

ACUTE TOXICITY OF GLYPHOSATE ON *CLARIAS GARIEPINUS* FINGERLINGS

Okayi, R.G., Annune P,A, Tachia, M.U. and Oshoke, O.J.

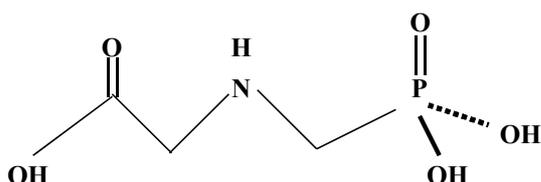
Department of Fisheries and Aquaculture
University of Agriculture, P.M. B. 2373,
Makurdi, Benue State.**ABSTRACT**

The effects of glyphosate on mortality rate and behavioural responses of *Clarias gariepinus* fingerlings were investigated under laboratory conditions for 96 hours exposure period. The lethal concentration (LC_{50}) value of glyphosate on fingerlings of *Clarias gariepinus* was 0.0018 ml/l for 96 hours of exposure. The regression equation was found to be $Y = 9.132 + 1.365 * \text{Log conc}$; $R^2 = 0.9277$ ($Y = \text{probit kill}$). Fingerlings also exhibited several abnormal behaviours, including restlessness, uncoordinated movement, loss of equilibrium, air gulping and staying motionless at tank bottom. Oxygen consumption by fingerlings decreased with increased concentration of glyphosate.

Key words: glyphosate, acute toxicity, *clarias gariepinus*, Makurdi

INTRODUCTION

Glyphosate is one of the herbicides used for the control of annual and perennial grasses, broad-based leafed weeds as well as many tree and species of crop in cropland and non-crop sites (Ronald, 1989). The chemical structure of glyphosate is as follows



IUPAC Name: [(phosphonomethyl) amino] acetic acid Rodosevich and Holt (1984)

observed that this herbicide is used in non-crop situations, in areas as industrial sites, road sides, ditch banks, irrigation canals, fence lines, power line rights of way, recreational areas and rail road embankments. According to Akobundu (1987), this chemical provides an effective and economical weed control in terms of reduced labour.

Ronald (1989) and Stefferud (1990) stated that this is one of the herbicides used in USA for effective control of weeds in corn, rice, and sorghum where crop production is fully mechanized. According to Ovie (1985) and

Ronald (1989), residue of this herbicide has been detected at phyto toxic concentration in ground water, lakes, and streams as a result of run-off from treated fields.

Many farmers in Nigeria of recent have been using indiscriminately various types of herbicides for the control of weeds in crop land areas especially in irrigated canals, rice fields e.t.c. The unpleasant development of this to fish as a result of run-off from treated fields to ground water, lakes, streams, rivers e.t.c. is yet to be fully quantitatively and qualitatively assessed. This study was therefore aimed at investigating the effect of the use of glyphosate on mortality rate and behavioural pattern of fingerlings of *Clarias gariepinus*.

MATERIALS AND METHODS

Fingerlings of *Clarias gariepinus*, mean weight $5.8 \pm 0.2\text{g}$ and $4.2 \pm 0.3\text{cm}$ of length collected from Tiddo Fish Farm, Makurdi, Benue State, were used for the investigation. The fish were acclimatized for seven days in glass aquaria tanks measuring 60cm x 30cm x 30cm containing de-chlorinated and aerated

tap water at room temperature of 28.43 ± 1.32 °C. During the acclimation period, fish were examined for pathogens and diseases. There was no mortality during the acclimation period. Water was changed at three days interval to prevent the build of metabolic wastes and was aerated to increase oxygen supply. Fingerlings were fed twice daily with fish meal at 3% body weight.

Feeding was stopped 24 hours prior to and during exposure period that lasted for 96 hours. This was necessary because feeding increases the rate of respiration and excretory products, which may influence the toxicity of test solution. Water was put into the bowls using a measuring cylinder and was made up with the test chemical (glyphosate) into the bowls making it up 30litres, the same method was applied to the duplicate, Ten fish were stocked per aquarium for experimental runs. The glyphosate concentration used was 0.015, 0.005, 0.0025, 0.0015, 0.0005 ml/l. and stock exposed for each of the five toxicant concentration. In each case, there was a control in which ten fish were exposed to the University of Agriculture (Makurdi) de-chlorinated tap water only. For each of the glyphosate concentration level, five aquaria acting as replicate for each treatment (concentration) were set up. Aeration was provided by means of aerators prior to and during exposures. Temperature condition was kept at room temperature, and all aquaria were exposed to equal amount of natural light. Fish were examined for abnormal behaviors and mortality for 6 hours, 12 hours, 48 hours, 72 hours, and 96 hours, during the period.

The 96 hour LC₅₀ toxicity for each glyphosate concentration was determined as a summary of percentage mortality data

following the method of Hoque *et al* (1993).

Which were immediately removed and counted in every aquarium at each observation time during the exposure periods. The temperature, total alkalinity, hardness, pH, dissolved oxygen, were monitored 24 hours using methods described by APHA (1985). The result obtained were subjected to regression statistical analysis and regression equation for probit obtain. Thereafter a graph was plotted to determine the relationship between mortality and toxicant concentration in order to determine the lethal concentration (LC₅₀).

RESULTS AND DISCUSSION

The physico-chemical parameters of the test solution in the experiment is as shown in table 1. The LC₅₀ values derived from the toxicity test revealed that *Clarias gariepinus* is sensitive to the herbicide. At higher concentrations, percentage mortality increased as shown in table 2 and 3 respectively. The LC₅₀ value for 96 hours was 0.0018 ml/l (figure 1). The computed regression equation was found to be $Y = 9.132 + 1.365 * \log \text{Conc.}$ ($R^2 = 0.9277$, $Y = \text{probit kill}$). The LC₅₀ value in the present study for *Clarias gariepinus* is similar to the findings of Haider and Inbarraji (1986) who used formulations of propan. Rao *et al* (1985) reported that LC₅₀ value of Elson for *Clarias gariepinus* was 0.0010 ml/l for 48 hours of exposure. Hoque *et al* (1993) observed that the LC₅₀ value of Ronil on *Clarias gariepinus* of 9.7cm in length was 1.67 ml/l exposed at 96 hours period.

At higher concentrations of the herbicide exposed to fish, several abnormal behaviors such as restlessness, erratic swimming, air gulping, or respiratory distress, loss of

equilibrium and resting motionless at the bottom of aquaria were observed, similar to Lovely (1998) observations. Avoaja and Oti (1997) reported these abnormal behavioral responses in fish exposed to toxicants.

The erratic swimming, restlessness gulping of air and resting motionless at the bottom of aquaria observed in this investigation are indications that mortality of the exposed fish is not only due to impaired metabolism, but could in addition be due to nervous disorder. This similar to the findings of Oti (2000), Annune, *et al* (1994) who reported these abnormal behavioral responses in fingerlings of the hybrid, Hetero-clarias exposed to toxicants at 96 hours period. The physio-chemical parameters of the solution fluctuated slightly during the bioassay. This fluctuation was not enough to have caused the mortality. Death of test fish exposed to herbicide may be attributed to the destruction of such organs as the gills, liver ,kidney ,brain , blood system and the pancreas. Shultz (1971) had demonstrated that several cell damage especially of pancreas can occur after contact with herbicides. Annune *et al* (1994) also reported that gill tissues are most

sensitive to water pollutants since bills are the primary site for osmo-regulation and respiration. They are highly vulnerable to lesions due to their immediate contact with aquatic pollutants while some pollutants enter the body, there is evidence that some of them exert their effects on the external surface of the fish especially the gills. The very high toxicity of the herbicide could probably be attributed to some possible synergistic effect likely to be produced by the active ingredients in the herbicide of which is the compound that exist separately as herbicide and likely to be equally toxic to fish. The herbicide is acutely toxic to fish *Clarias gariepinus*. Temperature, free carbon dioxide, PH, alkalinity, and other physiological status of the test animals have profound effects, on the toxicity of agro-chemicals.(Boyd 1979, Rand and Petrocelli 1985). Meleter *et al* (1971) reported that herbicide affect gas exchange of fish and other aquatic organisms. This might probably be out of the reasons for the decline of oxygen concentrations. Because the parameters had little variation it was evident that physical and chemical properties of the water holding tanks were within the desirable range of fish culture (Boyd, 1979).

Table 1: Physico chemical parameters of test solution

Parameters	Range	Mean S.E.
Temperature °C	28.9- 29.50	29.43- ±1.12 .32
Total Alkalinity mg/l CaCO ₃	22.00- 28.50	25.2975±0.666
pH	5.5- 7.50	6.535 ±0.366
Dissolved oxygen mg/l	16.50- 7.8	6.775±0.777
Water hardness mg/l CaCO ₃	35- 40	37 ±1155 8

Table 2: Mortality Rate of Fingerlings of *Clarias gariepinus* exposed to varied concentrations of glyphosate

No. of tanks	No. of test fish	Concentration ml/l	Mortality					No. of mortality	Percentage mortality
			12 hours	24 hours	48 hours	72 hours	96 hours		
Control	10	0	0	0	0	0	0	0/10	0
1	10	0.0005	0	0	0	1	3	4/10	40
2	10	0.0015	0	0	0	2	3	5/10	50
3	10	0.0025	0	0	0	3	4	7/10	70
4	10	0.005	0	0	0	2	6	8/10	80
5	10	0.015	0	1	3	6	0	10/10	100

Table 3: Percentage Mortality Rate of Fingerlings of *Clarias gariepinus* Exposed to varied concentration of Glyphosate.

Concentration ml/l	Number of deaths at 96 hours			Mortality	Percentage Mortality	Probit
	Replicates					
	1	2	3			
Control	0	0	0	0/30	0	-
0.0005	4	4	4	12/30	40	4.75
0.0015	5	6	5	16/30	53	5.08
0.0025	7	8	7	21/30	70	5.52
0.005	8	9	9	26/30	87	6.13
0.015	10	10	10	30/30	100	-

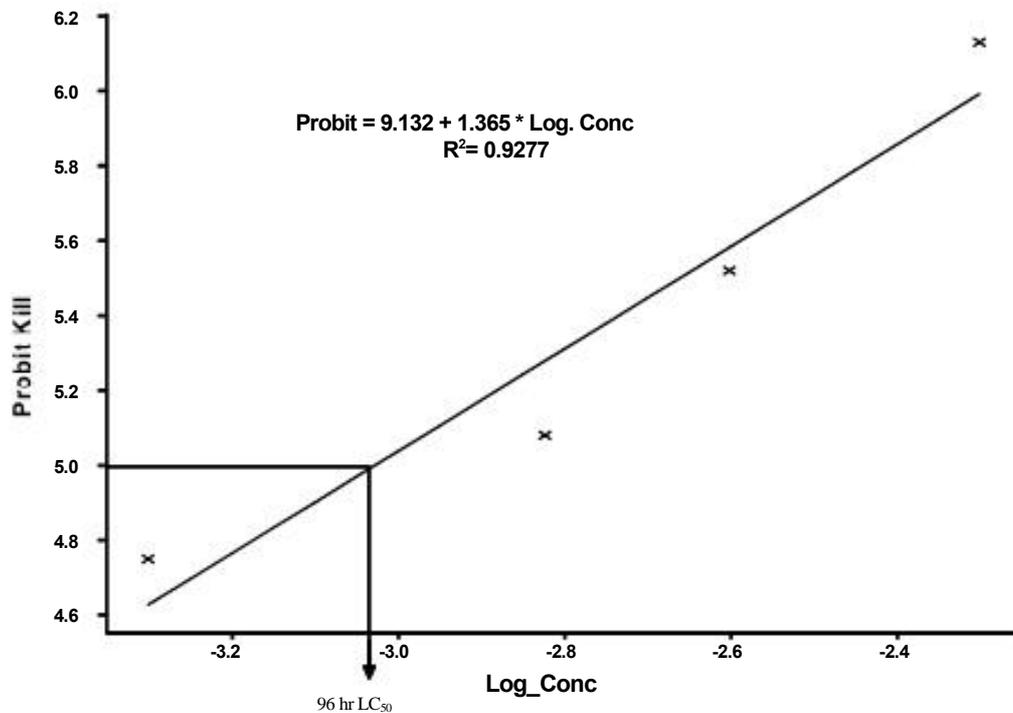


Figure 1: Relationship between probit kill and log of concentration of glyphosate for determination of 96 hrs Lc₅₀

From the investigation, it is found that glyphosate, was toxic to *Clarias gariepinus* and the effect increase with increase in concentrations. Environmental authorities need to set quality standards on use of glyphosate in aquatic ecosystem. This reduces the deleterious effects on the environment, other living aquatic organisms and man.

REFERENCES

- APHA (American Public Health Associations) (1985) Standard Methods for Examinations of Water (15th Edition), APHA, Washington D.C. 1076 pp.
- Akobundu, I.O (1987): Weed Science in the Tropics. Principles and Practices. 110 320 pp.
- Annune, P.A Ebelle, S.O and Oladimeji A.A. (1994) Acute toxicity of cadmium to juveniles of *clarias gariepinus* (Burchell) and *Oreochromis niloticus* (Trewavas), Journal of Environmental science Heath, A29 (7), 1357-1365.
- Avoaja, D.A. and Oti, E.E. (1997). Effect of Sublethal Concentrations of some Pesticides on the Growth and Survival of the Fingerling of African Freshwater Catfish "Heteroclaris" (Hybrid). Nig. Journal Biotech. 8(1): 40 47.
- Castillo, G.T. (1976): The Farmer Revisited: Towards a return to the Food Problem, Proceedings: The Worlds a Return Conference of 1979. IOWA State

- University, Ames, IOWA, USA. The IOWA State University Press, Ames, pp. 33-53.
- Cole, D.J. (1987): The Mode of Action of Glyphosate. Proc: 1982 British Crop Prot, Conf. pp. 309-15.
- Duncan, D.B. (1955). Multiple Range and Multiple F. Test. Biometrics 11 : 1-42
- Haider, S. and Inbaraji, R.M. (1986). Relative Toxicity of Technical Material and Commercial Formulation of Propan to a Freshwater Fish *Clarias gariepinus*. Ecotoxicol. Environ Saf. 11(3): 347-351.
- Hoque, M.M. Mirja, M.J.A. and Miah, M.S. (1993). Toxicity of Ronil to Fingerlings of *Clarias gariepinus*. Bangladesh J. Tran. Dev. 6(1): 19-26.
- Lovely, F. (1998). Toxicity of Three Commonly used Organophosphorous Herbicide to their Sharpute (Borbodes gonionotus) and African Catfish (*Clarias gariepinus*) Fry. Department of Fisheries and Genetics. Bangladesh Agricultural University, Mymensingh, Bangladesh. M.Sc. Thesis. 83 pp.
- Meleter, V.V. Kanaly A.S and Sokotovo N.G. (1971) Water toxicity American publishing co, PVT.Ltd 216p.
- Oti, E.E. (2000). Acute toxicity of water Extracts of Bark of *Thevetia peruviana* to the African Freshwater Catfish. "Heteroclaris" (Hybrid) fingerlings. J. Fish. Tech. vol. 2, 124-130 pp.
- Ovie, S.I. (1985). Zooplankton Study of Round Valley Reservoir, New Jersey, USA, Rutgers University New Brunswick, New Jersey, USA M.Sc. Thesis. 10 pp.
- Rao, K.R. Sumbasiva, S. Basu, K.S and Ramanarao, K.V. (1985) Toxicity of Elsan to *Clarias gariepinus*. Indian J. Fish 32 : 153-157.
- Ronald, E. (1989). Atrazine hazard to Fish, Wildlife and Invertible. A Synoptic Reviews, Springer Verlag. New York 107-125 pp.
- Rodosevich, S.R. and Holt, J.S. (1984). Weed Ecology: Implication for Vegetation Management. Wiley Interscience, New York. 265 pp.
- Shultz, D (1970,1971) C ited In. Fish pathology by Heinzherman Reichenback Klinke in collaboration with Marsha landolt T.F.H publication Inc. New York Jersey. 512pp.
- Stefferd, A. (ed.) (1990). The 1990 Yearbook of Agricultural Soils, USDA, Washington D.C. USA Government Printer, 784 pp.