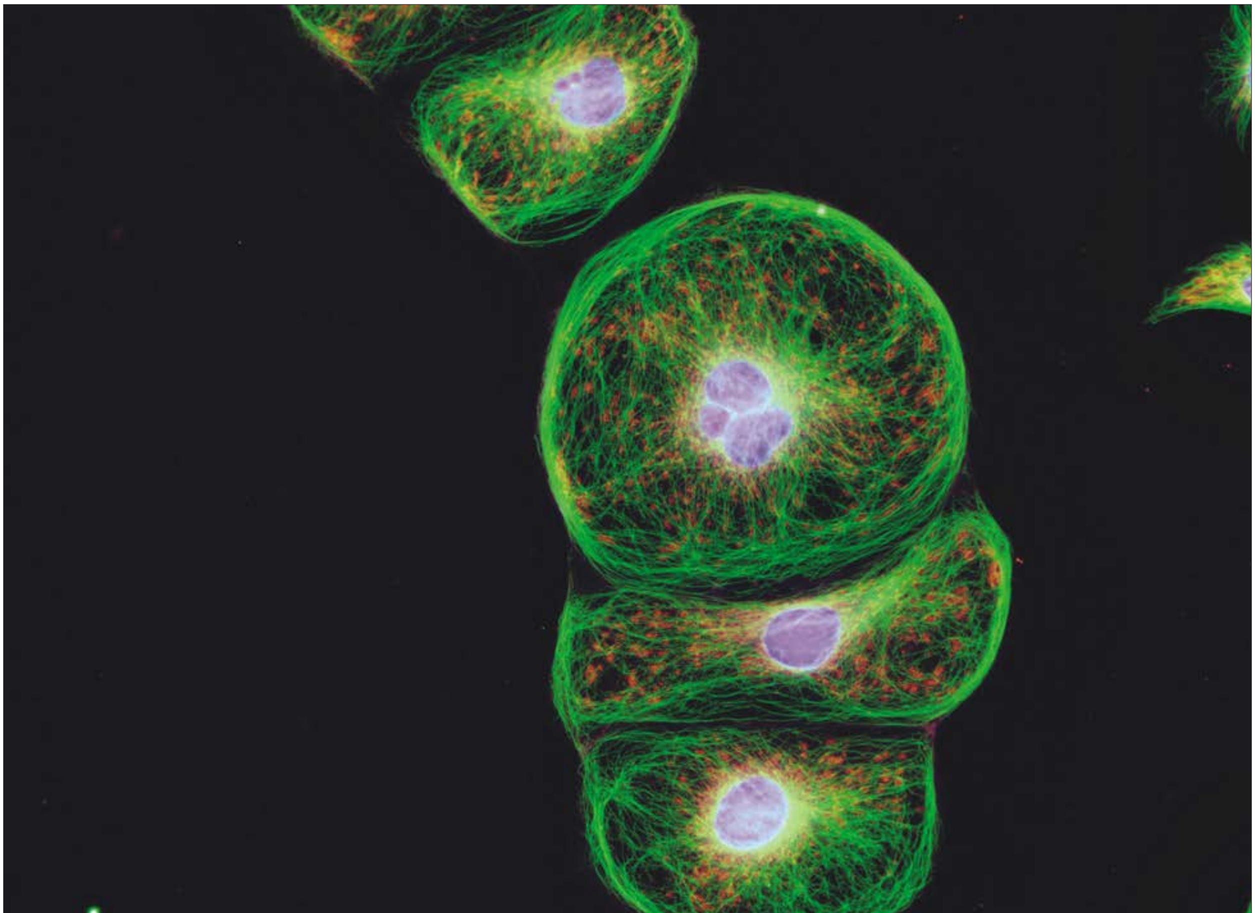

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Effect of Toxic Metal Mercury on Histomorphology of *Cymopsis tetragonoloba*

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Abstract: Histomorphology of the hypocotyl of *Cymopsis tetragonoloba* were studied following, treatment with toxic mercury (Hg) metal. The data was collected after 15 days and compared with control. It was observed that seed germination process of *Cymopsis tetragonoloba* was totally inhibited at all experimental condition of applied Hg. The germinated seeds showed abnormal growth. Development of xylem was effected, straight T-shaped tissues was formed and phloem was of small patch in cortical region. Parenchymatous cells, cortex and pith were large with intercellular spaces.

Key words: Hg, hypocotyl, *Cymopsis tetragonoloba*, vascular tissues

Introduction

Mercury is a naturally occurring element. Metallic mercury is highly toxic, non essential, persistent, immutable and non-biodegradable metal that undergoes many changes during transfer through different torphic levels of the food chain (Botkin and Keller, 1995). Heavy metals are known to causes irreversible damage to a number of vital metabolic constituents and important bimoloecules and also cause irreversible injury to the plants cell membrane, especially vascular bundles (Assche and Clijsters, 1990). Zn, Ni, Cu, V, Co, W and Cr are toxic elements with high or low importance as trace element. As Hg, Ag, Cd, Pb and U have no known function as nutrients and seems to be more toxic to plants and ecosystem (Koplik and Nemcova, 1983). The toxic threshold level of the metal in the tissue is defined by the stress point for metal toxic and beyond this level the physiological state of the cell will be irreversibly damaged (Chanda and Parmer, 2003). This change is reflected by change in the histomorphology of the plants (Reddy and Prasad, 1992). Parmar and Chanda (2005) reported that seedlings treated with lower concentration of mercury had similar activities to those treated with higher concentration of chromium. Heavy metals are observed to be growth inhibitors, as in the leaves shows decrease in the number of open stomata and thereby O_2 deficiency in plant (Filter and Hay, 1981). Stoyanova (1998) reported that heavy metals bring about anatomical changes in primary leaves hence treatment induced changes in the shape of pallisade cells. Moreover, effect of heavy metals on plants include reduction in growth parameters (Dalal *et al*., 1985) and disruption in the metabolic processes of living organism (Moran *et al*.,1986). Cadmium and mercury showed drastic effects at high concentration and longer duration with regards to seedling growth and metabolism (Neelima and Reddy, 2003).Metal toxicity also effects on reduction of uptake of basic nutrient mineral ions, which may attributed to the weakness of plant and ultimately death (Azmat and Ravida, 2005). Physiological processes of bean plants were also effected and amino acids were found to be reduced in presences of toxic metal and accumulation of metal appears in the from of yellow leaves (Azmat *et al*.,

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2005). The main objective of the present study was to investigate the effect of Hg on germination and histomorphology of the *Cymopsis tetragonoloba*.

Materials and Methods

Seeds of *Cymopsis tetragonoloba* were soaked in tap water for 3 h. They were thoroughly washed with tap water, followed by distill water and strelized by tween 20 and than transferred into pots containing different concentration of mercuric chloride (5, 25, 50, 75 and 100 ppm) based hoagland's solution. Fifteen seeds in each pot were placed on the net. The seeds were allowed to germinate at 25±°C in growth chamber for two weeks in December 2004. The hypocotyls were demarcated into two segments with marker. The segment near the root was designated as the lower segment and that near the cotyledon was designated as the upper (Nayan *et al*., 2005). Transverse Section (T.S.) of the hypocotyl of mercury treated seedlings and observed under a compound light microscope and histomorphology of hypocotyl was checked and drawn by pencil on to the paper. Seed germination processes and tissue structure of *Cymopsis tetragonoloba* compared with control one.

Results and Discussion

The term heavy metal includes more than one definitions changing according to the branches of science where it is included. From the agroindustrial point of view, it can be cultures or chemical elements that decrease agricultural productivity, due to its presence at high concentration in soil. Elements like Pb, Cd, Hg, As, U are non essential elements and found in several effluents originating from batteries, electronic materials, plastics industries, as well as the electroplating industry. The uncontrolled discharge of these in soil and water bodies, highly contaminates environmental sites. Also burning residues for industrial activities may causes partial volatization of those elements contaminating atmosphere. Even today the contaminated water of changer lake (Sindh) is discharged into Indus water which is used for drinking purpose and causes the death and severe gastric disease in the Sindh region.

Germination

Mercuric chloride caused greater reduction on germination percentage as compared to control. Germination was inhibited significantly as the concentration of mercury increased (Table 1). Figure 2-5 shows considerable decrease in germination up to 100 ppm of Hg. Where as no leaf formation was observed at 75 ppm of Hg even after passing six weeks and roots of plant were more effected. Figure 4 shows that only one reduced size leaf was formed in one seedling of *Cymopsis tetragonoloba* at 75 ppm of Hg where as in second seedling roots were more affected.

Table 1: Percentage germination of seeds in control and treated seedlings of *Cymopsis tetragonoloba*

Hg (ppm)	Germination of seeds (%)
0	75.0±0.01
5	33.3±0.03
25	31.2±0.03
50	26.6±0.02
75	13.3±0.01
100	12.1±0.01

Fig. 1: Growth of hypocotyl of control plant

Fig. 2: Growth of hypocotyl of 5 ppm of Hg

Fig. 3: Growth of hypocotyl of 50 ppm of Hg

External Morphology

In control after two weeks almost all the seeds were germinated and showed normal growth. Figure. 1 shows the hypocotyl of *Cymopsis tetragonoloba* after one week. At 5 ppm of Hg (Fig. 2) the growth of hypocotyl was effected and at 50 ppm of mercury abnormal elongation of hypocotyl was observed (Fig. 3) Seedlings observed in mercury treated plant were weak and no leaves formation in pots were observed after passing six weeks of growth. Roots were more effected due which plants experienced more rapid death.

Fig. 4: Growth of Seedlings after six weeks in 75 ppm of Hg

Fig. 5: Growth of Seedlings after six weeks in 100 ppm of Hg

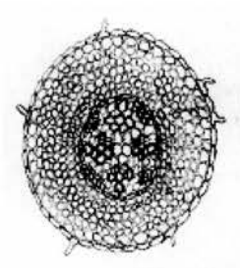

Fig. 6: T.S. of hypocotyl of *Cymopsis tetragonoloba* of control plant

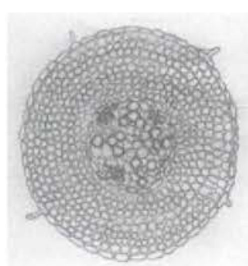

Fig. 7: T.S. of hypocotyl of *Cymopsis tetragonoloba* of 25 ppm of Hg

Table 2: Effect of different experimental conditions of mercury on histomorphology of hypocotyls of *Cymopsis tetragonoloba*

Treatments	Epidermis	Cortex	Endodermis	Phloem	Xylem	Pith
Control (ppm Hg)	Single layered	13 layered	1 barrel shaped	4 small patches	4 umbrella shaped	Large, rounded cells
5	-Do-	15 layered	-Do-	-Do-	-Do-	-Do-
25	1 unicellular hair	15 small cells	-Do-	-Do-	y-shaped small cell	-Do-
50	bilayered	17 small sized cells	1 hexagonal cells	-Do-	-Do-	-Do-
75	-Do-	-Do-	-Do-	-Do-	T-shaped	-Do-
100	-Do-	-Do-	-Do-	-Do-	Flying bird	-Do-

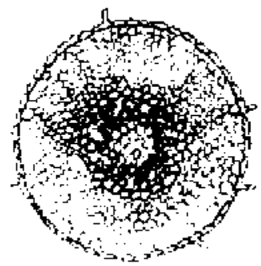

Fig. 8: T.S. of hypocotyl of *Cymopsis tetragonoloba* of 75 ppm of Hg

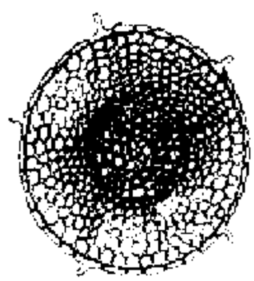

Fig. 9: T.S. of hypocotyl of *Cymopsis tetragonoloba* of 100 ppm of Hg

Histomorphology

The present investigation of applied mercury on plant growth showed inhibition in growth parameters as well as on tissue structure (Chanda and Parmer, 2003) of hypocotyls. It is also observed that mercury reduced the area of stem and leaves (Fig. 2-5). T.S. of treated hypocotyl showed the effect of mercury on the thickness of cortical region and shape of xylem tissues. T.S. of treated hypocotyl with increased concentration of Hg showed an increase in the cortical region with reduced cell size as compared with control (Fig. 7-9) i.e., in T.S. of hypocotyl of control Fig. 6 there are more or less twelve layers of large parachymetous cell (Table 2). Water absorbed by the root hair is directed towards the cortex from where it entered into the xylem. But accumulation of Hg change the tissue structure and T.S. of hypocotyl shows different structure of xylem. In 25 ppm of Hg it is Y-shaped while in 75 ppm Hg T-shaped. This change in number of cells and structure may relate with accumulation of Hg which effect on rate of absorption of water and mineral which ultimately attributed to the growth inhibitory effect of germination of seedling (Stoyanova, 1998).

Conclusions

Mercury is a metal with no known biological function and carcinogenic to the biological system. Here we conclude that accumulation of Hg effect the structure of the cell due to which transportation of water from the roots to the tips of new leaves were reduced which shows pronounced effect on germination, growth, stem and leaves of the plant and also damaged the root system of the seedling which attributed to the germination process.

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