No. 2

2012

Original Research Paper

Sublethal Effect of Quinalphos on Peripheral Haematology of *Oreochromis mossambicus* (Peters)

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Nat. Env. & Poll. Tech. Website: www.neptjournal.com Received: 14/11/2011 Accepted: 24/12/2011

Key Words:

Oreochromis mossambicus Peripheral haematology Quinalphos Sublethal effects

ABSTRACT

Exposure of *Oreochromis mossambicus* (Peters) for 48 hours to a sublethal concentration of an organophosphorus insecticide Quinalphos EC25 exhibited a significant reduction in total erythrocyte count (TEC) and significant elevation of haemoglobin (Hb), haematocrit (Ht) and erythrocyte sedimentation rate (ESR) values. The erythrocyte constants mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) and erythrocyte indices volume index (VI), colour Index (CI) and saturation Index (SI) of the experimental fish were significantly higher than that of the control fish. The results of the present study showed that the effect of sublethal concentration (30 ppm) of Quinalphos for 48 hours on the peripheral blood of *O. mossambicus* was macrocytosis and hyperchromia.

INTRODUCTION

Peripheral haematology is a reliable indicator of health status, diagnosis of diseases and evaluation of the physiological effects of environmental toxicants in human and veterinary medicine (Davidsohn & Henry 1977). In recent years, in fishery science also haematology has attained same status as that in human and veterinary medicine. As a result of rapid industrialization and constant use of chemical fertilizers, insecticides and sewage flow into the aquatic ecosystem, aquatic pollution has become one of the most disastrous problems all over the world. Moreover, organophosphorus and carbamate insecticides are now extensively used in plant protection operations on account of their less persistence in the environment. But their excessive and indiscriminate use produce more hazards to non-target and useful fauna of freshwater environment. The injurious pesticides finally affect human beings through the food chain.

The data on LC_{50} values of fish with different insecticides will be highly useful in the final evaluation of the extent of pollution of aquatic environment by agricultural chemicals. Furthermore, with the knowledge of tolerance limits in fish and other aquatic organisms, it would be easy to establish limits and levels of acceptability of toxic agents by the biotic components of the aquatic environment (Koundinya & Ramamurthi 1979).

Therefore, the present study was undertaken to evaluate the effect of Quinalphos on certain peripheral haematological parameters of the freshwater teleost, *Oreochromis* mossambicus (Peters).

MATERIALS AND METHODS

Live and healthy fish *Oreochromis mossambicus* were collected from Mathsya Fed (a fish culturing centre under the Central Government), Njarakkal. Fish were transported to the laboratory in large plastic containers and acclimatized in the laboratory conditions for two days.

Apparently healthy fish ranging from 15-22 cm were selected for the present study. Ten fish were maintained as control and ten others were used for experimental purpose.

Determination of median tolerance limit (TLm)/LC₅₀ (48 hours): The TLm (48 hours) was determined prior to the experimental study by employing the method proposed by Doudoroff et al. (1951). LC₅₀ 48 hour was found to be 40 ppm and a sublethal concentration of 30 ppm was selected for the present study.

The blood sample was collected directly from the caudal artery by severing the caudal peduncle. Haematological analyses were carried out by employing standard technique described by Hesser (1960) and Blaxhall & Daisley (1973).

From the values of the TEC (million/mm³), Hb (g/100 mL) and Ht (%), the following three erythrocyte constants MCV, MCH, MCHC were calculated by using standard formulae (Dacie & Lewis 1991). In the case of experimental fish, the three erythrocyte indices VI, CI and SI were also calculated.

Haematological parameters	Control fishes (n=10)	Experimental fishes (n=10)	't' values
TEC (× 10 ⁶ /mm ³) Range Hb (g%) Range Ht (%) Range ESR (mm/hr) Range MCV (m ³) Range	$\begin{array}{l} 3.69 \pm 0.02 \ (3.60\mathchar`-3.81) \\ 10.30 \pm 0.06 \ (10.00\mathchar`-10.50) \\ (32.02 \pm 0.19 \ (31.03\mathchar`-32.91) \\ 0.59 \pm 0.01 \ (0.55\mathchar`-0.65) \\ 86.58 \pm 0.34 \ (85.22\mathchar`-88.56) \end{array}$	$\begin{array}{l} 2.85 \pm 0.02 \; (2.75 \hbox{-} 2.93) \\ 11.54 \pm 0.03 \; (11.40 \hbox{-} 11.70) \\ 34.04 \pm 0.17 \; (33.18 \hbox{-} 34.68) \\ 0.70 \pm 0.01 \; (0.65 \hbox{-} 0.75) \\ 119.29 \pm 0.43 \; (117.43 \hbox{-} 121.15) \end{array}$	27.359* 21.472* 7.695* 7.570* 54.573*
MCH (Pg) Range MCHC (%) Range VI Range CI Range SI Range	$\begin{array}{c} 27.82 \pm 0.21 \ (26.66\text{-}28.72) \\ 32.14 \pm 0.22 \ (30.93\text{-}32.99) \\ 1.00 \\ 1.00 \\ 1.00 \end{array}$	$\begin{array}{l} 40.45 \pm 0.34 \ (39.04 - 42.18) \\ 33.90 \pm 0.22 \ (33.16 - 34.96) \\ 1.37 \pm 0.00 \ (1.35 - 1.39) \\ 1.44 \pm 0.01 \ (1.39 - 1.51) \\ 1.05 \pm \ 0.00 \ (1.03 - 1.08) \end{array}$	29.669* 5.447*

Table 1: Haematological parameters (mean ± S.E, range and 't' values) of control and Quinalphos exposed O. mossambicus.

*P< 0.01; Ranges are indicated in parenthesis; n = number of fish

RESULTS

The results of the haematological analyses of the control and experimental fish are presented in Table 1 and Fig. 1.

TEC (× 10⁶/mm³): TEC of the control fish ranged from 3.60-3.81 and that of the experimental from 2.75-2.93. The mean values for the two groups were 3.69 ± 0.02 and 2.85 ± 0.02 respectively. The results indicated that the exposure to Quinalphos significantly reduced (p<0.01) the erythrocyte count of the fish.

Hb (**g%**): The Hb content of control fish ranged from 10.00 -10.50 with a mean value of 10.30 ± 0.06 ; whereas Hb content of experimental fish ranged from 11.40-11.70, and the mean value for this category was 11.54 ± 0.03 . The results show that the exposure to Quinalphos significantly increased the Hb content (p<0.01).

Ht (%): The Ht values of control fish recorded a mean value of 32.02 ± 0.19 with a range of 31.03-32.91. For experimental fish, the Ht value recorded a mean value of 34.04 ± 0.17 with a range of 33.26-34.68. The Ht value for the experimental fish was significantly (p<0.01) higher than that of control fishes.

ESR (mm/hr): ESR values of the control fish recorded a mean value of 0.59 ± 0.01 with a range of 0.55-0.65. For the experimental fish, the ESR values recorded a mean of 0.70 ± 0.01 with a range of 0.65-0.75. Here also, the ESR values were significantly (p<0.01) higher in experimental fish as compared to control fish.

Erythrocyte constants: Erythrocyte constants like MCV, MCH and MCHC recorded significantly (p<0.01) higher values in Quinalphos exposed fish than that of control. The MCV of the experimental fish was 119.29 ± 0.43 mm³ with a range of 117.43 - 121.15 mm³, and that of control was 86.58 ± 0.34 with a range of 85.22-88.56 mm³.

MCH was also significantly (p<0.01) altered by

exposure to Quinalphos. In control fish, the MCH recorded an average value of 27.82 ± 0.21 Pg with a range of 26.66-28.72 Pg, whereas in experimental fish it was 40.45 ± 0.34 Pg with a range of 39.04-42.18 Pg.

MCHC was also significantly higher (p<0.01) in the experimental fish. In control fish, the average value of MCHC was 32.14 ± 0.22 % with a range of 30.93-32.99 % and that of experimental fish it was 33.90 ± 0.22 % with a range of 33.16-34.96 %.

Erythrocyte indices: From the mean MCV values of the control and experimental fish, VI can be calculated. VI of experimental fish recorded an average value of 1.37 ± 0.00 . It clearly shows that exposure to sublethal concentration of Quinalphos resulted in swelling of the erythrocytes. The mean value of CI of the experimental fish recorded was 1.44 \pm 0.01, and the SI was 1.05 ± 0.00 .

DISCUSSION

On exposure to the pesticide, TEC of the experimental fish showed significant reduction when compared to the control fish, while the Hb content, Ht values and ESR of the experimental fish showed significant increase. Moreover, the erythrocyte constants MCV, MCH, MCHC and erythrocyte indices CI, VI, SI were also significantly higher than that of control fish.

Many toxicants are known to lower TEC, Hb or Ht and cause anaemia in fishes (Das & Mukherjee 2001, Sampath et al. 2003). The anaemic state developed in the pesticide treated fish may be due to the stress induced by these toxicants. In general, the stress response is the result of haemodilution, a mechanism that reduces the presence of an irritating factor such as the pesticide in the circulatory system of fish. Razia Beevi & Radhakrishana (1987) studied harmful effect of sublethal concentration of formalin on *Sarotherodon mossambicus*. According to them exposure of



Fig. 1: Haematological parameters of control and exposed O. mossambicus.

S. mossambicus to 80 ppm formalin induced lowering of TEC and increase in Hb and Ht. Erythrocyte constant and indices of exposed fish were higher than that of control fish.

According to Radhakrishnan & Prasad (1994) exposure of tilapia to a sublethal concentration of an organophosphorus insecticide, Ekalux EC 25 (25 % W/W, Quinalphos) elevated TEC count, Hb and Ht. This enabled the fish to compensate for the insecticide induced reduction in gas exchange through the gills. *Labeo rohita* exposed to sublethal concentration of nuvan recorded reduction in erythrocyte count and anaemia (Das & Mukherjee (2001).

In the present study, unlike the results obtained by the former workers TEC was significantly lowered by Quinalphos treatment. It may be noted that MCV of exposed fish was much higher than that of the control fish. This naturally would result in the reduction of the number of erythrocytes in unit volume of blood. Therefore, it is reasonable to conclude that the direct effect of Quinalphos has been on the erythrocyte volume and not on its numerical abundance. Inhibition of erythropoiesis and increase in the rate of erythrocyte destruction in haematopoietic organs were the cause of decrease in RBC count (Joshi & Deep 2002).

Pesticides are supposed to cause a catalysing action on the incorporation of body iron store into Hb, thereby increasing the Hb% (Abidi & Srivastava 1988). Haemoconcentration resulting from water loss under the stress of pesticide is a major and perhaps the most important factor which explains the per unit volume increase in Hb%. Exposure to Quinalphos was found to result in statistically significant increase in Ht of *O. mossambicus*. Since, the TEC of exposed fish recorded a lower value than that of the control fish, presumably the increased Ht is the result of the increased MCV.

Radhakrishnan & Prasad (1994) reported an increase in Hb and Ht of *O. mossambicus* following exposure to Ekalux. But in their study TEC was also higher, and an increase in Hb and Ht might have been due to increased number of blood cells. In present study, high Ht and ESR recorded may be considered as an indicative of haemoconcentration. The increase in ESR may be correlated with the increased MCV of experimental fish, since size of the RBC is a critical factor affecting the sedimentation rate (Jayachandran & Ignatius 2009).

In conformity with the increased MCV, VI of the exposed fish recorded an average value of 1.37. It clearly indicates that exposure to Quinalphos induced swelling of erythrocytes of the fish. The increase in MCV of the experimental fish points to macrocytosis (increase size of RBC), a compensatory mechanism to combat the hypoxia developed due to loss of haemoglobin and decrease in TEC by increasing the size of RBC. The present results showed that the erythrocytes of Quinalphos exposed fish became hyperchromic

(high CI) and macrocytic (high VI). The observed high CI is not because of supersaturation of erythrocytes with Hb but because of increased cell volume. The larger cells have accumulated more Hb, compared with normal erythrocyte, resulting both in increased Hb and MCH.

The present study reveals that the contamination of sublethal concentration of Quinalphos to water causes haematological disturbances in fish, which in turn affects normal health of fish. Through food chain, pesticides reach higher trophic levels and affect the health of higher organisms including human beings. Therefore, adequate precautionary measures have to be taken to avoid the mixing of the pesticides into the nearby freshwaters and estuarine ponds or other water bodies where edible fishes are cultivated in large numbers.

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