

Accumulation and tolerance characteristics of chromium in nine jute varieties (*Corchorus* spp. and *Hibiscus* spp.)

Muhammad Kamrul Islam¹, Iftekhar Alam^{2,3}, Mst Salma Khanam⁴, Si Young Lee¹, Tatoba R Waghmode², Moo Ryong Huh^{1,2*}

¹Department of Horticulture, College of Agriculture and Life Science, Gyeongsang National University, Jinju 660-701, Korea

²Institute of Agriculture & Life Science (IALS), Gyeongsang National University, Jinju 660-701, Korea

³National Institute of Biotechnology, Ganakbari, Savar, Dhaka 1349, Bangladesh

⁴Department of Agronomy, Bangladesh Agricultural University, Mymensingh 2302, Bangladesh

*Corresponding author: huhmr@hanmail.net

Abstract

This study investigates the effect of chromium (Cr) on seed germination, growth characteristics and the Cr accumulation in nine jute varieties. Two separate experiments *viz.*, germination and vegetative growth were analyzed. Plants were exposed to 0-500 ppm Cr for 30 days and were evaluated for growth, gas exchange, photosynthetic activity, tolerance index and Cr accumulation. Excess chromium negatively affects on germination parameters such as the germination frequency (GF), germination index (GI) and vigor index (VI). There was a clear difference in germination properties across the varieties particularly in O-795 and O-9897. All varieties were moderately tolerant to Cr toxicity, with O-795, CVE-3, and BJC-7370, showed small inhibitions in plant growth and photosynthetic characteristics than the others. Furthermore, O-795, CVE-3 and BJC-7370 exhibited higher Cr concentration in the root, stem, leaf, higher bio-concentration factor as well as higher biomass than other jute varieties. Therefore, O-795, CVE-3 and BJC-7370 jute varieties can be used as excellent annual crop candidates for phytoremediation of Cr polluted soils.

Keywords: chromium, Cr uptake, phytoremediation, bioaccumulation, jute.

Abbreviations: GF_germination frequency; GI_germination index; VI_vigor index; Pn_net photosynthesis; Gs_stomatal conductance; BCF_bioconcentration factor.

Introduction

Heavy metal contamination is one of the major ecological problems worldwide, leading to losses in agricultural yields and harmfully affects human health when contaminants enter the food chain. Cr is the seventh most abundant element on earth; and the second largest contributor of ground water, soil and sediment contamination (Shrivastava and Thakur, 2006). Contamination of agricultural fields with Cr is very toxic to both human being and plants and has been led a major environmental concern over the last few decades (Tiwari et al., 2013). Release of Cr compounds to the environment is mainly due to electroplating, leather tanning, metal finishing, corrosion control and pigment manufacturing industries (Liu et al., 2011). Annually about 3×10^4 , 1.42×10^5 and 8.96×10^5 tons of Cr are released to the atmosphere, water and soil, respectively (Shen, 2002). Cr has two stable forms, i.e. trivalent Cr (III) and hexavalent Cr (VI) form, later one is more toxic. These two forms are interconvertible in soil due to various microbial activities. High amount of Cr in the soil reduces plant growth. Moreover, at high concentrations, Cr acts as a mutagen, teratogen and carcinogen. Cr also causes deleterious effects on physiological processes of plants such as the photosynthesis and mineral nutrition (Diwan et al., 2010). Thus, there is an urgent and imperative need to develop efficient techniques for Cr removal from the environment. Physico-chemical method for Cr removal from the contaminated soils is very difficult, expensive and not

feasible for large scale application (Danh et al., 2009). However, some plants are able to withstand a very high level of Cr through their physiological mechanism. Phytoremediation has recently attracted a great deal of attention as an alternative means of soil decontamination. This process is cost-effective, eco-friendly and can be applied to large areas. In practical phytoremediation using hyperaccumulator plants has a number of drawbacks including slow plant growth and no practical use of their biomasses (Khan et al., 2000). Thus, attention goes to moderately heavy metal accumulating plants which produces a large amount of economically valuable biomass such as the poplars (Pietrini et al., 2010) and *Miscanthus* (Sharmin et al., 2012).

Several annual and perennial species have been evaluated for their phytoremediation potential. These include *Typha angustifolia* (Dong et al., 2007), *Convolvulus arvensis* and *Brassica juncea* (Lim et al., 2004), Water Lettuce, *Pistia stratiotes* L (Das et al., 2014) and *Prosopis* sp (Buendia-Gonzalez et al., 2010). Poplars are among the tree species has been tested and promoted for phytoremediation (Sebastiani et al., 2004). Many of these species are capable of sequestering high amount of Cr from the soil; however, has least or no economic value. Until now, Cr-response has been poorly documented compared to other heavy metals. Perennial herbaceous species are potential serious weed. On the other

hand, tree species has a long growing period and not suitable for agricultural crop field. These limitations positioned economically valuable annual crop species in an important place for the removal of Cr from the soil. However, though the reuse of the harvest biomass in non-food industries is being investigated (Citerio et al., 2002). In this regard, jute can be an excellent candidate for phytoremediation as it is non-edible and most of the parts can be used industry as well as for cooking purposes and unused parts can be disposed by incineration. Sas-Nowosielska et al. (2004) also proposed that incineration is the most feasible, economically acceptable and environmentally sound method for the disposal of metal contaminated crop.

Jute (*Corchorus* spp. and *Hibiscus* spp.) is a natural fiber crop and is second in the world after cotton in terms of global production, consumption and availability (Ranjit et al., 2013). It is an annual fiber crop with tall stem and deep penetrating taproot. The plant grows fast and easily in nutrient-poor soil and makes a heavy quantity of valuable biomass. It produces soft, shiny and long fiber for wide usages. It is a completely biodegradable, recyclable and eco-friendly lingo-cellulose fiber. Therefore, we assessed the Cr phytoremediation potential of jute with the idea of combining phytoremediation and the production of fibers and cellulosic biomass for practical use. The objectives of this study were: (1) to analyze the growth of plants in experimental Cr polluted soil; (2) to evaluate the Cr tolerance and accumulation capacity of jute and to select the better variety that can be used for phytoremediation of Cr contaminated soils.

Results and Discussion

Effects of chromium on seed germination

The GF, GI and VI decreased with increasing Cr concentrations (Fig. 1). At 50 ppm Cr, the GF and GI was not decreased significantly in all varieties, whereas VI was significantly decreased except CVE-3, BJC-7370, O-4 and O-795 compared to control. On the other hand, 100 ppm and 300 ppm Cr caused significant reduction of GF and GI except of CVE-3, BJC-7370, O-4 and O-795, whereas VI decreased significantly for all varieties except O-795 compared with control. The GF, GI and VI were significantly reduced drastically at 500 and 700 ppm Cr treatment except in O-795. For concentrations > 500 ppm, the germination parameter was significantly different ($p < 0.05$) among species at chromium treatments and control (Fig. 1). At all chromium treatments and control, the germination frequency of O-9897 was the lowest compared to other varieties. O-795 had the highest germination frequency, GI and VI followed by BJC-7370, CVE-3, O-72, VM-1, O-4, HS-24, CVL-1 and O-9897 (Fig. 1). Plants show a great variation in their tolerance to Cr in the environment and Cr toxicity depends upon the plant species and the source (Lopez-Luna et al., 2009). In our experiment, germination and viability of seeds were negatively affected by elevated Cr concentration. Excess Cr applications significantly inhibited the germination properties like GF, GI and VI, which are important seed germination parameters. On the basis of germination results, the tested varieties can be considered as moderately tolerant to Cr. There is a variation in Cr response, among the tested varieties, O-795 ranked highest in terms of Cr tolerance during germination, while O-9897 appeared as the sensitive. These changes might be related to genetic aberration in tested varieties by Cr heavy metal. In the current study, 50 ppm Cr showed no significant effect on seed germination, whereas, in

alfalfa 15 to 55% inhibition was found at 5 to 40 mg/l Cr and 17 to 44% in pea at 25 to 100 mg/l Cr in comparison with control (Peralta et al., 2001). Result documented previously stated that the high level (500 ppm) of Cr (VI) in soil reduced germination up to 48% in the bush bean *Phaseolus vulgaris* (Panday and Kumar, 2008) but current study shows high concentration (500 ppm) did not decrease germination of O-795, CVE-3 and BJC-7370 significantly. Thus, the Cr sensitivities of different parameters are species specific. Based on the measures of GF, GI, and VI, O-795, CVE-3 and BJC-7370 were supposed to be tolerant to Cr.

Plant growth and tolerance index

In addition to study of germination response, we tested plant growth in soil pots. Variable level of Cr was found in the contaminated sites around the globe. For instance, up to 1500 ppm Cr was reported in several sites in USA (Palmer et al., 1991; Sharmin et al., 2012). Based on earlier observations (Sharmin et al., 2012), in this experiment, we have chosen non-lethal toxic doses of Cr to understand the toxicity as well as phytoremediation potential. As shown in the Table 1, After 30 days of treatment, O-795 had the largest biomass, followed sequentially by CVE-3, BJC-7370, VM-1, HS-24, O-72, O-4, CVL-1 and O-9897. Treatment with low concentrations of Cr (100 ppm) significantly suppressed root length in CVL-1, O-9897, VM-1 and HS-24 but did not suppress root length in CVE-3, BJC-7370, O-4, O-72 and O-795. Treatment with 300 ppm Cr repressed root growth, whereas with high Cr concentration (500 ppm) markedly reduced root growth in all plant species except O-795. The reduction in root growth was the lowest in O-795 (18%), followed by BJC-7370 (31%), VM-1 (35%), O-4 (38%), HS-24 (42%), CVE-3 (44%), O-72 (47%), O-9897 (51%) and CVL-1 (52%), at the highest treatment (Table 1). In all species except O-795 stem length decreased as Cr concentration increased ($p < 0.05$). At a Cr concentration of 500 ppm, the smallest reduction in stem length was observed in O-795 (8%), followed by CVE-3 (10%), O-4 (16%) VM-1 (17%), BJC-7370 (18%), HS-24 (20%), CVL-1 (25%), O-72 (26%) and O-9897 (30%) (Table 1). Furthermore, the total biomass of the nine jute varieties decreased when soil Cr concentration increased. At low concentration (100 ppm) total biomass was not significantly repressed in five varieties out of the nine jute varieties. Treatment with 100 ppm Cr significantly reduced total biomass in CVL-1, O-9897, VM-1 and HS-24 but not in CVE-3, BJC-7370, O-4, O-72 and O-795 (Table 1). Application of 300 ppm and 500 ppm Cr significantly reduced total biomass in any of variety compared to control. The least reduction in total biomass was seen in O-795 (17.56%), followed by CVE-3 (26.67%), VM-1 (30.15%), BJC-7370 (30.68%), HS-24 (36.11%), O-72 (40.21%), O-4 (41.19%), CVL-1 (46.61%) and O-9897 (52.30%). Among the tested varieties, O-795, CVE-3 showed better performance in total biomass. Also, tolerance index (TI) of nine jute varieties exposed to four concentrations of Cr is shown in Fig. 2. A metal tolerance index based on biomass indicated significant difference in relation to metal treatment. The TI of the total biomass decreased significantly when soil Cr concentration increased. The tested varieties O-795 and CVE-3 had the higher tolerance indices at all treatment levels, indicating that these varieties had a higher tolerance to Cr. The lowest TI was found in O-9897 at all Cr concentrations. Earlier studies reported that plants can suffer to toxicity effects if soil total Cr concentration reaches to >2 mg/kg, the soluble (bioavailable) Cr concentration >0.001

Table 1. Determination of stem length (m), root length (m) and total biomass (g/p) of fiber crops treated with Cr (Different letter indicates significant difference at p <0.05 level among different species, mean± SE, n=3).

Treatment (PPM)		CVE-3	CVL-1	BJC-7370	O-4	O-72	O-795	O-9897	VM-1	HS-24
Root length (m)	0	0.41±0.01a	0.33±0.01a	0.42±0.02a	0.37±0.01a	0.34±0.01a	0.38±0.02a	0.40±0.02a	0.49±0.02a	0.42±0.02a
	100	0.39±0.01a	0.28±0.02b	0.39±0.01a	0.34±0.01a	0.32±0.01a	0.37±0.01ab	0.29±0.01b	0.41±0.01b	0.37±0.01b
	300	0.30±0.01b	0.22±0.02c	0.34±0.01b	0.29±0.02b	0.26±0.02b	0.33±0.01bc	0.24±0.01c	0.33±0.01c	0.32±0.01c
	500	0.23±0.02c	0.16±0.01d	0.29±0.01c	0.23±0.02c	0.18±0.01c	0.31±0.01c	0.19±0.01d	0.30±0.03c	0.24±0.01d
Stem length (m)	0	2.34±0.01a	2.25±0.01a	2.28±0.01a	2.16±0.03a	2.29±0.02a	2.21±0.02a	2.08±0.02a	1.49±0.02a	1.64±0.04a
	100	2.31±0.01a	2.04±0.02b	2.23±0.01b	2.14±0.01a	2.26±0.02a	2.17±0.02ab	1.81±0.01b	1.37±0.02b	1.50±0.04b
	300	2.21±0.02b	1.87±0.04c	1.96±0.01c	1.99±0.03b	2.08±0.04b	2.09±0.04ab	1.65±0.01c	1.30±0.01c	1.40±0.02bc
	500	2.1±0.03c	1.69±0.03d	1.88±0.02d	1.82±0.02c	1.70±0.01c	2.04±0.04b	1.45±0.01d	1.24±0.01c	1.24±0.02c
Total biomass (g/p)	0	33.28±1.00a	33.53±0.34a	34.80±0.63a	28.62±0.26a	32.38±0.54a	32.33±0.39a	23.19±0.20a	33.89±0.21a	32.42±0.18a
	100	32.12±1.38a	25.97±0.17b	32.69±0.82a	27.54±0.36a	30.63±0.66a	30.88±0.47a	17.17±0.17b	29.64±0.11b	27.43±0.15b
	300	28.08±0.53b	22.66±0.43c	27.49±0.71b	22.24±0.39b	24.28±0.35b	28.77±0.64b	13.63±0.22c	27.09±0.21c	23.85±0.13c
	500	24.41±0.93b	17.89±0.36d	24.12±0.30c	16.83±0.24c	19.36±0.59c	26.65±0.27c	11.06±0.24d	23.67±0.05d	20.71±0.22d

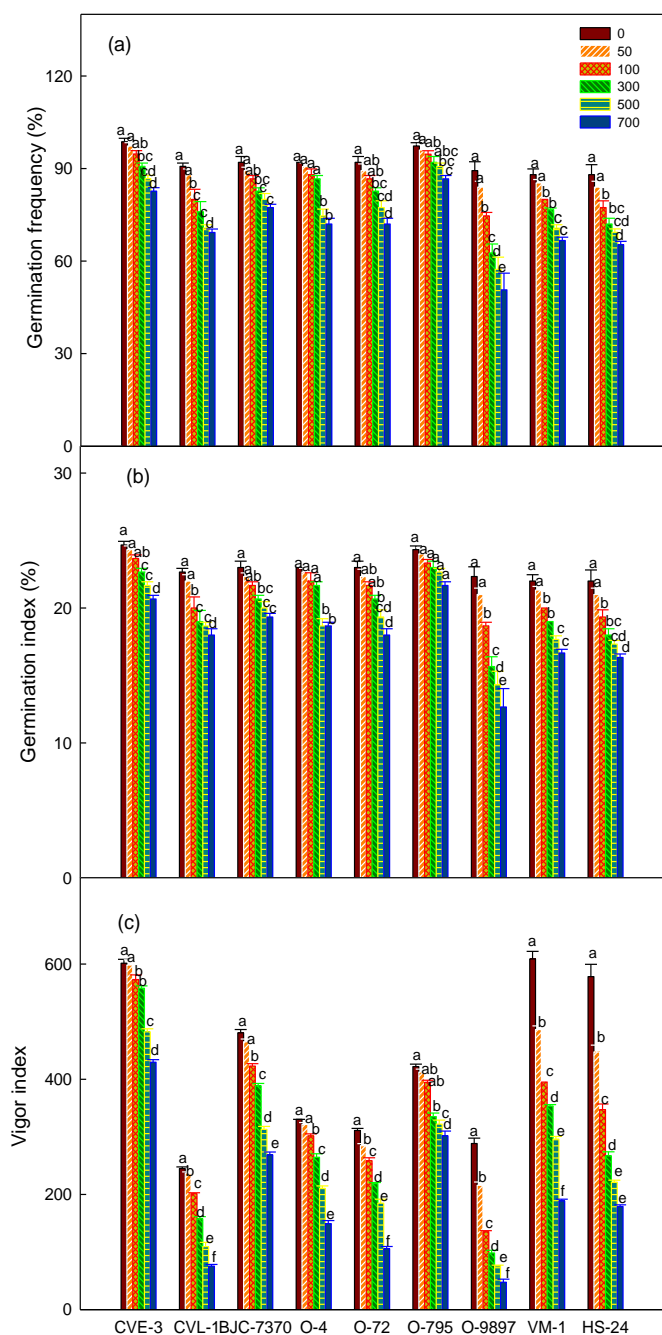


Fig 1. (a) Germination frequency, (b) germination index and (c) vigor index of nine jute varieties subjected to different Cr concentrations. Each value represents the mean \pm standard error (n=3). Means for each crop followed by the same letter are not significantly different at the level of $p < 0.05$ based on LSD test.

mg/kg, or tissue Cr concentration 2-8 mg/kg dry weight (Magnicol and Beckett, 1985). We observed that the tested jute cultivars were moderately tolerant to Cr. All plants survived, even at soil Cr concentrations of 500 ppm. Our results suggest that the tested plant varieties had an innate tolerance to Cr stress. Several indicators such as root, stem and leaf biomass and plant growth rate were considered to evaluate metal toxicity. The tolerance index indicated that the all varieties could grow well under Cr stress (100-500 ppm

Cr concentration) (Fig. 2). Lima et al. (2006) found that root was more sensitive than shoot to heavy metals in terms of elongation and biomass. The above results indicated that biomass could be a better indicator of Cr toxicity than shoot and root elongation but that roots are more sensitive than shoots. Our result was consistent with previously reported results (Shi et al., 2012). Tolerance index based on the root length, stem length and biomass in the selected jute varieties were chosen as indicators of the toxic effects of metals on plants. TI values normally ranged from 10 to 200. Lower the TI value, the greater the toxic effects of metals on plants (Yang et al., 2010). The variations in tolerance indices to various heavy metals indicate that genetically based tolerance may exist in populations that could survive heavy metal contaminated habitats. The Cr sensitivities of different parameters are species-specific. Therefore, the shoot and root biomass, as well as their TI were used as indicator to evaluate Cr toxicity to nine jute varieties. Among the tested varieties, O-795, CVE-3 and BJC-7370 were more tolerant to Cr than other varieties, based on measures of plant growth (Table 1) and TI (Fig. 2). However, O-795 was the most tolerant and O-9897 the most sensitive varieties.

Chlorophyll fluorescence and gas exchange

The measurement of chlorophyll a fluorescence (Fv/Fm) is shown in Fig. 3. Low concentrations of Cr (100 ppm) did not inhibit the Fv/Fm in all tested varieties except CVL-1, VM-1, HS-24 and O-9897. In the treatment with 300 ppm Cr, Fv/Fm was significantly repressed in all varieties except CVE-3, BJC-7370 and O-795 while in 500 ppm Cr treatment, it was reduced in most of the varieties except CVE-3 and O-795 at 10 days after treatment (DAT) (Fig. 3a). Although at 20 DAT Fv/Fm was significantly decreased in all varieties except O-795 at all Cr concentrations (Fig. 3b). The highest inhibition of chlorophyll fluorescence was noticed in O-9897, while O-795 was least affected. The response of different parameters of photosynthesis to Cr toxicity is shown in Table 2. Usually, leaf net photosynthesis (Pn), stomatal conductance (Gs), intercellular CO₂ concentration (Ci) and transpiration rate (E) were decreased in all Cr treatments and varieties. In all varieties Pn declined with increasing Cr concentration during the course of the experiment and the lowest values were recorded in O-9897 (Table 2). Low concentrations of Cr (100 ppm) significantly inhibited the Pn in all tested varieties except CVE-3, BJC-7370 and O-795. In the treatment with 300 ppm Cr, Pn was significantly repressed in all varieties except O-795 while in 500 ppm Cr treatment; it was reduced in most of the varieties except O-795 at 10 DAT (Table 2). Although at 20 DAT Pn was significantly decreased in all jute varieties except O-795 at different concentrations of Cr (Table 2). At the highest Cr concentration (500 ppm), the smallest reduction in Pn was seen in O-795 (19.49%) and the highest reduction was observed in O-9897 (54.72%) at 10 DAT. The photosynthetic rate was markedly reduced at 20 days with higher magnitude compared to 10 day observation with similar manner. Stomatal conductance, inter cellular CO₂ concentration and transpiration rates exhibited similar trends (Table 2). This result correlates with the biomass accumulation pattern. Among the tested varieties CVE-3 and O-795 showed less reduction in all gas exchange parameters compared to other varieties in all treatment label, indicating that these two varieties had a higher tolerance to Cr. There was a very clear effect of Cr treatment on Pn, Ci, E and Gs on nine jute varieties during two stages of growth period, after 10 and 20 days. The decline in Pn might be attributed due to

Table 2. The effect of chromium on net photosynthesis (Pn), stomatal conductance (Gs), transpiration rate (E) and intercellular CO₂ concentration (Ci), in leaves of jute varieties measured at irradiance on the topmost fully expanded leaf after 20 days of treatment. (Different letter indicates significant difference at p <0.05 level among different species, mean ± SE, n=3).

variety	Treatment (ppm)	Pn ($\mu\text{mol m}^{-2}\text{s}^{-1}$)		Gs ($\text{mol m}^{-2}\text{s}^{-1}$)		E ($\text{m mol m}^{-2}\text{s}^{-1}$)		Ci ($\mu\text{mol mol}^{-1}$)	
		10 DAT	20 DAT	10 DAT	20 DAT	10 DAT	20 DAT	10 DAT	20 DAT
CVE-3	0	26.97C1.01a	28.62±0.82a	0.47±0.005a	0.49±0.009a	4.83±0.080a	4.93±0.091a	700±9.60a	811.33±11.34a
	100	24.82±0.73a	23.87±0.50b	0.44±0.006a	0.44±0.006b	4.67±0.046a	4.68±0.061a	677±20.55a	672.67±11.05b
	300	20.16±1.03b	17.30±0.55c	0.37±0.014b	0.32±0.014c	3.47±0.049b	3.14±0.087b	504±5.45b	581.33±6.33c
	500	18.33±0.67b	14.48±0.45d	0.34±0.015b	0.25±0.015d	3.26±0.018c	2.56±0.217c	464±6.49c	533.67±3.76d
CVL-1	0	20.40±0.56a	22.56±1.66a	0.41±0.006a	0.41±0.030a	3.69±0.019a	4.06±0.143a	533±7.05a	662.00±17.39a
	100	15.16±0.59b	10.95±0.75b	0.27±0.006b	0.19±0.012b	2.47±0.015b	1.86±0.090b	396±2.88b	428.67±5.24b
	300	11.56±0.41c	7.95±0.37bc	0.18±0.008c	0.13±0.011c	1.83±0.090c	1.42±0.049c	326±14.62c	362.67±21.40c
	500	10.17±0.35c	6.60±0.55c	0.17±0.011c	0.12±0.010c	1.57±0.204c	1.16±0.032c	275±8.65d	326.33±27.33c
BJC-7370	0	24.93±0.56a	26.63±0.92a	0.47±0.010a	0.49±0.012a	4.48±0.065a	4.60±0.078a	637±9.75a	760.67±22.58a
	100	23.76±0.69a	23.01±0.21b	0.45±0.012a	0.45±0.009a	4.24±0.044a	4.10±0.059b	618±6.98a	650.67±10.53b
	300	17.20±0.52b	15.95±0.85c	0.33±0.006b	0.28±0.015b	3.05±0.145b	2.70±0.020c	551±11.25b	519.33±10.65c
	500	15.00±0.47c	12.94±0.43d	0.29±0.006c	0.23±0.009c	2.72±0.046c	2.21±0.038d	453±6.35c	459.67±22.88d
O-4	0	21.40±0.69a	24.18±1.14a	0.39±0.011a	0.44±0.020a	3.90±0.059a	4.21±0.090a	560±2.72a	799.00±4.51a
	100	19.93±0.32a	14.00±0.58b	0.34±0.012b	0.25±0.010b	3.04±0.055b	2.41±0.043b	449±18.35b	551.33±34.06b
	300	15.01±0.84b	11.49±0.28c	0.30±0.012c	0.20±0.009c	2.53±0.015c	1.93±0.042c	386±6.08c	501.67±12.81b
	500	12.58±1.26b	9.29±0.27d	0.25±0.012d	0.15±0.006d	2.18±0.046d	1.55±0.066d	305±7.50d	412.33±16.02c
O-72	0	17.33±0.46a	19.56±0.76a	0.32±0.011a	0.36±0.012a	3.07±0.067a	3.47±0.204a	452±15.85a	696.00±7.37a
	100	12.22±0.22b	9.57±0.83b	0.24±0.010b	0.17±0.006b	2.05±0.080b	1.69±0.136b	334±8.14b	395.67±8.09b
	300	9.77±0.35c	6.77±0.56c	0.18±0.012c	0.11±0.005c	1.50±0.030c	1.17±0.065c	270±2.18c	320.67±8.25c
	500	8.59±0.74c	5.51±0.31c	0.15±0.012c	0.10±0.006c	1.28±0.015d	0.97±0.094c	229±12.97d	266.00±9.54d
O-795	0	22.01±0.59a	22.99±1.10a	0.39±0.006a	0.42±0.015a	3.71±0.177a	3.89±0.047a	540±24.57a	744.67±12.46a
	100	20.95±0.66ab	21.81±0.62ab	0.37±0.012ab	0.39±0.014ab	3.58±0.205ab	3.74±0.043ab	527±21.94a	703.67±3.84ab
	300	19.30±0.37bc	20.55±0.46b	0.34±0.012bc	0.35±0.012bc	3.37±0.086ab	3.60±0.080b	486±5.60a	675.67±4.06b
	500	17.72±0.35c	17.00±0.11c	0.32±0.013c	0.33±0.009c	3.17±0.032b	3.27±0.056c	419±6.22b	555.67±12.41c
O-9897	0	16.10±0.52a	18.73±0.79a	0.30±0.006a	0.34±0.015a	2.93±0.055a	3.31±0.164a	447±8.56a	685.67±6.17a
	100	11.48±0.75b	8.29±0.40b	0.23±0.006b	0.15±0.006b	1.90±0.089b	1.44±0.012b	289±12.66b	336.33±7.75b
	300	8.97±0.98bc	6.56±0.70bc	0.17±0.012c	0.10±0.016c	1.32±0.050c	1.10±0.050c	256±21.57b	296.00±11.53c
	500	7.29±0.81c	4.95±0.51c	0.14±0.012d	0.09±0.012c	1.15±0.035d	0.85±0.046c	208±1.52c	237.33±2.19d
Vm-1	0	26.90±1.18a	28.06±1.56a	0.50±0.010a	0.50±0.012a	4.66±0.265a	4.96±0.276a	692±5.48a	811.00±6.25a
	100	22.27±0.90b	16.63±0.57b	0.41±0.006b	0.30±0.017b	3.91±0.087b	2.88±0.112b	583±24.05b	668.00±27.53b
	300	17.26±0.27c	13.85±0.60bc	0.33±0.020c	0.24±0.009c	3.20±0.128c	2.34±0.096c	487±5.50c	524.00±42.14c
	500	16.03±0.92c	11.00±1.06c	0.29±0.017c	0.19±0.007d	2.76±0.165d	1.87±0.047c	411±40.37c	464.33±7.88c
HS-24	0	24.30±0.99a	25.51±0.81a	0.45±0.006a	0.47±0.020a	4.37±0.048a	4.53±0.195a	642±17.50a	806.33±8.97a
	100	19.20±0.47b	15.23±0.67b	0.36±0.011b	0.27±0.012b	3.42±0.030b	2.60±0.069b	519±28.67b	55.67±8.67b
	300	17.00±0.63bc	12.34±0.92c	0.32±0.006c	0.23±0.006b	2.91±0.085c	2.15±0.051c	447±26.02b	516.67±8.01c
	500	14.83±1.04c	9.85±0.25d	0.28±0.005d	0.17±0.005c	2.50±0.042d	1.64±0.070d	374±12.89c	440.00±11.72d

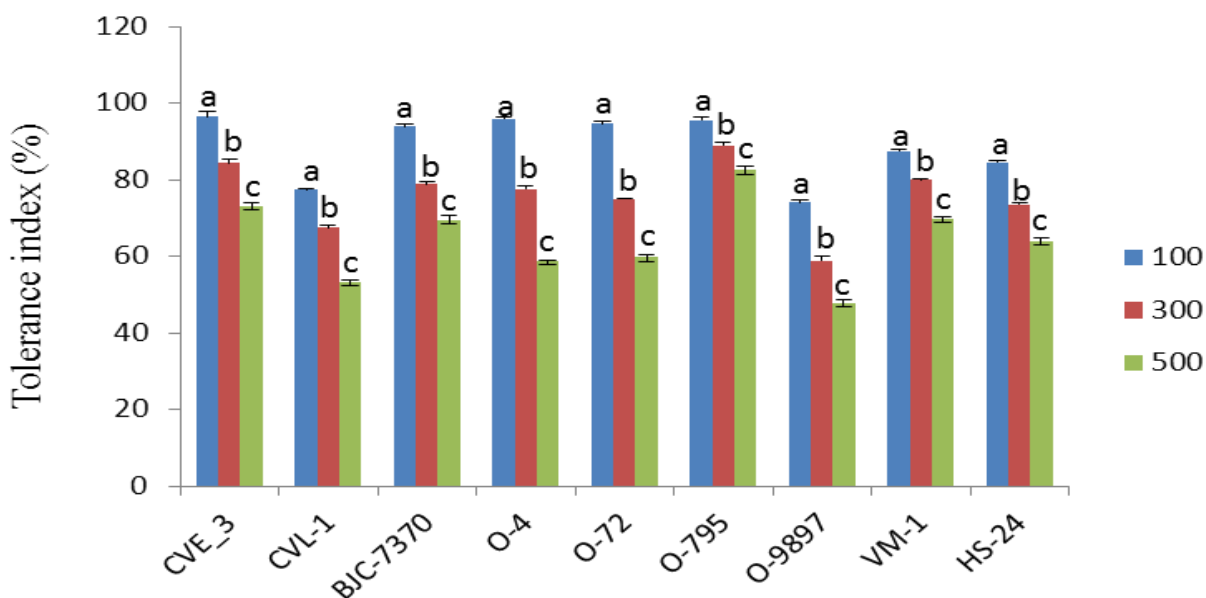


Fig 2. Tolerance index of nine jute varieties subjected to different Cr concentrations. Each value represents the mean \pm standard error (n=3). Means for each crop followed by the same letter are not significantly different at the level of $p < 0.05$ based on LSD test.

stomatal and/or non-stomatal limitations, thus Cr stress can affect photosynthesis in terms of CO_2 fixation, electron transport, photophosphorylation and enzyme activities (Shanker et al., 2005). Therefore, if the limitation of Pn in all varieties due to Gs, there would be a reduction in intracellular CO_2 concentration. Paiva et al. (2009) reported that the decrease in Pn caused by Cr (IV) probably the damage suffered by the photosynthetic system based on the decrease of the maximum quantum efficiency of PSII photochemistry (Fv/Fm). Also, Appenroth et al. (2001) reported that, Cr has been found to severely affect the performance index of PSII by reducing the total number of active reaction centers per absorption and damage to the oxygen-evolving complex. Another study has revealed that PSII, particularly core complex and connecting antenna, are more sensitive to Cr than PSI and light harvesting complex of PSII (Appenroth et al., 2003). In our study, Fv/Fm showed a significant reduction in all tested varieties in all growth stages compared to control. Moreover, Fv/Fm was the most sensitive parameter to Cr concentration in all stages of the experiment, denoting that excess Cr enhances photoinhibition induced by light stress. Liu et al. (2008) found that higher concentration of hexavalent Cr decreased Pn, Ci, Gs and E in *Amaranthus viridis*. It was also reported that Cr had a negative impact on photosynthetic parameter in other plants (Subrahmanyam, 2008).

Chromium accumulation in plants

Chromium concentrations in the roots, stems and leaves of the nine jute varieties are presented in Table 3. In general, plant Cr content increased as Cr concentration increased in the soil. Most of the Cr absorbed by the plants was present in the roots. The root Cr concentration was the highest in O-795; in contrast VM-1 accumulated the lowest. The stem Cr content was the highest in O-795 and the lowest in HS-24. However, the highest Cr content in leaf was found in O-795 and lowest in O-9897. The bioconcentration factor (BCF) for all other crops decreased as the supplied Cr increased (Table

3). The root BCFs were high in all varieties, while the highest in O-795, followed by CVE-3, HS-24, O-9897, O-72, CVL-1, O-4, BJC-7370 and VM-1. The stem BCF was the highest in O-795 followed by CVE-3, BJC-7370, O-4, O-72, O-9897, CVL-1, VM-1 and HS-24. The highest leaf BCF was observed in O-795, followed by CVE-3, BJC-7370, VM-1, O-4, CVL-1, O-9897, HS-24 and O-72. Translocation factors of the nine varieties were low (Table 3) suggesting that these varieties have a low capacity to move Cr from root to shoot. Translocation factors (TFs) of all cultivars were ranged from 27 to 82 % (Table 3), indicating that the studied jute varieties have relatively low capacity to translocate Cr from root to shoot compared to the hyperaccumulator plants. BJC-7370 had the highest TF (29-82%), while O-9897 showed the lowest (27-40%). Shoot Cr uptake is an indicator of the ability of phytoextraction. Although O-795, CVE-3 and BJC-7370 bio concentration factors was not as good as that of hyperaccumulator plants, these three varieties still had high extraction potentials because of their large biomass. Among the tested varieties CVE-3, BJC-7370 and O-795 had a higher Cr accumulation because of their high root, stem and leaf Cr content and large biomasses. Metal concentrations in plants vary with plant species and varieties, as well as by various soil conditions. Plant uptake of heavy metals from soil occurs either passively with the mass flow of water into the roots, or through active transport crosses the plasma membrane of root epidermal cells. Roots generally accumulated much higher amount of heavy metals than shoots. The results of the present study also showed that the most Cr was found in the roots, substantial amounts were found in the aerial parts (shoots), especially in leaf of O-795, CVE-3 and BJC-7370, where the Cr concentration was >800 mg/kg dry mass, indicating low mobility of Cr from the roots to the shoots and immobilization of heavy metals in the roots. This observation warrants further study of these varieties in field level. In contrast, in stems of O-795, CVE-3, BJC-7370 and O-4 where the Cr concentration was < 100 mg/kg dry mass, higher than other varieties. In leaf Cr was found higher than in the stem. The bio-concentration factor (BCF) is considered

Table 3. Content of Cr in plant tissues, bioconcentration factor (BCF) and translocation factor (TF) of jute varieties grown in Cr treated substrates. (Different letter indicates significant difference at p <0.05 level among different species, mean± SE, n=3).

Treatment (ppm)	Cr content (mg/kg)	Biococentration factor(BCF)			Translocation factor(TF)			
		Root	Stem	leaf	Root	stem	Leaf	
CVE-3	0	34.33±0.72c	2.97±0.36d	6.64±0.14d				0.28±0.000c
	100	1022.72±15.28b	53.95±0.64c	465.57±4.94c	10.23±0.18a	0.54±0.005a	4.65±0.05a	0.51±0.012b
	300	1280.19±22.84a	62.47±1.49b	573.87±2.32b	4.27±0.09b	0.21±0.006b	1.91±0.01b	0.50±0.012b
	500	1320.41±21.68a	71.42±1.07a	656.54±18.43a	2.64±0.05c	0.14±0.003c	1.31±0.03c	0.55±0.017a
CVL-1	0	32.37±0.88d	2.51±0.26d	6.65±0.22d				0.28±0.009d
	100	569.15±3.32c	40.81±0.67c	153.36±4.32c	5.71±0.03a	0.41±0.006a	1.54±0.04a	0.34±0.009c
	300	739.20±2.14b	49.78±0.39b	262.71±1.61b	2.46±0.01b	0.17±0.003b	0.88±0.01b	0.42±0.003b
	500	924.86±1.02a	55.74±0.39a	368.79±0.95a	1.85±0.00c	0.11±0.000c	0.73±0.00c	0.46±0.000a
BJC-7370	0	32.66±0.88d	2.65±0.15d	6.65±0.17d				0.29±0.007d
	100	550.68±1.19c	49.69±0.87c	370.88±0.68c	5.51±0.01a	0.50±0.007a	3.71±0.01a	0.76±0.003b
	300	716.10±6.83b	56.08±1.18b	475.99±1.58b	2.38±0.02b	0.18±0.003b	1.59±0.01b	0.74±0.007c
	500	801.78±2.86a	67.83±1.56a	589.89±2.25a	1.60±0.01c	0.13±0.003c	1.18±0.01c	0.82±0.006a
O-4	0	33.33±1.76d	2.24±0.26d	6.86±0.09d				0.27±0.010d
	100	555.41±3.24c	45.54±0.33c	159.37±3.13c	5.55±0.03a	0.46±0.003a	1.59±0.03a	0.37±0.006c
	300	722.09±1.75b	53.42±0.51b	267.00±5.25b	2.41±0.01b	0.18±0.00b	0.88±0.02b	0.44±0.009b
	500	925.75±7.01a	62.88±1.82a	379.58±8.61a	1.85±0.02c	0.12±0.003c	0.76±0.02c	0.48±0.010a
O-72	0	31.66±0.66d	1.52±0.41d	6.45±0.12d				0.25±0.010c
	100	582.16±19.91c	43.65±0.76c	136.62±9.04c	5.82±0.20a	0.44±0.007a	1.37±0.09a	0.35±0.031b
	300	761.03±11.96b	50.74±2.41b	251.06±0.75b	2.53±0.04b	0.17±0.007b	0.84±0.01b	0.40±0.006ab
	500	946.29±24.40a	57.76±0.33a	363.13±2.80a	1.89±0.05c	0.12±0.003c	0.73±0.01b	0.45±0.012a
O-795	0	31.33±2.18d	1.51±0.35d	6.38±0.16d				0.25±0.017c
	100	1074.92±46.74c	56.49±1.13c	679.62±2.67c	10.75±0.17a	0.57±0.012a	6.80±0.03a	0.68±0.012a
	300	1551.27±16.02b	62.91±2.27b	770.69±13.61b	5.17±0.05b	0.21±0.006b	2.57±0.04b	0.54±0.017b
	500	1779.41±31.06a	74.56±2.34a	832.40±7.36a	3.57±0.07c	0.15±0.006c	1.66±0.01c	0.51±0.006b
O-9897	0	32.00±2.00d	2.29±0.36d	6.41±0.05d				0.27±0.012c
	100	590.06±7.70c	41.91±0.52c	149.81±5.56c	5.90±0.08a	0.42±0.006a	1.50±0.05a	0.32±0.007b
	300	770.20±5.66b	51.65±0.57b	253.03±4.13b	2.57±0.02b	0.17±0.003b	0.84±0.01b	0.40±0.003a
	500	1074.57±20.64a	56.20±1.08a	354.87±4.93a	2.15±0.04c	0.11±0.003c	0.71±0.01c	0.38±0.102b
VM-1	0	32.00±0.57d	1.65±0.29d	6.76±0.17d				0.26±0.020d
	100	517.87±9.18c	38.64±0.70c	166.82±1.36c	5.18±0.09a	0.39±0.007a	1.67±0.01a	0.39±0.003c
	300	575.24±5.95b	47.93±0.78b	270.06±2.09b	1.92±0.02b	0.16±0.003b	0.90±0.01b	0.55±0.003b
	500	680.51±8.40a	51.83±0.66a	381.90±1.76a	1.36±0.02c	0.10±0.003c	0.76±0.01c	0.63±0.007a
HS-24	0	31.33±0.66d	1.59±0.33d	6.39±0.24d				0.25±0.007c
	100	613.82±13.24c	38.23±1.59c	146.45±4.29c	6.14±0.13a	0.38±0.015a	1.46±0.04a	0.30±0.015b
	300	714.29±15.54b	46.69±1.75b	241.40±15.78b	2.38±0.05b	0.16±0.007b	0.80±0.05b	0.40±0.029a
	500	928.39±14.99a	51.04±1.24a	361.81±1.84a	1.86±0.03c	0.10±0.003c	0.72±0.01b	0.44±0.007a

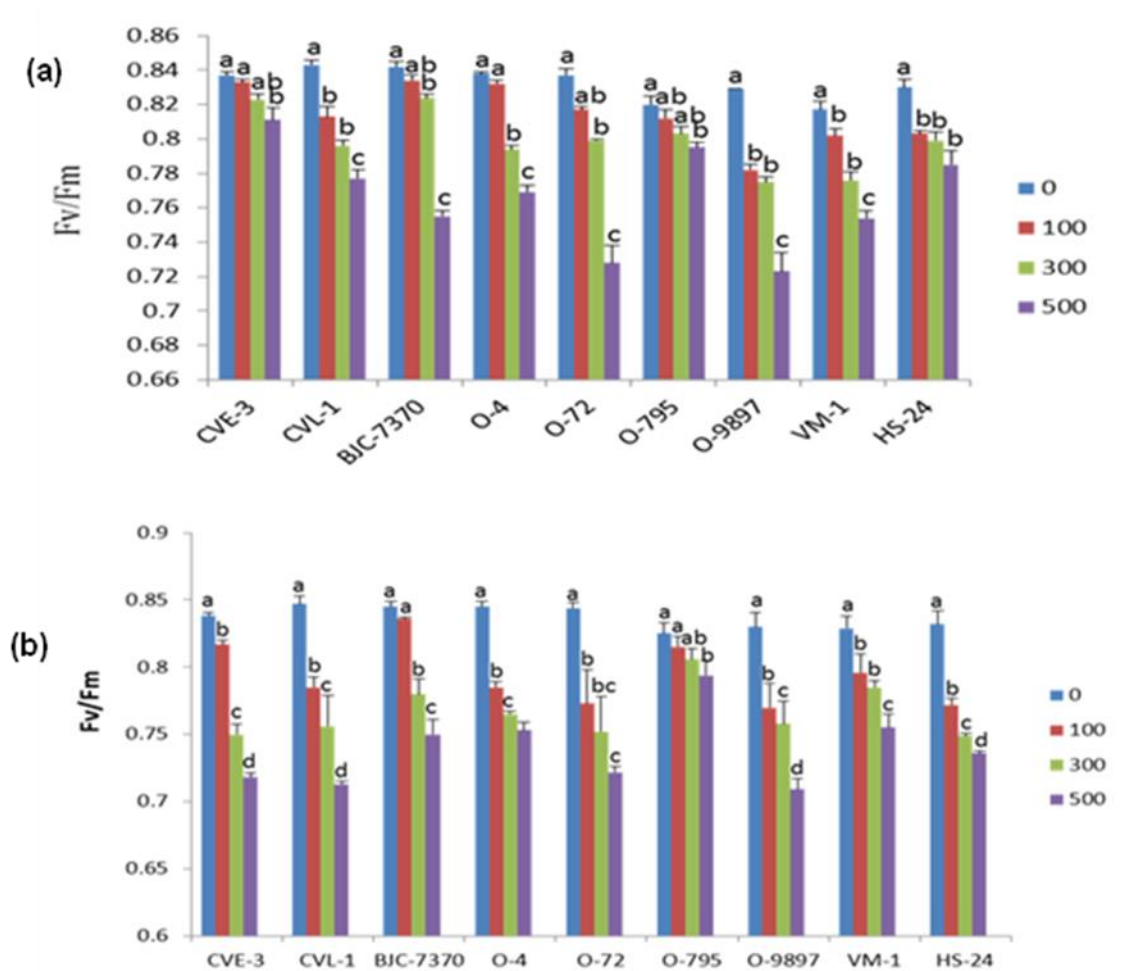


Fig 3. Chlorophyll fluorescence of nine jute varieties subjected to different Cr concentrations. Each value represents the mean \pm standard error (n=3) (a) at 10 days after exposure (b) at 20 days after exposure. Means for each crop followed by the same letter are not significantly different at the level of $p < 0.05$ based on LSD test.

a better indicator to classify the metal accumulating ability of a particular plant because it takes into account the trace element concentration in the substrate (Zayed et al., 1998). Zayed et al. (1998) recommended that a good metal accumulating plant should have the ability to bioconcentrate the element in its tissue to a BCF > 1000 (%). The present result showed that the shoot BCF in all jute varieties were lower than 1000 (%), indicating that these plants were not accumulators. However, at a Cr concentration of 100 ppm, root BCFs of O-795 and CVE-3 exceeded 1000 (%). These results suggest that these jute varieties have a high ability for Cr phytostabilization. We found that the TF of all crops lower than 1. This is consistent with the results reported elsewhere (Shi and Cai, 2009, 2010; Meng et al., 2013). They found eight potential energy crops and *Agropyron cristatum* exposed to Cd and Zn had the TFs lower than 1. These results suggest that jute could be regarded as potential candidate varieties for the phytostabilization of Cr-contaminated soil, which not only improves the environment but also reduces the risk of Cr entering the food chain. Phytoremediation efficiency is determined by the amount of metal transported to the above-ground tissues, and the above-ground biomass of the plant.

Schnoor (1997) suggested that any plant which is useful for phytoremediation should be vigorously growing, easily harvestable and should exhibit a biomass of more than 3 tons dry weight/ha/year. The present study showed that all the tested jute varieties have biomass yield more than 3 tons/ha/year (considering 420,000 plants/ha). The ideal plant used in phytoextraction should be tolerant to high levels of the metal, and accumulate high levels of the metal in its harvestable parts (Salt et al., 1998). Phytoremediation efficiency is determined by the amount of metal transported to the aboveground tissues, and the aboveground biomass of the plant. The results presented here demonstrate that O-795, CVE-3 and BJC-7370 showed high shoot BCF and Cr uptake, as well as high biomass. These crops, therefore, were more efficient than the others for the remediation of Cr contaminated soils. Our study suggests that theoretically, about 28 kg of Cr can be removed from one ha area contaminated with about 300 ppm in each crop cycle. Nevertheless, actual amount of Cr removal will vary depending on many factors. Phytoremediation process is quite slow and usually takes several years, or even decades, to halve the levels of heavy metal completely removed from contaminated soil (McGrath and Zhao, 2003). However, our

data showed that jute is a promising plant for phytoremediation and actual field trial is necessary to calculate the exact rate of Cr removal in a Cr contaminated site.

Materials and Methods

Collection of seed materials

Seeds of different jute varieties were collected from Bangladesh Jute Research Institute (BJRI), Dhaka. The selected varieties used for this study was Tossa Jute; *Corchorus olitorius* (O-4, O-72, O-795, O-9897), White Jute; *Corchorus capsularis* (CVE-3, CVL-1, and BJC-7370), and Mesta Jute; *Hibiscus sabdarifa* (VM-1, HS-24).

Germination experiments

The study was carried out on May 2012 at Gyeongsang National University, South Korea. Seeds were surface sterilized in a 0.5% aqueous solution of sodium hypochlorite for 1 min, rinsed five times with distilled water and then dried on filter papers. Twenty Five seeds were placed in a petri dish (90 mm) containing a thin layer of cotton. Fifteen milliliter of distilled water (control) and different concentration of $K_2Cr_2O_7$ solutions (50, 100, 300, 500 and 700 ppm) were applied to each petri dish. The plates were incubated in a growth chamber at 30 °C/25 °C (light/dark temperatures), with a photoperiod of 16 h light/8 h dark and light intensity of $225 \pm 25 \mu\text{mol/m}^2/\text{s}$ and humidity was kept at 85%. Seed germination was considered with the emergence of radical and counted to 96 h after incubation. All the experiment was performed in triplicate. The germination frequency (GF), germination index (GI) and vigor index (VI) was calculated as follows:

$$GF = (\text{Germinated seeds} / \text{total seeds}) \times 100$$

$$GI = Gt / Dt$$

$$VI = (\text{Mean shoot length} + \text{Mean root length}) \times \text{germination frequency}$$

Where, Gt and Dt are the number of seeds germinated and germination time, respectively.

Growth experiment

The pot experiment was conducted on May 2012 in a greenhouse of Gyeongsang National University campus, Jinju, South Korea (35° 12' 17" N and 28° 07' 13" E). The average temperature during the test period was $25.6 \pm 0.4^\circ\text{C}$ (day) and $22.0 \pm 0.4^\circ\text{C}$ (night), and the relative humidity was $61.5 \pm 1.3\%$ (daytime) and $68.0 \pm 1.9\%$ (night). Uniform seeds were directly sown into pots (30 cm × 25 cm) filled with a mixture of acid-washed sand and perlite (5:4, v/v). Uniform seedlings were allowed to grow in each pot. The pots received natural sunlight and irrigated with drinking water to maintain 60% field capacity. Thirty days after seed sowing, plants were irrigated with drinking water (control) and different concentrations of $K_2Cr_2O_7$ (100, 300 and 500 ppm) solution. Cr solutions were applied to the soil surface at 500 ml/pot/day. At every 7th day, the plants were fertilized with 100 ml of Hoagland's nutrient solution (H2395, Sigma, USA). The pots containing the plants were placed in drip trays which prevent any leachate from being lost as described by Giordani et al. (2005).

Gas exchange measurement

Three mature leaves were selected for the gas exchange measurement. The measurement was carried out after 10 and 20 days of treatment. Net photosynthetic rate (Pn), stomatal conductance (Gs), transpiration rate (E) and intercellular CO_2 concentration (C_i) were determined using a portable photosynthetic system (LiCor-6400, Nebraska, USA) with an attached LED light source (6400-02B). The measurements were carried out between 10:00 am to 12:00 pm.

Chlorophyll fluorescence measurement

The plant leaves were analyzed for photosynthetic activity by monitoring chlorophyll fluorescence after 10 and 20 days of treatment (Mini PAM (Walz, Effeltrich, Germany). The leaves were dark-adapted for 30 min and then subjected to a 1 second pulse of red light. The minimum chlorophyll fluorescence (F_0) after dark adaptation and maximum fluorescence (F_m) after the pulse of red light were measured. F_v/F_m (the ratio of variable to maximal fluorescence, which is a measure of the quantum yield of photosystem II photochemistry) values were obtained based on these measurements.

Morphological data

At the end of the experiment (31st days after Cr treatment), plants were collected for their weight estimation and accumulation of Cr. Roots were washed carefully with 10 mM Na_2EDTA , and then washed with distilled water to eliminate any residual salt from the surface. Plants were separated into roots and tops (stems and leaves), dried at the 105 °C for 2 h and subsequently at the 70 °C for 48 h in an oven, until they reached a constant weight. The root and stem length, root, stem and leaf weight were calculate and total biomass were measured. The tolerance index (TI) was expressed on basis of total biomass and calculated by using the following formula (Wilkins, 1978).

$$TI (\%) = 100 \times (\text{Growth parameters}_{Cr \text{ treated}}) / (\text{Growth parameters}_{control}).$$

Estimation of Cr accumulation

The dried root, leaves and stem tissues were ground into powder using a blender (Wonder blender, Osaka chemical Co. Ltd. Japan). The 0.5 gram of each sample was digested with HNO_3-HClO_4 (3:1, v/v). The Cr contents in leaves, stems and roots were determined by inductively coupled plasma-atomic emission spectroscopy (ICP-AES). The translocation factor (TF) of Cr from root to shoot and bioconcentration factor (BCF) were calculated as described by Ali et al. (2002):

$$TF = Cr_{shoot} / Cr_{root}$$

$$BCF = Cr_{shoot \text{ or } root} / Cr_{soil}$$

Statistical analysis

The experiment consisted of pots in a randomized complete block design with three replications. The results of the germination, growth parameters and accumulation of heavy metals were statistically analyzed by using analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) to determine significant differences among group means. Statistical analyses were performed using SPSS Version 11.5 software (SPSS Inc., USA).

The data were subjected to ANOVA, and differences between means were determined using the least squares deviation (LSD) test. The P value was 0.05. The other calculation was performed by using Microsoft excel 2010.

Conclusion

This study was conducted to screen plant growth on an artificial contaminated soil to determine their germination ability, tolerance index, and their potential for Cr metal accumulation. Among the nine jute varieties, O-795, CVE-3 and BJC-7370 were higher Cr tolerant than others for germination experiment. In the case of vegetative growth, all tested crops were tolerant at 0-500 ppm Cr stress. O-795, CVE-3, and BJC-7370, showed a minor reduction in plant growth and photosynthetic activities than the others. However, O-795, CVE-3 and BJC-7370 showed higher Cr concentration in root, stem, leaf, higher bio-concentration factor and higher total Cr uptake as well as higher biomass, suggesting that these crops can be good candidates for phytoremediation of Cr contaminated soil. Future, studies will focus on the use of O-795, CVE-3, and BJC-7370 for assessing their capability for phytoremediation of Cr contaminated field.

Acknowledgment

The authors are thankful to the Bangladesh Jute Research Institute for supplying seeds for this research. M.K. Islam was a beneficiary of a doctoral scholarship from the Gyeongsang National University, Republic of Korea.

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