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Effect of endosulfan on hematological indices of albino mice

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ABSTRACT

Endosulfan ($C_9H_6Cl_6O_3S$) belongs to organochlorine group of pesticide, under the cyclodiene subgroup. It used against insects and mites in agriculture and allied sectors. The effects of chemical endosulfan pesticide were investigated on Albino mice. The test was carried out using 1, 2, 3 and 4 mg/kg/d of endosulfan pesticide. Chronic bioassays were evaluated on hematological indices of the Albino mice. Blood sample was collected on weeks 2, 4, 6 and 12 for hematological analysis. There was significant reduction ($P<0.05$) in red blood cell (RBC) at days 15, 30 and 45, hemoglobin (Hb) at days 15 and 45, and packed cell volume (PCV) at all days of the evaluation. White blood cell (WBC) values however showed significant increase ($P<0.05$) at days 45 and 60. There were variations in mean values of mean cell volume (MCV), mean cell hemoglobin (MCH) and mean cell hemoglobin concentration (MCHC) with significant increase observed only at day 15 of MCHC

Key Words - Albino mice, Endosulfan, Haematological, Haemoglobin, Red blood cell

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INTRODUCTION

Endosulfan is a widely used acaricide and broad-spectrum chlorinated hydrocarbon pesticide for the management of agricultural pests and mites on fields, fruits, and vegetable crops (Naqvi and Vaishnavi, 1993). Concerns about environmental pollution were greatly heightened when residual levels of EN and other pesticides were found in the soil, water bodies, vegetables, grains, and other food products. (IARC, 1983; Smith, 1991; USEPA, 2002; WHO, 2002). Recognized as a persistent organic pollutant having variable half-life ranging from 3 days to about 5 months in water (Howard 1991). EN exhibits high bioavailability levels that can bioaccumulate and biomagnify themselves in the food chain. The food chain can bio-accumulate and bio-amplify EN's high levels of bio-availability. (Bhalerao and Puranic, 2007; Rivas *et al.,* 2007; Tan *et al.,* 2007). Earlier works on animals indicate that EN toxicity may be influenced by the species tested and the level of protein in the diet (Smith, 1991). The kidneys, liver, parathyroid glands, and blood are among the organs that EN metabolites are most likely to influence through stimulation of the central nervous system (ATDSR 2000). Fish (Johnson and Finley, 1980; Siang *et al*., 2007), birds (Hill and Camardese, 1986), and bees (Kidd and James, 1991; USNLM, 1995) have all been demonstrated to be extremely to moderately toxic when exposed to EN.

In addition, studies on the long-term toxicity, abnormalities in reproduction, and carcinogenicity of this substance in mice and rats [NCI, 1978; Hurt, 1991; Hack *et al*., 1995; Manzula *et al*., 2000; Paul *et al*., 2000; Dalsenter *et al.,* 2003] as well as exposure to EN in young male humans have demonstrated that it delays sexual maturity and interferes with the synthesis of sex hormones. The

kidneys, liver, parathyroid glands, and blood are among the organs that EN metabolites are most likely to influence through stimulation of the central nervous system.

The present study investigates the effect of Endosulfan on hematological parameter in *Mus musculus.*

MATERIALS AND METHODS

Experimental animals and care: Swiss albino mice weighing 20–25 g and aged 6–8 weeks were procured from the animal house of University Department of Zoology, B.N.M.U. Madhepura. They were kept in 45 cm x 30 cm x 30 cm polypropylene cages with weekly replacements of sawdust bedding. A constant room temperature of 28-30°C and a controlled light: dark cycle was maintained in the laboratory.

Reagents and Chemicals: Technical grade EN ($C_9H_6Cl_6O_3S$, 97% pure) was purchased from Scientific store Madhepura. All additional chemicals and reagents were of the highest purity and quality for analytical usage.

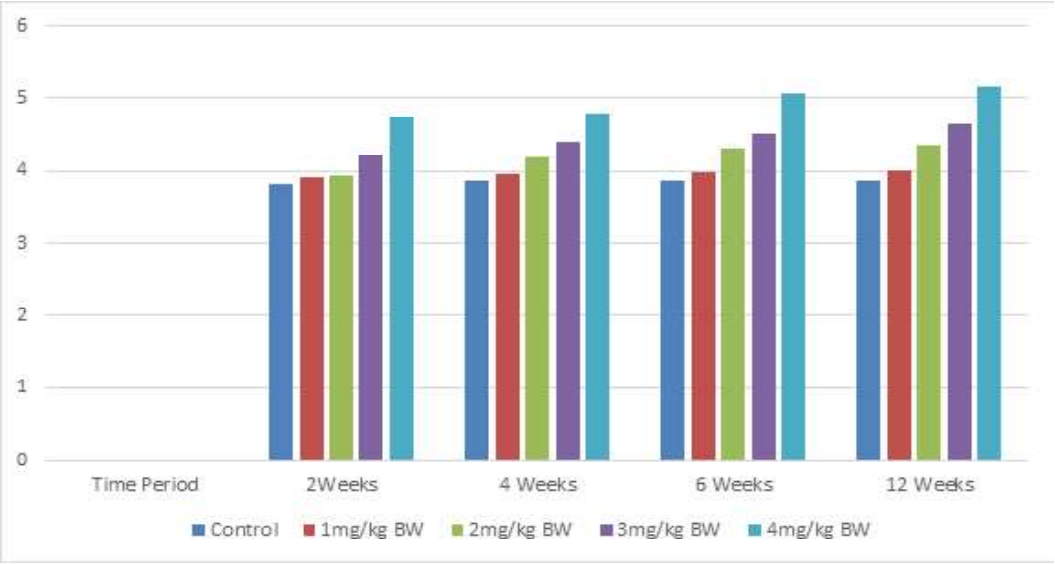
Working schedule: 60 mice were divided into five groups. The first group was considered as control group and rest are considered as experimental group received 1, 2, 3 and 4 mg/kg of body weight/ day of Endosulfan orally. Treatments were continued for 2, 4, 6 and 12 weeks. Blood was collected after 2, 4, 6 & 12 weeks from the treatment started and hematological parameter were done.

RESULT AND DISCUSSION

The Following Results were obtained after experiment.

Table A: Number of leukocytes (thousand/mm³) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

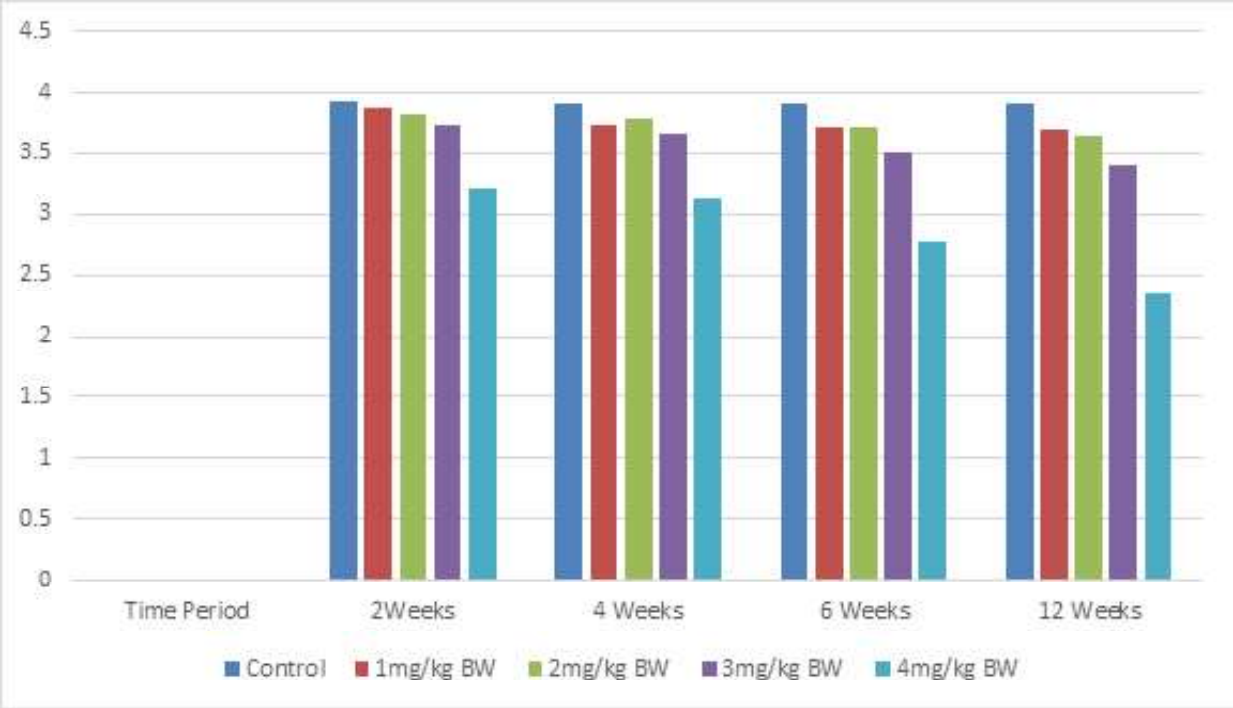
Doses Time Period	Control Group	1mg/kg BW Gr.1	2mg/kg BW Gr. 2	3mg/kg BW Gr.3	4mg/kg BW Gr. 4
2 Weeks	3.82± 1.65	3.91	3.93	4.21	4.75
4 Weeks	3.87± 1.57	3.95	4.20	4.39	4.79
6 Weeks	3.87± 1.55	3.99	4.31	4.51	5.07
12 Weeks	3.86± 1.56	4.01	4.35	4.66	5.15



Graph A: Number of leukocytes (thousand/mm³) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

Table B: Number of RBC (million/mm³) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

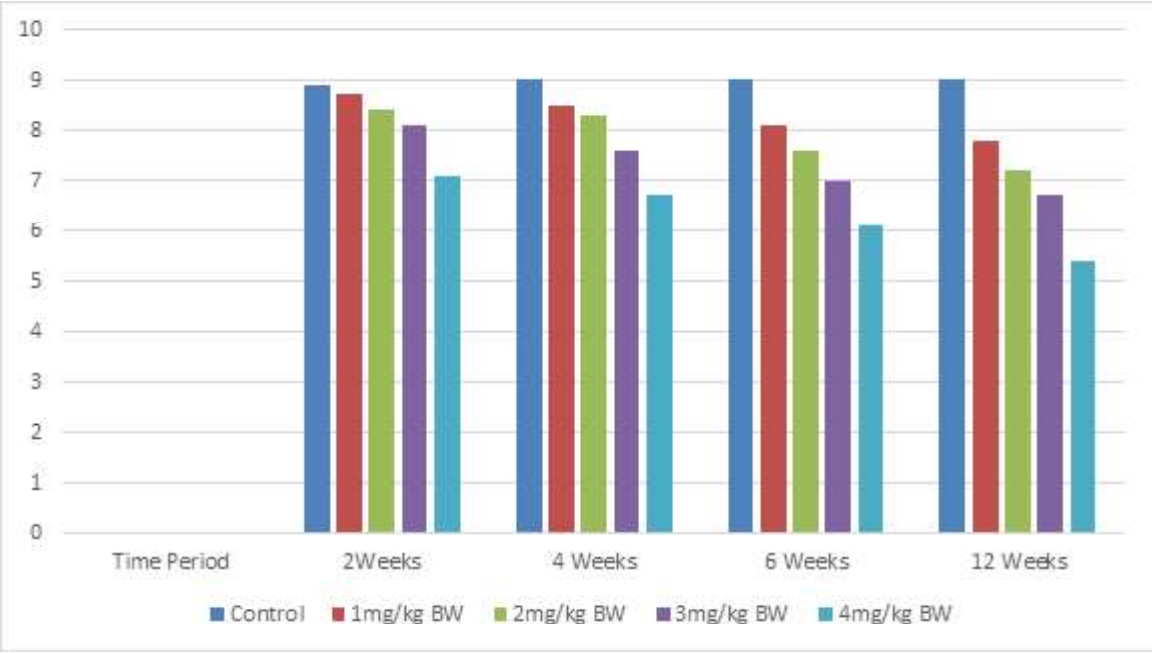
Doses Time Period	Control Group	1mg/kg BW Gr.1	2mg/kg BW Gr. 2	3mg/kg BW Gr.3	4mg/kg BW Gr. 4
2 Weeks	3.92± 0.92	3.87	3.82	3.74	3.21
4 Weeks	3.90± 0.91	3.74	3.78	3.67	3.12
6 Weeks	3.90± 0.87	3.71	3.71	3.50	2.78
12 Weeks	3.90± 0.88	3.69	3.65	3.41	2.34



Graph B: Number of RBC (million/mm³) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

Table C: Amount of Hb (gm%) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

Doses Time Period	Control Group	1mg/kg BW Gr.1	2mg/kg BW Gr. 2	3mg/kg BW Gr.3	4mg/kg BW Gr. 4
2 Weeks	8.91± 0.65	8.7	8.4	8.1	7.1
4 Weeks	9.01± 0.57	8.5	8.3	7.6	6.7
6 Weeks	9.03± 0.55	8.1	7.6	7.0	6.1
12 Weeks	9.03± 0.56	7.8	7.2	6.7	5.4



Graph C: Amount of Hb (gm%) in peripheral blood in Swiss mice poisoned with Endosulphan statistically significant differences compared to Control Group *(p<0.05).

Number of erythrocytes in mice: In Endosulfan poisoning, a statistically significant decrease was noted in the number of erythrocytes in the blood of animals, compared to the control groups ($p > 0.05$). The average no of erythrocyte 3.90 million/ mm^3 were obtained in control group. Lowest RBC count (2.34 million/mm^3) was found in group 4 at 12 weeks.

Hemoglobin level in mice: In the case of Endosulfan there is significant differences in the level of haemoglobin were observed between the control and experimental groups. The average amount of haemoglobin in mice is 8.99gm%. 5.4 gm% was the least amount of Hb detected in mice treated with dose 4mg/kg BW of Endosulfan till 12 weeks.

WBC in mice: In the experimental mice, an increase in the number of leukocytes (WBC -white blood cells) in peripheral blood occurred only after administration of the dose of Endosulfan, compared to the control.

Endosulfan poisoning in animals, which were administered high dose (4mg/kg BW) resulted in a statistically increase in the number of leukocytes (5.12 thousand/mm^3) in peripheral blood, compared to the control groups (3.85 thousand/ mm^3) and animals (4.75 thousand/mm^3) poisoned with the lower dose(1mg/kg/BW) of the pesticide ($p < 0.05$). After poisoning with four different doses of Endosulfan, a statistically significant increase was observed in the number of leukocytes in peripheral blood in experimental animals, compared to the control. Anemia might be attributed to low nutrition intake, reduced protein synthesis and increased protein excretion in urine.

Kumar *et al*., 2015 showed Endosulfan caused the lymphocytopenic leukocytopenia, which were reported in mice. Reduction in hemoglobin and PCV values, suggestive of anemia in the present study are in accordance with earlier reports in poultry, mice and pregnant rat.

Packed cell volume (PCV) decreases at days 15 and 45, after administration of Endosulfan. There were variations in mean values of mean cell volume (MCV), mean cell hemoglobin (MCH) and mean cell hemoglobin concentration (MCHC) with significant increase observed only at day 15 of MCHC.

It is true in that the quantity of erythrocytes, haemoglobin levels, in Swiss mice differ depending

on the sex of the animals. In comparison to male mice, all of the studied metrics are higher in female mice (Haratym, 2002). In Endosulfan poisoning, very significant changes were noted concerning the analyzed erythrocytic parameters in albino male. In all experimental mice group inhibition of erythrocytic system was observed. The amount and dose of the preparation also plays an important role in the response of the erythrocytic system. In Endosulfan poisoning, the higher dose strongly inhibits the hemopoietic system, whereas the lower dose causes its inhibition in animals but in lower extent. Other writers that studied the impact of pyrethroid poisoning on the quantity of erythrocytes in mice according to sex and dose noted similar connections (WHO, 1989, 90 & 91). Haemoglobin levels were similarly affected by Endosulfan poisoning in mice, depending on the sex and dose (Desi, 1986, & Eil, 1990).

In mice poisoned with fenvalerate a decrease in haemoglobin level and the number of erythrocytes were noted, with a clear increase in the number of thrombocytes (WHO, 1991 & International Agency for Research on Cancer, 1991) Studies conducted by Desi *et al*. (1986) showed a decrease in hematocrit level and an increase in the level of haemoglobin in animals poisoned with cypermethrine.

With respect to the number of leukocytes, the groups examined differed according to the Endosulfan dose. There are significant differences were noted among all the experimental groups. Endosulfan poisoning in mice caused an increase in the number of leukocytes in peripheral blood. The information may indicate that Endosulfan poisoning has a mobilising effect on mice's leukocytic system.

Other studies show similar data: in fluvalinate poisoning in rats of both sex (Gerg 1992) an increase was noted in the number of leukocytes in peripheral blood, especially of neutrophilic granulocytes, whereas in.-cypermethrin poisoning in female mice (Luty, 2000) an increase in the number of monocytes was observed. According to some authors, the administration cypermethrin causes a decrease in the number of leukocytes (Varshneya, 1992).

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