Phytoremediation of chromium (Cr) using *Typha angustifolia* L., *Canna indica* L. and *Hydrocotyle umbellata* L. in surface flow system of constructed wetland

T Taufikurahman*, M A S Pradisa, S G Amalia, and G E M Hutahaean

Bioengineering study program, School of Life Sciences and Technology, ITB, Jl. Let. Jend. Purn. Dr. (HC) Mashudi, Nomor 1 / Jalan Raya Jatinangor KM 20.75 Sumedang-Jawa Barat 45363, Indonesia

*Email: taufik@sith.itb.ac.id

Abstract. Leather tannery industry discharged wastewater containing chromium to the nearby river. The presence of chromium in excess of the limit pollutes the environment and could harm animal and human health. In this study, we examined wetland plants i.e. *Typha angustifolia* L., *Canna indica* L. and *Hydrocotyle umbellata* L. for their effectiveness in remediating chromium in an artificial wetland system within nine days period. Chromium compound, $K_2Cr_2O_7$, was used with a variation in concentration of 0, 10, 30 and 50 mg/L. All three plants survived until the 9th day. The plant with the highest relative growth was *C. indica*, whereas plants which showed highest chromium accumulation and bioconcentration factor (BCF) was *H. umbellata*. Chromium accumulated more in the root than in the shoot (TF <1). Plant with highest efficiency in reducing chromium in medium was *T. angustifolia* (99.78%) grown in 50 mg/L chromium. The efficiency of *C. indica* and *H. umbellata* were 99.67% and 86.36%, respectively. The three plants showed a good potential of phytoremediation for wastewater containing chromium.

1. Introduction

Leather tannery industry produces wastewater containing chromium. Chromium is a toxic substance, and excess Cr in the body causes skin diseases, respiratory, kidney and liver [1]. In the environment, Cr is often found in valences of 3 and 6. Chromium in trivalent form is less hazardous than hexavalent chromium, but when Cr (III) coincides the oxidant, Cr (III) has same toxic properties with Cr (VI) [2]. The hexavalent chromium is more soluble and mobile in water. The substance is more dangerous because of its high toxicity, its mutagenic nature and can cause carcinogenic effects to living organisms including microorganisms [3].

Heavy metals are generally indigestible (not biodegradable) and accumulates in living organisms and the environment. Various physical and chemical methods have been used in the removal of heavy metal ions [4]. Another method that can be used to remediate chromium-bearing wastes is phytoremediation, using live plants as a means of restoring contaminants in water. The use of artificial wetlands in phytoremediation methods is known to be less expensive than other methods [5]. In this experiment we used three terrestrial plants i.e. *Typha angustifolia*, *Canna indica* and *Hydrocotyle umbellata* to remediate water contaminated with chromium.
2. Materials and Methods

The plants used in this study were *T. angustifolia*, *C. indica* and *H. umbellata* with an average weight of 160 g, 100 g and 20 g, respectively. Plant media used was soil. The media used is andosol soil (average pH: 5.4; C-organic concentration: 1.24%-22.46%). Chromium used was in the form of K$_2$Cr$_2$O$_7$ salt dissolved in distilled water. The concentration used were 10, 30 and 50 mg/L. The plants were harvested on the 3rd, 6th and 9th day. Plant samples were cleaned and all parts of the organs (root, stems and leaves or shoot) were separated. The samples were dried in an oven at a 105°C. Measured parameters were relative growth based on fresh plant weight, and chromium concentration in plants and media. We calculated bioconcentration and translocation factors for each plant. Plant and water samples were processed by wet ashing method using 30% HNO$_3$ and H$_2$O$_2$. The chromium concentrations were analyzed using an atomic absorption spectrophotometer. The relative growth (RG) was calculated using equation (1) [5]. The efficiency of decreasing chromium concentration was calculated using equation (2), and bioconcentration and translocation factor values was calculated using equations (3) and (4) [7].

$$\text{Relative Growth} = \frac{\text{Final fresh weight (g)}}{\text{Initial fresh weight (g)}}$$  \hspace{1cm} (1)

$$\text{Plant Efficiency} = \frac{[\text{metal}]_{\text{initial}} - [\text{metal}]_{\text{final}}}{[\text{metal}]_{\text{initial}}}$$  \hspace{1cm} (2)

$$\text{Bioconcentration Factor} = \frac{[\text{metal}]_{\text{plant tissue}}}{[\text{metal}]_{\text{medium}}}$$  \hspace{1cm} (3)

$$\text{Translocation Factor} = \frac{[\text{metal}]_{\text{shoot}}}{[\text{metal}]_{\text{root}}}$$  \hspace{1cm} (4)

3. Results and discussion

3.1. Relative growth (RG)

Relative growth represents the ability of plants to grow and survive under stress conditions due to metals exposure [8]. It is known that chromium can cause leaf chlorosis, and inhibit the growth of biomass [9]. Thus, the presence of chromium is expected to inhibit the growth of plants. The relative growth of *T. angustifolia*, *C. indica* and *H. umbellata* is shown in figure 1. The highest relative growth value occurred in *C. indica* with a value of 1.309 ± 0.003, while the lowest value in *T. angustifolia* occurred with a value of 0.760 ± 0.055. The relative growth value of less than 1 indicates that the plant had reduced biomass at the end of the experiment. The result of the analysis of variance showed that the different concentration of chromium exposure had a significant effect on the relative growth of *T. angustifolia*, *H. umbellata* and *C. indica*. 
Figure 1. Relative growth of *T. angustifolia* L. (b) *C. indica* L. (c) *H. umbellata* after 9 days exposure to chromium.

3.2. Chromium concentrations
Chromium concentrations in *T. angustifolia*, *C. indica* and *H. umbellata* is shown in figure 2. The highest chromium concentrations in each plant were 0.143 mg/g for *T. angustifolia*, 0.542 mg/g for *C. indica*, and 1.895 mg/g for *H. umbellata*. *H. umbellata* accumulated the highest chromium concentration compared to *T. angustifolia* and *C. indica*. This could be related to the smaller biomass of *H. umbellata* compared to the other two plants. The translocation of chromium into plants progresses slowly [10], so that in plants with larger biomass, the chromium is not fully distributed in plant parts. Chromium essentially cannot be decomposed and therefore tend to accumulate either in living organisms or in the environment. Heavy metals transported by plants are generally stored in cell compartments such as vacuoles and lignocellulosic materials (cell walls). The chromium concentration accumulated in *H. umbellata* and *C. indica* showed a significant difference.
Figure 2. Chromium concentration in (a) *T. angustifolia* L. (b) *C. indica* L. (c) *H. umbellata* L. after 9 days exposure to chromium.

The accumulation of chromium in the root and shoot of each plant (*T. angustifolia*, *C. indica* and *H. umbellata*) is shown in figure 3. In general, the accumulation of chromium in the root of the plant is greater than the accumulation of chromium in the shoot. For *H. umbellate*, chromium concentration in root five time higher than in its root for treatment 50 mg/L. Chromium accumulates more in the root probably due to the translocation process of chromium from the roots to the shoot was very slow. The chromium is stored in the vacuole cells at the root of the plant to reduce the toxicity of the chromium, and reducing the mobility of the chromium and making the chromium translocation from the root to the shoot not optimum [10]. High concentration of chromium in root also showed plant ability to accumulate chromium and this may play a role in plant mechanism in protecting the whole plant i.e. by isolating chromium accumulation in the root will prevent chromium from damaging other parts of the plant.
Figure 3. Chromium concentration in root and shoot after 9 days exposure to chromium (a) *T. angustifolia* L. (b) *C. indica* L. (c) *H. umbellata* L.
The efficiency of chromium reduction by *T. angustifolia*, *C. indica* and *H. umbellata* is shown in figure 4. The plant roots have the ability to absorb heavy metals in large quantities [11]. One of the phytoremediation techniques, rhizofiltration, uses the roots of plants to absorb and precipitate metal contaminants from surface water and groundwater [12][13]. In general, the decrease in chromium concentration occurs as exposure time increased.

![Graphs showing chromium removal efficiency](attachment:graph.png)

**Figure 4.** Chromium removal efficiency of (a) *T. angustifolia* L. (b) *C. indica* L. (c) *H. umbellata* L. after 9 days of exposure to chromium.

Some chromium in water will be sedimented in the soil so that the chromium concentration in the soil medium increases as the duration of the exposure increased. The highest concentration of chromium in soil was 0.066 mg/g in *T. angustifolia*, 0.051 mg/g in *C. indica* and 0.124 mg/g in *H. umbellata* in chromium concentration of 50 mg/L on day 9. The concentration of chromium in the soil media is shown in figure 5.
Figure 5. Chromium concentration in soil after 9 days exposure to chromium, using (a) *T. angustifolia* L. (b) *C. indica* L. (c) *H. umbellata* L. as phytoremediator.

Bioconcentration factor (BCF) and translocation factor (TF) values of *T. angustifolia*, *C. indica* and *H. umbellata* are shown in table 1. BCF and TF are parameters that are used to determine the ability of plants to accumulate heavy metals. Bioconcentration factors are important parameters for determining the potential of plants to accumulate metals, the values are based on dry weight [14]. BCF value > 1, means that the plant has the potential as a phytoremediation agent of heavy metals chromium. The translocation factor shows the percentage of metals absorbed in the shoot with metals absorbed in the root of the plant [15]. TF value <1, means that more metals accumulate in the root than in the shoot.
The highest BCF was found in *H. umbellate* (up to 53.512), followed by *C. indica* (up to 13.325), and the least is in *T. angustifolia* (up to 9.248). However *H. umbellate* showed lowest TF, ten times lower than TF of *H. umbellate* and *C. indica*. Low TF in *H. umbellate* while its chromium concentration was the highest in root showed plant’s ability to retain high chromium concentration in its root.

### Table 1. BCF and TF values

| [Cr] in medium (mg/L) | Exposure time (days) | *T. angustifolia* | *C. indica* | *H. umbellata* |
|-----------------------|----------------------|------------------|-------------|----------------|
|                       |                      | BCF TF           | BCF TF      | BCF TF         |
| 10                    | 3                    | 2.968 0.203      | 13.325 0.47 | 29.557 0.053   |
|                       | 6                    | 6.7 0.154        | 8.783 0.415 | 53.512 0.027   |
|                       | 9                    | 9.248 0.166      | 11.677 0.185 | 56.532 0.118   |
| 30                    | 3                    | 2.408 0.15       | 10.139 1.57 | 22.651 0.08    |
|                       | 6                    | 3.162 0.274      | 10.256 0.976 | 29.631 0.057   |
|                       | 9                    | 4.751 0.666      | 10.483 0.439 | 37.147 0.101   |
| 50                    | 3                    | 0.707 2.479      | 7.891 2.186 | 15.632 0.138   |
|                       | 6                    | 1.496 0.881      | 9.153 1.968 | 29.478 0.055   |
|                       | 9                    | 2.571 1.035      | 10.85 1.173 | 37.893 0.191   |

### 4. Conclusion

The three plant species show that all three has potential as a phytoremediation agent of chromium. *T. angustifolia* showed the highest chromium concentration reduction efficiency of 99.783% at 50 mg/L compared to the other two species, which is 99.67% for *C. indica* and 86.367% for *H. umbellata*. *H. umbellate* showed highest BCF since it accumulated chromium in its roots more than 5 times higher than *T. angustifolia* and *C. indica*.

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