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Interactions between Chromium and Plant Growth Regulators on the Growth of *Spirodela polyrrhiza* (L.) Schleiden

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ABSTRACT

Chromium is one of the most toxic heavy metals which reduces the growth of plants at relatively low concentrations, whereas IAA, kinetin, GA_3 and 24-epibrassinolide (24-epiBr) are well known plant growth regulators (PGRs) which enhance the growth of plants. The present study was undertaken to determine the effect of binary combinations of Cr(VI) and PGRs on *Spirodela polyrrhiza* (L.) Schleiden, an aquatic macrophyte, grown on Hoagland's E-media. We observed that Cr(VI) at 1, 2 and 3 mM in the absence of exogenous PGRs significantly decreased the growth of plants, whereas IAA, kinetin, GA_3 (0.1, 10, 1000 nM) and 24-epiBr (0.01, 1, 100 nM) in the absence of Cr(VI) significantly increased the growth. Multiple regression models revealed that Cr(VI) inhibits the activity of exogenous PGRs leading to a further decrease in the growth of *S. polyrrhiza* cultured in binary combinations of Cr(VI) and PGRs.

Keywords: metal-PGR interaction, IAA, GA_3, 24-epibrassinolide
Abbreviations: 24-epiBr, 24-epibrassinolide; **mM**, millimolar; **nM**, nanomolar; **PGR**, plant growth regulator

INTRODUCTION

Chromium is one of the most widespread contaminants of human environments originating from various industries, e.g. metallurgical (ferro- and non-ferrous alloys), refractories (chrome and chrome-magnesite) and chemical (pigment, electroplating, tanning), etc. (Zhang *et al*. 2004; Shtiza *et al*. 2008). As a transition element, it occurs in a number of oxidation states, the most stable and common forms being Cr(III) and Cr(VI). Cr(III) in trace doses is essential for animal and human glucose and lipid metabolism (Cornelis *et al*. 1984). But Cr(VI) is a highly toxic carcinogen and may cause death to animals and human beings at higher concentrations (Zayed and Terry 2003; Panda and Choudhury 2005). The oral LD_{50} for Cr(VI) for human beings is 50 mg kg^{-1} body weight and for fresh water fish the LC_{50} is 250-400 mg L^{-1}. The WHO permissible limit for total Cr in drinking water is 100 µg L^{-1}. Cr(VI) can be actively transported across biological membranes of prokaryotes but Cr(III) is transported passively by cation exchange sites of cell walls (Skeffington *et al*. 1976; Sirko *et al*. 1990).

Aquatic macrophytes are a good remediation option because a few species have already been reported to accumulate Cr from waste waters (Zayed *et al*. 1998; Zurayk *et al*. 2001; Zhang *et al*. 2007). Duckweeds have been considered as promising prospective scavengers of heavy metals from polluted waters. *Lemna gibba* and *L. minor* L. are the most studied species of the family Lemnaceae with respect to toxicity and phytoremediation (Mkandawire *et al*. 2004; Mkandawire and Dudel 2005, 2007; Uysal and Taner 2007). Rahman *et al*. (2007) reported *Spirodela polyrrhiza* to be a good arsenic phytofiltrator. Duckweeds possess relatively high tolerance to Cr toxicity and are capable of active uptake and accumulation of this element against a concentration gradient (Staves and Knaus 1985; Tripathi and Chandra 1991). Chandra and Kulshreshta (2004) and Jaglarz *et al*. (2004) reported that *S. polyrrhiza* is a potential accumulator of Cr(VI).

Several PGRs like auxins, gibberellins, cytokinins and 24-epiBr are well known to increase the growth of plants. These are present in wastewaters as natural biomolecules of plants, biodegradation products of urine and fecal matter or as leachates of commercial applications of PGRs. Discerning how PGRs interact, and how their quantities are affected by environmental factors, such as nutrient supply, will be important for the improvement of traits of plants of economic importance (Buchanan *et al*. 2000). Therefore, it could be interesting to study the effect of binary concentrations of Cr(VI) and PGRs on the growth of plants. It has been reported that brassinosteroids ameliorate aluminium toxicity in mungbean (*Phaseolus aureus*) seedlings and promote seedling growth and chlorophyll content under metal stress (Abdullahi *et al*. 2003). Alleviation of the toxic effect of Cd, and an increase in seedling growth and seed germination under the effect of brassinosteroids was also reported by Anuradha and Rao (2007). Moya *et al*. (1995) reported that the addition of gibberellic acid with Cd or Ni partially reversed the effects of heavy metals, stimulating growth as well as mobilization of carbohydrate reserves in seeds from which seedlings had developed. The present study was undertaken to determine the effects of PGRs on the growth of *S. polyrrhiza* cultured in solutions containing Cr(VI) so as to study metal-PGR interactions and to determine the cause of Cr(VI) toxicity.

MATERIALS AND METHODS

S. polyrrhiza (giant duckweed) is a free-floating macrophyte with a world-wide distribution and grows luxuriantly in tropical and subtropical regions. The plant body lacks true leaves and stem, instead consists of frond or thallus which is 4-10 mm long. The underside of the frond bears 4-16 adventitious roots. The duckweeds are a high protein source for ducks, geese, fish and cattle. These make a good experimental material for toxicity bioassays, and can be cultured on semi-solid inorganic media. Plants used for the present study were collected from wild habitats of Amritsar (31°63′N, 74°87′E). The tiny plants were gently washed under running tap water and rinsed with sterile distilled water. Each plant was cleaned of foreign particles, if any, with brush and placed on a filter paper for 5 min. to soak water adhering to it. Three sets of 15

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Table 1 Composition of Hoagland's E-medium per litre of the final volume.

Salt	Stock solution	Volume (ml L^{-1} medium)
$MgSO_4.7H_2O$	24.6 g/100 ml	1.0
$CaNO_3.4\ H_2O$	23.6 g/100 ml	2.3
KH_2PO_4	13.6 g/100 ml	0.5
KNO_3	10.1 g/100 ml	2.5
Micronutrients	H_3BO_3 (2.86 g L^{-1})	0.5
	$MnCl_2.4\ H_2O$ (1.82 g L^{-1})	
	$ZnSO_4.7H_2O$ (0.22 g L^{-1})	
	$Na_2MoO_4.2H_2O$ (0.09 g L^{-1})	
	$CuSO_4.5\ H_2O$ (0.09 g L^{-1})	
Fe.EDTA	$FeCl_3.6H_2O$ (0.121 g/250 ml)	20.0
(Freshly prepared)	EDTA (0.375 g/250 ml)	
Agar		1.5 %

filter-soaked plants for each treatment were weighed and cultured in Hoagland's E-medium autoclaved at a pressure of 15 psi for 20 min. The composition of the medium as defined by Cowgill and Milazzo (1989), and available online (ASTM-STP 1989), is given in **Table 1**. Salts used in the present study were procured from Qualigens, Loba chemie, Sd-fine-chem and Central drug house, India. The pH of the medium was set at 6.00, and the medium was poured in sterilized Petri plates and kept at 25 ± 1°C under fluorescent light (1700 Lux) with a light:dark period of 14:10 h for 14 days. Since the plants were cultured on inorganic medium, and the experiment was a toxicity bioassay, the plants were not sterilized but cultured under aseptic conditions.

Salt used for chromium treatment was K_2CrO_4 (Merck). The plants were cultured in Hoagland's E-medium containing 0, 1, 2 and 3 mM concentrations (1 mM = 51.99 mg L^{-1} of Cr) of Cr in binary combinations with different concentrations of PGRs. For IAA, kinetin and GA_3, 0 nM, 0.1 nM (10^{-10} M), 10 nM (10^{-8} M) and 1000 nM (10^{-6} M) concentrations were used, whereas for 24-epiBr 0 nM, 0.01 nM (10^{-11} M), 1 nM (10^{-9} M) and 100 nM (10^{-7} M) concentrations were used. PGRs were procured from Sigma-Aldrich (24-epiBr) and other sources mentioned above. Each treatment was given in triplicate. The growth of plants was calculated as change in fresh weight per g initial weight after 14 days of culturing. Statistical analysis was carried out for descriptive statistics, 2-way ANOVA, Tukey's multiple comparison test (Daniel 1991), and linear, curvilinear and multiple linear regressions (Sokal and Rohlf 1981). Self-coded software was developed in MS-Excel.

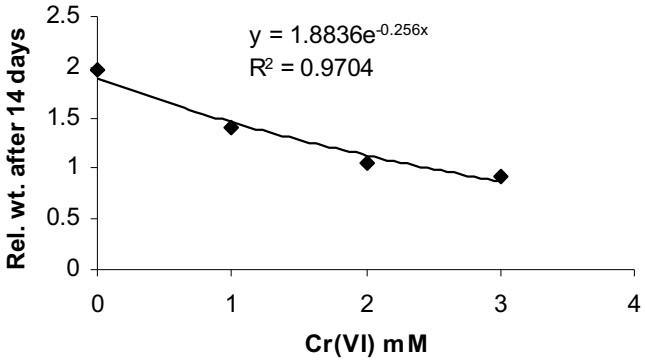

Fig. 1 Effect of Cr(VI) on the relative growth of *S. polyrrhiza* cultured on Hoagland's E-medium after 14 days.

RESULTS AND DISCUSSION

It was observed from the present study that Cr(VI), when applied at concentrations 1 mM to 3 mM, is toxic to *Spirodela polyrrhiza* and inhibits its growth varying from 34.1% to 64.4% at 3 mM concentration with reference to control (**Fig. 1**). The growth of plants reduces exponentially with Cr(VI) concentration in the medium. 50% inhibitory concentration as calculated from the exponential decay curve of Cr(VI) for *S. polyrrhiza* is 2.707 mM.

Application of PGRs, IAA, kinetin and GA_3 increased the growth of *S. polyrrhiza* significantly with reference to control. 24-epiBr was most effective in enhancing the growth by 43.6% at 10^{-7} M concentration. The 2-way analysis of variance revealed that Cr(VI) significantly affects the growth of plants in all the treatments at $p \leq 0.001$. The effect of IAA on the growth of *S. polyrrhiza* was weekly significant ($p \leq 0.1$). The interactions between Cr(VI) and PGRs were weekly significant for kinetin and 24-epiBr (**Table 2**). The linear correlation analysis revealed that there was a significant positive correlation between the increase in growth of the plants and application of IAA, kinetin and 24-epiBr (**Table 3**).

Effect of binary combinations of Cr and PGRs on the growth of *S. polyrrhiza*

Figs. 2-5 give the effect of growth regulators at different concentrations of Cr(VI). When *S. polyrrhiza* was cultured on media containing both Cr(VI) and PGRs, the growth of the plants decreased with reference to the control except for

Table 2 Two-way ANOVA and Tukey's multiple comparison test for fresh weight of *S. polyrrhiza* after 14 days of culturing in media containing Cr(VI) and PGRs.

Treatment	Source of variation	SS	df	MSS	F-ratio	HSD
IAA	Cr(VI)	6.052	3	2.017	86.470***	0.459
	IAA	0.195	3	0.065	2.791*	
	Cr(VI) X IAA	0.367	9	0.040	1.749	
	Error	0.746	32	0.023		
	Total	7.361	47			
Kinetin	Cr(VI)	2.075	3	0.691	36.096***	0.418
	Kinetin	0.008	3	0.002	0.146	
	Cr(VI) X Kinetin	0.369	9	0.041	2.139*	
	Error	0.613	32	0.019		
	Total	3.066	47			
GA_3	Cr(VI)	13.099	3	4.366	153.956***	0.507
	GA_3	0.030	3	0.010	0.358	
	Cr(VI) X GA_3	0.119	9	0.013	0.467	
	Error	0.907	32	0.028		
	Total	14.157	47			
24-EpiBr	Cr(VI)	41.595	3	13.865	110.200***	1.071
	24-EpiBr	0.792	3	0.264	2.099	
	Cr(VI) X 24-EpiBr	2.075	9	0.230	1.833*	
	Error	4.026	32	0.125		
	Total	48.490	47			

Significant at ***p≤ 0.001, * p≤ 0.1

Table 3 Linear regression analysis of the effect of PGRs (nM) in the absence of Cr on the relative growth (Y) of *S. polyrrhiza* after 14 days of culturing (g g^{-1} initial wt.)

PGR (X)	Regression equation	r
IAA	Y = 0.1181 X + 1.461	0.9762*
Kinetin	Y = 0.1223 X + 1.055	0.9843**
GA_3	Y = 0.0547 X + 2.245	0.7458 (ns)
24-EpiBr	Y = 0.4070 X + 2.325	0.9673*

Significant at ** p≤ 0.02, * p≤ 0.05, ns = not significant at p≤ 0.05.

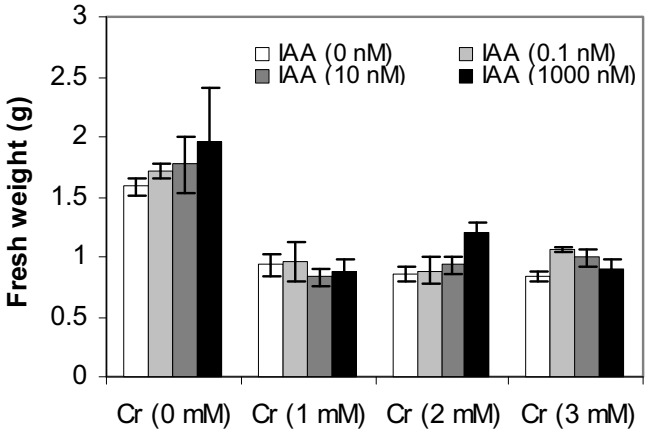

Fig. 2 Relative growth of *S. polyrrhiza* in binary combinations of Cr(VI) and IAA.

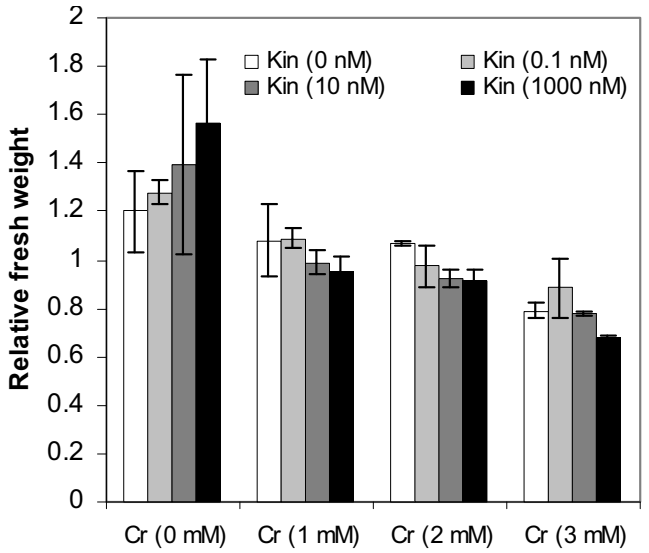

Fig. 3 Relative growth of *S. polyrrhiza* in binary combinations of Cr(VI) and kinetin.

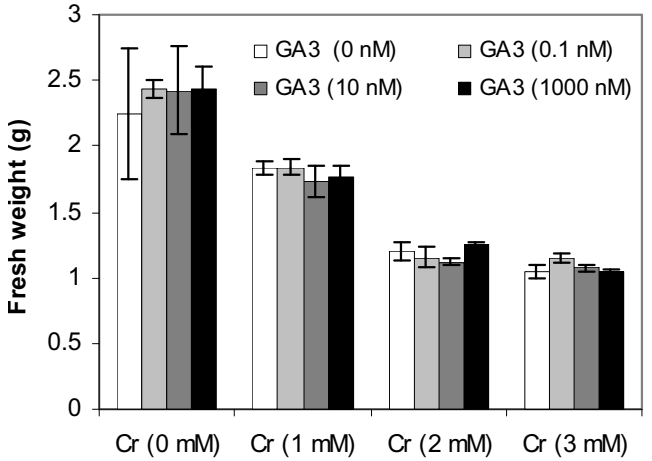

Fig. 4 Relative growth of *S. polyrrhiza* in binary combinations of Cr(VI) and GA_3.

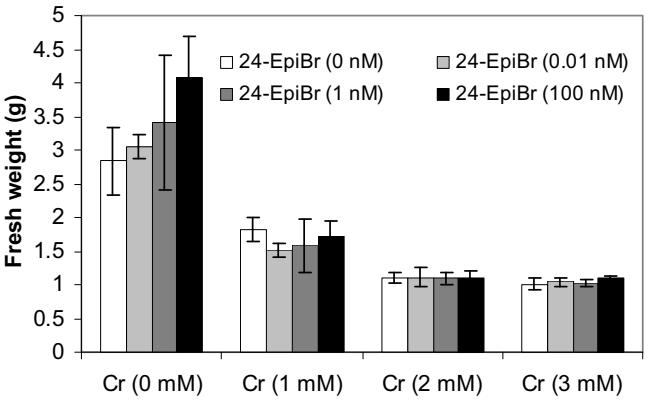

Fig. 5 Relative growth of *S. polyrrhiza* in binary combinations of Cr(VI) and 24-epiBr.

Cr 2 mM : IAA 1000 nM combination.

In order to find out the effect of interaction between the PGRs and Cr(VI) on the growth of *S. polyrrhiza* plants, data was analysed using multiple regression equation with interaction. General model used was

$$Y = a + b_1X_1 + b_2X_2 + cX_1X_2$$

where Y is the growth of *Spirodela* plants, X_1 and X_2 represent Cr(VI) and PGR concentrations and b_1, b_2 and c are partial regression coefficients. As given in **Table 4**, the correlation coefficients of multiple regression models for kinetin, GA_3 and 24-epiBr were found to be statistically significant at p≤0.001, and for IAA at p≤0.01. All the four PGRs had positive effect on the growth of *S. polyrrhiza* in the multiple regression models, whereas Cr(VI) had negative effect on its growth. Interaction between Cr(VI) and PGRs, as represented by partial interaction regression coefficient 'c', was negative in all the cases. In order to determine the relative effects of Cr(VI) and PGRs on the growth of *S. polyrrhiza*, β-regression coefficients were computed. It was observed that in the binary combinations of Cr(VI) with IAA, the factor influencing the growth most is Cr(VI) (negative), followed by IAA (positive), followed by interaction factor (negative). The same trends were observed for GA_3 and 24-epiBr. In case of kinetin, Cr(VI) affected the growth most, followed by interaction factor, followed by PGR.

It is, therefore observed from the present study that IAA, kinetin, gibberellic acid and 24-epiBr increase the growth of *S. polyrrhiza*. PGRs bring about growth and morphogenetic responses and each PGR is pleiotropic in its effects. Besides, several PGRs may affect the same response, e.g., cell elongation is affected by auxins, gibberellins and brassinosteroids, cell division is affected by auxins, cytokinins and gibberellins, implying thereby a redundancy in the control of the same response (Srivastava 2002). Koch and Durako (1991) showed that cytokinin supplements increased rhizome and shoot growth in *Ruppia maritima* L. explants grown in axenic cultures. In terrestrial angiosperms, auxins cause apical dominance in stems and promote root initiation and elongation; gibberellins reverse genetic dwarfs, and promote the growth of intact plants and dormant buds (Salisbury and Ross 1992). In terrestrial plants, externally applied gibberellins have been shown to promote stem elongation, leaf enlargement and elongation, phenotypic reversion of monocotyledonous genetic dwarfs and inhibition of root formation (Letham *et al*. 1978). Brassinosteroids, when applied exogenously at nM to µM concentrations, evoke cell elongation or proliferation and affect a number of physiological processes (Mandava 1988; Clouse 1996). Sharma and Bhardwaj (2007) reported that 24-epiBr increases seed germination, shoot and root length and fresh weight of seedlings of *Brassica juncea* L.

Another observation in the present study is that Cr(VI) affects the growth of plants. Chromium can be absorbed as Cr^{3+} or CrO_4^{2-} by the roots of higher plants although availa-

Table 4 Multiple regression equation models for relative growth of *S. polyrrhiza* in binary combinations of Cr(VI) and PGRs

PGR	Multiple Regression Equation with Interaction	r	β regression coefficients		
			Cr (X_1)	PGR (X_2)	Cr X PGR Interaction
IAA	$Y=1.4404 – 0.219\ X_1 + 0.0002\ X_2 – (7x10^{-5})\ X_1X_2$	0.731*	- 0.6581	0.2623	- 0.1618
Kinetin	$Y=1.2596 – 0.148\ X_1 + 0.0002X_2 – (1x10^{-4})\ X_1X_2$	0.921**	- 0.7293	0.3333	- 0.4647
GA_3	$Y=2.2748 – 0.447\ X_1 + 4x10^{-5}\ X_2 – (2x10^{-5})\ X_1X_2$	0.960**	- 0.9503	0.0349	- 0.0291
24-EpiBr	$Y=2.7237 – 0.674\ X_1 + 0.0072X_2 – 0.003\ X_1X_2$	0.888**	- 0.7830	0.3215	- 0.2554

Significant at ** $p \leq 0.001$, * $p \leq 0.01$.

ble data are still contradictory (Babula *et al*. 2008). One of the possible explanations of Cr(VI) toxicity could be that it produces damaging $^{-}$OH radicals which inactivate the PGRs. It has been reported by Davies *et al*. (2002) that chromium is toxic to most of the higher plants at 100 μmol kg^{-1} dry weight. Chromium toxicity is caused by the reaction of chromium with reducing agents such as NAD(P)H, which in turn react with H_2O_2 to generate damaging $^{-}$OH radicals, as well as, chromium reacting with the carboxyl and sulfhydryl groups of enzymes, thereby inhibiting their activity (Cervantes *et al*. 2001). Also the production of H_2O_2, $^{-}$OH and $^{-}O_2$ under chromium stress has been demonstrated in many plants, leading to damage of DNA, proteins and pigments, besides indicating lipid peroxidation (Bagchi *et al*. 2000; Panda and Patra 2000; Panda 2003). The most significant part of the study is that Cr(VI) toxifies the PGRs as revealed by interaction factor. The mode of action of PGRs involves several cellular components. Auxins induce the cell to excrete protons by stimulating the membrane bound ATPase-proton pump, resulting in turgor induced cell expansion. Cytokinins promote the onset of mitosis by activating the phosphatase that removes an inhibitory phosphate group from the cyclin-dependant kinase/cyclin B complex, thus moving the cell from the G2 to the M phase of cell division. Gibberellins induce the release of enzymes and carbohydrates during germination of barley (Hopkins and Hüner 2004). Brassinosteroids increase the H^+-ATPase activities of plasma membrane and tonoplast with a consequent cell elongation and division (Haubrick and Assmann 2006). Since Cr(VI) is capable of damaging several components of the cell and generate free radicals, the effect of PGRs is nullified.

It can, therefore, be concluded from the present study that the growth of *S. polyrrhiza* is retarded in cultures containing binary combinations of Cr(VI) and PGRs due to the negative interactions between the two.

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REFERENCES

Abdullahi BA, Gu XG, Gan, QL, Yang YH (2003) Brassinolide amelioration of aluminium toxicity in mung bean seedling growth. *Journal of Plant Nutrition* **26**, 1725-1735

Anuradha S, Rao SSR (2007) The effect of brassinosteroids on radish (*Raphanus sativus* L.) seedlings growing under cadmium stress. *Plant Soil Environment* **11**, 465-472

ASTM-STP (1989) Hoagland's E-medium. Available online: http://mobot.org/jwcross/duckweed/media.htm

Bagchi C, Joshi SS, Bagchi M, Balmoori J, Benner EJ, Kuszynski CA (2000) Cadmium and chromium induced oxidative stress, DNA damage and apoptotic cell death in cultured human chronic myelogenous leukemic K562 cell, promyelocytic leukemic HL–60 cells and normal human peripheral blood mononuclear cells. *Journal of Biology and Molecular Toxicology* **14**, 3-41

Babula P, Adam V, Opatrilova R, Zehnalek J, Havel L, Kizek R (2008) Uncommon heavy metals, metalloids and their plant toxicity: A review. *Environmental Chemistry Letters* **6**, 189-213

Buchanan BB, Gruissem W, Jones RL (2000) *Biochemistry and Molecular Biology of Plants*, American Society of Plant Physiologists, Rockville, Maryland, 1367 pp

Cervantes C, Campos-Garcia J, Devars S, Gutierrez-Corona F, Loza-Tavera H, Torres-Guzman JC, Moreno-Sanchez R (2001) Interaction of chromium with microorganisms and plants. *Microbiology Reviews* **25**, 335-347

Chandra P, Kulshreshta K (2004) Chromium accumulation and toxicity in aquatic vascular plants. *Botanical Review* **70**, 313-323

Clouse SD (1996) Molecular genetic studies confirm the role of brassinosteroids in plant growth and development. *The Plant Journal* **10**, 1-8

Cornelis R, Wallaeys B (1984) Chromium revisited. In: Bratter P, Schramel P (Eds) *Trace Element Analytical Chemistry in Medicine and Biology* (Vol 3), Walter de Gruyter & Co., Berlin, pp 219-233

Cowgill UM, Milazzo DP (1989) The culturing and testing of two species of duckweed. In: Cowgill UM, Williams LR (Eds) *Aquatic Toxicology and Hazard Assessment* (Vol 12), American Society of Testing and Materials, Philadelphia, pp 379-391

Daniel WW (1991) *Biostatistics: A Foundation for Analysis in the Health Sciences*, John Wiley & Sons, New York, 740 pp

Davies FT, Puryear JD, Newton RJ, Egilla JN, Grossi JAS (2002) Mycorrhizal fungi increase chromium uptake by sunflower plants: Influence on tissue mineral concentration, growth and gas exchange. *Journal of Plant Nutrition* **25**, 389-407

Haubrick LL, Assmann SM (2006) Brassinosteroids and plant function: some clues, more puzzles. *Plant Cell and Environment* **29**, 446-457

Hopkins WG, Hüner NPA (2004) *Introduction to Plant Physiology* (3rd edn), John Wiley & Sons Inc., Hoboken, NJ, 560 pp

Jaglarz A, Appenroth K-J, Gabryś H, Kaszycki P, Koloczek H, Sędziwy S, Walczac T (2004) Hexavalent chromium transport and reduction by *Spirodela polyrrhiza* upon sulfate competition. Development of a kinetic model. *Federation of Europian Biochemical Societies Journal*, Abstract no. P4.9-52

Koch EW, Durako MJ (1991) *In vitro* studies of the submerged angiosperm *Ruppia maritima*: auxin and cytokinin effects on plant growth and development. *Marine Biology* **110**, 1-6

Letham DS, Goodwin PB, Higgins TJV (1978) *Phytohormones and Related Compounds-A Comprehensive Treatise. (Vol II) Phytohormones and the Development of Higher Plants*, Elsevier/North-Holland, Amsterdam, 648 pp

Mandava NB (1988) Plant growth-promoting brassinosteroids. *Annual Review of Plant Physiology and Plant Molecular Biology* **39**, 23-52

Mkandawire M, Lyubun YV, Kosterin PV, Dudel EG (2004) Toxicity of arsenic species to *Lemna gibba* L. and the influence of phosphate on arsenic bioavailability. *Environmental Toxicology* **19**, 26-35

Mkandawire M, Dudel EG (2005) Accumulation of arsenic in *Lemna gibba* L. (duckweed) in tailing waters of two abandoned uranium mining sites in Saxony, Germany. *Science of Total Environment* **336**, 81-89

Mkandawire M, Dudel EG (2007) Are *Lemna* spp. effective phytoremediation agents? *Bioremediation, Biodiversity and Bioavailability* **1**, 56-71

Moya JL, Ros R, Picazo I (1995) Heavy metal - hormone interaction in rice plants: Effects on growth, net photosynthesis and carbohydrate distribution. *Journal of Plant Growth Regulation* **14**, 61-67

Panda SK, Patra HK (2000) Does Cr(III) produce oxidative damage in excised wheat leaves? *Journal of Plant Biology* **27**, 105-110

Panda SK (2003) Heavy metal phytotoxicity induces oxidative stress in *Taxithelium* Sp. *Current Science* **84**, 631-633

Panda SK, Coudhury S (2005) Chromium stress in plants. *Brazilian Journal of Plant Physiology* **17**, 95-102

Rahman MA, Hasegawa H, Ueda K, Maki T, Okumura C, Rahman MM (2007) Arsenic accumulation in duckweed (*Spirodela polyrrhiza* L.): A good option for phytoremediation. *Chemosphere* **69**, 493-499

Salisbury FB, Ross CW (1992) *Plant Physiology*, Wadsworth, Belmont, CA, 682 pp

Sharma P, Bhardwaj R (2007) Effect of 24-epibrassinolide on seed germination, seedling growth and heavy metal uptake in *Brassica juncea* L. *General and Applied Plant Physiology* **33**, 59-73

Shtiza A, Swennen R, Tashko A (2008) Chromium speciation and existing natural attenuation conditions in lagoonal and pond sediments in the former chemical plant of Porto-Romano (Albania). *Environment and Ecology* **53**, 1107-1128

Sokal RR, Rohlf FJ (1981) *Biometry: The Principles and Practice of Statistics in Biological Research*, WH Freeman and Co., San Francisco, 859 pp

Sirko A, Hryniewicz M, Hulanicka D, Bock A (1990) Sulfate and thiosulfate transport in *Escherichia coli* K-12: Nucleotide sequence and expression of the cysTWAM gene cluster. *Journal of Bacteriology* **172**, 3351-3357

Skeffington RA, Shewry PR, Peterson PJ (1976) Chromium uptake and transport in barley seedlings (*Hordeum vulgare* L.). *Planta* **132**, 209-214

Srivastava LM (2002) *Plant Growth and Development – Hormones and Environment*, Academic Press, California, 772 pp

Staves RP, Knaus RM (1985) Chromium removal from water by three species of duckweeds. *Aquatic Botany* **23**, 261-273

Tripathi RD, Chandra P (1991) Chromium uptake by *Spirodela polyrrhiza* L. Schleiden in relation to pH and metal chelators. *Bulletin of Environmental Contamination and Toxicology* **47**, 764-769

Uysal Y, Taner E (2007) The effect of cadmium ions on the growth rate of the freshwater macrophyte duckweed *Lemna minor*. *Ekoloji* **16**, 9-15

Zayed A, Gowthaman A, Terry N (1998) Phytoaccumulation of trace elements by wetland plants: I. Duckweed. *Journal of Environmental Quality* **27**, 715-721

Zayed AM, Terry N (2003) Chromium in the environment: Factors affecting biological remediation. *Plant and Soil* **249**, 139-156

Zhang XH, Wang DQ, Huang M (2004) Development of electroplating sludge technology. *Journal of Guilin University of Technology* **24**, 502-506

Zhang XH, Liu J, Huang HT, Chen J, Zhu YN, Wang DQ (2007) Chromium accumulation by the hyperaccumulator plant *Leersia hexandra* Swartz. *Chemosphere* **67**, 1138-1143

Zurayk R, Sukkariyah B, Baalbaki R (2001) Common hydrophytes as bioindicators of nickel, chromium and cadmium pollution. *Water, Air, and Soil Pollution* **127**, 373-388