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Comparative Efficiency of *Cyperus papyrus* and *Phragmites australis* for Bioaccumulation of Heavy Metals

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ABSTRACT

The present study intended to compare the efficiency of the wetland plants *Phragmites australis* (cav.) Trin. ex Steudel and *Cyperus papyrus* L. for the accumulation of heavy metals. Maximum accumulation of the heavy metals Pb (132.5-175 µg/g DW), Zn (97.5-100 µg/g DW) and Cr (80.5-90 µg/g DW) was recorded in roots of the two wetland plants *C. papyrus* and *P. australis*, respectively. Cadmium showed the lowest metal accumulation in the tissues of both plants (0.55-5.5 µg/g DW). *C. papyrus* roots accumulated higher levels of most metals than those of *P. australis*. Heavy metals were shown to be not only accumulated in roots of both plants but also were translocated to the shoots and accumulated in the harvestable plant parts. The results showed variation in the levels of the heavy metals accumulated in the different parts of *C. papyrus* and *P. australis*, in spite of being planted at the same site and subjected to the same conditions. The potential use of *Phragmites australis* and *Cyperus papyrus* in phytoremediation is also discussed.
Key words: Phytoremediation, *Cyperus papyrus*, *Phragmites australis*, common reed, heavy metals.

INTRODUCTION

Constructed wetlands are inexpensive systems for wastewater treatment (Butler and Dewedar, 1991; Dewedar *et al*., 2005). These wetlands are used not only to degrade organic pollutants and nutrients from domestic sewage and agricultural runoff, but also to remove metals from domestic, agricultural and industrial wastewater (Obarska-Pempkowiak and Klimkowska, 1999; Cheng *et al*., 2002). Recently, there has been much interest in the use of constructed wetlands for the removal of heavy metals from contaminated soils, sediments and water (Batty and Younger, 2004; Deng *et al*., 2004; Bragato *et al*., 2006). Plants play an important role in constructed wetland for the removal of pollutants (Deng *et al*., 2004). They not only take up nutrients, but are also able to absorb and accumulate metals (Rashad, 2005; Mant *et al*., 2006). Plants usually remove metals via filtration, absorption, cation exchange and through plant-induced chemical changes in the rhizosphere (Maine *et al*., 2006). Wetland plants such as *Phragmites australis*, *Typha latifolia* (Laing *et al*, 2006; Maine *et al*., 2006) and some *Cyperus* species (Deng *et al*., 2004) can accumulate heavy metals in their tissues. *Phragmites australis* and *Typha latifolia* have been successfully used for the phytoremediation for Pb/Zn mine tailings under waterlogged conditions (Peltier *et al*., 2003; Peterson and Teal, 1996).

Metal accumulation by wetland plants is affected by many factors: variations in plant species, growth stage of the plants and element characteristics control absorption, accumulation and translocation of metals (Groudeva *et al.,* 2001). Metal concentration, pH and nutrient status in substrata are also physical factors that affect metal availability to plants (Scholes *et al*., 1998). Furthermore, physiological adaptations also control heavy metal accumulation by sequestering metals in the roots (Bragato *et al*., 2006). Information about the abilities of different wetland plant species or tissues to absorb and transport metals under different conditions will provide insight for choosing appropriate plants for wetland phytoremediation systems.

Pollution with heavy metal in water bodies (both surface water and groundwater) is a serious environmental problem, threatening not only the aquatic ecosystems, but also human health, through contamination of drinking water. Unlike organic pollutants, heavy metals are not degraded through biological processes (Peterson and Teal, 1996).

The present study aims to determine the ability of two wetland plant species, *Cyperus papyrus* and *Phragmites australis*, to tolerate cadmium (Cd), copper (Cu), nickel (Ni), cobalt (Co), zinc (Zn), and lead (Pb) in root tissues, and to investigate the transportation capacity of various heavy metals tested to the shoot systems of the two plants. The potential use of wetland plants for heavy metal removal from wastewater in constructed wetlands is also discussed.

MATERIALS AND METHODS

Constructed wetland

A biological wastewater treatment system was established at Abu-Attwa experimental station, Ismailia, Egypt in 1998 for treatment of the primary treated domestic wastewater collected from Ismailia city. Primary treatment at Abu-Attwa experimental station is usually achieved through sedimentation in a large lagoon for 24-48 hours with continuous stirring.

The constructed wetland system consists of six-parallel treatment beds. Each bed is 20 m length, 2.5 m width and 1.0 m depth, and filled with gravel and/or sand. Two emergent-rooted wetland plants,

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papyrus *Cyperus papyrus* (Cyperaceae) and common reed *Phragmites australis* (Gramineae) were planted at a density of 1 plant/m^2 in the treatment beds. Both plants were propagated through rhizomes. Two treatment beds were planted with papyrus, while the other four treatment beds were planted with common reed. Aboveground shoot systems of both plants produced very dense mass in a short time. Also, plant rhizomes increased in size and produced very thick underground root system in few weeks.

Water samples

Six water samples were seasonally collected from the system during the period from June 2000 to May 2001. The samples represented the primary treated wastewater (influent) and five samples form treated beds (effluents). Water samples were collected in two replicates of clean, wide-mouthed, plastic bottles. One bottle was used for physicochemical analyses in which turbulence was carefully avoided. The second bottle was used for microbiological analyses. Samples were stored in an ice box while being transported to the laboratory.

Physicochemical analyses

The water quality of both influents and effluents of the treatment beds was monitored through determination of various physicochemical parameters according to the Standard Methods for the Examination of Water and Wastewater by the American Public Health Association (APHA, 1998). The flow rate of the influent feeding the treatment beds was regularly measured and manually adjusted to 10 L/min to achieve a final volume of 9.6 m^3/day (16 h flow/day).

Microbiological analyses

Microbiological analyses were carried out according to the Standard Methods for the Examination of Water and Wastewater (APHA, 1998). Total viable bacteria (TVB), total coliforms (TC), fecal coliforms (FC) and fecal enterococci (FE) were determined using the pour plate method. Suitable dilutions of the water samples were used. Triplicate plates were used for each dilution. Plates giving 30-300 cfu/ml were selected to count colonies.

Plant samples

The sampling was carried out at the late growing season in May 2001. Six shoots of the two wetland plant species *Cyperus papyrus* and *Phragmites australis* were randomly collected from plants near the outlet of the treatment beds. The shoots were cut off 3 cm above ground level, gently cleaned with paper towels and then quickly transferred in plastic bags to the laboratory for analyses. Fresh and dry (80°C for 48h) weights were determined for each sample.

Heavy metal analysis

Collected plant samples were dried at 105°C. Plant samples (1 g roots, stems or leaves) were then digested in 10 cm^3 concentrated nitric acid in a warm (60°C) water bath for 2 h., filtered through a Whatman no. 540 filter paper, made up to volume and then analyzed by the atomic absorption spectrophotometer (APHA, 1998).

Data analysis

Analysis of variance (ANOVA) test was used to evaluate the significance of differences between groups with a level of significance set to $p < 0.05$. When the ANOVA test gave a significant result, a pairwise Tukey's HSD test (Zar, 1984; Lentner and Bishop, 1986) was carried out to evaluate the differences between each pair.

RESULTS

Wastewater characteristics of the primary treated effluent are presented in Tables (1 and 2). The hydrogen ion concentration (pH) of the effluent from the treatment beds is slightly higher than that of the influent. The dissolved oxygen (DO) usually increase in

Table (1): Average physicochemical parameters of constructed wetland before and after treatment for treatment beds.

Parameter	Before treatment (influent) (± SD)	After treatment (effluent) (± SD)	Removal efficiency
pH	7.2 ± 0.11	7.8 ± 0.16	
Dissolved oxygen (mg/l)	0.28 ± 0.15	1.3 ± 0.41	
Biochemical oxygen demand (mg/l)	141.65 ± 3.85	48.28 ± 2.69	65.92 %
Chemical oxygen demand (mg/l)	657.18 ± 6.28	210.84 ± 5.66	67.92 %
Total suspended solids (mg/l)	52.78 ± 3.08	25.78 ± 2.04	51.16%
Total dissolved salts (mg/l)	797.55 ± 8.11	674.8 ± 6.50	15.39 %
Organic matter (mg/l)	42.73 ± 2.17	21.9± 1.37	48.75 %
Ammonia (mg/l)	28.63 ± 1.31	33.24 ± 1.67	
Oxidized nitrogen (mg/l)	0.4 ± 0.14	1.21 ± 0.44	
Total nitrogen (mg/l)	32.83 ± 1.46	28.56 ± 1.97	13.01 %
Total phosphorus (mg/l)	1.1 ± 0.06	1.05 ± 0.08	4.55 %
Calcium (mg/l)	48.9± 2.55	37.74 ± 3.02	
Boron (mg/l)	0.24 ± 0.12	0.20 ± 0.04	
Sodium (mg/l)	4.07 ± 0.02	3.64 ± 0.02	
potassium (mg/l)	2.27 ± 0.03	2.16 ± 0.04	

Table (2): Average bacterial count of constructed wetland before and after treatment for 20 m long treatment beds.

Parameter	Before treatment (influent)	After treatment (effluent)	Removal efficiency
Total viable bacteria (cfu/ml)	400 – 6.1 x 10^3	41.5 – 4.5 x 10^3	81 %
Total coliform (cfu/ml)	20.07 – 349.1 x 10^2	13.54 – 49.92 x 10^2	86 %
Fecal coliform (cfu/ml)	16.07 – 106.7 x 10^2	1.57 – 5.07 x 10^2	82 %
Fecal *Streptococci* (cfu/ml)	5.87 – 52.27 x 10^2	2.51 – 9.33 x 10^2	80 %

Table (3): Heavy metal concentration in the constructed wetlands of Abo-Attwa station, Ismailia, Egypt, concentrations before and after treatments.

Water sample	Heavy metals (µg/g DW)							
	Pb	Cd	Cu	Ni	Zn	Cr	Co	Mn
Before treatment	0.37	0.07	0.21	5.6	0.27	0.23	0.18	0.15
After treatment	0.001	0.04	0.001	0.55	0.07	0.03	0.02	0.02
Removal %	99.73	42.86	99.52	90.18	74.07	86.96	88.89	86.67

the treatment beds. The removal percentages of biochemical oxygen demand (BOD) and chemical oxygen demand (COD) were 65.92% and 67.92%, respectively. Both total suspended solids (TSS) and organic matter were removed by 51.16% and 48.75%. However, 15.39% only of the total dissolved salts (TDS) were removed from the influents. Bacterial indicator levels of the influent and the effluents are presented in Table (2). Removal percentage of bacterial indicators is relatively high; ranging between 80-86 %.

The concentrations of heavy metals in the influent and in the effluent are shown in Table (3). The best percentage removal was recorded for Pb (99.73), Cu (99.52) and Ni (90.81). The lowest removal percentage of metals was recorded for Cd. Removal percentage of Zn, Cr, Co and Mn were relatively high, ranging from 74.1-88.9 %.

Figure 1 show the bioaccumulation levels of the heavy metals Cd, Cu, Ni, Co, Zn, Pb, Cr and Mn in leaves, stems and roots of *Cyperus papyrus* and *Phragmites australis.* Pb and Zn recorded higher level of accumulation by the root system of both *Cyperus papyrus* and *Phragmites australis* in compare to other contaminated heavy metal.

Levels of Cd in all parts of the two plants were low (0.55-5.5 µg/g DW). *Cyperus papyrus* accumulated higher levels of Mn, Cr, Zn, Cu, Cd, and Pb in their root and shoot tissues than *Phragmites australis* (Figures 1 and 2). However, *P. australis* showed high levels of Ni and Co accumulation in root than those of *C. papyrus*.

Heavy metals accumulated by the two wetland plants under study were not only settled in root tissues, but were also translocated to the stems and leaves (Table 4). Most of the heavy metals tested were translocated from the roots to the stems and leaves of both plants (Table 4). The ratio of shoot to root metals indicates internal metal transportation. The rate and extent of translocation rate within the two plants depended on the metal and the plant species concerned. High levels of Cd, Ni, Zn, Cr and Mn accumulated in leaves of *Phragmites australis*. Also Cu, Ni, Cr and Mn accumulated at higher levels in the leaves of *Cyprus papyrus* than stems.

Analysis of variance (ANOVA) followed by the main differences Tukey's test reveals that the concentrations of the various heavy metals in different plant parts of the two wetland plants are significantly different (Table 5). Different levels of heavy metal accumulation were recorded in different organs of both plants.

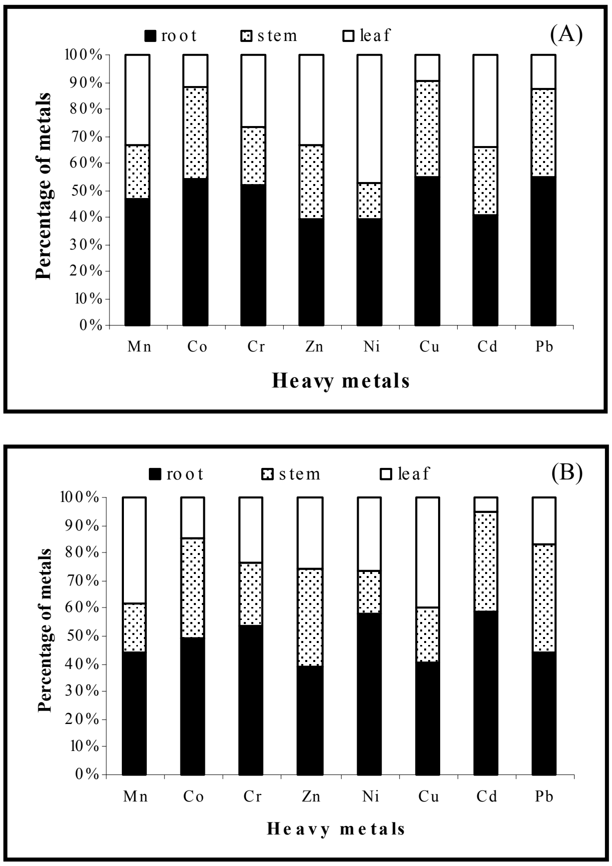

Figure (1): Distribution of various heavy metals in different tissues of (A) *Phragmites auatralis* and (B) *Cyprus papyrus*.

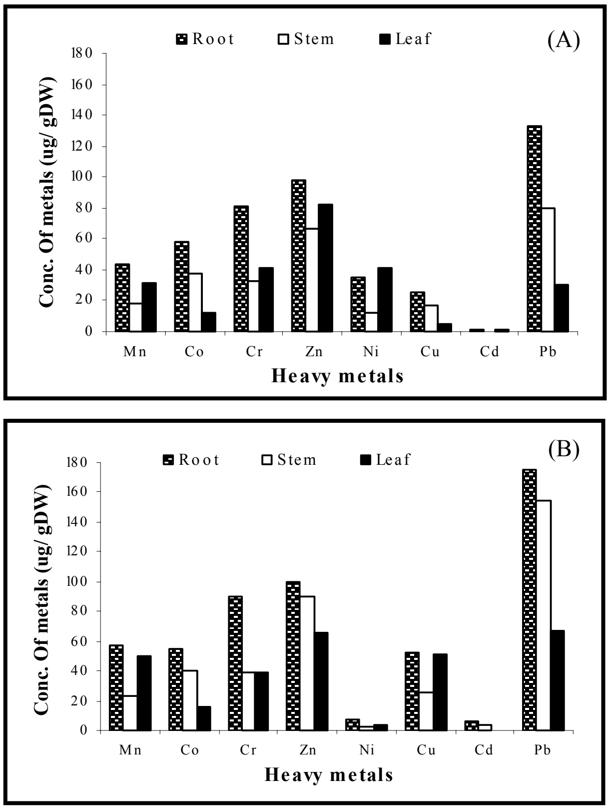

Figure (2): Concentrations of various heavy metals in roots, stems and leaves of (A) *Phragmites auatralis* and (B) *Cyprus papyrus*.

Discussion

Pollution with heavy metal represents an important environmental problem due to their toxicity effect, which is leading to serious ecological and health problems. Soil contaminated with heavy metals not only influence of fauna and flora, but also affect environmental parameters such as pH, oxygen and organic matter (Ansola *et al*., 1995; Laing *et al*., 2006; Maine *et al*., 2006). Generally, heavy metals affect the nervous system, liver and bones and block some vital enzymes (Samecka-Cymerman and Kempers, 2001).

The results presented in this study proof the ability of both *Cyperus papyrus* and *Phragmites australis* to accumulate the heavy metals Cd, Cu, Ni, Co, Zn, Pb, Cr and Mn in their roots, stems and leaves. High levels of most metals were accumulated in the roots of both plants. Levels of these metals were higher in the roots *Cyperus papyrus.* High accumulation levels of heavy metals in root tissues may reflect a strategy for metal tolerance in the root cells. On the other hand, some plant species could accumulate relatively high metal concentrations (far above the toxic concentration of higher plants) in their shoot tissues indicates that internal detoxification metal tolerance mechanism(s) are also included (Weis and Weis, 2004).

The results also suggeste an internal translocation scheme from roots to stems and from stems to leaves of both plants. The translocation ratio from roots to stems showed higher accumulation levels in roots. On the other hand, these ratios from stems to leaves indicate higher accumulation in leaves of both plants. Similar results were reported with other wetland plants for bioaccumulation of approximately similar levels of heavy metals (Deng *et al*., 2004; Weis and Weis, 2004; Bragato *et al*., 2006; Fritioff and Greger, 2006).

Long treatment beds (100 m) are usually excellent for the removal of nutrient and pathogenic bacteria from domestic wastewaters (Butler and Dewedar, 1991). However, short treatment beds (20 m), as those designed in the present study, could remove nutrients at 50-65% and bacteria at 80-86%. Fortunately, short beds are not a problem for the accumulation of heavy metals, as metals are usually accumulate in the tissues of higher plants more and more by time. High removal rates of heavy metals are recorded in the present study with the same constructed beds achieving moderate nutrient removals. Removal efficiency reached 99 % Pb and Cu. Removal percentages for Ni, Cr, Co and Mn were also relatively high (86-90%). Our results could be supported by other studies proving the efficiency of constructed wetland systems for the removal of not only nutrient and pathogenic bacteria (Dewedar *et al*., 2005), but also heavy metals (Ansola *et al*., 1995; Scholes *et al*., 1998; Batty and Younger, 2004; Deng *et al*., 2004; Maine *et al*., 2006).

Metal remediation through common physical and chemical technologies is expensive and unsuitable in case of voluminous effluents containing complex organic matters and metal contamination (Groudeva *et al*., 2001). Alternatively, the degradation or stabilization of contaminants by higher plants is rather safe and effective as an economic alternative to traditional methods of remediation (Cheng *et al*., 2002). Pollutants are taken up by plant roots and then are either sequestered or translocated to stems and leaves (Weis and Weis, 2004). Ultimately, metals are released within the plant to cell vacuoles by conjugation to glutathione or more frequently its derivative phytochelatin (Field and Thurman, 1996). Sequestration of pollutants within plants is the basis for phytoextraction of soils and water contaminated with heavy metals (Raskin *et al*., 1997). Several plant species including *Cyprus papyrus* and *Phragmites australis* have been shown to accumulate high levels of various heavy metals (Deng *et al*., 2004). Plant shoots and roots containing metals are subsequently harvested and treated as hazardous wastes (Peterson and Teal, 1996; Bragato *et al*., 2006) or the metals are recovered as ore recycling of metals from plant residues. Phytoremediation processes usually require relatively long periods of time and often require the disposal of toxic vegetation. In this connection, diverse plant species such as wetland plants (Deng *et al*., 2004), grasses (Nedlkoska and Doran, 2000), trees (Pulford and Watson, 2003) and several other monocots and dicots (Peterson and Teal, 1996) are extremely promising for phytoremediation of heavy metals in

Table (4): Translocation ratios (stems/roots and leaves/stems) in wetland plants grown for two years in the constructed wetlands of Abu-Attwa station, Ismailia, Egypt.

Plant species	Heavy metals (µg/g DW)							
	Pb	Cd	Cu	Ni	Zn	Cr	Co	Mn
				(stem/root)				
Phragmites australis	0.60	0.61	0.64	0.33	0.69	0.40	0.64	0.43
Cyprus papyrus	0.89	0.62	0.49	0.26	0.91	0.43	0.73	0.40
				(leaves/stem)				
Phragmites australis	0.38	1.36	0.28	3.61	1.23	1.28	0.34	1.68
Cyprus papyrus	0.44	0.13	1.99	1.75	0.72	1.01	0.4	2.15

Table (5): One way analysis of variance of metal accumulation by different parts of *Cyperus papyrus* and *Phragmites australis*.

Heavy metals	*Phragmites australis*		*Cyprus papyrus*	
	Mean square	**F value**	**Mean square**	**F value**
Pb	5851.70	5852 ***	9060.83	9061 ***
Cd	8.50	16.99 ***	8.95	10.74 ***
Cu	231.09	231.1 ***	1024.36	1024 ***
Ni	897.13	897.1 ***	811.51	811.5 ***
Zn	1388	1388 ***	3003.60	3004 ***

*** : high significance.

contaminated soils and wastewater. Phytoremediation technologies can be further directed to above or below ground contaminants to remove pollutants from affected areas.

In conclusion, the results of the present work revealed that *Cyprus papyrus* and *Phragmites australis* are promising for accumulating heavy metals. Both plants can be used together in the treatment beds of the constructed wetland systems for efficient removal of heavy metals, as the concentrations of various heavy metals accumulated in wetland plants at the same sites and receives the same levels of pollutants are different.

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مقارنة كفاءة نباتى البوص والبردى فى إستخلاص المعادن الثقيلة

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الملخص العربى

تتناول هذه الدراسة مقارنة كفاءة نباتى البوص والبردى فى استخلاص المعادن الثقيلة من مياه الصرف الصحى. وقد أثبتت النتائج أن أعلى نسبة استخلاص للرصاص (132.5- 175 ميكروجرام/ جرام وزن جاف)، والزنك (97.5- 100 ميكروجرام/ جرام وزن جاف)، والكروم (80.5- 90 ميكروجرام/ جرام وزن جاف) قد تمت من خلال جذور نباتى البوص والبردى. أما الكادميوم فقد استخلصه النباتين بكمية قليلة (0.55- 5.5 ميكروجرام/ جرام وزن جاف). جذور نبات البردى استخلصت كميات أكثر من معظم المعادن الثقيلة بالمقارنة بنبات البوص. لم يتوقف دور النباتين على استخلاص المعادن الثقيلة من الجذور فقط، بل نقل النباتين المعادن الثقيلة المستخلصة من الجذور الى السيقان ومنها الى الأوراق. كما أظهرت النتائج أن هناك تفاوت فى مقدرة الأعضاء النباتية المختلفة للنباتين على إستخلاص المعادن الثقيلة، بالرغم من تواجدهم فى نفس المنطقة وخضوعهم لنفس الظروف. ويعكس ذلك أهمية إستخدام أكثر من نبات للتخلص من المعادن الثقيلة الملوثة لمياه الصرف. ويناقش البحث قيمة نباتى البوص والبرى فى المعالجة البيولوجية للمعادن الثقيلة من مياه الصرف الصحى.