

## POTENTIAL OF WATER HYACINTH (*EICHHORNIA CRASSIPES* (MART.) SOLMS) FOR THE REMOVAL OF CHROMIUM FROM TANNERY EFFLUENT IN CONSTRUCTED POND SYSTEM

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**ABSTRACT:** The objective of the present study was to evaluate the potential use of water hyacinth for the removal of chromium (Cr) from tannery wastewater. This experiment was performed using healthy, young and acclimatized water hyacinth collected from unpolluted Awash River. Cr concentrations of 3, 5, 7, 10 and 20 mg/L were added to five different polyethylene tanks, containing 40 litre tap water cultured with Hoagland's solution. A sixth tank containing Cr-free water was used as a control group. Six plants of equal wet mass (each  $12.5 \pm 2$  g), shoot length ( $11 \pm 2$  cm) and root length ( $6 \pm 2$  cm) were transferred into each tank and allowed to grow in a greenhouse for 42 days. From each tank plants were harvested randomly every week. Bioaccumulation factor, translocation factor, shoot and root length; wet biomass and dry weight of the shoot and root were measured, and relative growth rate, tolerance index of the root and tolerance index of wet mass were analyzed. The maximum accumulation of  $2.52 \times 10^3$   $\mu\text{g/g}$  of water hyacinth was achieved in the plants exposed to 20 mg/L Cr solution. The root part of the plant accumulated 2.42 to 3.82 times higher than the shoot part. An overall Cr removal efficiency of up to 91% was achieved in this study, but the efficiency decreased as the concentration of Cr in water increased. The growth of the plant was inhibited at high concentration due to Cr toxicity. Therefore, the application of water hyacinth for Cr removal will be sustainable, if the concentration of Cr in wastewater does not exceed about 10 mg/L. The relative growth rate (RGR) of the plant decreased with increasing Cr concentration and the growth was inhibited above 15.3 mg/L Cr in water. Based on the above data, it could be concluded that water hyacinth can potentially be used for the removal of Cr from tannery effluents which is a major environmental problem in Ethiopia. However, further investigation is needed to ascertain the optimum conditions for maximum removal.

**Key words/phrases:** Bioaccumulation factor, chromium, phytoremediation, tannery effluent, water hyacinth

### INTRODUCTION

Water hyacinth, one of the rapidly growing and very productive free-floating aquatic plant, originated in the Amazon, South America (Bolenz *et al.*, 1990). Outside its native range, it was introduced into many countries during the late 19<sup>th</sup> and early 20<sup>th</sup> centuries (Cock *et al.*, 2000). In Ethiopia this plant was officially reported in the year 1956 in Koka Dam and Awash River (Stroud, 1991). Although water hyacinth is often seen as an invasive weed, studies showed that it has useful applications. Due to its fibrous tissue and high energy and protein content, water

hyacinth can be used for a variety of useful applications. A number of possible uses of the plant, some of which have been developed and others that are still in their infancy are summarized in an excellent review article by Anushee (2005).

One of its prime utilities that has found world-wide acceptance is in treating wastewaters (Tchobanoglous *et al.*, 1989). Water hyacinth has exceptionally high affinity and accumulation capacity for several metals (Zaranyika *et al.*, 1994; Zhu *et al.*, 1999). Natural wetland colonized by water hyacinth could serve as "nature's kidney" for proper effluent treatment to preserve the

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earth's precious water resource from getting polluted (Tchobanoglous *et al.*, 1989). Together with its capacity for metal ion accumulation, water hyacinth has attracted considerable attention because of its ability to grow in heavily polluted water (Anushee, 2005).

Water hyacinth has exceptionally high affinity and accumulation capacity for several metals. Therefore, it is used as a biomarker and is introduced in wetlands for water phytoremediation (Satyakala and Jamil, 1992). Zhu *et al.* (1999) investigated the ability of water hyacinth to take up and translocate six trace elements namely As(V), Cd(II), Cr(VI), Cu (II), Ni(II) and Se(VI) under controlled conditions. Cd, Cr, Cu, Ni and As were more accumulated in roots than in shoots whereas Se was accumulated more in shoots than in roots at the most external concentrations. Moreover, water hyacinth had high trace element bioconcentration factors (BCF) when supplied with low external concentrations of all six elements, particularly Cd (highest BCF=2150), Cr (1823) and Cu (595). Thus, it seems to be most efficient at phytoextracting trace elements and toxic pollutants from wastewater containing low concentrations of these elements. Recently, heavy metal phytoremediation by water hyacinth was demonstrated in constructed wetlands in Taiwan (Liao and Chang, 2004) where a high absorption capacity for Pb, Cu, Zn was recorded. A significant metal removal (over 90%) from aluminium industry effluents has also been obtained by employing water hyacinth (Roldán, 2002). Apart from this, investigations demonstrate that it can also tackle several recalcitrant organic chemicals such as herbicides (Roy and Hanninen, 1994).

In recent years, contamination of the environment by Cr, due to its huge industrial use, has become a major area of concern. Cr has been one of the important contaminant released into the environment (Nriagu and Nieboer, 1988). Cr, a non-essential micronutrient for normal plant metabolism, has been reported to be one of the most toxic heavy metals present in waste water discharges from electroplating, dye and pigment manufacturing, wood preserving and leather tanning industries. Often wastes from such industries are used as a fill material at numerous locations to reclaim marsh lands, and for backfill at sites following demolition. In many such sites, leaching and seepage of Cr from the soils into the

groundwater poses a considerable health hazard. In addition to being highly toxic, Cr is mobile, and has a long residence time in surface water and groundwater. As a result, it poses severe health risk to human beings, aquatic animals, and impairs the development and growth of plants (Chandra and Kulshreshtha, 2004; Mishra *et al.*, 2008 a and b). The tanning industry is a particularly large contributor of Cr pollution to water resources (Salunkhe *et al.*, 1998).

Ethiopia is one of the largest producers of leather in Africa and other industries that use Cr are growing in number. Tannery wastewaters are characterized by being strongly alkaline with a high oxygen demand and high content of salts and nutrients, one of which is Cr (Bajza and Vrcek, 2001). Nowadays chrome tanning is favoured by the majority of the leather industry because of the speed of processing, low cost, colour of leather and greater stability of the resulting leather (Hafez *et al.*, 2002). However, uptake of the Cr into the leather is not complete and relatively large amounts are found in the effluent. Estimates range from 2000–3000 mg/dm<sup>3</sup> (Bajza and Vrcek, 2001) to 3–350 mg/dm<sup>3</sup> (Vlyssides and Israilides, 1997). In Ethiopia, there is increasing concern about water pollution due to discharge of tannery effluents and paint factories without proper treatment. More than 80% of industries in the city of Addis Ababa and its vicinity discharge their effluent into the environment without treatment (AAEPA, 2004). As treatment of Cr-containing wastewater is expensive, many developing countries employ primary treatment alone which cannot reduce Cr to the level below legal discharge limit (Alves *et al.*, 1993). Advanced processes such as ion exchange, reverse osmosis, electrolysis and chemical precipitation are recommended (Vlyssides and Israilides, 1997; Hafez *et al.*, 2002). However, these methods are expensive and are often not considered cost effective for small sized industries.

Thus, in view of the seriousness of Cr pollution, considerable efforts have to be made to develop suitable methods for the remediation of Cr contaminated wastewater (Nriagu and Nieboer, 1988). Water hyacinth is one of the plant species that attracted considerable attention because of its ability to grow in heavily polluted water together with its capacity for metal ion

accumulation. Water hyacinth, whose increased affinity for nutrients uptake and highly explosive growth rate could be harnessed to remove Cr from the environment (Salt *et al.*, 1995). In general, the metal removal efficiency of water hyacinth is high at higher metal concentration but at the same time toxicity symptoms such as inhibition of chlorophyll synthesis and necrosis are serious drawbacks that require detailed investigation prior to field applications. Therefore, because of phytotoxicity problems at higher metal concentrations, optimum concentration range should be determined to utilize water hyacinth for wastewater treatment. In addition, information on the applicability of water hyacinth for the treatment of Cr contaminated water is very limited in the scientific literature and nonexistent in Ethiopia. The objective of this study is, therefore, to evaluate the potential of water hyacinth for the removal of Cr from wastewater and to determine the effect of Cr on the growth of water hyacinth.

## MATERIALS AND METHODS

### *Experimental site and the plant material*

The experiment was conducted in a greenhouse located at Addis Ababa University, over a period of 7 weeks between May and July 2008. The average temperature and relative humidity of the greenhouse were 26.9°C and 22.1%, respectively. The average water temperature and pH were 22.4°C and 6.4, respectively. The plant material, *Eichhornia crassipes* was obtained from uncontaminated Awash River water body in order to evaluate optimum uptake capacity.

### *Chromium removal experiment*

Hexavalent Chromium stock solution was prepared from 3.734 g  $K_2Cr_2O_7$  (dried at 100°C for 1 h) transferred to 1,000 mL volumetric flask containing 5 mL 1:1  $HNO_3$  and de-ionized water added to the mark (Hashintoni *et al.*, 1987). Cr(VI) concentrations of 3, 5, 7, 10 and 20 mg/L were added to 5 different polyethylene tanks containing 40 litre tap water cultured with Hoagland's solution. From the collected plant samples, healthy and young water hyacinth of similar shape and size were selected. Those plants were first acclimatized for a period of 7

days in a greenhouse without addition of nutrients. After 7 days, equal wet mass (wet mass of each plant  $12.5 \pm 2$  g), shoot and root length (aerial part  $11 \pm 2$  cm and root  $6 \pm 2$  cm) and equal number (6 plants in each pot) were transferred into round-bottom polyethylene containers of 60 cm depth and 50 cm width containing various concentrations of Cr and tap water cultured with Hoagland's nutrient solution. One control group was prepared in which the same volume of water cultured with Hoagland solution in a similar way but Cr was not added. The total volume of the solution in each container was kept constant by adding tap water cultured with Hoagland's solution every 5 days for water lost through plant transpiration and evaporation. Each experiment was performed in triplicates.

### *Sampling and sample preparation*

To determine Cr accumulated in the plant parts, water hyacinth samples were randomly harvested each week, from 9 to 11 A.M for six consecutive weeks (42 days) from all polyethylene tanks with different Cr concentrations (0, 3, 5, 7, 10 and 20 mg/L) and the control tank.

After harvesting the plant samples (water hyacinth), volumes of the water in the growth tank were adjusted to the original level. Water sample (500 mL) from all treatment tanks (3, 5, 7, 10 and 20 mg/L) were collected and filtered with Whatman filter paper No 41 (0.45  $\mu$ m pore size). Percent removal efficiency of the plant in each treatment tank was determined. The harvested plants were washed three to four times with tap water to remove any adsorbed materials on the plant external part. The shoot and root parts were oven dried at 85–90°C for 24 hours and then weighed, powdered and homogenized separately.

Well homogenized plant parts of 0.25 g were weighed and transferred into a round bottom flask (150 mL). 5 mL  $HNO_3$  and 2 mL  $H_2O_2$  was added and wet digested overnight. The digested solutions were then fitted to micro Kjeldahi digestion apparatus by setting the temperature dial to 150°C for 3 hours and 30 min. The digested solution was then cooled for 15 min at room temperature. After cooling, the solution was filtered into 50 mL volumetric flask fitted with Whatman filter paper (0.45  $\mu$ m) and the

samples were diluted with de-ionized water to the mark (Klumpp *et al.*, 2002).

From each collected water sample, 100 mL was transferred into 250 mL round bottom flask containing 2 mL HNO<sub>3</sub> and 1 mL HCl. The flask was placed on hot plate covered with an elevated watch glass. The temperature was adjusted to approximately 85°C. After 2 hours and 30 min the volume of the sample was reduced to 20 mL. The flask was allowed to cool for 15 min and transferred to 50 mL volumetric flask, the volume was adjusted to the mark with de-ionized water (Klumpp *et al.*, 2002).

The blank solution was prepared by digesting the mixture of reagents (69–72% HNO<sub>3</sub> and H<sub>2</sub>O<sub>2</sub>) following the same digestion and dilution procedures.

### Determination of chromium

The accumulation of metal in plant part is expressed in microgram (µg) of metal per gram (g) of dry matter. To determine the original Cr concentration in the plant part (before exposure to different Cr solutions), 6 plants were randomly chosen, their shoot and root parts were separated and Cr concentrations were analyzed at the Department of Chemistry, Addis Ababa University, using Atomic Absorption Spectrophotometer (AAS) equipped with deuterium arc (BUCK SCIENTIFIC MODEL 210VGP East Norwalk, USA).

The prepared sample solutions were aspirated into Flame Atomic Absorption Spectrometer and readings of Cr concentrations of the separate plant part (shoot and root) and water samples were recorded. Average values of three replicates were reported. Blank was also analyzed in the same manner as samples. The calibration curve was linear in the concentration ranges used in this study. The volumetric concentration was converted into actual mass concentration of metal in the sample by considering amount of sample digested and dilution factor.

### Data analysis

The Bio Concentration Factor (BCF) was determined using equation 1 (Zayed *et al.*, 1998).

$$\text{BAF} = \frac{\mu\text{g/g of Cr in plant body (dry mass)}}{\mu\text{g/g of Cr in external solution}} \dots\dots\dots(1)$$

The Translocation Factor (TF) which gives the root/leaf Cr concentration and depicts the ability of the plant to translocate the metal species from roots to shoot at different concentrations was estimated using equation 2 (Zayed *et al.*, 1998).

$$\text{TF} = \frac{\text{Cr contents of the root } \mu\text{g/g}}{\text{Cr content of the shoot } \mu\text{g/g}} \times 100 \dots\dots\dots(2)$$

Every week the shoot and root length of the harvested plant samples from all tanks were separated and measured using a ruler. Wet and dry masses of the shoot and root part were measured using analytical balance. Root tolerance index (RTI) of root length and wet biomass tolerance index (WMTI) of the total fresh weight were determined using equations 3 and 4, respectively (Zhu *et al.*, 1999). The higher the RTI the better is the tolerance.

$$\text{RTI} = \frac{\text{Root growth in metal containing solution (cm)}}{\text{Root growth in control group (cm)}} \dots\dots(3)$$

$$\text{WMTI} = \frac{\text{Wet biomass in metal containing solution (g)}}{\text{Wet biomass in control group (g)}} \dots\dots(4)$$

Plant relative growth rate (RGR) was evaluated according to Radford (1967) using equation 5.

$$\text{RGR} = \frac{\ln dm_2 - \ln dm_1}{t_2 - t_1} \dots\dots\dots(5)$$

where,  $dm_1$  and  $dm_2$  = Initial and final total dry mass;  $t_1$  and  $t_2$  = Time interval between two samplings (days).

Statistical analysis of the data was carried out using ANOVA. Statistical differences between treatments were determined by analysis of variance. Results were considered significant at  $p < 0.05$ .

## RESULTS AND DISCUSSION

### Accumulation of chromium in plant tissue

The data for accumulation of Cr in plant parts (shoot and root) grown in different Cr concentrations and exposure time are summarized in Table 1. The Cr concentration in shoot

and root increased with increasing Cr concentration and exposure time. Highest accumulation of Cr  $4.18 \pm 0.019$  mg/g (shoot + root) in water hyacinth was noted in plant exposed to 20 mg/L at the 42<sup>nd</sup> day (6<sup>th</sup> week) (Table 1). The analytical results in this study pertaining to Cr concentrations in root and shoot of plants in the study period revealed that, the highest accumulated concentration of Cr in the shoot was  $1.01 \pm 0.082$  mg/g on the 35<sup>th</sup> day and in the root was  $3.17 \pm 0.078$  mg/g on the 42<sup>nd</sup> day in the plant grown in 20 mg/L Cr solution. The results suggest that the plant accumulates more Cr in its root than in its shoot. Similar results were reported with other metals; water hyacinth grown in water containing Cd and Zn, showed higher accumulation of these metals as a result of increasing metal concentration in growing medium and exposure period (Hasan *et al.*, 2006).

Table 2 presents the mean Cr accumulation in shoot, root and the whole portion of the plant after exposure to 3, 5, 7, 10 and 20 mg/L Cr for 42

days. The mean Cr accumulation increased with increasing Cr concentration in both the shoot and root part. Deducting Cr content of the plant in the control group, the plant in 20, 10, 7, 5 and 3 mg/L Cr-containing water accumulates 2.52, 2.20, 2.04, 1.7 and 1.47 mg/g of Cr from first to fifth tank, respectively.

The present study shows that accumulation of Cr by water hyacinth is superior compared to other aquatic plants. For example, Sarital *et al.* (2001) reported that *Alternanthera sessilis* (rooted emergent) accumulated 1.21 mg/g; *Najas indica* (submerged) accumulated 473  $\mu$ g/g of Cr, when exposed to 8 mg/L Cr solution for 9 days. It should be noted however, that uptake of metals by plants is affected by several parameters such as pH, temperature and chemical constituents. In this study only exposure time and concentration of Cr were varied and we believe other variables that were not included could have played some role.

**Table 1. Cr uptake by water hyacinth ( $\mu$ g/g) exposed to different Cr concentrations in water.**

Conc (mg/L)	Plant part	Days of treatment						
		Initial	7	14	21	28	35	42
3	Shoot	6 $\pm$ 1	175 $\pm$ 1	302 $\pm$ 32	366 $\pm$ 33	437 $\pm$ 27	414 $\pm$ 25	507 $\pm$ 71
	Root	16 $\pm$ 1	751 $\pm$ 9	1,028 $\pm$ 176	1,390 $\pm$ 109	1,536 $\pm$ 72	1,721 $\pm$ 234	1,985 $\pm$ 135
	Total	22 $\pm$ 1	926 $\pm$ 10	1,329 $\pm$ 144	1,756 $\pm$ 78	1,972 $\pm$ 48	2,134 $\pm$ 259	2,491 $\pm$ 206
5	Shoot	5 $\pm$ 0.14	241 $\pm$ 5	385 $\pm$ 17	260 $\pm$ 26	493 $\pm$ 5	531 $\pm$ 22	661 $\pm$ 281
	Root	14 $\pm$ 1	918 $\pm$ 18	846 $\pm$ 70	1,647 $\pm$ 81	1,814 $\pm$ 129	2,148 $\pm$ 39	2,292 $\pm$ 70
	Total	19.49 $\pm$ 1	1,158 $\pm$ 15	1,232 $\pm$ 86	1,907 $\pm$ 55	2,307 $\pm$ 123	2,679 $\pm$ 29	2,953 $\pm$ 21
7	Shoot	6 $\pm$ 0.42	456 $\pm$ 7	473 $\pm$ 12	613 $\pm$ 32	893 $\pm$ 37	905 $\pm$ 35	941 $\pm$ 21
	Root	17 $\pm$ 1	1,144 $\pm$ 22	1,193 $\pm$ 34	1,507 $\pm$ 158	2,017 $\pm$ 19	2,173 $\pm$ 67	2,306 $\pm$ 91
	Total	23 $\pm$ 1	1,600 $\pm$ 14	1,666 $\pm$ 25	2,120 $\pm$ 129	2,910 $\pm$ 41	3,078 $\pm$ 39	3,247 $\pm$ 78
10	Shoot	6 $\pm$ 0.47	427 $\pm$ 21	471 $\pm$ 30	707 $\pm$ 33	841 $\pm$ 31	865 $\pm$ 45	948 $\pm$ 39
	Root	14 $\pm$ 0.39	1,140 $\pm$ 70	1,302 $\pm$ 46	1,592 $\pm$ 120	2,391 $\pm$ 136	2,502 $\pm$ 95	2,571 $\pm$ 92
	Total	20 $\pm$ 0.65	1,567 $\pm$ 85	1,773 $\pm$ 68	2,299 $\pm$ 148	3,232 $\pm$ 106	3,367 $\pm$ 56	3,519 $\pm$ 68
20	Shoot	6 $\pm$ 0.12	142 $\pm$ 10	616 $\pm$ 38	945 $\pm$ 51	825 $\pm$ 52	1,014 $\pm$ 83	1,011 $\pm$ 98
	Root	16 $\pm$ 0.30	1,152 $\pm$ 47	1,665 $\pm$ 67	2,663 $\pm$ 58	1,756 $\pm$ 102	3,000 $\pm$ 63	3,173 $\pm$ 78
	Total	22 $\pm$ 0.18	1,294 $\pm$ 37	2,281 $\pm$ 29	3,608 $\pm$ 6.35	2,582 $\pm$ 50	4,014 $\pm$ 20	4,184 $\pm$ 20
Control	Shoot	5 $\pm$ 0.33	9 $\pm$ 0.35	10 $\pm$ 0.27	17 $\pm$ 0.33	12 $\pm$ 0.09	15 $\pm$ 1	16 $\pm$ 1
	Root	15 $\pm$ 0.00	8 $\pm$ 0.43	15 $\pm$ 0.43	37 $\pm$ 0.59	35 $\pm$ 0.14	67 $\pm$ 0.29	55 $\pm$ 0.87
	Total	20 $\pm$ 2	17 $\pm$ 3	25 $\pm$ 4	54 $\pm$ 4	47 $\pm$ 5	82 $\pm$ 1	70 $\pm$ 2

**Table 2. Mean Cr accumulations in shoot, root and whole portion of water hyacinth exposed to different Cr concentrations for 42 days.**

Conc (mg/L)	Shoot ( $\times 10^2 \mu\text{g/g}$ )	$\pm$ SE	Root ( $\times 10^3 \mu\text{g/g}$ )	$\pm$ SE	Total ( $\times 10^3 \mu\text{g/g}$ )	$\pm$ SE
3	3.15	65	1.20	252	1.52	317
5	3.68	83	1.38	310	1.75	386
7	6.12	128	1.48	300	2.09	428
10	6.09	125	1.52	351	2.25	475
20	6.51	159	1.92	425	2.57	576
Control	0.11	2	0.031	9	0.044	10

SE= Standard error

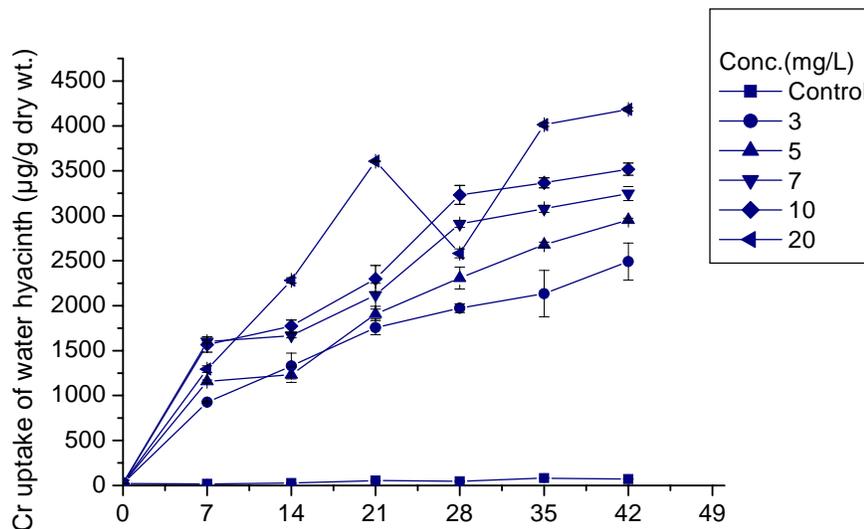
### *Effect of exposure time on chromium uptake*

The pattern of Cr uptake of water hyacinth with respect to exposure time and exposure level is shown in Figure 1. In this study, the maximum growth of water hyacinth was observed on the 35<sup>th</sup> and 42<sup>nd</sup> days of exposure time. During this period, the maximum accumulation of Cr for all treatment groups were noted and the plant in 20 mg/L accumulates best ( $4.18 \pm 0.019$  mg/g on the 42<sup>nd</sup> day). The plant growth declined and began to decay after the 42<sup>nd</sup> day (data not shown). The plant grown under 20 mg/L Cr condition showed a sharp peak uptake of  $3.60 \pm 0.006$  mg/g on the 21<sup>st</sup> day. This is due to the peak biomass growth observed during this period (Fig. 1). Other studies also reported that the uptake of metals by water hyacinth is high during the period of optimum growth (Soltan and Rashed, 2003; Jyaweera and Kasturiarachchi, 2004). The calculated regression coefficient between Cr uptake and its exposure time were found to be 0.919, 0.945, 0.883, 0.901 and 0.841, for the plant

grown in 3, 5, 7, 10 and 20 mg/L Cr-containing water, respectively. The plant Cr accumulation versus duration of exposure showed a linear relation for all exposure conditions. Thus, this observation shows that the extent of Cr uptake is highly dependent on exposure time. The slight inconsistency observed on day 21 for 20 mg/l Cr could be due to the observed variation in plant growth at this particular condition.

### *Effect of initial chromium concentration on uptake*

The uptake patterns of water hyacinth grown in different concentration of Cr containing water solution are given in Figure 2. The uptake increased with increasing concentration of Cr in water in the lower concentration range but as concentration increased further, reduced uptake was observed as shown in Figure 2. This may suggest that at higher concentration the plant may be susceptible to Cr toxicity. The biomass amount was also decreased during this period.



**Fig. 1. Concentration of Cr in all parts of water hyacinth treated at 3, 5, 7, 10 and 20 mg/L and Cr exposure condition for 42 days.**

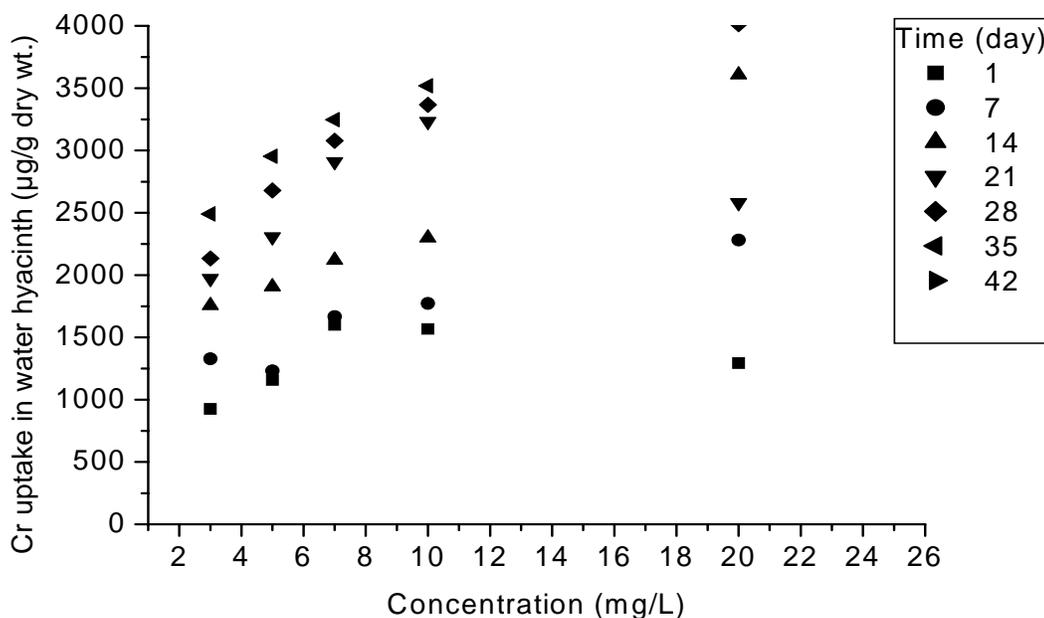


Fig. 2. Cr accumulation of the plant exposed to 3, 5, 7, 10 and 20 mg/L Cr containing water solution.

**Bioaccumulation factor (BAF) and translocation factor (TF)**

Data on BAF and translocation factor (TF) of water hyacinth resulting after exposure for 42 days to various concentration of Cr (3, 5, 7, 10 and 20 mg/L) are shown in Table 3. The BAF decreased with increasing Cr concentration and the accumulation of Cr in root was higher than in the shoot. The root tissue is in direct contact with the water and plants may develop mechanisms to minimize the accumulation of harmful ions in the shoot part (Rai and Chandra, 1992).

Table 3. BAF and TF of water hyacinth exposed to Cr for 42 days.

Cr conc. (mg/L)	BAF			TF (× 10 <sup>2</sup> )
	Shoot	Root	Total	
3	105	401	506	3.82
5	74	277	350	3.76
7	87	211	299	2.42
10	61	164	225	2.50
20	33	96	128	2.94

Metal accumulation by macrophytes can be affected by metal concentration in water or soil medium as indicated by Zayed *et al.* (1998). The concentration of Cr in water is the major factor influencing the metal uptake efficiency by the

plant. When the metal concentration in water increases, the amount of metal accumulation in plants increases, whereas the BCF values decrease (Sarital *et al.*, 2001). The present study also showed that high BAF value was achieved when the concentration in growth medium is low (3 mg/L).

The results in Table 3 also show that TF of Cr decreased with increasing Cr concentration up to 7 mg/L and increased again in 10 and 20 mg/L. High TF implies a poorer translocation capability (Zayed *et al.*, 1998). The maximum TF of 382 was observed for the plant exposed to 3 mg/L Cr. Water hyacinth absorbed Cr into the root first and translocated 20–29% into the shoot. The maximum translocation capacity (29%) which has the lowest TF (242) in this study was obtained when the plant is exposed to 7 mg/L Cr in water. The translocation capability of 20, 21, 23 and 27% was recorded for the water hyacinth grown in 3, 5, 20 and 10 mg/L Cr-containing water, respectively. This result is in close agreement with another similar study, which reported that water hyacinth accumulated heavy metals mostly to the roots and translocated only 6 to 25% to the shoots (Soltan and Rashed, 2003). Fine lateral roots of the water hyacinth reduce highly toxic Cr(VI) to the less toxic Cr(III), and then translocate relatively non-toxic Cr(III) to leaf

tissues (Lytle *et al.*, 1998). Lower accumulation of metals in shoot than root can be associated with the plant's ability to protect its photosynthetic tissue from toxic levels of trace elements (Baker, 1981; Landberg and Greger, 1996). This mechanism of partitioning from root to shoot is a common strategy of the plant that concentrates harmful ions in the roots in order to prevent toxicity to the leaves, the sites of photosynthesis and other metabolic activities (Sarital *et al.*, 2001).

Plants must have the ability to translocate Cr from the root to the shoot, or to compartmentalize it, in order to continue absorption of Cr from the external solution. Better translocation is advantageous for phytoextraction because it can reduce Cr concentration and thus reduce toxicity potential to the root (Baker, 1981). The differential localization of metals within the plant tissues may also be important in determining how well the metals may be bound and released from the plants (Suren, 1989). Translocation of trace elements from roots to shoots could be a limiting factor for the bioaccumulation of elements in the shoots.

As the concentration increased, the plant was not able to take up as much Cr which might be due to the fact that the Cr concentration in water exceeded the tolerance limit and the plant tissue may be injured. This suggests that Cr can best be removed from wastewater containing Cr concentration up to 10 mg/L. This is an indication that high removal efficiency could be achieved at lower Cr concentrations. Maine and Durate (2001) also reported that water hyacinth effectively removes appreciable quantity of heavy metals (Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn) from wastewater, especially at low concentrations.

#### *Chromium removal efficiency of water hyacinth*

Table 4 presents the Cr removal efficiency of water hyacinth grown in 3, 5, 7, 10 and 20 mg/L Cr containing water for 42 days. It is apparent that the removal efficiency decreased with increasing Cr concentration. The removal efficiency ranged from 69% (high concentration) to 91% (low concentration). Similar results have been

reported, with removal efficiency of 70% for water hyacinth exposed to 7 mg/L Cr concentration, for a period of 17 days (Keith *et al.*, 2006).

**Table 4. Variation of Cr removal efficiency as a function of initial concentration in water.**

Cr Conc in growth medium (mg/L)	Mean Cr Conc in growth medium after 42 days (mg/L)	Removal potential (%)
3	0.26±0.03	91
5	0.72±0.1	85
7	1.35±0.2	80
10	1.94±15	80
20	5.69±17	69

#### *Growth of water hyacinth in chromium containing water*

Results of mean shoot and root lengths (cm) and the root tolerance index (RTI) of water hyacinth after exposure to 3, 5, 7, 10 and 20 mg/L Cr for 42 days are given in Table 5. It was noted that the shoot and root length of the water hyacinth decreased with increasing Cr concentration. However, there is no significant difference between the plants grown in 3 and 5 mg/L Cr containing solution and the control group. The shoot and root length of the plant exposed to 7, 10 and 20 mg/L Cr solution significantly decreased ( $P<0.05$ ) compared to the control group.

**Table 5. Mean shoot and root length and root tolerance index of water hyacinth after 42 days exposure to Cr.**

Conc (mg/L)	Mean length (cm)				RTI
	Shoot	± SE	Root	± SE	
3	12.23	1.07	9.25	1.09	0.78
5	11.14	0.60	8.8	0.84	0.75
7	10.05	0.68	8.02	0.44	0.68
10	9.83	0.70	7.62	0.58	0.64
20	7.36	1.02	6.57	0.20	0.56
Con	13.02	1.11	11.83	1.55	

Con=Control; RTI = Root Tolerance Index; SE = Standard error.

Phytotoxicity symptoms are usually expressed as a percentage growth inhibition (Cervantes *et al.*, 2001). Figure 3 shows the shoot and root length growth of water hyacinth exposed to 3, 5, 7, 10 and 20 mg/L Cr-containing water solutions. The shoot length of the plant exposed to 20, 10 and 7 mg/L Cr concentration decreased by 43, 25 and 23% and the root length decreased by 44, 36 and 32% compared to the control group. It was reported that at higher Cr concentration (25 to 50 mg/L Cr), plants were stunted and had narrow and brownish red leaves with small necrotic areas and with poorly developed root system (Hunter and Vergnano, 1953). Growth change is often the first and most obvious reaction of plants under heavy metal stress (Hagemeyer, 1999). Wilting and plasmolysis of water hyacinth exposed to 10 and 20 mg/L Cr solutions was observed in this study. It was also reported that Cr can affect roots of plants causing wilting and plasmolysis (Roy and Hanninen, 1994). The finding of our study partly agrees with Hunter and Vergnano (1953) that they observed chlorotic leaves and normal roots at low Cr concentrations (5-10 mg/L).

Root tolerance index (RTI) of the plant decreased with increasing Cr concentration. This is in agreement with results of Shewry and Peter

son (1974) who further reported that the first toxic effect of Cr was inhibition of root growth. Shoot growth is only being affected at higher levels. This might be due to degradation of protein in plants which results in the inhibition of nitrate reductase activity (Panda and Choudhury, 2004). High production of  $H_2O_2$  and  $O_2^{1-}$  radicals were reported in many plant species exposed to Cr and the metal has been implicated in the generation of oxidative stress (Roy and Hanninen, 1994; Dixit *et al.*, 2001). It is also known that Cr is a non-essential heavy metal, and has inhibitory effects on plant growth.

The mean wet biomass of the shoot, the root and WMTI of the water hyacinth grown in various Cr concentrations for 42 days are given in Table 6. As can be seen from the table, both the shoot biomass and the root biomass decreased with increasing Cr concentration. However, the shoot and root biomass of the plant grown in 3 and 5 mg/L Cr solution did not significantly decrease but, the plant exposed to 7, 10 and 20 mg/L Cr solution significantly decreased ( $P < 0.05$ ), compared to the controlled group. It was also reported by other studies that Cr toxicity was evident in form of reduction of biomass of the plant (Hagemeyer, 1999).

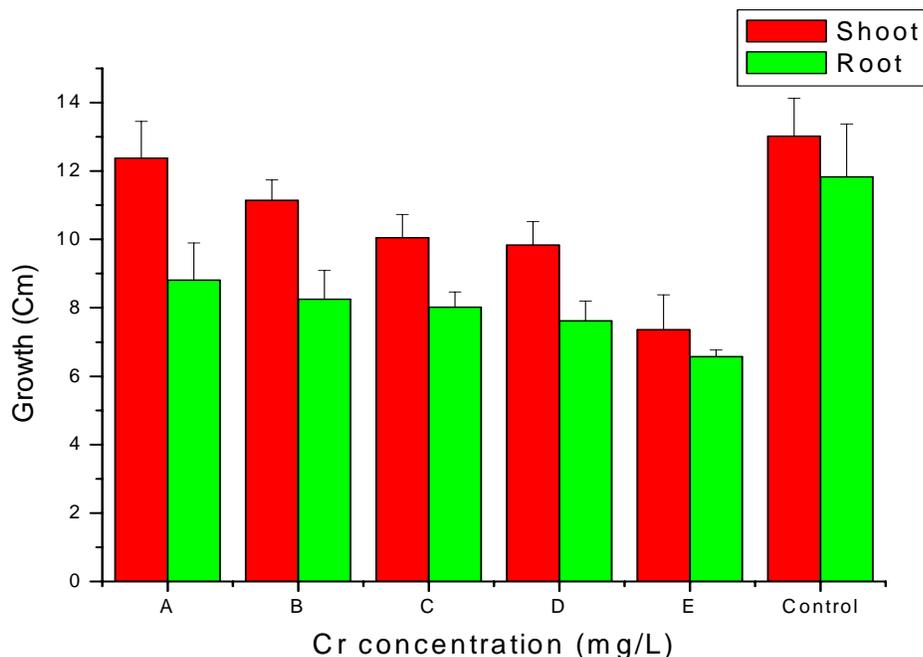


Fig. 3 Mean length of shoot and root (cm) of water hyacinth after 42 days exposure (A=3 mg/L; B=5 mg/L; C=7mg/L; D=10 mg/L; and E=20 mg/L).

Table 6. Mean wet biomass and wet mass tolerance index (WMTI) of water hyacinth after 42 days exposure to Cr.

Conc (mg/L)	Wet biomass (g)					WMTI
	Shoot	± SE	Root	± SE	Total	
3	24.29	5.80	4.18	0.76	28.47	6.52
5	22.68	5.46	4.13	1.44	26.81	6.78
7	19.55	4.08	4.05	1.02	23.60	5.07
10	17.17	3.43	3.65	0.30	20.82	3.73
20	9.82	0.48	2.72	0.23	12.54	0.52
Con	35.18	10.72	10.23	3.13	45.42	13.56

WMTI = Wet Mass Tolerance Index.

The WMTI of the plant decreased with increasing Cr concentration (Table 6). Visible damage symptom, like wilting and chlorosis was observed on plants exposed to 10 and 20 mg/L Cr solutions. It was reported that the initial symptoms of Cr toxicity appeared as severe wilting and chlorosis in water hyacinth (Turner and Rust, 1971). As a result of root damage, water and nutrient uptake is diminished and this is evidenced by wilting and reduced wet biomass and by visual symptoms that look like mineral deficiencies (*e.g.*, Fe deficiency chlorosis) in leaves. Reduced shoot wet biomass probably results in reduced leaf expansion. This was observed in this study, particularly at higher Cr concentrations. Turner and Rust (1971) proposed that chlorosis appeared in the upper leaves of the plant, as an indirect effect of Cr, probably due to the retardation of Fe and Zn translocation. In this study, the following sequence of sensitivity of the symptoms of Cr toxicity was observed: root wet mass > shoot wet mass > root length > shoot length. Other studies also observed the following sequence of sensitivity of symptoms of Cr toxic-

ity: root growth > visible damage symptoms > leaf growth (Hauschild, 1993).

#### *Variation of relative change in wet mass growth with time*

Variation of the total biomass growth of water hyacinth grown for 42 days under different Cr concentrations is shown in Figure 4. The growth increased with time with the exception of the plant grown in 20 mg/L Cr. The maximum wet mass growth was measured on the 42<sup>nd</sup> day (6<sup>th</sup> week) for all treatment groups, except for 20 mg/L Cr concentration. This may be attributable to the toxic effect of Cr at higher concentration. Other studies also reported that optimum growth of water hyacinth was observed when the plant attained 4 to 6 weeks of age (Jayaweera and Kasturiarachchi, 2004).

The effect of Cr on the growth rate of water (RGR) hyacinth is summarized in Table 7. The results show that the RGR decreased with increasing Cr concentration and at exposure to 20 mg/L of Cr the growth was negative. The negative value of RGR may be taken to indicate zero growth. It was considered that a decrease in the growth was induced by metal toxicity.

Table 7. Effect on the growth of water hyacinth following the exposure of Cr after 42 days.

Conc (mg/L)	Initial dry weight (g)	Dry weight after 42 days (g)	Growth	Growth rate ( $\times 10^{-2}$ )	RGR ( $\times 10^{-2}$ )
3	0.53	2.78	2.25	5.35	4.20
5	0.61	2.78	2.17	5.15	3.50
7	0.56	2.19	1.63	3.87	0.90
10	0.59	1.82	1.23	2.93	0.30
20	0.67	0.61	-0.06	-0.14	-0.20
Con	0.62	4.93	4.32	10.27	4.90

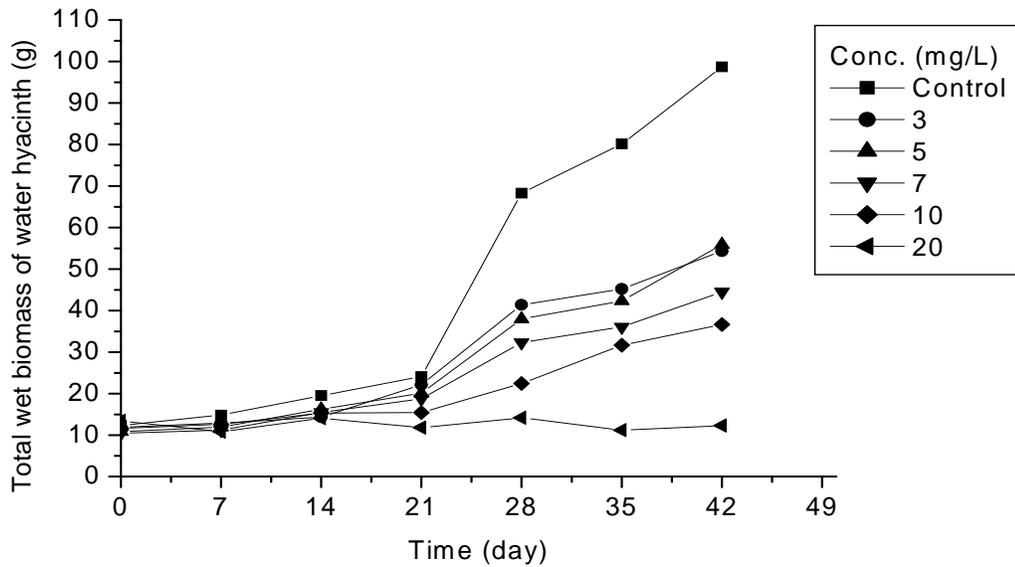


Fig. 4. Wet biomass growth pattern for water hyacinth exposed to different concentration of Cr.

The RGR curve of water hyacinth grown in 3, 5, 7, 10 and 20 mg/L Cr concentrations for 42 days is shown in Figure 5. It is apparent that the RGR curve intercepts the concentration axis approximately at 15.32 mg/L. This indicates that the

growth stopped after this concentration, due to the metal toxicity (Fig. 5). A study reported that the toxicity level of Cd to water hyacinth was 3.3 mg/L (Hasan *et al.*, 2006). This shows that Cr is less toxic than Cd.

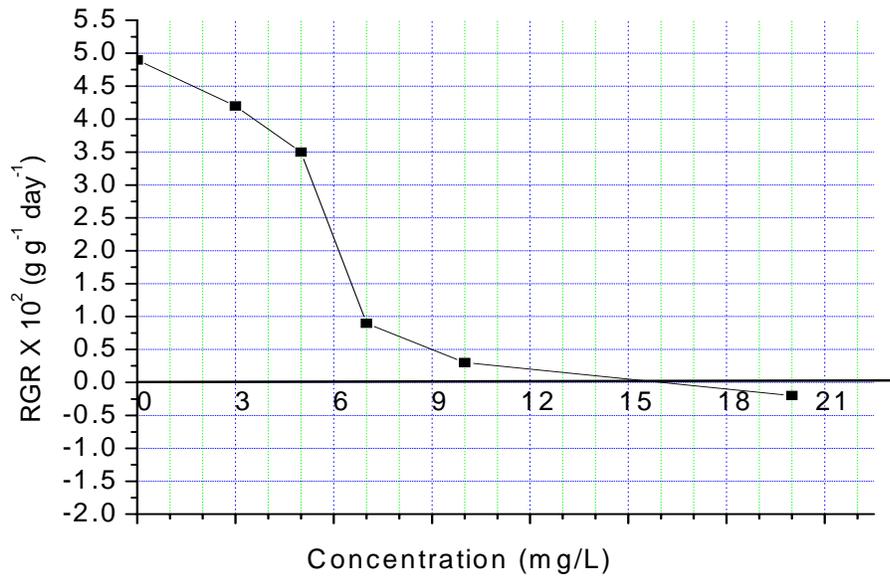


Fig. 5. Relative growth rate of water hyacinth exposed for 42 days in different initial concentration of Cr.

## CONCLUSIONS AND RECOMMENDATION

In this study, the potential of water hyacinth in the remediation of Cr-contaminated water has been demonstrated. Water hyacinth accumulated 2.52, 2.20, 2.04, 1.70 and 1.47 mg/g from 20, 10, 7, 5 and 3 mg/L Cr containing water in respective order and with BCF of 128, 225, 299, 350 and 506, respectively. As the concentration increased, the bioaccumulation potential decreased but the amount accumulated was still significant. The root part accumulated more Cr than the shoot. The Cr translocation factor of water hyacinth was 382, 376, 242, 250 and 294 for the plants grown in 3, 5, 7, 10 and 20 mg/L Cr solution, respectively. The poor translocation of Cr from root to shoot is a major hurdle in harvesting the plant after accumulation of Cr in plant. An overall Cr removal efficiency of up to 91% was achieved in this study, but the efficiency decreased as the concentration of Cr in water increased. The growth of the plant was inhibited at high concentrations due to Cr toxicity. Therefore, the application of water hyacinth for Cr removal will be sustainable, if the concentration of Cr in wastewater does not exceed about 10 mg/L.

Due to its wide industrial use, particularly in tanneries, Cr is considered a serious environmental pollutant in Ethiopia. Contamination of soil and water by Cr is of recent concern. Biological systems, such as constructed wetlands, are often very cost efficient and, therefore, the development of this experimental system into a large-scale working unit may offer an attractive alternative in Ethiopia where the cost of advanced technologies is prohibitive.

The problem of disposal of potentially large quantities of contaminated biomass is a hindrance to the implementation of developing phytoremediation systems. It is considered that disposal of chromium-contaminated water hyacinth could be achieved by initial desiccation, to reduce transport costs, and then incineration or ashing process where subsequent recovery of the Cr from the ash might be possible.

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