

Full Length Research Paper

Acute toxicity of *Lepidagathis alopecuriodes* (Vahl) to *Heterobranchus bidorsalis* fingerlings

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The acute toxicity of *L. alopecuriodes* to *H. bidorsalis* fingerlings of mean body weight, $1.99 \pm 0.38\text{g}$ and total length $6.32 \pm 0.93\text{cm}$ was assessed in a renewal bioassay for 96 h. Five graded concentrations of *L. alopecuriodes* were prepared as 0.5, 1.0, 1.5, 2.0 and 2.25mg/L and a control experiment (0.00 mg/L). There were no significant variations observed in the water quality parameters of the test solutions. Toxic reactions exhibited by the fish include hyperventilation, erratic swimming, incessant gulping of air, loss of reflex and death. Opercular beat frequency (OBF) and Tail beat frequency (TBF) decreased with increase in time of exposure but increased with concentration. There were significant interactions between concentration and time of mortality and OBF. Concentration of the extract shows a positive linear correlation with OBF, TBF and mortality. No death was recorded in the control (0.00mg/L) throughout the period of the experiment. The 96 h LC_{50} (the concentration required to kill 50% of the fish after 96 h of exposure) value computed was 0.59 mg/L. The median LT_{50} (the time needed to kill 50% of the fish at various concentration applied) was 83.60 and 19.05 h for 0.50 and 2.25 mg/L, respectively. The results show that *L. alopecuriodes* is highly toxic to *H. bidorsalis* fingerlings.

Key words: Acute toxicity, *Lepidagathis alopecuroides*, opercular beat frequency, tail beat frequency, mortality.

INTRODUCTION

Lepidagathis alopecuroides (vahl) (Family Acanthaceae) commonly known as 'Ogwuaru' among the Ikwere and 'Esie-ebua' among the Bosis in Rivers and Cross River States of Nigeria, respectively, have been reportedly used by artisanal fishers in fishing (Adediran, 2004). The extract of the plant shows larvicidal action against *Anopheles gambiae*, *Culex quinquefasciatus* (Obomanu et al., 2006) and tadpoles (Suileman, 2005). The plant is also used for treatment of abdominal pains and diarrhea, suggesting its antimicrobial contents (Obomanu et al., 2005). Photochemical screening of the plant shows the presence of alkaloids, saponins, tannins, glycosides and flavonoid (Obomanu et al., 2005).

Heterobranchus bidorsalis is an African catfish common in Nigerian waters that has not been well studied as other catfish of the same family Clariidae. Members of the genus *Heterobranchus* have been reported to have a higher growth rate and more resistance to environmental stress factors than other catfish especially with regards to disease conditions (Offem and Ikpi, 2013). The use of fish ichthyotoxins when estimating fish stocks has been mentioned by Brandt, 1984; Seyani and Chiotha, 1991; Ibrahim et al., 2000.

The effective concentration and time of toxicity of some toxicants have not been studied deeply. The toxicity of

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L. alopecuroides to mosquito larvae and tadpoles has been established (Adediran, 2004; Suleiman, 2006). In Rivers State, *L. alopecuroides* is used as quick kill for mudskippers (Obomanu et al., 2007). Before death, fish shows a number of measurable behaviours that are indicative of internal disturbances and stress responses, characterized by physiological changes (Dutta et al., 1994). The effect of pollutants on fish is assessed by acute and chronic toxicity tests (Heath, 1991). Toxicity of *L. alopecuroides* has been reported on *C. gariepinus* and catfish hybrid fingerlings (Gabriel et al., 2008a; 2008b; Gabriel and Okey, 2009) and Nile tilapia, *O. niloticus* (Okey et al., 2013) but none yet on *H. bidorsalis* fingerlings.

This study was conducted to determine the behavioural responses, Opercular beat frequency (Obf), Tail beat frequency, (Tbf) and 96 h lethal concentration (LCs), Mean Lethal Time (MLTs) for freshwater catfish *H. bidorsalis* that are actively fished using poisonous plant materials.

MATERIALS AND METHODS

Bioassay was conducted in the Fisheries Wet Laboratory, Rivers State University of Science and Technology. Two hundred and fifty *H. bidorsalis* fingerlings (mean body weight, 1.99 ± 0.38 g and length, 6.32 ± 0.93 cm) were procured from Idomor Farm Ltd., Olomoro in Delta State, Nigeria and acclimated to Laboratory conditions using glass tanks ($45 \times 40 \times 40$ cm) of 40 L capacity, filled with 30 L of tap water. The water in the containers was renewed daily and the fish were fed with commercial fish feed (Pellets) containing 40% crude protein at 1% of their body weight. Unconsumed feed and faecal wastes were removed; and the water replenished regularly as recommended by Oyelese and Faturoti (1995).

Feeding was discontinued 24 h before the commencement of and during experiment, to minimize the contamination of the test aquaria. *L. alopecuroides* were procured from Ogbakiri in Emohua Local Government Area in Rivers State, identified in the Department of Applied Biology Rivers State University of Science and Technology. The leaves were sun dried, then pulverized with a sterile grinding machine and sieved with a 100-micron sieve to obtain a fine powder.

A range finding test was carried out as described by Omitoyin et al. (2006) to determine the concentrations of *L. alopecuroides* used in the definite test. Based on the results of the range finding test, 2 g of *L. alopecuroides* powder was obtained and dissolved in distilled water to form a stock solution of 200mg/L of the material, the following concentrations were prepared and introduced into the experimental glass tanks 0.00 (control), 0.50, 1.00, 1.50, 2.00 and 2.25 mg/L in replicates of three.

A completely randomized design was used with 10 fish/ 20 L of tap water. The experiment lasted for 96 h and test solutions were renewed daily. The water quality parameters such as dissolved oxygen, temperature and the pH of the test media were measured and recorded daily before and after the toxicant, was added for the 96 h exposure period following the standard method on water quality assessment by APHA (1998). The behaviour (Obf and Tbf), mortality and other external changes in the body of the test fish were observed and recorded at 12, 24, 48, 72 and 96 h of exposure time as described by Gabriel and Kparobo (2003) and Omitoyin et al. (2006).

Dead fish were promptly removed to avoid contamination of the

test media. Data obtained from the experiments were subjected to a multivariate analysis for Two- way ANOVA using Statistical Package for the Social Sciences, (SPSS) version 15. Where differences exist, Tukey –Honest Significant different was used to separate them and F- test used for significant differences at ($p < 0.05$) between the various treatments (Wahua, 1999). Correlation and regression between OBF min^{-1} , TBF min^{-1} and cumulative mortality against concentrations and time for the species was done according to Zar (1996). An analysis of the lethal concentration (LCs) and median lethal time (MLTs) with associated confidence interval were done using Probit Analysis. Safe concentrations at the various time intervals were obtained by multiplying the lethal concentration (LC_{50}) by a factor of 0.1 (EIFAC, 1983; Koesoemadinata, 1980).

RESULTS

Water quality parameters

The mean water quality parameter of temperature, dissolved oxygen, alkalinity, total hardness and pH of the various concentrations did not vary significantly ($P < 0.05$) from those of control. In the control it showed that temperature was 27°C , dissolved oxygen 5.70 mg/L, alkalinity 18.00 meq/L, total hardness 17.97CaCO_3 mg/L and pH 8.12. The mean value recorded for the various concentrations were similar to those of the control as shown in Table 1.

Behavioural responses (OBFmin^{-1} and TBFmin^{-1}) and mortality of *Heterobranchus bidorsalis* exposed to *L. alopecuroides* for 96 h

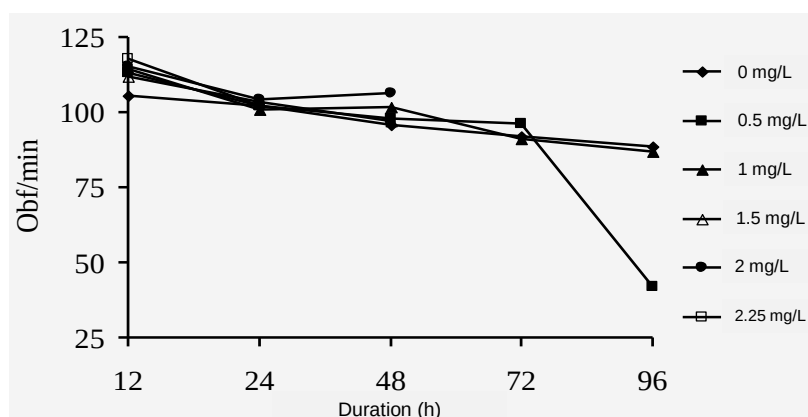
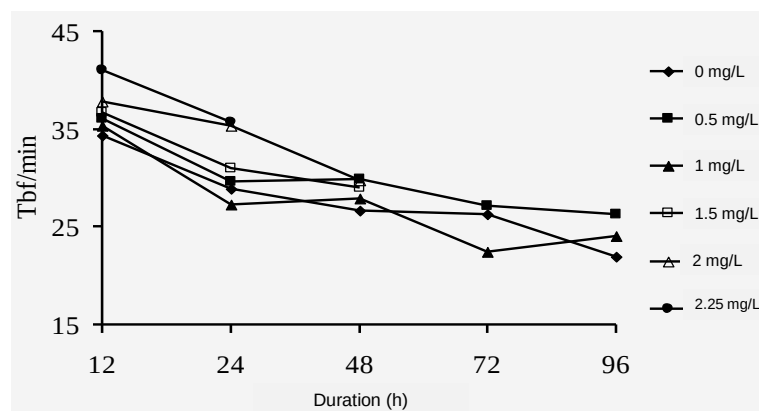
Heterobranchus bidorsalis fingerlings exposed to *L. alopecuroides* at different concentrations shows initial distress swimming movements, rapid opercular movements, loss of balance, incessant gulping of air, excessive mucus secretion, unusual lethargy and fish settling at the bottom motionless with slow opercular movement and erratic swimming before death. At lower concentrations (1.00mg/L and less) such changes were minimal and the control group showed no such signs. The abnormal behaviour displayed by the fish increased with increasing concentrations of *L. alopecuroides* in water but decreased with increase in time of exposure which then gradually reduced at higher concentrations.

The pattern of response of OBF/min and TBF/min to time of exposure and concentration of the toxicant decreased with time, but increased with increase in concentration while cumulative mortality increased with both increase in concentration and duration of exposure (Figures 1- 3). Differences in the responses of OBF to the interactive effects of the various concentrations of *L. alopecuroides* and time of exposure were narrowest between 12^{th} – 72^{nd} h, and more disperse at the 96^{th} h. Changes in cumulative mortality were narrowest at the 12^{th} h and becoming more variable with increase in exposure time. No mortality was recorded in the control.

Table 1. Mean and standard deviation of water quality parameters of various exposure solutions.

Parameter	Concentrations of <i>L. alopecuroides</i> (mg/L)					
	0	0.50	1.00	1.50	2.00	2.25
Temp.	26.71±1.92 ^a	26.66±0.63 ^a	25.75±1.30 ^a	26.00±7.42 ^a	26.61±3.16 ^a	27.00±2.01 ^{ab}
DO	4.7±1.37 ^a	4.62±1.63 ^a	4.66±0.31 ^a	4.57±0.17 ^a	4.37±3.11 ^a	4.55±2.13 ^a
pH	7.12±1.21	7.42±0.17	6.76±1.62	6.71±0.00	6.93±0.81	7.33±1.62
Tol. Alk	18.3±2.32	17.10±1.76	17.31±0.42	17.00±2.77	17.42±2.06	18.00±3.43
Tol. Hardness	17.77±1.44	17.00±2.06	16.89±0.59	17.01±1.0	16.88±1.7	16.96±0.97

Temp. = Temperature (°C), DO = Dissolved Oxygen (mg/L) Tol. = Total, Alk = Alkalinity (mg/L) and Hardness (mg/ L). Mean with different letters differs significantly (P < 0.05).

**Figure 1.** Effect of the interactions between concentrations of *L. alopecuroides* and time of exposure on the opercular beat frequency per minutes of *H. bidorsalis* at 96 h.**Figure 2.** Effect of the interactions between concentrations of *L. alopecuroides* and time of exposure on the tail beat frequency per minutes of *H. bidorsalis* at 96 h.

About 60% mortality was recorded for 0.50 mg/L by the 96th h, while the concentration that killed 100% of exposed fish were 1.5, 2.0 and 2.25 mg/L at the 96 h, 48th h and 24th h respectively. However, a more defined

time and concentration dependent trend in mortality was obvious at 0.5 to 1.0 mg/L concentrations from the 24th to the 96th h.

ANOVA showed that various concentrations of *L.*

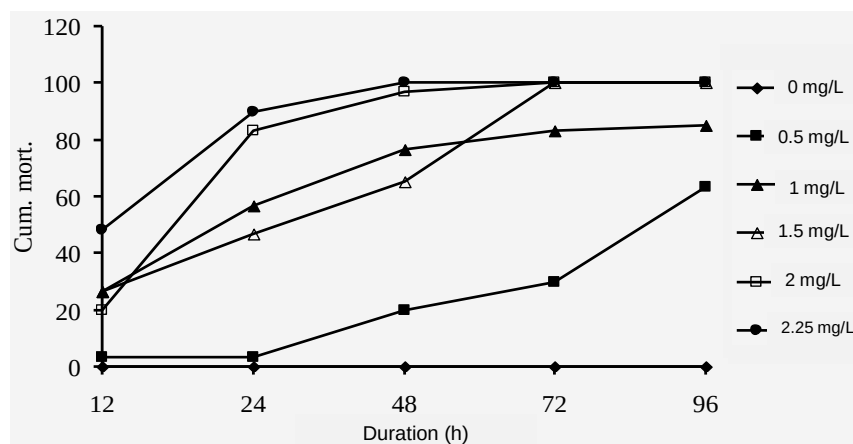


Figure 3. Effect of the interactions between concentrations of *L. alopecuroides* and time of exposure on the cumulative mortality of *H. bidorsalis* at 96 h.

Table 2. Mean squares from ANOVA of OBF/min., TBF/min. and cumulative mortality of *H. bidorsalis* fingerlings exposed to *L. alopecuroides* for 96 h.

Source of variation	Dependent variable	df	Mean square
Conc.	OBF	5	147.210 ^{ns}
	TBF	5	37.622*
	MORT	5	9480.677***
Time.	OBF	4	1821.038***
	TBF	4	193.610***
	MORT	4	3803.924***
Conc. x Time.	OBF	13	289.132**
	TBF	13	5.099 ^{ns}
	MORT	13	785.210**

TBF-tail beat frequency, OBF-opercular beat frequency, mort.-cumulative mortality (%), ns-non-significant, F-test significant level : * 0. 05,** 0.01,*** 0.001.

alopecuroides did not produce significant differences in the values of OBF ($p < 0.05$), but did in the values of TBF ($p < 0.01$) and mortality ($p < 0.001$). The time of exposure produced significant changes in OBF ($p < 0.001$), TBF, ($p < 0.001$) and mortality ($p < 0.001$). The OBF appeared to be more responsive to the treatment than TBF. Interactions between the exposure time and concentration of the toxicant did not produce any significant changes in the TBF ($p < 0.05$), while OBF ($p < 0.01$) and mortality ($p < 0.01$) were significant (Table 2). Changes in behavioural responses (obf and tbf/min) differed slightly from the control on both concentration and time of exposure however; the concentration between 1.0 to 2.25 mg/L did not show any significant difference at ($p > 0.05$) on the OBF (Figures 4 and 5). No significant differences existed in the mean Opercular beat frequency per minutes at 2.00 and 2.25 mg/L, but the former differ from those of 1.5, 0.5mg/L and control respectively. The mean value of tbf/m at 2.25 mg/L was

significantly higher at ($p < 0.05$) whereas cumulative mortality at 2.25 mg/L was not significantly different from that of 2.0 mg/L at ($p > 0.05$). The mean obf and tbf /m at 12 h were significantly higher while cumulative mortality was significantly lower in 96 h at ($p < 0.05$).

The relationship between obf/min, tbf/min and cumulative mortality on concentration and time shows a positive correlation with concentration and negative with exposure time except cumulative mortality, which is positive with time. For any increase in concentration, the opercular beat frequency is expected to increase by a constant value of 9.02 (Table 3). Opercular beat frequency is highly related to the change in time of exposure ($r^2 = 0.91$) and accounted for 95% of the total variability in the data ($r^2 = 0.95$). This indicates that it was more variables than tbf/m ($r^2 = 0.38$) that accounted for only 62% variability.

The lethal effects of *L. alopecuroides* on *H. bidorsalis* fingerlings expressed as LC₅, LC₅₀ and LC₉₅ respectively

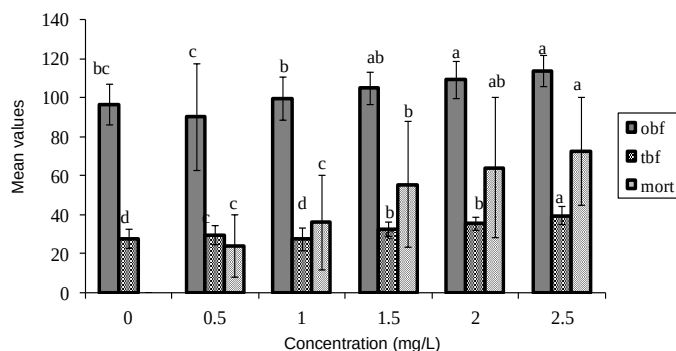


Figure 4. The mean values of behavioural (obf and tbf/min) and mortality of *H. bidorsalis* fingerlings on the various concentrations of *L. alopecuroides*. Bars represent mean $\pm$ S.D. Mean with different letters differs significantly ($p < 0.05$; Tukeys honest-significant different test).

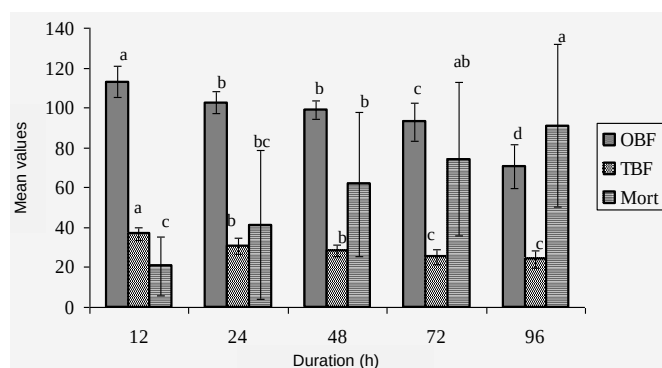


Figure 5. The mean values of behavioural (obf and tbf/min) and mortality of *H. bidorsalis* fingerlings on the duration of exposure to *L. alopecuroides* for 96 h. Bars represent mean $\pm$ S.D. Mean with different letters differs significantly ($p < 0.05$; Tukeys honest-significant different test).

for 24, 48, 72 and 96 h LC_{50} and associated 95% confidence limits of the toxicant concentrations shown in Table 4, indicated that the range of the values between the 24 h LC_{50} (1.15 mg/L) and 96hr LC_{50} (0.59 mg/L) were narrow. Safe concentrations at LC_{50} of *L. alopecuroides* exposed to *H. bidorsalis* fingerlings were also low (0.12 mg/L for 24 h and 0.06 mg/L for 96 h). For any increase in the values of concentrations at the 24 and 96 h of exposure, percentage mortality is expected to increase by a constant amount of 1.62 and 3.71 respectively. The toxicity factor (T.F) of the various concentrations increased with time of exposure from 24 to 96 h as indicated by the observed decrease in LC_{50} value. At 24 h, the LC_{50} value was 1.95 times greater than the 96 h LC_{50} value as shown in Table 5.

The Mean lethal (MLT) effects of the bioassay tests with various concentrations of *L. alopecuroides* on *H. bidorsalis* fingerlings were expressed as MLT_{50} and

MLT_{95} (the time required to kill 50 and 95% of the fish) respectively (Table 5). The time it took for half of the exposed fish to die at the various exposure concentrations decreased with time with the lowest concentration (0.5 mg/L) killing half of the exposed fish about 4.4 times greater than the time it took for 2.25 mg/L of the toxicant. The 2.0 and 2.25 mg/L concentration had the same MLT. For any increase in time of exposure at the concentrations of 0.5 and 2.25 mg/L, percentage mortality is expected to increase by constant values of 0.026 and 0.035 respectively.

The lethal concentration and MLT values appeared to have an inverse relationship with concentration and time. The LC_{50} value decreased from 1.15mg/L (24 h) to > 0.59mg/L (96 h), while the mean lethal time (MLT_{50}) values decreased with increase in concentration of the toxicant. The least concentration (0.5mg/L) had the highest time of 83.57(48.95-76.00) h while the highest concentration (2.25mg/L) had 19.05 h.

DISCUSSION

The results of the water quality of the media used in the present study are within the range reported by Viveen et al. (1985) as optimal requirement for African catfishes and did not vary significantly ($p < 0.05$) from those of the control. All were within the tolerance ranges of warm water fish species (Boyd, 1979; Adeniji and Ovie, 1989). This suggested that the parameters did not seem to alter the toxicity of *L. alopecuroides* to test fish; hence, it might not have contributed to the observed behavioural changes and the mortality of the test fish species in this study. This agrees with the work of Onusiriuka and Ufodike (1994) who exposed *C. gariepinus* to Akee apple and sausage plant extracts and reported no significant difference ($p < 0.05$) in the water quality parameters analyzed.

The study revealed that exposed fish showed stress responses such as increase opercula rate, erratic swimming, gasping for air and general hypersensitivity, which is demonstrated to be a sensitive indicator of physiological stress in fish subjected to lethal and sub-lethal concentration of pollutants (Ayoola, 2008). Omitoyin et al. (1999) reported similar observation in *Sarotherodon galilaeus* (Tilapia) and *Clarias gariepinus* fingerlings exposed to piscicidal plant extracts of *Tetrapleura tetraptera*; and Omitoyin et al. (2006) for *Clarias gariepinus* fingerlings exposed to Lindane. Fafioye (2001) also reported similar changes in fish exposed to *Parkia bioglobosa* and *Raffia vinifera*; and Obomanu et al. (2007) on Mudskipper exposed to *Lepidagathis alopecuroides*. Behavioural responses of fish to most toxicants and differences in reaction times have been observed to be due to the effect of chemicals, their concentrations, species, size and specific environmental conditions. Reed et al. (1967) reported that

Table 3. Regression lines for the prediction of the values of OBF/min., TBF/min. and cumulative mortality of *H. bidorsalis* exposed to *L. alopecuroides* for 96 h.

Independent variable	Dependent variable	Regression equation	(r ²)	(r)	Significance level
Time	OBF	y=117.61-0.44x	0.90	0.95	***
Time.	TBF	y=34.21-0.08x	0.38	0.62	ns
Time	Mort.	y=16.78+ 0.33x	0.42	0.65	*
Conc.	OBF	y=91.64 +9.02x	0.82	0.91	***
Conc.	TBF.	y=26.12+5.00x	0.81	0.90	**
Conc.	Mort.	y=10.38+26.30x	0.70	0.78	**

Where y= independent variable (concentration, Time), x= dependent variable (OBFmin⁻¹, TBFmin⁻¹ and mortality. F- test significance level: * 0.05, ** 0.01, *** 0.001 and ****0.0001, r² = coefficient of determination, r= coefficient of correlation.

Table 4. Relative toxicity of *L. alopecuroides* on *H. bidorsalis* fingerlings for 96 h.

Exposure time(h)	Lethal conc. and associated 95% C.L			Safe conc.	D/f	Probit equation	T.F	S.L.
	LC ₅	LC ₅₀	LC ₉₅					
24	0.147 (-4.27-0.76)	1.15 (0.10-1.67)	2.17 (1.94-7.30)	0.12	3	Y=-1.87+1.62x	1	*
48	-0.41 (0.00-0.00)	0.88 (0.00-0.00)	2.16(0.00-0.00)	0.09	3	Y=-1.12+1.28x	1.32	*
72	-0.34 (0.00-0.00)	0.66 (0.00-0.00)	1.64(0.00-0.00)	0.07	3	Y=-1.09+ 1.66x	1.76	*
96	0.15(-0.26-0.33)	0.59 (0.46-0.70)	1.04 (0.89-1.36)	0.06	3	Y=-2.20+ 3.71x	1.95	*

C.L= Confidence Limit, D/F = Degree of freedom, LC= Lethal Concentration, S.L = Significant level, T.F= Toxicity factor =LC₅₀ value at 24h ÷ LC₅₀ value of any other periods.

Table 5. Mean lethal time and associated 95% confidence limit of *H. bidorsalis* exposed to *L. alopecuroides* for 96 h.

Conc. (mg/L)	MLT and associated 95% C.L		D/f	Probit equation	S.L	R.T
	MLT ₅₀	MLT ₉₅				
0.5	83.57(48.95-76)	146.87(103.99-59.76)	4	Y=-2.172+0.026x	***	1
1.0	25.96 (11.27-36.08)	98.30 (70.31-102.01)	4	Y=-0.590+0.023x	ns	3.22
1.5	26.02 (11.46-35.98)	96.24 (79.60-128.74)	3	Y=-0.609+0.023x	ns	3.21
2.0	19.049 (0.000-0.00)	65.61 (0.00-0.00)	3	Y=-0.673+0.035x	**	4.39
2.25	19.049 (0.000-0.00)	65.61 (0.00-0.00)	3	Y=-0.673+0.035x	**	4.39

C.L= Confidence Limit, D/F = Degree of freedom, S.L =Significance Level. MLT=mean lethal time, R.T=Relative Time=MLT₅₀values at 0.5 mg/L ÷ MLT₅₀ value at any other concentrations.

plants poison, affect the gills of fish such that they cannot breathe. The increased mucous secretion in fish seen after treatment is a defense response

by which they try to reduce entrance of the toxicant through their body surface. The periods of motionlessness of the exposed fish may be due to

loss of muscular contraction due to the interference of the poison with the normal functioning of the nervous system and

consequently the coordination of muscular activities (Gbem et al., 1990). The trend in the OBF of this study and several others shows that, it usually peaks and then falls with time for the various concentrations of the toxicants tested (Agwuigwo, 1998; Gabriel and Okey, 2009). Increased opercular rate might result from decreased efficiency in oxygen uptake, transport or increased metabolic rate.

The response pattern of tail beat frequency (TBF) of exposed fish with respect to time and concentration of the toxicant in this study agrees with the findings of Koffi (2005) who studied the acute effects of 2, 4-D, an organophosphate, on *C. gariepinus* and hybrid. But it differed from the findings of Ekweozor et al. (2001), Gabriel and Kparobo (2003) and Onusiriuka and Ufodike (1994) on *C. gariepinus*, *O. niloticus*, *H. bidorsalis* and catfish hybrid exposed to fertilizer effluents, gramoxone, the bark and floral part of Akee apple respectively.

The stressful behaviour exhibited by the fish may be as a result of respiratory impairment due to the effect of rotenone and alkaloid on the gills and general metabolism of the exposed fish. These abnormal behavioural responses in fish exposed to *L. alopecuroides* were similar to that earlier reported by Ufodike and Omoregie (1994); Onusiriuka and Ufodike (1998); Oti (2002); Koffi (2005) and Obomanu et al. (2007) in fish exposed to various toxicants.

These were in a bid to avoid the toxicants. Exposure to increasing concentrations of toxicant was earlier observed by Dutta et al. (1994) and Tiwari and Singh (2004) to cause fatigue due to utilization of the energy substances (carbohydrate, protein and lipid). Similar observations were made in *C. gariepinus* exposed to acute concentrations of cypermethrin and 2,4-D exposed to *C. gariepinus* and hybrid post-fingerlings (Gabriel and Kparobo, 2003; Koffi, 2005). The significant negative correlation between LCs values and exposure periods that is, LC₅₀ value decreased from 1.15mg/L (24 h) to > 0.59mg/L (96 h) agrees with the work of Tiwari and Singh (2003) when they exposed *Channa punctatus* to leaf extracts of *Nerium indicum*.

The 96 h LC₅₀ for *H. bidorsalis* exposed to *L. alopecuroides* was found to be 0.59mg/L and the safe concentrations of the toxicant to the fish species was 0.06mg/L much lower than those reported by Onusiriuka and Ufodike (2000); Nwanna et al. (2000); Oti (2002); Omoniyi et al. (2002), when they exposed African catfish fingerlings to various toxicants. Olaifa et al. (2004) and Omitoyin et al. (2006) reported a 96 h LC₅₀ of copper as 0.67mg/L and Lindane 0.38mg/L for *C. gariepinus* respectively stating that they are highly toxic. Hence, *L. alopecuroides* is highly toxic to *H. bidorsalis*. Obomanu et al. (2006) reported that 0.22mg/L of *L. alopecuroides* produce 100% mortality for mosquito larvae and Suleiman (2005) reported same percentage for tadpoles within 24 h of exposure. Tadpoles and adult amphibians

are major competitors and predators in fishponds (Nguenga et al., 2000; Tiwari and Singh, 2003), hence the plant can be used for their control in ponds.

The 96 h LC₅₀ and safe concentration values of the plant *L. alopecuroides* recorded for the catfish suggest that it is highly toxic. The 96 h MLT₅₀ value for *H. bidorsalis* at 2.25 mg/L was 17.25 h. The inverse relationship between MLT and lethal concentrations, the toxicant concentrations and length of exposure time for *H. bidorsalis*, indicate that the survival time for the fish declined with increase in the concentration of the toxicant and time of exposure. The increased mortality rate recorded in this study was due to the impairment of normal metabolism by the inhibitory components in the toxicants.

This study revealed that *L. alopecuroides* exerts piscicidal property and are highly toxic to *H. bidorsalis*. It may act as respiratory poison affecting the gills, impairing respiration, various abnormal behaviours and eventually death. Increased opercular rate might result from decreased efficiency in oxygen uptake, transport or increased metabolic rate. These effects on the fish are directly proportional to the concentrations of the toxicant. The use of this toxicant in aquatic environment needs proper control to avoid reduction in fish production and non-target aquatic fauna. More study on the effect of these plants to tissues, is necessary to ascertain the extent of damage to the tested fish.

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