Title: Study of the Genotoxic and Pathological effects of the Domestic Sewage and Agriculture Waste in Blood of the Fish Labeo rohita Applying Micronucleus Assy.

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Abstract— The present study is to determine the genotoxic and pathological effects of domestic sewage and agriculture waste on the blood parameters of the fish Labeo rohita. For this micronucleus assy were applied**.**

In these studies, I have found formation of micronuclei, binuceated cells, lobed nucleus, as genotoxic effects.

Pathological effects are development of many neutrophils, formation of thrombocytes, highly increased number of lymphocytes, and development of macrophages. .

Index Terms— Agriculture, Binucleated**,** Clastogenic, Chromosomes, Domestic, Genotoxic, Micronucleus, Macrophages, Neutrophils, Pathogenic, Sewge, Sticky, Thrombocytes.

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1 INTRODUCTION

He present study is to determine the clastogenic and Tpathogenic effects of agriculture waste and domestic sewage. The fish selected for following studies is a teleost fish named as Labeo rohita. This fish is cultured in the selected pond. The pond in which fish is cultured is surrounded with fields used for crop culture and for the production of vegetables.

In these fields for protecting crops from insect/pests, and for increasing crop production formers used various chlorinated nd phosphorylated insecticides/pesticides and fertilizers. These chemicals and their remnants invade ponds when it rain. Flow off from from the fields enters directly into the ponds affecting living organisms.

Chlorinated compounds are known to increase pH where as phosphorylated compounds are known to decrease pH of waters(Russel E. Train, 1979.). A wide range of pH change also cause alterations in blood parameters (Mahdi Ghanbari & Mansoureh Jami, 2012).

Higher level of BOD results in depletion of available oxygen for aquatic organisms, while high amount of COD in effluent is toxic to biological life (Victoria A. Oriacu et al, 2011).

Small quantities of zinc are required for the normal development and metabolism, but if its level exceeds the physiological requirement , it can act as a toxicant(P.Gupta and N.Srivastava,2004).

The micronucleus assay is a simple and sensitive assay for evaluation of genotoxic properties of various agents. Since teleost erythrocytes are nucleated, micronucleus assay provides information about the measure of clastogenic activities (N.S. Nagpure et al, 2007).

In fishes chromosomes are small in size but large in numbers. Thus micronucleus in fishes could be smaller in size. The formation of micronucleus depends on the rate of proliferation of the cells, which in turn depends on fish species, environmental conditions and target tissues.

The micronuclei are small, extranuclear bodies that are formed during mitosis from accentric chromosomal fragments, or whole chromosomes that are not included in daughter nucleus.

Thus, micronucleus may contain a fragment of chromosome or it may contain a whole body of chromosome that is unable to travel to the spindle pole during anaphase.

2 Material and methods:

2.1- Water samples are collected from the ponds and physico-chemical properties of sample water are determined using standard analytical methods.

2.2- Blood were collected by caudal puncture. For this purpose plastic syringes of 1 ml capacity and needle (1.0 inch, 22 gauge) were used. The blood is directly drawn from caudal vein. Mixture of dry ammonium oxalate and potassium oxalate in the ratio of 3:2 is used as blood anti- coagulant.

2.3- Blood smears were made on the glass slides. Slides are air dried over night at room temperature in a dust free moisture free environment. Then they were fixed by dipping in absolute methanol for 10 minutes. Slides are then air dried for 1 hour. Then the slides are stained with 10% Giemsa stain for 30 minutes. Then they are washed for 3 times with double distilled water, so as to remove every Giemsa particle. Slides are dried overnight in dust and moisture free environment. Then the slides are mounted with DPX to make them permanent. These permanent blood slides are observed under the microscope with the help of the eye piece 10X and the objective lence of 100X. Drop of immersion oil is used.

3- Result:

The result of the physico-chemical analysis of water revealed higher values of most parameters (COD, BOD, Nitrates, Ammonia, Potassium, Phosphate, Sulphate, Chloride and Zinc), than the standards set by Environmental Protection Agency.

The pH value of the pond ranges between 7.00-9.00, through out

the study year.

Mean of the pH values of the ponds is 8.00

Figure 1 shows normal RBCs., and nucleus in the blood obtained from normal fish L. rohita.

Figure 2 shows formation of neutrophils, formation of thrombocytes, highly increased number of lymphocytes, and development of macrophages.

Figure 3 shows micronuclei, binuceated cells, lobed nucleus.

4- Conclusion:

The results obtained from this study indicates that the pond water is highly polluted and it have genotoxic potential. Nuclear anomalies and formation of micronucleus in blood erythrocytes of the fish L. rohita were identified as good genotoxic biomarker.

5- References:

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6-Figures:

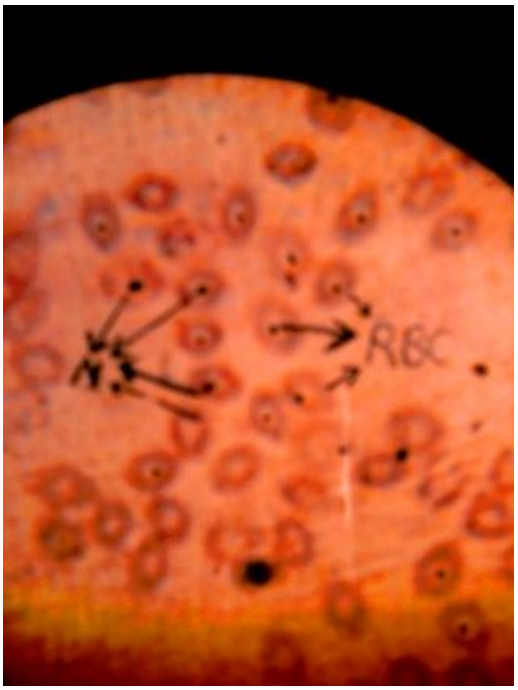

Figure-1 Showing RBc (Red blood corpuscles) and N(nucleus of RBcs.) of fish blood.

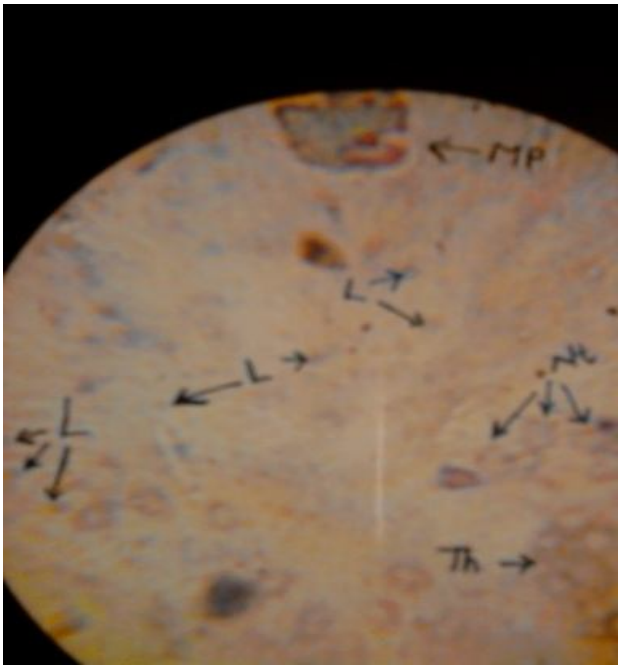

Figure-2 showing Mp (Macrophages), L (Lymphocytes), Nt (Neutrophils), Th (Thrombocytes) in fish blood.

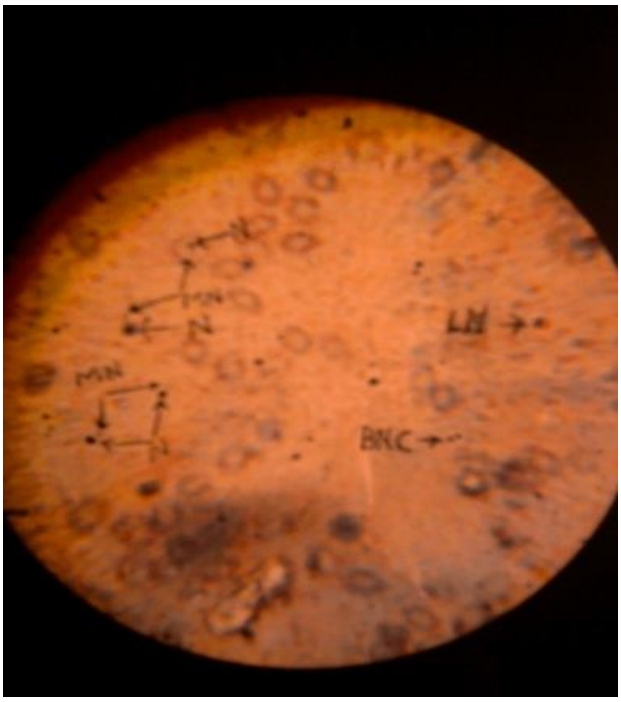

Figure- 3 showing bi-nucleated cells(BNC), nucleus(N), micronucleus(MN), lobed nucleus(LN).