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Preliminary Investigation of Permethrin Toxicity to Fry of African Mud Catfish, *Clarias gariepinus* (Burchell, 1822)

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Abstract: Permethrin-based insecticides are amongst first-line household insecticides in Nigeria, applied against common insect vermin such mosquitoes and cockroaches. They are affordably applied at homes, stores and gardens, including near fish culture setups. Acute static bioassay was carried out under laboratory conditions to determine the toxic effect of permethrin to fry of *Clarias gariepinus* (African mud cat fish). Hatchery-bred fry of *C. gariepinus* weighing 0.1g to 0.2g were exposed to different concentrations (0.05mg/l, 0.1mg/l, 0.15mg/l, 0.2mg/l and 0.25mg/l) of permethrin for 96 hours. Fry in the treatment media showed prolonged active swimming responses and the mobile severity was concentration and exposure-time dependent. Mortal damage was also concentration dependent. The low LC_{50} (median lethal concentration) value of 0.08mg/l recorded indicates that permethrin was acutely toxic to the species early life stage. Extreme care should be taken in applying permethrin based insecticides as the pesticide residue can be washed in run-off to natural fluvial habitats of *C. gariepinus*, as well as near residential fish culture enclosures, causing mortality of the fish critical fry stage with overall consequences on protein availability.

Key words: Acute toxicity · Pesticides · Mortality · Bioaccumulation · Synergistic effect

INTRODUCTION

The high rate of insect pest infestation in the tropics has led to frequent use of toxic chemicals in the form of insecticides which, in the long term, has significant toxic effect on non-target organisms. The effects range from outright mortality to subtle disruption of physiological processes [1, 2]. The toxic properties of many chemicals are unknown even when they are widely applied. In addition, varieties of chemicals combined together can have unknown synergistic effects even when the toxicity of each individual chemical is well-known [3, 4].

Permethrin-based insecticides are amongst the commonly used household insecticides in Nigeria, since permethrin has the ability to kill varieties of insect [5]. They are used against a number of pests, on nuts, fruits, vegetables, cotton, potatoes and cereal crops. It also controls animal ectoparasites, biting flies and cockroaches. It has a high insecticidal property which can persist up to 12 weeks after application [6]. These synthetic chemical, can also be processed into different forms ranging from dust, emulsifiable concentrates, smokes, ULV (Ultra Low Volume) and wettable powder formulations [5]. In particular, permethrin is the active ingredient of a common household insect powder used in Nigeria called "Rambo"; same as ''otaapiapia'', a widely locally applied insect killer liquid.

Aquatic ecosystems are vulnerable to the impact of insecticides, as they have multifaceted uptake pathways. The indiscriminate application of insecticides can generate sufficient residues that can contaminate inland waters. Inland fresh water swamps, rivulets and rivers serve as natural habitats for diverse fauna, including the highly relished African mud cat fish *C. gariepinus* [7-9]. *C. gariepinus* is known to be susceptible to environmental degradation and pollution [10, 11]. Recently, the Fisheries Society of Nigeria (FISON)

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emphasized the benefits of culture fishery, in terms of domestic fish production and income generation derivable from rearing fish in low-cost mobile fishponds [12]. They recommend fish culture in collapsible plastic tanks that, can be used by landless individuals, including the frontage of living apartments of urban tenants. Such near-living apartment homestead culture facilities would be particularly vulnerable to direct contact or residues of household routinely applied insecticides.

Dearth of primary information exists on permethrin toxicity on aquatic fauna [13, 14]. More so, there is no published data on the toxic impact of permethrin on the survival of the different life stages of *C. gariepinus*, an important widely cultured fish in Nigeria. This study was aimed at providing preliminary information on the effect of permethrin on survival of the fry of *C. gariepinus*.

MATERIALS AND METHODS

Experimental Fish: Hatchery bred post-yolk fry of *C. gariepinus* (0.1g to 0.2g in weight) were obtained from the Faculty of Agriculture Demonstration Farm, University of Port Harcourt where the experiment was conducted. The tested fry were first acclimatized to ambient conditions and fed for a period of 7days. Feeding was avoided 24 hours prior to the toxicity test. Dead fry and those with signs of poor state were removed from the holding tanks.

Exposure Toxicant: Sample bottle of Liquid permethrin was purchased from a chemical shop in Port Harcourt, Rivers State, Nigeria and stored in a cool dry place at temperature ranging from 26°C - 27°C.

Experimental Conditions and Procedures: Preliminary (Experiment 1) dose-range finding test [15] was carried out as static bioassay at room temperature. Three concentrations viz; 0.25mg/l, 0.20mg/l and 0.15mg/l were made from the stock solution prepared from serial dilution using a 500ml conical flask. Water used as control and in diluting the test media was obtained from the same source (borehole water) from which the fry were hatched. Each medium had three replicates and the experiment lasted for 72 hours.

Experiment 2 was carried out one week after experiment 1. Five (5) concentrations were prepared namely; 0.05mg/l, 0.1mg/l, 0.15mg/l, 0.20mg/l and 0.25mg/l. Each treatment and the control had 3 replicates. The fry were randomly and carefully introduced into 1000ml of each concentration in a 2litre plastic container using a small mesh-sized dip net. Each test medium had 10 fry making a total of 30 fry per concentration [16]. Observations were made for loss of balance and the death of fry. The experiment lasted for 96 hours (4 days).

Death of a fry was assumed to occur when it laid motionless at the bottom of the container and showed no response to gentle prodding. No feed was given to fry during the experiments. The behaviour of the tested specimens was observed and death was recorded for the 96hours. The median concentration, LC_{50} was determined by the arithmetic graph method (a plot of percentage death against concentration and extrapolating to the concentration at which 50% of test organisms were killed). Daily temperature, pH and dissolved oxygen concentration were measured in the test media with mercury in-glass thermometer, pH meter (Hanna pHep pocket-sized pH meter) and Milwaukee DO meter (model MW600) respectively.

RESULTS

Behavioural Observation: Save for the control, high intensity of vertical swimming was observed for fry in all the treatments, which was concentration dependent; more intense swimming in treatment IV and V (that is, 0.2mg/l and 0.25mg/l respectively). Succeeding intense swimming was loss of balance, followed by the fry remaining motionless at the bottom of the experimental containers.

Mortality: No mortality was recorded in the control replicates (Table 1). Mortal damage occurred in all the treated media. However, mortality rate was apparently concentration dependent. While 70% of the tested population survived in the 0.05mg/l, no survivor was recorded in the 0.25mg/l medium (Table 2). Figure 1 shows a plot of 50% mortality recorded in experiment 1 against time taken in hours. MT50 values (Fig. 1) indicate a direct relationship between fry mortality and concentration of the test media.

The three concentration 0.25mg/l, 0.2mg/l and 0.15mg/l had MT50 value of 3,6 and 10 hours respectively. Figure 2 shows the results of 50% mortality recorded in experiment 2. The plot also shows an inverse relationship between the time to death and concentration of the test media.

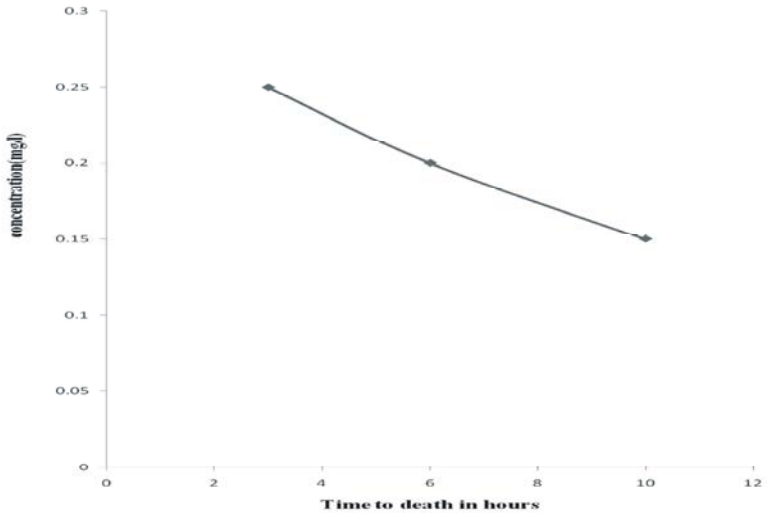

Fig. 1: A plot of 50% mortality of *C. gariepinus* fry recorded in the different concentrations of Experiment 1 against time taken in hours

Consequently, 0.25mg/l, 0.20mg/l, 0.5mg/l, 0.01mg/l and 0.05mg/l killed 50% of the tested population in 5, 8, 15, 30 and 55 hours respectively. The 96hours $LC_{50}$was 0.08mg/l as indicated in Figure 3.

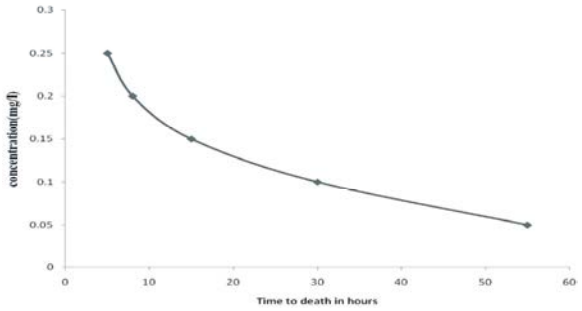

Fig. 2: A plot of concentration of test media against time taken to record 50% mortality (Experiment 2)

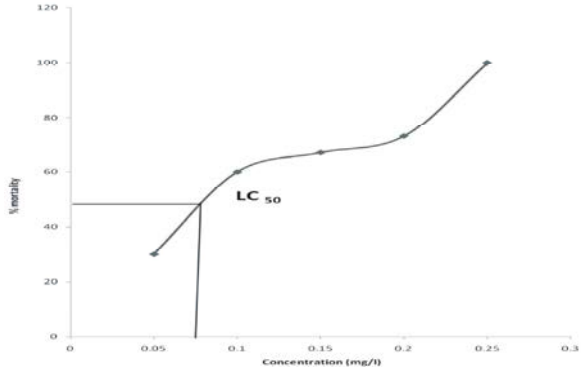

Fig. 3: A plot of percentage mortality against concentration to determine 96hours LC_{50} of *Clarias gariepinus*(experiment 2)

Temperature varied between 26.5 °C to 26.7 °C, pH varied between 6.58 to 6.70 and dissolved oxygen varied between 3.55 to 4.12 mg/l. Table 3 shows mean values of temperature, pH and DO.

Table 1: shows the no. of fry survivors at the end of 96hours

Conc. Mg/l	Rep 1	Rep 2	Rep 3
control	10	10	10
0.05	5	8	8
0.01	6	4	2
0.15	3	4	3
0.2	2	3	3
0.25	0	O	0

Table 2: Shows the percentage fry mortality recorded in each replicate medium (1-3) for each treatment and control at the end of 96 hours.

Conc. Mg/l	Rep 1	Rep 2	Rep 3
control	0	0	0
0.05	50	20	20
0.01	40	60	80
0.15	70	60	70
0.2	80	70	70
0.25	100	100	100

Table 3: Mean ± S.D of pH, temperature and dissolve oxygen monitored during experiment 2.

Concentration mg/l	PH	Temperature	Dissolve oxygen mg/l
Control	6.60±0.04	26.7±0.05	3.98±0.06
0.05	6.69±0.03	26.7±0.05	3.12±0.07
0.10	6.68±0.04	26.7±0.05	3.93±0.05
0.15	6.70±0.05	26.5±0.04	3.75±0.06
0.20	6.55±0.04	26.6±0.04	3.78±0.06
0.25	6.58±0.04	26.6±0.05	3.55±0.05

DISCUSSION

The need to determine the toxicity effect of permethrin to vulnerable non-target species, particularly food species, cannot be over-emphasized. As recorded in

this study, reasonably low concentration of permethrin can kill *C. gariepinus* fry. Such toxicity mediated mortality is dependent on a number of factors, the most important factor being the dose-time relationship [16, 17]. Secondly, mortality could be as a result of indirect effects whereby the toxicant distorts the fry gills respiratory function, leading to asphyxiation. The extent of depletion of oxygen in water is often proportional to the concentration of pollutants present in water [18].

Although fin fishes may have inherent mechanism of secreting detoxifying enzymes to counteract toxic effects when exposed to toxicant such as permethrin, the physiological response could only be achievable over a long term chronic exposure period [19].

Comparing Fig 1 and 2, it was obvious that permethrin toxicity to *C. gariepinus* fry was exposure time dependent. The fry in experiment 2 which were tested with the same stock of toxicant one week after experiment 1 showed higher tolerance and resistance to the treated concentrations. This implies that as the toxicant ages, toxicity decreases. In addition, the toxicity effect of permethrin was inversely related to the time of exposure. This is because degradation has an important function of reducing the toxicity of the toxicant. Degradation is time dependent and also depends greatly on the ambient temperature and light intensity [6]. Other major factors influencing degradation are pH, the level of dissolved oxygen and the presence of micro-organisms [20]. Consequently, much death occurred within the first 10hours beyond which degradation sets in and the concentration reduced such that it took longer time for another death to occur again. It was observed by Afolabi *et al*., [19] that almost all the damage done to their experimented organisms took place in the early hours of exposure and death declined as the concentration decreases due to evaporation.

Fry that survived in the treatment media may have accumulated reasonably amount of toxin or had their physiology impaired already that could possibly lead to post test death [10]. In addition, there are food safety repercussions for human consumption of toxic bio-accumulated fish [2].

Following the toxicity criteria of the OECD [21] hazard classification system, permethrin was highly acute toxic to *C. gariepinus* fry. Further toxicity test is recommended to complement the present data, which will serve as a basis for policy makers to set out local guidelines for the application of permethrin-based pesticides in riparian areas.

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