# A Study on the Effect of an insecticide "Encounter" (Herbal Plant Extract) on the Haematology of the Freshwater fish "Labeo rohita"

Binukumari.S<sup>1</sup>, Anbalagan.M<sup>2</sup> and Vasanthi.J<sup>3</sup> <sup>123</sup>PG and Research Department of Zoology,Kongunadu Arts and Science College,Coimbatore-641029, Tamil Nadu, India

Abstract: The onset of the industrial revolution has led to massive exploitation of a vast amount of our natural resources within a period of a few hundred years at unimaginable rates. Large amounts of wastes generated seriously damaging its natural processes. Fishes are aquatic and poikilothermic animals and perform best under a narrow range of optimum environmental conditions. A slight diversion from these, results in stress leading therby to diseases. The acute and sublethal toxicity of the insecticide 'Encounter' on the fingerlings of Labeo rohita was evaluated to determine its effect on the haematological values. The fish was exposed to varying levels of the toxicant concentrations using static bioassay to determine the median lethal concentration. The  $LC_{50}$ value is 0.11 ppm. The fish was exposed to different hours (24,48,72 and 96) in sublethal concentration 0.11 ppm and parameters like RBC,WBC,Hb,MCV,MCH,MCHC,PCV has been analysed.All haematological parameters except WBC were found to be decreased from control and the WBC was increased in all exposure periods. The values were statistically analyzed and most values were found to be significant at 5% level. *Keywords:* haematological, insecticide, lethal toxicity, sublethal, WBC.

#### I. Introduction

Water is the first and foremost need, without which there would not be any kind of life on earth.Adequate supply of clean water is essential for the survival of plants and animals and also fine quality drinking water is an essential human requisite. Good quality water is essential for all living being ([1]). The need to produce food in large quantities to cater for the ever increasing human population in the developing parts of the world has to increase in the use of agrochemicals (fertilizers and pesticides). Pesticides are used to control pests of food crops, livestock and human health. Due to their injudicious and indiscriminate usage, water bodies like ponds, lakes and low lying water filled areas are continuously polluted ([2]).

Fishes can serve as bioindicators of environmental pollution and can play significant role in potential risk associated with contamination in aquatic environment since they are directly exposed to chemicals resulting from agricultural production via surface run-off or indirectly through the food chain of ecosystem.

The extensive use of pesticides insecticides herbicides and fungicides are being promoted by Government of India to enhance the crop production to meet the demand of the growing population. The rationale behind this is the eradication of undesirable insects, pests, weeds and herbs for increased yield but on the other hand, these lead to large scale mortality of one of the most important aquatic fauna, the fishes forming stable food to the mankind. Hence the present study showed the effect of Herbal plant extract, Encounter on the haematology of the fresh water fish, Labeo rohita.

#### П. **Materials And Methods**

Fishes were maintained in a large tank and acclimatized to laboratory conditions for 21 days.Water was changed daily to maintain the oxygen content and to remove the excreta of fishes. Fishes were maintained at room temperature and fed with ad libitum daily at least one hour prior to the replacement of the tank water. Feeding was stopped two days prior to the experiment in order to keep the animal more or less in the same state of metabolic requirement.

The toxicant Encounter (Herbal Plant Extract) has been used for the study. It is dark brown in colour and liquid form.Encounter is a natural formulation derived from various herbals.It controls mites, scales, thrips and sucking pests effectively on all crops. Encounter is a mixture of Herbal extracts containing matrine, having excellent pest repellent property. It is effective on Caterpillars, Bollworms, Cabbage army worm, leaf folders on all agricultural, horticultural spices and plantation crops. It is primarily irritant to skin, eyes, respiratory tract and also affect the normal metabolism.

Batches of 10 healthy fishes were exposed to different concentrations of Encounter to calculate the  $LC_{50}$  value. One more set of fishes are maintained as control in tap water. To find the wide range of concentration 10 to 50 ml of Encounter were chosen and the number of dead or affected fish in each set up was counted at regular intervals upto 24 hours. The level of the dissolved oxygen, pH, alkalinity and hardness were monitored and maintained constant. The tanks were continuously aerated with electrically operated aerator. Appropriate narrow range of concentration 1-5 ppm was used to find the median lethal concentration, using a minimum of 6 fishes for each concentration and the mortality was recorded for every 24 hours upto 96 hours. It was found as 1.14 ppm for 96 hours. For this stock solution various sub lethal concentrations were prepared for bioassay study.

Four groups of fishes were exposed to 0.11 ppm(sub lethal concentration of 96 hours  $LC_{50}$  value)concentration of the Encounter for 24,48,72 and 96 hours respectively. Another group was maintained as control at the end of each exposure period, the blood was collected from gills using syringe and anticoagulants(ammonium oxalate,EDTA)were added and the haematological parameters such as Hb,RBC,WBC,MCV,MCH,MCHC and PCV were analyzed. The haemoglobin content was estimated by acid hematin method ([3]). Total RBC count and WBC count were counted using an improved Neubaur haemocytometer([4]). The mean corpuscular volume was calculated by using values of PCV% and the red blood cell counts expressed in  $\mu m^{-3}$  ([5]). The mean corpuscular haemoglobin content was calculated by using the value of haemoglobin concentration was calculated by using the values of haemoglobin content and the red blood cell counts and expressed in pg ([5]). The percentage of mean corpuscular haemoglobin content and the red blood cell counts and expressed in pg ([5]). The percentage of mean corpuscular haemoglobin content and the red blood cell counts and expressed in pg ([5]). The percentage of mean corpuscular haemoglobin content and the PCV% ([5]). The PCV percentage was calculated employing standard method and formulae ([6]).

## III. Results And Discussion

The amount of RBC in the blood of the fishes exposed to 0.11 ppm herbal plant extract for 24,48,72 and 96hrs was found to contain  $1.76,1.46,1.15,0.90 \times 10^{6}$ /mm<sup>3</sup> and mean control was found to be  $1.99 \times 10^{6}$ /mm<sup>3</sup>. The amount of WBC were found to be increased from the control. The values were 11.00,13.00,18.00,19.00,23.00 x  $10^{6}$ /mm<sup>3</sup> in control,24,48,72 and 96hrs respectively. The level of haemoglobin in the fish *Labeo rohita* on exposed to 24,48,72 and 96hrs was found to contain 4.3,3.7,0.02,2.9% and mean control was found to be 5.02 g%. The value of MCV in fishes exposed to 0.11 ppm Encounter for 24,48,72 and 96hrs was found to be 46.5µm3. The amount of MCH in the blood of the fishes exposed to 0.11 ppm Encounter was recorded as 18.0,17.0,14.0,10.0 and the control was found to be 22.0 Pg. The amount of MCHC recorded as 19.2,17.2,11.2,9.2,7.2 g/dL in control,24,48,72 and 96hrs was found to be 22.0 Pg. The amount of MCHC recorded as 19.2,17.2,11.2,9.2,7.2 g/dL in control,24,48,72 and 96hrs was found to be 22.0 Pg. The amount of MCHC recorded as 19.2,17.2,11.2,9.2,7.2 g/dL in control,24,48,72 and 96hrs was found to be 22.0 Pg. The amount of MCHC recorded as 19.2,17.2,11.2,9.2,7.2 g/dL in control,24,48,72 and 96hrs was found to be 22.0 Pg. The amount of MCHC recorded as 19.2,17.2,11.2,9.2,7.2 g/dL in control,24,48,72 and 96hrs exposures respectively. The amount of PCV in the blood of the fishes exposed to 0.11 ppm herbal plant extract for 24,48,72 and 96hrs was found to contain 13.2,10.2,10.08,0.92% and mean control was found to be 16.2%.

	Exposure	Exposure Periods				
Parameters	concentration 0.11ppm	Control	24 Hrs	48 Hrs	72 Hrs	96 Hrs
RBC	Mean±SD	1.99±0.56 <sup>a</sup>	1.76±0.46 <sup>ab</sup>	1.46±0.31 <sup>ab</sup>	1.13±0.32 <sup>b</sup>	0.90±0.17 <sup>b</sup>
(10 <sup>6</sup> /mm <sup>3</sup> )	%		11.55↓	26.63↓	43.21↓	54.77↓
WBC	Mean±SD	11.00±1.58 <sup>d</sup>	13.00±2.05°	18.00±1.69 <sup>b</sup>	19.00±1.15 <sup>b</sup>	23.00±1.37 <sup>a</sup>
(10 <sup>6</sup> /mm <sup>3</sup> )	%		18.18↑	63.63↑	72.72↑	109.09↑
Haemoglobin	Mean±SD	5.02±1.03ª	4.3±0.097 <sup>b</sup>	3.7±0.72 <sup>b</sup>	0.02±0.001 <sup>d</sup>	2.9±0.63°
(gm %)	%		14.34↓	26.29↓	99.60↓	44.23↓
MCV	Mean±SD	46.5±3.26 <sup>a</sup>	32.1±4.49 <sup>b</sup>	36.1±3.26°	21.6±1.62 <sup>d</sup>	19.1±1.05 <sup>e</sup>
(μm <sup>3</sup> )	%		30.96↓	22.36↓	53.54↓	58.92↓
MCH	Mean±SD	22.0±2.17 <sup>a</sup>	18.0±1.82 <sup>b</sup>	17.0±1.35°	14.0±1.74 <sup>d</sup>	10.0±1.18 <sup>e</sup>
(pg)	%		18.18↓	22.72↓	36.36↓	54.54↓
MCHC	Mean±SD	19.2±1.82 <sup>a</sup>	17.2±1.54 <sup>b</sup>	11.2±0.97 <sup>c</sup>	9.2±1.39 <sup>d</sup>	7.2±0.95°
(g/dL)	%		10.41↓	41.66↓	52.08↓	62.50↓
PCV	Mean±SD	16.2±1.36 <sup>a</sup>	13.2±0.98 <sup>b</sup>	10.2±0.85°	10.08±1.55°	0.92±0.14 <sup>d</sup>
(%)	%		18.51↓	37.03↓	37.77↓	94.32↓

	IV.	Table
Effect of ENCOUNTER on haematological pa	arameters	in blood of the fish <i>Labeo rohita</i> .

Results are mean (±SD) of 5 observations

% = Parenthesis denotes percentage increase/decrease over control.

In a column, means followed by a common letter or not significant at 1% level by using DMRT.

To get an insight into nature of changes that are taking place in the blood parametes in an organism as a result of insecticidal pollution by Herbal plant extract. Encounter haematological studies were carried out in the fish, Labeo rohita. Decrease in RBC may be due to the disruptive action of the pesticides on peripheral cell due to which viability of the cells was affected. Anaemia could be due to the effect of insecticide on haemopoisis or attraction of cell membrane. Similar reports have been given by ([7]). An increase in lymphocytes suggests that the immune mechanism of fish gets stimulated and becomes adapted under pesticide stress to fight against the pollutants in the environment. This significant increase in total leucocyte count might be due to immunological reactions to produce more antibodies to cope with the stress induced by these toxicants ([8]). The reduction in Hb content can be related to decrease in RBC number, which indicates haemolysis, haemorrhage and reduced erythropoesis in fishes on exposure to insecticides. In the present study, MCV and MCH was found to increase and then decrease gradually in all exposure periods. Increase in MCV values in 24 hours may be considered as an index of RBC destruction. Increase in MCH also indicates endosmosis where involves passage of solvent from concentration solution to more concentration solution.Decrease in MCHC reveals that loss of Hb is comparatively at higher rate than that of the PCV. Increase in PCV shows the magnitude of shrinking of cell size([9]).([10]) reported that the acute exposure to LC50 values of diazinon induced haematological alterations in Indian carp and offers a tool to evaluate toxicity derived alterations.

## V. Conclusion

The haematological parameters except WBC were found to be decreased from control and the WBC has increased in all exposure periods. From the above investigation it can be inferred that the aquatic animals are affected by the Encounter. So we should create awareness among people to use biocides instead of pesticides and herbicides.

### References

- [1] Azhi Bin Indris, Abdullah Almanamum Mohammad Amin, Mohamod soon and Wan Noor wan Azmin, Review of water quality standards and practices in Malaysia, *Poll.Res*, 22(2), 2003, 145-155.
- K.Kumar,N.Saradhamani, Effect of insecticide, AVAUNT on glycogen content of the freshwater fish *Cirrhinus mrigala.Nature* Environ.Pollut.Technol,3(4),2004,515-518.
- [3] T.Sahli, Text book of clinical pathology, (Ed.Scward, Eimiller). Williams and Williams and Co., Baltimore, 1962, pp. 35.
- [4] S.L.Shah, and A.Altindag,Haematological parameters of tench (Tinca tinca L.) after acute and chronic exposure to lethal and sublethal mercury treatments.Bull.Environ.Contam.Toxicol,73,2004,911-918.
- [5] D.Anderson, and G.W.Klontz, Basic haematology for the fish culturist, Ann. Northwest Fish Culture Conference, 16, 1965, 38-41.
- [6] G.S.Sandhu, Research Technique in Biological Sciences. Anmol Publications, 1990, New Delhi. 209
- [7] S.Binukumari, and M.C.Subisha, Haematological responses in a Freshwater fish *Oreochromis mosambicus* exposed to Chlorpyrifos. *The Ekologia*, 10 (1-2), 2010, 1-8.
- [8] K.Shanthi,N.Saradhamani,S.Binukumari andM.Manimegalai, Effect of Detergent Surf Excel on oxygen consumption and Haematological Parameters in Freshwater Fish, *Tilapia mossambica.The Ekologia*, 9 (1-2),2009, 11-17.
- [9] A.R.Shakoori,A.L.Mughal,M.J.Iqbal, Effect of sublethal doses of fenvalarate (A synthetic pyrethroid) administered continuously for 4 weeks on the blood, liver, muscles of freshwater fish *Ctenopharyngodon idella.Bull.Environ.Contam.Toxicol*,57,1996,487-494.
- [10] Abdul RAUF and Naeemuddin ARAIN, Acute toxicity of diazinon and its effect on haematological parameters in the Indian carp, *Cirrhinus mrigala*(Hamilton). *Turk J Vet Anim Sci*, 37, 2013, 535-540.