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Haematological response of the African catfish, *Clarias gariepinus* (Clariidae) exposed to manganese

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Abstract

African catfish, *Clarias gariepinus* were exposed to two sub-lethal concentrations of manganese (T_1=6.48mg/l and T_2= 8.64mg/l) for 144-hours and the blood samples were collected at 24h intervals to determine the effects of the exposure on the haematological parameters. Packed cell volume (PCV), haematocrit (Hb), red blood corpuscles (RBC) and white blood cell (WBC) values increased significantly while erythrocyte sedimentation rate (ESR) and lymphocyte count showed a drastic decline compared with the control. The short-term manganese exposure had no significant effect on mean cell volume (MCV), mean cell haematocrit (MCH), mean cell haemoglobin concentration (MCHC), and Neutrophil values. There was no significant difference in the blood response in the two concentrations. The study showed that short-term exposure of *Clarias gariepinus* to sub-lethal concentrations of manganese caused haematological changes.

INTRODUCTION

Manganese is an essential nutrient for optimal cellular function and is known to cause neuro-toxicity by increasing oxidative stress and also disturbing neurotransmitter metabolism [1]. Manganese is typically used in the manufacture of steel, stainless steel and other metal alloys. Individuals may be exposed to manganese dust by inhalation or to manganese salts dissolved in water. Manganese compounds exist naturally in the environment as solids in the soils and small particles in the water. Manganese concentration is enhanced in the air by anthropogenic activities such as industrial activities and through burning of fossil fuels. Manganese that derives from human sources can also enter surface water, ground water and sewage water. Manganese is one of the three essential trace elements, which is not only necessary for human survival but also toxic when high concentrations are present in the human body. Manganese effects occur mainly in the respiratory tract and in the brain. Symptoms of manganese poisoning in man include hallucinations, forgetfulness and nerve damage[2]. Manganese can also cause Parkinson disease, lung embolism and bronchitis [3]. Manganese substances have been reported to cause lung, liver and vascular disturbances, decline in blood pressure, failure in development of animal foetus and brain damage[4]. Information on haematological effects of tropical freshwater fish species to heavy metal exposure is [5]. Haematological changes in fishes depend on fish species, age, physiological stress and diseases [6]. However, the indicators of blood reveal the effect of pollutants on fishes and forecast the consequences of long-term exposure to chemical[4]. This study elucidates the blood response of the African catfish *Clarias gariepinus* to 144-hour exposure to manganese.

Materials and Methods

Forty-eight sub-adults African catfish *Clarias gariepinus* Clariidae (mean body length 200±0.9 mm and mean body weight 500±1.5g) were obtained from a local fish farm and transported in an oxygenated polythene bag half-filled with pond water. the specimens were divided into eight experimental aquaria containing fresh water (pH 7.7) and were fed daily with pelleted floating feed and allowed to acclimatize for 14 days before the commencement of the experiment. Manganese (Mn) was introduced directly into the water in form of manganese sulphate ($MnSO_4$). Two treatments viz: T_1 (6.76mg/l $MnSO_4$) and T_2 (8.45mg/l $MnSO_4$) were used based on [7] guidelines with each treatment having three replicates. Two aquaria served as the control.

Two fish specimens were randomly selected from each tank at 24-hour interval. Blood sample was collected with the use of a 5 ml plastic syringe and a No. 21 swg needle. The needle slightly aspirated was inserted at right angle to the ventral surface close to the genital area of the fish. The needle was pushed gently down the flesh until it reached the vertebral column where the ventral aorta lies. 2ml of blood was drawn from the fish into a heparinised (EDTA) bottle and kept for further investigation. Standard haematological procedures [8]were employed in the assessment of the various blood parameters.

Results

The exposure of *C. gariepinus* to two concentrations of manganese (T_1=6.76mg/l and T_2=8.45mg/l) resulted in variations in some haematological parameters monitored in this study. Mean minimum-maximum values for each parameter are presented in Table1. There was an increase in the values of PCV, Hb, RBC, WBC and neutrophil from 24-hour to 144-hour exposure (figures 1-4, 10), ESR and Lymphocytes values decreased compared with the control (figures 5, 9) while MCV and MCH showed a slight increase after 24-hour exposure but dropped after 48-hour and only showed an increase in value after 144-hour (Fig. 6,7). MCHC gave a fairly regular pattern of increase in the treatment and adjustment to the control value at 24-hour interval (Fig. 8). There was no significant difference in the haematological responses of *C. gariepinus* to the two concentrations of manganese.

Discussion

The Mn-induced increment in the PCV, Hb, RBC, WBC and neutrophil values, compared with the control throughout the experimental period agrees with the earlier report of [9]who reported an increase in Hb and PCV concentrations after 96-hour

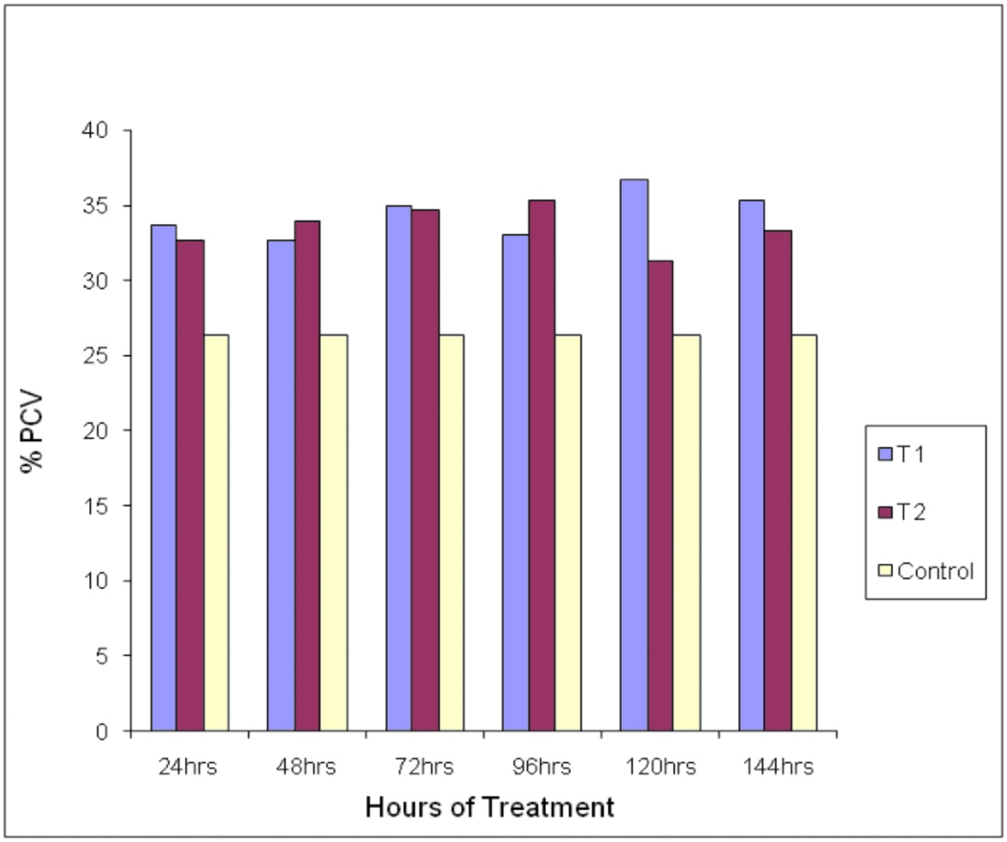

Figure 1. Variation in packed cell volume (%PCV) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

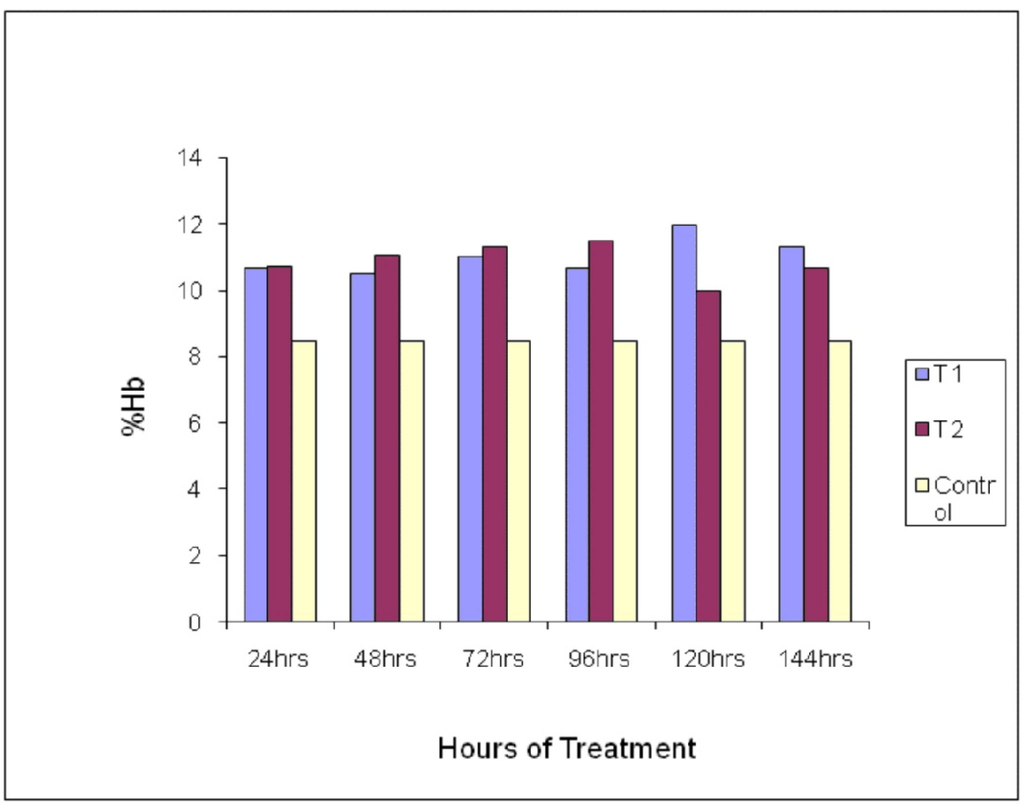

Figure 2. Variation in haemoglobin (%Hb) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

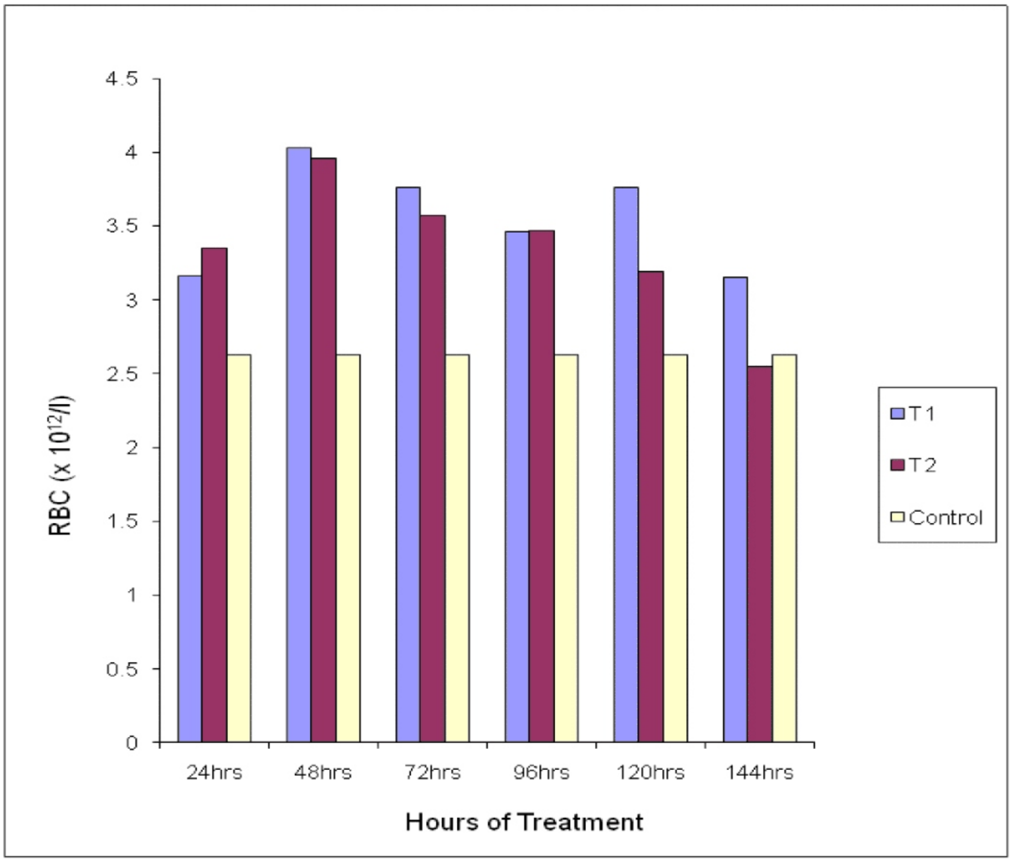

Figure 3. Variation in red blood corpuscles (RBC) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

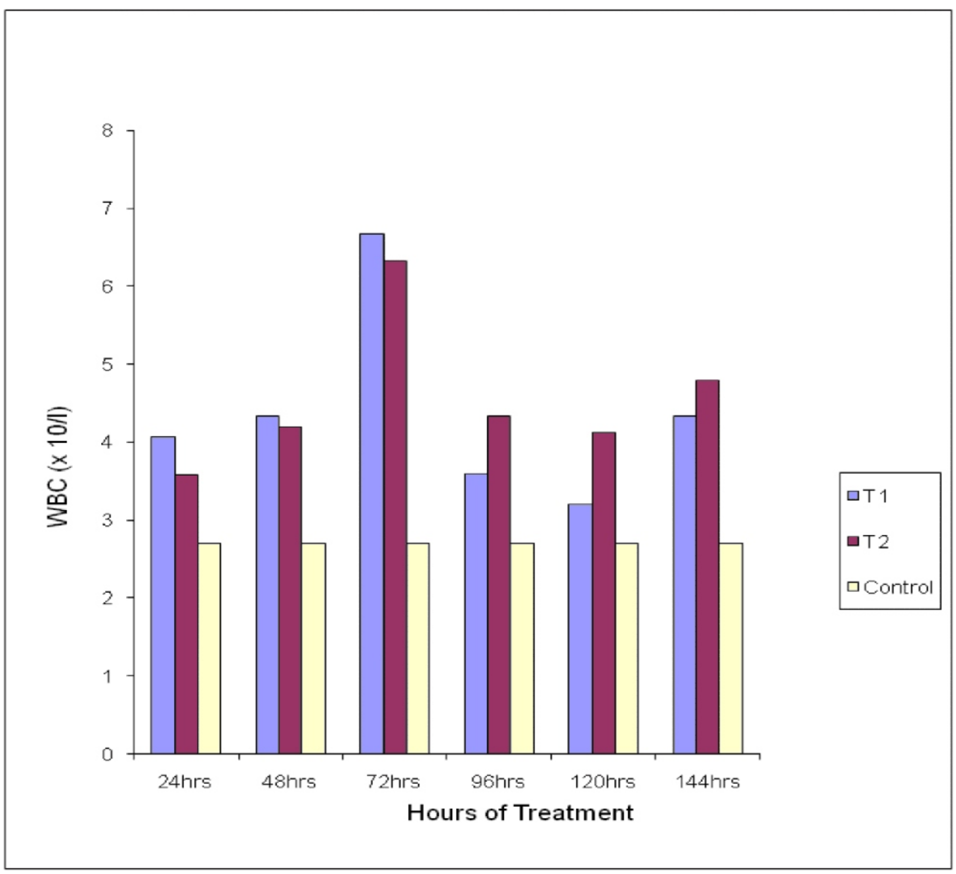

Figure 4. Variation in white blood cells (WBC) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

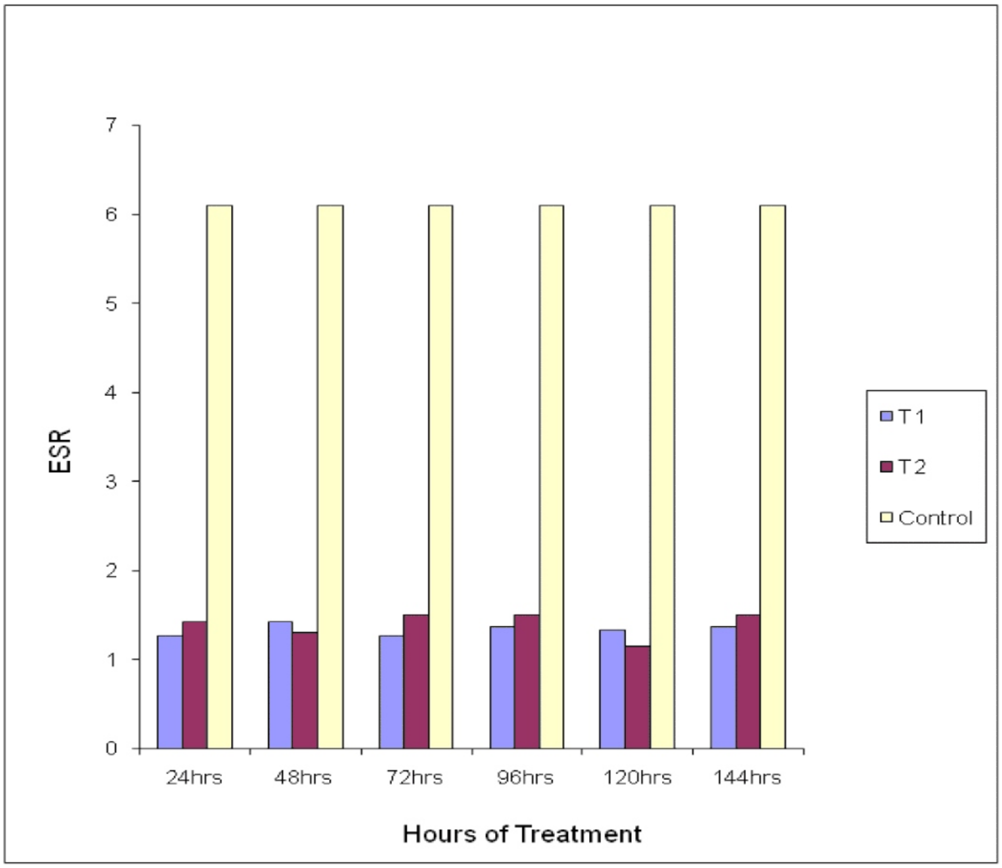

Figure 5. Variation in erythrocyte sedimentation rate (ESR) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

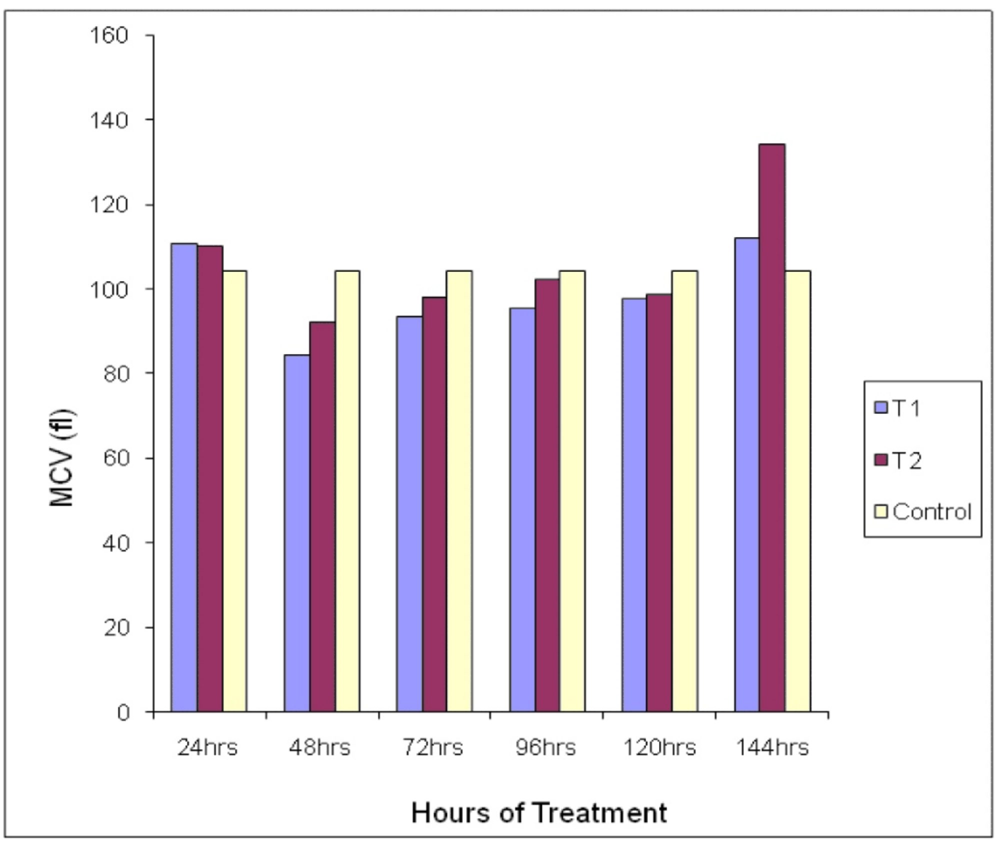

Figure 6. Variation in mean cell volume (MCV) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

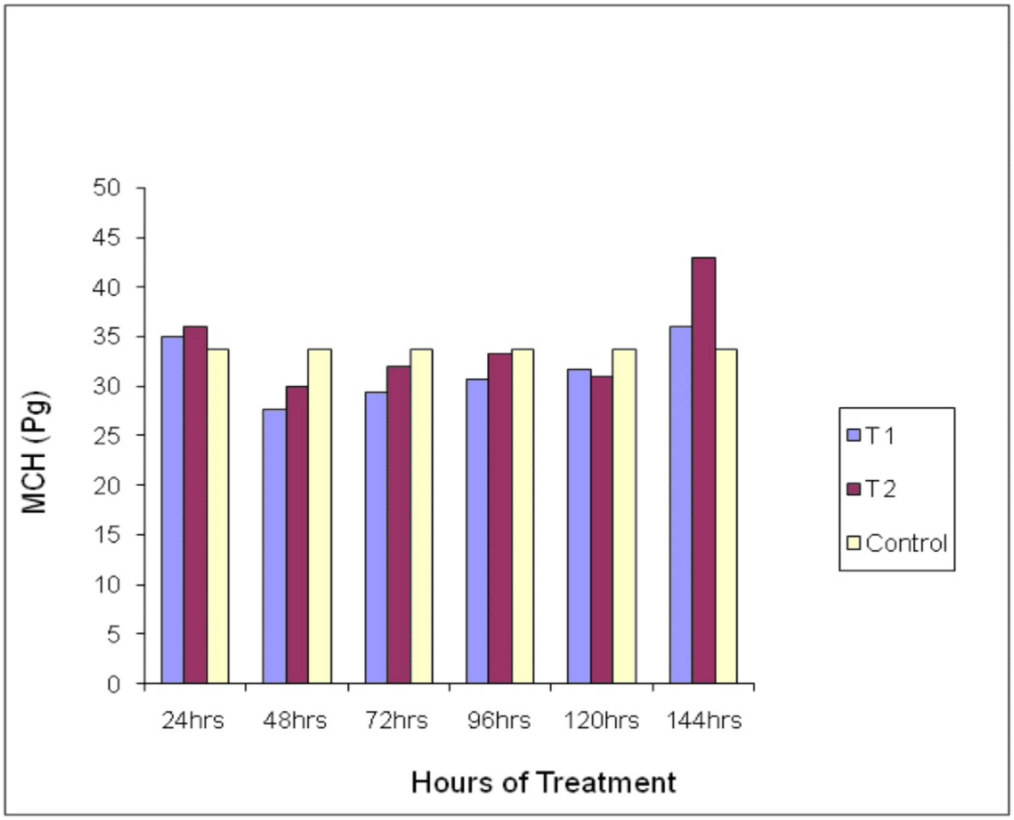

Figure 7. Variation in mean cell haemoglobin (MCH) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

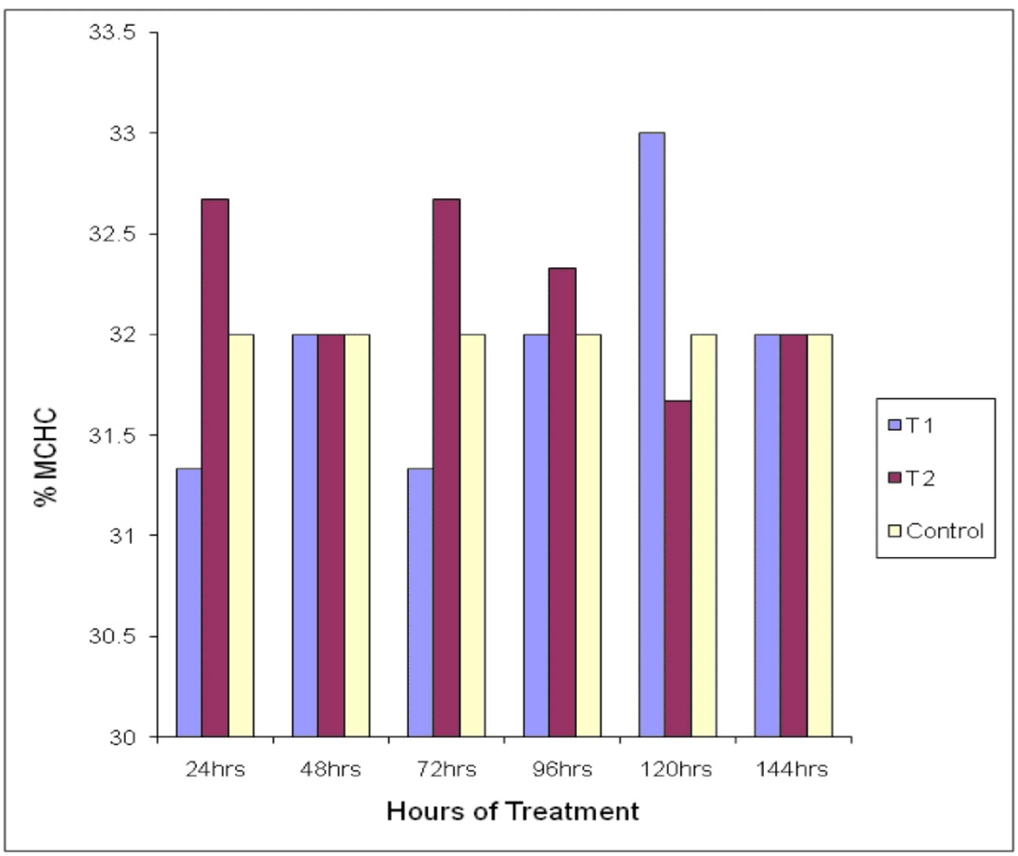

Figure 8. Variation in mean cell haemoglobin concentration (%MCHC) of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

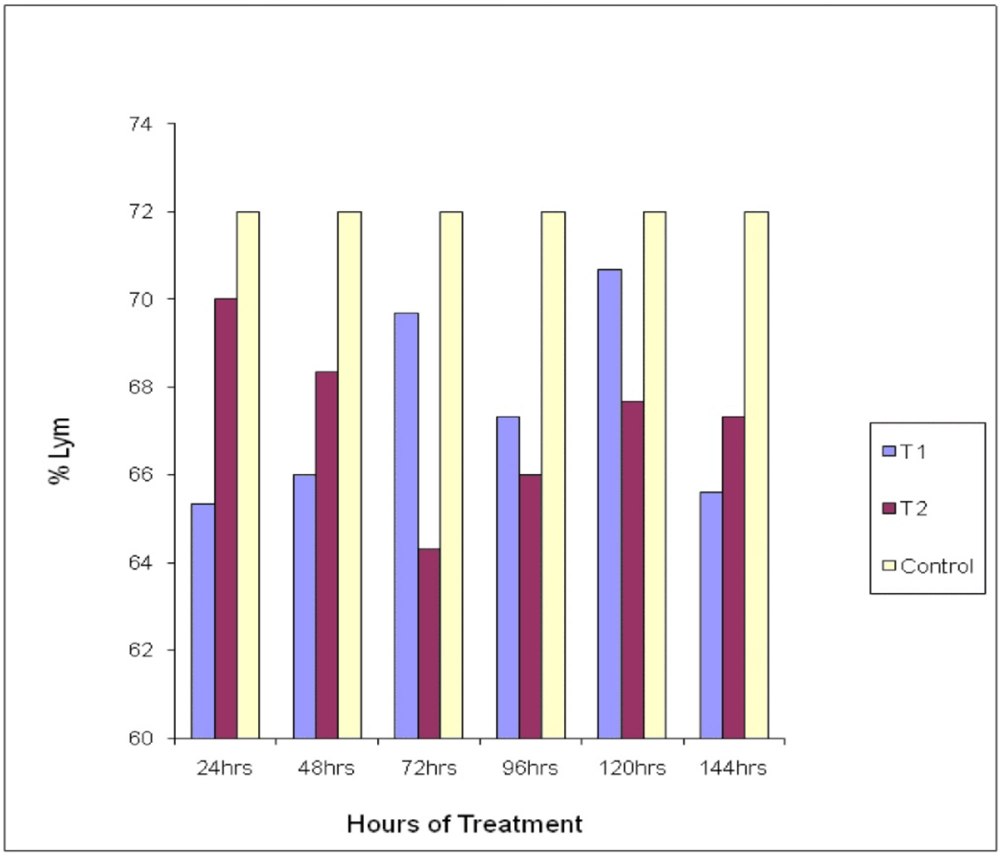

Figure 9. Variation in Lymnophil of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

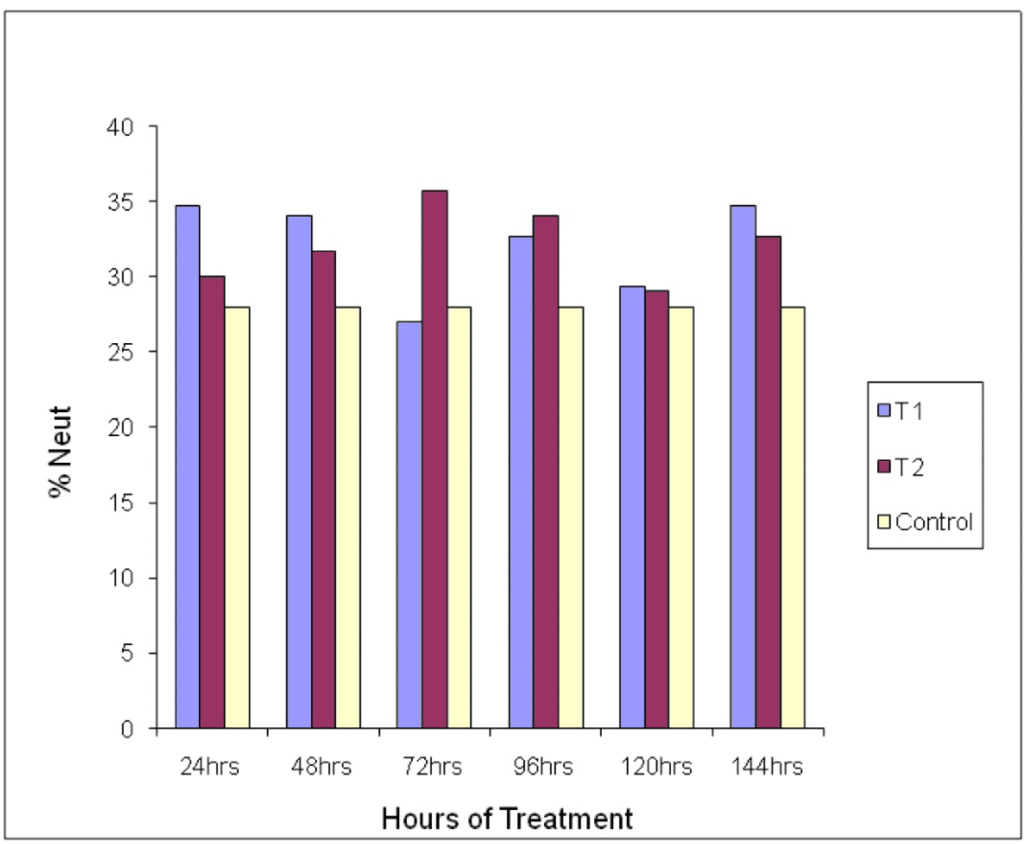

Figure 10. Variation in neutrophil of *Clarias gariepinus* exposed to two sub-lethal concentrations of manganese

exposure to lead. Lower concentration of haemoglobin to long-term exposure of trout to copper has been reported[10] . Other authors have reported a significant decrease in the erythrocyte count as well as haemoglobin concentration under acute 48-hour exposure of catfish *C. gariepinus* to background copper concentrations [11]. [12]reported haemolysis and anaemia in catfish (*C. lazera*) after 96-hour exposure to 3.2mg/l of copper while a decrease of erythrocytes in the blood of trout (*Salmo gairdneri*) after 24-hour exposure to 0.301mg/l of copper has been reported [13]. Studies have shown that there was an increase in erythrocyte, haemoglobin and haematocrit in fish exposed to various metals such as copper[9,14,15], cadmium[9,16,17], zinc [18,19] and chromium [14,20-23]. However, other authors have reported a significant decrease in erythrocytes as well as haemoglobin concentration in *C. gariepinus* under acute 48-hour exposure to background copper concentrations [11]. Fish stress reaction causes an osmotic imbalance and changes in the regulatory system of ionic interchange, which can diminish pH of blood and increase the volume of erythrocytes and subsequently the percentage of heamatocrit. Increase in the WBC count throughout the period of exposure showed the effort of the fish to protect the body against stressor agent. WBC also plays a role in inflammation and allergic reactions. The white cell count reached a crescendo after 72-hour exposure reflecting the immunological response of the body to the metal exposure. Contrary to reports of workers [13,14] who reported a decrease in WBC count on treatment with various metals, treatment of *C. gariepinus* with manganese was found to induce an increase in WBC count, a report supported by[8]. They however noted that a decrease in leukocyte count following an acute metal exposure is rather a non-specific stress reaction caused by a metal-induced stimulation of kidney chromaffine cells and cortisol secretion than a specific toxic action of metals upon the cells. An increase in leucocyte count has been reported for fish under short-term exposure [11] and long-term exposure to copper [10,24,25] while [13]reported a decrease in leucocytes due to decrease in count of lymphocytes and decrease in Neutrophil. A 2-hour exposure to copper led to increase in leucocyte count while long-term exposure led to decrease in copper. Increase in leucocyte was reported in fish exposed to cadmium [26] while a decrease in leucocyte was reported by[27-29]. Lymphocyte count showed consistent reduction after 24-hour exposure when the fish tried to restore disturbed homeostasis. The sustained leukocyte count showed the ability of the immune system to adjust to the stress agent. Lymphocytes are the most numerous cells compared to leucocytes, which function in the production of antibodies and chemical substances serving as body defense against infection. The primary consequence following changes in leucocytes count in stressed fish is suppression of the immune system and increased susceptibility to disease[30]. Decrease in Lymphocyte value under similar conditions has been reported by[31-34]. [35] reported that heavy metal exposure caused an increased cortisol level in fish, which was responsible for a decrease in WBC, particularly lymphocyte count and their activities. For MCV, macrocytosis, a value higher than the normal range was observed at 24-hour and 144-hour exposure while microcytosis, an indication of a value lower than the normal range was observed on 48-hour, 72-hour, 96-hour and 120-hour respectively. A short-term exposure of low concentration of heavy metal mostly induces an increase of these haematological indices.

[14] reported a decrease in the count of lymphocytes with an increase in the amount of neutrophils while it decreased with chromium[23].

Conclusion

The effect of short-term exposure of *C. gariepinus* to manganese was investigated and the result presented in this study. The importance of the effects of heavy metals such as Mn on aquatic biota cannot be over-emphasized since the aquatic organisms such as fish take up metals in the water and serves as a transportation route to man through ingestion. The results of this study provides information on the physiological disturbances that can arise as a result of exposure to manganese when fish is exposed to levels higher than required for normal physiological functioning of the fish body. Since manganese has been incriminated in the cause of diseases such as Parkinson, lung embolism and bronchitis in man, its effects on fish physiology especially blood is very crucial to the field of toxicology. With further research in this area, haematological responses of fish to metals may serve as a bio-indicator of metal contamination in the aquatic environment.

References

1. Erikson, K.M,, Dobson, A.W, Dorman, D.C. and Aschner, M., Manganese exposure and induced oxidative stress in the rat brain. *Sci. & Tot. Environ*. 334-335. 409-416. 2004.

2. Verity,M.A., Manganese neurotoxicity; a mechanistic hypothesis. *Neurotoxicity*. 20 (2-3); 489-497. 1999.

3. McMillan, D.E., A brief history of the neurobehavioral toxicity of manganese. *Neurotoxicity*, 20; 499-507. 1999.

4. Vosyliene, M.Z., Haematological parameters of rainbow trout (*Oncorhynchus mykiss*) during short-term exposure to copper. *Ekologija* 3:12-18. 1999.

5. Etim, L., Ekanem, S.B. and Uti, N.A., Haematological profile of two species of catfish, *Chrysichthys nigrodigitatus* (Lacepede) and *Chrysichthys furcatus* (Gruther) from the Great Kwa River, Nigeria. *Global Journal of Pure and Applied Science.* 5(1):1-4. 1999.

6. Luskova, V., Annual cycles and normal values of hematological parameter in fishs. *Acta Sc. Nat. Brno*.31c(5): 70. 1997.

7. USEPA (United States Environmental Protection Agency) Assessment and Control of Bio-concentration Contaminants in Surface Waters. Office of Health and Environmental Assessment, US Environmental Protection Agency. Cicinnati, Ohio. 1991.

8. Blaxhall, P.C. and Daisley, K.W., Routine haematological methods for use with fish blood. *J. Fish Biol.* 5 : 771-781. 1973.

9. Mishra, S. and Srivastava, A.K., The acute toxic effect of copper on the blood of the teleost. *Ecotoxicol. Environ. Saf.* 4:191-194. 1980.

10. Vosyliene, M.Z., The effect of heavy metals on haematological indices of fish (Survey). *Acta Zoologica Lithuanica. Hydrobiologia.* 9(2):76-82. 1996.

11. Van Vuren, H., Van Der Merwe, M. and Du Preez, H.H., The effect of copper on the blood chemistry of *Clarias gariepinus* (Clariidae). *Ecotoxicol.* 189-196. 1994.

12. El-Doriaty, N.A., Stress response of juvenile *Clarias lazera* elicited by copper. *Comp. Bioch. And Physiol.* C88 (2): 259-262. 1987.

13.] Dick P.T and Dixon D.G., Changes in circulating blood cell levels of rainbow trout, (*Salmo gairdneri*) Richadson, following acute and chronic exposure to copper. *J. Fish Biol.* 26;475-481. 1985.[14] Svobodova, Z., Vykusova, B. and Machova, J., Sub-lethal chronic effects of pollutants on freshwater fish. Ed. R. Muller Jr R. Lloyd. Lugano, 39-52pp. 1994.

15. Singh, H.S. and Reddy, T.V., Effect of copper sulfate on hematology, blood chemistry and hepato-somatic index of an Indian catfish, *Heteropneustes fossilis* (Bloch), and its recovery. *Ecotoxicol. Environ. Saf.* 20: 30-35. 1990.

16. Witeska, M., and Jiezerska, B., The effect of cadmium and lead on selected blood parameters of common carp. *Arch. Ryb. Pol.* 2(1):123-132. 1994.

17. Witeska, M., Changes in selected blood indices of common carp after acute exposure to cadmium. *Acta ver.Brno.* 67:289-293. 1998.

18. Tishinova, V. and Illieva, N., *Zoology* 85:111-123 (In Bulgarian). 1994.

19. Hilmy, A.M., El-Doriaty, N.A., Daabees, A.Y., and Abdel Latife, H.A., Some physiological and biochemical indices of zinc toxicity in two freshwater fishes, *Clarias lazera* and *Tilapia zilli. Comp. Biochem. Physiol.* 87C:297-301. 1987.

20. Van der Putte, I., van der Galien, W. and Strik, J.J.T.W.A., Effects of hexavalent chromium in rainbow trout (*Salmo gairdneri*) after prolonged exposure of two different pH levels. *Ecotoxicol. Environ. Saf.* 6: 246-257. 1982.

21. Gill, T.S. and Part, J.C., Haematological and pathological effects of chromium toxicosis in freshwater fish *Barbus conchonius* Ham. *Water Air Soil Pollut.* 35:241-250. 1987.

22. Strik, J.J.T.W.A., de Longh, H.H., van Rijn, van Alkenade, J.W.A. and Wuite, T.P., Toxicity of chromium(VI) in fish, with special reference to organ weights, liver and plasma enzyme activities, blood parameters and histological alterations. In. Koeman, J.H. and Strik, J.J.T.W.A., eds. *Sublethal Effects of Toxic Chemicals on Aquatic Animals.* Elsevier, Amsterdam, The Netherlands, 31-41. 1975.

23. Al-Akel, A. S., & Shamsi, M. J .K., Hexavalent chromium: toxicity and impact on carbohydrate metabolism and haematological parameters of carp (*Cyprinus carpio* L.) from Saudi Arabia. Aquat. Sci. 58: 24-30. 1996.

24. Dethloff, G.M. & Daisley, H.C., Effects of copper on immune system parameters of rainbow trout, (*Onchrhyncus mykiss*). *Environ. Toxicol. Chem.* 1(9): 1807-1814. 1998.

25. Dethloff, G.M, Schlenk, D. Khan, S. and Bailey, H.C., The effect of copper on blood and biochemical parameters of rainbow trout (*Oncorhynchus mykiss*). *Arch. Environ. Contam. Toxicol.* 36: 415-423. 1999.

26. Tort, L. and Hernandes-Pascal, M.D., Haematological effects in dogfish (*Scyliorhinus canicula*) after short-term sub-lethal cadmium exposure. *Acta hydrochem. Hydrobiol. 18: 379-383.* 1990.

27. Palackova, J., Pravda, D., Fasaic, K. and Celekchovska, O., Sub-lethal effects of cadmium on carp (*Cyprinus carpio*) fingerlings. Sub-lethal chronic effects of pollutants on freshwater fish. Ed. R. Muller and R. Lloyd. Lugano, 53-58. 1994.

28. Johansson-Sjobeck, M.L. and Larsson, A., The effects of cadmium on the haematology and on the activatity of delta-aminolevulic acid dehydratase (ALA-D) in blood and haematopoietic tissues of the flounder *Pleuronectes flessus. Environ. Res.* 17: 1991-2004. 1978.

29. Ruparella, S.G., Verma, Y., Saiyed, S.R. and Rawal, U.M., Effect of cadmium on blood of tilapia *Oreochromis mossambicus* (Peters) during prolonged exposure. *Environ. Contam. Toxicol.* 45: 305-312. 1990.

30. Wedemeyer, G.A. and Wood, J., *Stress: A predisposing factor in disease* .U.S Fish / Wildlife Service Fish Diseases leafet.399Pp. 1974.

31. Jiezerska, B. and Witeska, M., Metal toxicity of fish Wydawnictwo Akademin Podlaskiej,Siedlce. 2001.

32. Dunier, M. and Siwicki, A.K., Study on the effect of pollutant on fish defense mechanism I. *In vitro* influence of heavy metals on the spleen and kidney lymphocytes and macrophages activities in the carp (*Cyprinus carpio*). A*rch. Pol. Fish*. 2:55-66. 1994.

33. Siwicki, A.K., Studnicka, M., Glabski, E., and Kazun, K., Wplyw metallic Ciezkich na. Proliferacje limfocytow I aktyrwmosc fagocytarna makrofagow w badaniach in vitro u ryb. *Immunologia Polska* 19:32 (In Polish). 1994.

34. Viola, A., Pregnolato, G. and Albergoni, V., Effect of *in vitro* cadmium exposure on natural killer (NK) cells of catfish, *Ictalurus melas*. *Fish & Shellfish Immunology.* 6:167-172. 1996.

35. Donaldson, E.M. and Dye, H.M., Corticosteroid concentration in sockeye salmon (*Oncorhynchus nerka*) exposed to low concentration of copper. *J. Fish Res. Body. Can. 32*: 533-539. 1975.