SUMMARY

To characterize heavy metal toxicity, studies were carried on the changes in growth and metabolism of germinating rice (Oryza sativa), mungbean (Phaseolus aureus), Lettuce (Lactuca sativa) and wheat (Triticum vulgare) seedlings under the influence of toxic concentrations of mercuric chloride ($HgCl_2$) cupric sulphate ($CuSO_4$, $5H_2O$) and zinc sulphate ($ZnSO_4$, $7H_2O$).

Rice and mungbean seeds were germinated in different concentrations of Hg, Cu and Zn solutions at a constant temperature of 30°C in moist atmosphere in dark. Lettuce was germinated in different concentrations of the above metal solutions in light of 1000 lux provided by fluorescent tubes at a constant temperature of 25°C. Wheat was germinated in dark and the coleoptiles were incubated in dark humid chamber at 25°C. In all the cases, seeds were also germinated in water and designated as control. Growth data were taken after 5 days in the case of rice and mungbean after 7 days in the case of lettuce and after 4 days in the case of wheat. Different hormones were tested for their ability to overcome growth inhibition induced by heavy metal solutions. Interaction of Hg, Cu and Zn with indole-3-acetic acid was studied with wheat coleoptile and interaction with gibberellic acid were studied with lettuce seedlings. Cytological observations were carried out using onion root tips. Respiration studies were made with rice and mungbean seedlings. Chlorophyll content and Hill activity was

measured with rice leaves and isolated leaf discs of Cephalandra sp. Carotene and xanthophyll contents were measured from isolated leaf discs of Cephalandra sp. Anthocyanin content was estimated from the isolated petaloid staminode discs of Canna indica. Induction of thermosensitivity in rice and mungbean seeds by copper pretreatment followed by heat treatment and the protecting effect of hormones were studied. Turbidity, water soluble protein and alkali soluble protein content of homogenates from heat damaged mungbean seeds were estimated. Turbidity was measured from homogenates of $ZnSO_4$ treated rice seedlings. Effects of Hg and Zn solutions on water gain or loss, leakage of UV absorbing materials and alteration of sugar and amino acid contents were studied with potato discs. Conductivity of the leachates from potato discs soaked in different concentrations of Hg, Cu and Zn solutions was measured. Copper uptake was determined from rice and mungbean seedlings at various stages of germination. Sugar and nitrogen estimations were performed with mungbean seedlings treated with Hg and Zn. Water soluble protein contents of Hg and Zn treated rice seedlings were measured. Nucleic acid and alkali soluble protein were estimated from mungbean seedlings. Assay of enzymes were carried on with rice seeds. Interactions of Hg, Cu and Zn with gibberellic acid on α-amylase activity were studied on deembryonated rice half seeds. Gel electrophoretic studies of buffer soluble protein, basic protein and peroxidase isoenzymes were made from rice seedlings grown in Hg, Cu and Zn solutions.

1. In rice seedlings, inhibition of elongation began at 10 mM $ZnSO_4$. The inhibition increased at higher dosage and the effect being more pronounced in root than shoot. Maximum inhibition was obtained at 40 mM. At this concentration both root and shoot suffered from same amount of elongation inhibition. Zn-induced inhibition of both shoot and root in rice seedlings could be reversed partially by GA_3 treatment.

   In mungbean seedlings, the magnitude of elongation inhibition was greater in root than hypocotyl. When GA_3, cAMP and kinetin applied in combination with $ZnSO_4$, partial relief of inhibition was obtained, GA_3 being more effective than cAMP. But kinetin showed no remarkable effect.

2. When Hg, Cu and Zn was applied alone, there was a gradual reduction in the length of wheat coleoptile sections as compared to control (sucrose phosphate buffer), the magnitude of reduction increasing with increasing concentration of the metal solutions. When IAA was combined with Hg, Cu and Zn solutions, variable degree of prevention of inhibition was obtained. At 10 mM $ZnSO_4$, however, IAA-reversal was not only complete, but the coleoptile section growth exceeded that of the control.

3. The cytological observations with onion root tip cells revealed that although Hg, Cu and Zn did not affect the number of cells

undergoing mitosis, it caused disturbed cell division showing a series of abnormalities.

4. There was always a decrease in the respiration rate by Hg and Zn treatment in both rice and mungbean seedlings. The rate of inhibition on per seedling basis was greater than that on fresh weight basis.

5. There was drastic reduction in total chlorophyll in leaves of Hg, Cu and Zn treated rice seedlings.

In isolated leaf discs of Cephalandra sp. a rapid decline in chlorophyll content within 3 days was the result of Hg-treatment whereas a lag phase of 3 days in chlorophyll decay was noticed in the case of Cu and Zn. The pattern of decrease of carotene was almost similar to that of chlorophyll while xanthophyll content changed but little in control and treatment sets.

Hill activity of chloroplasts obtained from Hg, Cu and Zn treated rice leaves and isolated leaf discs of Cephalandra sp. suffered a great reduction in presence of metals.

6. Rice and mungbean seeds can be made more sensitive to heat shock by treatment with $CuSO_4$ than with pure water prior to heat treatment. In rice the magnitude of inhibition was more severe in dark than in light but the reverse was obtained in the case of mungbean. Hormones, when applied simultaneously

with copper, can prevent the effect of heat treatment, GA_3 being more effective than either cAMP or kinetin.

Turbidity, water soluble protein and alkali soluble protein content of homogenates, obtained from heat damaged mungbean seedlings were always increased with increasing concentration of $CuSO_4$.

7. Turbidity of homogenates obtained from $ZnSO_4$ treated rice seedlings was always increased over control.

8. In control, water uptake increased progressively with increasing soaking periods and Hg treatments induced significant loss of water. The water loss increased with increasing imbibition period. While Zn treatment showed no water loss but the rate of water gain was always less than control.

9. The quantity of UV absorbing materials leached out from potato discs soaked in Hg and Zn was always higher than that of the corresponding in water. The UV absorption spectra after imbibition for different hours were basically of the same nature in quality, differences being noticed in the higher optical density values after longer imbibition periods. Hg and Zn treatment increased both amino acid and sugar leachings from potato discs. Amino acids leached out was always in excess than sugar irrespective of control and treatment sets.

10. As compared to control, the conductivity of leachates obtained from potato discs was progressively increased with increasing soaking periods and concentrations of metal solutions.

11. Changes in growth of rice and mungbean seedlings and the uptake of copper as influenced by various concentrations of $CuSO_4$ showed a clear positive correlation between the amount of Cu-uptake and seedling age on one hand and the intensity of treatments on the other.

12. In axis, total sugar was reduced by Hg and Zn treatment. But in cotyledon total sugar was marked by a slight increment. Similarly, reducing sugar contents of Hg and Zn treated axis were always maintained at distinctly lower level than control, but cotyledons showed slight stimulation of reducing sugar content. Non reducing sugar decreased in $HgCl_2$ treated axis but increased in Zn treated axis. In cotyledons, nonreducing sugar contents were reduced in both Hg and Zn treatments.

13. Total nitrogen content in axis progressively declined with increase in Hg and Zn concentrations and in cotyledon this was maintained at higher level than control. Soluble nitrogen content of Hg and Zn treated mungbean axis and cotyledon also showed similar trend after Hg and Zn treatment.

14. Hg and Zn treated rice seedlings showed a declining trend of water soluble protein content when expressed as per seedling basis, but a progressive increment occurred on per g fresh wt. of seedling. Endosperm protein was increased in both per endosperm and per g fresh weight of tissue. The relative rate of increment of protein content in endosperm of Zn treated seeds was greater than Hg treated ones.

15. Hg and Zn treatments decreased the DNA and RNA content in mungbean axis. But in cotyledons a progressive increment was the result. The alkali soluble protein content of mungbean axis decreased when expressed as per 10 axis basis, but slightly stimulated on per gm fresh axis. A progressive accumulation of the amount of protein in Hg and Zn treated cotyledons was obtained on both per g fresh tissue and per 10 cotyledons basis respectively.

16. Catalase activity showed progressive increase with increasing concentration of Hg and Zn when expressed as per mg protein basis. Hg treated seedlings showed a slight fall in catalase activity. Zn treatment showed decreasing trend of catalase activity when expressed as per seedling basis.

Peroxidase and IAA oxidase activity was increased both on per seedling and mg protein basis. But IAA synthase activity on per mg protein basis showed increment after Hg and Zn treatment.

Hg treated seedling showed a slight fall in activity at 0.4 mM concentration, but a gradual increment occurred at the next higher doses, whereas Zn treated seedlings showed a progressive diminution of activity with the increasing concentrations. α-amylase activity of endosperm, ATPase activity of embryo and phytase activity of endosperm declined with increasing Hg and Zn concentrations when expressed both as per plant part and mg protein.

RNase activity of Hg treated embryo showed a progressive diminution with the increasing concentration both on per mg protein and seedling basis. But in Zn treated embryo a declining trend was obtained when expressed as per seedling basis. On the other hand, the activity on per mg protein was gradually increased after $ZnSO_4$ treatment. RNase of Hg treated endosperm was decreased both on per endosperm and mg protein basis, whereas RNase activity of Zn treated endosperm showed a progressive increment on per endosperm basis but a gradual diminution was obtained when expressed as per mg protein.

Ascorbic acid oxidase activity, when expressed as per mg protein and seedling showed progressive increase consistent with increased concentration of Hg and Zn. Chlorophyllase activity of rice leaves from seeds germinating in presence of Hg, Cu and Zn solutions showed an increasing trend proportional to

concentrations of Hg, Cu and Zn solutions.

17. There were some changes in the gel electrophoretic band pattern of buffer soluble, basic protein and peroxidase isoenzymes in Hg, Cu and Zn treated rice seedlings.