

**EFFECT OF CHROMIUM AND MANGANESE METAL ON
BIOMASS AND GROWTH RATE OF SOME PULSES****K. V. SONI* AND B. D. BHUVA***School Of Sciences, R. K. University, Rajkot, India***ABSTRACT**

Metals are essential micro molecules required by living cell for biochemical reactions. Several metals are essential and useful to cells while few show adverse effects on living forms. Chromium and manganese are commonly counted as important metals to plant cells but sometimes their higher concentration is harmful to plant cells. Present study focused on of effects of chromium and manganese on biomass and growth rate of three widely cultivated pulses i.e. *Glycine max*, *Vigna unguiculata* and *Vigna aconitifolia*. It was found that higher concentration of manganese showed reduction in biomass and growth rate of all three investigated plants. However, chromium poses adverse effect on biomass and growth rate of experimental plants even at low concentrations.

KEY WORDS: Chromium, Manganese, Effects, Growth Rate, Biomass

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INTRODUCTION

Life on this planet has evolved in the presence of metals. Metals have been mined and used since ancient times. Cells learned to make use of the more abundant metals in the Archean oceans at an integral component in their structure and function. Today, we inherit these as the essential metals. The industrial era has seen a sharp increase in both the amounts and variety of metals that have application in industry¹⁰. All things in nature ultimately succumb to decay. Much of this is a natural consequence of the laws of thermodynamics. Many molecules degrade by the action of oxygen, halogens and radicals naturally found in the environment⁶³. Modern industry is to a large degree, responsible for contamination of the environment. Lakes, rivers and oceans are being overwhelmed with bacteria, and wastewater. Among toxic substances reaching hazardous levels are heavy metals⁶⁷. Many uses of heavy metals in several applications lead to their wide distribution in soil, slit, waste and wastewater. Such a pollution of the environment by toxic metals and radio nucleotides arises as a result of many human activities, largely industrial, although sources such as agriculture and sewage disposal also contribute¹³. Heavy metals are among the conservative pollutants that are not subject to bacterial attack or other breakdown or degradation process and are permanent additions to the environment^{16, 34}. These metal contaminants pose adverse health effects to those who live near these polluted sites. Breathing, eating, drinking, and skin contact are all possible exposure routes for metal contaminants. Metals such as mercury, lead, and arsenic, potentially can be toxic to the kidneys, decrease mental capabilities, and cause weakness, headaches, abdominal cramps, diarrhea and anemia⁶⁶. Chronic exposure to these pollutants can cause permanent kidney and brain damage^{66, 1}. To solve the water pollution problem by toxic heavy metal contamination resulting from human's technological activities has for long presented a challenge⁶⁷. A key factor to the remediation of metals is that metals are non-biodegradable, but can be transformed through sorption, methylation, and complexation, and changes in valence state. These transformations affect the mobility and

bioavailability of metals. Adsorption, ion exchange, precipitation and complexation with organic matter are mechanisms that limit the amount of metal leaching through surface water or groundwater¹¹. There are about 50 metals that are studied with respect to the toxicological importance to plants, animals and man. Such metals accumulate in soil to reach the plant through roots during water absorption and cause serious adverse effect on plants viz., inhibition of seed germination, growth of seedlings and reduction of yield. Though some investigations have been carried out in India throwing light on various aspects of the accumulation and effect of heavy metals in plants, yet such study is not sufficient especially in certain agricultural plants of Gujarat state. Although a number of techniques have been developed to remove metals from contaminated soils, many sites remain contaminated because economic and environmental costs to clean up those sites with the available technologies are too high⁵². As a rule in nature anything that is present on this earth should be either degraded out or recycled. Heavy metals cannot be degraded out but they can be recycled by changing their ionic stage. Any kind of biomass can be easily degraded out in nature. Several modes of biotechniques are named as biosorption, phyto-sorption, bioaccumulation, phyto-accumulation, bio-extraction, phyto-extraction, rhizofiltration and rhizodegradation, microorganism stimulation and mobilization, phyto-stabilization and phyto-volatilization etc. Phytosorption is the technique where plants or plant materials are used to absorb heavy metals. In phyto-accumulation technique plants are used to absorb heavy metals and they are stored in plant parts. Contamination of heavy metals in biosphere increased drastically since 1900 and expressed severe health and the environmental problems throughout the world^{53, 18}. The plants that are used for phyto-extraction have tolerance towards the metal(s) targeted and efficient to translocate them from below ground parts to areal parts⁵. The present work was focused towards the toxic effects of chromium and manganese metal on biomass and growth rate on widely cultivated pulses

Glycine max, *Vigna unguiculata* and *Vigna aconitifolia*.

MATERIALS AND METHODS

The study was carried out in Rajkot city area (22° 17' Lat. and 70° 49' Lon.). Experiments on seedling emergence and seedling growth were performed on a coarse loam soil found in the natural habitats where the selected plants cultivated by seed germination. Soil was collected from natural habitats, air dried and passed through a 2 mm sieve. For the study of the effect of chromium and manganese on plant growth rate and biomass development, the soil was mixed with heavy metal salts and prepared for the cultivation of experimental plants. The chromium and manganese metals were used in the form of potassium dichromate salt (K₂Cr₂O₇) and manganese sulphate (MnSO₄ H₂O) respectively. The metal salts were mixed in eight different lots of soil (each lot of 10kg) at 3.3, 6.6, 10.0, 13.3, 16.6, 19.9, 23.2 and 26.6 grams for chromium and 1.04, 2.0, 3.1, 4.2, 5.2, 6.24, 7.3 and 8.3 gram for manganese to get 0.2, 0.4, 0.6, 0.8, 1.0, 1.2, 1.4 and 1.6 mM concentrations of metal salts. The soil mixed with metal salt was placed in polyethylene bags and cultivation of experimental plants was carried out in these bags. The soil without metal salt was control. The initial metal concentration of control soil was negligible and considered as zero. Tap water was added to the soil in polyethylene bags to field capacity and then allowed to dry for 6 days. The seeds of *Glycine max*; *Vigna unguiculata* and *Vigna aconitifolia* were collected from Sanjiv Agro Center, Rajkot. Metal salt mixed soils were then raked with fingers and seeds were sown after surface sterilization with H₂O₂. Ten seeds were sown in each bag at the depth of about 8-10 mm in evening. Immediately after sowing soils were watered and then after watering was carried out at alternate days. All the seedlings in each bag for each metal concentration were allowed for germination. The study was carried out twice. The results are average of the study of these two sets of germination. After specific time duration plants were harvested in such a way that the tap root and root hairs were not damaged or damage was minimum. Soil particles were removed from the root by gentle washing. The plants

collected for the study brought in the laboratory, washed with water and carefully blotted on the blotting sheets after washing to remove moisture on their surface. The length of entire plant was measured. The mean of 20 measurements was calculated as final reading. The growth rate of control and treated plant was studied on the basis of length of entire plant five weeks after the germination. The method of Hunt (1978) was used to study the biomass of experimental plants. The fresh weight of root, stem and leaves was determined separately after blotting in the laboratory. They were cut into small pieces after weighing and placed in brown paper bags separately and kept in oven at 80° C for a period of 8 days for uniform drying. The dry weight of these organs was recorded.

RESULTS

Presence of heavy metal affects growth performance of plants. They showed marked differences in fresh weight and dry weight of root, stem and leaf. The data regarding the fresh weight and dry weight are presented in Table 1 to 12.

Effect of chromium on fresh weight

The fresh weight in root, stem and leaf of *Glycine max* was 9.97, 13.29 and 4.21 gm respectively which was reduced to 6.19 gm in root, 8.21 mg in stem and 2.86 gm in leaf by 0.2 mM chromium treatment (Table 1 - 3). The fresh weight of root, stem and leaf of *Vigna unguiculata* was lower than control at 0.2 mM chromium concentration (Table 1 - 3). In *Vigna aconitifolia* the root, stem and leaf fresh weight was decreased at 0.2 mM concentration of chromium in the treatment. Germination was not recorded at 0.4 - 1.6 mM concentrations of chromium in all three investigated plants (Table 1 - 3).

Effect of manganese on fresh weight

The proportion of fresh weight of *Glycine max* root, stem and leaves was reduced by manganese treatment and was lower than control (Table 4 - 6). In *Vigna unguiculata* root and stem fresh weight was lower than control in all treatments of manganese. In leaf the fresh weight was gradually decreased by increasing the manganese concentration in

treatment. The lowest fresh weight was reported at 1.6 mM manganese concentration in all three organs (Table 4 - 6). In *Vigna aconitifolia* the root fresh weight was gradually decreased by increasing the concentration of

manganese and remained lower than the fresh weight of control (Table 4). Similar results were found for the stem and leaf (Table 5 and 6).

Table 1
Effect of chromium on root fresh weight

Concentration (mM)	Glycine (gm)	max	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	9.97		11.34	10.26
0.2	6.19 ± 0.01		7.58 ± 0.01	7.37 ± 0.01
0.4	N.G.		N.G.	N.G.
0.6	N.G.		N.G.	N.G.
0.8	N.G.		N.G.	N.G.
1.0	N.G.		N.G.	N.G.
1.2	N.G.		N.G.	N.G.
1.4	N.G.		N.G.	N.G.
1.6	N.G.		N.G.	N.G.

Table 2
Effect of chromium on stem fresh weight

Concentration (mM)	Glycine (gm)	max	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	13.29		16.45	17.35
0.2	8.21 ± 0.01		10.75 ± 0.01	10.13 ± 0.01
0.4	N.G.		N.G.	N.G.
0.6	N.G.		N.G.	N.G.
0.8	N.G.		N.G.	N.G.
1.0	N.G.		N.G.	N.G.
1.2	N.G.		N.G.	N.G.
1.4	N.G.		N.G.	N.G.
1.6	N.G.		N.G.	N.G.

Table 3
Effect of chromium on leaf fresh weight

Concentration (mM)	Glycine (gm)	max	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	4.21		5.12	5.63
0.2	2.86 ± 0.01		3.29 ± 0.01	2.75 ± 0.01
0.4	N.G.		N.G.	N.G.
0.6	N.G.		N.G.	N.G.
0.8	N.G.		N.G.	N.G.
1.0	N.G.		N.G.	N.G.
1.2	N.G.		N.G.	N.G.
1.4	N.G.		N.G.	N.G.
1.6	N.G.		N.G.	N.G.

N.G. = no germination

Table 4
Effect of manganese on root fresh weight

Concentration (mM)	Glycine (gm)	max	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	11.25		15.85	13.28
0.2	10.63 ± 0.16		15.32 ± 0.04	13.25 ± 0.01
0.4	10.56 ± 0.16		14.89 ± 0.04	12.18 ± 0.01
0.6	10.87 ± 0.16		14.29 ± 0.04	11.60 ± 0.01
0.8	11.11 ± 0.16		12.99 ± 0.04	10.65 ± 0.01
1.0	11.04 ± 0.16		12.63 ± 0.04	10.20 ± 0.01
1.2	10.70 ± 0.16		12.70 ± 0.04	9.47 ± 0.01
1.4	9.39 ± 0.16		12.54 ± 0.04	8.95 ± 0.01
1.6	8.31 ± 0.16		12.47 ± 0.04	8.59 ± 0.01

Table 5
Effect of manganese on stem fresh weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	14.92	20.67	18.10
0.2	13.64 ± 0.28	19.71 ± 0.04	17.37 ± 0.01
0.4	12.70 ± 0.28	19.47 ± 0.04	16.79 ± 0.01
0.6	13.69 ± 0.28	18.74 ± 0.04	16.27 ± 0.01
0.8	13.72 ± 0.28	17.09 ± 0.04	15.67 ± 0.01
1.0	12.68 ± 0.28	17.07 ± 0.04	14.97 ± 0.01
1.2	12.46 ± 0.28	16.68 ± 0.04	14.36 ± 0.01
1.4	12.39 ± 0.28	17.09 ± 0.04	13.84 ± 0.01
1.6	12.35 ± 0.28	16.63 ± 0.04	13.34 ± 0.01

Table 6
Effect of manganese on leaf fresh weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	3.70	6.30	5.09
0.2	3.58 ± 0.04	6.01 ± 0.01	4.89 ± 0.01
0.4	3.55 ± 0.04	5.85 ± 0.01	4.68 ± 0.01
0.6	3.26 ± 0.04	5.59 ± 0.01	4.41 ± 0.01
0.8	2.86 ± 0.04	5.15 ± 0.01	4.21 ± 0.01
1.0	2.80 ± 0.04	5.02 ± 0.01	3.96 ± 0.01
1.2	2.83 ± 0.04	4.94 ± 0.01	3.71 ± 0.01
1.4	2.86 ± 0.04	4.89 ± 0.01	3.47 ± 0.01
1.6	2.67 ± 0.04	4.66 ± 0.01	3.14 ± 0.01

Table 7
Effect of chromium on root dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	8.91	9.02	8.79
0.2	5.40 ± 0.01	6.85 ± 0.01	6.54 ± 0.01
0.4	N.G.	N.G.	N.G.
0.6	N.G.	N.G.	N.G.
0.8	N.G.	N.G.	N.G.
1.0	N.G.	N.G.	N.G.
1.2	N.G.	N.G.	N.G.
1.4	N.G.	N.G.	N.G.
1.6	N.G.	N.G.	N.G.

Table 8
Effect of chromium on stem dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	10.36	11.23	11.52
0.2	5.86 ± 0.01	6.17 ± 0.01	6.76 ± 0.01
0.4	N.G.	N.G.	N.G.
0.6	N.G.	N.G.	N.G.
0.8	N.G.	N.G.	N.G.
1.0	N.G.	N.G.	N.G.
1.2	N.G.	N.G.	N.G.
1.4	N.G.	N.G.	N.G.
1.6	N.G.	N.G.	N.G.

Table 9
Effect of chromium on leaf dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	2.92	3.94	3.97
0.2	1.81 ± 0.01	1.95 ± 0.01	1.73 ± 0.01
0.4	N.G.	N.G.	N.G.
0.6	N.G.	N.G.	N.G.
0.8	N.G.	N.G.	N.G.
1.0	N.G.	N.G.	N.G.
1.2	N.G.	N.G.	N.G.
1.4	N.G.	N.G.	N.G.
1.6	N.G.	N.G.	N.G.

N.G. = no germination

Table 10
Effect of manganese on root dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	9.68	13.63	11.42
0.2	9.14 ± 0.11	13.59 ± 0.02	11.40 ± 0.01
0.4	9.08 ± 0.11	12.80 ± 0.02	10.47 ± 0.01
0.6	9.35 ± 0.11	12.29 ± 0.02	9.98 ± 0.01
0.8	9.33 ± 0.11	10.91 ± 0.02	8.94 ± 0.01
1.0	9.16 ± 0.11	10.48 ± 0.02	8.47 ± 0.01
1.2	8.77 ± 0.11	10.41 ± 0.02	7.76 ± 0.01
1.4	7.51 ± 0.11	9.95 ± 0.02	7.16 ± 0.01
1.6	6.48 ± 0.11	9.80 ± 0.02	6.70 ± 0.01

Table 11
Effect of manganese on stem dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	12.83	17.78	15.57
0.2	11.73 ± 0.21	16.95 ± 0.02	14.94 ± 0.01
0.4	10.92 ± 0.21	16.74 ± 0.02	14.44 ± 0.01
0.6	11.77 ± 0.21	16.12 ± 0.02	13.99 ± 0.01
0.8	13.20 ± 0.21	14.35 ± 0.02	13.16 ± 0.01
1.0	13.01 ± 0.21	14.16 ± 0.02	12.42 ± 0.01
1.2	10.22 ± 0.21	13.68 ± 0.02	11.78 ± 0.01
1.4	9.91 ± 0.21	13.67 ± 0.02	11.07 ± 0.01
1.6	9.63 ± 0.21	12.97 ± 0.02	10.40 ± 0.01

Table 12
Effect of manganese on leaf dry weight

Concentration (mM)	<i>Glycine max</i> (gm)	<i>Vigna unguiculata</i> (gm)	<i>Vigna aconitifolia</i> (gm)
Control	3.18	5.41	4.38
0.2	3.08 ± 0.21	5.17 ± 0.02	4.20 ± 0.01
0.4	3.05 ± 0.21	5.03 ± 0.02	4.02 ± 0.01
0.6	2.80 ± 0.21	4.81 ± 0.02	3.80 ± 0.01
0.8	2.40 ± 0.21	4.33 ± 0.02	3.54 ± 0.01
1.0	2.32 ± 0.21	4.17 ± 0.02	3.29 ± 0.01
1.2	2.32 ± 0.21	4.05 ± 0.02	3.04 ± 0.01
1.4	2.29 ± 0.21	3.91 ± 0.02	2.78 ± 0.01
1.6	2.08 ± 0.21	3.63 ± 0.02	2.45 ± 0.01

Effect of chromium on dry weight

The root, stem and leaf dry weight of *Glycine max* was 8.91, 10.36 and 2.92 gm respectively which was reduced to 5.40, 5.86 and 1.81 gm at 0.2 mM chromium treatment (Table 7 - 9). The root, stem and leaf dry weight of *Vigna*

unguiculata and *Vigna aconitifolia* was reduced and found lower than control at 0.2 mM chromium treatment. No germination was observed due to 0.4 - 1.6 mM chromium treatments (Table 7 - 9).

Effect of manganese on dry weight

The root dry weight of *Glycine max* was reduced and was lower than control due to the treatment of different concentrations of manganese (Table 10). The stem dry weight was also lower than control due to manganese treatments (Table 11) except at 0.8 and 1.0 mM manganese concentrations (Table 11). The leaf dry weight was lower than the control (Table 12). In *Vigna unguiculata* and *Vigna aconitifolia* root, stem and leaf dry weight was decreased by increasing the manganese concentration in treatment (Table 10 - 12). The lowest dry weight of all three organs was

reported at 1.6 mM manganese concentration (Table 10 - 12).

Effect of chromium on growth rate

In *Glycine max*, *Vigna unguiculata* and *Vigna aconitifolia* the length of entire plant was reduced at lower chromium concentration in the treatment. Germination was not observed at 0.4 - 1.6 mM chromium concentrations in these plants (Table 13 - 15).

Effect of manganese on growth rate

Manganese showed reduction in growth of *Glycine max*, *Vigna unguiculata* and *Vigna aconitifolia* (Table 13 - 15).

Table 13
Effect of heavy metals on *Glycine max* growth rate

Concentration (mM)	Length of plant (cm)		
	Control	Chromium	Manganese
Water	52.00	-	-
0.2	-	4.90 ± 0.01	49.20 ± 0.17
0.4	-	N.G.	48.80 ± 0.17
0.6	-	N.G.	50.30 ± 0.17
0.8	-	N.G.	51.70 ± 0.17
1.0	-	N.G.	51.10 ± 0.17
1.2	-	N.G.	49.60 ± 0.17
1.4	-	N.G.	43.90 ± 0.17
1.6	-	N.G.	38.40 ± 0.17

Table 14
Effect of heavy metals on *Vigna unguiculata* growth rate

Concentration (mM)	Length of plant (cm)		
	Control	Chromium	Manganese
Water	70.00	-	-
0.2	-	3.60 ± 0.01	64.00 ± 0.27
0.4	-	N.G.	67.50 ± 0.27
0.6	-	N.G.	62.70 ± 0.27
0.8	-	N.G.	48.80 ± 0.27
1.0	-	N.G.	53.70 ± 0.27
1.2	-	N.G.	58.60 ± 0.27
1.4	-	N.G.	68.10 ± 0.27
1.6	-	N.G.	67.90 ± 0.27

Table 15
Effect of heavy metals on *Vigna aconitifolia* growth rate

Concentration (mM)	Length of plant (cm)		
	Control	Chromium	Manganese
Water	61.20	-	-
0.2	-	47.5 ± 0.01	61.1 ± 0.01
0.4	-	N.G.	54.3 ± 0.01
0.6	-	N.G.	51.3 ± 0.01
0.8	-	N.G.	47.5 ± 0.01
1.0	-	N.G.	44.8 ± 0.01
1.2	-	N.G.	41.9 ± 0.01
1.4	-	N.G.	39.5 ± 0.01
1.6	-	N.G.	37.1 ± 0.01

N.G. = no germination

DISCUSSION

Dry matter yield decrease has generally been accepted as the standard measure for comparisons of toxicity. However, occasionally other measures such as fresh weight, commencement of symptoms¹⁷ and metabolic responses have been used²⁵.

Effect of chromium on growth rate and biomass

Chromium plays an important role in growth and development of plants^{54, 3}. It is toxic to plants when present in higher concentration and affects the growth^{68, 4, 12, 23}. Chromium toxicity results in to the decrease in root and shoot length^{32, 22, 4, 14, 19, 57, 8, 35, 59}, biomass^{65, 64}, plant weight^{57, 61} seedling height, killing of seeds⁶, root weight^{8, 35}, fresh weight and dry weight⁵⁹. In the present work only 30 - 40 % seed germination was observed and biomass of all investigated plants was reduced at lower concentration of chromium. Adverse effects of chromium on growth rate have been reported in different plants^{26, 59, 36}. In the investigated plants also chromium has adverse effect on the growth.

Effect of manganese on growth rate and biomass

Manganese is an essential micronutrient in most organisms^{46, 43}. In plants, it participates in the structure of photosynthetic proteins and enzymes. Its deficit is dangerous for chloroplasts because it affects the water-splitting system of photosystem II, which provides the necessary electrons for photosynthesis⁷. However, its excess seems also to be particularly damaging to the photosynthetic apparatus⁴⁹. Thus, manganese has two roles in plant metabolic processes: as an essential micronutrient and as a toxic element when it is in excess^{40, 15}. Plant species differ considerably in their normal or adequate manganese concentrations⁹ and in their susceptibility to manganese deficiency^{55, 43, 45}. The studies of several workers have indicated that the excess manganese inhibits the plant growth^{49, 20, 62, 24} and decreases root, stem and leaf dry weight / biomass. This reduction in fresh / dry weight / biomass of various plant organs was observed in *Lolium perenne*^{56, 47}, *Trifolium repens*⁵⁶, *Juncus*

*effuses*⁵¹ and *Populus cathayana*⁴². In this study decrease in fresh weight and dry weight of root, stem and leaf of *Glycine max*, *Vigna unguiculata* and *Vigna aconitifolia* was observed by increasing the concentration manganese in the treatment. Reduction in shoot growth and dry weight of various organs of sugar maple seedlings with increasing manganese level in the treatment was noticed⁴⁴. The sensitivity of other seedlings of several plant species to excess manganese was reported^{31, 37, 38, 39, 41, 44, 48, 58, 60}. Excess manganese may induce nutrient deficiencies in plants by interfering with the adsorption, translocation and / or utilization of nutrient elements such as calcium and magnesium^{27, 28, 29, 58}. Manganese toxicity may play an important role in poor growth of plants. In several plants reduction in growth rate⁴⁶, decrease in plant or shoot height^{51, 42} and root length⁵⁰ was found due to effect of excess manganese. The reduction in growth rate was noticed in all three investigated plants by higher manganese concentration in the current work. The reduction in nutrient elements by excess manganese may affect the growth rate⁴⁴. The reduced growth during manganese treatment probably is due to the effect of manganese on physiological processes, for example, inhibition of DNA replication² and protein synthesis²¹. The tolerance to an excess of manganese is highly dependent on the plant species and cultivars or genotypes within a species^{21, 29}.

CONCLUSION

The main points of conclusion which can be derived from the results obtained in present study are as follow: Reduction in fresh weight and dry weight was observed when the concentration of heavy metals increased. Decrease in fresh and dry weight was observed in following manner, chromium > manganese. The growth rate was reduced by increasing concentration of manganese in the treatment. The growth was decreased even at lower concentration of chromium. Higher concentration of manganese reduced the growth rate of all three investigated plants. Chromium has adverse effect on growth and biomass at lower concentration.

REFERENCES

1. Adeniji A. Bioremediation of arsenic, chromium, lead and mercury. United States Environmental Protection Agency for Toxic Substances and Disease Registry, ATSDR, (2004).
2. Baranowska H., Ejchart A. and Putrment A. Manganese mutagenesis in yeast, V. on mutation and conversion induction in nuclear DNA. *Mutat. Res.*, 42 : 343-345, (1977).
3. Bertrand D. and De Wolf A. Le Chrome. Oligoelements doivent etre utilises comme engrais complementaires? *Academic d' Agriculture de France. Comptes Rendus des Sciences* : 113-117, (1965).
4. Bishnoi N. R., Chugh L. K. and Sawhney S. K. Effect of chromium on photosynthesis, respiration and nitrogen fixation in pea (*Pisum sativum* L.) seedlings. *J. Plant Physiol.*, 42 : 25-30, (1993).
5. Blaylock M. J. and Huang J. W. Phytoextraction of metals. In: Raskin, I. and Ensley, B. D. (eds.), *Phytoremediation of toxic metals: Using plants to clean-up the environment*. John Wiley and Sons, New York : 53-70, (2000).
6. Bradshaw D., Mc Neilly R. S. and Gregory R. P. G. Industrialization evaluation and the development of heavy metal tolerance in plants. *Symp. Br. Eco. Sco.*, 5 : 327-343, (1965).
7. Buchanan B., Grusen W. and Jones R. Biochemistry and molecular biology of plants. *Ame. Soc. Plant Physiol.*, Maryland : 1367, (2000).
8. Chen N. C., Kanazawa S., Horiguchi T. and Chen N. C. Effect of chromium on some enzyme activities in the wheat rhizosphere. *Soil Microbio.*, 55 : 3-10, (2001).
9. Clarkson D. T. The uptake and translocation of manganese by plant roots. In: Graham, R. D., Hannam R. J. and Uren, N. C. (eds.), *Manganese in soils and plants*. Kluwer Academic Publishers, Dordrecht, Netherlands : 101-111, (1988).
10. Clarkson T. *Environ Health Perspect.* United States : 9-12, (1995).
11. Cossich E. S., Tavares C. R. G. and Ravagnani T. M. K. Biosorption of chromium (III) by *Sargassum* sp. biomass. *Elect. J. Biotech.*, 5 : 133-137, (2002).
12. Davis F. T. Jr., Puryear J. D., Newton R. J., Egilla J. N. and Grossi J. A. S. Mycorrhizal fungi enhance accumulation and tolerance of chromium in sunflower (*Helianthus annuus*). *J. Plant Physiol.*, 158 : 777-786, (2001).
13. Diels L., Van Der Lelie N. and Bastiaens L. New developments in treatment of heavy metal contaminated soils *Rev. Environ. Sci. Biotech.*, 1 : 75-82, (2002).
14. Dube B. K., Tewari K., Chatterjee J. and Chatterjee C. Excess chromium alters uptake and translocation of certain nutrients in *Citrullus*. *Chemosphere*, 53 : 1147-1153, (2003).
15. Ducic T. and Polle A. Transport and detoxification of manganese and copper in plants. *Braz. J. Plant Physiol.* 17 : 103-112, (2005).
16. El - Nady F. E. and Atta M. M. Toxicity and bioaccumulation of heavy metals to some marine biota from the Egyptain coastal waters. *J. Environ. Sci. Health*, 31 : 1529-1545, (1996).
17. Elamin O. M. and Wilcox G. E. Nitrogen form ratio influence on muskmelon growth, composition and manganese toxicity. *J. Ame. Soc. Hortic. Sci.*, 111 : 320-322, (1986).
18. Ensley B. D. Rational for use of phytoremediation. In: Raskin I. and Ensley B. D. (eds.), *Phytoremediation of toxic metals: Using plants to clean-up the environment*. John Wiley and Sons, New York : 3-12, (2000).
19. Faisal M. and Hasnain S. Chromate resistant *Bacillus cereus* augments sunflower growth by reducing toxicity chromium (VI). *J. Plant Bio.*, 48 : 187-194, (2005).
20. Feng J. P., Shi Q. H. and Wang X. F. Effects of exogenous silicon on photosynthetic capacity and antioxidant enzyme activities in chloroplast of cucumber seedlings under excess manganese. *Agric. Sci. China*, 8 : 40-50, (2009).

21. Foy C. D., Chaney R. L. and White M. C. The physiology of metal toxicity in plants. *Ann. Rev. Plant Physiol.*, 29 : 511-566, (1978).
22. Gardea-Torresdey J. L., Peralta-Videa J. R., Montes M., De La Rosa G. and Corral-Diaz B. Bioaccumulation of cadmium, chromium and copper by *Convolvulus arvensis* L.: Impact on plant growth and uptake of nutritional elements. *Biores. Tech.*, 92 : 229-235, (2004).
23. Gbaruko B. C. and Friday O. U. Bioaccumulation of heavy metals in some fauna and flora. *Intern. J. Environ. Sci. Tech.*, 4 : 197-202, (2007).
24. Gherardi M. and Rengel Z. Genotypes of lucerne (*Medicago sativa* L.) show differential tolerance to manganese deficiency and toxicity when grown in bauxite residue sand. *Plant Soil*, 249 : 287-296, (2003).
25. Gherardi M. J., Dell B. and Huang L. Functional copper requirement for catechol oxidase activity in plantation *Eucalyptus* species. *Plant and Soil*, 210 : 65-81, (1999).
26. Hanus J. and Tomas J. An investigation of chromium content and its uptake from soil in white mustard. *Acta Fytotech.*, 48 : 39-47, (1993).
27. Hecht-Buchholz C., Jorns C. A. and Keil P. Effect of excessive aluminum and manganese on Norway spruce seedlings as related to magnesium nutrition. *J. Plant Nutr.*, 10 : 1103-1110, (1987).
28. Heenan D. P. and Campbell L. C. Influence of potassium and manganese on growth and uptake of magnesium by soya beans (*Glycine max* L. Merr. cv Bragg). *Plant Soil*, 61 : 447-456, (1981).
29. Horst W. J. and Marschner H. Effect of silicon on manganese tolerance of bean plants (*Phaseolus vulgaris* L.). *Plant and Soil*, 50 : 287-303, (1978).
30. Horst W. J. The physiology of manganese toxicity. In: Graham R. D., Hannam R. J. and Uren N. J. (eds.), *Manganese in soil and Plants*. Kluwer Academic Publishers, Dordrecht, Netherlands : 175-188, (1988).
31. Hoyle M. C. Manganese toxicity in yellow birch (*Betula alleghaniensis* Britton) seedlings. *Plant Soil*, 36 : 229-232, (1972).
32. Huffman E. W. D. Jr. and Allaway W. H. Growth of plants in solution culture containing low levels of chromium. *Plant Physiol.*, 52 : 72-75, (1973).
33. Hunt R. *Plant growth analysis*. 1st (ed.) Edward Arnold, London, (1978).
34. Igwe J. C. and Abia A. A. A bioseparation process for removing heavy metals from waste water using biosorbents, *Review. Afr. J. Biotech.*, 5 : 1167-1179, (2006).
35. Iqbal M. Z., Saeeda S. and Shafiq M. Effects of chromium on an important arid tree (*Caesalpinia pulcherrima*) of Karachi city, Pakistan. *Ekol. Bratislava.*, 20 : 414-422, (2001).
36. Joseph G. W., Merrilee R. A. and Raymond E. Comparative toxicities of six heavy metals using root elongation and shoot growth in three plant species. The symposium on environmental toxicology and risk assessment, Atlanta, G. A., USA : 26-29, (1995).
37. Kavvadias V. A. and Miller H. G. Manganese and calcium nutrition of *Pinus sylvestris* and *Pinus nigra* from two different origins. I. Manganese. *Forestry*, 72 : 35-45, (1999).
38. Keil V. P., Hecht-Buchholz C. and Ortman U. Zum Einfluss von erhöhten Manganangebot auf Fichtensammlinge. *Allgem. Forstzeitschrift* 34/35 : 855-858, (1986).
39. Kitao M., Lei T. T. and Koike T. Effects of manganese in solution culture on the growth of five deciduous broad-leaved tree species with different successional characters from northern Japan. *Photosyn.* 36 : 31-40, (1999).
40. Kochian L., Hoekenga O. and Pifleros M. How do crop plants tolerate acid soils? Mechanisms of aluminum tolerance and phosphorus efficiency. *Ann. Rev. Plant Bio.*, 55 : 459-493, (1992).
41. Langheinrich U., Tischner R. and Godbold D. L. Influence of a high manganese supply on Norway spruce *Picea abies* (L.) Karst. seedlings in relation to nitrogen source. *Tree Physiol.*, 10 : 259-271, (2004).

42. Lei Y., Korpelainen H. and Li C. Physiological and biochemical responses to high manganese concentrations in two contrasting *Populus cathayana* populations. *Chemosphere*, 68 : 686-694, (2007).
43. Marschner H. Mineral nutrition of higher plants. Academic Press, San Diego : 325-329, (1995).
44. McQuattie C. J. and Schier G. A. Response of sugar maple seedlings to manganese. *Can. J. For. Res.*, 30 : 456-467, (2000).
45. Mengel K. and Kirkby E. A. Principles of plant nutrition. 5th edn. Kluwer Academic Publishers, Dordrecht, Netherlands, (2001).
46. Millaleo R., Reyes-Dláz M., Ivanov A. G., Mora M. L. and Alberdi M. Manganese as essential and toxic element for plants: Transport, accumulation and resistance mechanisms. *J. Soil Sci. Plant Nutr.*, 10 : 476-494, (2010).
47. Mora M. L., Rosas A., Ribera A. and Rengel Z. Differential tolerance to manganese toxicity in perennial ryegrass genotypes: Involvement of antioxidative enzymes and root exudation of carboxylates. *Plant Soil*, 320 : 79-89, (2009).
48. Morrison I. K. and Armson K. A. Influence of manganese on growth of jack pine and black spruce seedlings. *For. Chron.*, 44 : 32-35, (1968).
49. Mukhopadhyay M. and Sharma A. Manganese in cell metabolism of higher plants. *Bot. Rev.*, 57 : 117-149, (1991).
50. Mumthas S., Chidambaram A., Sundaramoorthy P. and Sankar Ganesh K. Effect of arsenic and manganese on root growth and cell division in root tip cells of green gram (*Vigna radiata* L.). *Emir. J. Food Agric.*, 22 : 285-297, (2010).
51. Najeeb U., Xu L., Shafaqat A., Jilani G., Gong H. J., Shen W. Q., Zhou W. J. Citric acid enhances the phytoextraction of manganese and plant growth by alleviating the ultrastructural damages in *Juncus effusus* L. *J. Hazard. Mater.*, 170 : 1156-1163, (2009).
52. Nascimento C. W. A., Amarasiriwardena D. and Xing B. Comparison of natural organic acids and synthetic chelates at enhancing phytoextraction of metals from a multi-metal contaminated soil. *Environ. Pollut.*, 140 : 114-123, (2006).
53. Nriagu J. O. Global inventory of natural and anthropogenic emissions of trace metals to the atmosphere. *Nature*, 279 : 409- 411, (1979).
54. Pratt P. F. Chromium in diagnostic criteria for plants and soils. In : Chapman, H. D. (ed.), University of California, California : 136-141, (1966).
55. Reuter D. J., Alston A. M. and McFarlane J. D. Occurrence and correction of manganese deficiency in plants. In : Graham, R. D., Hannam, R. J. and Uren, N. C. (eds.), Manganese in soils and plants. Kluwer Academic Publishers, Dordrecht, Netherlands : 205-224, (1988).
56. Rosas A., Rengel Z. and Mora M. Manganese supply and pH influence on growth, carboxylate exudation and peroxidase activity of ryegrass and white clover. *J. Plant Nutr.*, 30 : 253-270, (2007).
57. Rout G. R., Samantaray S. and Das P. Different chromium tolerance among eight mung bean cultivars grown in nutrient culture. *J. Plant Nutr.*, 20 : 341-347, (1997).
58. Safford L. O. Effect of manganese level in nutrient solution on growth and magnesium content of *Pinus radiata* seedlings. *Plant Soil*, 42 : 293-297, (1975).
59. Sankar G. K., Sundaramoorthy P. and Chidambaram A. L. A. Chromium toxicity effect on blackgram, soybean and paddy. *Pollut. Res.*, 25 : 257-261, (2006).
60. Schweitzer C. J., Sharpe W. E. and Edwards P. J. The effect of soil manganese on Japanese larch (*Larix leptolepis* Sieb and Zucc.) seedlings in the greenhouse: In: Stringer, J. W. and Loftis, D. L. (eds.), Proc. 12th central Hardwoods conf., Gen. Tech. Rep. SRS-24, USDA Forest Service, Southern Research Station, USA, 240-244, (1999).
61. Sharma D. C. and Sharma C. P. Chromium uptake and its effects on growth and biological yield of wheat. *Cereal Res. Commun.*, 21 : 317-321, (1993).
62. Shi Q. and Zhu Z. Effects of exogenous salicylic acid on manganese toxicity, element contents and antioxidative

- system in cucumber. Environ. Exp. Bot., 63 : 317-326, (2008).
63. Shmaefsky B. R. and Tucker G. Phytoremediation: Bioremediation: Panacea or fad? In: Zynda, T. (Writer) Factsheet. TAB Program, Michigan State University, Michigan, (2001).
64. Soni K. V. Effect of increasing concentration of chromium on emergence and growth of *Pithocolobium dulce* (Mimosae). M.Sc. Dissertation, Saurashtra University, Rajkot, (2004).
65. Tripathi A. K. and Tripathi S. Change in some physiological and biochemical characters in *Albizia lebbek* as bioindicators of heavy metal toxicity. J. Environ. Bio., 20 : 93-98, (1999).
66. USEPA (United States Environmental Protection Agency), Wastewater technology fact sheet. Chemical precipitation. USEPA 832-F-00-018, Washington, DC, (2000).
67. Vieira R. H. S. F. and Volesky B. Biosorption: A solution to pollution? Intern. Microbiol., 3 : 17-24, (2000).
68. Volcker J. A. Pot culture experiments. J. Royal Agric. Soc., 82 : 286-297, (1921).