Sublethal Effects of Industrial Effluents on the Haematology of Freshwater Fish *H.Fossilis*During 10 Days Exposure

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Abstract

The general haematological tests are used to establish the normal health status and to diagnose the diseases caused by various factors viz. nutritional deficiencies, environmental stress, parasitic infections in human and veterinary medicine. The changes in composition of blood can therefore be used as a tool to detect the onset of physiological changes caused by environmental pollutants.

Alterations in the chemical composition of the natural aquatic environment by industrial effluents induce changes in behavioural and pathological aspects of fish. In fish pollutants are known to induce biochemical changes before the more drastic cellular and systemic dysfunction manifest themselves.

The composition of blood of fish is under greater influence of aquatic environment as compared to terrestrial warm blooded vertebrates.

In this context in present research work freshwater catfish *Heteropneustesfossilis* is exposed to sublethal doses ($1/10^{th}$ of 96 Hr. LC50) of raw (0.4%v/v) and neutralized pharmaceutical wastewater (0.9%v/v)producing ayurvedic products for 10 days duration and haematological parameters were investigated.

Significant changes were recorded in the blood composition of *Heteropneustesfossilis*in present study in both the industrial effluents at sublethal doses of exposure in 10 days duration at raw and neutralized levels.

KEY WORDS: Haematological parameters, *Heteropneustesfossilis*, sublethal doses, industrial effluents, pharmaceutical wastewater.

Introduction

Blood is a specialized connective tissue and a transporting medium in which there is a liquid intercellular substance the plasma and formed elements. It is a known fact that most diseases traumatic, inflammatory, infective, organic and neoplastic are accompanied by alterations in the composition of blood (Sood, 1996). These alterations may be in the plasma, serum or in the blood cells.

The general haematological tests are used to establish the normal health status and to diagnose the diseases caused by various factors viz. nutritional deficiencies, environmental stress, parasitic infections etc. in human and veterinary medicine, but such faculties are not well documented in poikilothermic organisms especially fish (WedemeyerandYasutake, 1977). Since blood takes part either directly or indirectly in all the biochemical processes of the body it is but natural to expect alterations in its properties when exposed to environmental toxicants. The changes in composition of

blood can therefore be used as a tool to detect the onset of physiological changes caused by environmental pollutants (Bansal*et*al , 1979).

The acute and chronic toxicity data by themselves have limited use in toxic control and hazard evaluation of industrial effluents (Sarkar*et* al, 1995). The ultimate aim of toxicity tests is to identify with reasonable accuracy a concentration that will protect and conserve the biological integrity in receiving water (Cairns, 1995; Rombke and Moltman*et al*, 1992).

Materials and Methods

The freshwater catfish *Heteropneustesfossilis* is used as a test organism for assessing the sublethal effect of pharmaceutical Industry effluent at raw and neutralized levels. This cat fish is readily available in freshwater dams and lakes of vidarbha region. The length of experimental fish varies from 12-14 cms. while weight varies from 30-40 gms.

The industrial effluent (Pharmaceutical wastewater) was collected from aayurvedic pharmaceutical company based at Nagpur producing a large number of ayurvedicproducts. The raw and neutralized wastewater from this industry was collected and characterized for heavy metals and toxicants and subsequently used.

After deriving 96 Hour LC50 for raw and neutralized wastewater, sublethal doses were prepared accordingly. The sublethal dose of raw pharmaceutical effluent was 0.40% v/v, while that of neutralized wastewater was 0.9%v/v. The selected doses of respective wastewater was prepared and rate of flow was kept at 24ml/min. approx..to give replacement in 24 hrs. from the test aquarium. This arrangement simulates the flow in streams and also ensures the removal of food particles and metabolites from the test chamber.

The experiments were performed in 20litre capacity glass aquaria having a capacity of 17 litres. A laboratory model constant dosing unit fabricated in National Environmental Engineering Research Institute (NEERI) workshop was used for dose delivery to the aquarium. The exposed fish were taken out after an interval of 10 days for haematological investigations and estimation of haematological parameters were done as per standard literature (Wintrobe, 1973; Dacie and Lewis, 1975; Sood, 1996).

Result and Discussion

Blood is an indicator of most of the physiological and biochemical functions. Variations in ambient environmental conditions which induce physiological stress have measurable influence on haematological parameters such as total erythrocyte count, haemoglobin, PCV, Corpusular constants and total and differential leucocyte counts.Marked variability in these values occur while poikilotherms e.g. fish adapt to changed physiological needs during pollution stress. Research on the application of haematological data for diagnosis of disease and toxicant induced stress in the aquatic environment has been recommended by authors of national and international recognition (Miller, 1983, Murty 1986, Evans, 1988).

The results of the present research are shown in table 1 and Table 2. The results indicate that prolonged exposure to industrial effluents of pharmaceutical industry at raw and neutralized levels induced significant alterations in the peripheral

blood parameters at sub-lethal dose of 1/10th of 96 hr LC50 values. The extent of changes were more in case of raw effluent as compared to neutralized one.

The decrease in erythrocytes might be due to increased erythrocyte destruction due to presence of toxicants in wastewater. Similar observations were reported by Rai and Qayyum (1984) in *Catlacatla* exposed to lead, supported by our findings.

Studies on acute effects of pollutants are useful in assessing the lethal thresholds of response to a particular toxicant. Acute toxicity end points are of use in screening of toxic wastewaters by regulatory agencies viz. Pollution Control Boards to impose limit on the toxic waste discharge. However continued sublethal exposures may produce a variety oof chronic symptoms not observed in short term tests (Hassler*et* al, 1967) or may result in adaptation by the fish to pollutant (Mckim*et al*, 1970).

The haematological studies on *H.fossilis* after 10 days exposure at sublethal doses confirm that sublethal doses of industrial effluents of ayurvedicpharmaceutlcal industry at raw and neutralized levels can change the blood profile of fresh water fish which can not be seen from external appearance and so haematological tests prove useful in these context to safeguard the aquatic ecosystems from further harm..

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References

- Bansal S.K.,Verma S,R., Gupta A.K. and Dalela R.C.(1979). Physiological dysfunction of the haematopoietic system in a freshwater teleost Labeorohita following chronic chloradane exposure I : Alterations in certain haematological parameters. Bull.Environ.Contam.Toxicol. 22: 666-673.
- Cairns J.Jr.(1995). Chemical Versus Biological Pollution Monitoring In" Pollution and Biomonitoring" Ed. B.C.Rana, Tata McGraw Hill Publishing Co.New Delhi.
- Dacie J.V and Lewis S.M.(1975). Practical Hematology, Churchill Livingstone, London, UK.
- Evans M.S.(1988). Toxic contaminants and Ecosystem Health : A Great lakes Focus, pp. 602, A Wiley InterscicencePubl.,New York.
- Hassler T.S.,Neuhold J.M and Sigler W.F.(1967). Effects of alkyl benzene sulfonate on rainbow trout, U.S.Dept.Interior, Bur.SportFish.Wildl.Tech.Paper 16: 1-15.
- MckimJ.M,Christensen G.M and Hunt E.P.(1970). Changes in the blood of brook trout (*Salvelinusfontinalis*) after short term and long term exposure to copper. J.Fish.Res.Bd.Can.27: 1883-1889.
- Miller W.R., Hendricks, A.C. and Greene J.C.(1983). Normal Ranges for Diagnostically important haematological and blood chemistry characteristics of rainbow trout (*Salmogairdneri*) Can.J.Fish.Aquat.Sci. 41 : 1829-1834.

- Murty A.S.(1986).Toxicity of Pesticides to Fish,Vol.II, CRC Press, Boca Raton, Florida, pp.143.
- Rai R and Qayyum M.A.(1984). Haematological responses in a freshwater fish to experimental lead poisoning. J.Environ.Biol. 5(1): 53-56.
- Raizada M.N, Singh C.P. and Maheshwari U.K.(1977). Haematological studies of Freshwater teleosts. Curr.Sci. 46(22) : 790-791.
- Rombke J and Moltmann J.F.(1996). Applied Ecotoxicology, Lewis Publishers, Boca Raton, pp.282.
- Sarkar R, Chaudhari P.R., Sitre S, and Gajghate D.G.(1995). Toxicity Testing Through Fish Bioassay And Its Application In India, in "Pollution and Biomonitoring" , Tata McGraw Hills Publishing Company Ltd. New Delhi.
- Sood R (1996). Haematology for students and Practioners, 4th Ed., Jaypee Brothers, New Delhi, pp. 397.
- Wedemeyer G.A. and Yasutake W.T. (1977). Clinical Methods for the assessment of the effects of environmental stress on fish health, Technical Paper, U.S.Fish.Wildlife Services, Washington D.C.1-18.
- Wintrboe M.M.(1973). Clinical Haematology, 7th edition, Lee and Fabiger, Philadelphia, pp.935.

Table 1
Alterations in the haematological parameters of *H.Fossilis* during 10 days exposure to raw pharmaceutical wastewater at sublethal dose of 0.4% v/v

Sr.No.	Parameter	Observation		Percentage Change Observed
		Control	Exposed	
1.	Total Erythrocyte Count ($x10^6$/cmm)	3.09 ±0.03	2.45±0.01$^{\#}$	20.71(-)
2.	Total Leucocyte Count ($x10^4$/cmm)	4.10±1.92	6.14±1.30$^{\#}$	49.75(+)
3.	Haemoglobin (Gm/100ml)	11.82±0.05	10.58±0.03$^{\$}$	10.49(-)
4.	Packed Cell Volume (%)	39.98±1.48	37.98±0.67$^{\$}$	5.00(-)
5.	Clotting time (Sec)	118.00±1.58	109.00±1.92$^{\#}$	7.62 (-)
6.	ESR mm/hr)	1.30± 0.21	1.70±0.10*	30.76(+)
7.	Mean Corpuscular Volume (cm3/cell)	129.38±1.19	144.20±0.98$^{\#}$	11.73(+)
8.	Mean Corpuscular Haemoglobin (pg/cell)	38.25±1.14	43.18±1.3$^{\$}$	12.88(+)
9.	Mean Corpuscular Haemoglobin Percentage (MCHC) (%)	29.55±0.45	28.63±0.55$^{\$}$	2.94(-)

Results are expressed as mean ±S.D. $^{\$}$P<0.01 *P<0.05 $^{\#}$P<0.001

Table 2
Alterations in the haematological parameters of *H.Fossilis* during 10 days exposure to neutralized pharmaceutical wastewater at sublethal dose of 0.9% v/v

Sr.No.	Parameter	Observation		Percentage Change Observed
		Control	Exposed	
1.	Total Erythrocyte Count ($x10^6$/cmm)	3.40±2.17	2.80±1.00*	17.64(-)
2.	Total Leucocyte Count ($x10^4$/cmm)	4.24±2.59	6.32±1.87*	49.05(+)
3.	Haemoglobin (Gm/100ml)	11.40±0.04	10.52±0.01*	7.71(-)
4.	Packed Cell Volume (%)	39.00±1.14	37.05±0.26$^{\$}$	5.00(-)
5.	Clotting time (Sec)	117.00±1.48	110.00±1.58$^{\$}$	6.00 (-)
6.	ESR mm/hr)	1.30± 0.27	1.63±0.41	23.38(+)
7.	Mean Corpuscular Volume (cm3/cell)	114.70±1.15	132.32±1.82*	15.36(+)
8.	Mean Corpuscular Haemoglobin (pg/cell)	3.52±1.26	37.57±1.18	12.08(+)
9.	Mean Corpuscular Haemoglobin Percentage (MCHC) (%)	29.23±0.27	28.39±0.41$^{\$}$	2.87(-)

Results are expressed as mean ±S.D. $^{\$}$P<0.01 *P<0.001