Available online at http://www.journalcra.com

International Journal of Current Research
Vol.2, Issue, 1, pp.176-180, January, 2011

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

IMPACT OF PESTICIDE CYPERMETHRIN ON HAEMATOLOGICAL ALTERATIONS IN THE FRESHWATER FISH *OREOCHROMIS MOSSAMBICUS*

***Kumaresan, T.**

Department of Zoology, Annamalai University, Tamil Nadu, India

ARTICLE INFO

Article History:
Received 7th October, 2010
Received in revised form
14th November, 2010
Accepted 30th December, 2010
Published online 30th January, 2011

Key words:
Fish Oreochromis mossambicus, Cypermethrin, Haematology, RBC, WBC, Hb, MCH.

ABSTRACT

Pesticide is common pollutants of freshwater ecosystems where they induce adverse effects on the aquatic biota. Fish, *Oreochromis mossambicus* is an important carp species in Tamil Nadu region having good nutritional values. Fishes living in close association with may accumulate pesticides. In the present observation, the toxic effects of pesticide cypermethrin LC_{50} 0.28 ppm on the total RBC, WBC, Hb and MCH in the fish, *Oreochromis mossambicus* were estimated. The sublethal concentrations of pesticide cypermethrin on (10% sublethal concentration and 20% sublethal concentration) showed a decreasing trend in the RBC, Hb and mean corpuscular haemoglobin (MCH) compared to controls and the WBC analysis revealed a significant increased compared to control for a period of 10, 20 and 30 days exposures. The results indicated the toxic nature of the sublethal concentrations pesticide cypermethrin.

INTRODUCTION

Cypermethrin is widely used against pests all over the world to increase the production of food grains and other agricultural-products (Usmani and Knowles, 2001) and there is increased risk of food being contaminated with the insecticide, which may harm humans and domesticated animals. Pesticides have been applied to fight against pests of plants, animals and humans. However, the introduction of pesticides to the natural environment has also some negative effects, including unintentional intoxication of useful insects, fish, birds, mammals, and other inhabitants of aquatic and terrestrial biocenoses (Senthil kumar *et al.,* 2001). Pesticides represent a relevant stressor for many aquatic and terrestrial species (Kale Monika *et al.,* 2006 and Leiss *et al.,* 2005b). The contamination affect all group of organisms in aquatic ecosystem like invertebrate (Castillo *et al.,* 2006) and non target aquatic biota like fishes (Prashanth and Neelagund, 2008).Cypermethrin is a synthetic pyrothroid pesticide that has been widely used over the past 30 year in India and other countries against pests, particularly Lepidoptera, cockroaches and termites. In animals, cypermethrin has been used as chemotherapeutic agent against ectoparasite infestations (Velisek *et al.,* 2006). Agricultural chemicals including nitrogen compounds, pesticides and brokedown products are commonly present in water bodies (Capel *et al.,* 2008). After use, cypermethrin is released directly into the environment, enters water sources and affects the aquatic ecosystem.

***Corresponding author:* **Kumaresan, T.,**

Large-scale population declines of many species of birds, mainly fish-eating and bird-eating species, have been attributed to exposure to insecticides through higher order food chain and upward biomagnification of residues (Muthu-viveganandavel *et al.,* 2008). The effect of major pesticides (cypermethrin and chlorpyrifos) used against soil insects of field and vegetable crops on circulatory fluids of a common Tilapine fish species *(Tilapia guineensis)* prevalent in the Niger Delta waters. Several ecotoxicological studies have implied that haematological changes such as increased levels of plasma enzymes occurred in some vertebrates after exposure to pesticides (Abou-Donia, 1990; Lapadula *et al.,* 1990). Blood is a pathological indicator of the whole body, and hence hematological parameters are important for analysis of the functional status of an exposed animal to suspected toxicant (Omitoyin, 2006). Moreover, blood cell profile has been considered as an important indicator of diseases and other toxicants. As pathomorphological changes are indicative of numerous diseases (Yawata, 2003) there is little doubt of the importance of elucidating the mechanism governing erythrocyte shape (Pawlowski *et al.,* 2006). In the present investigation, tested variations of haematological changes in fish (*Oreochromis mossambicus*) were exposed to pesticide cypermethrin.

MATERIALS AND METHODS

Fish, *Oreochromis mossambicus* was collected from Chidambaram area and were brought to the laboratory in large plastic troughs and acclimatized for one week. Healthy, fish having equal size (length 5 to 10 cm) and weight (50 to 100 g) were used for experimentation. Stock solution of cypermethrin was prepared by dissolving appropriate amount of salt in distilled water. The physico-chemical characteristic of test water have analyzed regularly during the test periods following the standard method describe by APHA (1998). Batches of 10 healthy fishes were exposed to different concentrations of pesticide cypermethrin to calculate the medium lethal concentration LC_{50} value (0.28 ppm) using probit analysis Finney method (1971). The fishes (Four groups) were exposed to the two sublethal concentrations ($1/10^{th}$ and $1/20^{th}$ mg/L) of cypermethrin for 10, 20 and 30 days respectively. Another group was maintained as control. Fish was collected and gently wiped with a dry cloth to remove water. Caudal peduncle was cut with a sharp blade and the blood was collected in a watch glass containing EDTA, an anticoagulant (6% Ethylene diamine tetra acetic acid). The blood was mixed well with the EDTA solution by using a needle and this sample was used for determining the Red Blood Corpuscle Count (RBC), White Blood Corpuscle Count (WBC), Haemoglobin count (HB) and Mean Corpuscular Haemoglobin(MCH).

RESULTS

Cypermethrin insecticide caused 50% mortality of fish *Oreochromis mossambicus* at 96 hours was 0.28 ppm. The LC_{50} values of Cypermethrin for 24, 48, 72 and 96 hours were 0.36, 0.34, 0.32 and 0.28 ppm respectively. The toxic effects of Cypermethrin is presented on the haematological parameters of *Oreochromis mossambicus* such as number of red blood corpuscles (RBC), white blood corpuscles (WBC), haemoglobin content (Hb)and mean corpuscular haemoglobin(MCH). The observations were made at the end of exposure periods 10, 20 and 30 days) to calculate the percentage of increase and decrease of different haematological parameters.

Red Blood Corpuscles (RBC): The sub lethal concentrations of cypermethrin on (10% and 20%) showed a decreasing trend in the RBC counts compared to controls (Table 1 and Fig. 1). The mean values of RBC in control fish were estimated to be 2.88 ± 0.38, 2.89 ± 0.4 and 2.86 ± 0.38 (106/mm3). The control fish, the maximum (2.89 ± 0.4) was recorded for 20 days. In the sublethal concentration (10%) values were recorded to be 2.79±0.45, 2.22 ± 0.27, and 1.84 ± 0.34 and the sublethal concentration (20%) values were recorded 2.5 ± 0.37, 2.12+0.33 and 1.68 ± 0.45 respectively.

White Blood Cells (WBC): The changes of haematological parameter like, WBC in the fish *Oreochromis mossambicus*, both in control as well as sublethal concentrations of cypermethrin exposed to (10% and 20%) for 10, 20, and 30 days (Table 1 and Fig 2). The WBC analysis revealed a significant increase compared to control fish. The White Blood Cells (WBC) count from 15.92 ± 0.58, 15.93 ± 0.4 and 15.82 ± 0.61 in control fish. The sublethal concentrations (10%) values were recorded from 16.67 ± 0.78, 18.18 ± 0.63, and 18.47 ± 0.53 and the sublethal concentrations (20%) values were recorded from 17.75 ± 0.63, 18.34 ± 0.30, 18.68 ± 0.39 respectively.

Haemoglobin (Hb): The changes of haematological parameter like Haemoglobin in the fish *Oreochromis mossambicus*, both in control as well as sublethal concentrations of cypermethrin exposed to (10% and 20%) for 10, 20 and 30 days (Table 1 and Fig.3). The haemoglobin analysis revealed a significant reduction compared to control fish. The haemoglobin count from 6.36 ± 0.68, 6.34 ± 0.75 and 6.33 ± 0.40 g/100ml in control fish. The control fish, the value maximum (6.36 ± 0.68) was recorded for 10 days. Sublethal concentration (10%) values were recorded from 6.26 ± 0.38, 5.78 ± 0.44, and 5.07 ± 0.47 and 20% sublethal concentration values were recorded from 6.02 ± 0.55, 5.5 ± 0.65 and 4.97 ± 0.35 respectively.

Table 1. The haematological parameters of fish *Oreochromis mossambicus* under sub lethal concentrations of cypermethrin.

Days	Exposure	RBC ($10^6/mm^3$)	WBC ($10^3/mm^3$)	Hb (g/100ml)	MCH (mg/dl).
10 days	Control	2.88 ± 0.38	15.92 ± 0.58	6.36 ± 0.68	22.35 ± 0.81
	10% SLC	2.79 ± 0.45	16.47 ± 0.78	6.26 ± 0.38	22.28 ± 0.50
	20% SLC	2.5 ± 0.37	17.75 ± 0.63	6.02 ± 0.55	21.85 ± 0.37
20 days	Control	2.89 ± 0.4	15.93 ± 0.4	6.34 ± 0.75	22.33 ± 0.49
	10% SLC	2.22 ± 0.27	18.18 ± 0.63	5.78 ± 0.44	21.59 ± 0.49
	20% SLC	2.12 ± 0.33	18.34 ± 0.30	5.5 ± 0.65	21.5 ± 0.73
30 days	Control	2.86 ± 0.38	15.82 ± 0.61	6.33 ± 0.40	22.38 ± 0.51
	10% SLC	1.84 ± 0.34	18.47 ± 0.53	5.07 ± 0.47	21.18 ± 0.77
	20% SLC	1.68 ± 0.45	18.68 ± 0.39	4.97 ± 0.35	20.65 ± 0.69

Values are mean ± SD – or + indicate present decrease or increase over control

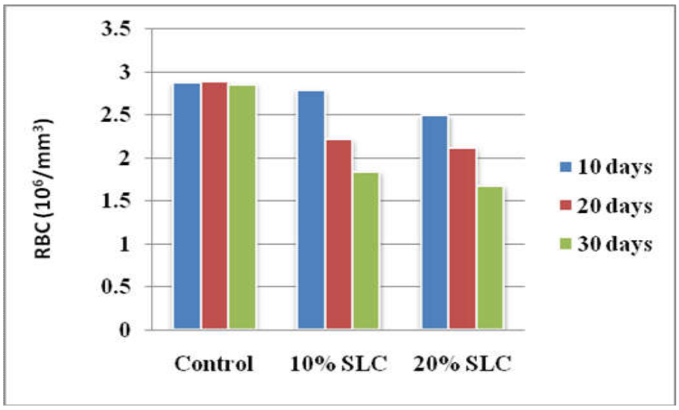

Fig. 1. The total RBC of *Oreochromis mossambicus* under sub lethal concentrations of cypermethrin

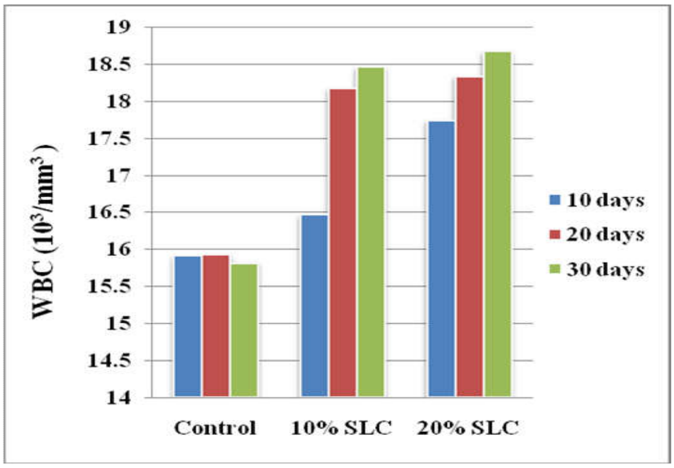

Fig. 2. Total WBC of *Oreochromis mossambicus* under sub lethal concentrations of cypermethrin

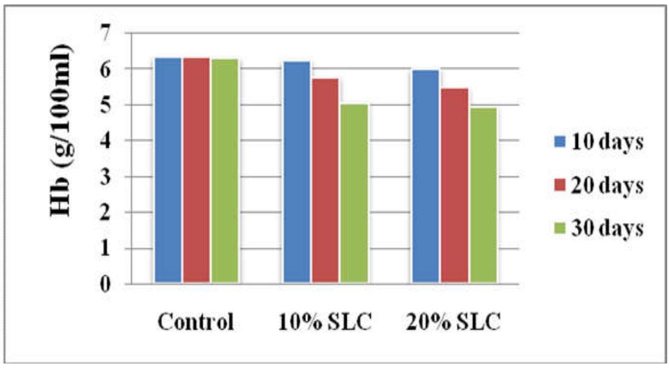

Fig. 3. The total Hb of *Oreochromis mossambicus* under sub lethal concentrations of cypermethrin

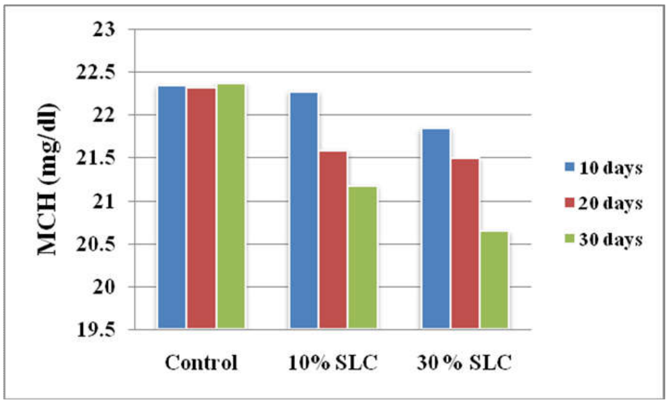

Fig. 4. Total MHC (mg/dl) of *Oreochromis mossambicus* under sub lethal concentrations of cypermethrin

Mean Corpuscular Haemoglobin (MCH): The changes of haematological parameter like, MCH in the fish *Oreochromis mossambicus*, both in control as well as sublethal concentrations of cypermethrin exposed to 10, 20, and 30 days (Table 1 and Fig. 4). The haematological analysis revealed a significant reduction compared to control, in MCH count from 22.35 ± 0.81, 22.33 ± 0.49 and 22.38 ± 0.51 (mg/dl) in control fish. The sublethal concentration (10%) values were recorded from 22.28 ± 0.50, 21.59 ± 0.49 and 21.18 ± 0.77 and the 20% sublethal concentration values were recorded from 21.85 ± 0.37, 21.5 ± 0.73 and 20.65 ± 0.69 (mg/dl).

DISCUSSION

In the present study LC_{50} values of cypermethrin of fish *Oreochromis mossambicus* at 96 hours was 0.28 ppm and sublethal concentrations namely 10% and 20% values were selected, studying their effects on various physiological, biochemical and histological aspects. During the acute toxicity tests, the fish were seen to exhibit several behavioural responses, such as fast jerking, frequently jumping, erratic swimming, spiraling, convulsions and tendency to escape from the aquaria. Following this state of hyper excitability, the fish became inactive and loss of orientation. There was loss of equilibrium and paralysis which ultimately resolved in death of the fish. These altered behavioral abnormalities were observed only at high concentration ranges (values higher than 96 h LC50). Rao *et al.,* 2005 reported that abnormal changes in behavior in mosquito fish *Gambusia affinis* in response to the sub-lethal exposure to chlorpyrifos. Acute toxicity of pesticides like Endosulfon, Malathion and Copper sulphate at different concentrations to fresh water prawns Macrobrachium rosenbergii were reported (Natarajan *et al.,* 1992). RBC to heamolytic crisis that results in severe anemia in fish exposed to heavy metals and herbicide respectively. Furthermore, the reduction of RBC also leads to development of hypoxic condition which in turn leads to increase in destruction of RBC or decreased in rate of formation of RBC due to non availability of HB content in cellular medium (Chen *et al.,* 2004). The damage of toxicant on erythrocyte may be secondary, resulting from a primary action of toxicant on erythropoietic tissues on which there exists a failure in red cell production and or due to increase in the erythorocyte destruction. These results are in affirmative agreement with that investigated by Wahbi *et al.,* (2004). A significant decrease in erythrocyte (RBC) counts, haemaglobin (HB), an increase of White Blood Corpuscles polluted waters can definitely be related to the pollution due to slaughter house wastes (Reo and Hymavathi, 2000). The reduction in RBC and HB, in *labeorohita* after exposure to arsenic trioxide has been suggested by Pazhanisamy and Indra, (2007). A point of interest noticed in the present investigation is the increase in WBC count after Rogor (Dimethoate) 30 per cent EC treatment. WBCs are involved in the adjustment of immunological work in many organisms and the saw increase in WBC count in mercury chloride treated fish shows a generalized immune reply and a defensive response to mercury chloride. Rousing effect of the toxicant on immune system and liberate of lymphocytes from lymphomyeloid tissue as a protection mechanism may also redound to expansion in WBC count in fish (Ates *et al.,* 2008). The decreased erythrocyte count in the experimental groups with increasing temperature and exposure period may be due to a decrease in the erythopoietic activity of the kidney as reported by Santhakumar *et al.,* (1999).

An increased in WBC suggested a compensatory erythropoiesis due to stimulatory effects. The enhancement of WBC in mice could be due to stimulated lymphoiesis and/or enhanced release of lymphocytes from lymphoid to tissues. The increase in lymphocyte number of intoxicated mice is also probably for the removal of cellular debris of necrosis tissue at a quicker rate (Mcleay and Brown, 1974). White blood cells (WBC) or leukocytes, are important component of the immune system involved in defending the body against both infectious diseases and foreign materials. Five different and diverse types of leukocytes exist, but they are all produced and derived from a multipotent cell in the bone marrow known as a hematopoietic stem cell. The increased total leucocytes count of the exposed fish in the present study indicates increased defensive reaction against the stressors. Increased WBC counts have also been reported in fish exposed to other certain xenobioties like endosulfan Abidi and Srivastova, (1988). Goel *et al.,* (1985) reported that the reduction in HB, RBC and PVC in Heteropenustes fossils exposed zinc. Similarly the decreased amount of RBC, HB, and PVC could be corroborated with previous investigations in *Oreochromismossambicus* exposed copper and zinc (Sampath *et al.,* 1998). Pamila *et al.,* (1990) have reported that the reduction in Hb content might be due to the inhibitory effect of toxic substance on the enzymes systems which involved in the synthesis of haemoglobin. Joshi *et al.,* (2002) have reported that heavy metal exposure decreased the RBC, Hb and PVC due to impaired intestinal absorption of iron. Karuppasamy *et* *al.,* (2005) observed that the significant decrease in Hb, RBC, PVC, MCHC in *Channa punctatus* exposed to cadmium. The haemoglobin (Hb) levels increased with increasing temperatures in the control group after 15 and 30 days of acclimation period, which may be due to the increase in oxygen demand as indicated by Verma *et al.,* (2007) in common carp *Cyprinus carpio*. In content, the decline in the chlorine treated groups may be due to the inhibitory effect of chlorine as stated by the above authors. The decrease in the haemoglobin content may be due to the inhibition of erythropoietin, chemosynthesis, osmoregulatory dysfunction ore due to an increase in the rate of erythrocyte destruction in the haematopoietic organ, which may lead to anemia in the exposed fish observed by Jenking *et al.,* (2003) in *Cyprinus carpio*.

Conclusion

The present study indicates that presence of low concentration of cypermethrin in the water is toxic to fishes and alters the haematological of the fish tissues. The results indicate that the usage of the cypermethrin in the agriculture fields may be a threat to aquatic fauna and flora as well as humans. Finally, concluded that the assured greater significance due to the increasing emphasis on fish culture and greater awareness of the pollution in aquatic ecosystem. Therefore, the information obtained may be useful for management and monitoring of agricultural insecticide contamination in aquatic ecosystem.

REFERENCES

Abidi, R., and Srivastava., U.S. 1988. Effect of endosulfan on certain aspects of hematology of the fish, Channa punctatus (Bloch). *Proc. Natl. Acad. Sci. India* 58: 55 – 65.

Abou-Donia, M.B., 1990. Absence of developed neurotoxicity and increased plasma butyryl-cholinesterase activity in triallate-treated hen. *Fundam. Appl. Toxic.,* 14, 191-198.

Adeyemo, O.K.., Agbede, S.A., Olaniyan A.O. and Shoaga, O.A.2003. The hematological response of *Clarias gariepinus* to changes in acclimation temperature. *Afr. J. Biomed Res*., 6: 105 – 108.

APHA, 1998. Standard methods for the examination of water and waste water, 20th Edition, Washington, DC.

Ates, B., Orun, I., Talas, Z.S., Durmaz, G., and Yilmaz, I., 2008. “Effects of sodium selenite on some biochemical and hematological parameters of rainbow trout (Oncorhynchus mykiss walbaum, 1792). Exposed to Pb^{2+} and Cu^{2+}”. *Fish physiology and Biochemistry,* 24: 53 – 59.

Capel, P.D., Mccarthy, K.A., and Barbash, J.E., 2008. National,holistic, water-shed approach to understand the sources, transport ans fate of agricultural chemicals. *J. Environ. Quality,* 37:983-993.

Castillo, LE., Martinez, E., Ruepert, C., Savage, C., Gilek, M., Pinnock, M. and Solis, E., 2006. Water quality and microinvertebrate community response following pesticide applications in a banana plantation Limon costa Rica. *Science.Total Environ.*, 367:418-432.

Chen, X., Yin, D., Hu, S. and Hou, 2004. “Immunotoxicty of penta chlorophenol on macrophage immunity and IgM secretion of the crucian carp (*Carassius auratus), J. Bull. Environ. Contam and Toxicol*., Vol. 73. PP. 153 – 160.

Finney, D.J. 1971. Probit analysis, 3rd (Ed.), Cambridge University Press, London, 333.

Goel, K.A., and G. Kalpana, 1985. Haematological characteristics of *Heteropneustes fossilis* under the stress of zinc. *Indian. J. Fish,* 36, 256 – 259.

Jenkins, F., Smith, J., Rajanna, B., Shameen, U., Umadevi, K., Sandhya, V., Madhavi, R. 2003. Effect of sublethal concentrations of endosulfan on hematological and serum biochemical parameters in the Carp *Cyprinus carpio. Bull Environ Contam Toxicol*., 70: 993 – 997.

Joshi, P.K., Bose, M., and Harish, D., 2002. Haematological changes in the blood of *Clarias batrachusn* exposed to mercuric chloride, *Ecotoxic. Environ, Monit*., 12, 199 – 122.

Kale Monika, K., Joshi, P.P., and Kulkarni, G.k., 2006. Effect of cadmium toxicity on biochemical composition of freshwater fish *Rasbora daniconicus*. In ecology and Environment (B.N. Pandey and M.K. Joyti Eds.) APH publicATION, New Delhi. Pp. 271-278.

Karuppasamy, R., S., Subathra and Puvaneswari, S. 2005. Haematological responses to exposure to sublethal concentration of cadmium in air breathing fish, *Channa punctatus* (Bloch). *J. Environ. Biol.,* 26(1), 123 – 128.

Lapadula, D.M., Johannsen, F. and Abou-Donia, M.B. 1990. Absence of developed neurotoxicity and increased plasma butyrylcholinesterase activity in triallate- treated hen. *Tundam, Appl. Toxic.,* 14, 191 - 198.

Liess, M., Brown, C., Dohmen, P., Heimbach, F., and Kreuger, J. 2005b. Effects of pesticides in field – EPIF.Brussels.SETAC press.136.

McLeay, D.J., and Brown, D.A. 1974. Growth stimulation and biochemical changes injuvenile cohosalmon *Oncorhyrchus kistuch* exposed to Bleached Kraft pulp mill effluent for 200 days. *J. Fish. Res. Biol.,* Canada, 31: 1043 – 1049.

Muthuviveganandavel, V., Muthuraman, P., Muthu, S. and Srikumar, K. 2008. A study on low dose cypermethrin induced histopathology, lipid peroxidation and marker enzyme changes in male rat. *Pestic Biochem Phys,* 91, 12–16.

Natarajan, E., Biradar, R.S., George, J.P.1992. Acute toxicity of pesticides to giant freshwater prawn *Macrobrachium rosenbergii* (De Man). *J. Aquacult. Tropics*., 7 (2):183-188.

Omitoyin, B.O. 2006. Haematological changes in the blood of *Clarias gariepinus* (Burchell1822) juveniles fed poultry litter. *Livestock Res. Rural Develop.,* 18:11.

Pamila, D., P. A., Subbaiyan, and Ramaswamy, M. 1991. Toxic effect of chromium, and cobalt on *Sartherdonmosambicus* (peters), *Indian J. Environ. Health*., 33, 218–224.

Pawloski, P.H., Burzynska, B., and Zielenkiewicz, P. 2006. Theoretical model of reticulocyte to erythrocyte shape transformation. *J. Theoretical Biol*. 243: 24- 38.

Pazhanisamy, K., and Indra, N. 2007. Toxic effects of arsenic on protein content in the fish, *Labeo rohita* (Hamilton). *Nat. Environ. Poll. Technol*., 6(1): 113-116.

Prashanth, M.S, and Neelagund, S.E. 2008. Impact of cypermethrin on enzyme activities in the freshwater fish *cirrhinus mrigala* (Hamilton) *Caspian. J. Environ. Sci*. 6(2): 91-95.

Rao, J. V., Begum, G., Sridhar, V., Reddy, N. C. 2005. Sublethal effects of monochrotophos on locomotor behavior and gill architecture of the mosquito fish, *Gambusia affinis. J. Environ. Sci. Health*, B. 40: 813 – 825.

Roe, J.H. 1955. The determination of sugar in blood and spinal fluid with anthrone reagent. *J. Biol. Chem*., 153: 373-380.

Sampath, K., and James, R., and Akbar, K.M. 1998. Effect of copper and zinc on blood parameters and prediction of their recovery in *Oreochromis mossambicus* (Posces: cichlidae). *Indian J. Fish*, 45, 129 – 139.

Santhakumar, M., Balaji, M., Ramudu, K. 1999. Effect of sublethal concentration of monocrotophos on erythropoietic activity and certain haematological parameters of fish *Anabus testudineus* (Bloch). *Bull Environ Contam Toxicol* 63: 379 – 384.

Senthil kumar, K., Kannan, K., Subramanian, A., and Tanabe, S. 2001. Accumulation of organochlorine pesticides and polychlorinated biphenyls in sediments, aquatic organisms, birds, bird eggs and bat collected from south India. *Environ. Sci Pollut. Res. Int*., 8, 35-47.

Usmani, K.A., and Knowles, C.O. 2001. Toxicity of pyrethroids and effect of synergists to larval and adult Helicoverpa Zea, *Spodoptera frugiperda*, and *Agrotis ipsilon* (Lepidoptera: Noctuidae). *J. Econ. Entomol*., 94: 868–73.

Velisek, J., Wlasow, T., Gomulka, P., Svobodova, Z., Dobsikova, R.., Novotny, L. Dudzik, M. 2006. Effects of cypermethrin on rainbow trout (*Oncorhynchus mykiss*), *Veterinarni Medicina,* 51(10): 469–476.

Verma, A.K., Pal, A.K., Manush, S.M., Das, T., Dalvi, R.S., Chandrachoodan, P.P., Ravi, P.M., Apte, S.K. 2007. Persistent sublethal chlorine exposure augments temperature induced immunosuppersion in *Cyprinus carpio* advanced fingerlings. *Fish shellfish Immunol*. 22: 547 – 555.

Wahbi, O. M., Shyma, S. M., and El – Dakar, A.Y. 2004. “Effect of pulp and paper industrial effluent on some blood parameters, gonads and flesh proteins in experimentally exposed striped seabream Lithongnathus mormyrus”. *Egyptian journal of Aquatic research*, Vol. 30, No. A. PP. 25 – 42.

Yawata, Y. 2003. Cell membrane: The red blood cell as a model. Wiley, VCH Verlag GmbH and Co., KGaA, Weinheim, p. 173- 260.

****