Nigerian Veterinary Journal Vol. 24 (1) 28-32

HAEMATOLOGICAL STUDIES OF *OREOCHROMIS NILOTICUS* EXPOSED TO PIG EFFLUENT

ONUOHA, GRACE C., NWOSU*, CHIKA I.. AND AMADI, J. N.

Department of Fisheries, Michael Okpara University of Agriculture
P.M.B.7267, Umuahia, Abia State

*National Veterinary Research Institute Laboratory Umudike
P.O. Box 2208 Umuahia Abia State

SUMMARY

Studies on the haematological response of the fresh water fish *Oreochromis niloticus* exposed to pig effluent were conducted in 1998 to determine the effect of various concentrations of the effluent on haemoglobin (Hb), haematocrit (Ht), red and white blood cells (RBC and WBC). It was observed that as the concentration of the effluent increased beyond 25 ppt, the water become toxic resulting in hypoxia. There were increased fin/tail and opercular movements at high concentrations and low feed intake. The blood parameters initially increased but after the 25ppt concentration the value decreased significantly (P>0.05).

KEY WORDS: Haematology, *Oreochromis niloticus*, pig effluent, toxicology.

INTRODUCTION

Haematological analysis provides a quick screening method for assessing the health of a fish, as changes in the blood parameters are often quick response to environmental or physiological stress (Blaxhall, 1972, Mc Leavy, 1973, Hickey, 1976, Chakrabarty and Banerjee, 1988). The range of these stressful effects include alteration in feeding strategies, reduced swimming activity, and low rate of reproduction (Little *et al.*, 1985). Osmoregulation may be disturbed (Lewis and Lewis, 1971) and there may be respiratory distress (Hughes, 1981) or outright tissue damage may occur (Tort *et al.*, 1984; Tort and Torres, 1988., Annune *et al.*, 1994). Haematological parameters are easier to measure and can provide an integrated measure of the physiological status of any animal. Blood is involved in respiration, defense mechanisms and movement of nutrients and metabolites. Thus any change in the blood parameter will affect the efficiency of the fish.

Although Nigeria is an oil producing country, it cannot neglect agriculture. With an increase in population, and available land being fragmented into parcels for industrialization, many farmers have adopted an integrated approach. In an integrated farming system a combination of different farming practices are incorporated into one system as only one of several potential strategies, which can help alleviate hunger, employment and poverty. The objective of this study is to determine any changes in the haematological values of fish exposed to pig effluent.

MATERIALS AND METHOD

Live specimens of *Oreochromis niloticus* with total length of 16.40 to 17.5 with a mean length of 16.9cm ± 0.32cm weighing

164.4 to 198.8g with a mean weight of 181.8g ± 16.5g were obtained from the fish farm of the Michael Okpara University of Agriculture Umudike using a dragnet. The fish were held in a large glass tank with aeration using an aerator for acclimation (Plate 1).

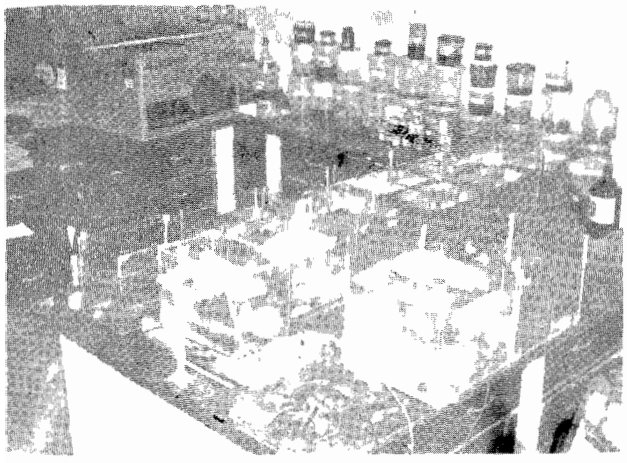

Plate 1: Acclimatization tank with aerator

Fish were fed with 25% crude protein feed, formulated in the University, for 2 weeks before the experiment. The fish were fed twice a day, in the morning and in the afternoon, at 5% biomass of the weight of fish samples. This therefore necessitated change of water every 2 days during the acclimation period. Fish were not fed during the test period. Eighteen glass aquaria of size 35x25x30 cm with 15 litres of borehole water were used in the experiments (Plate 2). A screening test was carried out in order to determine the various concentrations to be used. Based on this, five concentrations of 10ppt, 15ppt, 25ppt, 30ppt, and 40ppt respectively with 3 replications and control were then prepared from the stock solution. The stock solution was obtained by dissolving 100g of the solid (pig) effluent from the University piggery farm in 1000mls of water which gave a hundred parts per thousand (ppt) of effluent stock concentration.

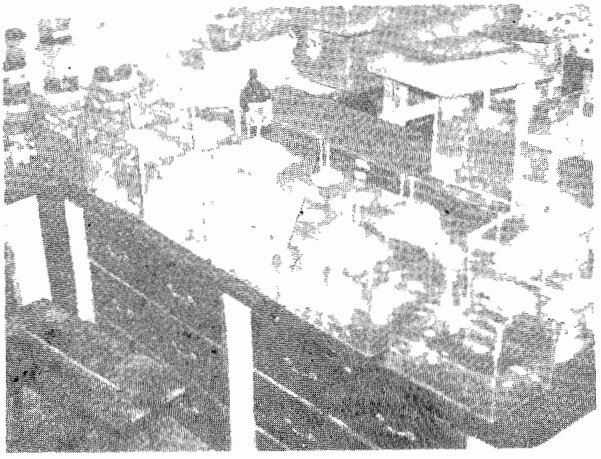

Plate 2: Experimental set up with glass aquaria and fish samples

Five acclimated fish were released into each aquarium containing different concentrations of the pig effluent as well as in the control. All tests were done at room temperature. Fish mortality at various intervals was recorded; also tail fin beat and opercular beat per minute of the fish were noted in each concentration.

The water was renewed every 2 days to maintain the strength of the effluent and the level of dissolved oxygen. The pH was taken twice a day. Blood samples were collected from the fish by puncture of the caudal artery. The haematocrit was measured in capillary tubes filled with blood and centrifuged in a haematocrity centrifuge for 15 minutes at 2000 RPM. The haemoglobin was measured with a spectrophotometer. The red blood cell count (RBCC) was performed with a Neubauer counter using Hayem's fluid (Conroy and Hermon, 1970) while the white blood cell count (WBCC) was carried out using Turk's solution as the diluent. The mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin

concentration (MCHC) were calculated according to Tort and Torres (1988) and Annune *et al*. (1994).

Statistical Analysis of variance (ANOVA) was used to determine the significance of mean difference of the various blood parameters for the different concentrations.

RESULTS AND DISCUSSIONS

The mean values of the haematological parameters obtained from the fish in the control and in the various concentrations of effluent are shown in Table I. Although the blood indices varied significantly ($P<0.05$) they did not follow any regular pattern.

Various results were obtained for the tail fin beat and opercular beat per minute for the different concentrations. Tail fin beat per minute was higher in the various concentrations than that of the control, although the increase at 10ppt and 15ppt concentrations were still within the range as compared with that of the control (20 beats per minute). Also the opercular beat was higher in the different concentrations than in the control.

The value of the control was found to be 22.5 beats per minute (Figs. 1 and 2).

TABLE I: Mean haematological values of *Oreochromis niloticus* exposed to pig effluent

Parameter	Control	10ppt	15ppt	25ppt	30ppt	40ppt
Haemoglobin (g/100ml)	6.7±0.09	6.81±1.83	7.93±1.33	8.46±1.25	6.32±0.18	5.26±1.25
Haematocrit (%)	20.3±0.26	20.43±1.65	26.19±1.98	28.08±1.28	19.96±2.65	14.78±0.15
RBC (cell mm^{-1} x10^{-3})	730.15±1.32	752.32±0.1	832.04±0.1	964.32±0.32	132.32±1.34	620.31±0.11
WBC (cell mm^{-1} x10^{-})	46.4±1.03	48.36±1.33	48.31±0.32	52.35±1.32	39.36±0.32	24.3±0.03
MCV (mm^{-3})	279±1.06	271.68±1.01	314.77±1.41	291.19±0.41	247.85±0.5	231.19±0.32
MCH (pg)	92.6±0.31	90.61±0.45	95.30±0.73	87.73±6.34	78.48±3.61	82.28±1.52
MCHC (g/100ml)	33.17±0.19	33.33±0.34	30.27±1.0	30.12±4.30	31.67±1.36	35.59±0.18

Abnormal behaviour such as restlessness, sudden quick movements, gulping of air, increased opercular activities, loss of balance and finally death were similar to the observations of Rahman *et al*. (2000). These behaviours suggested respiratory impairment, probably due to the effect of the effluent on the gills and general metabolism. This agrees with the findings of Kela *et al*. (1979), Duncan and Strock (1983) on their use of molluscidal formulations. Haematological values of fish have a relationship with energetic, respiratory and defense mechanism. The behaviours agree with the response of the haematologic parameters as shown in Table I.

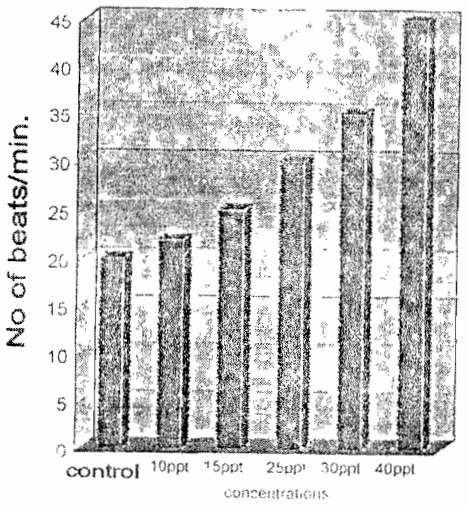

Fig. 1: Tail fin beats per min.

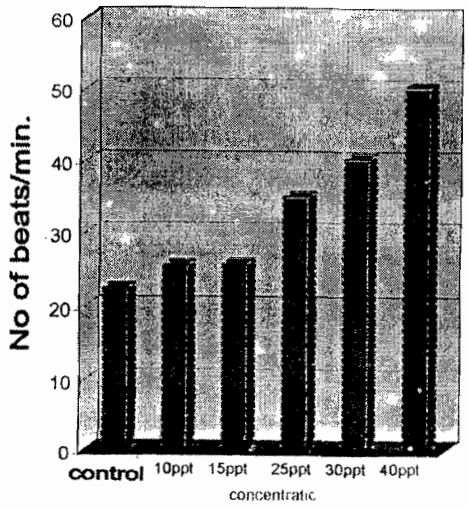

Fig. 2: Opercular beats per min.

Changes in the blood parameters often are quick response to environmental or physiological changes (Mc Leavy, 1973; Annune and Ahuma 1999). In this study, haematological values increased significantly ($P<0.05$) up to 25ppt but subsequently reduced ($P<0.05$). Fish fed on the pig effluent in lower concentrations had high blood values, but at higher concentrations of 30ppt and 40ppt the pig effluent was toxic and dissolved oxygen content was lowered. This led to hypoxia and severe stress and increased tail fin beat and opercular beat. Since the fish were stressed, they avoided feeding. Poor feeding will affect haemopoeisis, hence a low Hb, and RBC. The finding is in agreement with Smirnova (1979). Flos *et al.* (1987) reported that the feeding pattern of fish decreased following intoxication. Joshi (1980) suggested that developing gonads through a gradual decreasing space in the abdominal cavity depletes feeding in female fish. The pig effluent contained a lot of protein, which enhanced the haematological values initially (up to 25ppt). On inquiry, it was noted that the pigs were fed mainly on spent grain and soya-bean meal, which may have contributed to the effluent enriching the water.

In an integrated far of fish and pigs, it will therefore be necessary to determine the threshold value of the pig effluent so that the fish will not be intoxicated by excess effluent.

REFERENCES

ANNUNE, P.A, IYANIWURA, T.T., EBELLE, S.T., and OLADIMEJI, A.A., (1994). Effects of sublethal concentrations of zinc on haematological parameters of freshwater fishes, *Clarias gariepinus* (Burch) and *Orechromis niloticus* (Trewavas). *J. Aquatic sci.* **9**: 1-6.

ANNUNE, P.A. and AHUMA, F.T.A. (1999). Haematological changes in mud fish, *Clarias gariepinus* (BURCH), exposed to sublethal concentrations of copper and lead. *J. Aquatic Sci.* **13**: 33-36.

BLAXHALL, P.C (1973). The haematological assessment of the health of freshwater fish. A review of selected literature. *J. Fish. Biol.*. **4**: 593-604.

CHAKRABARTY, P. and BANERJEE, V. (1988). Effect of sublethal toxicity of three organophosphorus pesticides on the peripheral haemogram of the fish *Channa punctatus*. *Environ. Ecol.* **6**: 151-158

CONROY, A. and HERMAN, P. (1970). Fish diseases. Oxford University Press. pp. 280-302.

DUNCAN, J. and STROCK, R. F. (1983). Laboratory evaluation of the plant molluscicides. In: Plant Molluscicides (K.E. Mott, ed.)

UNDP/World Bank/WHO Report. 289pp

HICKEY, C.R. (1976). Fish haematology, its uses and significance. *N. Y. Fish Game J.* **23**: 170-285.

HUGHES, G.M. (1981). Effects of low oxygen and pollution on the respiratory systems of fish. In: Stress and Fish (A.D. Pickering ed.). Academic Press, London. pp 121-146.

JOSHI, B.D. (1980). Sex related variation in some haematological values of certain freshwater teleosts. *Japanese J. Ichthyology*. **27**: 139-143.

KELA, S.L., OGUNSUSI, R.A., OGBOGULI, I. and NWUDE, N. (1979). Susceptibility of two weeks old *Lymnaea natalensis* to some plant extracts. *Revue Lt. Med. Vet. Phys. Trop*. **42**: 286-287.

LEWIS, S.D. and LEWIS, W.M. (1971). The effect of zinc and copper on osmolality of blood serum of channel catfish, *Ictalurus punctatus. Trans. Am. Fish. Soc*. **4**: 639-643.

LITTLE, E.E., FLEROV, B.A. and RUZHINSKAYA, N.N. (1985). Behavioural approaches in aquatic toxicity investigations – a review. In: Toxic substances in the aquatic environment. *Am. Fisheries Soc.* Water Quality Section, Bethesda, MD. Pp. 72-98.

MC LEAVY, D.L. (1973). Effects of 12hr. and 25days exposure to kraft pulpmill effluent on the blood and tissue of juvenile Coho Salmon. *J. Fish Res. Bd. Canada*. **30**: 395-400.

MISHRA, S. and SRIVASTAVA, A.K. (1979). Haematology as an index of sublethal toxicity of zinc in freshwater teleost. *Bull. Environ. Con. Toxicol*. **22**: 695-698.

RAHMAN, M.Z., HOSSAIN, Z., MOLLAH, M.F.A. and AHMED, G.U. (2002). Effect of Diazinon 60 EC on *Anabas testudineus, Channa punctatus and Barbodes gonionotus. Naga Iclam Quarterly*. **25**: 8-12.

SMIRNOVA, L.I. (1965). Blood indices of barbot during prolonged total fasting and subsequent feedings. *Proc. Acad. Scient. USSR*. **190**: 170-175.

TORT, L., HODSON, P.V. and SPRY, D.J. (1984). Resistance to zinc sulphate of zebra fish Brachydanio rerio at different phases of its life history. *Ann. Appl. Biol*. **56**: 47-53.

TORT, L. and TORRES, P. (1988). The effect of sublethal concentration pf cadmium on haematological parameters of dogfish, *Scylorhynus canicula. J. Fish Biol*. **32**: 227-282.