Paleamonetes africanus were 25.44, 2.43 and 0.92ppt respectively, 44.71, 2.93, and 0.27ppt respectively were recorded for grey water. The ballast water had LC_{50} values of 83052.90, 3030.27, and 690.69ppt respectively, 52.50, 0.71, and 0.47ppt respectively was recorded for slop water. Against Tilapia guinensis the values recorded were 335.98, 4.46, and 4.27ppt for black water, 39.81, 25.84, and 2.28ppt for grey water, 19380.11, 1301.31, and 270.36ppt for ballast water 68.83, 18.30, and 4.14ppt for slop water. Grey water demonstrated the highest toxic effect on Paleamonetes africanus and Tilapia guineensis, However, ballast water had the least toxic effect on both test organisms. The study further revealed that continuous disposal of house boat effluent untreated into aquatic system will negatively impact the aquatic biota.*

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I. Introduction

Houseboat generally refers to a boat that has been engineered, designed or modified to be used primarily for housing purposes. Some houseboats are not self propelled, because they are usually towed, positioned at a particular location and often anchored to land to provide utilities. As a result of human activities in the house boat, waste are generated and generally referred to as effluents or waste water.

According to the United States Environmental Protection Agency Effluent is defined as "wastewater (treated or untreated) from a treatment plant, sewer, or industrial outfall. Effluents are an out flowing of liquid wastes or gas from anthropogenic activities (Blinova, 2000). Effluent generally is considered to be a source of water pollution, and could involve cases such as discharge of sewage from a treatment facility or the wastewater discharged from industrial facilities (Joel *et al*, 2009).

Some commonly generated effluents from house boats include:

- Grey water
- Black water
- Ballast water
- Slop water

These above named effluents are mostly discharged into the receiving environment (aquatic system) usually untreated and they act as a source of contamination of the receiving waters. The indiscriminate disposal of effluent into recipient aquatic waters result in environmental pollution which is the unfavorable change in the earth's physical, chemical, and biological components in such a degree that normal environmental processes are affected negatively (Odokuma and Akponah, 2008). Since the site of pollution is the aquatic system, it could be further said that indiscriminate disposal of effluents leads to aquatic pollution, which is the contamination of the natural water bodies by chemicals, physical and biological contaminants affecting the entire biosphere. Different marine pollutants such as oil, industrial effluents, domestic effluents and agricultural materials contribute to unhealthy marine lives and even extinction of many aquatic organisms.

However, Department of Petroleum Resources (DPR) requires biological treatment, preferably by a standard sewage Wastewater Treatment Plant (WWTP). This will be followed by mandatory monitoring of dissolved oxygen (DO), 5day-biochemical oxygen demand (BOD_5), total suspended solids (TSS), residual chlorine and faecal coliform in treated sewage before discharge into the sea (DPR, 2002).

Effluents (Grey water, Black water, Ballast water, and Slop water), from house boats are source of contaminant to the recipient environment when discharged into them untreated. In Nigeria, where proper sewage and effluent treatment systems are lacking and the disposal of waste is indiscriminate especially into aquatic system, serious environmental risk often is the case. Possible mortality of the aquatic biota could occur as a result of this discharge. Heavy metals which are also a component of these wastes can also bioaccumulate in organisms. The aquatic organisms affected by such indiscriminate disposals include Fishes e.g. *Tilapia guineensis,* Crustaceans e.g. *Paleamonetes africanus* e.t.c. This study gears towards determining the toxicity of the effluents discharged from Houseboat on aquatic/marine biota.

Toxicity is the extent to which a substance can cause damage to an organism (Odokuma and Akponah, 2008). Toxicity can refer to the response of a whole organism, such as an animal, bacterium, or plant (Okpokwasili and Odokuma, 1994).

II. Materials And Methods

Sample Collection

The effluents (Gray water, Black water, Ballast water, and Slop water) were obtained using sterile ten (10) liter rubber cans for each of the effluent from a House Boat in Asaramatoru/Bonny Estuary of Andoni Local Government Area, Rivers State, Nigeria.

Sources of microorganisms for toxicity test

(a) The brackish water juvenile shrimps (*Palaemonetes africanus*) were collected from Nigerian Institute for Oceanography and Marine Research (NIOMR).
(b) The brackish water fish (*Tilapia guineensis*) were collected from Nigerian Institute for Oceanography and Marine Research (NIOMR).

The choice of these species was based on their availability throughout the year and ease of handling. This is in accordance with the recommendation of DPR, (2002).

Acclimatization procedure for *Palaemonetes africanus* and *Tillapia guineensis*

All test organisms were initially acclimatized for ten days at room temperature (28-30^{o}C). The organisms were set in a holding tank, and circulated air through by the assistance of an aerator (APHA, 1998). The holding brackish water was changed on a daily bases to increase nutrient availability and removed unwanted pollutants.

Semi-static bioassay techniques for *Palaemonetes africanus* and *Tillapia guineensis*

Acute toxicity tests were done with Aquatic organisms (*Palaemonetes africanus* and *Tillapia guineensis*) by presenting the organisms to the toxicants at different concentrations, utilizing the semi-static agitation test procedure, suggested by the Department of Petroleum Resources (DPR) for a time of 96hours (DPR, 2002).

Selection of test organisms

After the acclimatization period, ten test organisms of fairly equal size were randomly caught with the aid of hand net and carefully transferred into the test container. Only healthy and active organisms were selected.

Test medium

Five different concentrations (0.01ppt, 0.1ppt, 1.0ppt, 10ppt, 100ppt and 1000ppt) of each of the effluent samples (Grey water, Black water, Ballast water, and Slop water), were prepared using the habitat water as the diluents.

Toxicity test procedure for *Tilapia guineensis* and *Paleamonetes africanus.*

The procedure used for toxicity was adapted from Odokuma and Akponah, (2008). Test containers with concentrations (0.01ppt, 0.1ppt, 1.0ppt, 10ppt, 100ppt and 1000ppt) prepared by diluting the effluent with the brackish water, and a control without any effluent concentration was setup. The containers were labeled appropriately according to their concentrations. Ten (10) of the active test organisms of fairly equal sizes were carefully transferred with the aid of hand net into each container. The same experimental setup was prepared for the two test organisms *Tilapia guineensis* and *Paleamonetes africanus.*

The setups were properly aerated with an aerator, and observations were made every 24hours for 96 hours to ascertain the mortality of the test organisms. At the end of each exposure period, dead organisms were removed. The rate log survival of the organisms in the toxicant was computed by getting the count in every toxicant concentration and dividing by the count in the zero toxicant concentration and then multiplying the product by 100.

Therefore %survival = $C \times \frac{100}{c}$

Where C = count in each toxicant concentration,

c = count in the zero toxicant concentration.

Determination of LC_{50}

Lethal dose of the toxicant that will execute half of the test organism (LC_{50}) was determined for the higher organism life forms (*Tilapia guineensis* and *Paleamonetes africanus).*

III. Results

Effects of concentration of effluents on the growth of the test organisms

The effect of different concentrations of black water (0.01, 0.1, 1.0, 10, 100, and 1000) ppt respectively on the test organism (*Paleamonetes africanus*) showed that there was a decrease in the percentage survival of the organism with increase in the concentration of the toxicant (black water) and contact time. Although at lower concentrations (0.01, 0.1, and 1.0) ppt, there was no mortality after 48hrs, however, at higher concentrations of the toxicant (100 and 1000) ppt respectively, the mortality was high with the highest mortality recorded at 1000 ppt concentration of the toxicant, in which all organisms died after 48hrs. Grey water expressed a decrease in percentage survival of *Paleamonetes africanus* at concentration of 100ppt after 24hrs of the toxicity testing. It was also observed that at concentration of 1.01ppt, even after 72hrs, the percentage survival was 100%. The higher concentration of the toxicant and the longer exposure period showed detrimental effects on the test organism. A 1000ppt concentration at 72hrs was able to kill the entire test organism, bringing the % mortality to 100 percent. Among all the toxicant utilized as a part of this review, ballast water was seen to have minimal impact on *Paleamonetes africanus*. Indeed, even at high concentrations of 1000ppt and 96hrs of exposure of the test organism to the toxicant, the rate survival of the organism was still at 50 percent. In any case, it was observed that there was a decline in the rate survival with increment in concentration of the toxicant and increment in the contact time. The different concentrations of 0.01, 0.1, and 1.0 ppt had no inhibitory effect on *Paleamonetes africanus* after 72hrs exposure period; however, concentrations of 10, 100, and 1000ppt had little effects at 96hrs exposure period. The inhibitory impact of slop water on the test organism (*Paleamonetes africanus*) was observed to increase the rate mortality with an increasing in the concentration of the toxicant (Slop water) and an increment in the contact time. Inhibitory impacts were seen at concentrations of 1.0ppt, which brought the %mortality after 24hrs to 10%. Be that as it may, a sharp decrease in the mortality rate was seen as concentration of 1000ppt after 48hrs contact period indicated 100% mortality. Concentration 10, 100, and 100ppt was seen to kill 100% of the test organism at 96hrs exposure period. Indeed, even at low concentrations of 0.01ppt, the percentage mortality after 96hrs exposure was as high at 60%.

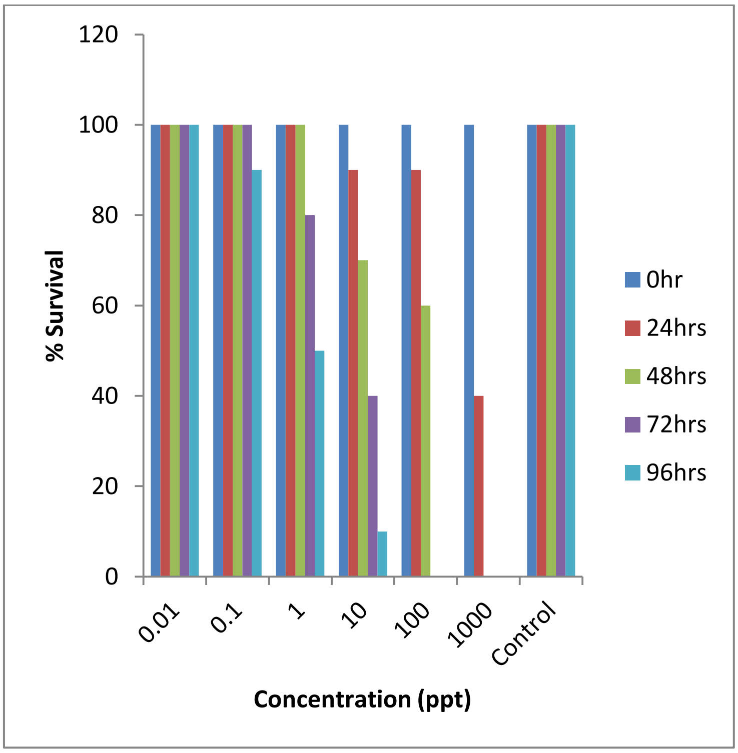

Figure 1: Effect of concentration of black water on survival of *Paleamonetes africanus*

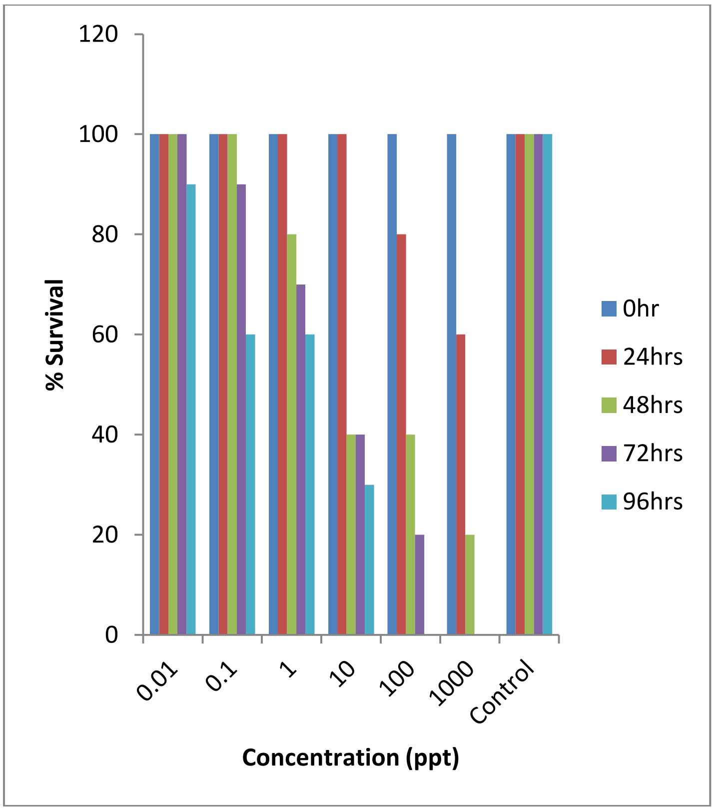

Figure 2: Effect of concentration of grey water on survival of *Paleamonetes africanus*

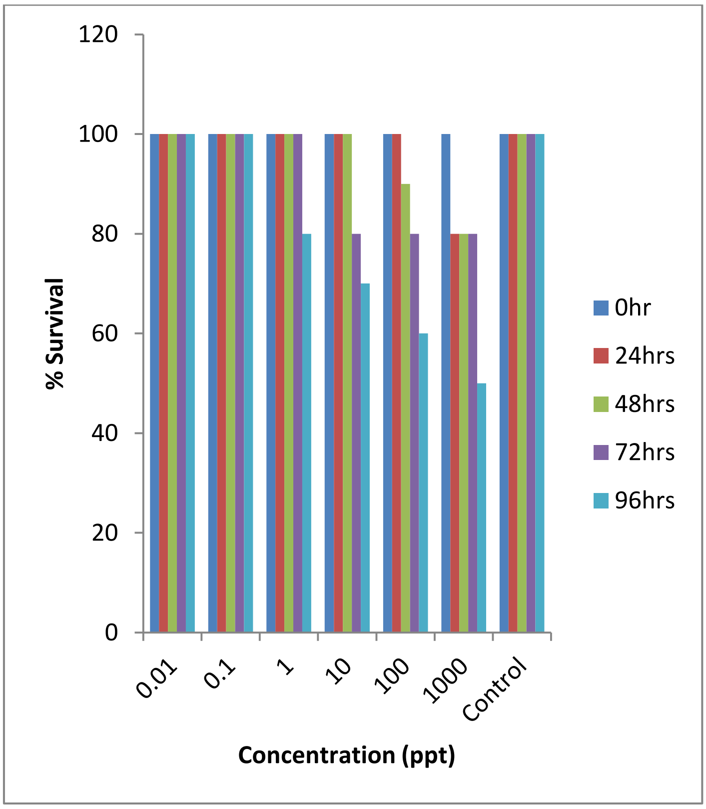

Figure 3: Effect of concentration of ballast water on survival of *Paleamonetes africanus*

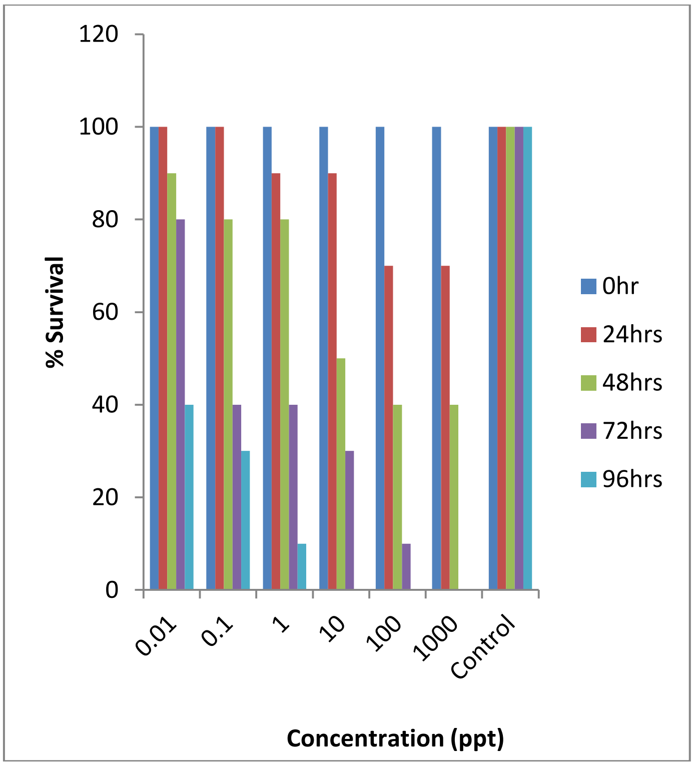

Figure 4: Effect of concentration of slop water on survival of *Paleamonetes africanus*

However, on the other hand there was a decrease in the percentage survival of the *Tilapia guineensis* with increment in concentration of the toxicant and expanding contact time (Exposure period). Concentrations of 0.01, 0.1, and 1.0ppt did not record any mortality at 48hrs exposure period; however concentrations 0.1 and 1.0 showed 10% and 20% mortalities recorded at 72hrs exposure period. The lowest concentration 0.01ppt, after 96hrs contact time had no inhibitory impact on the test organism (*Tilapia guineensis*). It was observed that there were no survivors in the highest concentration of 1000ppt after 72hrs contact time. Higher concentrations of 10, 100, and 1000ppt had the highest negative effect on *Tilapia guineensis.* As compared with toxic effects of grey water on *Paleamonetes africanus*, grey water had a less negative effect on *Tilapia guineensis*. Concentration f 10ppt was observed to demonstrate some rate mortality on *Paleamonetes africanus* after 24hrs of exposure, while at a similar concentration and contact time, no mortality was recorded for *Tilapia guineensis*. In any case, at increasing concentrations of the toxicant and expanding contact period, the rate survival of the test organism (*Tilapia guineensis*) diminished, without any survivors at 1000ppt concentration after 96hrs exposure period. Concentrations of 0.01, 0.1, and 1.0ppt showed little or no mortality rate after 48hrs exposure of the toxicant. The increasing concentration of the contaminant (ballast water) and furthermore expanding contact time (exposure period) affected the test organism (*Tilapia guineensis*). It was observed that a diminishing in the rate survival of the organism with increasing concentration of toxicant and contact time was the pattern. Despite the fact that 50 percent of the organism survived concentration as high as 1000ppt after 96hrs exposure period, concentration as low as10ppt after 48hrs presentation period negatively affected the test organism showing with 10% mortality. At lower concentrations of the toxicant (0.01, 0.1, and 1.0)ppt, and 72hrs exposure period, there was no decrease in the percentage survival and lower concentrations of 0.01 and 0.1ppt after 96hrs exposure period recorded 100% survival. Finally, after 24hrs contact time, there was a diminishing in rate survival in the setup containing 1000ppt concentration of the toxicant. In any case, there was a general diminishing in the rate survival with increasing concentrations of the toxicant and contact time. The most elevated mortality of 100% was recorded in concentrations of 1000ppt at 96hrs contact period. The lower concentrations of 0.01, 0.1, and 1.0ppt had minimal negative impact, though higher concentrations of 10, 100, and 1000ppt demonstrated the highest decrease in the rate survival of the test organism with increasing time.

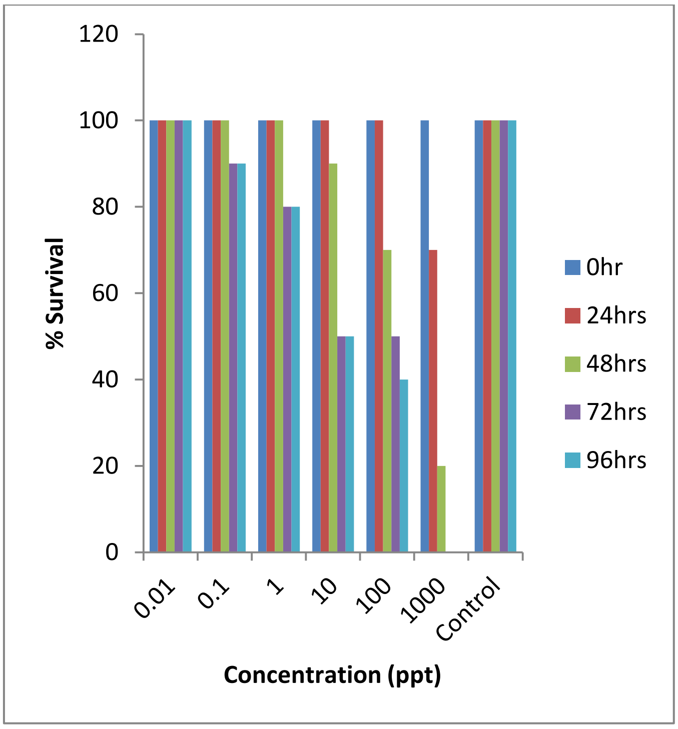

Figure 5: Effect of concentration of black water on survival of *Tilapia guineensis*

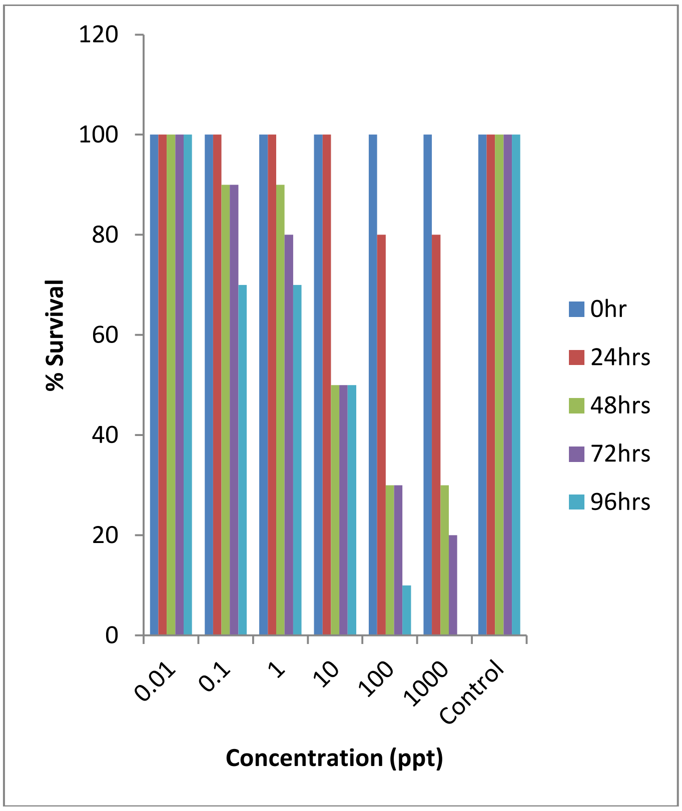

Figure 6: Effect of concentration of grey water on survival of *Tilapia guineensis*

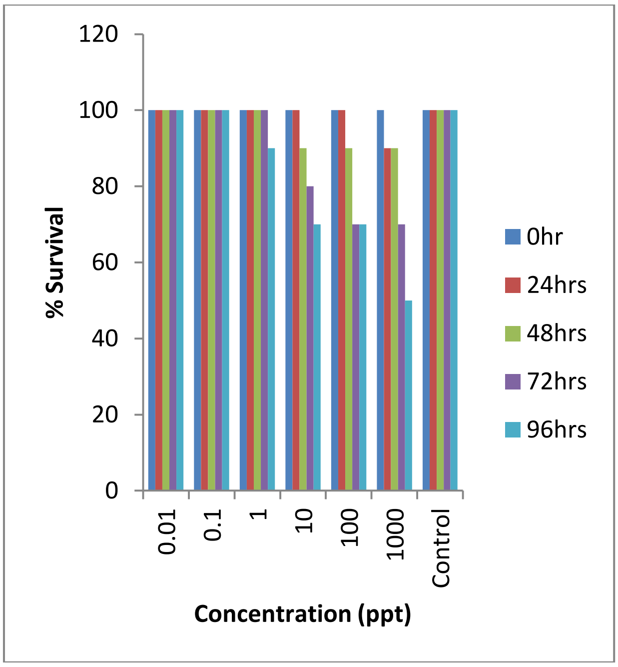

Figure 7: Effect of concentration of ballast water on survival of *Tilapia guineensis*

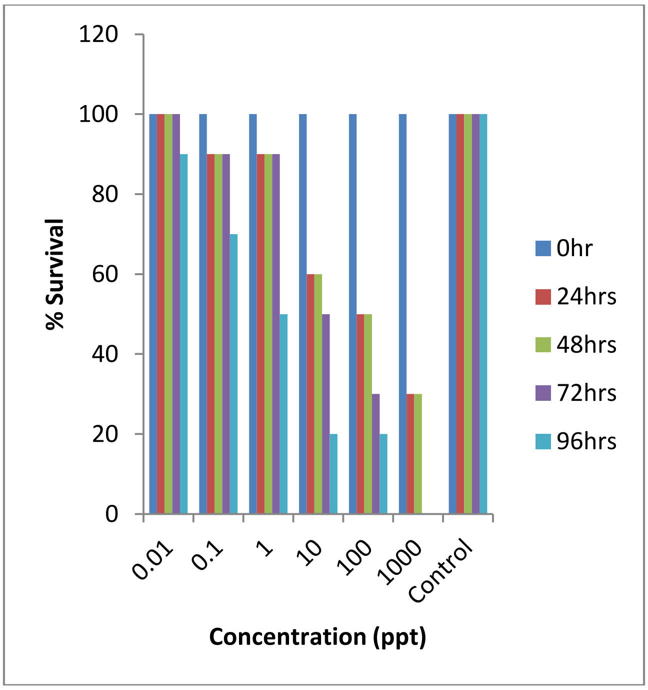

Figure 8: Effect of concentration of slop water on survival of *Tilapia guineensis*

Lethal Concentration (LC_{50}) of the effluents on the test organisms.

Probit was used to determine the lethal concentration (LC_{50}) of the toxicants on the test organisms. Black water for *Paleamonetes* expressed 48, 72, and 96 hours LC_{50} values to be 25.44, 2.43, and 0.92ppt respectively. The LC_{50} values are represented in table 1. At 48hours, the LC_{50} of black water for *Paleamonetes africanus* was 25.44ppt, at 72hours it was 2.43ppt, while at 96hours the LC_{50} reduced to 0.92ppt. The result revealed that the LC_{50} of black water decreased with increasing contact time of toxicant for *Paleamonetes africanus*. Grey water for *Paleamonetes africanus* had LC_{50} values for 48, 72, and 96 hours respectively of 44.71, 2.93, and 0.27ppt. After 96hrs of exposure period, it was observed that a very low concentration (0.27ppt) of the grey water was able to kill 50% of *Paleamonetes africanus.* The (LC_{50}) of water used for ballast purposes for *Paleamonetes africanus* at for 48, 72, and 96 hours respectively were 83052.90, 3030.27, and 690.69ppt. From the result obtained ballast water used in this study expressed the least toxicity to the test organism (*Paleamonetes africanus).* However, there was a decrease in the LC_{50} value of ballast water with increasing exposure time to the ballast water for *Paleamonetes africanus*. The LC_{50} values of slop water on *Paleamonetes africanus* for 48, 72, and 96 hours respectively were 52.50, 0.71, and 0.47ppt. A lower value for LC_{50} of slop water after 72 and 96hrs exposure period was recorded. This result indicates that at even vey minute concentrations of the slop water, expressed toxic effects on *Paleamonetes africanus*.

On the other hand, the lethal concentration (LC_{50}) of black water for *Tilapia guineensis* for 48, 72, and 96 hours respectively were 335.98, 4.46, and 4.27ppt. The data obtained demonstrated a decrease in the LC_{50} of black water as exposure time to the toxicant increased. Grey water for *Tilapia guineensis* had LC_{50} values for 48, 72, and 96 hours respectively of 39.81, 25.84, and 2.28ppt also expressing a decrease in the LC_{50} of grey water with increase in the exposure time of the toxicant for *Tilapia guineensis*. Ballast water for *Tilapia guineensis* had LC_{50} values for 48, 72, and 96 hours respectively of 19380.11, 1301.31, and 270.36ppt. From the results which are dominated by high values of $LC_{50,}$ indicates that the test organism was tolerant to the ballast water. However, the data obtained, demonstrated a decrease in the LC_{50} of ballast water with increasing exposure time of the toxicant for *Tilapia guineensis* was observed. A low LC_{50} was recorded after 96hrs contact time for slop water suggesting that the toxicant at concentration 4.14ppt killed 50% of the *Tilapia*. From the data obtained, a decrease in the LC_{50} of slop water with increase in the exposure time of the toxicant for *Tilapia guineensis* was observed.

Table 1: Lethal concentrations (LC_{50}) of effluents on *Paleamonetes africanus.*

Effluent/ Organism	46 hrs (LC_{50})	72 hrs(LC_{50})	96 hrs(LC_{50})
Black water on *Paleamonetes africanus*.	25.44	2.43	0.92
Grey water on *Paleamonetes africanus*	44.71	2.93	0.27
Ballast water on *Paleamonetes africanus*	83052.90	3030.27	690.69
Slop water on *Paleamonetes africanus*	52.50	0.71	0.47

Table 2: Lethal concentrations (LC_{50}) of effluents on *Tilapia guineensis*

Effluent/ Organism	46 hrs (LC_{50})	72 hrs(LC_{50})	96 hrs(LC_{50})
Black water on *Tilapia guineensis*	335	4.46	4.27
Grey water on *Tilapia guineensis*	39.81	25.84	2.28
Ballast water on *Tilapia guineensis*	19380.11	1301.31	270.36
Slop water on *Tilapia guineensis*	68.83	18.30	4.14

IV. Discussion And Conclusion

It is impossible to unequivocally attribute the toxicity of complex mixtures such as effluents to one or few compounds (Nrior and Odokuma., 2015). However, for the toxicity testing of the effluents, abatement in the rate survival of *Paleamonetes africanus* and *Tilapia guineensis* with increasing contact time and exposure of the effluents was noticed. This outcome is in concurrence with perceptions of Odokuma and Akponah, (2008).

Several indices for toxicity of a few toxicants have been illustrated, be that as it may, used was lethal toxicity (LC_{50}) able to kill half of the test organism. A static batch system was utilized for the microorganisms and static batch with renewal utilized for the higher organisms (Odokuma and Akponah, 2008).

The LC_{50} of the toxicants for various test organism at 48hrs - LC_{50}, 76hrs - LC_{50}, and 96hrs - LC_{50}, to decide the concentration of the toxicant that will kill half of the test organism after 48, 72, and 96hrs separately. The LC_{50} was figured using probit in Microsoft excel at 95% confidence limit.

The response of *Paleamonetes africanus* to black water on the principal day (0h) exhibited 100% survival. In any case, this has been credited to the lag period in which the organisms need to acclimatize to the toxicant. With increment in the contact time and concentration of the toxicant, a decrease in the rate survival of the organism was recorded. The most astounding mortality was recorded after 96hrs of exposure of the organism to the toxicant. The 48hrs - LC_{50} for black water on *Paleamonetes africanus* was calculated to be 25.44ppt, 72hrs - LC_{50} was 2.43ppt, and 96hrs - LC_{50} was 0.92ppt. The outcome in LC_{50} shows that with increment in contact time and concentration of the toxicant led to decrease in LC_{50}, which means a lower concentration of the toxicant could kill half of the test organism after 96hrs exposure period. A concentration as low as 0.92ppt was found to kill the test organism after 96hrs, showing that the black water is toxic to *Paleamonetes africanus*.

The toxicity of grey water on *Paleamonetes africanus* was established by calculating the 48hrs - LC_{50}, 72hrs - LC_{50}, and 96hrs - LC_{50} respectively. The result obtained were 44.7ppt, 2.93ppt, and 0.27ppt respectively. The result however portrays that grey water was more toxic to *Paleamonetes africanus* than black water, as lower concentrations of grey water was able to kill the test organism than those observed in black water after 48hrs, 72hrs and 96hrs.

Another toxicant tested for was ballast water which had the least toxic effect on *Paleamonetes africanus*. The 48hrs - LC_{50} was 83052.90ppt, 72hrs - LC_{50} was 3030.27ppt, and 96hrs - LC_{50} was 690.69ppt.

The LC_{50} pattern of ballast water declined at 96hrs to a value less than 1000. The results obtained suggest that the ballast water did not exhibit toxic effects to *Paleamonetes africanus*. On the other hand, slop water demonstrated a reverse in LC_{50} pattern. 48hrs - LC_{50} value was 52.50ppt, after which a decrease in the value to 0.71ppt as the 72hrs - LC_{50} ensued. 96hrs - LC_{50} was 0.47ppt.

This simply indicates that very minute amount of slop water was inhibitory to the organism as 65.4ppt of the slop water was able to kill 50% of the test organism after 96hrs.

Grey water had the highest negative effect on *Paleamonetes africanus*, as 0.27ppt of the toxicant killed 50% of the test organism after 96hrs. Ballast water had the least negative effect, suggesting that the organisms were more tolerant to the toxicant. However, looking at the physicochemical parameters of both toxicant, it was discovered that some physicochemical parameters such as BOD, Do and COD values of the ballast water was within regulatory limits, while grey water was observed to have values higher that stimulatory regulatory limits. This report agrees with the findings of Nrior and Odokuma, (2015), that suggests that high BOD, DO, and COD values have been found to have deleterious effects on aquatic organisms

Another organism for which the toxicants were tested was *Tilapia guineensis.* The test organism however proved to be more tolerant to the toxic effect of the toxicant then *Paleamonetes aficanus*, as higher LC_{50} values was observed for all the toxicants at various exposure times. For black water, 48hrs - LC_{50} values was calculated to be 335.98ppt, 72hrs - LC_{50} was 4.46ppt, and 96hrs - LC_{50} was 4.27ppt.

The pattern of LC_{50} followed by *Paleamonetes africanus* was almost the same for *Tilapia guineensis,* in which the grey water had more deleterious effect on the test organism. The 48hrs - LC_{50} was determined to be 39.81ppt, 72hrs - LC_{50} was 25.84 and 96hrs - LC_{50} was 2.28ppt.

The 48hrs - LC_{50} for ballast water on *Tilapia guineensis* was determined to be 19380.11ppt, 72hrs - LC_{50} was 1301.31ppt and 96hrs - LC_{50} was 270.36ppt. the same pattern was recorded for slop water in which the 48hrs - LC_{50} was determined to be 68.83ppt, 72hrs - LC_{50} was 18.30ppt and 96hrs - LC_{50} was 4.14ppt respectively.

The LC_{50} of the toxicants on both *Paleamonetes africanus* and *Tilapia guineensis* followed a general pattern, in which the amount of toxicant that was able to kill 50 percent of *Tilapia guineensis* was always higher than that required to kill 50 percent of *Paleamonetes africanus*. This suggests that *Tilapia guineensis* had more tolerance to the toxicant at the several concentrations and exposure time than *Paleamonetes africanus.*

It has been observed by some researchers that the toxicity of some fluids tested depends partly on their water solubility, chemical composition, concentration and genetic constitution of the organism (Zheng *et al*., 2003). However it should be noted that the quality of waste water could be quite different due to culture, custom, nutrition, health, education, etc. (Metcalf and Eddy., 2003).

The semi-static bioassay demonstrated that at low concentration and increasing exposure period, the toxicants also had lethal effects on aquatic organisms. A decrease in the number of survivors of the test organism with increase in exposure time and concentration of the toxicants was the general pattern observed for all the test organisms. However, the LC_{50} for the different effluents (toxicants) decreased with increase in exposure time as well as increasing concentrations of the effluents. *Tilapia guineensis* was more susceptible to grey water and more tolerant to ballast water, while *Paleamonetes africanus* was more sensitive to grey water and more tolerant to ballast water. Slop water however demonstrated the most toxic effect when compared with other effluents studied. Black water also demonstrated very toxic effects on test organisms, while grey water demonstrated the highest toxic effect on *Paleamonetes africanus*.

It will be concluded that efficient effluent management will help meet up with the long term challenges of sound environmental management, improve standards of living and health, enhance economic opportunities, and make for sustainable development.

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