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Remediation of Mercury Induced Stress on *Vigna mungo* (L.)Hepper Using *Martynia annua* L. Leaf Powder as Biosorbent

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Abstract

Heavy metals are a threat to human health and ecosystem. These days, great deal of attention is being given to green technologies for remediation of metal contaminated soil. Biosorption is one among such emerging technologies, which utilizes naturally occurring waste materials to sequester heavy metals from contaminated soil. In this present study the impact of mercury chloride was analysed. Seedlings of *Vigna mungo* (L.) Hepper were treated with various concentration of mercury chloride such as 5 mM, 10 mM, 15 mM, 20 mM and 25 mM. After 10 days of treatment various biochemical and enzyme characteristics were analysed. Apart from the biochemical such as glucose, protein, amino acid, the activity of nitrate reductase was gradually decreased with increasing concentration of mercury chloride. But the content of proline, leaf nitrate, catalase and peroxidise activity was in reverse. When optimal concentration 15 mM of mercury chloride was treated with various amounts of the leaf powder of a weed plant namely *Martynia annua* L. *viz*., 2 gm, 4 gm and 6 gm, and the filtrate was applied on the same plant. The reduced biochemical and enzyme characteristics due to metal toxicity were found improved considerably. From this study, it was inferred that the biosorbent.

Keywords: Biosorption; *Vigna mungo*; Mercury; Proline; Leaf nitrate; Catalase; Peroxidise

Introduction

Heavy metal pollution of soils is of serious concern as they are persistent, non-biodegradable and become toxic to living organisms [1]. They have a tendency to accumulate in soft tissues of living organisms. The deposition may then show biochemical or physiological changes in them [2]. Extreme values may cause growth inhibition and loss in net production, prominently seen in plants [3]. Growth reduction as a result of changes in physiological and biochemical processes in plants growing on heavy metal polluted soils has been recorded [4]. Heavy metal salts are water-soluble and get dissolved in wastewater, which means they cannot be separated by physical separation methods [5]. Additionally, physico-chemical methods are ineffective or expensive when the concentration of heavy metals is very low. Alternately, biological methods like biosorption and/or bioaccumulation for removal of heavy metals may be an attractive alternative to physico-chemical methods [6]. Use of plants and plant by-product for remediation purposesis thus a possible solution for heavy metal pollution since it includes sustainable remediation technologies to rectify and re-establish the natural condition of soil.

Materials and Methods

The seeds were procured from Tamil Nadu Agricultural University, Coimbatore. The plant *Martynia annua* L dried leaf powder used as a biadsorbent. The various concentration of mercury chloride (5 mM, 10 mM, 15 mM, 20 mM and 25 mM) were prepared. Both control and experimental plants were allowed to grow in soil mixture (red: black: garden soil) in the ratio of 1:1:1. After 10 days, the seedlings of *Vigna mungo* (L.) Hepper were treated with heavy metal solution aforesaid. Various biochemical and enzymatic characteristics were analysed on the treated plants. The optimal concentration mercury (15 mm) was mixed with various amounts of *Martynia annua* L dried leaf powder (2 g/L, 4 g/L and 6 g/L w/v) and kept in shaker for 24 hours. The filtrate was used to treat plants. After 10 days of treatment, the same biochemical and enzymatic characteristics were analyzed as follows: protein [7], glucose [8], amino acid [8], proline [9], *in vivo* nitrate reductase [10], peroxidise and catalase activity in Tables 1-6 [11].

Results and Discussion

The results obtained that morphometric characters such as root length, shoot length, fresh weight, dry weight and leaf area were decreased with increasing the concentration of mecury. The photosynthetic pigments of chlorophyll and carotenoids were decreased but the level of anthocyanin content was increased. The total soluble sugar content was decreased with the increase in the concentration of mercury. The mercury has caused a considerable increase in the free amino acid content than the control. The Leaf nitrate, proline, catalase activity and peroxidase activity content was increased with the increasing concentration of mercury chloride.

There was a decrease in protein content and total soluble sugar with increasing concentration of mercury chloride. This result coincides with [12]. Decline in protein content under metal stress can be related to the inhibition of protein synthesis or increase in protein degradation Zn [13]. The free proline has also been shown to protect plants against free radical induced damage by quenching of singlet oxygen [14]. Similar increase in leaf nitrate content, reduction *in vivo* NR activities with increased in concentration of cadmium chloride on Vigna radiata has been reported earlier [15]. Similar observation was noticed by [16] under the barium treatment in *Amaranthus caudatus* L. Catalase is another-antioxidant scavenging enzyme. It is also analysed in the present study and was found to increase with the increasing concentration of mercury. Catalase is a special type of peroxidase enzymes, which catalyses the degradation of H_2O_2, which is a natural metabolite and also toxic to plants [17]. The removal of heavy metals from metal contaminated soil is carried out using biosorbents

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Growth Parameters	Control	2 mm	4 mm	6 mm	8 mm	10 mm
Root length (cm)	9.66 ± 0.066 (100)	9.13 ± 0.088 (91)	8.7 ± 0.11 (85)	8.2 ± 0.057 (73)	7.26 ± 0.073 (66)	6.46 ± 0.056 (58)
Shoot length (cm)	23.91 ± 0.145 (100)	19.8 ± 0.152 (92)	19.16 ± 0.043 (89)	18.13 ± 0.088 (74)	17.23 ± 0.145 (62)	15.16 ± 0.088 (51)
Leaf area (cm2)	7.4 ± 0.152 (100)	6.32 ± 0.115 (92)	5.3 ± 0.057 (78)	4.6 ± 0.152 (57)	3.16 ± 0.033 (41)	2.53 ± 0.176 (38)
Fresh weight(gm)	0.98 ± 0.041 (100)	0.91 ± 0.005 (89)	0.78 ± 0.017 (71)	0.69 ± 0.005 (54)	0.55 ± 0.008 (36)	0.45 ± 0.009 (21)
Dry weight (gm)	0.73 ± 0.053 (100)	0.63 ± 0.005 (88)	0.42 ± 0.057 (65)	0.26 ± 0.034 (49)	0.14 ± 0.034 (31)	0.11 ± 0.066 (28)
+: Values are an average of ten observations. Values in parentheses are percentage over control. Mean ± SE n=10						

Table 1: Impact of various concentrations of mercuric chloride on the morphometric characteristics of *Vigna mungo* (L.) Hepper.

Growth Parameters	Control	2 mm	4 mm	6 mm	8 mm	10 mm
Chlorophyll .a (mg/gLFW)	3.34 ± 0.094 (100)	3.02 ± 0.078(85)	2.78 ± 0.092 (71)	2.57 ± 0.019 (57)	1.84 ± 0.066 (37)	1.24 ± 0.026 (25)
Chlorophyll .b (mg/gLFW)	4.84 ± 0.036 (100)	4.62 ± 0.140 (81)	3.21 ± 0.208 (73)	2.94 ± 0.138 (53)	2.15 ± 0.105 (32)	2.02 ± 0.089 (21)
Total.Chlorophyll (mg/gLFW)	8.18 ± 0.039 (100)	7.64 ± 0.137 (85)	5.89 ± 0.029 (76)	5.51 ± 0.124 (51)	3.99 ± 0.022 (36)	3.26 ± 0.136 (23)
Carotenoids (mg/gLFW)	3.98 ± 0.062 (100)	3.13 ± 0.227 (88)	2.76 ± 0.224 (63)	2.11 ± 0.164 (58)	1.98 ± 0.028 (43)	0.98 ± 0.114 (31)
Anthocyanin (µg/gLFW)	3.02 ± 0.129 (100)	3.73 ± 0.066 (131)	4.05 ± 0.137 (153)	5.89 ± 0.131 (182)	6.47 ± 0.165 (218)	6.98 ± 0.036 (234)
+: Values are an average of five observations. Values in parentheses are percentage over control. Mean ± SE n=5						

Table 2: Impact of various concentrations of mercuric chloride on the pigment contents of *Vigna mungo* (L) Hepper.

Parameters	Control	2 mm	4 mm	6 mm	8 mm	10 mm
Total soluble sugar (mg/gLFW)	9.46 ± 0.129 (100)	8.56 ± 0.081 (88)	7.62 ± 0.153 (78)	7.12 ± 0.060 (73)	4.22 ± 0.164 (43)	3.32 ± 0.136 (34)
Total soluble protein (mg/gLFW)	5.83 ± 0.043 (100)	4.64 ± 0.013 (83)	3.70 ± 0.021 (68)	3.12 ± 0.057 (53)	2.32 ± 0.086 (39)	2.32 ± 0.086 (39)
Amino acid (µ mole/gLFW)	3.32 ± 0.120 (100)	4.82 ± 0.136 (121)	5.42 ± 0.46 (146)	6.94 ± 0.158 (163)	7.25 ± 0.172 (188)	8.23 ± 0.123 (204)
Proline (µ mole/g LFW)	3.03 ± 0.064 (100)	4.57 ± 0.118 (118)	4.42 ± 0.036 (146)	6.19 ± 0.037 (168)	6.92 ± 0.037 (188)	7.86 ± 0.173 (205)
Leaf nitrate (µg/gLFW)	4.29 ± 0.120 (100)	5.12 ± 0.138 (114)	5.71 ± 0.184 (133)	6.98 ± 0.114 (154)	7.08 ± 0.228 (178)	8.30 ± 0.177 (195)
+: Values are an average of five observations .Values in parentheses are percentage over control. Mean ± SE n=5						

Table 3: Impact of various concentrations of mercuric chloride on the biochemical characteristics of *Vigna mungo* (L.) Hepper

Parameters	Control	2 mm	4 mm	6 mm	8 mm	10 mm
Nitrate Reductase (µ mole nitrite formed/g LFW)/hr	7.68 ± 0.045 (100)	7.03 ± 0.254 (83)	6.54 ± 0.173 (74)	5.46 ± 0.066 (59)	4.23 ± 0.149 (47)	3.23 ± 0.103 (38)
Catalase activity (µ mole/g LFW)/1 min.	1.37 ± 0.022 (100)	1.91 ± 0.024 (140)	2.55 ± 0.021 (151)	2.06 ± 0.038 (188)	3.04 ± 0.058 (224)	3.73 ± 0.015 (273)
Peroxidase activity (µ mole/g LFW)/3min.	1.48 ± 0.017 (100)	2.21 ± 0.067 (149)	3.06 ± 0.053 (206)	3.69 ± 0.102 (248)	4.61 ± 0.025 (310)	5.29 ± 0.103 (356)
+: Values are an average of five observations. Values in parentheses are percentage over control. Mean ± SE n=5						

Table 4: Impact of various concentration of mercuric chloride on the enzyme activity characteristics of *Vigna mungo* (L) Hepper.

Growth Parameters	Control water	+Mercury 6 Mm	6 mM mercuric chloride +		
			2 gm/100 ml leaf powder	4 gm/100 ml leaf powder	6 gm/`100 m leaf powder
Root length (cm)	9.66 ± 0.066 (100)	8.20 ± 0.057 (73	7.21 ± 0.145 (78)	8.01 ± 0.057 (89)	8.86 ± 0.120 (96)
Shoot length (cm)	23.91 ± 0.145 (100)	18.13 ± 0.088 (74)	19.77 ± 0.213 (79)	20.36 ± 0.202 (84)	22.54 ± 0.167 (95)
Leaf area (cm2)	7.40 ± 0.152 (100)	4.6 ± 0.152 (57)	5.21 ± 0.031 (63)	6.63 ± 0.173 (81)	7.14 ± 0.218 (98)
Fresh weight (gm)	0.98 ± 0.041 (100)	0.69 ± 0.005 (54)	0.36 ± 0.009 (68)	0.47 ± 0.085 (73)	00.81 ± 0.017 (86)
Dry weight (gm)	0.73 ± 0.053 (100)	0.26 ± 0.034 (49)	0.20 ± 0.060 (58	0.35 ± 0.006 (78)	0.48 ± 0.028 (83)
+: Values are an average of ten observations. Values in parentheses are percentage over control. Mean ± SE n=10					

Table 5: Effect of *Martynia annua* L. leaf powder treated mercuric chloride on the morphometric characteristics of *Vigna mungo* (L.) Hepper.

Parameters	Control water	+ Mercury 6 mM	6 mM mercuric chloride +		
			2 gm/100 ml leaf powder	4 gm/100 ml leaf powder	6 gm/`100 m leaf powder
Chlorophyll a mg/gLFW	3.34 ± 0.094 (100)	2.57 ± 0.099 (57)	2.16 ± 0.012 (78)	3.40 ± 0.810 (92)	4.81 ± 0.073 (110)
Chlorophyll b mg/gLFW	5.84 ±0.036 (100)	2..94 ± 0.138 (53)	3.44 ± 0.012 (76)	4.75. ± 0.017 (84)	5.23 ± 0.472 (108)
Total chlorophyll mg/gLFW	8.18 ± 0.039 (100)	5.51 ± 0.124 (51)	5..60 ± 0.219 (84)	8.15 ± 0.059 (93)	9.04 ± 0.194 (110)
Carotenoids mg/gLFW	3.98 ± 0.062 (100)	2.11 ± 0.164 (58)	2.24 ± 0.081 (67)	3.23 ± 0.093 (88)	4.95 ± 0.082 (107)
Anthocyanin mg/gLFW	3.02 ± 0.129 (100)	5.89 ± 0.131 (182)	5.43 ± 0.073 (153)	4.41 ± 0.131 (124)	3.61 ± 0.163 (105)
+: Values are an average of five observations. Values in parentheses are percentage over control. Mean ± SE n=5					

Table 6: Effect of *Martynia annua* L. leaf powder treated mercuric chloride on the pigment contents of *Vigna mungo* (L.) Hepper.

instead of conventional adsorbents. In the recent years, many low cost biosorbents such as algae, fungi, bacteria and agricultural byproducts have been investigated for their biosorption capacity towards heavy metal removal [18]. Plants applied with bioadsorbent treated metal solution showed increase in protein content, total soluble sugar and activity of nitrate reductase (Tables 7 and 8). In contrary amino acid content, activity of catalase and peroxidise was decreased in Tables 7 and 8.

Biochemical parameters	Control water	+ Mercury (6 mM)	6 mM mercuric chloride		
			2 gm/100 ml leaf powder	4 gm/100 ml leaf powder	6 gm/`100 m leaf powder
Total soluble sugar (mg/g LFW)	9.46 ± 0.129 (100)	7.12 ± 0.060 (73)	7.62 ± 0.057 (74)	8.54 ± 0.031 (96)	9.84. ± 0.026 (110)
Total soluble protein (mg/g LFW)	5.83 ± 0.043 (100)	3.12 ± 0.057 (53)	3.93. ± 0.070 (81)	5.24 ± 0.045 (93)	6.30 ± 0.053 (112)
Amino acid (µ mole/g LFW)	3.23 ± 0.120 (100)	6.94 ± 0.158 (163)	6.34. ± 0.05 (157)	4.75 ± 0.046 (133)	3.98 ± 0.58 (106)
Proline (µ mole/g LFW)	3.32 ± 0.064 (100)	6.19 ± 0.013 (188)	5.14 ± 0.024 (163)	4.63 ± 0.031 (133)	3.68 ± 0.187 (109)
Leaf nitrate (µg/gLFW)	4.29 ± 0.120 (100)	6.98 ± 0.114 (204)	7.34.± 0.092 (179)	6.34 ± 0.137 (135)	4.82 ± 0.076 (112)
+: Values are an average of five observations. Values in parentheses are percentage over control. Mean ± SE n=5					

Table 7: Effect of *Martynia annua* L. leaf powder treated mercuric chloride on the biochemical characteristics of *Vigna mungo* (L.) Hepper.

Enzymatic parameters	Control water	+ Mercury (6 mM)	6 mM mercuric chloride		
			2 gm/100 ml leaf powder	4 gm/100 ml leaf powder	6 gm/`100 m leaf powder
Nitrate reductase µmole/ gLFW|hr	7.68 ± 0.045 (100)	5.46 ± 0.066 (59)	3.63 ± 0.041 (74)	6.71 ± 0.063 (86)	7.93 ± 0.114 (104)
Catalase activity µmole/gLFW1min.	1.37 ± 0.115 (100)	2.06 ± 0.038 (188)	3.01 ± 0.093 (172)	2.63 ± 0.081 (148)	1.96 ± 0.023 (114)
Peroxidase activity µmole/gLFW|3 min	1.48 ± 0.017 (100)	3.69 ± 0.102 (248)	2.73 ± 0.086 (176)	2.06 ± 0.028 (136)	1.74 ± 0.036 (112)
+: Values are an average of five observations. Values in parentheses are percentage over control. Mean ± SE n=5					

Table 8: Effect of *Martynia annua* L. leaf powder treated mercuric chloride on the enzymatic characteristics of *Vigna mungo* (L.) Hepper.

Conclusion

From the present investigation, it is confirmed that the plant *Martynia annua* L dried leaf powder is nullifying the toxicity of heavy metal. Since it restores the suppressed biochemicals and enzyme activity due to metal toxicity.

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References

1. Tomas J, Arvay J, Toth T (2012) Heavy metals in productive parts of agricultural plants. J Microbiol Biotechnol Food Sci 1: 819-817.
2. Srinivas J, Purushotham AV, Murali Krishna KVSG (2013) The effects of Heavy metals on Seed Germination and Plant Growth on Coccinia, Mentha and Trigonella Plant Seeds in Timmapuram, E.G. District, Andhra Pradesh, India. Int Res J Environment Sci 2(6): 20-24.
3. Nath TN. (2013) The Status of Micronutrients (Mn, Fe, Cu, Zn) in Tea Plantations in Dibrugarh district of Assam, India. Int Res J Environment Sci 2(6): 25-30.
4. Chatterjee J, Chatterjee C (2000) Phytotoxicity of cobalt, chromium and copper in cauliflower. Environ Pollut 109(1): 69-74.
5. Hussein H, Farag S, Moawad H (2004) Isolation and characterization of Pseudomonas resistant to heavy metals contaminants. Arab J Biotechnol 7(1): 13–22.
6. Kapoor A,Viraraghvan T (1995) Fungal biosorption-An alternative treatment option for heavy metal bearing wastewater:A review. Bioresour Technol 53(3): 195–206.
7. Lowry OH, Rosenbury NJ, Farr AL, Randall RJ (1951) Protein measurement with folin phenol reagent. J Biol Chem 193(1): 262-275.
8. Jayaraman J (1981) Laboratory manual in Biochemistry. Wiley Eastern.
9. Bates LS, Waldren RP, Teare ID (1973) Rapid determination of the proline in water stress studies. Plant and Soil 39: 205-207.
10. Jaworski EG (1971) Nitrate reductase assay in intact plant tissues. Biochem Biophy & Res Commun 43(6): 1274-1279.
11. Kar M, Mishra D (1976) Catalase, peroxidase and polyphenol oxidase activities during rice leaf senescence. Plant Physiol 57(2): 315-319.
12. Periyanayagi G, Sevugaperumal R, Ramasubramanian V (2015) Assessment of the biosorptive properties of Padina commersonii (Seaweed).Int J Innov Res Sci Eng Technol 1(3): 45-49.
13. Gautam S, Kannaujiya P, Srivastava MN (2015) Growth and biochemical responses of spinach (Spinacea oleracea L.) grown in Zn contaminated soils. Int J Rec Biotech 3(1): 7-12.
14. Matysik J, Bhalu BA, Mohanty P (2002) Molecular mechanisms of quenching of reactive oxygen species by proline under stress in plants. Curr Sci 82(5): 525-532.
15. Jayakumar S, Ramasubramanian V (2009) Bioremoval of chromium using seaweeds as biosorbents. J Basic and App Biol 3(3&4): 121-128.
16. Marisamy K, Duraipandian M, Sevugaperumal R, Ramasubramanian V (2015) Estimation of Barium Toxicity Mitigating Efficacy of *Amaranthus caudatus* L. Univers j environ res technol. 5(6): 295-305.
17. Balasinha D (1982) Regulation of peroxidase in higher plants. Ann Rev Plant Physiol 25: 225-228.
18. Volesky B (1986) The Mechanism of Metal Cation and Anion Biosorption. Biosorbent materials, Biotehnol Bioeng Symp Ser 16: 121-126.