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AQUATIC TOXICITY BIOASSAY OF CASSAVA MILL EFFLUENT USING TADPOLES (*SCLEROPHRYS REGULARIS*)

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ABSTRACT

During the production of garri, starch and tapioca from cassava, cassava effluents are usually produced as by-product and this by-product contains cyanide and other substances. This study was intended to investigate a 96-hour toxicity test of cassava effluent on tadpoles to check the aquatic environmental health status. Two hundred and ten (210) tadpoles with length and weight ranging from 2.8 -3.5 cm and 0.24 -0.47 mg respectively were collected from a puddle, 150 - 200m from the cassava mill shed and were randomly divided in tens and in duplicate into group A,B,C,D,E and F. Group A is the control. Group B, C, D, E and F were treated with 5ml/L, 10ml/L, 15ml/L, 20ml/L and 25ml/L of the cassava mill effluent with an LC50 of 7.6ml/L. Result showed that there was no death in 5ml/L group B in the 96 hours toxicity test compared with the control. However, in the 10ml/L group C there was a total death of 8 tadpoles in 24, 48, 72 and 96hours of exposure compared with the control. In 15ml/L, 20ml/L and 25ml/L doses there was no survival throughout the period of the experiment $p<0.05$. This study indicated that untreated cassava mill effluent should not be discharge into water body without treatment to save the aquatic lives in the water body.

Keywords: Toxicity, Tadpole, Cassava Effluent, Cyanide, Garri

INTRODUCTION

Untreated waste effluent contributes major health hazard especially in areas where they are not properly managed. In most developing nations of the world, liquid waste generated through agricultural, industrial and domestic activities are discharged into water sources causing environment health problem (Onifade *et.al.* 2015). The UN Water, 2023 recorded that about 80 to 90% of most wastewater generated in developing countries is discharged directly into surface water bodies. Water quality is being drastically affected by untreated waste water, sewage, industrial waste dumping and agricultural run-off. (Ahmad and Saadieh, 2007).

In most locality of the Niger Delta in Nigeria, one of their major occupations is farming and cassava is a major crop. Cassava is a staple food for the local people and all over Nigeria (Eruvbetine *et al.,* 2003). Apart from food, cassava is very versatile and its products such as starch, gum, glues, confectionery sweeteners, biodegradable products and drugs are used in animal feed and alcohol production (Olaniyi *et al*., 2013). In the course of production of garri and tapioca (processed cassava products) by the locals, cassava waste effluent is discharged into the aquatic environment because the local normally establish the cassava mill close to aquatic environment as a reserviour or sink without knowing the environmental implication of the waste effluent on aquatic life. Most cassava variety contains cyanide a substance that makes the crop toxic to the aquatic ecosystem if they are not well processed for proper removal of the cyanide chemical. (Adeyemo, 2005; Olaniyi *et al.,* 2013).

Histopathological studies of cassava effluent carried out by (Olaniyi *et al*., 2013; Olufayo and David 2013; Adeyemo 2005), on a 96-hour toxicity test of cassava effluents on Catfish and Nile tilapia, reveal that the gill was the primary target tissue affected by cassava mill effluents.

The effects of pollutant release indiscriminately into nearby streams and wells, emanating from cassava processing sites are necessary for evaluation (Onifade *et.al* 2015). This research tends to further enhance the use of tadpoles as aquatic toxicity bioindicator because of it sensitivity to aquatic environmental pollution compared to fish.

MATERIALS AND METHODS

The following are materials and methods use in the research

Collection of cassava effluent

The cassava effluent was collected from a local cassava processing mill shed in Dennis Osadebay University, Asaba Delta State, Nigeria. The mill is owned by a private person and not the school. Dennis Osadebay University is located in Delta State, Nigeria with a geographical location coordinate of $6^0$15'10''N and 6^0 42'12''E.

Collection of tadpoles

The tadpoles of *Sclerophrys regularis* were randomly collected from a puddle in the university premises 150 – 200 m away from the cassava mill shed. The tadpoles were collected with a hand filter net into a clean water of the puddle in a plastic container and taken to the laboratory immediately.

Acclimation of the tadpoles

The tadpoles were acclimated for 7 days in their natural water in the laboratory so that they will get use to the laboratory environmental condition prior to the treatment.

Experimental design

A total of 110 tadpoles of length 2.8 to 3.5cm were randomly distributed in tens into 11 toxicity tanks and in duplicate except the control which was a single tank. The experiment was done in duplicate to avoid bias and for accuracy purpose. The average number of death of each duplicate concentration was used as the final mortality of the tadpoles in each treated group. Group A is the control and group B, C, D, E and F were the treatment groups. Group B, C, D, E and F were treated with 5ml, 10ml, 15ml, 20ml and 25ml of the cassava mill effluent. Each tank contains 1L of the tadpoles natural puddle water. The cassava effluent was introduced into the static tanks containing the water puddle and the tadpoles mimicking their natural environment. In the course of the experiment, the behavior and the activity of the tadpole were recorded

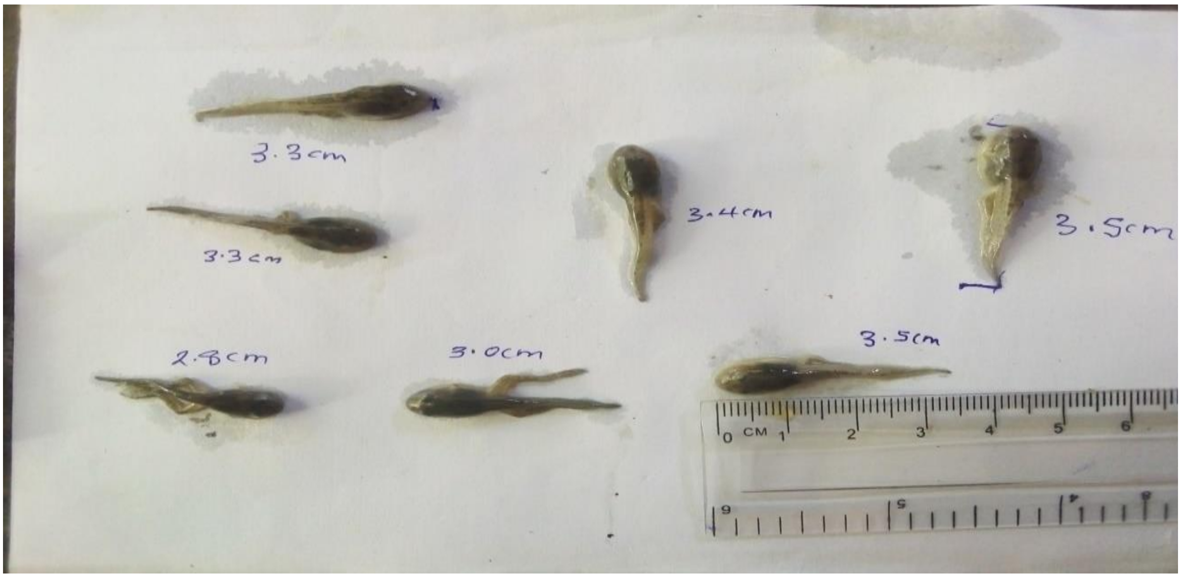

Plate 1: This plate is showing the length in centimeter of the tadpoles which ranges between 2.8 – 3.5cm

Method of Data Analysis

All data collected were subjected to a One-way ANOVA using computer based excel. SPSS version 20 was used to determine the Probit analysis for LC_{50} of the effluent on the tadpoles.

RESULTS AND DISCUSSION

This research tends to emphasize on the use tadpoles in toxicological bioassay experiment especially the aquatic environment. The result of the 96 hours' mortality test in each day from control 0.0ml/L to treatments concentration of 5ml/L, 10ml/L, 15ml/L, 20ml/L and 25ml/L over time (time in hour) as shown in Figure1, Figure2, Figure3, Figure4 and Figure5. Figure6 showed the 96 hours mortality over time in one graph. Table 1 described the behaviour of the tadpoles when exposed to the different concentration of the cassava mill effluent. Plate 1 gives the length of the tadpoles in centimeter in the range of 2.8 – 3.5cm.

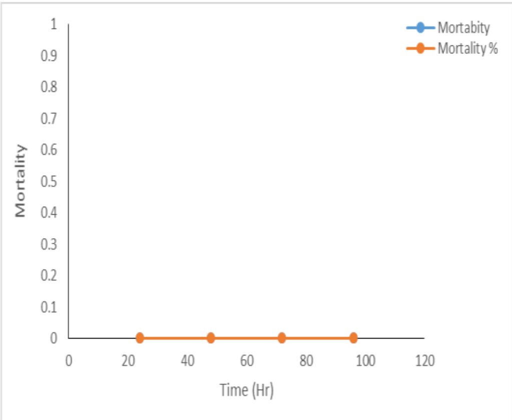

Figure 1: Mortality at 5 mg/L concentration level over time

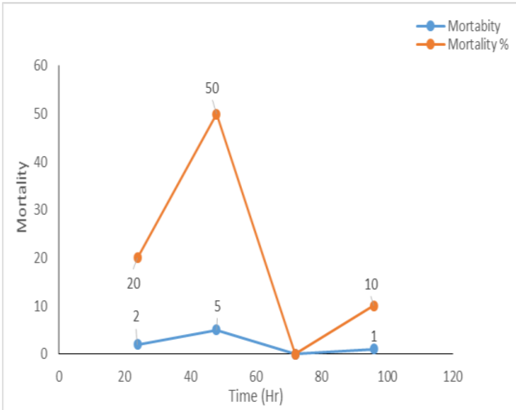

Figure 2: Mortality at 10 mg/L concentration level over time

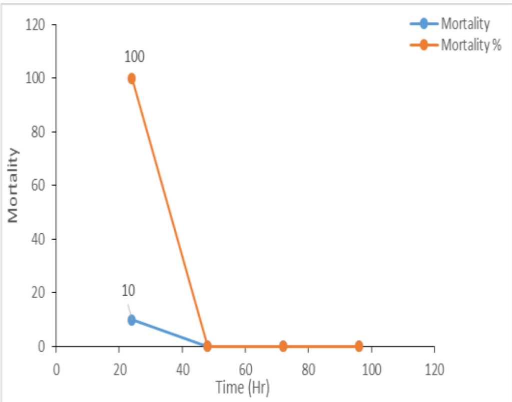

Figure 3: Mortality at 15 mg/L concentration level over time

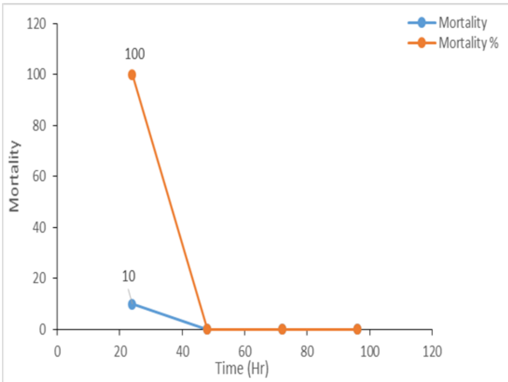

Figure 4: Mortality at 20 mg/L concentration level over time

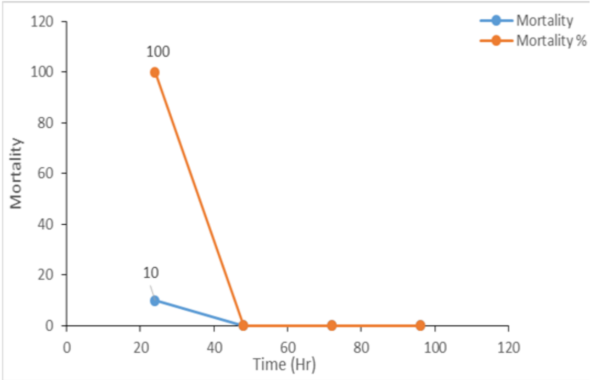

Figure.5: Mortality at 25 mg/L concentration level over time

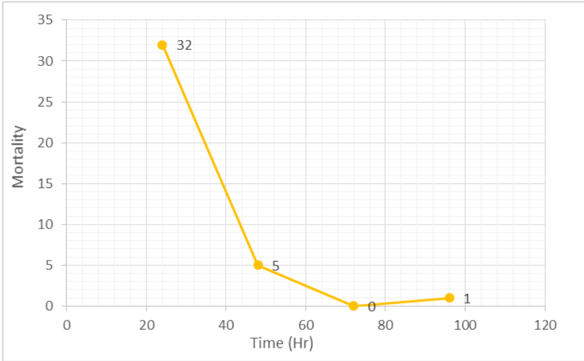

Figure 6: Numbers of mortality at different concentration level over time

Table 1: Behaviour of tadpoles exposed to different concentrations of cassava mill effluent

Treatment	Activity/Behaviour
Control 0.0ml/L	The tadpoles were active throughout the experimental period and no death was recorded in 96hours
5mg/L	There was little erratic movement but calmed almost immediately and no death was recorded in 96 hours of treatment.
1mg/L	There was movement of the tadpoles to the surface of the tank probably to inhale oxygen. Retard movement was observed after about 10 to 15 minutes and after 24 hours mortality of 8 tadpoles were recorded.
15mg/L	After 24 hours of treatment the tadpoles exposed to 15 mg/L experienced sluggishness in locomotion and after 24hours of treatment all the tadpoles died.
20mg/L	There was abrupt inactivity of the tadpoles when exposed to 20mg/L and after 24 hours there was total death of the tadpoles.
25mg/L	There was abrupt inactivity of the tadpoles when exposed to 25mg/L and after 24 hours there was total death of the tadpoles.

Discussion

To examine the toxicological effects of cassava mill effluent on the environment especially the aquatic environment, tadpoles of length within 2.80 to 3.50cm of the *Sclerophrys regularis* species were exposed to 96 hours toxicity test. Tadpoles was used for this analysis because they are very sensitive to variation from normal environmental condition as a bioindicator of pollutant expose to the aquatic environment which support the work of (Wang *et al* 2019) where tadpoles served as surrogates for fish in the safety evaluation of organic pollutants in the environment. Apart from fish fingerlings and crayfish that are usually used to examine the effects of toxicant on the health status of the aquatic environment, tadpoles are equally used to ascertain the effects of toxicant in the aquatic body. The usage of tadpoles as a test organism to examine the 96hours toxicity test by the researchers of this work corresponding to the work of (Shuo *et.al* 2019; Sandra *et.al* 2021) using tadpoles as bioindicator.

Throughout the period of the research, no death was recorded from the control group. This is an indication that the tadpoles used for the experiment were viable and healthy and therefore gives credit to this experiment. At 5 ml/L, no death was also recorded throughout the experimental period. This implies that in the treatment of cassava waste effluent, the environmental authority of the local government of that community or communities involved in cassava processing, should advocate that cassava mill effluent should be treated to a concentration that should be between 0.00mg/l to 5mg/l before discharging the effluent to the environment to avoid deleterious consequences in the aquatic environment. In Figure 2, 10 mg/l, a total of 80% mortality was recorded and only 20 % survived at the end of 96 hour acute toxicity test. This result was as a result of the toxic potency of the cyanide in the cassava mill effluent. This result was statistically significant $p<0.05$ when compared with the control. This finding revealed that tadpoles are very sensitive to environmental contaminant and the use of it as bioindicator should be encouraged. In Figure 3 to Figure 5, there was no survival from the very day of exposure to the end of the 96 hours toxicity test. This was as a result of the cyanide concentration in the test cassava mill effluent and this lead to reduction in oxygen (anoxic condition) and the external gills defect of the tadpoles. This report coincided with the work of (Olufayo and David, 2013 ; Ural and Simsek, 2006) that reported the dysfunction of behavior and respiration as an index of cassava mill effluent toxicity in *Claria gariepinus*.

Table 3 showed the activities of tadpoles exposed to varying concentrations of cassava effluent and how they were stressed progressively with time before death. The stress signs included erratic swimming, increased opercular ventilation, air gulping, and increased mucus secretion on the skin and gills. These behavioural changes have also been reported in other studies where fish were exposed with various toxicants (Ariyomo *et.al* 2017). Cassava mill effluent has a very high content of cyanide which leads to the mortality of the tadpoles in this acute toxicity test. This fact of high content of cyanide in cassava mill effluent support several works of other researchers on fishes and other aquatic organisms (Ebhoaye and Dada, 2004; Orji and Ayogu, 2018; Adomi and Morka, 2020 and (Ariyomo *et. al*. 2017) etc.

Plate 1 gives the sizes and lengths of the tadpoles. This sizes and lengths were not different from the sizes of fingerlings of catfish and tilapia use as test organisms in toxicity test. This study tends to emphasize and to add the too few literature in environmental toxicity test using tadpole as the bioindicator.

CONCLUSION

In summary because of the sensitivity of tadpoles to cassava mill effluent and other chemical toxicants to the aquatic environments, it would be concluded that tadpoles should be used as bioindicator in aquatic toxicity. This implied that apart from fish been used as a toxicity test organism to access the effects of toxicant on the environment, tadpoles can equally be used to evaluate toxicant effects on the aquatic environment.

RECOMMENDATIONS

i. It is therefore recommended that cassava mill effluent should be treated before discharge to the environment.
ii. Local people that are garri producers should be educated about the environmental consequences of discharging untreated cassava mill effluent to the environment.
iii. Further test such as subacute and chronic toxicity cassava mill effluent analysis should be carried out.

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