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Laboratory study on the survival of water hyacinth under several conditions of heavy metal concentrations^{\ddagger}

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Abstract

To study the survival and behaviour of water hyacinth [*Eichhornia crassipes* (Mart.) Solms] under varying conditions of heavy metal concentrations, groups of the plants were grown in different media (distilled water, Nile water, wastewater and different concentrations of heavy metals). Simultaneously, blank experiments were carried out for comparison. Visual changes in the plants observed during each experiment were noted. The heavy metal (Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn) concentrations, pH and conductivity of the media were measured before, during and at the termination of experiments. In addition, analyses for heavy metals were carried out on the plant samples after termination of the experiments to determine the effect of different media on metal accumulation by the plants. The results showed that water hyacinth can survive in a mixture of heavy metal concentrations up to 3 mg 1^{-1} and in 100 mg Pb 1^{-1} solution, whereas higher concentrations of metals as mixtures and 100 mg Cd 1^{-1} led to rapid fading of the plants. Water hyacinth exhibited a deprotonation reaction during the uptake of metal ions, which was detected as a result of a decrease in pH of the growth media. The results indicated that water hyacinth plays an outstanding role as a heavy metal decontaminator; in addition, its role as a pollutant by releasing metal ions into the aquatic environment was also noted. Elevated pH and ammonia concentrations, along with the low dissolved oxygen content in the microenvironment around the root hairs, are suggested as the main factors for the rapid wilting of plants growing in Kima drain wastewater.

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1. Introduction

Aquatic plants are known to accumulate metals from their environment (Outridge and Noller, 1991; Ali and Soltan, 1999) and affect metal fluxes through those ecosystems (Jackson et al., 1994; St-Cyr et al., 1994). Water hyacinth [*Eichhornia crassipes* (Mart.) Solms] is highly effective in removing excess nutrients (Desougi, 1984; Vora and Rao, 1988; Goel et al., 1989; Oke and Elmo, 1990; Soltan, 1999), heavy metals (Wolverton and McDonald, 1976; Yahya, 1990; Vesk and Allaway, 1997; Vesk et al., 1999; Ali and Soltan, 1999; Soltan, 1999), toxic metals (Kelley et al., 1999, 2000), minerals and organic chemicals, and herbicides from polluted water (Boyd, 1967; Reay, 1972). Water hyacinth has variable chemical contents (Gopal and Sharma, 1981) which have been related to differences in study site, season, plant parts, and the chemical nature of the habitat (Peters, 1986; Ahmed et al., 1992; Zaranyika and Ndapwadza, 1995; Vesk and Allaway, 1997). Laboratory studies of the water hyacinth have demonstrated the potential use of this species in removing metals

 $^{^{\}star}$ Dedicated to one who gave us everything (science, love and guidance), our Professor, the late R.M. Awadallah (1943–1999).

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Metal	Culture solution	Source	Molecular	Assav	Wt. $\equiv 100 \text{ mg } 1^{-1}$
			weight	(%)	(g)
Cd	$Cd(NO_3)_2 \cdot 4H_2O$	Merck	308.47	99.99	0.2745
Со	$Co(NO_3)_2 \cdot 6H_2O$	Merck	291.03	99.99	0.4939
Cr	$Cr(NO_3)_3 \cdot 9H_2O$	Merck	400.15	99	0.7774
Cu	$Cu(NO_3)_2 \cdot 2.5H_2O$	Merck	232.55	98	0.3695
Mn	$Mn(NO_3)_2.\cdot 4H_2O$	Merck	251.01	98	0.4615
Ni	$Ni(NO_3)_2 \cdot 6H_2O$	Merck	290.81	99.9	0.4959
Pb	$Pb(NO_3)_2$	Merck	331.2	99.99	0.1599
Zn	$Zn(NO_3)_2 \cdot 6H_2O$	Merck	297.47	98	0.4643

Table 1Composition of metal culture solutions

from polluted water and have shown that metal concentrations of the plant and the water column are correlated (Muramoto and Oki, 1983; Lee and Hardy, 1987). The deprotonation reaction of water hyacinth during the uptake of metal ions from aqueous solution, as shown by a decrease in pH, has been explained on the basis of chelation with amino acid, carboxylic and hydroxyl groups (subunits) of macrocyclic molecules, such as ionophores present in the mitochondria of water hyacinth. The process of uptake of chemical species by water hyacinth may take place through the cell membrane via diffusion and osmosis (Haider et al., 1983).

Accumulation of metals in the roots and leaves of water hyacinth has also been shown in field studies in which water hyacinth was used as a biological monitor of metal pollution (Ajmal et al., 1987; Zaranyika and Ndapwadza, 1995). Heavy metals (e.g. Cu and Cd) were found to have accumulated in roots more than petioles and least in leaves (Cooly and Martin, 1979). Carboxylic acids are responsible for binding the intracellular proportion of Eu(III) in the roots (Kelley et al., 2000). Metal accumulation is of interest for basic research into the physiology and ecology of plant survival in flooded conditions and under elevated metal levels (Otte et al., 1989; St-Cyr and Campbell, 1996). Investigations on tropical eutrophic lakes that receive domestic sewage and are infested with water hyacinth show that they support very low species diversity and production rates of phytoplankton. However, the reasons for low phytoplankton variety and production rates during the period of water hyacinth decay are far from clear (Agarkar, 1984; Sandhu, 1986; Sharma 1986, 1990). Water hyacinth litter leaches substantial quantities of inorganic and organic nutrients, particularly nitrogen, phosphorus and carbon, during its initial, as well as later phases of decay (Reddy and Sacco, 1981; Varghese, 1991; Gaur et al., 1992). Sharma et al., (1996) offered an explanation as to why the poor variety and low production rates of phytoplankton in the water hyacinth-infested wetlands are maintained, even during the period of water hyacinth decay. They found that decaying water hyacinth litter releases some toxins, which cause acute toxicity to *Scenedesmus* growth. The characteristics of water hyacinth are prominent and interesting and are useful in investigating its role as a source of water contamination or purification. Therefore, the present study was targeted at studying the effect of different media (distilled water, wastewater and solutions containing different concentrations of heavy metals) on the survival of water hyacinth and to investigate its role in the purification or pollution of water.

2. Experimental

2.1. Sample collection

Plant samples were collected from the river Nile water at Abu El Riesh village (At site, 3×3 m², approx. 1 m deep, 1.5 m from the bank), Aswan city (24° 2' N and 32° 9' E), Egypt in September 1999 and were transferred to the laboratory in polyethylene bags. Plants of similar shape, size (weight of each plant, 200 ± 20 g wet mass) and height (roots, 20-22 cm; aerial parts, 26-27 cm) were selected and washed several times using tap and bidistilled water. Three of these plant samples were chosen as a blank to determine their initial metal ion concentrations before the experiments. The other plant samples were inserted in an upright position in glass boxes of $40 \times 40 \times 40$ cm³ containing 30 1 of media, i.e. Nile water (from the Nile river), distilled water, wastewater (from Kima drain wastewater, Aswan, Egypt), solutions with several metals at different concentrations (1, 3, 5, 7, 10, 50 and 100 mg 1^{-1} of Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn) and individual solutions with 100 mg l^{-1} Pb and 100 mg l^{-1} Cd. The plants were left to grow for 240 h or until complete wilting occurred. The total volume of the solution in each glass box was kept constant by adding deionized water to compensate for water lost through plant transpiration and evaporation.

2.2. Preparation of growth solutions

Metal culture solutions were prepared for each metal as given in Table 1. All the chemicals used were of

Media	Duration of experiment (h)										
	0	24	48	72	96	120	144	168	192	240	
Distilled water	Н	Н	Н	Н	Н	Н	Н	Н	Н	Н	
Nile water	Н	Н	Н	Н	Н	Н	Н	Н	Н	Н	
Wastewater	Н	UP	UB	UD	_	_	_	_	_	_	
Heavy metal solution											
$1 \text{ mg } 1^{-1}$ (mixture)	Н	Н	Н	Н	Н	Н	Н	Н	Н	Н	
$3 \text{ mg } 1^{-1}$ (mixture)	Н	Н	Н	Н	Н	Н	Н	Н	Н	Н	
5 mg l^{-1} (mixture)	Н	Н	Н	Н	UY	UY	UY	UM	UC	_	
7 mg l^{-1} (mixture)	Н	Н	Н	UY	UY	UY	UY	UM	UC	_	
$10 \text{ mg} 1^{-1} \text{ (mixture)}$	Н	Н	UY	UP	UP	UP	UM	UC	-	_	
50 mg 1^{-1} (mixture)	Н	Н	UB	UM	UC	_	_	_	_	_	
$100 \text{ mg} \text{ l}^{-1}$ (mixture)	Н	UB	UM	UC	_	_	_	_	_	_	
$100 \text{ mg } 1^{-1} \text{ Pb}^{2+}$	Н	Н	Н	Н	Н	Н	Н	Н	Н	Н	
$100 \text{ mg} \text{ l}^{-1} \text{ Cd}^{2+}$	Н	Н	UP	UM	UC	_	_	_	_	_	

Table 2 Visual changes observed in water hyacinth growing in different media for the duration of the experiments

H, the plant looked quite healthy with green leaves; UY, the plant looked unhealthy with yellow leaves; UB, the plant looked unhealthy with brown leaves; UP, the plant looked unhealthy with partial wilting; UM, the plant looked unhealthy and almost all leaves were dead; UC, the plant looked unhealthy with complete wilting; and UD, the plant looked unhealthy and deteriorated with a very delicate stolon.

Analar grade (98–99.99%, E. Merck). The solutions were analysed before starting the experiments (0 h) and after 24, 48, 96, 144, 192 and 240 h to determine the change in concentration during the experiments. However, plants were analysed only at the termination of the experiments. The fact that the amount of metals measured at time=0 was often slightly less than the amount dissolved in solution suggests that some metals were adsorbed to the walls of the containers.

2.3. Digestion of plants

After the termination of each experiment, the plants were washed well using tap water and distilled water. All plant samples (including blanks) were then divided into roots and aerial parts, dried to a constant weight in an electric furnace at 105 °C, and the subsamples were ground using an agate mortar and sieved through a nylon sieve to obtain the size fraction <150 μ m. A 1-g portion of each subsample was digested with a concentrated H₂SO₄-HClO₄-HNO₃ acid mixture (1:2:10 by volume). Digestion was continued until the solution became clear. After cooling, 20 ml of 20% HNO₃ was added and the mixture was rewarmed at 80 °C for 20 min. The solution was filtered after cooling through Whatman No. 42 filter paper and made up to 50 ml with deionized water (Allen, 1989).

2.4. Analysis of samples

The pH (pH meter, Orion Research, model SA 520, USA) and conductivity (conductivity meter, Jenway, model 4010, UK) values of the media were measured

before and after inserting the plants into the glass containers and at each measurement for trace elements. Anions and cations released into the media from root tissues were determined using standard procedures (APHA, 1992); HCO_3^- (titrimetry), SO_4^{2-} (turbidimetry), Cl^- (Mohr's method) and Ca^{2+} and Mg²⁺(complexometrically). Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn were determined with a SP 1900 Pye Unicam flame atomic absorption spectrophotometer. Atomic absorption spectroscopic standard solution (1 mg ml^{-1}) for elements Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn was purchased from BDH (England). Working standard solutions with different concentrations of each element were prepared by diluting the stock solution using deionized water. The precision of the measurements was estimated by taking three replicates from the sample, and a blank was included in each batch. USGS-G2 geological standard (Flanagan, 1973) was used as reference material for the determination of metals.

3. Results and discussion

3.1. Visual changes observed in water hyacinth over the duration of experiments

The visual changes observed in water hyacinth growing in different media over the duration of experiments are summarised in Table 2. Water hyacinth plants growing in distilled water and Nile water, as well as 1 and 3 mg 1^{-1} metal solutions and 100 mg 1^{-1} Pb²⁺, solution looked quite healthy, with green leaves and strong stolons during the period under study (240 h). Plants growing in wastewater medium appeared to be unhealthy after 12 h, with partial wilting and brown leaves, and the wilting and brown leaf colour increased with time (48 h). The wilting continued until all plants were deteriorated after 72 h. In 5 and 7 mg l^{-1} metal solutions, the plants looked healthy with green leaves until 72 and 48 h, respectively. A yellow colour then developed on the plant leaves and continued until 144 h. At 168 h, almost all leaves were dead, with complete wilting for plants recorded at 192 h. Water hyacinth plants growing in 10 and 50 m gl^{-1} metal solutions looked healthy with green leaves for 48 h. After 48 h, the plants in the 10 mg l^{-1} solution exhibited yellow leaves, followed by partial wilting until 120 h, and almost all leaves were dead at 168 h. Finally, complete wilting was recorded at 192 h. On other hand, plants in the 50 mg 1^{-1} solution looked unhealthy with brown leaves after 48 h, and almost all leaves were dead (wilting with brown colour) at 72 h. At 96 h, plants in the 50 m gl⁻¹ solution exhibited complete wilting with brown colour. Quick wilting combined with brown leaf colour appeared after 12 h on water hyacinth plants growing in the 100 mg l^{-1} solution (as a mixture), and almost all leaves were dead at 48 h, followed by complete wilting with brown leaf colour at 72 h. The plants of water hyacinth exposed to 100 m gl^{-1} Cd²⁺ solution appeared healthy with green leaf colour for 48 h, but thereafter showed partial wilting. The wilting continued until almost all leaves were dead at 72 h, with complete wilting observed at 96 h. From the above observations, survival of water hyacinths growing in distilled water may be due to high accumulation of nutrients in the roots (Soltan, 1999), which compensated for the lack of nutrients in distilled water. Roots have been reported to contain trace metals in addition to Fe, notably Mn, Cu and Zn (Otte et al., 1989; St-Cyr and Campbell, 1996). Deposition of iron at the root surface does not immobilise trace metals, which are found deep within the root. This demonstrates that uptake is significant and the roots do not just provide a surface for particulate adsorption and microbial growth (Vesk et al., 1999). The visual changes observed in plants growing in metal solutions indicated that the toxicity of metals on the plants increases with increasing metal concentrations. In addition, the toxicity of Cd has a very strong effect on water hyacinth in comparison with Pb. This indicates that the water hyacinth is tolerant to Pb. The quick wilting of water hyacinth exposed to Kima drain wastewater can be related to the wastewater composition (Soltan, 1995; Awadallah et al., 1998). The deficiency of dissolved oxygen and the high ammonia content may be the main reasons for water hyacinth deterioration.

3.2. Concentrations of heavy metals investigated in water hyacinth (blank)

Analysis of water hyacinths showed higher concentrations of heavy metals in the roots than in the aerial

Table 3

Concentrations of heavy metals investigated in water hyacinth (mean of three samples)

Metal	Concentration $(\mu g g^{-1} dry matter)$					
	Root	Aerial part				
Cd	1.5 ± 0.8	0.3 ± 0.15				
Co	36.5 ± 8	16 ± 5				
Cr	100 ± 9	41 ± 6.5				
Cu	142 ± 19	43 ± 7				
Mn	1485 ± 110	615 ± 65				
Ni	97 ± 13	33 ± 7				
Pb	185 ± 13	41 ± 8.5				
Zn	295 ± 10	11 ± 14				

parts of the plants (Table 3). The co-precipitation of metals in the plaques of iron and manganese on the roots is one process that can explain the elevated concentrations found in the roots (Vesk and Allaway, 1997). Plaques of iron and manganese oxyhydroxides often form on waterlogged roots (St-Cyr et al., 1993). Oxidation is believed to be a mechanism for avoiding toxicity of reduced forms of Fe and Mn to roots under flooded conditions (Green and Etherington, 1977). Compared with other metals, root concentrations of Mn were high $(1485 \pm 110 \ \mu g \ g^{-1} \ dry \ matter)$, followed by Zn, Pb, Cu, Cr, Ni, Co and Cd in decreasing order $(295 \pm 10, 185 \pm 13, 142 \pm 19, 100 \pm 9, 97 \pm 13, 36.5 \pm 8,$ 1.5 ± 0.8 µg g⁻¹ dry matter, respectively). The root surface was often covered with a reddish/brown layer, which may include mucilage, particulate such as clays, and a diversity of micro-organisms, including bacteria, protozoans and diatoms (Vesk et al., 1999). High levels of Mn may also be due to its high concentration in the above components. The high bioaccumulation of Zn and Cu in water hyacinth may be due to their role as micronutrients (Etherington, 1974). Low cadmium content in roots $(1.5\pm0.8 \ \mu g \ g^{-1} \ DM)$ leads to minimisation of its uptake rate compared with the other metals. The cadmium uptake rate increases with increasing cadmium content of the roots (Petit et al., 1978). This might reflect the induction of synthesis of compounds such as metallothioneins or phytochelatins, which have a specific binding affinity for heavy metals. The nontoxic effect of Pb to plants (H) exhibited by relative high concentrations of Pb in the roots $(185 \pm 13 \ \mu g)$ g^{-1} DM) may be attributed to the absorbed lead being mostly bound to the cell wall of the plant, thus rendering lead ineffective in acting as a strong metabolic inhibitor (Buddhari et al., 1983). Pb is still regarded as an important ecotoxic element with chronic or latent toxicity effects (Breckle and Kahle, 1992).

Regarding the other metals (Cr, Ni and Co), their moderate concentrations are due to the selective uptake of metal ions (Bannister, 1981). Plants may avoid

Media	Duration of	Concentration ($\mu g g^{-1}$ dry matter)								
	experiment (h)	Cd		Со		Cr		Cu		
		Root	Aerial part	Root	Aerial part	Root	Aerial part	Root	Aerial part	
Distilled water	240	65	5	52	26	135	115	164	109	
Nile water	240	10	2	63	17	160	64	141	53	
Wastewater	96	15	5	64	30	155	95	142	68	
Heavy metal solution										
$1 \text{ mg } 1^{-1}$ (mixture)	240	615	30	1300	37	450	52.5	1750	57	
$3 \text{ mg } 1^{-1}$ (mixture)	240	865	50	1530	49	1950	58	2110	68	
5 mg 1^{-1} (mixture)	192	630	95	1680	169	1550	48	2710	252	
7 mg 1^{-1} (mixture)	192	640	280	1355	570	2300	236	2750	1105	
$10 \text{ mg } 1^{-1}$ (mixture)	168	635	485	2000	800	2500	495	2900	700	
50 mg 1^{-1} (mixture)	96	1010	930	2680	2225	3000	1500	2950	1525	
$100 \text{ mg } 1^{-1} \text{ (mixture)}$	72	620	1200	1915	2475	2150	3000	2800	1900	
100 mg 1 ⁻¹ Pb ²⁺	240	25	15	117	40	165	45	268	89	
$100 \text{ mg } l^{-1} \text{ Cd}^{2+}$	96	2060	325	58	10	140	54	129	47	
		Mn		Ni		Pb		Zn		
Distilled water	240	1950	755	95	40	65	30	445	225	
Nile water	240	1875	785	140	55	90	31	470	180	
Wastewater	96	1945	1290	125	60	105	46	275	230	
Heavy metal solution										
$1 \text{ mg } 1^{-1}$ (mixture)	240	1950	635	1100	107	1800	85	1850	325	
$3 \text{ mg } 1^{-1}$ (mixture)	240	1965	765	1210	121	4900	125	4350	345	
5 mg 1^{-1} (mixture)	192	2110	1230	960	260	3800	90	3850	1000	
7 mg 1^{-1} (mixture)	192	1965	1155	900	560	3850	155	3850	2800	
$10 \text{ mg } 1^{-1}$ (mixture)	168	1990	915	1090	945	1600	95	4700	4000	
50 mg 1^{-1} (mixture)	96	2010	1840	1400	1300	2250	170	5000	4850	
$100 \text{ mg } 1^{-1} \text{ (mixture)}$	72	1995	1900	1035	1500	1090	105	3800	5400	
$100 \text{ mg} \text{ l}^{-1} \text{ Pb}^{2+}$	240	1915	1005	140	70	34950	1030	650	280	
100 mg l ⁻¹ Cd ²⁺	96	1955	640	115	65	2400	80	700	245	

 Table 4

 Effect of external solutions (growth media) on the accumulation of metals in water hyacinth

toxicity by colonising areas of lower metal concentration within the contaminated area, or may avoid, through some exclusion mechanism or selective uptake, excessive cellular concentrations of metallic ions (Antonovies et al., 1971). The concentrations of trace elements in Nile water are very low (Ali and Soltan, 1999) compared with their concentrations in the plants. This observation indicates that water hyacinth absorbs and accumulates a large number of heavy metals (Wolverton and McDonald, 1976).

3.3. Influence of the external solutions and duration of exposure on metal accumulation by the water hyacinth

The concentrations of heavy metals in water hyacinth samples grown in different media are given in Table 4. These data are indicative of two important findings. First, the uptake of heavy metals by plants generally increased with increasing metal concentrations. Second, the water hyacinth has a higher capacity to accumulate heavy metals in the roots than in the aerial parts (Table 4).

The metal contents in water hyacinth plants decreased with increasing growth media concentrations (Table 4). This observation was correlated with the reduction in exposure time (240 h for 1 and 3 mg 1^{-1} ; 192 h for 5 and 7 mg 1^{-1} ; 96 h for 50 mg 1^{-1} ; and 72 h for 100 mg l^{-1} metal solutions) for the plants growing in high concentration media. However, metal accumulation continued during the whole duration of experiments. A marked decrease in metal content for the plants growing in 100 mg 1^{-1} solution compared to other media was paralleled by the relative wilting of plants after 12 h and complete wilting after 72 h. Wilting minimises the uptake of metals by plants, where the process of uptake of chemical species by water hyacinth may take place through the cell membrane through diffusion and osmosis (Haider et al., 1983).

Accumulation factor (Af) ratios for metal concentrations in plant tissue (roots or aerial parts) are plotted in Figs. 1 and 2 to show how the metal accumulation factor decreases with increasing metal content in the growth medium (Wang and Lewis, 1977). The accumulation factors for metal content in the roots and aerial



Fig. 1. Accumulation factor (Af) ratios for metal concentrations in roots/growth media for water hyacinth plants.

parts of plants growing in 100 and 50 mg l^{-1} metal solutions markedly decreased compared to the plants growing in lower concentration media (Figs. 1 and 2), indicating the influence of exposure time and plant wilting on accumulation factors. In addition, the marked decrease in accumulation factor for plant tissue growing in higher concentration media may be due to transpiration of root metals into the external solution, resulting from the effect of toxic metal doses on the survival of water hyacinth. Increased media concentrations (50 and 100 mg l^{-1}) at the termination of experiments as compared to their initial concentrations indicated the above interpretation (Table 5i,j). A common effect of heavy metal accumulation by plants is reduced growth. This is often a result of inhibited photosynthesis and has been shown for plants cultivated at enhanced concentrations of Cd, Pb, Ni, Sn, Hg and Zn (Bazzaz et al., 1974; Schlegel et al., 1987; Greger and Ögren, 1991).

The root/aerial part metal accumulation ratios (Fig. 3) are generally used to show movement of metal within the plant (from roots to aerial parts). The roots/aerial parts ratio markedly decreased for plants growing in 100 and 50 mg l^{-1} metal solutions compared to plants growing in lower concentrations (Fig. 3). This may be due to delicacy and wilting of plant tissues that are responsible for transportation of chemical species from the roots to aerial parts.

The complete lack of toxic effects of lead on plants growing at 100 mg l^{-1} Pb during the period of the experiment (240 h) may be due to the absorbed lead being bound to the cell wall of the plant, thus rendering



Fig. 2. Accumulation factor (Af) ratios for metal concentrations in aerial parts/growth media for water hyacinth plants.

lead ineffective in acting as a strong metabolic inhibitor (Buddhari et al., 1983). When the external concentrations were high (at 10 and 100 mg l⁻¹ Pb), more lead was accumulated in the roots than in the shoots, indicating a relatively low mobility of lead. The immobilised portion is bound to inactive parts of plant cells (Sharpe and Denny, 1976), and the unbound portion is rapidly swept up through transpiration to the upper parts of the plants (Buddhari et al., 1983). This observation was indicated in the present study by the increase in the root/aerial part lead ratio with increasing Pb concentrations in the media compared with other metals (Fig. 3). In addition, a higher concentration of lead was accumulated in the roots (34950 μ g g⁻¹ DM) than the aerial parts (1030 μ g g⁻¹ DM) of water hyacinth growing in 100 mg l⁻¹ Pb²⁺ solution (Table 4). The

plants grown in 100 mg l⁻¹ Cd solution absorbed greater amounts of Cd, with visible wilting starting after 48 h, but the aerial parts of plants exhibited complete wilting with brown colour after 96 h. Cadmium is extremely toxic to organisms because it inhibits a large number of metabolic enzyme systems, complexes with amino acids, peptides and proteins, and affects the conformation of polyriboadenyl acid and the physical properties of DNA (Conway, 1978). Currently, cadmium is considered one of the five most toxic metals in aquatic environments (Nalewajko, 1995). The water hyacinth grown in Kima drain wastewater medium exhibited low concentrations compared to the plants grown in media with lower metal contents (Table 4). The complete wilting of these plants and the reduction in exposure time led to minimisation of the uptake of

Table 5

Effect of water hyacinth growing in different media (a-h) on the concentration of metals in aqueous solution (media) during the study period

Metal	Concentration (mg l^{-1})									
	Time (h)									
	0	24	48	96	144	192	240			
(a) Distille	ed water									
Zn	0	0.36	0.39	0.39	0.43	0.0	0.0			
Cu	0	0.0	0.0	0.0	0.0	0.0	0.0			
Mn	0	0.02	0.0	0.02	0.0	0.0	0.0			
Co	0	0.0	0.0	0.0	0.0	0.0	0.0			
Cd	0	0.0	0.0	0.0	0.0	0.0	0.0			
Ni	0	0.0	0.01	0.0	0.01	0.01	0.02			
Pb	0	0.01	0.0	0.02	0.01	0.0	0.01			
Cr	0	0.0	0.01	0.0	0.0	0.0	0.01			
(b) Nile we	ater									
Zn	0.12	0.0	0.15	0.0	0.0	0.0	0.05			
Cu	0.01	0.01	0.01	0.0	0.01	0.0	0.0			
Mn	0.02	0.0	0.01	0.0	0.0	0.0	0.0			
Co	0.008	0.01	0.03	0.0	0.03	0.01	0.0			
Cd	0.001	0.0	0.0	0.0	0.0	0.0	0.0			
Ni	0.002	0.01	0.0	0.04	0.05	0.03	0.0			
Pb	0.01	0.0	0.02	0.01	0.04	0.02	0.01			
Cr	0.0	0.0	0.0	0.0	0.0	0.0	0.0			
(c) Wastew	vater									
Zn	0.07	0.01	0.0	0.0	0.0	-	_			
Cu	0.03	0.0	0.02	0.0	0.0	-	-			
Mn	0.0	0.45	0.81	0.87	0.42	_	_			
Co	0.03	0.0	0.0	0.01	0.03	-	-			
Cd	0.01	0.0	0.01	0.0	0.0	_	_			
Ni	0.08	0.03	0.0	0.0	0.0	_	_			
Pb	0.29	0.0	0.0	0.0	0.0	_	_			
Cr	0.08	0.0	0.03	0.0	0.0	_	-			
(d) 1 mg l	$^{-1}$ mixed metals so	lution								
Zn	0.99	1	1.20	0.79	0.35	0.45	0.29			
Cu	0.98	0.72	0.52	0.43	0.11	0.15	0.15			
Mn	1	1.20	1.10	0.85	0.48	0.22	0.20			
Co	0.97	0.38	0.45	0.32	0.17	0.11	0.13			
Cd	0.97	0.90	0.60	0.40	0.33	0.16	0.25			
Ni	0.95	0.45	0.55	0.55	0.40	0.23	0.30			
Pb	0.90	0.55	0.45	0.55	0.27	0.22	0.16			
Cr	0.94	0.65	0.52	0.57	0.43	0.38	0.37			
(e) 3 mg l	$^{-1}$ mixed metals so	lution								
Zn	2.95	2.40	3.31	3.63	2.71	1.63	2.45			
Cu	2.98	2.21	2.13	1.85	0.97	0.45	0.63			
Mn	2.93	2.41	2.05	1.74	1.01	0.97	1.51			
Со	2.90	2.35	2.42	1.65	1.42	1.21	1.35			
Cd	2.91	2.15	2.01	1.65	1.43	0.99	1			
Ni	2.85	1.85	1.93	1.86	1.73	1.064	2.53			
Ph	2.76	2.63	2 35	1.12	1 15	1 23	0.68			
Cr	2.91	2.15	1.85	1.97	1.25	0.95	0.15			
(f) 5 mg 1-	⁻¹ mixed metals sol	ution								
Zn	5.03	4.11	4.35	3.42	1.95	1.27	_			
Cu	4.93	4.73	4.66	3.11	1.82	0.51	_			
Mn	5.05	4 21	3 97	3 24	2 53	1.25	_			
Co	4 91	4 22	3 34	3 24	2.55	2.51	_			
Cd	5 05	4 74	4 33	3.24	3 71	2.99	_			
Cu	5.05	7.77	т.55	5.75	5.71	2.77	_			

Metal	Concentration (mg l^{-1})									
	Time (h)									
	0	24	48	96	144	192	240			
Ni	4.90	4.35	3.73	3.51	3.37	3.62	-			
Pb	4.95	3.11	2.95	2.12	2.09	2.85	_			
Cr	4.93	3.20	3.70	2.77	2.47	2.69	-			
(g) 7 mg l	$^{-1}$ mixed metals so	olution								
Zn	6.85	6.85	5.41	5.80	5.81	5.93	_			
Cu	6.79	5.92	4.85	4.91	2.11	1.10	_			
Mn	7.02	6.88	6.75	6.68	6.71	6.15	_			
Co	6.83	6.38	6.55	6.60	6.71	7.03	_			
Cd	6.92	6.45	6.33	6.35	6.61	6.90	_			
Ni	6.88	6.57	6.48	6.35	6.71	6.95	_			
Ph	6 90	6.95	615	6 37	6.45	6 50	_			
Cr	6.92	6.39	5.64	5.71	5.48	5.44	_			
(h) 10 mg	l^{-1} mixed metals s	solution								
Zn	9 78	7 31	7 39	6 90	6 10	5 90	6 40			
Cu	9.81	5.81	6.21	5 50	5.01	4 95	5.81			
Mn	10.08	6.01	5.21	1 43	3.02	2.05	6.36			
Co	0.67	7.05	7.12	4.45	5.02	2.95	0.30			
Cd	9.07	7.93	7.12	0.15	6.12	0.4J 5.22	0./4 6.12			
Cu N:	9.65	/.00	7.20	0.05	0.82	J.25 7 55	0.15			
IN1 Dh	9.95	8.11	8.02	7.85	7.95	/.55	8.99			
Pb	9.73	6.14	6.91	5.44	5.83	6.81	9.01			
Cr	10.1	8.33	6.97	7.25	6.88	8.01	8.12			
(<i>i</i>) 50 mg	l^{-1} mixed metals s	olution	41.25	20.15	40.10	25.40				
Zn	48.91	45.31	41.35	38.15	40.18	35.40	_			
Cu	49.15	44.43	45.61	47.23	48.13	49.85	-			
Mn	49.61	48.13	47.24	46.91	46.13	47.18	-			
Co	48.32	45.94	46.15	47.19	48.11	52.13	-			
Cd	48.79	48.01	49.15	50.13	51.21	53.86	_			
Ni	49.31	49.45	48.13	48.15	49.35	56.14	_			
Pb	49.58	49.59	49.12	49.01	49.25	49.75	-			
Cr	48.91	48.05	48	48.13	48	48.50	-			
(j) 100 mg	l^{-1} mixed metals	solution								
Zn	98.15	98.17	96	92	99.91	-	-			
Cu	98.97	99.10	95.3	95.6	97.15	-	_			
Mn	100.1	98.40	98.35	97.51	96.12	-	_			
Co	98.91	94.31	95.32	94.42	98.27	-	_			
Cd	99.10	95.21	96.43	89.27	93.15	_	_			
Ni	98.45	99.15	100.5	100	103	_	_			
Pb	99.11	99.55	99.67	98.14	95.13	_	_			
Cr	98.73	97.25	96.31	97.18	98.95	_	_			
(k) 100 mg	$l^{-1} Pb^{2+}$ solution	ı								
Zn	0.0	0.47	0.17	0.58	1.35	0.21	1.04			
Cu	0.0	0.0	0.0	0.01	0.0	0.0	0.01			
Mn	0.0	0.09	0.01	0.01	0.0	0.0	0.0			
Со	0.0	0.04	0.09	0.24	0.13	0.22	0.20			
Cd	0.0	0.02	0.0	0.0	0.0	0.0	0.01			
Ni	0.0	0.05	0.02	0.0	0.02	0.03	0.06			
Ph	90.15	80.15	81 35	72 32	65 10	59.15	58.15			
Cr	0.0	0.0	0.17	0.15	0.17	0.15	0.15			
(1) 100 -	$1^{-1} C d^{2+} - 1 \cdots$		5.17	5.15	5.17	0.12	0.15			
(1) 100 mg Zn	$i = Ca^{-1}$ solution	1 15	1 85	2.03	1 47	3.45	_			
2 11	0.0	1.15	1.05	2.05	1.7/	5.75	—			

Table 5 (Continued)

Metal	Concentration (mg 1 ⁻¹) Time (h)											
	Cu	0.0	0.0	0.0	0.0	0.0	0.0	_				
Mn	0.0	0.0	0.0	0.0	0.0	0.0	_					
Co	0.0	0.18	0.20	0.26	0.24	0.19	_					
Cd	99.35	90.13	81.91	75.21	63.95	61.11	_					
Ni	0.0	0.09	0.08	0.08	0.07	0.08	_					
Pb	0.0	0.34	1.49	0.36	0.30	0.41	_					
Cr	0.0	0.18	0.18	0.18	0.18	0.19	_					

Table 5 (Continued)

metals by the plants during the period of the experiment. Table 5 c shows the effect of plants on the concentration of metals in aqueous solution (wastewater). The increase in metal concentrations at the termination of the experiment as compared to the initial concentrations may be attributed to the release of these metals from dead plant tissue into the solution medium. In order for ions and other solutes to accumulate against a concentration gradient, an expenditure of energy is required, either directly or indirectly. The main source of energy in non-photosynthesising cells and tissues (including roots) is respiration. Thus, all factors that affect respiration may also influence ion accumulation. Water hyacinth litter leaches substantial quantities of inorganic and organic nutrients (Reddy and Sacco, 1981).

3.4. Influence of growing plants on different media as a function of pH and conductivity

The different media solutions exhibited increasing conductivity with increasing metal concentrations and duration of exposure to plants (Table 6). pH changes did not follow this trend for the solutions with increasing metal concentrations; however, pH levels generally decreased with increasing metal concentrations from 50 to 100 mg^{-1} and with duration in the growth solution (Table 6). This observation may be explained by the ion-exchange capability of water hyacinth and the marked proton secretion from this plant during metal accumulation. The root system of water hyacinth has pH-dependent charge sites, which are responsible for the absorption and accumulation of large amounts of cations by the roots (Yahya, 1990). Both in roots and in the cell wall continuum of other plant tissue, the socalled apoplasm, carboxylic groups (R·COO⁻) act as cation exchangers. In roots, therefore, cations from the external solution can accumulate via a non-metabolic step in the free space, whereas anions are repelled (Marschner, 1997). With respect to the distilled water medium, the increase in conductivity may be the result of diffusion (Table 5a). Solutes may be more concentrated on one side of the membrane, and thus diffuse

from a higher to a lower concentration. As a rule, ion uptake by plant cells and roots has features of saturation kinetics. This is in accordance with the assumption of control, as for example by the number of binding sites of ions (carrier, fermeases), or the capacity of the proton efflux pumps, in the plasma membrane and tonoplast. At the termination of the experiments, the media were analysed to determine the anions and cations present in the solutions which led to the lowering of pH and the increase in conductivity, in addition to acidic groups. Soltan (1999) stated that the roots of water hyacinth accumulated high concentrations of SO_4^{2-} $(679 \pm 40 \ \mu g \ g^{-1} \ DM), \ Cl^{-} \ (635 \pm 29 \ \mu g \ g^{-1} \ DM),$ Ca²⁺ (88±10 µg g⁻¹ DM) and Mg²⁺ (109±7 µg g⁻¹ DM). Results of the present study showed considerably lower amounts of these ions (Table 7). The decrease in pH (from 9.52 to 6.65) and the increase in conductivity with increasing exposure time were also observed in the wastewater medium (Table 6). The wastewater of Kima drain is characterised by high concentrations of ammonia $(31\pm 5 \text{ mg } l^{-1} \text{ NH}_4^+)$ and a high pH value (9.52 \pm 0.5), resulting from the discharge from Kima fertiliser manufacturing (Soltan, 1995). The lowering of pH is related to the deprotonation of NH_4^+ and the increase in conductivity to the release of ions from wilting root tissues of plants growing in this medium (Table 7). Mengel et al. (1976) reported that at least a substantial proportion of ammonium nitrogen is not taken up in the form of NH_4^+ , but that NH₃ permeates the plasma membrane, indicating deprotonation, leaving H^+ in the external solution. Studies with lower plants indicate that deprotonation before uptake (i.e. permeation as NH₃) may become increasingly important at higher substrate concentrations of NH⁺₄ (Berti et al., 1984). Water hyacinth litter leaches substantial quantities of inorganic and organic nutrients, particularly nitrogen, phosphorus and carbon, during both its initial and later phases of decay (Varghese, 1991). Considerable amounts of Mn (0.42 mg l^{-1}), Pb $(0.09 \text{ mg } 1^{-1})$ and Co (0.03 mg^{-1}) (more than the initial concentrations) were detected in the wastewater medium at the termination of the experiment (Table



Fig. 3. The root/aerial parts metal accumulation ratios for water hyacinth growing in media containing different metal concentrations.

Media	Duration of	pН	pH							
	experiment (h)	0 h	24 h	48 h	96 h	144 h	192 h	240 h		
Distilled water	240	7.35	7.47	6.73	6.50	7.11	7.09	7.02		
Nile water	240	8.11	7.74	7.78	7.49	7.30	7.54	7.73		
Wastewater	96	9.52	8.72	8.38	8.02	6.65	_	_		
Heavy metal solution										
$1 \text{ mg } 1^{-1}$ (mixture)	240	5.21	5.73	5.69	6.04	6.42	6.78	6.75		
$3 \text{ mg } 1^{-1}$ (mixture)	240	5.27	5.65	5.56	5.87	6.36	6.69	6.67		
5 mg 1^{-1} (mixture)	192	5.35	5.37	5.20	5.42	5.87	5.82	_		
7 mg l^{-1} (mixture)	192	5.43	5.12	5.13	5.33	5.64	5.61	_		
$10 \text{ mg } 1^{-1} \text{ (mixture)}$	168	6.13	6.22	6.22	6.30	6.66	_	_		
50 mg l^{-1} (mixture)	96	6.21	6.25	6.10	5.99	_	_	_		
$100 \text{ mg } 1^{-1} \text{ (mixture)}$	72	6.10	5.99	5.75	_	_	_	_		
$100 \text{ mg } 1^{-1} \text{ Pb}^{2+}$	96	6.52	6.65	6.60	6.65	_	_	_		
$100 \text{ mg } l^{-1} \text{ Cd}^{2+}$	72	6.06	6.18	6.26	6.45	6.70	6.96	7.10		
		Conduct	tivity (µS cr	n^{-1})						
Distilled water	240	0 h	24 h	48 h	96 h	144 h	192 h	240 h		
Nile water	240	3	11	17	35	44	45	47		
Wastewater	96	159	207	212	246	258	278	293		
Heavy metal solution		1186	1223	1309	1338	_	_	-		
$1 \text{ mg } 1^{-1}$ (mixture)	240									
$3 \text{ mg } 1^{-1}$ (mixture)	240	78	96	114	111	131	134	158		
5 mg 1^{-1} (mixture)	192	114	143	163	178	192	225	259		
7 mg 1^{-1} (mixture)	192	163	186	207	218	236	249	-		
$10 \text{ mg } 1^{-1} \text{ (mixture)}$	168	213	270	309	332	370	387	_		
50 mg 1^{-1} (mixture)	96	249	340	364	375	435	_	_		
$100 \text{ mg } 1^{-1} \text{ (mixture)}$	72	1253	1270	1353	1442	_	_	_		
$100 \text{ mg } 1^{-1} \text{ Pb}^{2+}$	96	2190	2260	2310	_	_	_	_		
$100 \text{ mg} 1^{-1} \text{ Cd}^{2+}$	72	198	270	294	298	_	_	_		
		191	247	281	288	313	333	357		

Table 6 Effect of growing plants on different media as a function of pH and conductivity

Table 7 Concentrations of anion and cation secretions resulting from water hyacinth growing in different media

Media	Concentration (mg 1^{-1})									
	HCO ₃ ⁻	SO_4^{2-}	Cl ⁻	Ca ²⁺	Mg^{2+}					
Distilled water	14.3 ± 0.2	3.5 ± 0.1	10.9 ± 0.3	5 ± 0.1	0.2 ± 0.1					
Nile water	35 ± 0.5	8.5 ± 0.3	20.6 ± 0.8	6.2 ± 0.1	3 ± 0.1					
Wastewater	36.7 ± 0.4	14.8 ± 0.4	29.8 ± 0.6	7.9 ± 0.1	2.1 ± 0.1					
Heavy metal solution										
$1 \text{ mg } l^{-1}$ (mixture)	24.9 ± 0.3	11.4 ± 0.2	17.1 ± 0.1	4.1 ± 0.1	1.8 ± 0.1					
$3 \text{ mg } 1^{-1}$ (mixture)	41.7 ± 0.6	20.9 ± 0.3	33.2 ± 0.3	8.2 ± 0.1	2.1 ± 0.1					
$5 \text{ mg } l^{-1}$ (mixture)	47.4 ± 0.3	28.7 ± 0.2	35.4 ± 0.3	9.7 ± 0.1	3.3 ± 0.1					
7 mg l^{-1} (mixture)	42.1 ± 0.5	26.6 ± 0.3	31.5 ± 0.5	9.1 ± 0.1	3.2 ± 0.1					
$10 \text{ mg } l^{-1}$ (mixture)	45.9 ± 0.6	28.1 ± 0.4	34.4 ± 0.3	9.8 ± 0.1	3.1 ± 0.1					
50 mg 1^{-1} (mixture)	44.8 ± 0.5	30.2 ± 0.3	33.8 ± 0.5	10.3 ± 0.2	3.6 ± 0.1					
$100 \text{ mg } 1^{-1} \text{ (mixture)}$	29.8 ± 0.2	17.6 ± 0.3	25.8 ± 0.4	7.3 ± 0.1	2.4 ± 0.1					
$100 \text{ mg } l^{-1} \text{ Pb}^{2+}$	28.1 ± 0.3	15.3 ± 0.2	21.3 ± 0.3	8.9 ± 0.1	2.9 ± 0.1					
$100 \text{ mg} \text{ l}^{-1} \text{ Cd}^{2+}$	22.4 ± 0.2	13.6 ± 0.2	16.9 ± 0.2	6.4 ± 0.1	1.9 ± 0.1					

4c). Similar investigations carried out on higher plants indicated the release of nutrients when the internal concentration was high (Gaur et al., 1992). Measurements of influx clearly indicate that besides efflux, other mechanisms contribute to the decline in net uptake when internal concentrations are high. In barley roots, with increasing internal nitrate concentrations, both I_{max} and K_{m} decrease by a factor of four–five, indicating effective feedback regulation of the influx component (Siddiqi et al., 1990).

4. Conclusion

Water hyacinth accumulates higher concentrations of heavy metals in the root than in the aerial part. It has relatively low nutrient requirements and can survive in distilled water (depending on the nutrients accumulated in its root and aerial part). The plants appeared healthy during experiments using 1 and 3 mg 1^{-1} solutions of metal mixtures and 100 mg 1^{-1} Pb solution, otherwise they exhibited gradual wilting when grown in 5, 7 and 10 mg 1^{-1} metal solutions and quick fading in 100 mg 1^{-1} solutions of metal mixtures, 100 mg 1^{-1} Cd solution, and wastewater characterised by a deficiency of dissolved oxygen (1.44 mg l^{-1}), high concentration of ammonia $(31\pm 5 \text{ mg } l^{-1} \text{ NH}_4^+)$ and high pH value (9.52 ± 0.50) . Intact membranes are effective barriers to the passage of ions and uncharged molecules, and thus the plants growing in 1 and 3 mg 1^{-1} metal solutions exhibited higher bioaccumulation factors than the plants growing in high concentrations of metals (50 and 100 mg 1^{-1} as mixtures). The non-toxic effect of lead on plants was marked up to 100 mg l^{-1} , which is in contrast to Cd; this is due to the adsorbed lead being mostly bound to the cell wall of the plants, thus rendering lead ineffective in acting as a strong metabolic inhibitor. Water hyacinth effectively removes appreciable quantities of heavy metals (Cd, Co, Cr, Cu, Mn, Ni, Pb and Zn) from freshwater (Table 5b), especially at low concentrations (below 10 mg l^{-1} as a mixture), whilst it releases small quantities of heavy metals at wilting (Table 5f-h), and thus the plants must be continuously removed from the water medium after the vegetation period. Generally, the role of water hyacinth can be regarded as a heavy metal decontaminator in aquatic environments, rather than as a pollutant.

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