SEM Study on Cytotoxic effect of Monocrotophos (MCP) on Blood of Mice

Khalid Hamdan Gathwan

ABSTRACT:

BACK GROUND:

Monocrotophos (MCP) is an organophosphorus insecticide has both systemic and contact actions and has been used against insects.

METHODS:

Thirty six male Balb/c mice were used. These were divided into six group (A, B, A1, B1, A2, B2,). MCP was given orally daily by gavage 1/5th of LD50 (0.28 mg/100gm of body wt.), vitamin E and vitamin C was given orally daily by gavage @ 100 mg /kg body wt., while MCP+ Vitamine E and MCP + Vitamine C was also given.

RESULTS:

The signs of MCP toxicity include shivering, salviation and Iacrimation. Scanining electron microscopy (SEM) revealed that of erythrocytes in control were perfect discocytes (D). MCP treatment resulted in drastic alteration in the topography of erythrocytes. Vitamin E and Vitamin C treated mice revealed normal erythrocyte like that in control. MCP+ Vitamin E and MCP + vitamin C treated mice revealed almost normal of topography of erythrocyte.

CONCLUSION:

Pesticide exposure could be prevented by CO- administration antioxidant Vitamin E or Vitamin C.

KEY WORDS: MCP, Vitamin E, Vitamin C, Erythrocytes.

INTRODUCTION:

Monocrotophos (MCP) is an organophosphorus insecticide has both systemic and contact actions and has been used against wide range of insects including mites, boll worms, leaf eating beetles, sucking insects and other larvae on variety of crops[1-3]. Its residual effect on different vegetable crops and on soil microflora[4]. Adecrease in Hb content and in erythrocyte (RBC) has also been reported (MCP) exposure in the marked elevation of bilirubin in serum[5-8]. Furthermore, its known that (MCP) had adverse on the bone marrow causing decreased production of RBC[9].

Scanining electron microscopy (SEM) study was carried out to understand the mode action of (MCP) on erythrocytes of mice in the presences of vitamin E and C. Vitamin E is the most important lipid- soluble antioxidant, it acts by allowing free radicles to abstract a hydrogen atom from it[10-11]. Ascorbic acide (vitamin C) is one of the major soluble antioxidants in the aqueous compartments of cells due to its strong reducing power[12], and plays vital role in maintaining cellular α - tocopherol levels during oxidative stress and plays a prominent part in the protection of extracellular fluids such as blood plasma[13-14]. Therefore, the optimum levels of vitamins E and C, simultanceously needed for protection against oxidative stress[15].

In the present report, SEM study was carried out to understand the mode of action of MCP on erythrocytes of mice.

MATERIAL AND METHODS:

Adult male Balb/c mice weighing 32-34 gm.

These were divided into six groups (A,B,A1,B1,A2,B2), each group constituted at least 6 animals.

These were provided with rat chow and water ad libitum. The animals were orally administered MCP by gavage 1/5th of LD50 (0.28 mg/100gm of body wt.), the value of MCP as calculated by Janardhan et al[16].

Animals in (groupA) was given tap water and served as control, in group B was given daily MCP, group A1 was given daily of vitamin E @ 100 mg /kg body wt., while group A2 was given 0.2 mg/kg body wt., MCP therough gavage along with vitamin E @ 100mg /kg body wt.

The group B1 was given daily of vitamin C @ 100mg /kg body wt., while group B2 was given 0.28 mg/kg body wt., MCP through gavage along with vitamin C @ 100mg /kg body wt.

After the treatment period (21 days) blood from all the six groups was collected, SEM study of

College of Dentistry , Department of Basic Science

erythrocytes was carried out to visualize topographical changes in erythrocytes as result of MCP, vitamin E and vitamin C treatment.
For this, the technique[17], was taken, and viewed under SEM (Jeol 2601).

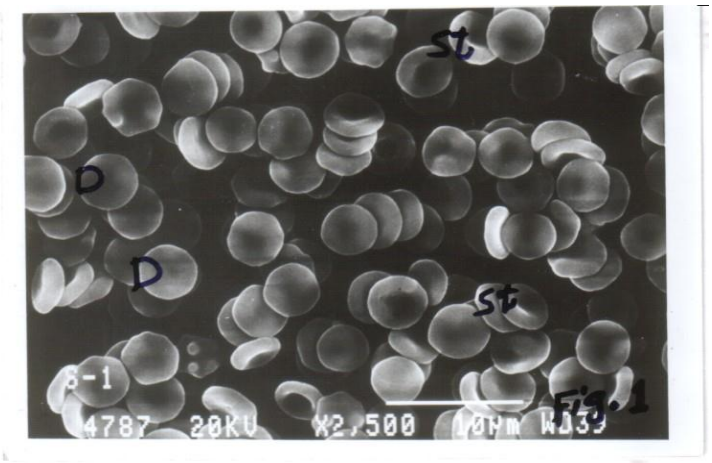
RESULTS:

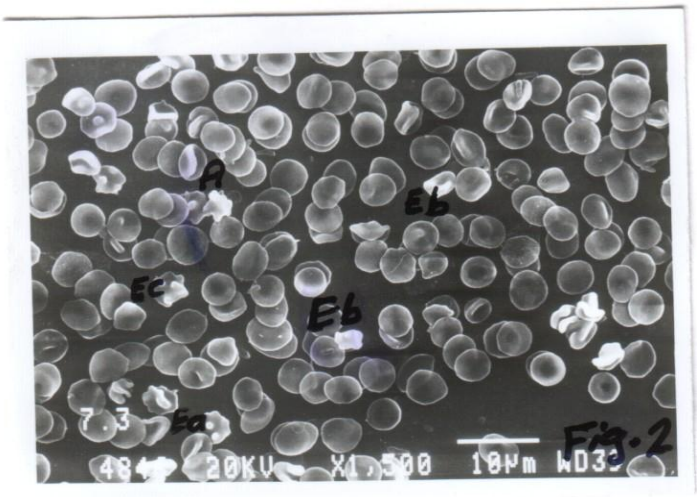
The signs of pesticide toxicity were observed within 30 minutes of administration of MCP.
These include shivering, salivation and lacrimation, urination was also found to be increased as shown by yellowish, these symptoms were more prominent during the initial 15-20 days.
The symptoms were found to be less server in MCP+ vitamin C treated (groupB2).
However, MCP + vitamin E treated (group A2) mice appeared to almost normal.
The present study revealed that of the most of the erythrocytes in control (groupA) were prefect disocytes (D).
A few cup shaped stomatocytes (St) were also observed Fig.1. MCP treatment to mice resulted in drastic alteration in the topography of erythrocytes. Most of the cells changed to stomatocytes (St) as well as echinocytes (E) (irregularty cernated with many projection). Echinocytes observed were of different stage viz. stage 1 (a) characterized by irregularity of edges ; stage 2 (b) characterized by spicules in still flat cells and stage 3 (c) characterized by spicules uniformly distributed over the surface of round cells.
One more type of cells called acanthocytes (A) were observed having spicules of varying length irregularly distributed over the erythrocyte surface, Figs.2-4.
Vitamin E treated (group A1) Fig.5 and vitamin C treated (group B1) Fig.6, revealed normal bioconcave discocyted like that of normal control (group a) . MCP + vitamin E trated (group A2) mice revealed almost normal topography of erythrocyte whereby most of the cells were found to be perfect discocytes, however a very few echinocytes or spherocytes were also observed Fig. 7. MCP+ Vitamin C treated (group B2) mice also revealed improved morphology of erythrocytes as compared to MCP treated (group B).
Howver, this improvement was found to be less when compared to MCP+ vitamin E (group A2), since the number echinocyted and acanthocytes were more in this group Fig.8.

DISCUSSION:

The study of the morphological features of cellular elements has great importance for assessing their functional state and vitality.
SEM having large depth of focus view to delineate the pathologic status of affected erythrocytes, the modification in the shape and size of erythrocytes represent the most common morphologic abnormalities that occur in pathological conditions[18]. It was revealed in the present study that MCP treatment (group B) to mice resulated in the drastic alterations in the topography of erythrocytes whereby the normal discocytes changed to stomatocytes, echinocytes in various stage and acanthocytes or exhibited central or peripherial protuberances.
These alteration in the surface morphology of erythrocytes may be due to abnormal erythropoiesis, deficient haemoglobin formation, damage to red cells after they leave the bone marrow or increased erythropoiesis to compensate for anemia[19].
Another possible reason for alterations / changes in erythrocyte morphology could be the formation of active Ioci on the erythrocyte membrane as a result of interaction of membrane lipids and proteins with free radicals, which might be produced due to the oxidative stress induced by MCP exposure.
In the present study, significant changes in the erythrocyte membrane have been observed following exposure to MCP.
Furthermore, the normalization of surface morphology of erythrocyte in MCP+ vitamin E and minimization of toxic effects in MCP+ vitamin C revealed that morphological changes in erythrocytes due to pesticide exposure could be prevented by CO- administration of antioxidant vitamin E or C, the SEM study confirms the protective role of vitamins E and C aginst MCP induced stress.
However, vitamin E has been found to be more beneficial since it could ameliorate MCP toxicity more effectively as compared to vitamin C in erythrocytes.

Fig .1 Showing biconcave discocytes (D) and stomatocytes (s t) in control .
Fig .2 Showing pathologically altered erythrocytes
echinoytes of various stages (Ea, Eb ,Ec), and acanthocytes (A) in MCP treated group.
Fig. 3 Showing pathologically altered echioncytes (E) and acanthocytes (A) at higher magnification in MCP treated group .


St
D
D
St
S-1
4787 20KV X2,500 10µm WD39
Fig.1


A
Eb
Ec
Eb
7.3
Ec
20KV X1,500 10µm WD39
Fig.2

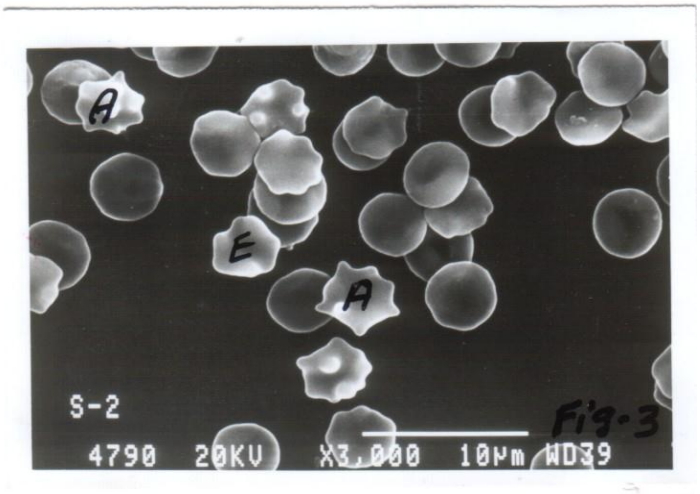
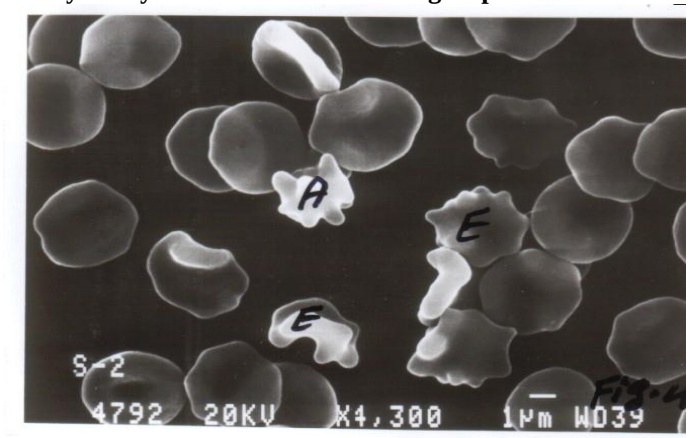
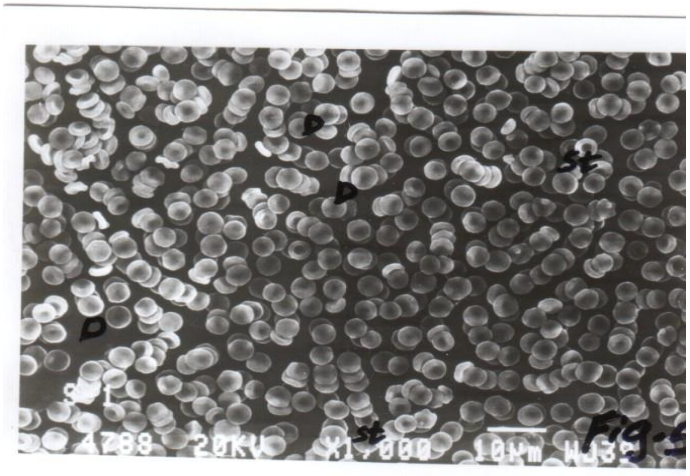

A
E
A
S-2
4790 20KV X3,000 10µm WD39
Fig.3

Fig . 4 Showing pathologically altered echioncytes (E) and acanthocytes (A) at higher magnification in MCP treated group .
Fig .5 Showing typical biconcave discocytes (D) and stomatocytes (st) in vitamin E treated group
Fig .6 Showing normal erythrocytes in vitamin C treated group .





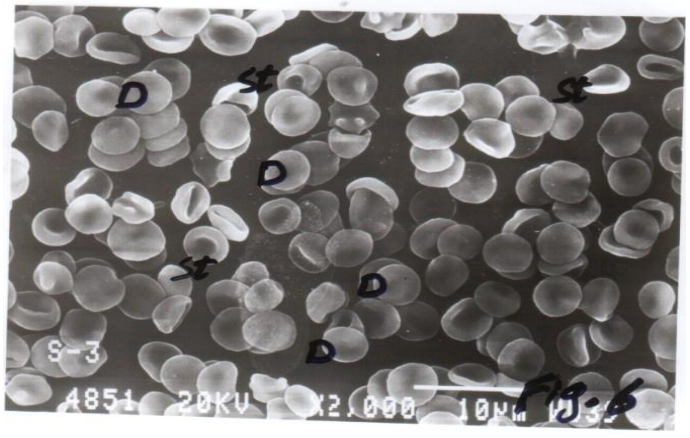
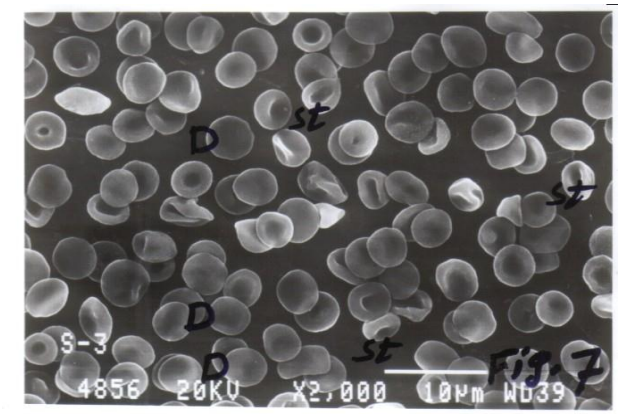
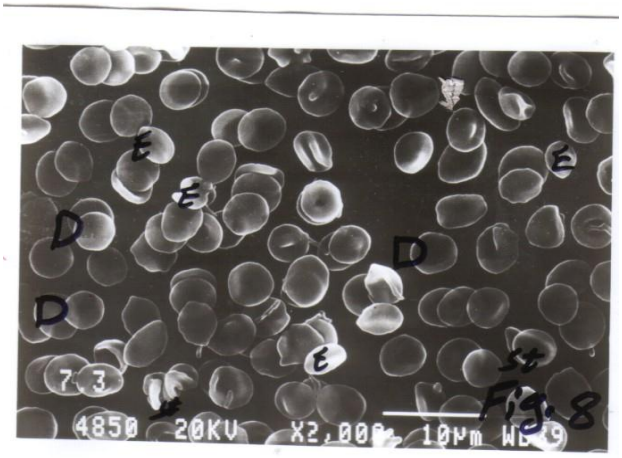


Fig .7 Perfectly normal erythrocytes in MCP +vitamin E treated group .
Fig .8 Showing normal erythrocytes and a few echinocytes (E) in MCP + vitamin C treated group.





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