Al-Azhar Bulletin of Science

Volume 18 | Issue 2 Article 8

12-1-2007

Section: Botany, Microbiology and Zoology

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How to Cite This Article

ABDALLA, S.; MAHDY, E.; SOLIMAN, M.; and RAMADAN, D. (2007) "EFFECT OF CERIUM AND CADMIUM IN LIQUID RADIOACTIVE WASTES ON THE MORPHOLOGY, PROTEINS, CARBOHYDRATES AND NUCLEIC ACIDS CONTENTS OF THE AQUATIC PLANT LUDURIGIA STOLONIFERA," *Al-Azhar Bulletin of Science*: Vol. 18: Iss. 2, Article 8.
DOI: https://doi.org/10.21608/absb.2007.11631

Al-Azhar Bull. Sci. Vol. 18, No. 2 (Dec.): pp. 25-35, 2007.

EFFECT OF CERIUM AND CADMIUM IN LIQUID RADIOACTIVE WASTES ON THE MORPHOLOGY, PROTEINS, CARBOHYDRATES AND NUCLEIC ACIDS CONTENTS OF THE AQUATIC PLANT *LUDURIGIA STOLONIFERA*

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Abstract

Aquatic plant *Ludurigia stolonifera* is used to absorb two toxic ions (Cd and Ce) from low level radioactive waste solutions. The effects of these toxic ions with three concentrations on the morphology, carbohydrates, proteins and nucleic acids (DNA, RNA) contents of the plant were studied. The results obtained in the present investigation showed a decrease in the weight of the plant, proteins, carbohydrates and nucleic acids contents with the increase in the concentration of toxic ions.

Introduction

Aquatic environment provides an ecosystem service to humans. Yet it often receives high levels of pollutants from both direct and indirect sources. Several methods are used to remove great amount of hazardous, industrial and radioactive wastes such as, treatment, storage and disposal of these wastes. The increased loading of heavy metals in water produces increase in human health hazards or cell death in plants Nyarai-Horvath et al., (1997); Jagetiya et al., (1998); Sahi et al., (1998); Anita et al., (1999) and Sanchez et al., (1999). The absorption of toxic or hazardous ions by the aquatic plant is proved to be economic, simple and easy method Wolverton and Donald (1986). The distribution of toxic elements or radioactive substances entering the plant is influenced by the chemical characteristic of ions and their interaction with constituents of plant tissues. The presence of excessive amount of Cd in the soil elicits many symptoms in the plant such as root growth Weigel and Jagar (1980) and Rubio et al., (1994). Nirupama et al., (1996) detected that the concentration dependent decrease protein, RNA and DNA and nutrient uptake by Lemna minor and Azolla pinnata resulting from exposure to some heavy metals. Also Saygideger (2000) investigated the absorption of Cd and its effects on growth, protein content of Veronica anagallis aquatic L.and Ranunculus aquatic L. Ramadan (1999) showed the changes in carbohydrates content of Potamogeton crispus plant immersed in Cd solution and observed that the treatment decreased carbohydrate contents of the plant. Hu et al., (2002) studied the effect of Ce on the vegetable growth, root length, dry weight of root and shoots. Zhang and Yan (2003) found that, many kinds of organic matter such as sugar, amino acids and organic acids excreted by root in wheat were affected by Cd stress.

The present work was devoted to investigate the changes that may occur in morphology, proteins, carbohydrates and nucleic acid contents after immersion of *Ludurigia stolonifera* plant in solutions of cerium or cadmium at different concentrations.

Materials and Methods

Materials:

The plant material

The floated plant *Ludurigia stolonifera* (Rhizophoraceae) consists of floating leaves, yellow small flowers and fibrous roots with pale pink color Fig.(1). The plants were distributed along the Ismailia Canal. A stock of plant was kept in glass aquaria of dimensions of 100x50x50 cm in the laboratory and filled with fresh water. The water temperature was 25±3°C.

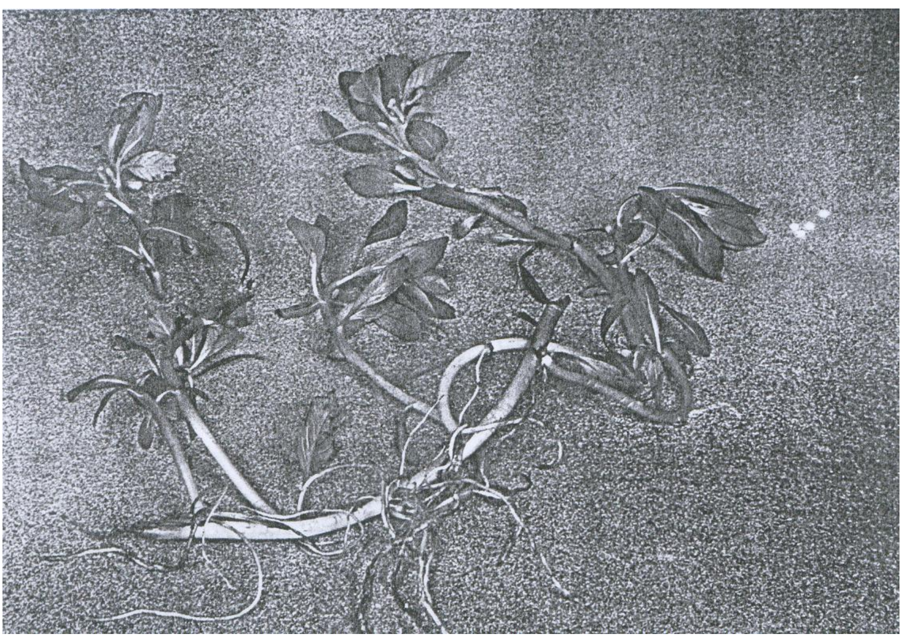

Fig.(1): *Ludurigia stolonifera* plant

Chemicals:

The used chemicals were of analytical pure in the form of cerium and cadmium chloride. The ions of such heavy metals were applied as aqueous solutions of concentrations of $7x10^{-4}$M and 10^{-4}M respectively. These chosen concentrations were based on the results obtained from an experiments carried firstly to determine the uptakes and the effective concentration and the lethal one. PH of solutions was adjusted using 1N. of hydrochloric acid or sodium hydroxide.

Treatment of the samples:

The plant *Ludurigia stolonifera* was immersed in solution of $7x10^{-4}$ of Ce(III) and 10^{-4}M of Cd(II).

Effect of toxic ion concentrations on the morphology of *Ludurigia stolonifera* plant:

A morphological changes for shoot and root systems after immersing the plant in solutions of Ce(III) of $7x10^{-4}$M, $1.4x10^{-3}$M and $2.8x10^{-3}$ or in solutions of 10^{-4}M . $5x10^{-4}$M and 10^{-3}M of Cd(II) were investigated .

Determination of total carbohydrates content:

The plant was immersed in solutions of Ce(III) and Cd(II) with concentration of $7x10^{-4}$M and 10^{-4}M respectively for two days. The stems and leaves were collected and dried. The total carbohydrates were estimated according to Bernsled and Miller (1959).

Determination of proteins and nucleic acids content:

Proteins and DNA were determined quantitatively by Agarose electrophoresis densitometry, while RNA was estimated quantitatively by Peppel and Baglioni (1962) by using freezing fresh green leaves after immersing the plant in solutions of $7x10^{-4}$M of Ce(III) and 10^{-4}M of Cd(II) for two days. A blank experiment was carried out by immersing the plant in distilled water for the same time.

Results:

Effect of toxic ions on the morphology of *Ludurigia stolonifera* plant:

Results presented in Table (1) and Figs.(2 and 3) revealed the morphological changes that occurred in the root and shoot systems of *L. stolonifera* plant after 10 days of immersion in different concentrations of Ce(III) of $7x10^{-4}$M .$1.4x10^{-3}$M and

2.8x10^{-3}M and 10^{-4}M, 5x10^{-4}M and 10^{-3}M of Cd(II). The results showed that, the weight and length of shoot and root systems decreased when the concentrations of toxic ions were increased as compared to control group.

Table (1): Morphological measurements after immersion of *L. stolonifera* plant in different concentrations of toxic ions.

Toxic ion	Conc. (M)	Weight (g) before immersion	Weight (g) after immersion	before immersion (Cm)			After immersion (Cm)		
				root length	Shoot length	root shoot ratio	root length	Shoot length	root shoot ratio
Ce III	7X10^{-4}	15	13.8	13.7	37	0.37	12.1	38.8	0.54
	1.4X10^{-3}	16	9.5	14.5	37.2	0.38	13.3	35.5	0.37
	2.8X10^{-3}	17	8	10.8	40.3	0.26	8.8	36.5	0.24
Cd II	10^{-4}	14	10	7.9	32	0.24	6.5	29.9	0.21
	5X10^{-4}	14	9	10.9	35.8	0.30	7.5	31.5	0.23
	10^{-3}	15	8	8.5	32.9	0.25	5.2	29.3	0.17
Control	-	15	16.7	9	38	0.23	10	38.5	0.26

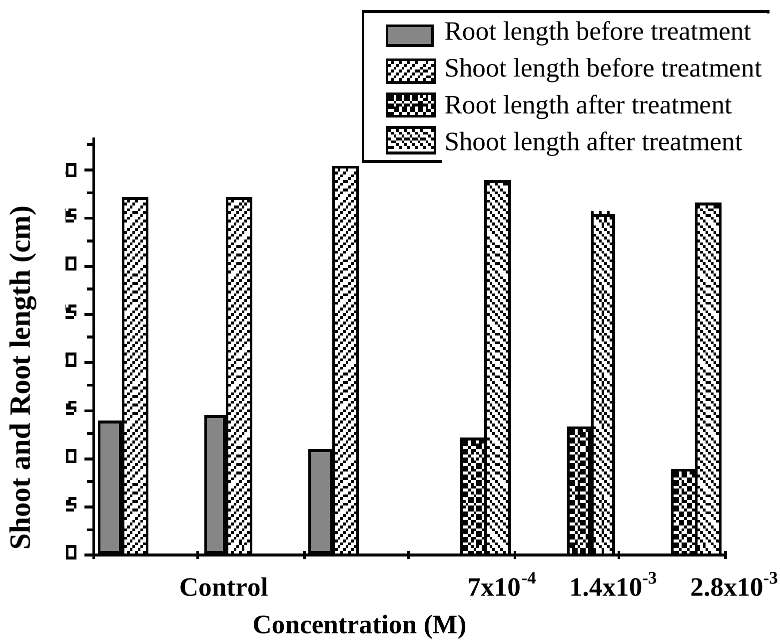

Fig. (2): Shoot and root length (cm) before and after treatment with different concentrations of Ce(III)

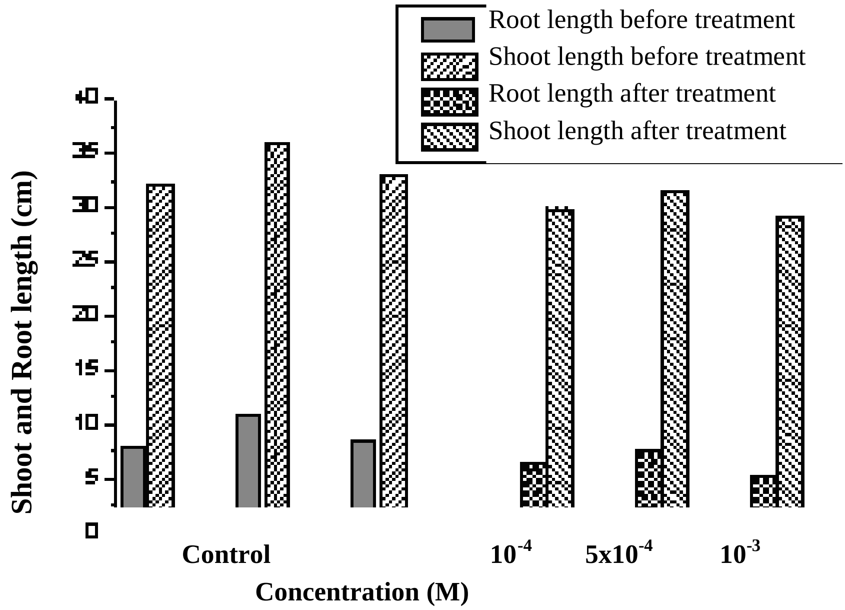

Fig. (3): Shoot and root length (cm) before and after treatment with different concentrations of Cd(II)

Effect of toxic ions on the total carbohydrate content:

Results presented in Table (2) and Fig. (4) showed the changes in the carbohydrate content of *Ludurigia stolonifera* plant after immersion of the plant in the lower concentration of Ce(III) $7x10^{-4}$M and 10^{-4}M of Cd(II). The total carbohydrate content was decreased by increasing the concentration of toxic ions (Ce and Cd) as compared to control group, where, the total carbohydrate contents of Cd(III) group showed the lowest values.

Table (2): Determination of total carbohydrate gm/100gm dry weight using the lowest concentration of toxic ion solutions after two days of immersion.

Toxic ions	Conc. M	Total carbohydrates	Non soluble sugar	Soluble sugar	
				Reducing	Non reducing
Ce(III)	$7X10^{-4}$	34.74	24.28	7.15	3.31
Cd(II)	10^{-4}	25.53	15.5	8.09	1.94
control	-	37.98	27.81	7.01	3.16

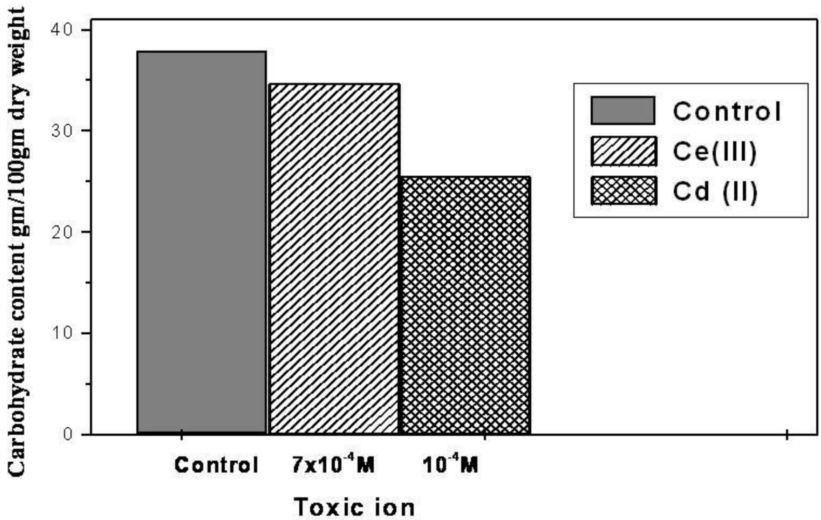

Fig. (4): Contents of carbohydrates after immersion of *L.stolonifera* in the low concentrations of toxic ions.

Effect of toxic ions on the proteins and nucleic acids content:

Results presented in table (3) and Figs.(5-7) showed the changes in the contents of proteins, RNA and DNA of ***L.*** *stolonifera* plant after immersion in low concentration of Ce(III) ($7x10^{-4}$M) and of Cd(II) (10^{-4}M). The results indicated that the above contents decreased by using the previous toxic concentrations as compared with control.

Table (3): Determination of proteins, DNA and RNA using the low concentrations of toxic ion solutions after two days of immersion.

Toxic ions	Conc. M	Proteins µg/1g fresh weight	DNA µg/1g fresh weight	RNA µg/1g fresh weight
Ce III	$7X10^{-4}$	19.1004	5387.9	82.8
Cd II	10^{-4}	5.0962	5104.7	60.4
Control	-	52.022	5911.8	141.6

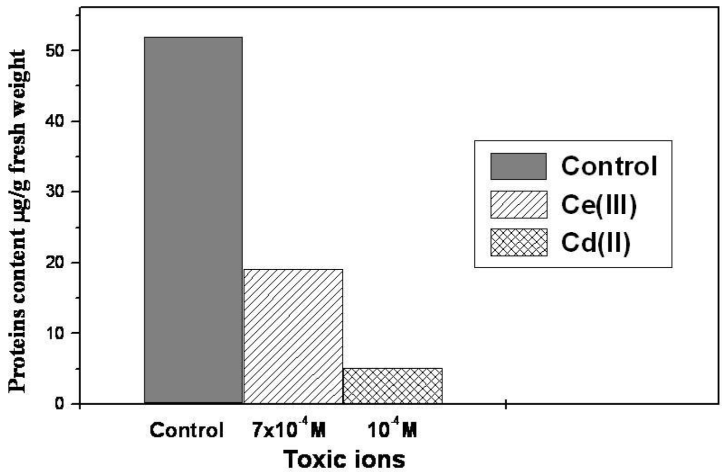

Fig. (5): Contents of proteins after immersion of *L.stolonifera* in the low concentrations of toxic ions.

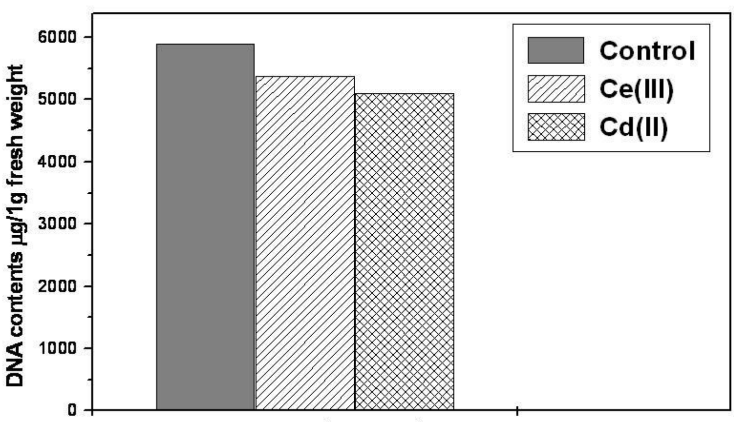

Fig. (6): Contents of DNA after immersion of *L. stolonifera* in the low concentrations of toxic ions for two days.

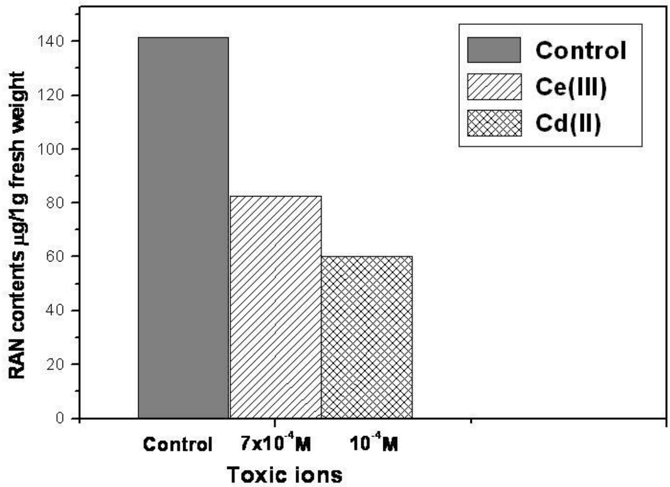

Fig. (7): Contents of RNA after immersion of *L. stolonifera* in the low concentrations of toxic ions for two days.

Discussion

Results of the present study showed that morphological changes occurred after immersing *L. stolonifera* plant in three different concentrations of toxic ions Ce(III) ($7x10^{-4}$M, 1.4 $x10^{-3}$M and 2.8 $x10^{-3}$M) and of Cd(II) (10^{-4}M , $5x10^{-4}$M and 10^{-3}M).

In this respect it was found that the low concentration of Ce(III) ($7x10^{-4}$M) caused minor changes in the morphology of the plant (root and shoot length) where the concentration of Cd(II) (10^{-4}M) caused changes in the leaf color from green to yellow while the root changed from pink to brown color. Increasing Cd(II) from(10^{-4}M) to 10^{-3}M showed decrease in shoot and root length.

On immersion of the plant in solutions of Ce(III) (1.4 $x10^{-3}$M and 2.8 $x10^{-3}$M), it was found that the leaves changed to yellow color and then dried with appearance of brown spots. The roots changed to brown color and the length for both root and shoot decreased. These results are in agreement with Hu. et al (2002), Wu. & Zhang (2002) and Wu. et al (2003).

On the other hand the effect of low concentration of Cd(II) decreased proteins, nucleic acid and total carbohydrates contents rather than that of the corresponding decrease in the same contents using Ce(III) due to its toxicity. These results agreed with Aslan et al (2003) where they showed that exposure of *Nasturtiom officinal* and Mentha *aquatica L.* to cadmium decreased protein concentration of leaves.

From the above results we can conclude that *L. stolonifera* plant can be used as a good phytoremediator or a good vehicle to clean up the waste water from toxic ions. These results are in complete accordance with Faisal (2005).

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تأثير بعض الأيونات السامة فى النفايات المشعة السائلة على الشكل الظاهرى والمحتوى البروتينى والكربوهيدراتى والأحماض النووية لنبات المداد المائى

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الملخص العربى

تم استخدام نبات المداد المائى لامتصاص الأيونات السامة لعنصري (السيريوم والكادميوم) المتواجدة فى محاليل النفايات المشعة السائلة ذات المستوى الضعيف ومدى تأثير تلك الأيونات بتركيزات مختلفة على الشكل الظاهرى والمحتوى البروتينى والكربوهيدراتى والأحماض النووية للنبات.

ولقد أظهرت الدراسة أنه بزيادة تركيز تلك الأيونات يقل وزن وطول المجموع الخضرى والجذرى للنبات وأن عنصر الكادميوم يكون أشد ضررا على الساق والأوراق مع ظهور بعض البقع السوداء وسرعة جفافها، كما ينخفض المحتوى الكربوهيدراتى والبروتينى والأحماض النووية للنبات المعامل بتلك الأيونات بالمقارنة بالنبات الغير المعالج.