Research Article

Acute Lethal Toxicity of N,P,K 15:15:15 Fertilizer on the Juveniles of *Clarias gariepinus* in Zaria, Nigeria

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ABSTRACT

The acute toxicity of N,P,K 15:15:15 fertilizer to juveniles of *Clarias gariepinus* of mean weight 11.6g, mean total length and mean standard length of 12.9cm and 10.9cm respectively were investigated under laboratory conditions. The effect of the N,P,K fertilizer were measured based on 2 levels of organization; organs and whole body. At the organ level, the acute concentration of N,P,K showed marked increase in the opercular and tail fin beats of the fish in the first 24 hours but further exposure resulted in decrease of the opercular and tail fin beats. This was concentration dependent. At whole body level, the 96 hours LC_{50} bio-assay showed symptoms of toxicity of N,P,K that altered the fish behaviour such as loss of equilibrium, agitated swimming, e.t.c. The 96 hour LC_{50} was extrapolated to be 239.85gl^{-1} that is, the concentration that will kill the typical fish. It is therefore recommended that the use of N,P,K 15:15:15 on farmlands with channels to lake, streams, ponds, e.t.c be minimized.

KEYWORDS: N,P,K 15:15:15, *Clarias gariepinus*, toxicity, opercular beat, tailfin beat, mortality

INTRODUCTION

Survey of agricultural, urban streams and groundwater have brought public attention to a wide spread chemical mixture contamination and the associate risk to aquatic life are being increasingly considered in marketing decisions along with the societal benefits gained from the use of new chemicals. Agrochemicals such as fertilizers and pesticides are routinely employed as part of the integrated farming practice to protect crops and animals from insect pest, weeds and diseases and encourage healthy growth of crops by making essential nutrients available to crops grown on poor agricultural soil.

N,P,K fertilizer, over the years, has been used alongside with other chemicals on agricultural lands. When the rains come much of these

chemicals are washed into the streams, lakes or rivers that form the aquatic environment. It our curiosity of the risks to aquatic organisms associated with the introduction of chemicals into the environment that has necessitated research interest in estimating the hazard of chemical substances to aquatic life. The aquatic ecosystem as a part of the natural environment is also faced with the threat of a shrinking genetic base and biodiversity owing to indiscriminate use of chemicals especially pesticides [1]. These chemicals become readily available in the food chain and subsequently bio-accumulate in both aquatic flora and fauna [2,3]. Several works done on pesticide effect on fish reveals that important organs, for example, the kidney, liver, gills, stomach, brain, muscle and genital organs are damaged in fish exposed to pesticides because of residential effect of these pesticides [1]. This study focuses on the toxic effect of N,P,K fertilizer on the juveniles of *Clarias gariepinus*, the African catfish.

MATERIALS AND METHODS

This study is based on the short term toxicity bio-assay method of [4] with some modifications.

FISH COLLECTION AND MAINTENANCE

The fishes were purchased from elegant fish farm in Kaduna and were transported to the Fisheries research laboratory of the Department of Biological Sciences, Ahmadu Bello University, Zaria by means of cold boxes with sufficient water from the fish farm. It was then transferred carefully into a large water bath containing deionised tap water. The fishes were acclimatized for five weeks prior to the commencement of the bio-assay. During this period, the fishes were fed with commercial feed pellets (Copens) twice daily, morning and evening feeding plan, with 12 hours illumination plan. Unconsumed feeds and faecal waste were removed by siphoning and water replenished regularly as recommended. Feeding was stopped 24 hours before the fishes were introduced to the test solutions.

PREPARATION OF TEST SOLUTIONS

N,P,K 15:15:15 fertilizer was bought from Samaru market in Zaria. In the laboratory, several concentrations of the test solutions were prepared by dissolving a weighed amount of the fertilizer and dissolving it in a litre of distilled water. A sensitive weighing balance was used for the weighing and the dissolution was done in a flat bottom flask to make the concentrations in gramme per litre (gl^{-1}).

EXPERIMENTAL FISH

A total of 180 juveniles of *C. gariepinus* were used with an average weight of 11.6g, average total length of 12.9cm and average standard length of 10.9cm. The fishes were maintained at 27±1˚C, PH of 6.07-7.06 and dissolved oxygen of 5.04-6.67mgl^{-1} throughout the static bioassay.

PILOT STUDY

A range finding test was carried out to determine the range of fertilizer concentration to be used in the definitive test that lasted for 96 hours. Geometric series was used to span a range of concentration from that causing zero

mortality to that causing complete mortality using three fishes per concentration. 0.1, 0.2 and 0.3g of N,P,K were chosen using a spacing factor (x10) to give concentrations 10, 20, 30, 100, 200 and 300gl^{-1} used for the pilot test to establish the range of concentrations 200.00, 221.32, 244.93, 271.04 and 299.94gl^{-1} used for the definitive test.

BIO-ASSAY

A total of 6 glass aquariums were used (5 experimental and a control) containing 20 litres of water in each used for the dilution of each concentration. These were replicated three times making a total of 18 tanks for the study. The different concentrations prepared were introduced into each tank apart from the control. The fishes were then randomly introduced into the tanks with each tank containing 8 fishes as described by [5] and [1]. The behaviour and general condition of the fishes were noted prior to, during and after the bioassay. The tail-fin beats and opercular ventilation rate were noted at the onset of exposure until 96 hours. Mortality rate was also observed at 24, 48, 72 and 96 hours respectively. Confirmation of dead fish was done by touching them gently with a glass rod and noticing no opercular movement and tail-fin beats. Dead fishes were promptly removed and mortality was recorded as described by [1].

STATISTICAL ANALYSIS:

The percentage mortality in each concentration was determined. The LC_{50} after 96 hours was also determined and a probit graph was drawn using Microsoft excel package. The observed tail-fin beats and opercular movement were presented in figures; Analysis of variance was used to test for significant difference between the observed means.

RESULTS AND DISCUSSION

TAILFIN BEAT RATE OF *CLARIAS GARIEPINUS*

The test fishes in the tank containing the highest concentration of the fertilizer (299.94gl^{-1}) showed the highest tail-fin beat of about 110 beats per minute (figure 1). About the same beat rate was recorded in the tank containing 271.04gl^{-1} of the fertilizer solution. The beat rate rose sharply in the 24th hour and fell slightly in the 48th hour. The beat rate fell continually till the end of the bioassay. The control, however, recorded about 60 beats per minute at the start of the experiment and this beat rate rose gradually with time owing to increased activity of the fishes in the tanks.

OPERCULAR BEAT RATE OF *CLARIAS GARIEPINUS*

The opercular beat rate of *C. gariepinus* exposed to the fertilizer solution is shown in figure 2. Again, the 24 hour period had the highest opercular beat rate in the tank containing the highest concentration of the fertilizer solution (299.94gl^{-1}). The opercular beat fell with time in tanks 2, 3, 4 and 5 respectively. The control tank recorded the lowest opercular beat at the onset of exposure that rose steadily at 24 hours and 48 hours to about 87 and 96 beats per minute due to increased cannibalism and aggression and fell to about 95 beats at the end of the study.

MORTALITY RATE

Mortality was recorded in all tanks except that of the control (Table 1) and the rate of mortality was concentration dependent. Tank 1 (200.00gl^{-1}) recorded the least mortality (1); tank 5 (299.94gl^{-1}) recorded complete mortality (8) at the end of 96 hours. Dead fishes removed from treatment tanks were observed to be covered with mucus much of which was over the operculum. The probit kill value was plotted against the log of concentrations of the toxicant (Figure 3) from which the 96 hours LC_{50} was determined to be 236.89gl^{-1}.

DISCUSSION

The operculum and the tail-fin activity are the main organ in focus in this study. The fall in the rate of opercular and tail-fin beats may be attributed to decreased activities of various respiratory enzymes. A similar finding was reported by [6] when *Tilapia mossanbica* were exposed to methyl parathion. After induction of pesticide, the intoxicated fish were aggregated at the corner of the aquarium resting at the bottom and frequently come to the surface followed by heavy breathing with stronger opercular movements and loss of equilibrium. Also the over secretion of mucus was observed on the fishes treated to pesticide. This has been observed in fishes treated with other pesticides also [7]. The same activity also reported in the gills of catfish treated with azadirakhtin [8]. [9] also noted that opercular movement increased and then decreased as time for death approached. Again, the changes observed in the rate of opercular and tail-fin beats may result from alteration of the blood composition either directly by destruction of erythrocyte cells or indirectly by disruption of osmotic and ion-regulation thereby affecting oxygen uptake to tissues resulting in fatigue, imbalance and eventually death of the fish. This agrees with the report of [10] and [11]. The sharp increase in the opercular ventilation and tail-fin beats noticed at the onset of exposure to the fertilizer solution is concentration dependent. Similar findings were reported by [12] and [13].

At the whole body level, the fishes exposed to the various concentration of N,P,K 15:15:15 fertilizer were observed to show uncomfortable responses to the medium. The fishes moved from one end to another of the tank in a jerky and erratic manner with hypersensitivity to touch at the early periods of exposure. Overtime, the fishes grew weak, stood vertically to the medium with their heads at the surface of the medium and even when touched with a glass-rod do not respond actively. This is a clear sign of fatigue. Loss of equilibrium and eventually death resulted in the fish. Similar alterations in behaviour were reported by [14] and [15]. The rapid jerky and erratic movement and hypersensitivity noticed may be attributed to the effect of the fertilizer on the central nervous system of the fish that principally may have resulted in impaired cerebral oxidative metabolism as suggested by [16].

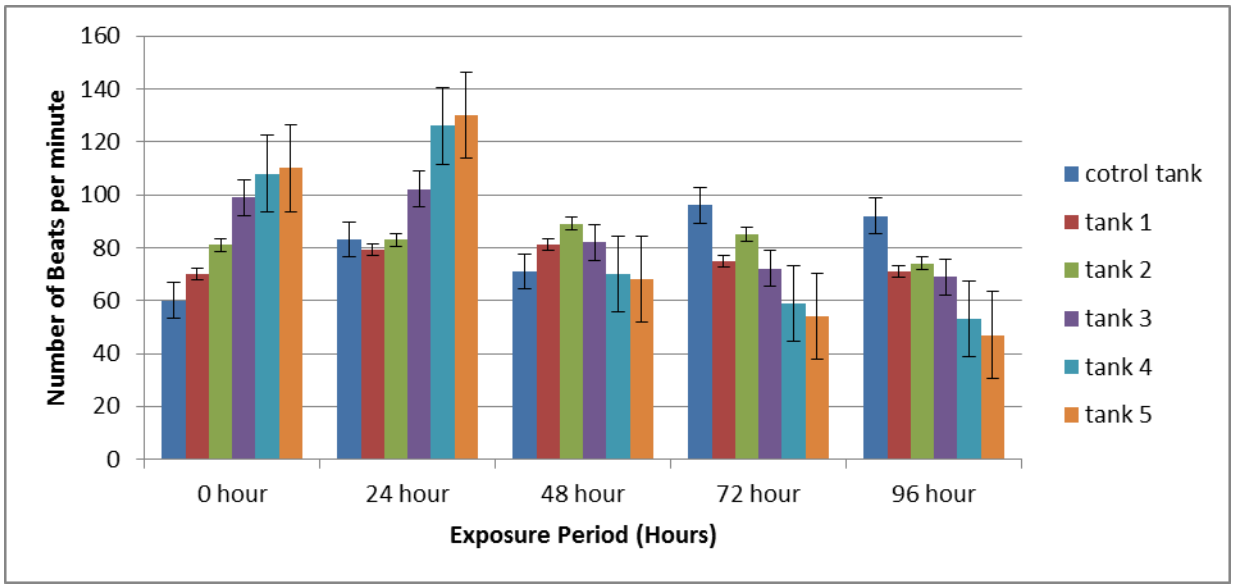

Figure 1: Tailfin beat of *Clarias gariepinus* juveniles exposed to various concentrations of N,P,K 15:15:15 Fertilizer

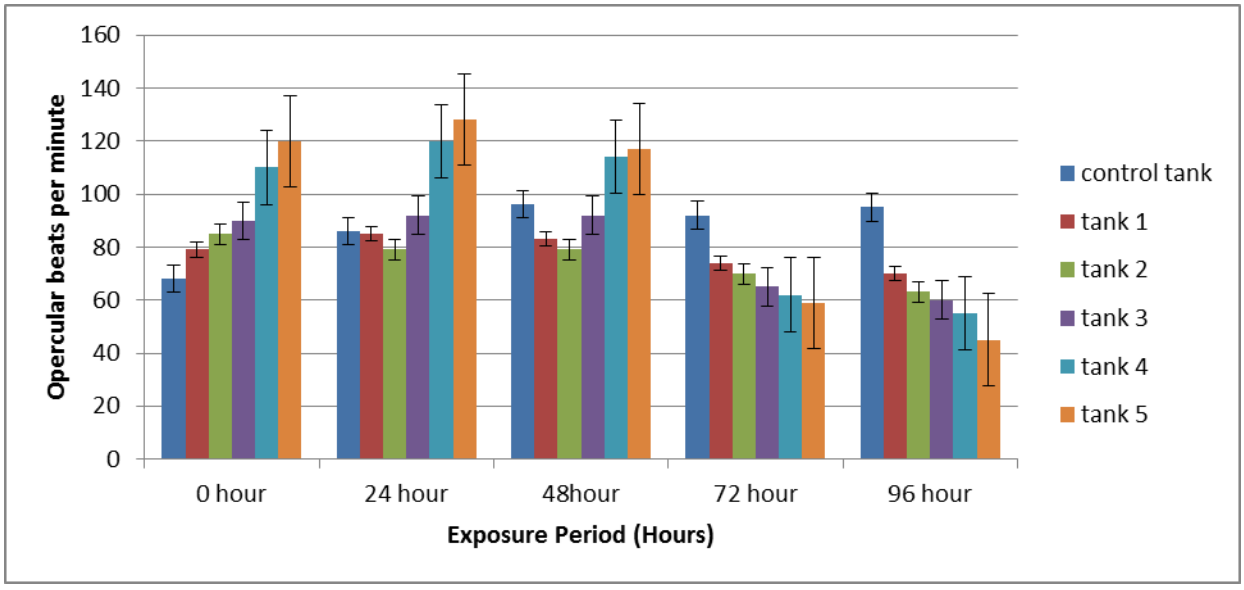

Figure 2: Opercular Ventilation rate of *C. gariepinus* juveniles exposed to various concentrations of N,P,K 15:15:15 Fertilizer

Table 1: Mortality rate of *C. gariepinus* juveniles exposed to various concentrations of N,P,K 15:15:15 Fertilizer

Tank	Concentration of N,P,K (gl^{-1})	Log of conc.	Mortality	% Mortality	Probit value
control	0.00	0.00	0	0	-
1	200.00	2.300	1	12.5	3.85
2	221.32	2.345	2	25	4.33
3	244.93	2.389	6	75	5.68
4	271.04	2.433	7	87.5	6.15
5	299.94	2.477	8	100	-

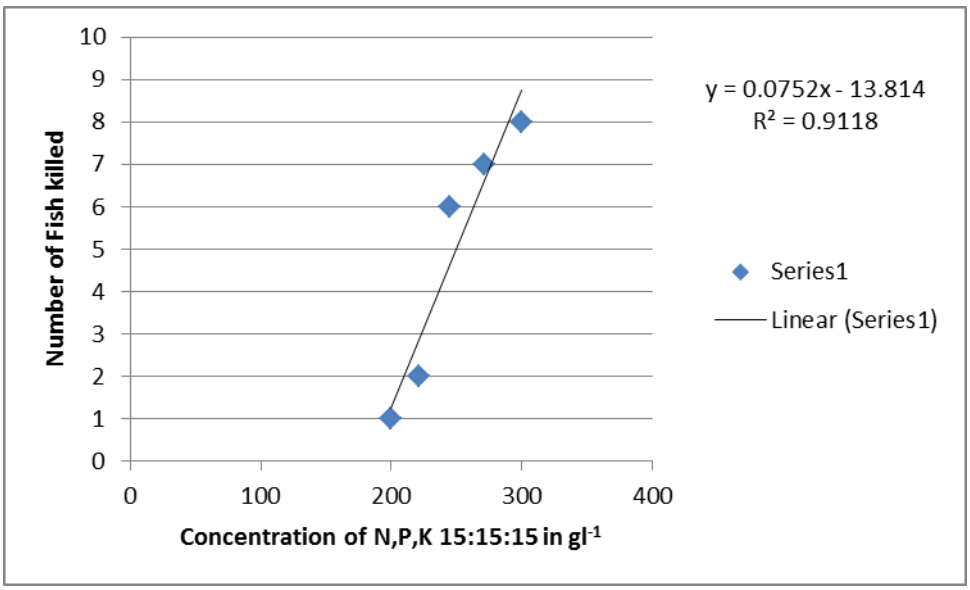

Figure 3: Profit plot showing the LC_{50} of N,P,K 15:15:15 Fertilizer on *C. gariepinus* juvenile

CONCLUSION

It is therefore concluded that N,P,K 15:15:15 fertilizer results in the alteration of tail-fin beat rate and opercular movement of the juveniles of *Clarias gariepinus* fish. The concentrations of N,P,K 15:15:15 fertilizer also killed juveniles of *Clarias gariepinus* implying that it has toxic

effect on this life-stage of the fish and 236.89gl-1 will kill half the population of fish exposed.

RECOMMENDATION

I hereby recommend that more work be done to determine the damage N,P,K 15:15:15 has on the internal organs of the fish. Further studies may also be carried out on other life stages of the fish, *Clarias gariepinus.*

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