

Impact of the Herbicide Glyphosate Roundup (41%) On The Haematology of the Freshwater Fish, *Catla Catla* (Hamilton)

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Abstract: Herbicide Glyphosate Roundup (41%), has been widely used in aquatic and agricultural weed control in several countries including India. Herbicide glyphosate roundup (41%) has been associated with pollution due to its prolonged persistence and quick accumulation in blood as well as tissues. The present study thus aims to review the works of toxic effects of Herbicide glyphosate roundup (41%), on haematological indices of fish, *Catla catla* for the experimental period of 24hrs, 48hrs, 72hrs and 96hrs. LC50 value of Herbicide glyphosate roundup (41%), for *Catla catla* comes out to be 4.60ppm /l. The studies revealed that Total Erythrocyte Count (TEC), Total Leucocyte Count (TLC), Haemoglobin (Hb), and Haematocrit (Hct), and exhibited marked decline while calculated values viz., Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Haemoglobin Concentration (MCHC) showed fluctuating pattern. The observed alterations were ultimately become the causative for affecting the general health status of the fish. The values were statistically analysed and most of values were found to be significant at 5% level of DMRT.

Keywords: *Catla catla*, Glyphosate Roundup (41%), Haematology, Herbicide.

I. Introduction

Environmental pollution by toxicants has become, one of the most important problems in the world [1]. Pesticide contamination of aquatic system has attracted the attention of researchers worldwide [2] and has increased in the last decades due to extensive use of them in agricultural, chemical and industrial processes which are becoming threats to living organisms. Fish are more frequently exposed to these pollutants because it is believed that regardless of where the pollution occurs, it will eventually end up in the aquatic environment[3].

In recent past, much literature has been generated about the pollutional effects of various chemicals on different organs and haematology of different fishes by [4], [5], [6], [7], [8], [9], [10], [11], [12], [13], [14], [15], [16]. The above survey of the literature reveals that a lot of work has been carried out on various aspects of haematological changes in different fishes due to herbicide and pesticide pollution but the effect of the herbicide glyphosate Roundup (41%) on the same is less studied. Therefore an attempt has been made to study the impact of the herbicide glyphosate roundup (41%) on the haematology of the freshwater fish, *Catla catla* (Hamilton).

II. Material And Methods

Catla catla, a commonly occurring freshwater major carp fish available in the local ponds and rivers was selected for experimental studies. Bulk of sample of fishes, (*Catla catla*) ranging in weight from 3-4 gms and measuring 3-5cm in length were procured from Bhavanisagar reservoir. Fishes were acclimatized to the laboratory conditions for 2 weeks in large cement tank (6×4×3) at 24±3°C. The fishes were fed with ad-libitum and minced boiled egg. The level of the dissolved oxygen, pH, Alkalinity and hardness were monitored and maintained constant. The tanks were continuously aerated with electrically operated aerator. Fishes of about the same size (5-8gms and 7-10cm) irrespective of sexes were selected for the experiment. Technical grades of glyphosate roundup (41%), a herbicide, was used in this investigation. Batches of 10 healthy fishes were exposed to different concentrations of herbicide glyphosate roundup (41%) to calculate the LC₅₀ value. It was found as 4.60 ppm for 24hrs using probit analysis method [17].

Four groups of fishes were exposed to 0.46ppm (1/10th of 24hrs LC₅₀ value) concentration of the herbicide glyphosate roundup (41%) for 24hrs, 48hrs, 72hrs, and 96hrs respectively. Another group was maintained as control. At the end of each exposure period, blood was collected using heparinized syringe from the gills and various haematological parameters were analysed by using standard methods, RBC and WBC [18], HB [19], HCT [20], and MCV, MCH, MCHC as described by [21].

III. Results And Discussion

The results of haematological parameters were given in the table (1 and 2) and the values were statistically analysed using one way ANOVA (SPSS version 16.0 .0)

Toxicity and the effects of the herbicide glyphosate roundup (41%) in blood parameters of *Catla catla* has been analysed and the results were presented in tables (1-2) and figures (1-7). Blood is a patho-physiological reflector of whole body and therefore, blood parameters are useful in diagnosing the structural and functional status of body organs exposed to toxicants. The toxic chemicals inhibited the protein synthesis in fish which resulted in the total body weight.

The freshwater fish *Catla catla* exposed to sub-lethal concentration of glyphosate roundup(41%) at 27°C (lab condition) resulted in a significant decrease of RBC's count from control exposed to 24 hrs, 48hrs, 72hrs and 96hrs. Maximum decrease was noted in 96hrs. similar reports have been made by [22] and [23] on *Heteropneustes fossilis* and *Oreochromis mossambicus* after exposed to zinc and copper.

Lowering of RBC count might be due to damaging action of herbicides on peripheral red cell due to which viability of the cell was affected. [24] are one of the view that developed anaemia, could be due to effects of herbicide on haemopoiesis of cell membrane and hydrolysis of acetylcholine in body fluids by cholinesterase of erythrocytes. However it may be concluded that damaging effects on erythrocytes, may be secondary results from a primary action of the herbicides on the erythropoietic tissue due to which there was a failure in the red cell production.

In the present investigation the WBC were found to be increased from control in all exposure periods. Increase in the number of leucocytes occurs due to immunological response. An increase in lymphocytes suggests that the immune mechanism of fish gets stimulated and becomes adapted under herbicide stress to against the pollution in the environment.

IV. Tables And Figures

Table : 1. Changes in the blood of *Catla catla* exposed to 0.46ppm of Herbicide Glyphosate roundup (41%) in Short term duration

Periods	RBC (10 ⁶ ×mm ³)				WBC (10 ³ ×mm ³)				HB (gm%)				HCT(%)			
	C	E	MD	%	C	E	MD	%	C	E	MD	%	C	E	MD	%
24 hrs	3.18 ± 0.015 ^a	3.13 ± 0.015 ^{ab}	0.05***	1.57	14.17 ± 0.015 ^a	14.71± 0.015 ^{ab}	-0.54**	-3.87	9.53 ± 0.015 ^a	9.39 ± 0.015 ^{ab}	0.14***	1.46	28.62 ± 0.015 ^a	28.17 ± 0.015 ^{ab}	0.45***	1.57
48 hrs	3.17 ± 0.015 ^a	2.89 ± 0.015 ^{ac}	0.28***	8.83	14.14 ± 0.015 ^a	15.89± 0.015 ^{ac}	-1.75***	-12.37	9.51 ± 0.015 ^a	8.67 ± 0.015 ^{ac}	0.84***	8.83	28.60 ± 0.015 ^a	25.99 ± 0.015 ^{ac}	2.61***	9.12
72 hrs	3.15 ± 0.015 ^a	2.67 ± 0.015 ^{ad}	0.48***	15.23	14.11 ± 0.015 ^a	16.02± 0.015 ^{ad}	-1.91**	-13.53	9.49 ± 0.015 ^a	7.99 ± 0.052 ^{ad}	1.50***	15.80	28.59 ± 0.015 ^a	24.03 ± 0.015 ^{ad}	4.56***	15.94
96 hrs	3.13 ± 0.015 ^a	2.56 ± 0.015 ^{ae}	0.57***	18.21	14.09 ± 0.015 ^a	15.54± 0.015 ^{ae}	-1.45*	-10.29	9.47 ± 0.015 ^a	7.68 ± 0.015 ^{ae}	1.79**	18.90	28.57 ± 0.015 ^a	23.04 ± 0.015 ^{ae}	5.53***	19.35
SE	0.049				0.728				0.149				0.449			

significance was expressed using F-value at P<0.05.

Table : 2. Changes in the blood of *Catla catla* exposed to 0.46ppm of Herbicide Glyphosate roundup (41%) in Short term duration

Periods	MCV (µm ³)				MCH (pg)				MCHC (%)			
	C	E	MD	%	C	E	MD	%	C	E	MD	%
24 hrs	90.06 ± 0.015 ^a	89.91 ± 0.015 ^{ab}	0.15***	0.16	90.09± 0.015 ^a	29.76 ± 0.015 ^{ab}	60.33***	66.96	33.36 ± 0.015 ^a	33.31 ± 0.015 ^{ab}	0.05***	0.14
48 hrs	90.05 ± 0.015 ^a	89.86 ± 0.015 ^{ac}	0.19***	0.21	90.07 ± 0.015 ^a	24.19 ± 0.015 ^{ac}	65.19***	72.37	33.33± 0.015 ^a	33.38 ± 0.015 ^{ac}	-0.05*	-0.15
72 hrs	90.02 ± 0.015 ^a	89.81 ± 0.015 ^{ad}	0.21***	0.23	90.04 ± 0.015 ^a	23.56 ± 0.015 ^{ad}	66.48***	73.83	33.31 ± 0.015 ^a	33.42 ± 0.015 ^{ad}	-0.11***	-0.33
96 hrs	90.00 ± 0.015 ^a	89.76 ± 0.015 ^{ae}	0.24**	0.26	90.02 ± 0.015 ^a	22.45 ± 0.015 ^{ae}	67.57***	75.06	33.29 ± 0.015 ^a	33.44 ± 0.015 ^{ae}	-0.15***	-0.45
SE	0.021				0.679				0.007			

significance was expressed using F-value at P<0.05.

Values are Mean ± SD of five observation

Duncan's multiple range test (DMRT) was used to evaluate the comparison between means as indicated by different case letters in a descending order a, b,c,d and e

Statistical significant : * - P < 0.05 , ** - P < 0.01 , *** - P < 0.001 , ns - Not significant

Unit : C- Control / E- Experiment / SE- Standard Error / MD – Mean Difference / % - Percentage increases (+) or decreases (-) over control

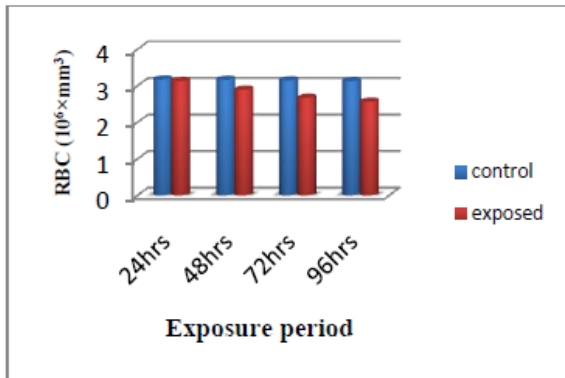


Fig.1. Changes in RBC of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

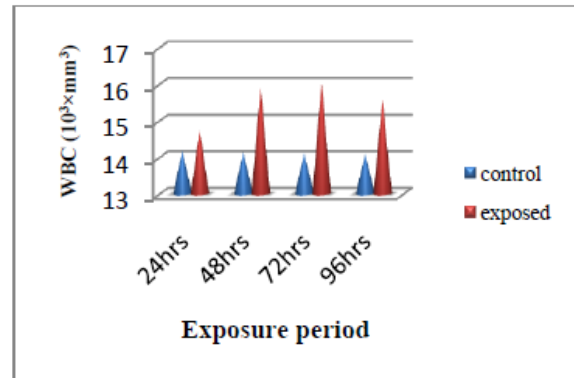


Fig.2. Changes in WBC of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

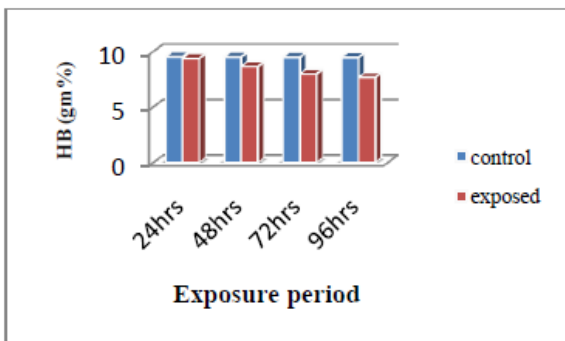


Fig.3. Changes in HB of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

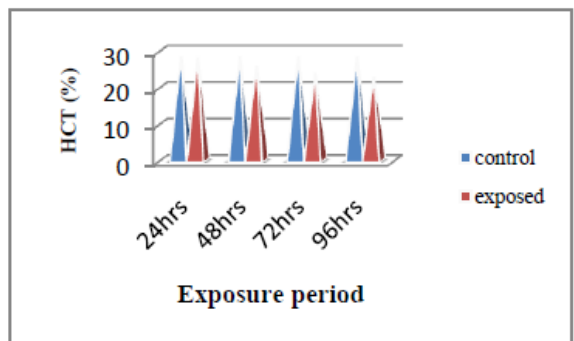


Fig.4. Changes in HCT of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

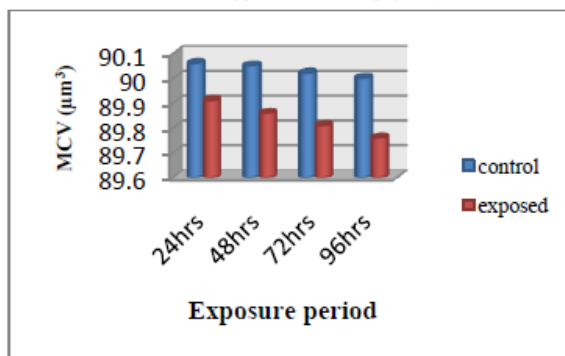


Fig.5. Changes in MCV of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

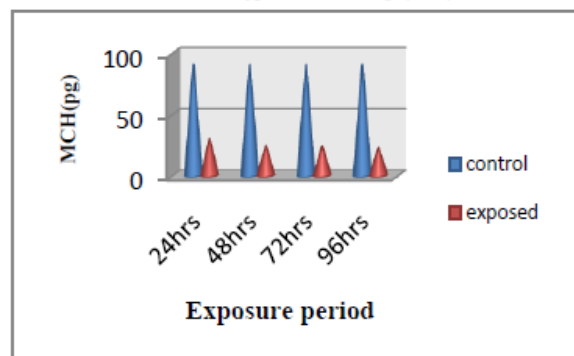


Fig.6. Changes in MCH of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

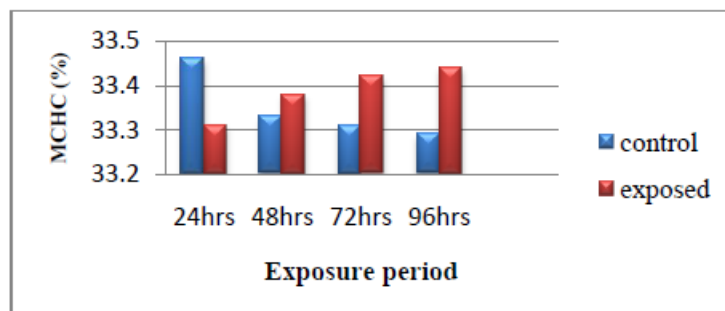


Fig.7. Changes in MCHC of *Catla catla* exposed to 0.46 ppm of Herbicide Glyphosate roundup (41%) .

Similar increase in total leucocytes count was observed by [25] in the fish *Channa striatus* on exposed to BHC and Malathion. [26] reported significant increase in the total leucocyte counts when they exposed to the

fish *Clarius batrachus* to sevin and *Heterophneustes fossilis* to chlordecone respectively. Similar increase in total leucocytes count has been observed by [27].

The Hb content of the exposed fish were found to be decreased when compared to normal fish upon exposure to 24hrs, 48hrs, 72hrs, and 96hrs to herbicide. It was supported by the findings of [28], and [29] in *C. carpio* on exposed to pesticide.

The reduction of HB might be attributed to the blood coagulation. [30] reported to decline in HB, HCT and RBC count by an organophosphorous insecticide in Japanese quail. The reduction can be related to decrease RBC number which indicates haemolysis, haemorrhage and reduced erythropoiesis in fishes on exposure to herbicide. Decrease in MCHC reveals that loss of HB is comparatively at higher rate than that of the HCT. The decreased MCV, MCH, and MCHC clearly indicate hypochromic microcytic anemia. The above findings are supported by [31]. Decrease HCT shows the magnitude of shrinking of cell size [32].

V. Conclusion

From the above study, it can be revealed that all the haematological parameters except WBC were decreased when compared to control on exposed to the herbicide glyphosate roundup (41%). This clearly indicates the toxic nature of the herbicide glyphosate roundup (41%) on the haematology of the fish *Catla catla*.

References

- [1]. R. Chandran, A.A.Sivakumar, S. Mohandass, M. Aruchami, Effect of cadmium and zinc on antioxidant enzyme activity in the gastropod, *Achatina fulica*, Comp. Biochem. Physiol, 140C, 2005, 422–426.
- [2]. H. M. Dutta, R. Dalal, The effect of endosulfan on the ovary of bluegill sunfish: a histopathological study (*Lepomis macrochirus* sp), Int. J. Environ. Res, 2, 2008, 215–224.
- [3]. F. Ajani and A. A. Awogbade, Hematological Changes of the African Catfish *Clarias gariepinus* (Burchell, 1822) Juveniles Induced by Diuron, British Biotechnology Journal, 2(4), 2012, 247–256.
- [4]. M.Santhakumar, M.Balaji, K.Ramudu, Effect of sublethal concentrations of monocrotophos on erythropoietic activity and certain haematological parameters of fish *Anabas testudineus*, Bull. Environ. Contam. Toxicol, 63, 1999, 379–384.
- [5]. M.Dorucu, and A. Girgin, The effects of cypermethrin on some haematological parameters of *Cyprinus carpio*, Aquaculture International, 9, 2001, 183–187.
- [6]. G.Affonso, V.L.P.Polezb, C.F.Correa, A.F.Mazon, M.R.R..Araujo, G.Moraes, and F.T. Rantin, Blood parameters and metabolites in the teleost fish *Colossoma macropomum* exposed to sulphide or hypoxia, Comparative Biochemistry and Physiology, Part C, 133, 2002, 375–382.
- [7]. M. Atamanalp, and T.Yanik, Alterations in hematological parameters of rainbow trout (*Oncorhynchus mykiss*) exposed to mancozeb, Turk J Vet Anim Sci, 27, 2003, 1213–1217.
- [8]. G. N. O. Ezeri, U. U. Gabriel and O. O. Opabunmi, Haematological response of cultured and wild *Clarias gariepinus* to acclimation, Environment and Ecology, 22, 2004, 628 – 632.
- [9]. O.K.Adeyemo, Haematological and histopathological effects of cassava mill effluent in *Clarias gariepinus*, African Journal of Biomedical Research, 8 (3), 2005, 179–183.
- [10]. P.C.Das, S.Ayyappan and J. K. Jena, Haematological changes in the three Indian major carps, *Catla catla* (Hamilton), *Labeo rohita* (Hamilton) and *Cirrhinus mrigala* (Hamilton) exposed to acidic and alkaline water pH, Aquaculture, 256, 2006, 80–87.
- [11]. U.U. Gabriel, P. E. Anyanmu and A. O. Akinrotimi, Haematological profile of black-chinned tilapia (*Sarotherodon melanotheron*) from Buguma creek., Niger Delta. Agri. J. 2 (3), 2007a, 348–387.
- [12]. U.R..Maheswaran, A.Devapaul, S.Muralidaran, B.Velmurugan and S.Ignacimuthu, Haematological studies of fresh water fish, *Clarias batrachus* (L.) exposed to mercuric chloride, International Journal Of Integrative Biology, 2, 2008a, 49–54.
- [13]. M.Ramesh, R..Srinivasan and M.Saravanan, Effect of atrazine (Herbicide) on blood parameters of common carp *Cyprinus carpio* (Actinopterygii: Cypriniformes), African Journal of Environmental Science and Technology, 3 (12), 2009, 453–458.
- [14]. A.Safahieh, A.Hedayati, A. Savari and A. Movahedinia, Experimental approaches of hematotoxic and immunotoxic effects of mercury chloride on Yellowfin Sea Bream (*Acanthopagrus latus*), American –Eurasian Journal of Toxicological Sciences, 2(3), 2010, 169–176.
- [15]. Z.Ahmed, Acute toxicity and haematological changes in common carp (*Cyprinus carpio*) caused by diazinon exposure, African Journal of Biotechnology, 10 (63), 2011, 13852–13859.
- [16]. F.B.Ada, E.Ekpenyong, and E.O.Ayotunde, Haematological, biological and behavioural changes in *Oreochromis niloticus* (Linne 1757) juveniles exposed to Paraquat herbicide, Journal of Environmental Chemistry and Ecotoxicology, 4(3), 2012, 64–74.
- [17]. D.J.Finney, Probit analysis, Cambridge Univ. Press, 3rd Edition, 1971, pp: 333.
- [18]. M. M. Wintrobe, Clinical hematology. Lea and Febiger, Philadelphia, Library of Congress, Print USA, 6th Eds, 1967.
- [19]. G.M.Dethloff, D. Schlenk, S. Khan and H.C. Bailey, The effects of copper on blood and biochemical parameters of rainbow trout (*Oncorhynchus mykiss*), Arch. Environ. Contam. Toxicol, Volume 36, 1999, pp. 415–423.
- [20]. E. F. Hesser, “Methods of routine fish hematology,” Progressive Fish Culturist, vol. 22, 1960, pp. 164–171.
- [21]. Dukova-Peneva, and M .Penev, 2007. Laboratory Hematology, Artik -2001, Sofia, 649 pp. (Bg).
- [22]. K.A.Goel, And G. Kalpana, Haematological characteristics of *Heteropneustes fossilis* under stress of zinc, Indian J. Fish, 36, 1985, 256–259.
- [23]. K.Sampath, R. James and K.M. Akbar Ali, Effects of copper and zinc on blood parameters and prediction of their recovery in *Oreochromis mossambicus*, Indian j. fish, 45(2), 1998, 129–139.
- [24]. R.G. Mac Farlaner And A.H.T. Robb Smith, Function of the blood, Academic Press, New York, 1981.
- [25]. S.Bhargava, R.S.Dixit And M. Rawat, BHC and malathion induced changes in TLC and DLC of *Channa striatus*, Proc. Acad. Environ. Boil. 8(1), 1999, 91–94.
- [26]. V. Garg, 2-4-Diamine, 3-Amino Azobenzene (DAAB) induced alteration in differential leucocyte in *Channa punctatus*, J. Environ. Boil, 3, 1982, 123–126.
- [27]. N. Mamta Kumari, P. Sing, And Gautam Ranjan, Effect of nickel on haematological parameters of an exotic carp, *Cyprinus carpio*, during the exposure and recovery periods, Natural Environ. And.Poll. Technology, 3(4), 2007, 491–494.

- [28]. S.Paulraj, Studies on the effect of paper factory effluent on the hydrography of the river Cauvery and its toxicity to common carp, *Cyprinus carpio* var. *communis* (linnaeus). Ph.D. thesis. Tamilnadu agricultural university, Coimbatore.
- [29]. G. Thomas Nithyanandam, C. Maruthanayagam, P. Viswanathan, Effects of sublethal level of a pesticide, monocrotophos, on haematology of *Cyprinus carpio* during the exposure and recovery periods, Nature Environment And Pollution Technology, Technoscience Publications, 6, 2007, 615-621.
- [30]. K. Germysz-Kathowaska, E. Szubartowska and E. Ecezanowaska, Pheripheral blood the Japanese quail (*Coturnix coturnix japonica*) in acode poisoning by different insecticides, comp. biochem. Physiol, 81c(1), 1985, 209-212.
- [31]. Ramalingam, V. Vimaladevi, R. Narmadaraji and P. Prabakaran, Effect of lead on the haematological and biochemical changes in the freshwater fish, *Cirrhinus mrigala*, Poll. Res., 19(1), 2000, 81-84.
- [32]. A.R. Shakoori, A.L. Mughal, M.J. Iqbal, Effect of sublethal doses of fenvalerate (a synthetic pyrethroid) administered continuously for four weeks on the blood, liver, and muscles of a freshwater fish, *Ctenopharyngodon idella*, Bull. Environ. Contam. Toxicol, 57, 1996, 487-494.