

Cytotoxicity of Certain Environmental Agents on Plant Systems

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The cytotoxic and clastogenic effects of essential trace element, manganese at concentration of 1 to 10,000 ppm *Allium Sativum* and *Vallisneria spiralis*. The metal caused disturbances in mitotic cell division with different types of chromosomal abnormalities. The frequency of deviation and of aberrant cells depended on the period of treatment and concentrations. Prolonged treatment led to lethality and pulverization with appearance of pycnosis and extrusion of chromatin matter. The cytotoxic effect was more pronounced in *V. spiralis* than in *A. sativum*.

Key Words Chromosome Clastogenic Cytotoxic Division Mitotic Pycnosis Manganese Trace element

The cytotoxic effect of metals as environmental pollutants have been studied extensively on mammalian systems (King *et al.*, 1983; Erikson *et al.*, 1984; Morganti *et al.*, 1985; Sharma & Talukdar, 1987). As information on plants is relatively meagre, the present work has been undertaken to observe the cytotoxic effects of a metal manganese on plant materials.

Mn is an essential trace element for plant, animal and man. It is widely distributed in soil, sediments, rocks, water and biological material. In addition, the metal is also present in all foodstuffs usually at concentrations below 5 mg/kg (WHO, 1981). The daily intake of Mn by man (about 40% of the body pool) is much higher than that of most cationic trace metals. The toxicity depends upon the chemical form and its oxidation state (Venugopal & Luckey, 1978). The morphological effects on plants include marginal chlorosis, necrosis of leaves, leaf puckering, chlorosis of young leaves (resembling Fe deficiency) and necrotic spots on leaves. In severe cases of Mn toxicity, plant roots turn brown (Vlams & Williams, 1973; Foy, 1973, 1984). Other physiological and morphological disorders associated with excess Mn are crinkle leaf, growth retardation, leaf tip burn and fruit cracking at the blossom end (Masui & Ishida, 1975; Keisling *et al.*, 1984).

The main objective of this investigation is to assess the cytotoxic effects of different salts of this element on different plant systems *in vivo*.

MATERIALS & METHODS - The two test chemicals chosen were $MnSO_4$, H_2O and $KMnO_4$ the cationic and bound anionic forms, respectively. The plants were bulbous *Allium sativum* L. and aquatic *Vallisneria spiralis* L.

Vallisneria plants from aquarium of the Departmental garden of the University were transferred to glass jars containing different concentrations of the metallic salts in Knop's nutrient medium. The concentrations used were 1,100 and 10,000 ppm.

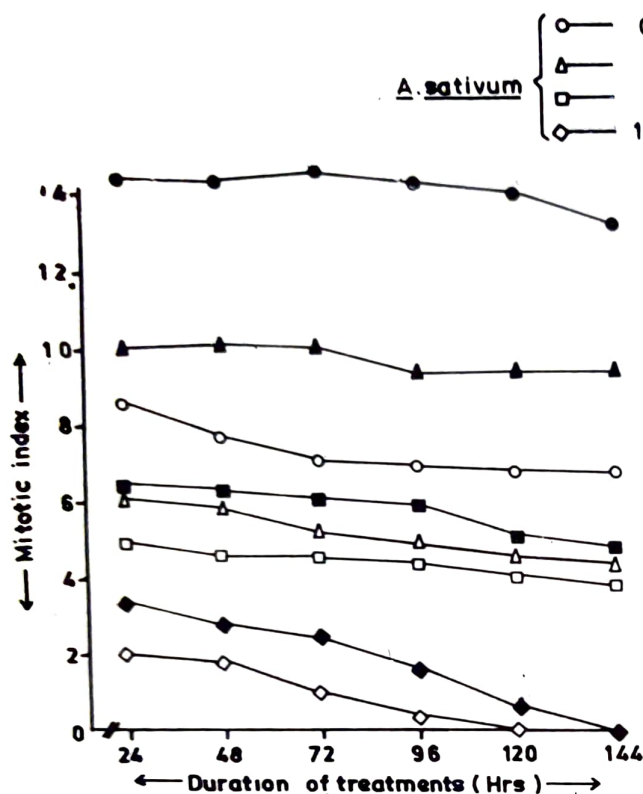
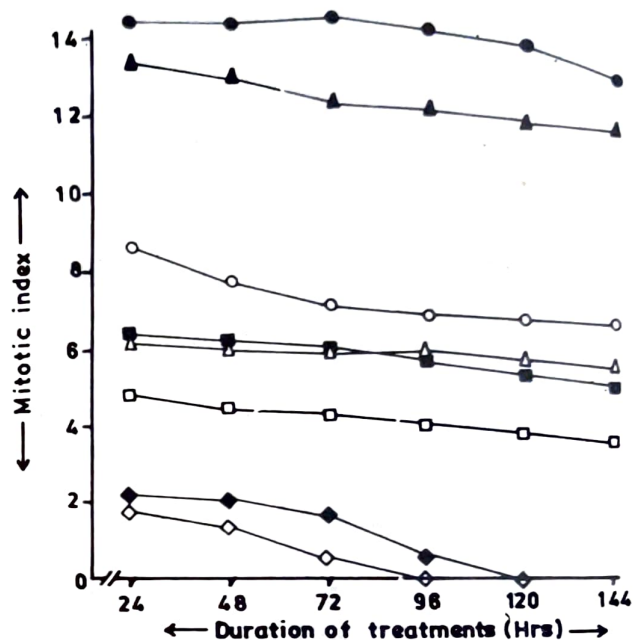
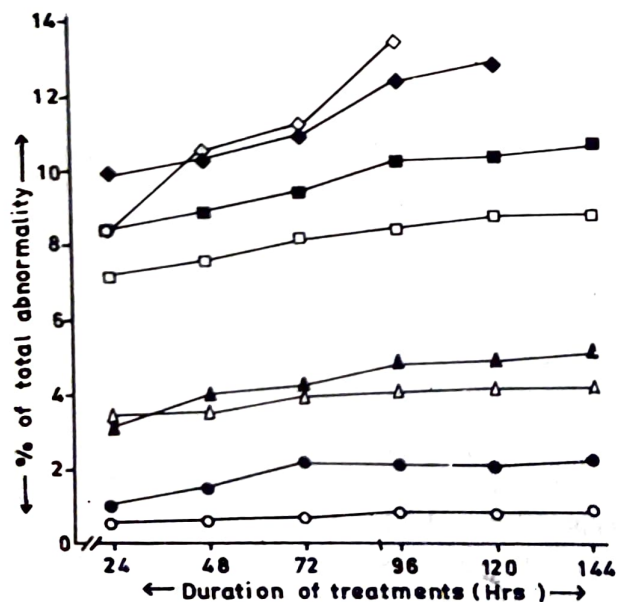
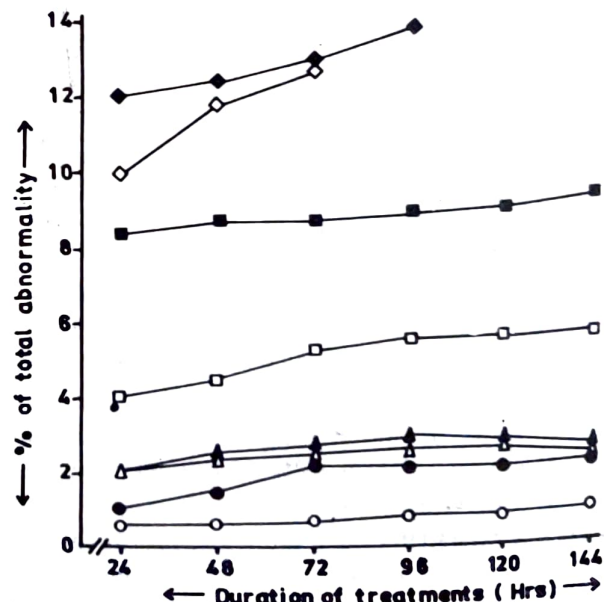
Bulbs of *A. sativum* with healthy roots were kept in wide mouth tubes containing the salt solution.

Meristem assay was carried out from excised *Vallisneria* leaf tips and *Allium* root-tips after every 24h interval for 144h. The tips were squashed following the usual acetic-orcein staining schedule after fixation in acetic-ethanol mixture (1:3). Leaf tips were fixed for overnight and root tips for 1.1/2 h (Sharma & Sharma, 1980). Controls were treated in Knop's solution. The experiments were repeated thrice.

The parameters used were frequency of mitosis and chromosomal abnormalities in 5000 cells per concentration per treatment. The data were analysed statistically following student's 't' test at 5% level.

RESULTS & DISCUSSION - In *Allium cepa* following treatment with 10,000 ppm, the roots were shorter than in the control. This effect was more pronounced with $MnSO_4$, indicating the effects on cell division and elongation contributing to root growth. Treatment for 72 h resulted in 20 mm and 14 mm roots in the bulbs treated with $MnSO_4$ and $KMnO_4$, respectively. In control plants the roots were 36mm. Following treatment with $MnSO_4$, mitotic division gradually decreased in plants. $KMnO_4$ caused more drastic effect on the plants, reducing significantly mitotic index following treatment with 10,000 ppm (Figures 1,2.)

The total abnormalities induced by 10,000 ppm in *Vallisneria sp.* increased in direct proportion with dosage and period of treatment.

Fig. 1 EFFECT OF MnSO_4 ON FREQUENCY OF MITOTIC DIVISIONFig. 2 EFFECT OF KMnO_4 ON FREQUENCY OF MITOTIC DIVISIONFig. 3 EFFECT OF MnSO_4 AS INDICATED BY PERCENTAGE OF ABNORMAL CELLSFig. 4 EFFECT OF KMnO_4 AS INDICATED BY PERCENTAGE OF ABNORMAL CELLS

In the bulbous species, both the salts caused significant effect on total chromosomal abnormalities and the value of aberrations was significantly more with 10,000 ppm than with 1 ppm and 100 ppm solutions.

KMnO₄ caused higher frequency of total abnormalities than MnSO₄ at 10,000 ppm (Fig 3.4.)

The metal at all concentrations induced abnormalities which included aggregation of chromosomes during metaphase, stickiness, diplochromatid, polyploidy, sticky bridge, erosion of chromosomes and pycnotic clotting.

It is clear that the cytotoxicity of the metal in both the plants is dose and time dependent. Though the toxicity is higher in KMnO₄, in some cases yet a steady increase is induced by MnSO₄.

Submerged plants are more affected possibly due to uptake of the metal both through leaf and root systems, as shown earlier by Mukherjee & Dhir. (1987).

In *Allium*, the uptake of the metal is through the root system indicating the tolerance of the plant systems to a metal must be dependent upon physiological conditions of a plant and mode of uptake of the metal.

Mn acts mainly on spindle apparatus as indicated by spindle disturbance and fall in mitotic index which may be attributed to its binding property with cytoplasmic proteins, the action of the chemical on thiol group and on osmotic concentration of the cell sap. The possible chelation of the metal with cyclic groups lead to alterations in the composition and viscosity of the cytoplasm disturbing spindle formations (Sharma, 1985).

It is suggested that different chemical forms of a metal must be used when assessing its cytotoxicity with multiple test systems.

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