

## **HAEMATO-BIOCHEMICAL PROFILE FOLLOWING SUB ACUTE TOXICITY OF MALATHION IN MALE ALBINO RATS**

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### **Summary**

Pesticide exposures cause disorders varying from straight forward topical irritant reactions, to complex systemic illness. Organophosphorus esters have the potential to produce several forms of toxicity. In the present study therefore we have sought to investigate the toxic effect of malathion (an organophosphate) on haematological and serum biochemical parameters at the dose level of 100, 300 and 500 mg/kg b.wt/day for 30 days. Total erythrocyte count, haemoglobin and haematocrit percentage were decreased whereas the total leucocyte count increased significantly in a dose dependent manner. The mean corpuscular volume showed non-significant changes whereas the mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration were decreased as compared to the corresponding controls at higher dose levels. The serum biochemical parameters viz alanine amino transferase, aspartate amino transferase and acid phosphatase were increased while alkaline phosphatase was decreased which showed the hepatotoxic effect of malathion. From the above mentioned findings it has been concluded that exposure of malathion has deleterious effects on physiology and biochemistry in albino rats. It is therefore the application of such pesticides should be limited to a designed program.

Key words: Malathion, Haematology, Serum, Biochemistry, Rat.

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## Introduction

Pesticides are chemical substances that are used to kill, repel, or regulate the growth of biological organisms (1). Pesticides occupy a unique position among the many hazardous chemicals that men and animals encounter daily (2). Pesticides are a diverse group of chemical compounds and consist of insecticides, fungicides, herbicides, and rodenticides. Pesticides have contributed to dramatic increases worldwide in crop yields and have helped to limit the spread of disease. But pesticides also have harmful effects and can injure human health as well as the environment. The range of these adverse health effects includes acute and persistent injury to the nervous system, lung damage, injury to the reproductive organs and dysfunction of the immune and endocrine system, birth defects and cancer (3).

Problem of pesticide contamination appears considerable not only in India but in many other countries. Among the three major groups of pesticides organophosphorous pesticides are more potent (4). Hundreds of Organophosphorus (OP) compounds are currently available to use as insecticides (5). Organophosphorus insecticides are used extensively to control agricultural, household and structural pests. These insecticides constitute a diverse group of chemical structures exhibiting a wide range of physicochemical properties, with their primary toxicological action arising from inhibition of the enzyme acetylcholinesterase (6). Their persistence and ubiquitous nature coupled with a tendency to concentrate in organism as they move up the food chain may increase their toxicity to man and cause other harmful effects on man's health and well being (7, 8).

This study is therefore undertaken to investigate the magnitude of toxicity of malathion (an organophosphate) on haematology and serum biochemistry of albino rat.

## Materials and methods

**Test animal:** Healthy adult male albino rats (*Rattus norvegicus*) weighing 150-200 gm were selected for experimentation. They were housed in individual polypropylene cages at room temperature ( $22 \pm 2^\circ\text{C}$ ) and uniform light (14:10: L: D). The animals were mostly maintained on standard pallet diet procured from Ashirwad food Industries Ltd, Chandigarh, India and occasionally on germinated / sprouted gram and wheat seeds as an alternative feed and fresh water *ad-libitum*.

**Testing dose and experimental design:** Malathion (Technical grade) was dissolved in groundnut oil and administered to albino rats by pearl point needle at the dose level of 100, 300 and 500mg/kg b.wt./day for 30 days. Healthy rats were divided into four groups of ten animals each. The control group I received only the vehicle (groundnut oil), whereas the animals of group II, III and IV were received malathion dissolved in groundnut oil. At the end of experimentation period i.e. on day 31st the animals were weighed and autopsied under light ether anesthesia and blood was collected by cardiac puncture. Serum was then obtained by centrifugation at 3000 rpm.

**Blood and Serum analysis:** Haemoglobin concentration was estimated by standard procedure (9), packed cell volume was determined by using wintrobe tube (10), total erythrocyte count and total leukocyte count were counted on improved Neubauer haemocytometer (11). Serum aspartate amino transferase, alanine amino transferase were assayed (12). Serum acid and alkaline phosphatases were estimated (13). Difference between the control and treated groups were evaluated statistically by ANOVA test and significance was set at  $P < 0.01$  and  $P < 0.001$ .

## Results

The exposure of rat to malathion was carried out at three dose levels viz 100, 300 and 500 mg/kg. b.wt. /day for 30 days.

### Haematology–

Table 1: Effect of oral administration of malathion on haematological parameters in male rats

S.No.	Parameters	Control	100	300	500
			mg/kg.b.wt./day	mg/kg.b.wt./day	mg/kg.b.wt./day
			30 days	30 days	30 days
1.	Total Erythrocyte Count(TEC) (million/mm <sup>3</sup> )	6.56 ± 0.21	5.87 <sup>ns</sup> ± 0.33	5.77** ± 0.19	4.72** ± 0.18
2.	Total Leucocyte Count(TLC) (million/mm <sup>3</sup> )	5300 ± 253	7937.50* ± 821.17	7087.50** ± 461.14	8312.50** ± 569.49
3.	Haemoglobin (Hb) (gm %)	15.25 ± 0.32	13.68** ± 0.31	11.28** ± 0.28	10.00** ± 0.20
4.	Haematocrit (%)	50.44 ± 1.59	50.75 <sup>ns</sup> ± 0.10	34.50* ± 3.01	38.50** ± 1.32

Values given are Mean of results obtained from 10 animals.

ns = non-significant values ± SEM 10 determinations.

\* = P ≤ 0.01 significant

\*\* = P ≤ 0.001 highly- significant

### Total erythrocyte count (TEC)

The malathion exposed rats showed highly significant decrease in total erythrocyte count at higher dose levels as compared to control rats.

### Total leukocyte count (TLC)

A highly significant increase in leukocyte count of malathion exposed rats was observed at higher dose levels as compared to control rats.

### Haemoglobin

The haemoglobin concentration of all malathion exposed rats were found to be decreased highly significant at all the dose levels.

**Haematocrit**

Decrease in the percentage of haematocrit was noticed at higher doses of malathion exposed rats as compared to control group.

**Haematological indices –**

Table 2: Haematological analysis of treated male rats

S.No.	Parameters	Control	100 mg/kg.b.wt./ day	300 mg/kg.b.wt./ day	500 mg/kg.b.wt./ day
1.	Mean Corpuscular volume (MCV) (cu)	76.89 ± 4.48	87.30 <sup>ns</sup> ± 6.10	81.0 <sup>ns</sup> ± 1.80	68.80 <sup>ns</sup> ± 5.79
2.	Mean Corpuscular Haemoglobin (MCH) (pg)	23.24 ± 1.31	23.40 <sup>ns</sup> ± 1.40	21.0 <sup>ns</sup> ± 0.70	19.57* ± 0.91
3.	Mean Corpuscular Haemoglobin concentration (MCHC) (gm/dl)	30.26 ± 0.38	26.97* ± 1.02	25.0 <sup>ns</sup> ± 7.50	28.00** ± 0.25

Values given are Mean of results obtained from 10 animals.

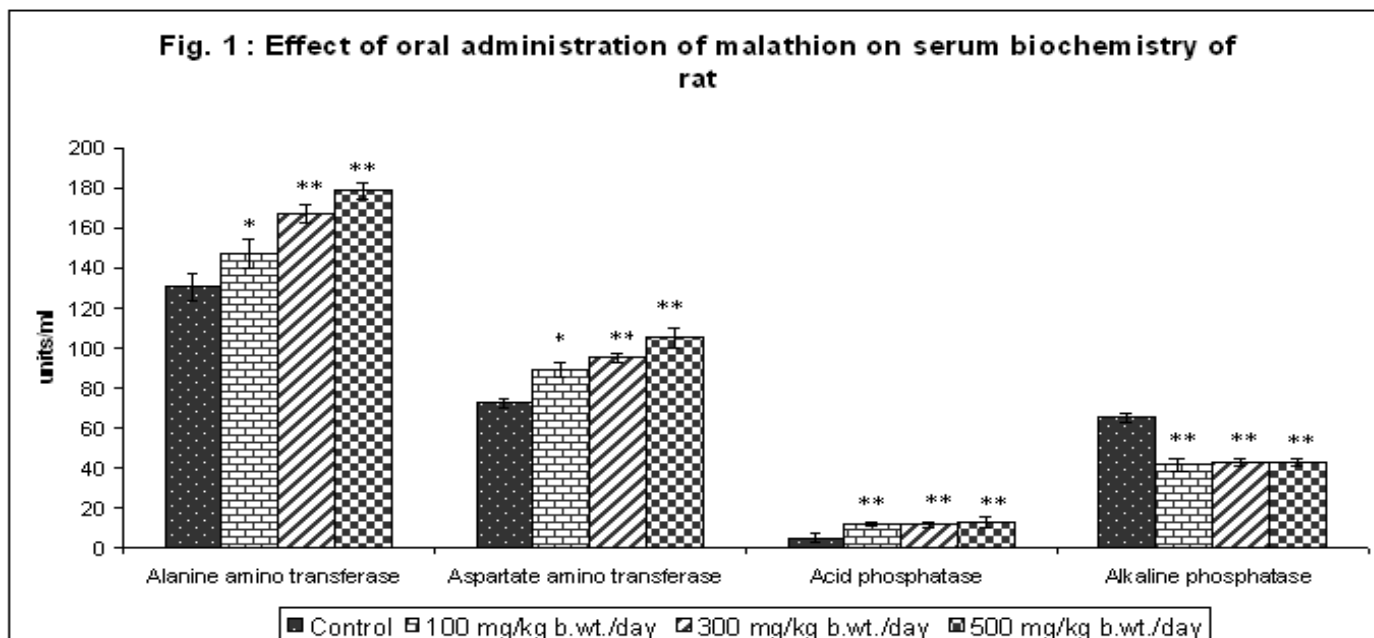
ns = non-significant values ± SEM 10 determinations.

\* =  $P \leq 0.01$  significant

\*\* =  $P \leq 0.001$  highly- significant

Exposure of rats to malathion revealed non significant changes in mean corpuscular volume (MCV) whereas mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) decreased significantly at higher dose levels.

## Serum biochemistry -



ns = non significant

\* =  $p \leq 0.01$  significant

\*\* =  $p \leq 0.001$  highly- significant

#### Alanine amino transferase and aspartate amino transferase (Fig. 1)

After exposure of malathion to rats for 30 days it was observed that all the three groups showed highly significant ( $P \leq 0.01$  and  $P \leq 0.001$ ) increase in both alanine amino transferase and aspartate amino transferases. The respective increase in aspartate amino transferase was 31.57%, 23.98% and 45.42% and that of alanine amino transferases was 28.28%, 24.44% and 37.11% in comparison to control animals.

#### Acid and alkaline phosphatase (Fig. 1)

A significant increase ( $P \leq 0.01$  and  $P \leq 0.001$ ) in the activity of acid phosphatase was observed after exposure to malathion to all the three groups of rats in comparison to control rats, whereas a highly significant decrease ( $P \leq 0.001$ ) in the activity of alkaline phosphatase was observed in all the three groups of malathion exposed rats in comparison to control rats.

### **Discussion**

Blood is a sensitive index of the physiological changes of an animal to any environmental pollutants and it is well known that toxic stress of any nature would show conspicuous and significant changes in the haematological parameters.

The present study revealed that administration of malathion to rats at different dose levels for 30 days produces significant changes in the haematopoietic system and in serum biochemical parameters. Exposure to malathion shows blood diseases viz aplastic anemia, which is a precursor to leukemia. It also caused blood disorders, seizures, reproductive problems.

In present study the decrease in erythrocyte count may be due to either inhibition of erythrocyte production or destruction of erythrocyte due to poisoning. Vitamin B12 and folic acid are essential for the proper maturation and production of nucleated red cells. Vitamin B12 deficiency is characterized by disturbance in erythropoiesis. Its deficiency leads to impaired synthesis of nucleic acid resulting into defective maturation of erythrocytes and their nuclei (14). Decrease in erythrocyte count leads to fall in haemoglobin concentration (15).

Synthesis of haemoglobin(Hb) begins in the polychromic normoblast stage. The synthesis of Hb requires iron, which is generally obtained from stored ferritin and from dietary sources. The reduction in general food intake by intoxicated albino rats and no supplementary supply of extra iron might be the reasons for the iron deficiency (16, 17). A fall in the rate of haemoglobin synthesis during all the stages of maturation of erythrocytes has also been reported, when the supply of iron is inadequate. A similar decrease in R.B.C. and haemoglobin has been reported in rats given sub acute doses of other organophosphate insecticides (18). Stimulation in leukocyte production was also observed as insecticides acts as chemical stressors causing increase in adrenaline level and consequently lymphatic leucocytosis (19). The decrease in the haematocrit percent may be due to either a decrease in the size of R.B.C. or a decrease in the number of erythrocytes (20). Decrease in the values of MCH, MCV and MCHC can be correlated with the decrease in the erythrocyte count and haemoglobin (21).

The elevation of aspartate amino transferase and alanine amino transferase levels may be due to the pathological changes such as necrosis of hepatocytes, which causes increase in the permeability of the cell membranes, resulting in the release of transferases in the blood stream (22).

A significant reduction in alkaline phosphatase may be attributed to the decrease osteoblastic activity of bone, since the alkaline phosphatase is formed and present in the osteoblasts. A significant increase in serum acid phosphatase activity was observed. Abnormal serum activity also indicates the liver injury. Pathological alteration in hepatic cells is the major contribution factor for this change (23, 24).

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