

## ZINC AND CHROMIUM REMOVAL MECHANISMS FROM INDUSTRIAL WASTEWATER BY WATER HYACINTH, *EICHHORNIA CRASSIPES* (MART.) SOLMS

GAKWAVU, R.J.<sup>1\*</sup> – SEKOMO, B.C.<sup>2,3</sup> – NHAPI, I.<sup>1,2</sup>

<sup>1</sup>*Department of Civil Engineering, Faculty of Applied Sciences, National University of Rwanda, P.O. Box 117 Huye, Southern Province, Rwanda, (phone: + (250) (0)788 52 34 30)*

<sup>2</sup>*UNESCO-IHE Institute for Water Education, P.O. Box 3015, Delft, the Netherlands*

<sup>3</sup>*Department of Chemistry, Faculty of Sciences, National University of Rwanda, P.O. Box. 117 Huye, Southern Province, Rwanda*

*\*Corresponding author  
e-mail: gakwavurjohn@yahoo.fr*

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**Abstract.** Zinc and chromium are environmental pollutants that are toxic even at very low concentrations. Domestic and industrial wastewater discharges are probably the two most important sources for chromium and zinc in water. In Rwanda, the discharge into natural ecosystems of untreated wastewater containing heavy metals by factories and households is a growing problem. A bench-scale study was therefore conducted from May to October 2007 to investigate the major mechanisms responsible for Cr (VI) and Zn (II) removal from industrial wastewater using water hyacinth. The pH effects, plant relative growth, trace metal remaining in water samples, translocation ability, bioconcentration factor, adsorption, bioaccumulation and uptake mechanisms were investigated. The pH slightly increased from the start time pH = 6.7 (0 hr) to pH = 7.64 to 7.86 (48 hr); but after 48 hours of experiment, the pH decreased due to the saturation of bond sites, resulting in some H<sup>+</sup> being released back into the water. The relative growth significantly decreased ( $P \leq 0.05$ ) from 1, 3 and 6 mg/L in 1 week but it slightly decreased linearly after 1 week with increasing metal concentrations ( $P \leq 0.05$ ). About 56.7 % of Zn (II) was accumulated in petioles, 27.0 % in leaves and 16.3 % in roots. For Cr (VI) 73.7 % was taken up in roots, 14.1 % in petioles and 12.2 % in leaves. It was observed that 17.6 %, 6.1 % and 1.1 % were adsorbed for 1, 3 and 6 mg/L of Zn (II) concentrations, respectively, by water hyacinth plants. For Cr (VI), 9.0 %, 36.4 % and 54.6 % were adsorbed for 1, 3 and 6 mg/L, respectively. The order of translocation ability for Cr (VI) was leaves<petioles<roots in water hyacinth whereas for Zn (II) it was leaves<roots<petioles.

**Keywords:** *Adsorption experiments, Chromium removal, industrial wastewater, metal removal mechanisms, water hyacinth, zinc removal*

### Introduction

Zinc and chromium are environmental pollutants that are toxic even at very low concentrations. The pollution of the biosphere with toxic metals has accelerated dramatically since the beginning of the industrial revolution (Nriego, 1979). The primary sources of this pollution include the burning of fossil fuels, mining and smelting of metals, municipal wastes, fertilizers, pesticides and sewage. Heavy metals are of great concern primarily due to their known toxicity to aquatic life and human health at trace levels (Lesage, 2000). It has been reported that domestic and industrial discharges are probably the two most important anthropogenic sources of metals in aquatic systems (Stephenson *et al.*, 1980). However, the lack of a reliable method to

predict metals distribution in treatment units is a key weakness in determining the fate of metals and their transportation in wastewater treatment processes, and therefore the development of effective pre-treatment guidelines (Memon, 2000). The removal of heavy metals from aqueous solutions has therefore received considerable attention in recent years. However, the practical application of physicochemical technology such as chemical precipitation, membrane filtration and ion exchange is sometimes restricted due to technical or economical constraints. For example, the ion exchange process is very effective but requires expensive adsorbent materials (Lehmann *et al.*, 1999; Volesky, 2001).

The use of low-cost waste materials as adsorbents of dissolved metal ions provides economically viable solutions to this global problem and could be considered an eco-friendly solution (Volesky and Holan, 1995; Mullen *et al.*, 1989). At present, emphasis is given to the utilization of biological adsorbents for the removal and recovery of heavy metal contaminants (Mullen *et al.*, 1989). A research study was therefore conducted in Rwanda, where there are no appropriate systems of heavy metals removal, particularly for zinc and chromium which are some of the main contaminants released from textile and other industries. The development of a system to remove these toxic contaminants was investigated using a pilot scale bench study and conducted from May to October 2007. The aim was to investigate the major mechanisms responsible for Cr (VI) and Zn (II) removal from industrial wastewater using water hyacinth. The water hyacinth plant is rapidly infesting many aquatic ecosystems in the country. However, there are possibilities for using the hyacinth plant in heavy metal removal, considering its reported successes in general wastewater treatment (Mullen *et al.*, 1989). Such a system could be inexpensive, providing Rwanda and other developing countries with an appropriate technology thus contributing to environmental sustainability.

## Materials and methods

### *Water hyacinth collection and preparation*

The pilot-scale experiments were performed using free-floating water hyacinth (*E. Crassipes*) plants that had been collected from a natural wetland area called Nyabugogo located in Kigali City. The plants were transported to the Chemistry Laboratory of the National University of Rwanda, Butare, in big plastic buckets as shown in *Fig. 1(a)*. They were then placed under natural sunlight for several days to let them adapt to the new environment in the laboratory. The plants with similar size, weight and shape were selected, rinsed with distilled water to remove any epiphytes and insect larvae grown on plants and then placed in small buckets for experiments as shown in *Fig. 1(b)*.



**Figure 1.** *Water hyacinth collection and preparation (a) plants in big container, (b) plan view of experimental set up*

All experiments were run in batch mode, using a nutrient solution containing 500 ml of tap water from the valley located at Butare near the Natural Science Centre, 500 ml of wastewater from the Nyabugogo wetland and 20 mg of  $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$  and  $\text{NH}_4\text{Cl}$ , and 40 mg of  $\text{K}_2\text{HPO}_4$ . The total fresh weight of plants in each bucket was measured before the start of each growing time, which was 1, 2 and 4 weeks.

### ***Description of the experimental setup in the laboratory***

The laboratory experimental setup for zinc and chromium removal mechanisms consisted of 12 small buckets as show in *Fig 1(b)*. The three buckets without water hyacinth plants served as controls (blanks) containing 1, 3 and 6 mg/L of zinc and chromium. Nine (9) small buckets with water hyacinth plants were used with 3 buckets each containing 1, 3 and 6 mg/L of zinc and chromium. The buckets were maintained for 1, 2 and 4 weeks which were the experimental periods. All experiments were performed in the laboratory at a constant temperature (25°C). A stock solution (1,000 mg/L) of each metal was prepared in distilled water which was later used as dilution water. Individual plants were initially rinsed with distilled water to remove epiphytes, microbes, and any nutrient that might be transferred and were then placed in 2 L small plastic buckets containing 1 L of solution (0.5 L from Nyabugogo wetland + 0.5 L from Butare valley ). The plants were maintained in water supplemented by the heavy metals by adding the required volume of zinc and chromium stock solutions to obtain final concentrations of 1, 3 and 6 mg/L of Cr ( $\text{K}_2\text{CrO}_4$ ) and Zn ( $\text{ZnCl}_2$ ), respectively.

Distilled water was added in order to compensate for water losses through plant transpiration, sampling and evaporation. Water samples and pH measurements were taken every 60 minutes for the first 6 hours and then one sample per day during 1, 2 and 4 weeks of exposure to the metal solution. All samples were filtered using 0.45  $\mu\text{m}$  cellulose acetate filters (Whatman papers) and acidified with 5 drops of nitric acid prior to the storage of samples. The samples were then analyzed using a Perkin Elmer Atomic Absorption Spectrometer. After each test duration (1, 2 and 4 weeks), the final fresh weight for each water hyacinth plant was taken and the plants were harvested for other analyses. They were separated into petioles, roots and leaves and analyzed for relative growth, metals accumulation, translocation ability, bioconcentration factor (BCF) and adsorption on the outer surface of roots. In addition, the metals remaining in the solution were measured to assess the removal capacity of the water hyacinth plants.

### ***Data analyses***

#### ***a) Relative Growth (RG)***

The relative growth of control and treated plants was calculated using *Equation 1* to assess the effects of zinc and chromium concentrations on water hyacinth plant growth.

$$\text{RG} = \text{FFW}/\text{IFW} \quad (\text{Eq.1})$$

Where *RG* denotes relative growth of water hyacinth plants during experimental period, dimensionless;  
*FFW* denotes final fresh weight in grams of water hyacinth plants taken at the end of each experimental period, and  
*IFW* denotes the initial fresh weight in grams of water hyacinth plants taken before starting experiment.

### b) Bioconcentration Factor (BCF)

The BCF provides an index of the ability of the plant to accumulate the metal with respect to the metal concentration in the substrate. The BCF was calculated using Equation 2. A larger ratio implies better phytoaccumulation capability.

$$BCF = (P/E)_i \quad (\text{Eq.2})$$

Where *i* denotes the heavy metal concerned,

*BCF* is the dimensionless bioconcentration factor,

*P* represents the trace element concentration in plant tissues ( $\text{mgL}^{-1}$ ),

*E* represents the initial concentration in water ( $\text{mg/L}$ ) or in the sediment ( $\text{mg/kg}$  dry wt).

### c) Metal Accumulation

Metals accumulation in plant and water samples were measured. Digestion of samples in this study was performed according to the Standard Methods (APHA/AWWA/WEF, 2005). Plant biomass samples were reduced to dry matter by heating at  $105^\circ\text{C}$  for 24 hours in a hot air oven and the ash was digested with nitric acid ( $\text{HNO}_3$ ) and hydrogen peroxide ( $\text{H}_2\text{O}_2$ ), filtered through a Whatman® filter paper into a volumetric flask before Atomic Absorption Spectrometer analyses. The following three mechanisms were investigated to differentiate the metal adsorption, bioaccumulation, and translocation by water hyacinth during the experimental period.

### Adsorption

The adsorption consists of metal attachment to the outer surface of the plant. Adsorption was quantified after the plant had been exposed to different concentrations of chromium and zinc in different periods of times (1 week, 2 weeks and 4 weeks). At the end of the experiment, adsorption was determined by placing roots of the water hyacinth plant in nine 100 ml of EDTA- $\text{Na}_2$  3, 24 mmolar respectively for 5, 10, 15, 20, 25, 30, 35, 40 and 45 minutes. This was to remove zinc and chromium trace elements on the outer surface of the roots. The EDTA- $\text{Na}_2$  solutions were then filtered, acidified by 5 drops of nitric acid ( $\text{HNO}_3$ ), and then analyzed by Atomic Absorption Spectrometer (AAS) to determine the zinc and chromium adsorbed by the plants. The most important parameter to consider is the pH (Keith *et al.*, 2006). Generally when the pH decreases, the toxicity of metal ions increases because the proportion of the adsorbed ion on the root system decreases (Kara, 2005).

### Uptake

The uptake process is a mechanism by which metal ions are transported across the cell membrane and can be used in the building of new biomass or stored in vacuoles. This mechanism was assessed at the end of the experimental period when water hyacinth plants were taken out from the small buckets and roots, petioles and leaves were separated, dried in an oven at  $105^\circ\text{C}$  for 24 hours. The plant samples were transformed into ash, digested and analyzed by AAS to determine the zinc and chromium concentrations in plant biomass (roots, leaves and petioles). The results from

AAS analyses show the plant parts which have higher metal accumulations. The presence of carboxyl groups at the roots system induces a significant cation exchange capacity and this could be the mechanism of moving heavy metal in the roots system where active absorption takes place.

### ***Translocation ability (TA)***

The translocation ability was calculated by dividing the concentration of a trace element accumulated in the root tissues by that accumulated in shoot tissues (Volesky and Holan, 1995). *TA* was calculated using *Equation 5*.

$$TA = (Ar/As)_i \quad (\text{Eq.3})$$

Where *i* denotes the heavy metal concerned,

*TA* is the translocation ability and is dimensionless,

*Ar* represents the amount of trace element accumulated in the roots (mgL<sup>-1</sup> dw),  
and

*As* represents the amount of trace element accumulated in the shoots (mgL<sup>-1</sup> dw)

### ***Statistical analysis***

In order to detect quantitative differences in the data, statistical analyses were performed.

#### **Standard deviation**

The standard deviation was obtained from the variance by extracting the square root and was expressed in the unit in which the measurements were taken:

$$S = \sqrt{\frac{\sum fd^2}{(n-1)}} \quad (\text{Eq.4})$$

#### **Regression analysis**

Regression is defined as the determination of statistical relationship between two or more variables. In simple regression one variable (defined as independent) is the cause of the behaviour of another one (defined as dependent variable). The correlation coefficient (*r*) is expressed by equation:

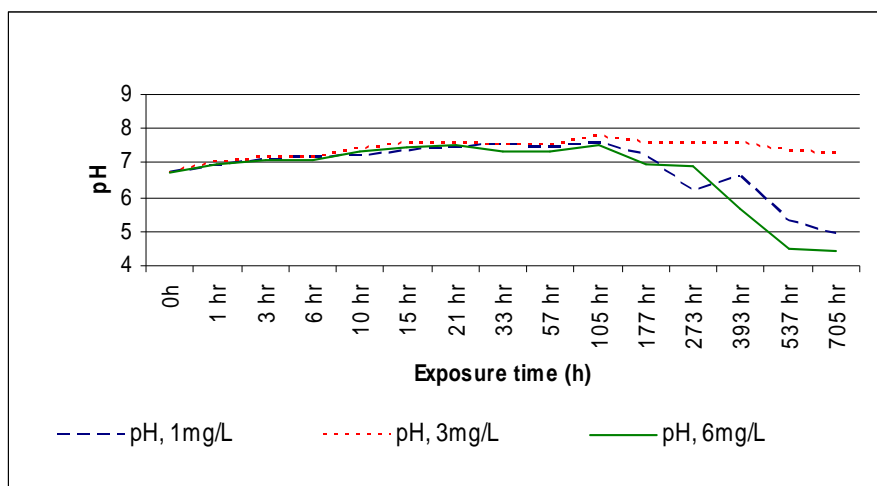
$$r = \frac{\sum dx.dy}{\sqrt{\sum dx^2 \cdot \sum dy^2}} \quad (\text{Eq.5})$$

## **Results and discussions**

### ***Effects of pH variations***

The initial solution pH was adjusted to 6.7 in the small plastic buckets using HCl or NaOH. This is the pH tested in Nyabugogo Wetland from where the hyacinth was collected. The pH variations over time for the water hyacinth-covered containers are

shown in *Figure 3*. The pH varied considerably in different buckets with water hyacinth plants over the exposure time.



**Figure 2.** Results for pH variations in water hyacinth covered containers

Metals will precipitate as hydroxides when the pH of the wastewater is raised to pH 8 to 11 (Barron *et al.*, 1982). As a result, the extent of adsorption was rather low at low pH values. However, in the equilibrium solid phase, Zn (II) and Cr (VI) ion concentrations increased with increasing pH because of increasingly negative charges on the surfaces of the roots at high pH values. This attracted positively charged Zn (II) and Cr (VI) ions more strongly. The ANOVA with replications showed that for 1 mg/L, 3 mg/L and 6 mg/L there was no effect of exposure time but high difference between pH effects and metal remaining in water samples ( $P \leq 0.05$ ). It was observed that the pH effect variations were due to the saturation of binding sites on root systems which affect the pH in water samples with water hyacinth plants by releasing  $H^+$  in water samples.

### Plant relative growth (RG)

The relative growth of water hyacinth plants at different concentrations of zinc and chromium is shown in *Table 1*. It can be seen that the relative growth of plants decreases with the increase of zinc and chromium concentrations.

**Table 1.** Relative changes in growth of plants vs. zinc and chromium concentrations

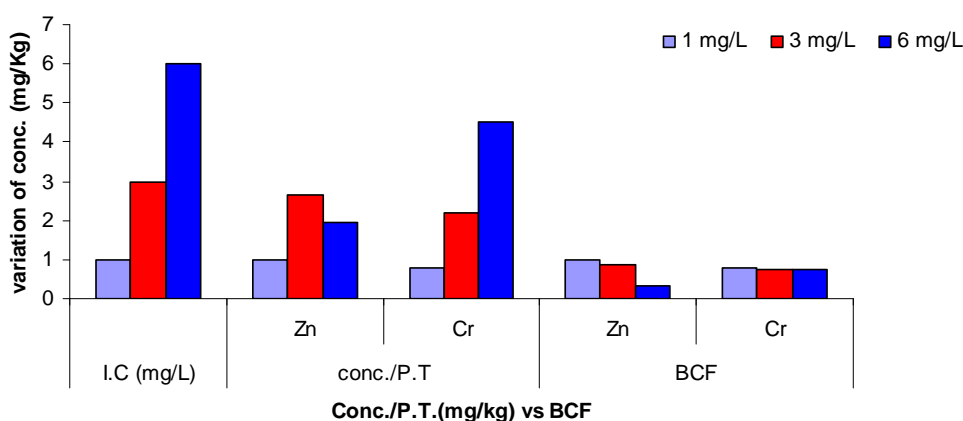
Exposure time (week) with Zn & Cr concentrations	Initial water hyacinth fresh weight (g)	Final water hyacinth fresh weight (g)	Relative growth
1 wk, 1 mg/L	32.33	85.23	2.64
1 wk, 3 mg/L	34.50	96.91	2.81
1 wk, 6 mg/L	26.38	50.07	1.89
2 wks, 1 mg/L	29.75	55.92	1.88
2 wks, 3 mg/L	42.90	80.18	1.87
2 wks, 6 mg/L	16.24	39.96	2.27
4 wks, 1 mg/L	41.15	85.57	2.08
4 wks, 3 mg/L	39.65	98.05	2.47
4 wks, 6 mg/L	34.54	96.16	2.78



For water hyacinth plants treated with Zn and Cr, the plant relative growth significantly decreased ( $P \leq 0.05$ ) from 1, 3 and 6 mg/L in 1 week but for 2 and 4 weeks, the relative growth decreased only slightly with increasing ( $P \leq 0.05$ ) metal concentrations. The relative growth exhibited a decreasing trend caused by relative increases in toxicity of chromium and zinc. The analysis of variance showed that for 1 week exposure time, there is a high effect (difference is significant) of initial concentrations (1, 3 and 6 mg/L) to the growth of the plants ( $P > 0.05$ ), but for 2 and 4 weeks related to initial concentrations, the difference is not significant ( $P > 0.05$ ). Xiaomei et al. (2004) reported that the relative growth of water hyacinth decreases when metal concentrations increase, confirming what was observed in this investigation.

### **Bioconcentration Factor (BCF) of zinc and chromium**

The bioconcentration factor (BCF) was calculated as the ratio of the trace element concentration in the plant tissues at harvest to the concentration of the element in the external environment (Zayed, 1998). The pattern of the bioconcentration factor of water hyacinth plants is shown in Fig. 3.



Note: I.C.: initial concentrations (mg/L); Conc./P.T.: concentration in plant tissues (mg/Kg); BCF: Bioconcentration Factor.

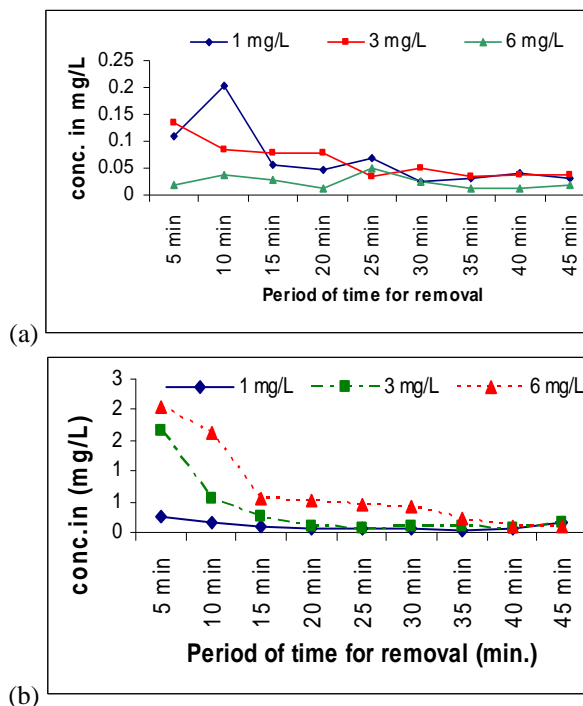
**Figure 3.** BCF for different initial concentrations of Zinc and Chromium

A high competition between zinc and chromium accumulation by the plants was observed due to the kinetics of these metals. The comparison of Zn (II) and Cr (VI) showed that the BCF of zinc was higher than the chromium's BCF for 1 and 3 mg/L, but very low for 6 mg/L for zinc. The plant accumulated more for low initial concentrations than for high ones. It was observed that there is no significant difference both for zinc and chromium when comparing initial concentrations to the concentrations in plant tissues and bioconcentration factors for zinc and chromium ( $P > 0.05$ ).

### **Adsorption**

Figure 4 shows the adsorption experimental results for zinc and chromium. The removal was highest for the first 15 minutes and the highest chromium concentrations observed for 6 mg/L was around 2 mg/L, for 3 mg/L was around 1.6 mg/L and for 1 mg/L was around 0.3 mg/L. The adsorption capacity of water hyacinth plants seemed to be different when zinc and chromium are compared. For zinc, 17.6 % of 1 mg/L was

adsorbed by the water hyacinth plants, 6.1 % of 3 mg/L was adsorbed and the plants adsorbed 1.1 % of 6 mg/L. For chromium, 9.0 % of 1 mg/L, 36.4 % of 3 mg/L and 54.6 % of 6 mg/L were adsorbed by the water hyacinth plants. Meggo(2001) has confirmed that aquatic plants are able to accumulate metals at low concentrations.



**Figure 4.** Desorption over time for various initial concentrations of (a) and (b) chromium

### **Uptake for zinc and chromium**

It was observed that 56.7 % of zinc was accumulated in petioles, 27.0 % in leaves and 16.3 % in roots. The analysis demonstrated no significant difference for different initial concentration and exposure time ( $p > 0.05$ ) in uptake mechanisms of zinc. There was a significant difference observed in plant parts in uptake processes ( $p \leq 0.05$ ). For chromium, 73.7 % was taken up in roots, 14.1 % in petioles and 12.2% in leaves, demonstrating the preference of plants to store chromium in roots. The trend was the same for zinc; no significant difference existed between plant parts ( $p > 0.05$ ) and also between initial concentrations in uptake processes ( $p > 0.05$ ).

### **Translocation ability (TA)**

The quantities of trace elements accumulated in the petioles exceeded those in the shoots. Roots of water hyacinth accumulated about 3 to 15 times more trace elements than did the shoots. It appears that chromium translocation is comparable to zinc as shown in *Table 2*. The ability of plants to translocate trace elements of chromium increased for roots/leaves (5.3 times for 1 mg/L, 6.5 times for 3 mg/L and 6 times for 6 mg/L). The number of times for roots/petioles decreases (4 times for 1 mg/L, 4 times for 3 mg/L and 7 times for 6 mg/L) because the order of storage was leaves<petioles<roots.



**Table 2.** TA of (a) zinc and (b) chromium

<b>(a) Initial conc. of Zn (II)</b>				
	<b>Roots/ Shoots</b>	<b>1 mg/L</b>	<b>3 mg/L</b>	<b>6 mg/L</b>
1 week	Roots/petioles	0.382 <sup>a</sup>	0.255 <sup>a</sup>	0.383 <sup>a</sup>
	Roots/leaves	1.114 <sup>a</sup>	0.732 <sup>a</sup>	0.164 <sup>a</sup>
2 weeks	Roots/petioles	0.478 <sup>a</sup>	0.485 <sup>a</sup>	0.439 <sup>a</sup>
	Roots/leaves	0.461 <sup>a</sup>	0.564 <sup>a</sup>	0.993 <sup>a</sup>
4 weeks	Roots/petioles	0.171 <sup>a</sup>	0.510 <sup>a</sup>	0.109 <sup>a</sup>
	Roots/leaves	0.241 <sup>a</sup>	1.041 <sup>a</sup>	0.255 <sup>a</sup>

<b>(b) Initial conc. of Cr (VI)</b>				
	<b>Roots/ Shoots</b>	<b>1 mg/L</b>	<b>3 mg/L</b>	<b>6 mg/L</b>
1 week	roots/petioles	4.104a	3.663a	6.831a
	roots/leaves	5.288a	6.487a	5.965a
2 weeks	Roots/petioles	2.012a	2.254a	5.870a
	Roots/leaves	2.365a	3.851a	4.564a
4 weeks	Roots/petioles	1.235a	3.258a	1.239a
	Roots/leaves	2.578a	3.851a	1.008a

An ANOVA was performed on the variability in translocation ability. It was observed that the difference between metal concentrations is not significant ( $p > 0.05$ ) and there was no significant difference between roots and shoots translocation ( $p > 0.05$ ). Stratford *et al.* (1984) found that metal accumulations in water hyacinth increased linearly with solution metal concentration in order *leaves*<*petioles*<*roots*. In this research, the following order was observed: *leaves*<*roots*<*petioles*. When the concentration is high, the water hyacinth plant can only accumulate a low concentration in plant cells. This is in agreement with the results of this study in the case of chromium concentration accumulation in water hyacinth plants, where the high concentration was accumulated in roots followed by petioles and then leaves. Water hyacinth materials were burned after all experiences to avoid the contamination.

## Conclusions and recommendations

From the above results, it is concluded that water hyacinth could effectively remove zinc and chromium from industrial wastewater. The removal mechanisms of zinc and chromium by water hyacinth plants showed that the plant concentrated a high quantity of metals. The experiments conducted under this study could be up-scaled to include treatment of actual wastewater from various industries. Essential design parameters such as hydraulic retention time needs to be determined so that the technology can be widely used in Rwanda and beyond.

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