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THE TOXICITY OF ESSENTIAL ELEMENT (Zn) IN THE BLOOD PROFILE OF FRESH WATER TELEOST, *CLARIAS BATRACHUS*

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Abstract: Minerals such as zinc (Zn) are abundant in the Earth's crust, serving as cofactors for a variety of proteins and modulating immune functions, however, in high doses, it might cause toxic effects on the body. This study examined the effects of excessive Zn concentrations on fish hematological parameters. The authors employed fresh water fishes *Clarias batrachus* as an experimental model and treated them with three different concentrations of Zn. The treatment with Zn was done for total 28 days and hematological parameters were assessed at different time intervals. As compared to control, Zn treated fishes showed considerable changes in hematological parameters such as, Haemoglobin (Hb), total erythrocyte counts (TEC), total leucocytes count (TLC), erythrocyte sedimentation rate (ESR), packed cell volume (PCV), mean corpuscular haemoglobin (MCH) and mean corpuscular volume (MCV). The present study showed that exposure to Zn could be linked to chronic and acute hematological toxicity in fishes in time and dose-dependent manner.

Keywords: Blood, *Clarias batrachus*, Haematology, Teleost, Toxicity, Zinc.

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INTRODUCTION

Heavy metal contamination of drinking water, such as arsenic, cadmium, nickel, mercury, chromium, zinc, and lead, has become a serious health concern (Rehman *et al.,* 2018). Heavy metals are present in essential and nonessential forms with relatively high density compared to water (Ferugusson, 1990). Every organism exposed to it through the air, water, or soil, experiences different effects (Jan *et al.*, 2015; Araujo and Cedeno-Macious, 2016). All living organisms require essential elements like Zn, Fe, Mn, Co, Cu etc. to grow and develop properly and their deficiency is also harmful to them. Nevertheless, due to their non-biodegradable nature, these metals are accumulating in an uncontrolled manner, which has made them toxic to organic life (Arruti *et al.*, 2010; Sani, 2011). Bhat *et al.* (2019) reviewed those heavy metals that may cause grievous changes in the

future and told that several health risks are associated with heavy metal toxicity. They affect the metabolism of organisms, even though they function as virtual components of the body.

All the fishes live in water, which makes them vulnerable to oppressive conditions because they breathe, eat, reproduce, and survive there. As a result of their high tolerance, these can escape the harmful effects of unwanted environments (Yarsan and Yipel, 2013; Saleh and Marie, 2014). The haematological parameters are important indicators also used to diagnose physiological disorders (Satheeshkumar *et al.,* 2011). With the help of heavy metals, morphological abnormalities, osmoregulation anomalies, genetic anomalies, physiological abnormalities, and transfigurations of the blood profile such as Hb, TEC, TLC, ESR, PCV, MCH, MCV, glucose, cholesterol, etc. are observed (Srivastava and Prakash, 2019).

The zinc plays an essential role in human growth and development. Zn is found in the body in amounts ranging from 1.4 -2.3 grams in adults (Bhomik *et al.,* 2010). There are many uses of this metal, including solar panels, paints, semiconductors, and waste water treatment plants (Zimmermann *et al.,* 2012). A small amount or up to permissible limit of this element is required for structural components of many enzymes, metabolism, and normal development. Nevertheless, when zinc concentrations exceed the physiological requirements of fish bodies, it becomes toxic (Shukla *et al*., 2002). Zn has been found to cause toxic effects in aquatic organisms, including fish (Ryan *et* a*l*., 2009; Mottin *et al.,* 2010).

Zn can also cause several fatal diseases hence it is imperative to understand how heavy metals, like Zn affect fish health and how to mitigate them (Saxena *et al.,* 2009). Previously, Celik *et al.* (2013), El-Sayed and Shokr (2015) evaluated how zinc altered the haematological and biochemical profiles of tilpia exposed to different concentrations. Therefore, the present study was conducted to determine the effect of sub-lethal doses of Zn on haematological parameters of fresh water teleost *C. batrachus* under controlled laboratory conditions.

MATERIALS AND METHODS

Fish collection and groups

Fresh water fishes *Clarias batrachus* were purchased from different fish markets of Bareilly, Uttar Pradesh, India, and randomized into four groups. The fishes were acclimatized for about 7-10 days in laboratory conditions. The groups were as follows:

Group 'A'- Control, fishes were kept in fresh water.

Group 'B'- Fishes were exposed to 1.0 ppm of Zn.

Group 'C'- Fishes were exposed to 3.0 ppm of Zn.

Group 'D'-Fishes were exposed to 6.0 ppm of Zn.

A total of 16 fishes weighed from 30-40 grams were included in each of the above groups.

Treatment of Zinc in fishes: Different zinc toxicants were prepared in laboratory by using 1 g pure Zn metal which was dissolved in H_2SO_4 and diluted to 1000 ml stock solution by using distilled water. Stock solution 1.0 ml had 1.0 mg of lead in concentration. Fishes were exposed to 1ppm, 3ppm and 6ppm of zinc for 15 days.

Collection of blood and hematological analysis: Blood was collected from the caudal vein of experimental fishes with the help of anticoagulated syringes. The collected blood from each group was used to assess Hb, TEC, TLC, ESR, PCV, MCH and mean MCV as reported earlier (Wintrobe 1981).

Statistical analysis: The data was analyzed by using one-way Anova test using GraphPad prism 9.5.0.

RESULTS AND DISCUSSION

Studies were conducted to determine the blood parameters like Hb, TEC, TLC, ESR, PCV, MCH and MCV in blood samples collected from experimental fishes from each group. Throughout the four-week period, different parameters were accessed on each fish from zinc-treated different groups.

Hb: The control group showed of Hb was 13.60%, and the maximum reduction in the haemoglobin was recorded at 10.50% with 22.6% decreased in

group D during maximum time exposure and concentration (6.0 ppm of zinc after 28 days exposure period) (Table-1, Fig.-1).

TEC: Zinc exposure resulted insignificant

Table 1: Haematological parameters of *C. batrachus* exposed to various concentration of zinc.

Sl. No	Parameters	Group A (Control)	Group A (Control)				Group A (Control)				Group A (Control)			
			7 Days	14 Days	21 Days	28 Days	7 Days	14 Days	21 Days	28 Days	7 Days	14 Days	21 Days	28 Days
1.	Hb %	13.60	12.50	12.50	11.80	11.50	12.50	12.00	11.50	11.00	12.00	11.00	11.00	10.50
2.	TECx106/Cumm	3.17	2.90	2.85	3.00	2.90	3.00	2.70	2.50	2.40	2.69	2.50	2.60	2.40
3.	TLC /Cumm	13100	15600	15800	16300	18300	15000	16400	16100	19500	16100	17100	18200	19900
4.	ESR (mm/h)	2.13	2.80	2.70	3.00	3.15	2.40	2.95	2.95	3.00	2.30	2.70	2.80	3.20
5.	PCV %	43.00	42.00	38.00	34.00	31.00	42.00	36.00	33.00	30.00	40.00	34.00	31.00	29.00
6	MCV (3)	135.64	144.82	133.33	113.30	106.89	140.00	133.30	132.00	125.00	148.69	136.00	119.23	120.83
7	MCH (Pg)	42.90	43.10	43.85	39.33	39.65	41.66	44.44	46.00	45.83	44.60	44.00	42.30	43.75

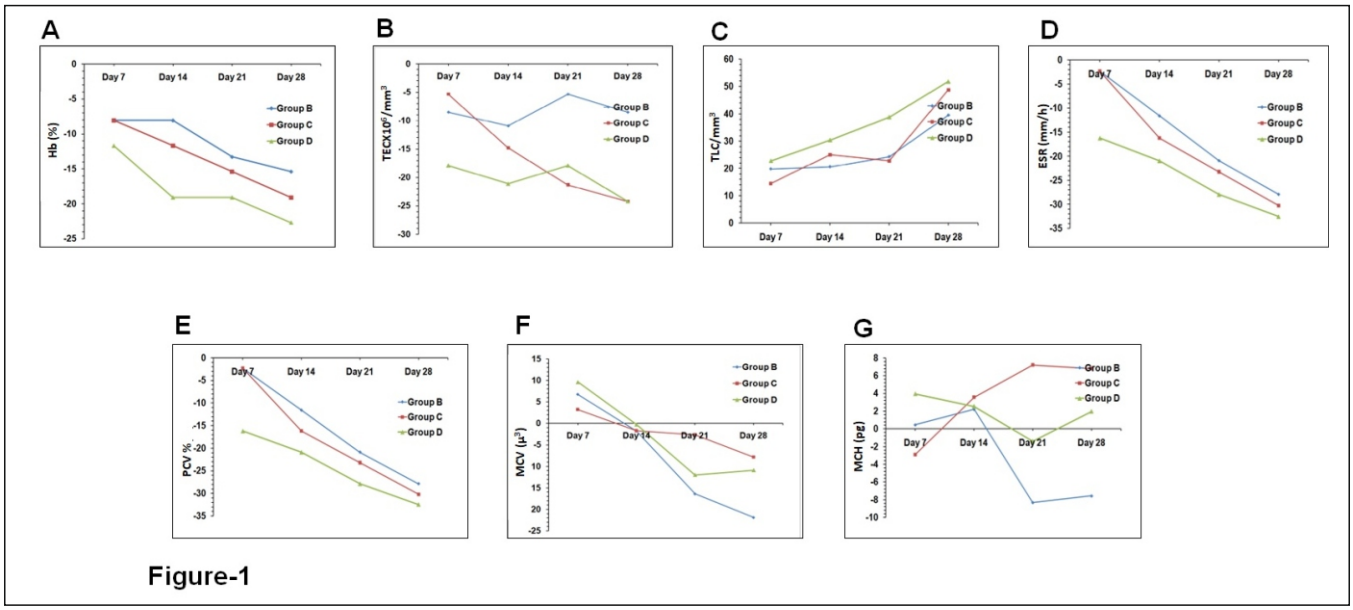

Fig. 1: Line graph shows the changes in (A) heamoglobin, (B) total erythrocyte counts, (C) total leucocytes count, (D) erythrocyte sedimentation rate (ESR), (E) packed total erythrocyte counts cell volume (PCV), (F) mean corpuscular haemoglobin (MCH), and (G) mean corpuscular volume in 1ppm, 3ppm, and 6ppm Zinc-treated *Clarias batrachus* groups each week for a total of four weeks.

decrease in the total erythrocytes. The maximum change in the TEC value was $2.40x10^6$/Cu mm. (28 Days: 3.0 ppm and 6.0 ppm). Maximum decrease was observed to 24.2% (Table-1, Fig.-2).

TLC: *C. batrachus* showed approximately the same negative response when compared with the control values. The highest increase was 19,900/mm^3 (28 days at 6.0 ppm) and minimum was 15,000 (7 days: 3.00 ppm). 51.9% increase observed at the maximum concentration and time exposure (Table-1, Fig.- 3).

ESR: The value of ESR in *C. batrachus* when exposed to sublethal concentration of Zn indicated maximum value of ESR (3.20 mm/h) at 28 days (6.0 ppm Zn) and minimum (2.40 mm/h) at 7 days (3.0 ppm) of exposure period. The maximum increased observed was 50.2% in Group D during 28 days exposure period. These observations indicate that the sublethal concentration of both the heavy metals produced

pronounced effects on ESR of all the experimental group of fishes (Table-1, Fig.- 4).

PCV: In PCV, the maximum effect was indicated by the values of 29% at maximum concentration during maximum exposure period. (28 days: 6.0 ppm of Zn) and minimum reduction was apparent from the values of 42% (7 days: 1.0 ppm and 3.0 ppm of Zn. The 32.5 percentage decreased were observed after 28 days exposure in Group D (Table-1, Fig.- 5).

MCV: The MCV values were 148.69 μ^3 during 7 days exposure period at 6.0 ppm Zn (Group D) whereas minimum value of MCV was 106.89 μ^3 during 28 days exposure period (Group B). The maximum percentage decrease was -16.4% in Group B during 21-day exposure period while the maximum increased recorded was 9.62% during 7 days in Group D (Table-1, Fig.- 6).

MCV: In case of MCH values, *C. batrachus*, reached its peak at 46.0 (pg) after 21 days exposure (Group C). Whereas minimum value of MCH was 39.33 (pg) during 1.00 ppm of zinc

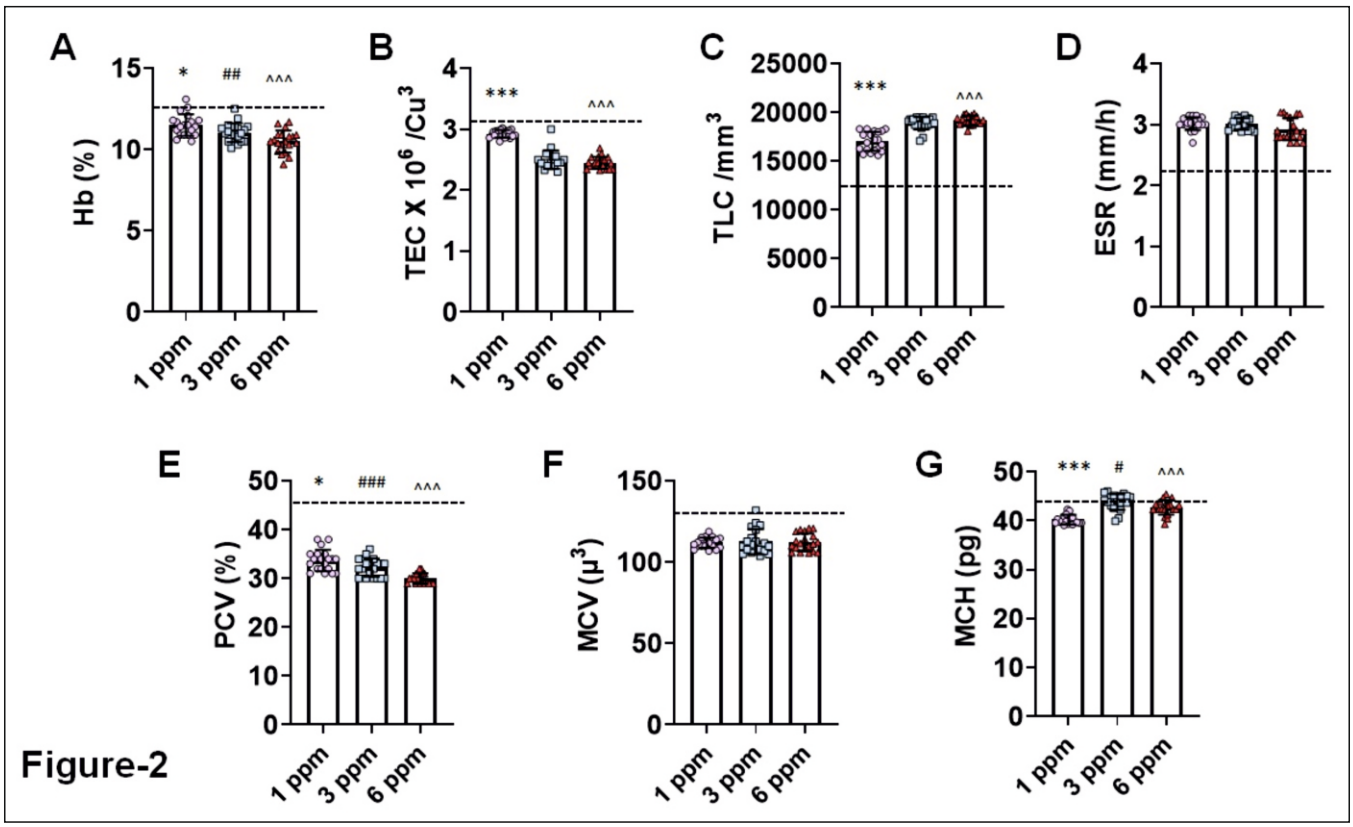

Fig. 2: Bar and scatter plot shows the changes in (A) Hb% (B) TEC (C) TEC (D) ESR (E) PCV) (F) MCH) and (G) MCV at 28th day in 6 ppm Zinc-treated *Clarias batrachus* groups. **P*<0.05, ****P*<0.0005-versus 3 ppm zinc; #*P*<0.05, ##*P*<0.005, ###*P*<0.0005-versus 3 ppm zinc; ^ ^ ^*P*<0.0005-versus 1 ppm zinc; data analyzed.

during 21 days post treatment with -8.32% changes (Table-1, Fig.- 7).

Table-2 depicts the results of Hb, TEC, TLC, ESR, PCV, MCH and MCV in fishes after 28 days exposure to Zn (6.0 ppm).

When heavy metals accumulate biologically in undesirable amounts, they can cause serious complications in living organisms and even lead to death (Bhat *et al.,* 2019). A severe alteration in the values of the experimental fishes as compared to the control fish occurred when they were exposed to various sublethal concentrations of zinc (1ppm, 3ppm, and 6ppm) at different time intervals (7, 14, 21 and 28 days). Physiological status of animals exposed to toxicants can be diagnosed by measuring blood parameters. The present study evaluated the functional status of *C. batrachus* exposed to zinc at various concentrations over a period of time. According to Joshi *et al.* (2002), heavy metal toxicity can lead

to anaemia in fish exposed to these metals. The results of this study were in agreement with those of Tyagi and Srivastava (2005), who tested *Channa punctatus* and Olurin *et al.* (2012), who examined *Clarias gariepinus* when treated with zinc chloride. Hoq and Haque *(*2014) found the correlation between zinc concentrations and blood glucose level in the blood of fish *Heteropneustes fossilis.* They precisely observed that Zn amount was increased and the blood glucose was significantly decreased. Ganesan and Karrupasami (2015) found that zinc toxicity induced alteration in the haematological values of *channa punctatus* with decrease in RBC, MCV and MCH and Increase in WBC and MCHC due to disturbance in metabolic and haemopoitic activities in fish. Rani *et al.* (2015) observed significant decrease in RBS, PCV% and HB%. However, the DLC, Platelets count and blood clotting time of the fish did not change significantly with reference to Zn and Cd. El-Sayed and Shokr (2015) evaluated reflection in haematological values of nile tilapia to sub lethal concentration of zinc sulphate (15, 30 and 45 ppm) during different exposure period (24, 48 and 72 hours). They concluded that zinc related stress in fish as shown by haematological parameters can be a good indicator.

A comparative study of Zn, Cd and Cu treatment with fresh water fish *Clarias batrachus* resulted the Zn and cadmium exposure decreased the TEC haemoglobin percentage and morphological changes in red blood cells while the Cu shows the slightly changes (Kumar and Ram, 2016). Thuraiswami (2020) observed the zinc toxicity affects the blood parameters of *Mystus mantanus,* which can be reduced by adding probiotics. The heavy metals such as lead and zinc can induce the biochemical changes in the blood of fresh water fishes *Heteropneustes fossilis and Clarias batrachus* (Verma *et al.,* 2020a, 2020b, 2020c, 2020d, 2021).

The results of the present work conclusively demonstrate that the experimental fish are sensitive to any forbidding changes in their aquatic environment. Although Zn is an essential element, it can have adverse effects on the blood physiology of an organism when it is in an undesirable condition. In the biosphere, this metal is regularly accumulating and magnifying due to human activities. Therefore, studying the haematological characteristics of *Clarias batrachus* can serve as an early warning sign of zinc pollution in aquatic systems.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1. **Araujo C.V.M. and Cedeno-Macias L.A.** (2016). Heavy metals in yellow fin tuna (*Thunnus albacares*) and common dolphin fish (*Coryphaena hippurus*) landed on the Ecuadorian coast. *Sci Total Environ.* 541:149-154. http://dx.doi.10.1016/ j.scitotenv.2015.09.090.
2. **Arruti A., Fernández-Olmo I. and Irabien A.** (2010). Evaluation of the contribution of local sources to trace metals levels in urban PM 2.5 and PM 10 in the Cantabria region (Northern Spain). *J Environ Monit.* 12(7):1451-1458. http://doi.10.1039/b926740a.
3. **Bhat D.S.A., Hassan D.T. and Majid P.S.** (2019). Heavy metal toxicity and their harmful effects on living organism-A review. *International Journal of Medical Science and Diagnosis Research.* 3 (1): 106-122.
4. **Bhowmik D., Chiranjib K.P. and Kumar K.P.S.** (2010). A potential medicinal importance of zinc in human health and chronic disease. *Int J Pharm Biomed Sci.* 1(1):05-11.
5. **Celik E.S., Kaya H., Yilmaz S., Akbulut M. and Tulgar A.** (2013). Effects of zinc exposure on the accumulation, haematology and immunology of *Mozambique tilapia, Oreochromis mossambicus. African Journal of Biotechnology*. 12(7):744-753. https://doi.org/10.5897/AJB12.1408
6. **El-Sayed A. and Shokr M.** (2015). Effects of zinc on hematology and biochemistry of Nile tilapia. J. *Chemic. Pharmaceut. Res.* 7(3): 1943-1950.
7. **Fergusson J.E.** (1990). The Heavy Elements: Chemistry, Environmental Impact and Health Effects. Pergamon Press, Maxwell House, Fairview Park, Elmsford, NY 10523. 614 p.
8. **Ganesan J. and Karuppasamy R.** (2015). Impact of zinc on haematological parameters of freshwater fish *Channa punctatus* (Bloch). *International Journal of Current Research.* 7 (03): 13626-13630.

9. **Hoq T. and Haque N.** (2014). Effects of sublethal concentrations of zinc (II) sulphate heptahydrate (ZnSO4.7H2O) on blood glucose level of freshwater fish *Heteropneustes fossilis*. *Intl. J. Scient. Techon Res.* 3 (5): 128-131.

10. **Jan A.T., Azam M. and Siddiqui K. *et al.*** (2015). Heavy metals and human health: mechanistic insight into toxicity and counter defense system of antioxidants. *Intl. J Mol Sci*. 16(12): 29592-29630.http://doi.10.3390/ijms161226183.

11. **Joshi P.K., Bose M. and Harish D.** (2002). Haematological changes in the blood of *Clarias batracus* exposed to mercuric chloride. *Ecotoxic. Environ monit*. 1:199-122.

12. **Kumar M. and Ram M.** (2016). Toxicity of some heavy metals on blood characteristics. of freshwater fish *Clarias batrachus*. IJFAS 24(1): 85-89.

13. **Mottin E., Caplat C., Mahaut M.L., Costil K., Barillier D., Lebel J.M. and Serpentini A.** (2010). Effect of in vitro exposure to zinc on immunological parameters of haemocytes from the marine gastropod *Haliotis tuberculata*. *Fish Shellfish Immun*. 29(5):846-853. http://doi.10.1016/j.fsi.2010.07.022

14. **Olurin K.B., Olojo E.A.A. and Tijani O.B.** (2012). Effects of zonc on haematological parameters of African catfish *(Clarias gariepinus)*. *Asian Journal of Pharmaceutical and Health Sciences*. 2(1): 266-272. https://ajphs.com/ article/2012/2/1/266-272.

15. **Rani S., Gupta R.K. and Rani M.** (2015). Heavy metal induced toxicity in fish with special reference to zinc and cadmium. *IJFAS*. 3(2): 118-123.

16. **Rehman K., Fatima F., Waheed I. and Hamid Akash M.S.** (2018). Prevalence of exposure of heavy metals and their impact on health consequences. *J Cell Biochem*. 119(1): 157-184. http://doi.10.1002/jcb.26234.

17. **Ryan A.C., Tomasso J.R. and Klaine S.J.** (2009). Influence of pH, hardness, dissolved organic carbon concentration and dissolved organic matter source on the acute toxicity of copper to Daphnia magna in soft waters: implications for the biotic ligand model. *Environ Toxicol Chem* 28(8):1663-1670. http://doi.10.1897/08-361.1

18. **Saleh Y.S. and Marie M.S.** (2014). Assessment of metal contamination in water sediment, and tissues of *Arius thalassinus* fish from the Red Sea coast of Yemen and the potential human risk assessment. *Environmental Science Pollutant Research*. 22(7): 5481-5490. http://doi.10.1007/s11356-014-3780-0.

19. **Sani U.** (2011). Determination of some heavy metals' concentration in the tissues of tilapia and catfishes. *Biokemistri*. 23(2):73-80.

20. **Satheeshkumar P., Ananthan G., Senthil Kumar D. and Jagadeesan L.** (2011). Haematology and biochemical parameters of different feeding behaviour of teleost fishes from Vellar estuary, India. *Comparative Clinical Pathology*. 21(6): 1187-1191. http://eprints.cmfri.org.in/ 8687/.

21. **Saxena M., Saxena H., Kaur P. and Kaur K.** (2009). Effect of heavy metal pollution of water on response of fish lymphocytes to mitogenic stimulation. *The Internet Journal of Veterinary Medicine*. 5 (2):1-4. https://ispub.com/IJVM/5/2/4714

22. **Shukla V., Rathi P. and Sastry K.V.** (2002). Effect of cadmium individually and in combination with other metals on the nutritive value of fresh water fish, *Channa punctatus*. *J. Environ. Biol.* 23:105-110. https://pubmed.ncbi.nlm.nih.gov/12602846/.

23. **Srivatava N.K. and Prakash S.** (2019). Effect of Zinc on the Histopathology of Gill, Liver and Kidney of Fresh Water Catfish, *Clarias batrachus* (Linn.). *International Journal of Biological Innovations*. 1 (1): 8-13. https://doi.org/10.46505/IJBI.2019.1102.

24. **Thuraiswami S.** (2020). Probiotic effect on zinc sulphate induced haematological toxicity in the fish *Mystus montanus. Intl. J.C.R.* 7(03):13626-13630.

25. **Tyagi A. and Srivastava N.** (2005). Haematological response of fish *Channa punctatus* (Bloch) to chronic zinc exposure.*J. Envl. Biol.* 26(2):429-432.https://pubmed.ncbi.nlm.nih.gov/16334279/.

26. **Verma V.K. Gupta N. and Gupta D.K.** (2021). Haematological responses of *Clarias batrachus* (Linn) to subleathal concentrations of lead. *IJPSR*. 12 (5): 1-10. http://doi.10.13040/IJPSR.0975-8232.12(5).2743-49.

27. **Verma V.K., Gupta N. and Gupta D.K.** (2020a). Zinc induced leucocyte anomalies in the fresh water teleosts*Clarias batrachus. Flora & Fauna*. 26 (1): 99-106. http://doi.10.33451/florafauna.v26i1pp99-106.

28. **Verma V.K., Gupta N. and Gupta D.K.** (2020b). Haematotoxicity of lead (Pb) on the blood parameters of fresh water teleosts *Heteropneustes fossilis* (Bloch*). IJPSR.*11(3): 1444-1451.

29. **Verma V.K., Gupta N. and Gupta D.K**. (2020c). Zinc induced changes in differential leucocyte counts of the fresh water teleost, *Heteropneustes fossilis* (Bloch). *Proc. Zool. Soc. India*. 19 (1): 91-100. https:// pubmed.ncbi.nlm.nih.gov/19436143/.

30. **Verma V.K., Gupta N., Gupta D.K. and Shalaby S.I.** (2020d). Heamotoxic assessment of differential leucocyte counts (DLC) in fresh water teleost *Heteropneustes fossilis* exposed to lead. *Egypt. J. Vet. Sci.* 51(3): 363-371. http://doi. 10.21608/EJVS.2020.28734.1169.

31. **Wintrobe M.M.** (1981). Clinical haematology. Lea and Febiger, Philadelphia, VIII Edition: 202.

32. **Yarsan E. and Yipel M.** (2013). The important terms of marine pollution bio makers and bio monitoring, bio accumulation, Bio Concentration, Bio Magnification. J *Molecular Biomarker & Diagnosis.* S1:003. http://doi.10.4172/2155-9929.S1-003.

33. **Zimmermann Y.S., Schaffer A., Hugi C., Fent K., Corvini P.F and Lenz M.** (2012). Organic photovoltaics: Potential fate and effects in the environment. *Environment International.* 49:128-140. https://doi.org/ 10.1016/j.envint.2012.08.015.