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A Study on the Effects of Glyphosate Toxicity on the Behaviour, Haematology, and Histopathology of Stinging Catfish, *Heteropneustes fossilis* (Bloch), of Assam, India

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Abstract

A variety of agrochemicals, such as the herbicide glyphosate N-(phosphonomethyl)-glycine, can adversely affect fish. The purpose of this study was to assess the effect of glyphosate on the behavioural, haematological, and histological parameters of *Heteropneustes fossilis,* a stinging catfish. The fish displayed abnormal behaviours during exposure, including increased movement of gill covers, jumping, breathing air, difficulty in movement, and changes in skin colour. The results were statistically significant ($p< 0.05$) and histological examination revealed enlargement of hepatocytes with pyknotic nuclei, extensive damage to the chloride cells as well as hypertrophy and fusion of secondary lamellae of gill. There was prominent deterioration to the columnar epithelial cells of the intestines, vacuolation and rupture into the lamina propria's connective tissue, as well as the brush border epithelium's elimination in contrast to the control. Therefore, it was suggested that the farmer should use a concentration that is suitable and won't harm non target organisms. As an alternative, the biological way of controlling weeds needs to be preferred over the chemical method.

Keywords: Glyphosate, Haematology, Herbicide, *Heteropneustes fossilis,* Histopathology

1. Introduction

Herbicides are frequently used globally to reduce the detrimental effects that weeds and pests have on fish farms and agricultural productivity[1,2]. Herbicides find their way into different aquatic ecosystems from the site of application and become highly toxic to non-target organisms, especially fishes[2]. Pesticides and herbicides at high concentrations are known to reduce the survival, growth and reproduction of fish, and produce many visible effects on fish[3]. Aquatic organisms physiology and ability to survive are impacted by herbicides, but they also interact with their genetic makeup and can cause mutations or carcinogenesis. Glyphosate, a prominent broad-spectrum herbicide in agriculture, eliminates a variety of grasses, sedges, broad-leaved weeds, and woody shrubs that are annual, biennial, and perennial, as well as aquatic weeds[4]. The use of glyphosate is widespread in weed control because it primarily targets annual broad-leaved plants and

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competing grasses. As an all-purpose herbicide, glyphosate has immediate effects on a range of plant species, with potential consequences for endangered species, nitrogen fixation reduction, and increased susceptibility to plant diseases.

Behaviour is the outcome of genetic, physiological, and central nervous system processes that assist organisms in adapting to their environment, including responses to stress and pollution. It connects physiology and ecology, making it useful for researching pollution effects[5]. Toxicant exposure frequently reduces the performance of important fitness and survival behaviours in natural ecosystems which has been recorded in different studies[6]. Because aquatic contaminants are toxic, improper behavioural responses to environmental and physiological stimuli can have major consequences for survival[7]. Histology enables a rapid method for identifying the effects of stressors in various tissues and organs. Histological alterations arise as a medium-term response to sub-lethal stressors[8]. Histopathological biomarkers in environmental monitoring provide significant advantages by allowing specialised organ evaluation, such as the stomach and intestine, which are important for food digestion and absorption in fish. Haematology serves as an index of immunological state, allowing for precise fish diagnoses following toxicant exposure. The haematological parameters are critical biomarkers for assessing the structural and functional health of fish after exposure to pollutants[2,9]. Toxicant-induced alterations in water quality manifest in the haematological parameters of aquatic organisms. Fish is a highly sensitive bio-indicator of aquatic contamination as an ecotoxicological study model[9] and haematology study, particularly when exposed to toxicants, crucial in assessing the structural and functional well-being of internal organs[6]. Due to its susceptibility to contaminants, the *H. fossilis*, an air-sac catfish commonly known as the stinging catfish, serves as a valuable model for toxicity studies[10,11]. Determining the toxicity of glyphosate on *H. fossilis* is the primary objective of the present research. Despite the manufacturer's claims that glyphosate is a low-toxicity, environmentally friendly pesticide since it is widely used and is suspected to affect non-target organisms; this study aims to thoroughly assess the impact it has. Specifically, it will focus on the aquatic environment in Assam, where paddy fields are frequently washed away by flooding and adjacent water bodies.

2. Materials and Methods

2.1 Herbicide

Glyphosate, available in various solid and liquid forms including acids and salts, was utilized in this study through a commercial formulation (Glyphosphate 41% SL) recognized by its trade name (Genki) in the Indian market[12].

2.2 Experimental Design and Acute Toxicity

From a neighbourhood market in Nagaon district of Assam, male and female specimens of the chosen species measuring an average length of 12.5±1.5 cm and weight of 15.5±1 gram were gathered (Latitude: 26.34430567, Longitude: 92.70993218). Before the experiment, fishes were collected in the month of February and acclimatized in the laboratory at room temperaturefor three days. Fish were put in 20-litre water containers and divided into six groups. The bubble diffuser with a circular airstone disc offered enough aeration. The acute toxicity tests were conducted following the static non-renewal technique, by the established procedures outlined by the US Environmental Protection Agency (EPA). Fish were not fed 24 hours before starting and 96 hours during the experiment. The test specimens were subjected to six separate concentration tests (0.04, 0.08, 0.16, 0.24, and 1.0 ml/l), with one serving as a control. After the determination of the LC_{50} value (0.22 ml/l), two test concentrations of glyphosate *viz.*, 1/10 of LC_{50} and 1/5 of LC_{50} were selected for the experiment. The experiment was performed in triplicates taking six fishes in each group. The fish mortalities at different test concentrations were recorded. The Organisation for Economic Cooperation and Development (OECD) and the US Environmental Protection Agency (EPA) provided reference criteria and guidelines that were used to evaluate the behavioural responses. Surface visits, jumping, air gulping, quick swimming, mucous secretion, opercular movement, colour fading, and inactivity were among the behavioural and morphological responses observed. For 10 to 15 minutes, each container was inspected to accurately assess each fish.

2.3 Assessment of Blood Parameters

Considering fish that had survived the chemical exposure, blood samples were obtained by severing the artery at the caudal region two centimeters from

the caudal peduncle, blood was drawn from three fish for each treatment. Medical syringes (2.5mL) and hypodermal needles prepared with the anticoagulant Ethylene Diamine Tetraacetic Acid (EDTA) were used for blood collection. For evaluation, several haematological parameters including haemoglobin concentration, total WBC count, and total RBC count were measured. The total count of RBC and WBC was done using a Neubauer haemocytometer and Haemoglobin content (Hb) was determined using Sahli's haemoglobinometer.

2.4 Histopathological Analysis

Fish were anaesthetized with tricaine methanesulphonate after the exposure phase (MS 222). The intended organs-the liver, gut, and gills-were excised and set aside for histological examination. After being fixed in Bouin's fluid for 24 h, the tissues were cleaned with 70% ethanol and allowed to dry, using ethanol grades. Then, the tissues were embedded in paraffin, sectioned at a thickness of 5µm, stained with haematoxylin and eosin, and checked for anomalies using an Olympus Vanox light microscope (Olympus, Tokyo, Japan)[13].

Table 1. Fish survival at various test doses and intervals of sampling

Concentration (ml/l)	24h	48h	72h	96 h		
					% Survival	% Mortality
Control	6	6	6	6	100	0
0.04	6	6	6	6	100	0
0.08	6	6	5	5	83.33	16.67
0.16	6	6	5	3	50.00	50.00
0.24	5	5	4	1	16.66	83.34
1.00	5	3	2	0	0	100

2.5 Statistical Analysis

SPSS version 21, was used to conduct the statistical analysis. All generated data are presented as means ± SD. To evaluate significance among exposure chemicals and concentrations with significance at $P<0.05$, one-way Analysis of Variance (ANOVA) was used to analyse the haematological parameters.

3. Results

3.1 Fish Mortality

Except for the control group, mortality of the fish was observed in all treated groupsas seen in Table 1 and Figure 1. The acute toxicity test results showed fish mortality at different concentrations of glyphosate and at varied times (24, 48, 72, and 96 hours).

3.2 Behavioural Response

After being exposed for 24 to 96 hours, the treated fish showed increased swimming, surfacing, and hyperactivity immediately after being exposed to the test solution. After 24 hours of exposure, there was agitation, fast surfacing, skin peeling, and colour fading while somewhat lower activity and a progressive rise in colour fading were observed after 48 hours of exposure along with a thin mucous coating over the body surface and gill adhesion was observed. After 72 hours, surfacing and gulping of air was vigorous and the fish exhibited a loss of balance and choppy swimming

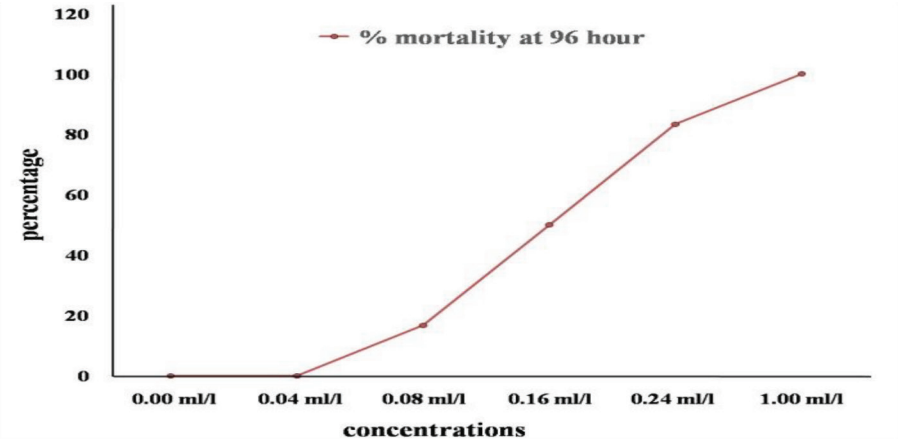


Figure 1. Percentage of mortality of *H. fossilis* at 96 hours at different test concentrations.

patterns. After 96 hours of exposure, ulcers on the trunk, caudal base, and pectoral fins were seen and almost all the test fish had a thick mucus coating covering their entire body and gills. The natural colouration of the test fish was gone and all of the test fish displayed a loss of equilibrium before dying and sinking to the bottom. Table 2 displays the behavioural responses of *H. fossilis* subjected to glyphosate doses below the fatal threshold.

3.3 Haematological Parameters of *H. fossilis* Exposed to Glyphosate

After 96 hours of exposure, the Red Blood Cell(RBC) count of the *H. fossilis* subjected to sub-lethal quantities of glyphosate (Table 3) was considerably ($P < 0.05$) lower than that of the control group. A significant decline in the mean RBC values was observed with a rise in glyphosate concentration, where the mean RBC values in the control group were $3.88\pm0.09x10^6$ mm^3 in comparison to treated groups where $3.70\pm0.07x10^6$ mm^3 in 0.022 ml/l and $3.55\pm0.06x10^6$ mm^3 in 0.044 ml/l. These results suggest dose-dependent toxicity. After 96 hours of exposure, there was a substantial ($P<0.05$) rise in the White Blood Cell (WBC) profile of the treated fish samples (Table 3), which was correlated with higher glyphosate concentrations in the different treatments. The average WBC value of the fish in the control group was $10.61 \pm 0.42x10^6$ mm^3, while the fish exposed to glyphosate at sub-lethal concentrations of 0.022 ml/l and 0.044 ml/l had average blood values of $11.50 \pm 0.50x\ 10^6$ mm^3 and $14.18 \pm 0.52x10^6$ mm^3, respectively. The fluctuations seen in the white blood cell count across the different treatments indicate that the exposed fish's elevated WBC count could be an adaptive defense mechanism reacting to acute exposure to glyphosate. Table 3 displays the mean values of Haemoglobin (Hb) content, which also indicate a substantial ($P<0.05$) dose-dependent decrease. The fish of the control group have a mean haemoglobin level of 28.41 ± 0.42 g/dl. The fish exposed to glyphosate at sub-lethal concentrations of 0.022 ml/l and 0.044 ml/l had mean haemoglobin values of 7.13 ± 0.29 g/dl and 7.02 ± 0.17 g/dl, respectively.The results demonstrated that the fish's haemoglobin concentration decreased with an increase in concentration following the exposure period in reaction to the acute glyphosate concentrations, which may be regarded as a biomarker of distress.

3.4 Histopathological Study

In histopathological studies, it wasfound that the chemical has deleterious effects on different internal organs.

Liver: The existence of polygonal hepatocytes with centrally positioned nuclei, Kupffer cells, and sinusoids was identified in the liver histology of the control fish (Figure 2 (A)). The most noticeable histopathological alterations caused by glyphosate toxicity in the liver of H. fossilis were enlarged and pyknotic hepatocytes, vacuolation in the cytoplasm, a distorted polygonal shape of the hepatocyte, hepatocytic inflammation, and the appearance of sinusoidal space in the treated fish of both sub-lethal concentrations (Figure 2 (B), 2 (C)).

Gills: Gills histology of the controlled fish revealed lamellae (primary and secondary) with cartilaginous skeletal framework, multi-layered epithelium, and a vascular system. The chloride cells of the primary epithelium were supported by closely packed pavement cells (Figure 3A). While both sub-lethal dosages of glyphosate caused secondary lamellae to fuse and

Table3. Impact of glyphosate at sub-lethal dosages on *H. fossilis*haematological parameters following 96 hours of exposure

Concentration (ml/l)	RBC ($x10^6$ mm^3)	WBC ($x10^6$ mm^3)	Hb(g/dl)
0.000 (Control)	3.88±0.09	10.61±0.42	28.41±0.42
0.022	3.70±0.07	11.50±0.50	7.13±0.29
0.044	3.55±0.06	14.18±0.52	7.02±0.17

Values are given as mean± standard deviation. Column indicates variation in the mean values that are significant at $p< 0.05$. RBC= Red Blood Cell; WBC= White Blood Cell; Hb= Haemoglobin.

Table 2. Behaviour responses of glyphosate exposed *H. fossilis*at control and two sub-lethal concentrations

Concentration ml/l	Surface Visit	Jumping	Air Gulping	Fast Swimming	Mucous Secretion	Opercula Movement	Colour Fading
0.00 (control)	+	-	+	+	-	+	-
0.022	++	+	+	++	+	++	++
0.044	++	++	++	++	++	+++	+++

-(none); + (mild); ++ (moderate); +++ (rapid, strong)

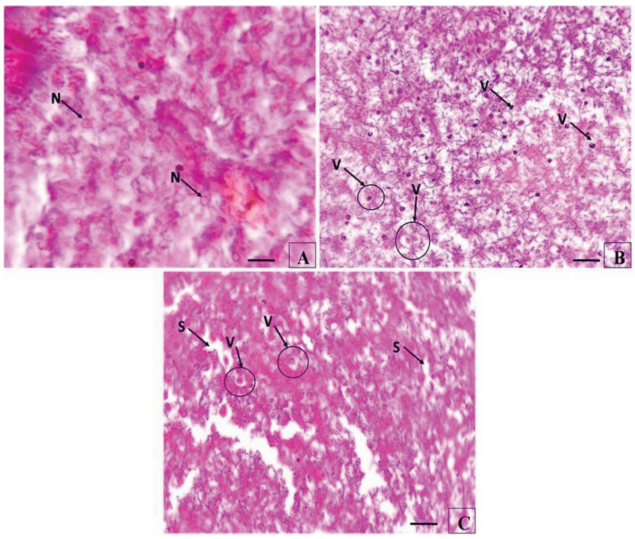


Figure 2. Photomicrographs of histology of liver of *H. fossilis* of both control and treated groups under light microscopy. **(A)** Normal appearance of hepatocytes (arrow) with distinct nucleus (N). **(B)** Treated with 0.022 ml/l showing vacuolation (V) with distorted polygonal shape of hepatocytes(Y) and appearance of sinusoidal space (oval). **(C)** Treated with 0.044 ml/l showing vacuolation (V) and appearance of sinusoidal space (S).

hypertrophise, chloride cells were severely damaged, and some lamellae were seen to be partially fused. After exposure to glyphosate, certain stratified epithelial cells in the gills completely lost all their micro ridges, while others showed enlarged and irregularly arranged. The treated fish displayed gill epithelium with deteriorating chloride cell alterations (Figure 3(B), 3(C)).

Intestine: Four histological layers, the mucosa, submucosa, muscularis, and athin serosa were seen in the control gut. The mucosa consists of simple, long, finger-like villi that are supported by columnar epithelial cells that absorb substances and have nuclei either in the centre or at the base of the cell. The villi are embraced by a narrow top plate with brush borders. Several mucous cells sustain the intestinal mucosa (Figure 4(A)). The submucosal lamina propria is made up of loose connective tissues, while the serosa layer is made up of a dense network of blood vessels. The most obvious histopathological changes observed in both sub-lethal concentrations of treated fish in comparison to the control were the disruption and vacuolation of the lamina propria's connective tissue and columnar epithelial cells, significant harm to these cells including epithelial lifting, and the total elimination of the brush border(Figure 4(B), 4(C)).

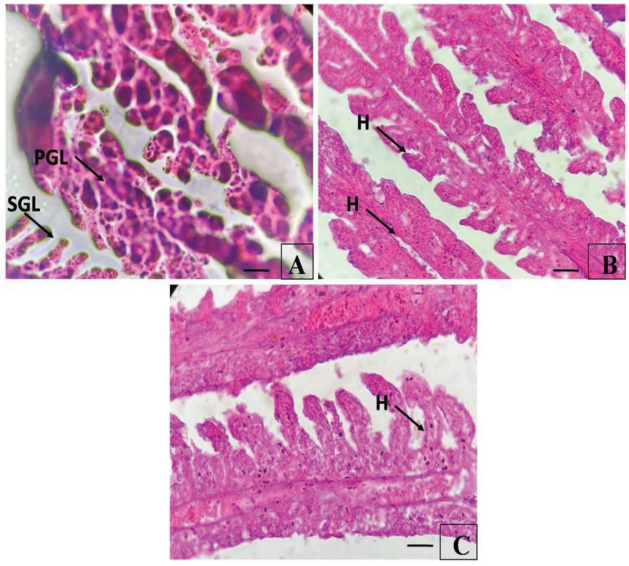


Figure 3. Photomicrographs of histology of gills of *H. fossilis* of both control and treated groups under light microscopy. **(A)** Normal appearance of Primary Gill Lamellae (PGL) and Secondary Gill Lamellae (SGL). **(B)** Treated with 0.022 ml/l showing Hypertrophy (H) and fusion of secondary lamellae (arrow). **(C)** Treated with 0.044 ml/l showing Hypertrophy (H), fusion of secondary lamellae (arrow) and distortion of chloride and pillar cells (oval).

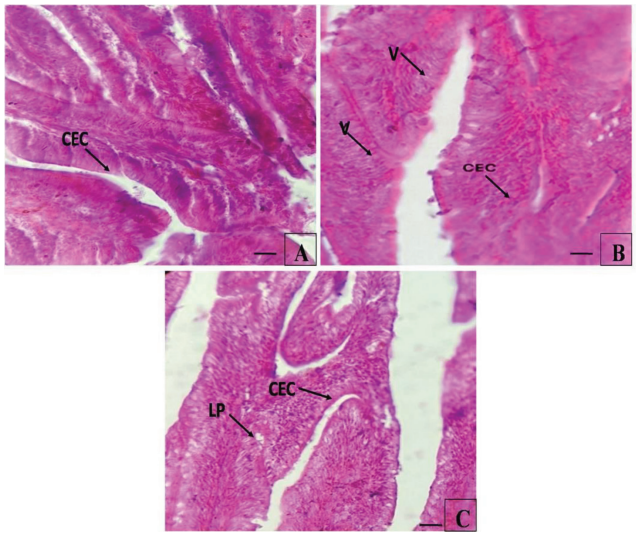


Figure 4. Photomicrographs of histology of intestine of *H. fossilis* of both control and treated groups under light microscopy. **(A)** Normal structure of CEC with prominent nucleus. **(B)** Treated with 0.022 ml/l showing disruption and Vacuolation (V) in lamina propria and damage in CEC. **(C)** Treated with 0.044 ml/l showing disruption of Lamina Propria (LP), damage in CEC (arrow) and disappearance of brush border (oval).

4. Discussion

Fish, as sensitive aquatic species, respond quickly to environmental changes. Fish perform a vital role in identifying potential threats in aquatic ecosystems and are frequently confronted with a range of environmental toxins, making them useful sentinel species in ecotoxicological research. As a result, their employment as indicator species for testing xenobiotic substances is critical[14,15].

Haematological variables, such as pesticides and heavy metals, are reliable indications of sublethal contaminant consequences in fish. According to research, when toxicants are introduced into aquatic creatures, haematological parameters change. *H. fossilis* exposed to sublethal glyphosate had lower RBC and haemoglobin levels but higher WBC levels. This increased white blood cell count in sub lethal trials could be connected to increased antibody synthesis, which would improve fish survival and recovery. Glyphosate appears to have an impact on fish immune systems, changing the synthesis of proinflammatory cytokines, phagocytic function, complement cascade, and lymphocyte responses[3,9].

Pesticide exposure produced fatty degeneration and liver necrosis in fish, most likely due to detoxifying strain and a lack of cell regeneration[6,16-18]. Since the gills are in direct touch with the water, they operate as a crucial barrier against outside influences getting into fish[19]. Waterborne agents have the potential to harm fish gills, disrupting gas exchange and causing tissue hypoxia. The gill structure of the control *H. fossilis* is like that of other freshwater catfish[20]. Sub-lethal glyphosate concentrations, on the other hand, cause visible ultrastructural changes such as hypertrophy, lamellae fusion, chloride cell damage, and micro-ridge alterations. These changes, according to Schwaiger *et al.*, may have an impact on respiratory function and overall fish health[15,16,21]. The intestine, a crucial organ, is frequently exposed to food-borne pollutants[22]. Histological analysis revealed tissue destruction, including deformation, vacuolation in the lamina propria, extensive CEC damage, and brush border loss[1]. Kumari and Kumar observed serosa, mucosa, submucosal layer degeneration, necrosis, proliferation, and villi desquamation in polluted water-derived *Channa striatus* and *H. fossilis*[23]. Braunbeck and Appelbaum discovered that endosulfan exposure caused necrosis of the intestinal epithelial lining in *Cyprinus carpio*, resulting in malabsorption[7]. CEC damage in *H. fossilis* is similar to that seen in glyphosate-exposed *Channa punctatus*[7,15,23,24].

5. Conclusion

The possibility of unintentional glyphosate discharge into aquatic environments is significant. The high concentration, prolonged exposure, and high temperature of the aquatic habitat create severely detrimental biochemical and physiological changes that increase fish mortality and impede fish production. The study's findings conclude that, despite its efficacy in weed control, glyphosate is harmful to fish when it accumulates in aquatic bodies over time. The current study effectively established glyphosate's poisonous nature and toxicity to experimental fish *H. fossilis* even at sub-lethal concentrations in behavioural, morphological, haematological, and histopathological aspects. Significant changes in the fish's behaviour were seen during the study's exposure period. Their severity varied with dose and duration. The physiological changes seen in the behavioural, haematological, and histopathological characteristics of *H. fossilis* exposed to glyphosate may have impacted the fish's overall health and condition, eventually resulting to fish mortality. Furthermore, to ensure the safe utilization of these pesticides, it becomes imperative to embark on additional experimental endeavours aimed at delineating the precise concentration and duration of exposure that averts the onset of appreciable sub lethal repercussions in fish populations.

6. Acknowledgement

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7. References

1. Samanta P, Pal S, Mukherjee AK, Ghosh AR. Pathological (histological and ultrastructural) study in stomach and intestine of *Heteropneustes fossilis* (Bloch) to excel Mera 71, a glyphosate-based herbicide. J Gastrointest Dig Syst. 2016; 6(6). https://doi.org/10.4172/2161-069x.1000479
2. George AD, Akinrotimi OA, Nwokoma UK. Haematological changes in African catfish (*Clarias gariepinus*) exposed to mixture of atrazine and metolachlor in the laboratory. J Fish Sci. 2017; 11(3):48. https://doi.org/10.21767/1307-234x.1000129
3. Annune PA, Ebele SO, Oladimeji AA. Acute toxicity of cadmium to juveniles of *Clarias gariepinus* (Teugels)

and *Oreochromis niloticus* (Trewavas). J Environ Sci Health A. 1994; 29(7):1357-65. https://doi.org/10.1080/10934529409376116
4. Ani LC, Nwamba HO, Ejilibe CO, Nwani CD. Acute toxicity of glyphosate-based herbicide glycot on juvenile african catfish *Clarias gariepinus* (Burchell 1822). J Fisheries Livest Prod. 2017; 5(3): 252. https://doi.org/10.4172/2332-2608.1000252
5. Sharma M, Thakur J, Verma S, Sharma P. Behavioural responses in effect to chemical stress in fish: A review. Int J Fish Aquat Stud. 2019; 7(1):1-5.
6. Ugah J. Toxic effects of glyphosate on the behaviour, haematology, histopathology and growth of the African catfish, *Clarias gariepinus* (burchell, 1822) juveniles (doctoral dissertation).
7. Braunbeck T, Appelbaum S. Ultrastructural alterations in the liver and intestine of carp *Cyprinus carpio* induced orally by ultra-low doses of endosulfan. Dis Aquat Organ. 1999; 36(3):183-200. https://doi.org/10.3354/dao036183
8. Edeh IC, Nsofor CI, Iheanacho SC, Ikechukwu CC, Nnachi IA, Nwankwo CG. Behavioural and haematological alterations in the African Catfish (*Clarias gariepinus*) exposed to varying concentrations of glyphosate. Asian J Fish Aquat Res. 2023; 21(1):32-42. https://doi.org/10.9734/ajfar/2023/v21i1527
9. Uchenna UB, Uka A, Obiahu OH. The impact of sub-lethal concentrations of glyphosate on growth and haematology of African catfish under aquatic ecological micro-climate. J Environ Chem Ecotoxicol. 2022; 4:164-70. https://doi.org/10.1016/j.enceco.2022.06.001
10. Akter R, Pervin MA, Jahan H, Rakhi SF, Reza AM, Hossain Z. Toxic effects of an organophosphate pesticide, envoy 50 SC on the histopathological, hematological, and brain acetylcholinesterase activities in stinging catfish (*Heteropneustes fossilis*). J Basic Appl Zool. 2020; 81:1-4. https://doi.org/10.1186/s41936-020-00184-w
11. Mehra S, Chadha P. Effect of 2 naphthalene sulfonate on biochemical stress markers as well as structural integrity of DNA in liver and kidney tissue of *Channa punctatus* after acuteexposure. Toxicol Int. 2021; 23:213-222. https://doi.org/10.18311 /ti/2021/v28i3/26733
12. Henderson AM, Gervais JA, Luukinen B, Buhl K, Stone D. Glyphosate general fact sheet. National Pesticide Information Center. Oregon State University Extension Services. 2010.
13. Singhal P, Singh, NN, Sreedhar, G, Banerjee, S, Batra, M, Garg, A. Evaluation of histo-morphometric changes in tissue architecture in relation to alteration in fixation protocol-An *invitro* study. J Clin Diagn Res. 2016; 10(8):ZC28-ZC32. https://doi.org/10.7860/jcdr/2016/19007.8236
14. Olurin KB, Olojo EAA, Mbaka GO, Akindele AT. Histopathological responses of the gill and liver tissues of *Clarias gariepinus* fingerlings to herbicide, glyphosate. Afr J Biotechnol. 2006; 5(24):2480-2487.
15. Samanta P, Mukherjee AK, Pal S, Kole D, Ghosh AR. Toxic effects of glyphosate-based herbicide, Excel Mera 71 on gill, liver, and kidney of *Heteropneustes fossilis* under laboratory and field conditions. J Microsc Ultrastruct. 2016; 4(3):147-155. https://doi.org/10.1016/j.jmau.2016.01.002
16. Selvanathan J, Vincent S, Nirmala A. Histopathology changes in freshwater fish *Clarias batrachus* (Linn.) exposed to mercury and cadmium. Int J Pharm Pract. 2012; 3(4):422-428.
17. Ayoola SO. Histopathological effect of glyphosate on juvenile African Catfish (*Clarias gariepinus*). Am-Eurasian J Agric Environ Sci. 2008; 4(3):362-367. https://ir.unilag.edu.ng/handle/123456789/6349
18. Erhunmwunse NO, Ewere EE, Ainerua M, Alohan FI. Biomarker of exposure: Alterations in GIT of post juvenile Africa cat fish (*Clarias gariepinus*) exposed to sub-lethal concentrations of glyphosate herbicide (IPA 360g/L). Int J Innov Sci. 2014; 4:1-5.
19. Lu C, Kania PW, Buchmann K. Particle effects on fish gills: An immunogenetic approach for rainbow trout and zebrafish. Aquac. 2018; 484:98-104. https://doi.org/10.1016/j.aquaculture.2017.11.005
20. Srivastava CBL, Seal M. Electroreceptors in Indian catfish teleosts. Sensory Physiology of Aquatic Lower Vertebrates. Satellite Symposium of the 28th International Congress of Physiological Sciences; 1981. p. 1-11. https://doi.org/10.1016/b978-0-08-027352-5.50006-9
21. Schwaiger J, Ferling H, Mallow U, Wintermayr H, Negele RD. Toxic effects of the non-steroidal anti-inflammatory drug diclofenac: Part I: Histopathological alterations and bio-accumulation in rainbow trout. Aquattoxicol. 2004; 68(2):141-150. https://doi.org/10.1016/s0166-445x(04)00096-7
22. Filippov AA, Golovanova IL, Aminov AI. Effects of organic pollutants on fish digestive enzymes: A review. Inland Water Biol. 2013; 6:155-160. https://doi.org/10.1134/s199508291302003x
23. Kumari AS, Kumar NSR. Effect of water pollution on histology of intestine of two fresh water fishes from Hussainsagar lake (AP). Indian J Environ Toxicol. 1997; 7(2):68-70.
24. Senapati T, Mukerjee AK, Ghosh AR. Observations on the effect of glyphosate-based herbicide on ultra structure (SEM) and enzymatic activity in different regions of alimentary canal and gill of *Channa punctatus* (Bloch). J crop weed. 2009; 5(1):233-242