

Full Length Research Paper

Lambda-Cyhalothrin induced hepato-nephro toxicity potentials and post treatment recovery in *Clarias gariepinus*

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This evaluates the 28-day toxicity and 7-day post treatment effect of LCT on the behaviour, liver and kidney of *Clarias gariepinus*. Prior to the experiment, fishes were acclimatized for two weeks. 120 fishes of standard length (SL) / weight (W) 10-12 cm, 8 - 17 g were used for median lethal concentration (LC₅₀) test and 120 fishes of SL / W 16 - 40 cm, 200 - 250 g were used for the behavioural, hepato-nephrotoxicity and 7-day post treatment tests. The behavioural response of *C. gariepinus* upon exposure to LCT was observed from 24 to 96 h. The experiment had four treatments with LCT concentrations of 0.00, 2.5×10^{-4} µg/L, 5.0×10^{-4} µg/L and 6.25×10^{-4} µg/L and 30 fishes per treatment in triplicates for 28 days. In days 1, 7, 14, 21 and 28 of treatment and 7 days after treatment, fishes were brought out for blood samples collected through caudal alteration for liver and kidney marker enzymes tests (alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), creatinine and urea) using standard methods. There was a concentration dependent increase in faster swimming movement, hyperactivity, jerky movement, gulping of air, repeated closing and opening of the mouth and percentage mortality of *C. gariepinus* exposed to LCT. ALT, AST, ALP, creatinine (CR) and urea levels showed concentration and duration significant increased ($p < 0.05$) while total protein significantly decreased ($p < 0.05$) compared with controls. After 7 days of depuration, ALT, AST, CR and total protein were not different from the control. This study has demonstrated that LCT caused hepato-nephrotoxicity in *C. gariepinus*. The severity of LCT hepato-nephro in *C. gariepinus* toxicity was evident in this studies because ALP and urea levels did not return to normal after 7 days of depuration.

Key words: Liver enzymes, kidney enzymes, toxicity, lambda-cyhalothrin, behavioural responses, *Clarias gariepinus*.

INTRODUCTION

Agrochemicals especially pesticides are indirectly consumed by human via food chain. Man's pursuit to meet up with the increase in food demand has led to various technologies and production of synthetic

chemical used for eliminating superfluous pests and scheming disease vectors. Consequently, there are notable cases of adverse environmental effects on the non-targeted organisms mainly in aquatic ecosystem

(Murthy et al., 2013).

Lambda cyhalothrin (LCT) is a synthetic pyrethroid (an insecticide) used for the eradication of several insects at home and agricultural fields (Mergel, 2000). The LCT is normally routed into the soil through discharge of remains of materials used for packaging and storage and accidental discharge during spraying, then the aquatic ecosystem through runoff resulting from its use in agricultural field (De Moraes et al., 2013). The major risk of synthetic pesticide is environmental contamination especially the natural water systems where it causes various deleterious effect in aquatic resources (fish) and ultimately in human. Researches have proven that fresh water fish diversity is threatened by a number of environmental stressors such as contaminants and nutrient loading, habitat degradation and climate change (Jelks et al., 2008). Accumulation of synthetic pesticides results in huge number of residues in the environment, thereby causing a substantial hazard in the environment due to its uptake and accumulation in the food chain and drinking water (Somdare, 2015). Edward et al. (2013) reported that though toxic chemicals in water may be below detectable levels when sampled, but the concentrations due to bioaccumulation found in examined fish parts were beyond tolerable levels. Ginebreda et al. (2014) established that organisms in aquatic environments are exposed to a complex mixture of chemicals including parent compounds and their transformation products which cause multiple damages in the organisms, population and ecosystem level due to effect on organ function, reproductive stages and biological diversity.

Based on the above background, the present study evaluated lambda-cyhalothrin induced hepato-nephro toxicity potentials and post treatment recovery in *Clarias gariepinus*.

MATERIALS AND METHODS

Collection of experimental fish

A total of 240 *C. gariepinus* (with standard length and weight that ranged from 16 to 40 cm and 80 to 250 g respectively) procured from Freedom Fisheries Ltd, University Market Road, Nsukka, Enugu State, Nigeria, was used for this study. They were transported to the Laboratory in aerated bags. The fish were disinfected in 0.05% potassium permanganate (KMnO₄) solution for two minutes to avoid dermal infections and later acclimatized for two weeks in plastic tanks of 300 L capacity. They were fed daily with food (Coppens commercial feed) containing 40% crude protein. The fecal matter and other waste materials were siphoned off and water changed daily to reduce ammonia content in

the water during experimentation. Dead fish was removed with forceps to avoid possible deterioration of the water quality. During acclimatization the water was changed after 48 h with well aerated tap water.

Procurement of the test compound

A commercial formulation of lambda-cyhalothrin (600 g l⁻¹), batch number 160227 marked by Amanik Agro Investment Limited Lagos, Nigeria, was purchased at Ogige Local Market Nsukka, Enugu state, Nigeria.

Physico-chemical parameters of the test water

Some physico-chemical parameters (temperature, dissolved oxygen, pH, nitrate and nitrite) of the test water were analysed following the protocol of APHA (1992).

Determination of median lethal concentration (LC₅₀)

Prior to experiment, determinations of the LC_{24-96 h} of LCT were conducted using 120 fish. Triplicate sets of 10 fish were randomly exposed to LCT at concentrations of 0.0, 5.0 x 10⁻⁴, 6.25 x 10⁻⁴, 7.5 x 10⁻⁴ and 8.75 x 10⁻⁴ µg/L derived from a range finding test using plastic tanks of 40 L capacity each. Ten L of water was poured into each tank. Another set of 10 fish (replicated three times) was simultaneously maintained with equal amount of tap water but without the test compound and considered as control. Fish were not fed throughout the experiment and toxicity of the toxicity end point was observed. Fish was physically examined daily and considered dead in the absence of respiratory movement and swimming in response to gentle touch. Dead fish were removed and mortality was recorded at intervals of 24, 48, 72 and 96 h. The LC₂₄₋₉₆ values of the insecticide for the species at 24, 48, 72 and 96 h were determined by Probit analysis (Finney, 1971).

Determination of safe levels

The safe levels of the test compound were estimated by multiplying the 96 h LC₅₀ with different application factors (AF) and were based on the methods of (Hart et al., 1948; Sprague, 1971; CWQC, 1972; NAS/NAE, 1973; IJC, 1977; CCREM, 1991).

Experimental design for sublethal exposure

The experiment consisted of four treatments of 0.00, 2.5 x 10⁻⁴ µg/L, 5.0 x 10⁻⁴ µg/L and 6.25 x 10⁻⁴ µg/L (A-D) in replicate. Each tank contained 10 L dechlorinated tap water with 10 fishes in each of the tank. The exposure period was 28 days during which the fish were fed with small quantity of feed approximately 1% of total body weight an hour before the test solution was renewed daily. On each sampling day, (7, 14, 21 and 28), three to five fish from each of the treatment group including the control were sacrificed after anesthetizing with tricaine methanesulfonate (MS 222) to minimize stress. Blood samples were collected (through caudal alteration)

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for liver and kidney marker enzymes test. After the end of the sublethal exposure, the fish in each of the concentrations were withdrawn from the exposure of the chemical and were placed in chemical-free water after which further observation was made after 7 days of the withdrawal.

Behavioural responses

Some behavioural responses (hyperactivity, swimming patterns, fin movement, buccal cavity and gills) of *C. gariepinus* upon exposure to different concentrations of LCT were observed from 24 h to 96 h of the exposure.

Determination of liver marker and kidney marker enzymes

Liver marker enzymes AST, ALP and ALT levels were determined using the standard method described by (Reitman and Frankel, 1957). The total protein and albumin levels were determined using the Biuret method as described by (Sood, 2006). Determination of kidney markers enzymes - blood urea (BUN) and creatinine (CR) levels were determined according to the method of (Bartels and Bohmer, 1972).

Statistical analysis

Data was analysed using Statistical Packages for Social Sciences (SPSS) 20.0 (IBM Corp, Armonk, USA) and Statplus v 5.9.8 (AnalystSoft Inc., Walnut, Canada). Probit regression analysis using the Finney Method (lognormal distribution) was for lethal concentration (LC). Two-way Analysis of Variance (ANOVA) was used to compare concentration of LCT and duration of exposure dependent effects. The means were separated using DMRT (Duncan Multiple Range Test). Level of significance was set at $p < 0.05$.

RESULTS

Physico-chemical parameters of the water used for the experiment

The physico-chemical parameters of the water used for the experiment at different concentration level of LCT are shown in Table 1. The pH level and dissolved oxygen of test water after exposure to different concentrations of LCT showed no difference from each other. The pH level and dissolved oxygen observed were all concentration-dependent as indicated in Table 1 while NO_2 and NO_3 were not present in the water used for the experiment as the values recorded were 0.0 both in control and treatment groups.

Behavioural responses of *C. gariepinus* exposed to lambda-cyhalothrin

Behavioural responses of *C. gariepinus* exposed to LCT at different concentration levels for 96 h are presented in

Table 2. In group A (control), from 24 - 96 h of exposure, no mortality and behavioural changes were observed as fish exhibited normal swimming patterns, normal body and fin movements. The treatment groups B, C, D and E displayed varied behavioural abnormalities as the concentration increased. Faster swimming movement, hyperactivity, jerky movement, rapid fin and opercula movement, gulping of air, repeated closing and opening of the mouth were more severe in groups D and E which led to loss of balance and finally death (Table 2).

Median lethal concentration (LC_{50})

Cumulative mortality of fish exposed to different concentration levels of Lambda cyhalothrin

Percentage mortality of *C. gariepinus* exposed to graded concentrations of LCT at 24 h increased with increase in toxicant concentration Table 3. Fishes exposed to 6.5×10^{-4} $\mu\text{g/L}$, 7.5×10^{-4} $\mu\text{g/L}$ and 8.75×10^{-4} $\mu\text{g/L}$ had 3.3%, 23.3% and 20.0% mortality respectively while no death was recorded in 5.0×10^{-4} $\mu\text{g/L}$ and the control. The percentage mortality at 48 h did not follow concentration gradient.

The highest mortality at the 96 h was recorded in the group D exposed to 7.5×10^{-4} $\mu\text{g/L}$ toxicant concentration with 66.7% death when compared to other concentration. No absolute (100%) mortality was observed at the end of the exposure.

Lethal concentration of Lambda-cyhalothrin (95% CI) $\times 10^{-4}$ $\mu\text{g/L}$ depending on exposure time for *C. gariepinus*

The concentration at 5% lethality (LC_{50}) to LC_{99} of LCT for 24, 48, 72- and 96-h exposure of *C. gariepinus* is presented in Table 4. The LC_{50} exposure of *C. gariepinus* to LCT at 24 h exposure gave 10.7234×10^{-4} $\mu\text{g/L}$ (95% CI, 0.00091364 – 0.00211) while LC_{50} at 48 and 72 h LCT was 9.8482×10^{-4} $\mu\text{g/L}$ (95% CI, 0.00043134 – 0.0022485) and 8.2218×10^{-4} $\mu\text{g/L}$ (95% CI, 0.00050983 – 0.0013259) respectively. Finally, the value at 96 h exposure to the highest graded concentration of LCT was 8.163×10^{-4} $\mu\text{g/L}$ (95% CI, 0.00049513 - 0.0013458). The toxicant concentration in all the groups exposed to LCT decreased as time progressed. LC_{99} of LCT at 24 h was 23.0092×10^{-4} $\mu\text{g/L}$ (95% CI, 0.0014729 - 0.018895) and at 96 h was 13.8741×10^{-4} $\mu\text{g/L}$ (95% CI, 0.00024477 - 0.00786417).

Estimation of safe level for *C. gariepinus* after 96 h exposure

The safe levels of LCT were obtained using different

Table 1. The physico-chemical parameters of the experimental water exposed to different concentration levels of Lambda-cyhalothrin .

S/N	Treatment ($\mu\text{g/L}$)	Temperature ($^{\circ}\text{C}$)	pH	DO (mg/l)	NO_2 (mg/l)	NO_3 (mg/l)
1	Control	23.11	6	5.8	0	0
2	5.0×10^{-4}	23.11	6.8	5.71	0	0
3	6.25×10^{-4}	22.12	6.6	5.55	0	0
4	7.5×10^{-4}	22.12	5.7	5.35	0	0
5	8.75×10^{-4}	22.12	5.7	5.35	0	0

DO = Dissolved oxygen; NO_2 = Nitrite; NO_3 = Nitrate.

Table 2. Behavioural responses of *C. gariepinus* exposed to Lambda-cyhalothrin at different concentration levels.

Concentration ($\mu\text{g/L}$)	Swimming rate	Fin movements	Hyperactivities	Jerky movement	Equilibrium status
24 h					
Control	+++	+++	-	-	+++
5.0×10^{-4}	+++	+++	-	-	+++
6.25×10^{-4}	+++	+++	-	-	+++
7.5×10^{-4}	++	++	-	-	++
8.75×10^{-4}	+	+	-	-	+
48 h					
Control	+++	+++	-	-	+++
5.0×10^{-4}	+++	+++	-	-	+++
6.25×10^{-4}	+++	+++	-	-	+++
7.5×10^{-4}	++	++	-	-	++
8.75×10^{-4}	+	+	-	-	+
72 h					
Control	+++	+++	-	-	+++
5.0×10^{-4}	++	++	-	-	++
6.25×10^{-4}	+	+	-	-	+
7.5×10^{-4}	-	-	+++	+++	-
8.75×10^{-4}	-	-	+++	+++	-
96 h					
Control	+++	+++	-	-	+++
5.0×10^{-4}	+	+	-	-	+
6.25×10^{-4}	+	+	-	-	+
7.5×10^{-4}	-	-	+++	+++	-
8.75×10^{-4}	-	-	+++	+++	-

None = -; Mild = +; Moderate = ++; Strong = +++.

Table 3. Mortality of *Clarias gariepinus* exposed to different concentrations of Lambda-cyhalothrin.

Group	Concentration ($\mu\text{g/L}$)	Treatment size		Mortality (%)			
		(n - 30)	24 h	48 h	72 h	96 h	
A	0	30	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
B	5.0×10^{-4}	30	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	
C	6.25×10^{-4}	30	1 (3.3)	1 (3.3)	1 (3.3)	1 (3.3)	
D	7.5×10^{-4}	30	7 (23.3)	11 (36.7)	19 (63.3)	20 (66.7)	
E	8.75×10^{-4}	30	6 (20.0)	7 (23.3)	13 (43.3)	13 (43.3)	

Table 4. Lethal concentration of Lambda-cyhalothrin.

Percentile	Concentration (95% CI) x 10 ⁻⁴ µg/L			
	24 h	48 h	72 h	96 h
5	6.25(4.16 - 7.02)	5.90(2.75 - 12.65)	5.65(2.49 - 12.79)	5.61(2.35 - 13.41)
10	7.04(5.66 - 7.84)	6.61(3.83 - 11.38)	6.14(3.23 - 11.66)	6.09(3.07 - 12.11)
20	8.14(7.31 - 10.05)	7.58(4.94 - 11.64)	6.79(4.23 - 10.88)	6.74(4.06 - 11.17)
25	8.60(7.74 - 11.50)	8.00(5.06 - 12.61)	7.05(4.58 - 10.85)	7.00(4.42 - 11.10)
30	9.03(8.07 - 13.09)	8.37(5.01 - 13.96)	7.30(4.84 - 11.00)	7.24(4.68 - 11.21)
40	9.87(8.63 - 16.71)	9.10(4.71 - 17.59)	7.76(5.10 - 11.81)	7.71(4.95 - 11.99)
50	10.72(9.143 - 21.10)	9.85(4.31 - 22.49)	8.22(5.10 - 13.26)	8.16(4.95 - 13.46)
60	11.65(9.65 - 26.71)	10.65(3.89 - 29.16)	8.71(4.93 - 15.39)	8.65(4.78 - 15.66)
70	12.74(10.21 - 34.44)	11.59(3.46 - 38.85)	9.26(4.65 - 18.45)	9.20(4.49 - 18.86)
75	13.38(10.54 - 39.66)	12.15(3.23 - 45.66)	9.59(4.48 - 23.20)	9.52(4.30 - 21.05)
80	14.13(10.91 - 46.42)	12.80(2.99 - 54.73)	9.96(4.28 - 23.20)	9.89(4.09 - 23.88)
90	16.33 (11.93 - 70.32)	14.67(2.43 - 88.59)	11.01(3.75 - 32.35)	10.93(3.55 - 33.68)
95	18.40(12.84 - 99.15)	16.43(2.04 - 132.28)	11.96(3.34 - 42.86)	11.88(3.13 - 45.08)
99	23.01(14.73 - 188.95)	20.31(1.46 - 281.74)	13.97(2.66 - 73.31)	13.87(2.45 - 78.64)

CI = confidence interval.

Table 5. Estimated safe level of Lambda-cyhalothrin for *C. gariepinus* after 96 h.

Chemical	96h LC ₅₀ (µg/L)	Method	Application factor	Safe level (µg/L)
Lambda-cyhalothrin	8.163 x 10 ⁻⁴	Hart et al. (1948) *	-	3.50291 x 10 ⁻⁷
		Sprague (1971)	0.1	8.163 x 10 ⁻⁵
		CWQC (1972)	0.01	8.163 x 10 ⁻⁶
		NAS/NAE (1973)	0.01 – 0.00001	8.163 x 10 ⁻⁵ – 8.163 x 10 ⁻⁹
		CCREM (1991)	0.05	4.0815 x 10 ⁻⁵
		IJC (1977)	5% of 96h LC ₅₀	4.0815 x 10 ⁻⁵

application factors as indicated in Table 5. The calculated safe levels of LCT ranged between 8.163 x 10⁻⁵ and 8.163 x 10⁻⁹ µg/L.

Effects of Lambda-cyhalothrin on biomarkers of hepatotoxicity

The biomarkers of hepatotoxicity followed a distinct trend dependent on concentration of LCT and exposure duration. The ALT, AST and ALP levels increased significantly ($p < 0.05$) on day 28 compared to day 1 in groups exposed to the three concentrations of LCT ($p < 0.05$); in the control, there was no significant increase in these enzymes at the same duration ($p > 0.05$). ALT and AST in groups exposed to the concentrations of LCT normalised to baseline level at the end of 7-day recovery period (Table 6). The ALP level dropped significantly in groups exposed to the three concentrations of LCT at the end of 7-day recovery period compared to its level at the

end of 28 days exposure, but baseline level was not attained. The AST and ALP levels at baseline were not different (at $p < 0.05$) between all the groups (control, 2.5 x 10⁻⁴, 5.0 x 10⁻⁴ or 6.25 x 10⁻⁴ µg/L). The ALT level of all the groups exposed to LCT was not different ($p > 0.05$) from control, except 6.25 x 10⁻⁴ µg/L group which was less at $p < 0.05$. But from day 7, 14, and 21, ALT, AST and ALP concentration respectively were higher ($p < 0.05$) than control till on day 28.

Effects of Lambda-cyhalothrin on total protein

Total protein concentration was reduced with increase in concentration of LCT and duration of exposure (Figure 1). Total protein level in all the groups exposed to concentrations of LCT was significantly less than the control from day 14 to day 28 ($p < 0.05$). At the end of 7 days recovery period, total protein in all groups exposed to LCT was not different from the control.

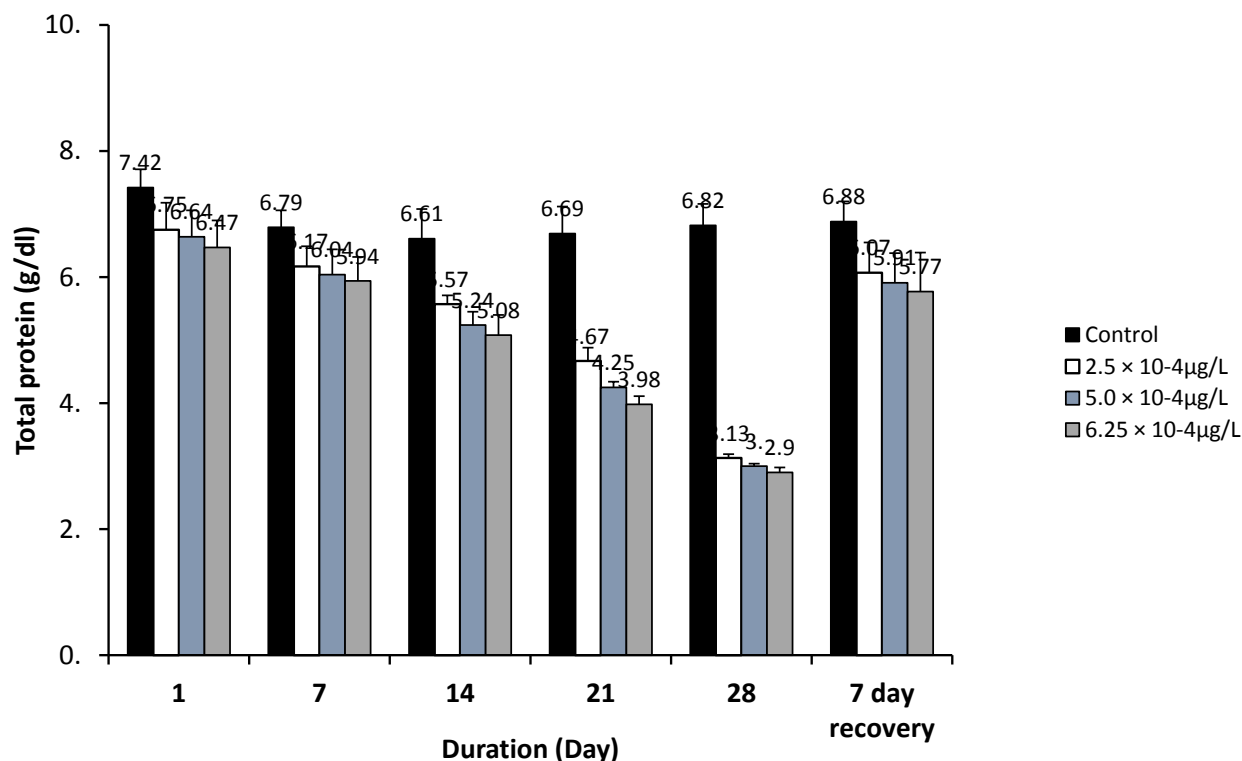


Figure 1. Variations in total protein on exposure of *Clarias gariepinus* to lambda-cyhalothrin.

Bars with different alphabet label for each group (control, 2.5×10^{-4} , 5.0×10^{-4} and $6.25 \times 10^{-4} \mu\text{g/L}$) were significantly between weeks of treatment ($p < 0.05$). Bars with different numeric superscript for each week were significantly different between treatment concentrations (control, 2.5×10^{-4} , 5.0×10^{-4} or $6.25 \times 10^{-4} \mu\text{g/L}$) ($p < 0.05$).

Effects of Lambda-cyhalothrin on biomarkers of nephrotoxicity

There was concentration and duration of exposure dependent effect of LCT on creatinine and urea concentrations. Both biomarkers of nephrotoxicity increased ($p < 0.05$) on day 28 compared to baseline level in 2.5×10^{-4} , 5.0×10^{-4} and $6.25 \times 10^{-4} \mu\text{g/L}$ LCT exposed groups ($p < 0.05$); CR and urea were significantly higher than baseline from day 21 and day 14 respectively in the treatment groups (Table 7). Concentration of CR in control had some variations which was not significant within the same duration; but urea increased significantly on day 28 compared to days 21, 14, 7 and 1 value ($p < 0.05$). Seven days post-exposure to the concentrations of LCT, CR and urea levels reduced significantly ($p < 0.05$) compared to day 28 exposure

levels ($p < 0.05$).

DISCUSSION

Variations in physicochemical parameters in water bodies' especially surface water bodies are indicative of the influence of anthropogenic activities (Nnamonu et al., 2018a). Physicochemical analyses serve as a sensitive tool for assessing the portability and vulnerability of water sourced for drinking and other domestic purposes (Nnamonu et al., 2018a). The temperature recorded in all groups was within the optimal range for fish production. This is in consonant with Keremi et al. (2014). Potential of hydrogen (pH) is a logarithmic scale for expressing the acidity or alkalinity of a solution. In water, it affects metabolism and physiological processes of fish and also exerts considerable influence on toxicity of ammonia. The pH observed (5.7- 6.0) was within desirable range and agreed with ICAR (2006). The DO values (5.35- 5.80 mg/L) recorded in this study align with Edward et al. (2013). Nitrite (NO_2) and nitrate (NO_3) are introduced into the water bodies through run off waters. They are also introduced into ponds through dead phytoplankton, uneaten feeds, dead and decaying organic matter Keremi et al., 2014). The findings were in agreement and within

Table 6. Variations in Mean \pm SE of selected biomarkers of hepatotoxicity on exposure of *Clarias gariepinus* to lambda-cyhalothrin.

Parameter	Concentration ($\mu\text{g/L}$)	Duration (Day)					7-day recovery
		1	7	14	21	28	
ALT (U/L)	Control	11.88 \pm 0.06 ^{a2}	11.85 \pm 0.08 ^{a2}	11.87 \pm 0.07 ^{a1}	11.61 \pm 0.32 ^{a1}	11.60 \pm 0.32 ^{a1}	11.48 \pm 0.24 ^{a1}
	2.5 $\times 10^{-4}$	11.87 \pm 0.07 ^{c2}	11.65 \pm 0.10 ^{c12}	12.80 \pm 0.29 ^{b2}	13.74 \pm 0.48 ^{a2}	13.99 \pm 0.02 ^{a2}	11.93 \pm 0.04 ^{c1}
	5.0 $\times 10^{-4}$	11.85 \pm 0.03 ^{c2}	11.57 \pm 0.11 ^{c12}	13.20 \pm 0.32 ^{b2}	13.60 \pm 0.20 ^{b2}	14.24 \pm 0.07 ^{a2}	11.82 \pm 0.07 ^{c1}
	6.25 $\times 10^{-4}$	11.53 \pm 0.16 ^{c1}	11.43 \pm 0.16 ^{c1}	13.35 \pm 0.29 ^{b2}	13.78 \pm 0.17 ^{ab2}	14.29 \pm 0.13 ^{a2}	11.76 \pm 0.07 ^{c1}
AST (U/L)	Control	11.61 \pm 0.21 ^{a1}	11.39 \pm 0.44 ^{a1}	11.79 \pm 0.18 ^{a1}	11.78 \pm 0.18 ^{a1}	11.86 \pm 0.07 ^{a1}	11.77 \pm 0.12 ^{a1}
	2.5 $\times 10^{-4}$	11.82 \pm 0.13 ^{b1}	11.20 \pm 0.42 ^{c1}	12.34 \pm 0.10 ^{b12}	13.41 \pm 0.11 ^{a2}	13.84 \pm 0.14 ^{a2}	11.88 \pm 0.05 ^{b1}
	5.0 $\times 10^{-4}$	11.11 \pm 0.58 ^{c1}	11.06 \pm 0.46 ^{c1}	12.56 \pm 0.24 ^{b12}	13.72 \pm 0.17 ^{a2}	14.08 \pm 0.12 ^{a2}	11.69 \pm 0.10 ^{bc1}
	6.25 $\times 10^{-4}$	10.81 \pm 0.81 ^{b1}	10.91 \pm 0.36 ^{b1}	13.00 \pm 0.43 ^{a2}	13.87 \pm 0.10 ^{a2}	14.16 \pm 0.15 ^{a2}	11.48 \pm 0.20 ^{b1}
ALP (U/L)	Control	49.62 \pm 5.65 ^{a1}	51.00 \pm 5.39 ^{a1}	49.61 \pm 5.41 ^{a1}	50.06 \pm 6.00 ^{a1}	50.28 \pm 6.70 ^{a1}	50.30 \pm 6.61 ^{a1}
	2.5 $\times 10^{-4}$	48.83 \pm 5.19 ^{c1}	65.83 \pm 2.96 ^{b2}	85.22 \pm 2.73 ^{a2}	89.31 \pm 3.27 ^{a2}	93.83 \pm 0.38 ^{a2}	61.49 \pm 0.59 ^{b2}
	5.0 $\times 10^{-4}$	49.50 \pm 5.09 ^{c1}	66.56 \pm 3.08 ^{b2}	87.00 \pm 3.28 ^{a2}	89.83 \pm 3.37 ^{a2}	94.52 \pm 0.10 ^{a2}	62.69 \pm 0.53 ^{b2}
	6.25 $\times 10^{-4}$	50.83 \pm 4.04 ^{c1}	67.89 \pm 2.99 ^{b2}	87.44 \pm 2.73 ^{a2}	93.56 \pm 5.18 ^{a2}	95.03 \pm 0.17 ^{a2}	63.00 \pm 0.93 ^{b2}

Values with different alphabet superscript across a row were significantly different; and values with different numeric superscript down a column were significantly different ($p < 0.05$). ALT, Alanine aminotransferase; AST, Aspartate aminotransferase; ALP, Alkaline phosphatase.

Table 7. Variations in selected biomarkers of nephrotoxicity on exposure of *C. gariepinus* to lambda-cyhalothrin.

Parameter	Concentration ($\mu\text{g/L}$)	Duration (Day)					7-day recovery
		1	7	14	21	28	
CR (mg/dl)	Control	0.52 \pm 0.08 ^{a1}	0.59 \pm 0.08 ^{a1}	0.52 \pm 0.08 ^{a1}	0.52 \pm 0.08 ^{a1}	0.44 \pm 0.13 ^{a1}	0.47 \pm 0.03 ^{a1}
	2.5 $\times 10^{-4}$	0.82 \pm 0.07 ^{c2}	0.59 \pm 0.08 ^{cd1}	0.74 \pm 0.07 ^{cd2}	1.24 \pm 0.07 ^{b2}	1.62 \pm 0.06 ^{a2}	0.52 \pm 0.08 ^{d1}
	5.0 $\times 10^{-4}$	0.82 \pm 0.07 ^{c2}	0.74 \pm 0.07 ^{c1}	0.89 \pm 0.00 ^{c23}	1.42 \pm 0.07 ^{b23}	1.84 \pm 0.06 ^{a23}	0.67 \pm 0.13 ^{c12}
	6.25 $\times 10^{-4}$	0.89 \pm 0.00 ^{c2}	0.82 \pm 0.07 ^{c1}	0.96 \pm 0.07 ^{c3}	1.59 \pm 0.10 ^{b3}	2.00 \pm 0.13 ^{a3}	0.82 \pm 0.07 ^{c2}
Urea (mg/dl)	Control	24.17 \pm 0.90 ^{b1}	24.60 \pm 0.89 ^{b1}	25.71 \pm 1.02 ^{b1}	24.04 \pm 1.16 ^{b1}	37.29 \pm 2.13 ^{a1}	36.92 \pm 1.70 ^{a1}
	2.5 $\times 10^{-4}$	26.14 \pm 1.13 ^{c12}	26.70 \pm 1.66 ^{c1}	36.31 \pm 4.13 ^{b2}	51.38 \pm 1.34 ^{a2}	54.49 \pm 0.66 ^{a2}	39.39 \pm 0.62 ^{b12}
	5.0 $\times 10^{-4}$	26.87 \pm 0.89 ^{c12}	27.51 \pm 1.94 ^{c1}	40.21 \pm 2.11 ^{b2}	52.59 \pm 1.61 ^{a2}	56.92 \pm 0.96 ^{a2}	41.23 \pm 0.49 ^{b2}
	6.25 $\times 10^{-4}$	27.73 \pm 1.09 ^{c2}	29.06 \pm 1.96 ^{c1}	41.48 \pm 1.92 ^{b2}	53.34 \pm 1.67 ^{a2}	57.62 \pm 1.14 ^{a2}	42.22 \pm 0.55 ^{b2}

Values as mean \pm S.E. For each parameter, values with different alphabet superscript across a row were significantly different; and values with different numeric superscript down a column were significantly different ($p < 0.05$).

desirable range of 0.0 to 10.00 as reported by WHO (2010). The behavioural alteration and loss of equilibrium exhibited by the *C. gariepinus* exposed to different levels of LCT is an indication that the region of the brain which is associated with the maintenance of (equilibrium must have been affected by LCT exposure. The supports Odo et al. (2016). This would result in prolonged neuromuscular depolarisation, culminating in the observed uncoordinated and jerky movement that was noticed in *C. gariepinus* exposed to LCT (Sarai et al., 2013). We also observed repeated opening and closing of the mouth and operculum covering accompanied by partially extended fin. These behavioural changes are caused by hypoxic conditions which hampers oxygen uptake in fish. Hypoxic conditions arise primarily due to damage of gills of fish exposed to insecticides. These reports were consistent with (Somdare, 2015).

There was increased mucus secretion by the experimental animals which could be an adaptive response to counter the irritating effects of the insecticides on body surface and mucus membrane. This is in agreement with the report of Odo et al. (2016). The observed behavioural changes as demonstrated in our study might have affected swimming behaviour, feeding activities, predation, competition and reproduction.

This study has demonstrated that mortality of *C. gariepinus* exposed to LCT was concentration and exposure duration dependent. By implication, LCT is highly toxic to fish and other aquatic animals. This report is in line with Taofeek et al. (2013). Remarkably, the LC₅₀ values of the present study decreased as the exposure time increased from 24 to 96 h due to effects of toxicant.

The variation in the safe level as demonstrated in this study showed that differences obtained were all dependent on concentration and duration to LCT exposure.

The increase in AST, ALP and ALT agrees with Marzouk et al. (2012) while the increase in ALP disagrees with Bhushan et al. (2010). These enzymes are secreted in to blood in hepatocellular injury and their levels increase. The enhanced activities of transaminases ALP, AST and ALT revealed the hepatic damage / degeneration in LCT- treated group. These increases may be mainly due to the leakage of these enzymes from the liver cytosol into the bloodstream (Odo et al., 2016; Nnamonu et al., 2018b).

The increase of soluble liver enzymes in blood serum may be useful as an indicator of hepatic dysfunction and hepatocellular damage (Sloss, 2009). The significant increase in creatinine and urea agrees with Donadio et al. (1997). The increased plasma creatinine and urea levels reflect the diagnosis of renal failure (Donadio et al., 1997). Moreover, elevated blood urea is known to be correlated with an increased protein catabolism in mammals and/or the conversion of ammonia to urea as a result of increased synthesis of arginase enzyme involved

in urea production. The significant increase ($p < 0.05$) in urea and creatinine levels depicts renal injury in the LCT-treated fish.

Conclusion

The high mortality rate of *Clarias gariepinus* exposed to LCT, significant elevations in liver and kidney marker enzymes confirm the severity of LCT toxicity. The severity of LCT hepato-nephro in *C. gariepinus* is so evident in our studies because ALP and urea levels did not return to normal after 7 days of depuration.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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