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Full Length Research Paper

Toxicity of dizensate (Glyphosate herbicide) on C*larias gariepinus* fingerlings

Akinsorotan A.M

Department of Fisheries Technology, Federal College of Fisheries and Marine Technology, PMB 80063, Victoria Island, Lagos-Nigeria. E-mail: demo4them@yahoo.co.uk

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Run-off containing a number of chemical solutions, which include herbicides, effluents, find their ways into the water bodies with a resultant negative effects on the aquatic ecosystem. This study sought to establish the effects of herbicide widely used in eradicating aquatic weed; Dizensate glyphosate (N-phosphonomethyl glycine) on Fingerlings of African catfish (*Clarias gariepinus)*. The four day static bioassay was used to determine the median lethal concentration (LC_{50}) and the value was 18.07 mg/l. Mean mortality was 0, 17, 58, 75 and 92% in the concentration of 9.6, 14.4, 19.2, 21.6 and 24.0 mg/l respectively, while there was no mortality in the control treatment. There were significant differences ($P<0.05$) on the effect of concentration. Fish responses to the toxicant include erratic movement, air gulping, loss of reflex, molting, barbell deformation and excessive mucus secretion. Mortality increased with increase in concentration of Dizensate glyphosate on catfish fingerlings and time of exposure. The physicochemical parameters also showed a slight increase as the concentration increased. It could be concluded that Dizensate glyphosate; an aquatic herbicide, has harmful effects on the physiology, biochemistry and histological structure of fish which in turn affects the growth rate.

Key words: Dizensate glyphosate, *Clarias gariepinus*, acute toxicity, median lethal concentration.

INTRODUCTION

Nigerian water bodies suffer from neglect and abuse because of non-enforcement of laws regulating their use. Environmental Impact Assessment (EIA) reports are not carried out as a perquisite for the establishment of industries, Ponds are dug indiscriminately even on dunghills that toxic materials has been dumped on for years without any restriction from government agencies. The constant flow of agricultural and industrial effluents into fresh water often leads to a variety of pollutant and accumulation of various metals, which becomes apparent when considering toxic pollution (Mason, 1991).

These herbicides and pesticides when applied in restricted areas are washed and carried away by rains and floods to nearby aquatic system, thereby affecting aquatic biota, especially fish, which serves as a rich protein supplement for man (Chattopadhay et al., 2006;

Farombi et al., 2008; Ndimele et al., 2010). The herbicides affect not only the physiology and survival of aquatic organisms but also interact with their genetic make-up leading to mutations and/or carcinogenesis (Goksoyr, 1991; Da-Fonseca et al., 2008; Nwani et al., 2010).

Toxicants contaminate freshwater bodies and affect non-target organisms. Various researchers have reported the effects of chemicals on aquatic organisms (Jiraunghkoorskul et al., 2002). Environmental factors such as pH, turbidity, alkalinity, dissolved oxygen, temperature and conductivity influence the rate of reaction of the pollutants entering the water or the lethal effects on the aquatic organisms (Fagbenro, 2002). Pollutants in water significantly affect the ability of fish to detect and respond to chemical stimuli, feeding, growth, and reproductive performances could also be seriously affected by such polluted habitat. Pollution of aquatic habitat may result in mass fish mortality or their failure to breed in the polluted environment.

This study determines the 96 h LC_{50} of glyphosate herbicide on fingerlings of *Clarias gariepinus* and reports the effects on the physiology and survival of fish.

MATERIALS AND METHODS

Description of the study area

The bioassay experiment was carried out using the laboratory of Federal College of Fisheries and Marine Technology, Victoria Island, Lagos.

The experiment was conducted using standard static bioassay procedure (Reish and Oshida, 1987; American Public Health Association, 1987). This involved controlled environmental conditions as to define the response of the test organism to Dizensate herbicide Dizensate herbicide with a trade name Dizensate 480_{SC} manufactured by Jiangshu Flag Chemical Industrial Park, China was used for this experiments. The study was conducted between November 2010 and July 2011.

One hundred and forty four (144) live, and apparently healthy fingerlings of *C. gariepinus* measuring between 9.3 and 10.6 cm total length, and weighing between 5.8 and 6.5 g, were collected from a private hatchery in Akute- Lagos State, Nigeria, transported in plastic bowl at 07 h and acclimated for 1 week in the laboratory, inside transparent rectangular glass tanks measuring 40 × 29 × 28 cm of 60 L capacity, filled with 20 L unchlorinated well water at the Fisheries Research Laboratory, Federal College of Fisheries and Marine Technology, Victoria Island, Lagos. The fish were fed diet containing 45% crude protein during the acclimation period. Feeding was discontinued 48 h before the commencement of the experiments, to minimize the production of waste in the test container.

The amount of herbicide which contained the require milligram of Dizensate herbicide was determined from the 480 g/L of Dizensate herbicide formulation and concentration of 9.6, 14.4, 19.2, 21.6 and 24 mg/l were introduced into each of the tanks in duplicate treatments and 0.0 mg/l (control). The fish were batch weighed and distributed randomly in duplicate treatments into 12 glass tanks (12 fish/tank). The pH of the solutions was measured with the pH meter, temperature with mercury-in-glass thermometer, dissolved oxygen with a digital Dissolved Oxygen. The exposure lasted for 96 h. Fish mortality was monitored and recorded hourly for the first four hours, 4 h for the next 24 h and subsequently every 24 h for the next 96 h. The inability of the fish to respond to external stimuli was used as an index of death. Dead fish were removed immediately with a scoop net to avoid contamination due to rotting. Apart from monitoring and recording fish mortality, the fish behavior such as erratic swimming, air gulping, loss of reflex, discoloration and molting were monitored.

Water temperature, pH and dissolved oxygen were determined every hour for the first 4 h and once every 4 h for the next 20 h and once every 24 h before the end of the experiment. The LC_{50} was determined graphically using logarithm transformation at the end of the exposure period. All data obtained were subjected to Analysis of Variance (ANOVA) and multiple range test using the SAS.

RESULTS AND DISCUSSION

The concentration of Dizensate herbicide that will bring about 50% mortality of the test organism in 96 h is referred to as 96 h $LC_{50.}$ The 96 h LC_{50} of Dizensate herbicide to fingerlings of *C*. *gariepinus* is presented in Table 1 and Figure 1.

This value (18.07 mg/l) is the concentrations of the treatments required to bring about 50% mortality of C. *gariepinus* fingerlings within 96 h period. The acute toxicity of Dizensate herbicide decrease with increase in time. Total mortality resulted at concentration of 28.8, 38.4 and 48 mg/l of Dizensate herbicide to fingerlings of *C*. *gariepinus*.

The result of the study revealed that fish exposed to the dizensate herbicide solutions showed erratic

Table 1. The lethal concentrations (LC_{50}) values of Dizensate herbicide to *C. gariepinus* fingerlings after several hours.

Time (h)	Log C value	LC_{50} (mg/l)
24	1.387	24.38
48	1.366	23.23
72	1.318	20.80
96	1.257	18.07

Key: LC_{50}: Leather Concentration that kills 50% of test organism.

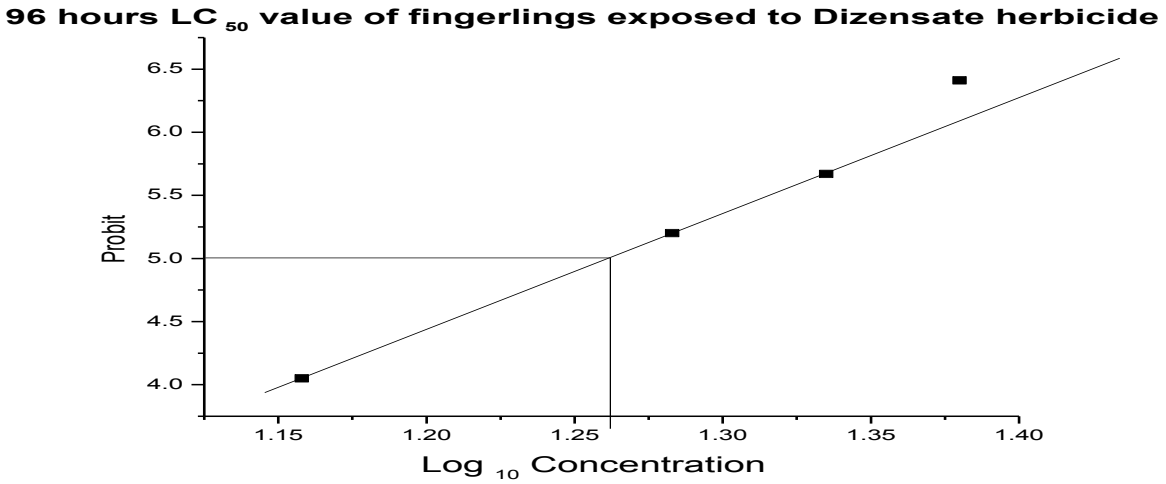

Figure 1. Shows the LC_{50} for fingerlings of C. *gariepinus* at 96 h exposure to Dizensate herbicide.

swimming, loss of reflex, moulting, discolouration, behavioural changes and increasing opercula ventilation and movement (Table 1). At higher concentration (19.2 – 24 mg/l) the fingerlings showed erratic swimming, air gulping, loss of reflex, barbell deformation, excessive mucus secretion and molting within 24 h. The fish finally settle at the bottom motionless with slow opercula movement. Lower level of Dizensate herbicide concentrations (9.6 and 14.4 mg/l) did not produce any serious change in the fish behavior within 24 h. Meanwhile, by 96 h, some of those in the 19.2 – 24 mg/l concentrations demonstrated loss of reflex and death occurred which was observed to be proportional to increase Dizensate herbicide concentration level and duration of exposure.

Dizensate herbicide was highly toxic to *C. gariepinus*. The toxicity rate of the organism is dose-dependent. The observation is confirmed by the significantly different ($P<0.05$) mortality rates obtained due to dosage, according to Ufodike and Omoregie (1994) and Aguigwo (1998). The mortality pattern recorded corroborates with that reported by Rand and Pectrocelli (1985) which stated that there should be less than 35% mortality in one of the concentration and at least more than 65% mortality in the highest concentration. The abnormal rapid movements of the fish subjected to high concentrations of Dizensate herbicide suggest that it acted on the nerves of the fish similarly reported by Okon (1991) of pesticides.

Water quality attributes are prime factors that influence fish survival, reproduction, growth performance, and overall biological production (King, 1998; king and Jonathan, 2003). They affect aquatic biotic integrity by directly causing mortality and / or shifting the equilibrium among species due to subtle influences such as reduced reproductive rates or alternations in competitive ability.

This inverse relationship is interesting and indicative of

Table 2. Physico-chemical parameters measured during the 96 h exposure to Dizensate herbicide.

Time	Parameter	0 mg/l	9.6mg/l	19.2mg/l	28.8mg/l
Before	Temp (0^oC)	25±0.1	26±0.2	26±0.1	25±0.1
	Ph	6.9±0.2	7.0±0.2	7.0±0.2	7.0±0.2
	DO_2 (mg/l)	5.9±0.1	5.8±0.1	5.8±0.1	5.8±0.2
8 h	Temp (0^oC)	25±0.1	27±0.2	27±0.2	27±0.2
	Ph	7.2±0.2	7.2±0.1	7.2±0.1	7.2±0.1
	DO_2 (mg/l)	5.9±0.1	5.6±0.1	5.5±0.2	5.4±0.2
24 h	Temp (0^oC)	25±0.0	27±0.1	27±0.1	27±0.1
	Ph	7.2±0.0	7.2±0.1	7.2±0.1	7.2±0.1
	DO_2 (mg/l)	5.8±0.1	5.5±0.1	5.2±0.1	4.5±0.1
48 h	Temp (0^oC)	25±0.1	27±0.2	27±0.2	27±0.2
	Ph	7.2±0.3	6.8±0.1	6.8±0.1	6.8±0.1
	DO_2 (mg/l)	5.8±0.1	5.5±0.1	4.9±0.1	4.2±0.1
72 h	Temp (0^oC)	25±0.1	27±0.1	27±0.1	27±0.1
	Ph	7.2±0.1	7.2±0.2	7.2±0.2	7.2±0.2
	DO_2 (mg/l)	5.5±0.1	4.5±0.1	4.7±0.1	3.9±0.1
96 h	Temp (0^oC)	25±0.2	27±0.1	27±0.1	27±0.1
	Ph	7.2±0.1	7.2±0.2	7.2±0.2	7.2±0.2
	DO_2 (mg/l)	5.4±0.1	4.4±0.1	4.2±0.1	3.3±0.1

higher demand for oxygen prompted by condition of hyperactivity as observed and explained by Ofojekwu et al. (2001) and Oti (2003).

Physicochemical parameter measured seemed to be within optimum range for fish culture as reported by Omitoyin et al. (2006) and Olaifa et al. (2003). There was a significant negative correlation between pH and dissolved oxygen values. There was a significant change in water quality resulting from increase in concentration of toxicant. This observation was in line with Okoli-Anunobi et al. (2002) who investigated the lethal effect of the elephant Blue detergent (R) on the Nile Tilapia *Oreochronis niloticus*. In case of dissolved oxygen, the treatments did not only show a dose dependent decline in concentration, but also rapid depletion of dissolved oxygen with time. Warren (1977) had earlier reported that the introduction of a toxicant into an aquatic system might decrease the dissolved oxygen concentration, which will impair respiration leading to asphyxiation. This was probably why the fishes were stressed progressively with time before death. Okoli-Anunobi et al. (2002) also reported pH shift slightly from 6.30 to the alkaline death point of 10.75. This finding agrees with pH values in the the present study.

Water qualities were also affected negatively in the exposed medium. The parameters measured increases with increase in concentration of the toxicant. Temperature varies between 26±0.2 to 27±0.1 in the 9.6 mg/l concentration. The pH reduced slightly from 7.0±0.2 in control to 6.4±0.1 in test treatment of concentration 48.0 mg/l after the whole experiment as shown in Table 2. The DO_2 decrease slight from 5.8±0.1 in control experiment to 4.6±0.1 in test treatment of concentration of 48.0 mg/l. This will invariably affect the optimum growth and development of the cultured fish. Bacterials and other germs will thrive with bad water quality. Dissolved oxygen was noticed to decline with increase of the toxicant, a situation refers to as oxygen sag which is characterized by high mortality within short time. Death recorded could therefore have occurred by this reason.

Conclusion and recommendation

The study was carried out to determine the toxicity of dizensate herbicide to the most cultivable fish species in

Nigeria, African catfish *C. gariepinus*. This test animal is important in capture and culture fisheries. Dizensate herbicide is toxic to *C. gariepinus.* This was revealed by the results of various investigation carried out in this study: the relatively very high values of LC_{50}; the abnormal behavior displayed in the exposed fish as well as various degenerative changes observed in the water parameters are pointer to this. It is time that the widespread use of this toxic chemical on ponds area, water ways, drains, streams, roadsides, footpaths, parks, gardens, schools, farms, forestry, national parks etc is stopped or highly restricted.

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