BIOLOGICAL SCIENCES

THE SUSCEPTIBILITY OF FINGERLINGS OF *OREOCHROMIS NILOTICUS* (L.) TO THREE SYNTHETIC INSECTICIDES COMMONLY USED IN GHANA

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ABSTRACT

The acute toxicities of three synthetic insecticides, Thiodan 35EC, Cymbush 10EC and Roxion 40EC, used in the control of agricultural pests in Ghana to the fingerlings of Oreochromis niloticus were investigated in freshwater at 26 - 28°C under static bioassay test conditions.

The 96-hr LC50's of Thiodan and Roxion were 2.66 ppb and 12,232.07 ppb respectively; the fingerlings of 0. niloticus are thus about 4600 times more susceptible to Thiodan, an organochlorine insecticide than to an organophosphate insecticide. The 12-hr LC50 of Cymbush was 446.68 ppb.

The general trend of sequential events in the reaction of O. niloticus to the various concentrations of the test chemical were: high excitability, erratic and uncoordinated movements, loss of equilibrium and finally death.

Keywords: Susceptibility, acute toxicity, synthetic insecticides, fingerlings, *Oreochromis niloticus.*

INTRODUCTION

To increase food production, insecticides, mostly synthetic are applied to crops to combat insect pests which contribute to crop losses on the field. However, these insecticides eventually find their way into water bodies, for example, ponds, lakes, streams, rivers, underground water and sea, through run offs from contaminated fields. Some of these insecticides, at high concentrations, can cause massive fish-kills directly or indirectly by detrimentally affecting the food-chain [1].

At very low concentrations, some insecticides may also cause fish mortality [2]. Additional consequences of sublethal concentrations of insecticides are the possible effect on growth and yield of fish and accumulation of the organic residues in fish flesh [3,4,5]. Work on the toxicity of various pesticides to fish has been extensively reviewed by Murty [6,7].

Thiodan 35EC, Cymbush 10EC and Roxion 40EC are synthetic insecticides, which are extensively used in Ghana for agricultural purposes. According to the manufacturers (e.g. Food Machinery and Chemical Corporation, F.M.C; Imperial Chemical Industries, I.C.I; and Celamerk GMbH and Co.K.G. Ingelheim/Rhein, (Germany), these three insecticides are toxic to fish, but there is apparently a dearth of quantitative information on their level of toxicity to fish and other aquatic fauna. The present work thus seeks to evaluate the toxicity of these synthetic insecticides on *Oreochromis niloticus*, a commercial fish species found in many inland water bodies in Ghana and also the most common species used for stocking of fish ponds.

MATERIALS AND METHODS

The fingerlings of *O. niloticus* used in the experiments were collected from the University of Science and Technology fish pond. They were acclimatised in the laboratory in aerated, dechlorinated tap water contained in 200 l disinfected glass aquarium tanks for at least six days. The fingerlings were not fed 24 hours before the tests were carried out and during the period of experiments.

The temperature, pH and dissolved oxygen content of test solutions were determined at the beginning and end of the experiment.

A series of preliminary tests were performed to establish the minimum concentration of each test chemical required to kill all the test fish within a period of 24 hours [8]. Consequently, various concentrations of each individual test chemical were prepared within the estimated range by diluting measured volumes with dechlorinated tap water.

The toxicity of insecticides was studied using the static bioassay test method as has been described by EIFAC [8] and Reish and Oshida [9] with some modifications.

Ten healthy fingerlings were put in each test solution of 10 l. Each experiment was replicated three times. Control experiments with 10 l of test solutions made up of only dechlorinated tap water were concurrently run with ten fingerlings for each insecticide tested. The experiments were conducted for 24, 48, 72 and 96 hours. During this period, behaviour, loss of equilibrium, mortality and abnormalities if any, were recorded. Individuals showing no respiratory movement and no response to a tactile stimulus were recorded as dead and were removed.

Observations were made initially at hourly intervals and then later at different time intervals throughout the experimental period..

Between experiments, test containers were cleansed with detergent, filled with tap water, allowed to stand for 24 hours, washed again and then rinsed carefully with tap water. This procedure removed insecticides, which adhered to the glass as indicated by periodic bioassays of cleansed aquarium tanks.

The LC 50 of each tested insecticide was calculated using probit analysis [10] and the 95% confidence limits around the LC 50 computed according to Busvine [11].


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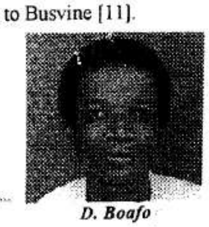
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TABLE 1: **Mean Weights of Fingerlings of *O. Niloticus* used in Definitive Test at Various Concentrations of Test Chemicals and the Susceptibility of Fingerlings to Test Chemicals**

TEST CHEMICAL	CONCENTRATION (ppb)	MEAN WEIGHT* OF FINGERLINGS (g) ± S.E.	L.S.D OF MEAN WEIGHTS	PERCENTAGE MORTALITY	LT 50 (hr)	LT 100 (hr)
THIODAN	0	1.001 ± 0.0174	-	0	-	-
	0.35	1.071 ± 0.0193	-	3.3	-	-
	0.70	1.063 ± 0.0155	0.612 NS	6.7	-	-
	1.40	1.039 ± 0.0191	-	3.3	-	-
	2.80	1.048 ± 0.0197	-	26.7	-	-
	5.60	1.029 ± 0.0222	-	100	15	26
CYMBUSH	0	1.012 ± 0.0241	-	0	-	-
	40	1.023 ± 0.0197	-	6.7	-	-
	80	1.033 ± 0.0202	0.586 NS	93.3	19	-
	160	1.052 ± 0.0230	-	100	21	61
	320	1.030 ± 0.0200	-	100	16	38
	640	1.055 ± 0.0238	-	100	10	34
ROXION	0	0.972 ± 0.0126	-	0	-	-
	2500	0.999 ± 0.0195	-	16.7	-	-
	5000	0.996 ± 0.0188	0.302 NS	13.3	-	-
	10000	0.992 ± 0.0157	-	13.3	-	-
	20000	0.997 ± 0.0177	-	73.3	35	-
	40000	0.999 ± 0.0176	-	100	43 min	1

* AVERAGE NUMBER OF FINGERLINGS PER CONCENTRATION = 10
TOTAL NUMBER OF FINGERLINGS PER CONCENTRATION = 30
NS - NOT SIGNIFICANT (P = 0.05)

RESULTS

The mean weights of fingerlings of *O. niloticus* used in each of the various test solutions were not significantly different (P = 0.05) (see Table 1). The temperature of the test solutions for all the test chemicals throughout the period was in the range of 26° - 28°C. The pH was 7. The dissolved oxygen content of the test solutions ranged from 6.4 mg/l to 7.8 mg/l.

The percentage mortality of *O. niloticus* generally decreased with decrease in concentration of the test chemicals (Table 1 and Fig 1). The median lethal time (LT 50) as well as the LT 100 decreased with increase in concentration of the test chemicals (Table 1).

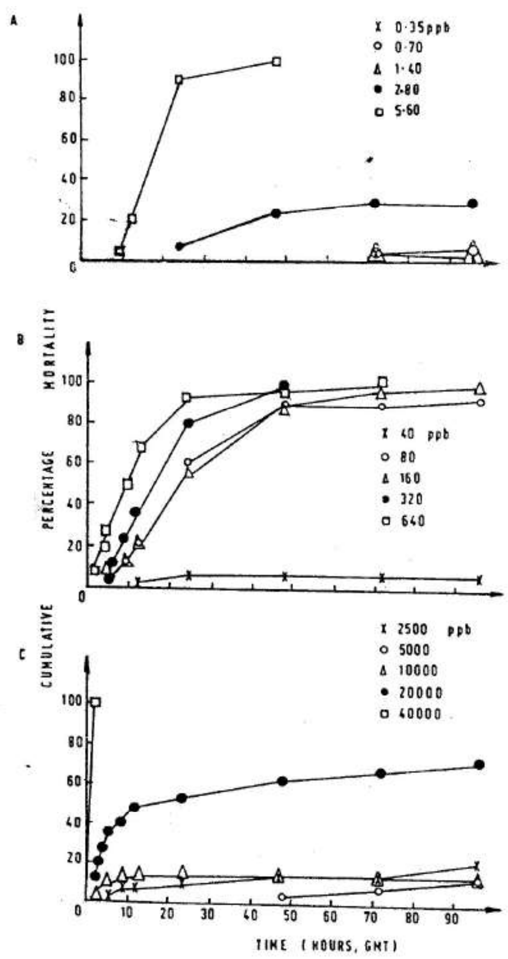

Fig. 1: Cumulative percentage mortality of *O. niloticus* versus time (hours) at different concentrations of (a). Thiodan (b). Cymbush and (c). Roxion

The manifestation time, which is the time that the toxic process was initiated, generally varied with the type of test chemical and the different concentrations of the test chemical. At the highest concentration of Thiodan, 5.60 ppb, the first effects were not apparent until after 5 hours of exposure of *O. niloticus*. The manifestation time in the case of Cymbush at the highest concentration of 640 ppb was 1 hour. The shortest manifestation time of 5 minutes was noted for Roxion at a concentration of 40,000 ppb.

During the first few minutes or hours depending on the concentration of a test chemical, fingerlings of *O. niloticus* remained calm and showed no sign of stress. Later they moved rapidly and tried to jump out of the test solutions. Death was preceded by aggressive erratic, and uncoordinated movements and spasms. These movements were followed by longer and longer periods of quiescence until respiratory movement ceased and fingerlings no more responded to mechanical stimuli. After death, most of the fish had their mouths and opercula wide opened.

The 96-hr LC 50 values of Thiodan and Roxion were found to be 2.66 ppb and 12,232.07 ppb respectively. The 12hr LC 50 of Cymbush was, however, 446.68 ppb [Fig 2].

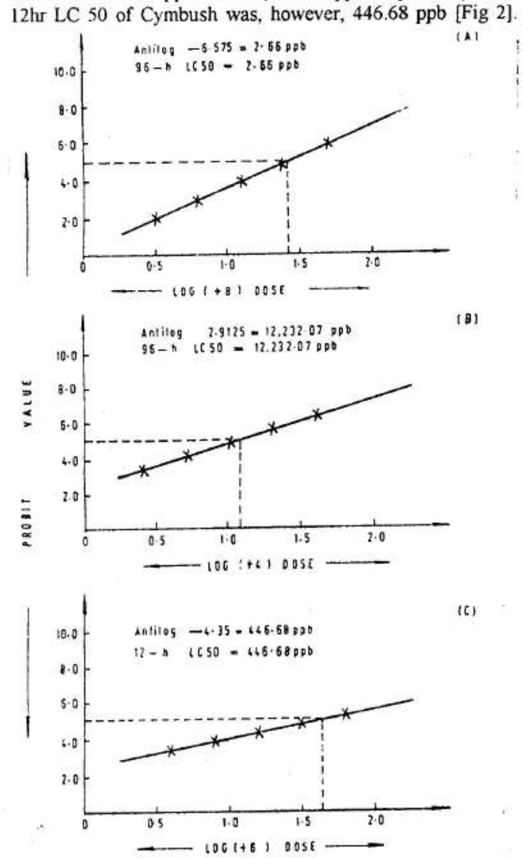

Fig. 2: Graph of: (a). 96-h LC50 of Thiodan 35EC, (b). 96-h LC50 of Roxion 40EC and (c). 12-h LC50 of Cymbush 10EC, for fingerlings of *Oreochromis niloticus*.

TABLE 2: **The Lc 50 Values of Insecticides for Fingerlings of *Oreochromis Niloticus*, Together with Lower and Upper Lc 50 Values Within 95% Confidence Limits**

Insecticide	Time (hr)	LC 50 (ppb)	95% confidence interval
Thiodan	96	2.66	2.60 to 2.71
Roxion	96	12,232.07	11,915.16 to 12,557.40
Cymbush	12	446.68	428.35 to 465.80

The lower and upper limits of LC 50's for the insecticides to *O. niloticus* within 95% confidence limits are given in Table 2.

DISCUSSION

Chemical preparations, such as Thiodan, Cymbush and Rioxion, used for agricultural purposes, have contributed to pollution of inland waters creating problems for freshwater fisheries. The effects of insecticides depend on a number of factors including the species and size of the organism, the concentration and persistence of the pollutants, temperature, pH, dissolved oxygen content, hardness of water, turbidity as well as the presence of other pollutants which may result in synergistic effects.

The fingerlings of *O. niloticus* used in the test were homogeneous in size ($p = 0.05$). The observed effects of the chemicals were thus independent of any variation in weight or size of the fingerlings. The observed temperature range (26° - 28°C) and pH value of 7 for the test solutions were within the optimum range for the development of *O. niloticus* [1, 12]. For standard tests the recommended dissolved oxygen content of the test solutions for warm water fish should be greater than 4 mg/l [8]; the dissolved oxygen content of test solutions in the present study varied from 6.4 to 7.8 mg/l.

The reaction of fish to most toxicants is similar. In acute poisoning, the first sign is an increase in activity due to irritability and hypersensitivity to tactile and sometimes to auditory stimuli [13]. Hepher and Pruginin [1] observed that the fish at first showed a brief period of high excitability, followed by alternate periods of muscular spasms, and then by complete loss of equilibrium, with the fish turning on their longer axes. In practice not much could be done to save the fish; death followed after a variable period. Henderson *et al.* [2] in their study of organochlorine insecticides also noted that the physiological reactions of their test fish were somewhat similar for all compounds tested except BHC (Lindane) where the fish sank slowly to the bottom of the aquarium expiring gradually. The reaction of *O. niloticus* to various concentrations of Thiodan, Cymbush and Roxion also confirmed that reported by earlier workers [1,2,13].

The manifestation time may be different with different chemicals and different concentrations of the chemicals as was observed in the current study and also reported by Schoettger [14]. In the present study, the manifestation time was found to decrease with increasing concentrations of the test chemical. At high concentrations of the test chemicals, the first effects were not apparent in *O. niloticus* until after 5-60 minutes, thus confirming reports by other workers such as Hepher and Pruginin [1] and Henderson *et al* [2].

Most of the dead fish were observed to have their mouths and opercula wide opened. Death might have been a result of respiratory arrest as also noted by Barnes [13] and Henderson *et al.* [2].

The 96-hr LC50's of Thiodan [2.66 ppb] and Roxion [12,232.07 ppb] for *O. niloticus* indicate that *O. niloticus* is about 4,600 times more susceptible to Thiodan, a chlorinated hydrocarbon than Roxion, an organic phosphorous insecticide. Similar observations on the relatively high levels of toxicity of organochlorine insecticides compared to organophosphate insecticides have been made by other workers such as Gruber [15], Kimura and Matida [16] and Lahav and Sarig [17]. Thiodan has also been shown to be highly toxic to a variety of species in laboratory and field trials [18, 19]. 46 ppb of Thiodan applied to a 27 acre pond killed all minnows, perch, sunfish, bullheads and suckers within 7 days [20].

There is apparently a dearth of data in the effects of synthetic pyrethroids including Cymbush on tropical fish. However, it is known that Cymbush is toxic to fish [21]. Duah [22] observed that the 96-hr LC50 of Cymbush to fingerlings of *O. niloticus* was 55.2 ppb. Coats *et al* [23] and Kumaraguru *et al.* [24] have also reported that some synthetic pyrethroids have high toxicities to fish (96-hr LC50's of 0.037 ppm to gold fish and 0.008 ppm to bluegeill).

The present study indicates that Cymbush is less toxic than Thiodan but more toxic than Roxion to fingerlings of *O. niloticus*. Golow *et al.* [25] have however, observed that Deltamethrin, a synthetic pyrethroid is two times more toxic (96-hr LC50 of 0.0145 mg/l), to *O. niloticus* than dieldrin (96-hr LC50 of 0.030 mg/l) a chlorininated hydrocarbon.

In Ghana, the demand for pesticides has grown substantially as a result of multiple cropping and other intensified agricultural practices as well as for uses in the control of disease vectors. Some of these pesticides indiscriminately disposed on land leach as surface run off into reservoirs and other inland water bodies. Concentrations of Thiodan, Cymbush and Roxion as applied to crops on the field against insect pests and as recommended by the manufacturers are quite high being 350,000 - 700,000 ppb, 30,000 - 1,167,000 ppb and 200,000-600,000 ppb respectively. Contamination of water body by these insecticides at high concentrations, through careless spraying or by accidental spillage could cause heavy mortalities to the fish, which form part of the aquatic ecosystem. Such contamination of lake water, through leakage of Thiodan from drums waiting to be transported by ferryboat to Northern Ghana, was found to cause heavy mortality to fish along the shores of the Volta Lake [26].

Exposure of adult fish even at sublethal levels for long periods may produce unfavourable effects influencing their behaviour, reproductive capacity and adaptability for survival. Indiscriminate use of the insecticides under study can exert adverse effects on the juvenile fish and thereby affect fish production.

CONCLUSION

Of the three insecticides tested on fingerlings of *O. niloticus*, Thiodan, an organochlorine compound was found to be the most Toxic. The fingerlings were about 4,600 times more susceptible to Thiodan than Roxion, an organophosphorus compound.

The insecticides induced behavioural changes in the exposed fish. Indiscriminate use of these insecticides can have unfavourable effects on the juvenile stages of fish life and could thus affect fish production. It remains necessary, therefore, to ensure by education and instruction that these insecticides are used properly.

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