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The Ability of Red Fountain Grass to Grow in Cadmium-Contaminated Soil

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ABSTRACT

A pot experiment was conducted during two successive seasons 2017 and 2018 at experimental farm of Faculty of Agriculture, Cairo University, Giza, Egypt. The aim of this study is to determine the ability of *Pennisetum setaceum* cv. rubrum plant to grow in cadmium (Cd) contaminated soil (100 mg/kg). Also, investigate the effect of citric acid application (1 and 2 mmol/kg) on phyto-extraction of cadmium-contaminated soil. The results clarified that the presence of cadmium (100 mg/kg) in the soil caused low reduction in the herb growth compared to unpolluted soil. The reduction rates in both seasons were 20.33 and 26.47% for herb fresh weight, 5.08 and 14.79% for herb dry weight, 8.86 and 0.0% for number of inflorescences/plant, respectively. The treatment of citric acid 1 mmol/kg increased the Cd content 149.3, 137.5% in the herb, and 221.25, and 170.81% within plant root in both seasons, respectively compared to the control. Also, such treatment raised phenols content in the herb to 102.3 and 93.0% compared to control plants in both seasons, respectively. The results showed that translocation factor (TF) was bigger than one which means that the plants had accumulated the cadmium in their tissues; however the plant tolerated Cd-contaminated soil accompanied with high content in both phenols and catalase enzyme activity. It could be concluded that *P. setaceum* cv. rubrum is a promising plant for phytoremediation.

Keywords: Cadmium, Citric Acid, polluted soil, *Pennisetum setaceum* cv. rubrum, phytoremediation

INTRODUCTION

Nowadays soil pollution has been increased as a side effect of the negative behavior of human beings to achieve prosperity. Hashim *et al.* (2017) studied the cadmium concentration of soils adjacent to Cairo – Alexandria Agricultural Highway, Egypt and they found that soil cadmium exceeded the permissible levels of the United Kingdom (1 mg/kg), Germany (2 mg/kg), and Poland (3 mg/kg) accordingly; those soils are considered contaminated with cadmium. Al Naggar *et al.* (2014) studied the concentration of the heavy metal in four different Egyptian regions; they found that the concentration of Cd in soil locations surpassed the maximum safety concentration (8 mg/kg, dry weight). The concentration of Cd ranged between 11.35-28.88 mg/kg dry weight. Also, concluded that cultivated such soils may result in hazards to human health. Also, using sewage sludge and phosphate fertilizers are known as important sources of Cd pollution.

The Nile Delta suffers from different pollution sources including heavy metals as a result of increasing the different types of industries, the use of drain water supply and increasing waste deposits, fertilizers, and agriculture irrigation. Abou El-Anwar *et al.* (2018) studied the quality of the sediments of the Rosetta Nile branch and they found that the elemental ratio and index of geo-accumulation of the Rosetta bottom sediments are classified as moderately polluted when contained copper, cobalt, nickel and iron (Cu, Co, Ni, and Fe), moderately to strongly polluted with chromium and zinc (Cr and Zn) and very strong polluted with lead and cadmium (Pb and Cd). They mentioned that the ecological risk comes mainly from sediment pollution with Pb and Cd.

Thus, the phytoextraction method is required as an environmental friend way to eliminate or at least reduce the pollution. Phytoextraction is a mechanism that removes heavy metals from the soil by plants that capable of concentrate the heavy metal inside their tissues.

Pennisetum setaceum cv. rubrum plant belongs to the family Poaceae and commonly known as Red fountain grass, it is purple-pigmented ornamental grass. It has multiple landscape uses such as border, erosion control, rock garden, urban garden. It needs a warm climate for showy leaf color. Red fountain grass was chosen because it is evergreen grass, thrives in hot climates, and has drought tolerance (González-Rodríguez *et al.*, 2010). It belongs to C4 grasses which have a special photosynthetic pathway that adapts them to hot climates. The plant has long blooming summer and fall. Plants grow in full sun, well-drained soil, and prefer dry locations. The plant has outstanding characteristics; spikes of purplish flowers gracefully spray out of its mass of long colored leaves (Williams and Black, 1996).

Contamination of soils with cadmium is a crucial agent affecting soil properties and plant growth. Cadmium is toxic to most plants at a low level, while other plants had varying capabilities to grow under relatively high cadmium levels (Coakley *et al.*, 2019). Plants have several techniques to overcome cadmium pollution, including synthesis of metal-chelating proteins, the exhibition of enzymatic and

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non-enzymatic antioxidants, organic acids and plant root-mycorrhizal association (Wahid *et al.*, 2009).

Citric acid can be used for enhancing phytoextraction of Pb with no environmental constraints, (Josangela *et al*., 2010). The rhizosphere contains low molecular weight organic acids that affect the mobility of cadmium, meanwhile, Cd exhibits high mobility in acid environments (Kabata-Pendias and Pendias, 1992).

Citric acid is biodegradable rapidly to carbon dioxide and water which makes it an ideal soil amendment for Cd phytoextraction and also helps prevent the possible movement of Cd-Citric acid complexes across the soil profile. The addition of organic acids could promote Pb and Cd accumulation in the plant tissues (Han *et al.* 2018). Also, Farid *et al.* (2019) studied the phytoextraction of chromium (Cr) by sunflower. They found that the application of Citric acid significantly reduced the production of reactive oxygen species in the plants.

The purpose of present research was to examine the ability of *Pennisetum setaceum* cv. rubrum plant to grow in cadmium contaminated soil. Also, to test the potential effect of citric acid application on facilitating the phytoextraction process in Cd-polluted soil.

MATERIALS AND METHODS

Pots Experiment:

The research was conductive during two successive seasons of 2017 and 2018, at the experimental nursery of Faculty of Agriculture, Cairo University, aiming to study the growth responses of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil. Also, assess the use of citric acid as a soil application on phytoextraction of cadmium. The research was conducted in Giza Governorate, as the weather is very hot in summer and warm most of the year. Soil mixture (2 sand: 1 peat moss) was mixed with cadmium nitrate [Cd $(NO_3)_2$ $.4H_2O$] at the dose of 100 mg/kg of soil mixture (about 2.45 folds that present in Egyptian soil according to AL Naggar *et al.*(2014) then equilibrate for 2 weeks. On 15th February 2017 and 2018 seasons, similar tillers (offsets) were planted in plastic pots with 28 cm in diameter and height, filled with the soil mixture (10 kg/pot).Watering was done when the plants needed using tap water. Care was taken to avoid leaching of cadmium from the pots to unpolluted soil using polyethylene sheets under the experimental pots. Plants were supplied with NPK chemical fertilizers during the growing season. Each pot supplied with 6.0 g of Enciabein commercial fertilizer (40.0% N), 6.0 g triple superphosphate (46.0% P_2O_5), and 3.0 g potassium sulphate (48% K_2O) divided into two equal doses, the first dose was added 30 days after planting and the second one, two months later. After the plants grown two months in Cd-contaminated soil, citric acid was added at the doses of 1 and 2 mmol/ kg of soil mixture. Data on plant length [cm], herb fresh and dry weights [g], number of tillers/plant, number of inflorescences/plant, and root fresh and dry weights [g] were recorded after eight months from planting.

The layout of the experiment was complete randomized block design, with three replications of six treatments in each season. The treatments were as the following: Control, citric acid at the dose of 1 mmol/kg of soil mixture(CA1), citric acid at the dose of 2 mmol/kg of soil mixture(CA2), cadmium at the dose of 100 mg/kg of soil mixture(Cd), cadmium at the dose 100 mg/kg of soil mixture plus 1 mmol citric acid per kg of soil mixture (Cd -CA1) and cadmium at the dose of 100 mg/kg of soil mixture plus 2 mmol citric acid per kg of soil mixture(Cd –CA2).

Soil and Chemical Analyses:

Pots filled with equal amounts of homogeneous soil mixture of sand and peat moss (2:1) with the following mechanical characteristics: coarse sand 21.3 %, medium sand 46.8, fine sand 28.1, clay+ silt 3.8 %, field capacity 17 (by volume) , wilting point 8.2 (by volume) and available water 8.8%. Chemical characteristics were shown in (Tab.1). Mechanical and chemical analyses of soil were analyzed as described by (Klute, 1986 and Jackson, 1973).

Table 1. Chemical analysis of the experimental soil

Chemical Analysis														
pH [1:25]	$CaCO_3$ %	E.C. [dS/m]	OM %	Soluble Cations [meq /L]				Soluble Anions (meq /L)			Available macronutrients [mg/kg]			Heavy metal [mg/kg]
				Na^+	K^+	Ca^{++}	Mg^{++}	HCO_3^-	Cl^-	$SO4^{--}$	N	P	K	Cd
7.6	2.23	2.64	0.41	18.1	0.95	5.64	4.30	2.00	14.8	12.20	377.3	2.4	320	0.1

Cadmium soil content was determined at the end of the experiment. Total cadmium accumulation [mg/kg] in dry herb and dry root was determined at the end of the experiment, using the atomic absorption spectrophotometry method recommended by (Helrich, 1990).

Chemical analysis of fresh leaves was conducted to determine the contents of total chlorophyll (a + b) [mg/g FW], using the method described by (Moran, 1982). The total carbohydrates percentage was determined according to (Herbert *et al.*, 1971).

Total phenolic contents [mg GAE/ g DW] of leaves were determined spectrophotometrically according to the Folin-Ciocalteu colorimetric method (Singleton and Rossi, 1965).

Nitrogen percentage determination was carried out using the modified micro Kjeldahl method, as described by Pregl (1945). The phosphorus content was estimated, as recommended by (King, 1951). Potassium was determined according to (Cottenie *et al.*, 1982).

Measuring catalase activity (Unit/g) in fresh herb in the second season was determined according to (Beyer and Fridovich, 1987) using UV/V Spectrophotometer, Jenway, England.

Translocation factor (TF) was calculated as ratio of heavy metal in plant shoot to that in plant root, according to (Mirecki *et al*., 2015).

$$TF = \frac{\text{Metal concentration in shoots}}{\text{Metal concentration in roots}}$$

If the ratios >1, the plants have accumulated elements, the ratios around 1 indicate that the plants are not influenced by the elements, and ratios <1 show that plants exclude the elements from the uptake. If the plants have higher TF values, they can be used for phytoremediation.

Statistical Analysis of Data:

Data recorded on vegetative growth traits were subjected to normal statistical analysis as shown by (Snedecor and Cochran, 1982). Comparison among means was done using "Least Significant Difference (LSD) "at $p < 0.05$.

RESULTS AND DISCUSSION

Growth parameters

The acquired data from *Pennisetum setaceum* cv. rubrum plant growth parameters represented in (Tab. 2) revealed that the use of citric acid resulted in a significant decrease in the growth of the Red fountain plant compared to control plants. In both seasons, the lowest values of herb growth were recorded with citric acid at the dose of 2 mmol/kg which gave the highest reduction rate (Fig.1C) (32.58%, 34.52%) for plant length, (39.46%, 44.29%) for herb fresh weight, (39.64%, 34.54%) for herb dry weight and (60.0%, 59.5%) for the number of inflorescences, in the two seasons, respectively, compared to control plants (Fig.1A). On the other hand, the control plants gave the lowest number of tillers/plant compared to all other treatments. The effect of citric acid on plants was also mentioned by (Kamel and Sakr, 2009). They found that treating soil with citric acid decreased vegetative growth characteristics as well as concentrations of total chlorophylls in leaves and total carbohydrates in the shoots of *Senna occidentalis* plants.

Concerning the root growth, data presented in (Tab. 2) showed that the presence of cadmium in the soil at the dose of 100 mg/kg combined with citric acid at the dose of 1 mmol/kg led to the highest reduction in root growth represented in (45.10%, 50.44%) for root fresh weight, (30.38%, 34.56%) for root dry weight compared to healthy control plants in the two seasons, respectively.

The lowest reduction rate in herb growth compared to healthy control plant was achieved with cadmium application at the dose of 100 mg/kg (Fig.1D). The reduction rate in both seasons represented in (23.33%, 29.18%) for plant length, (20.33%, 26.47%) for herb fresh weight, (5.08%, 14.79%) for herb dry weight, and (8.86%, 0.00%) for number of inflorescences/plant, respectively. The disorders in plant growth due to the presence of cadmium can be attributed to reduced photosynthetic activities, plant mineral nutrition, and reduced activity of some enzymes. The results showed that the Red fountain plant has the potential to grow well in cadmium contaminated soil because there were no sever phytotoxic symptoms and had medium tolerance to cadmium. In this respect, Shah *et al*. (2017) on *Tagetes erecta* plant reported that cadmium accumulation had not noticeable passive effect on morphological plant growth.

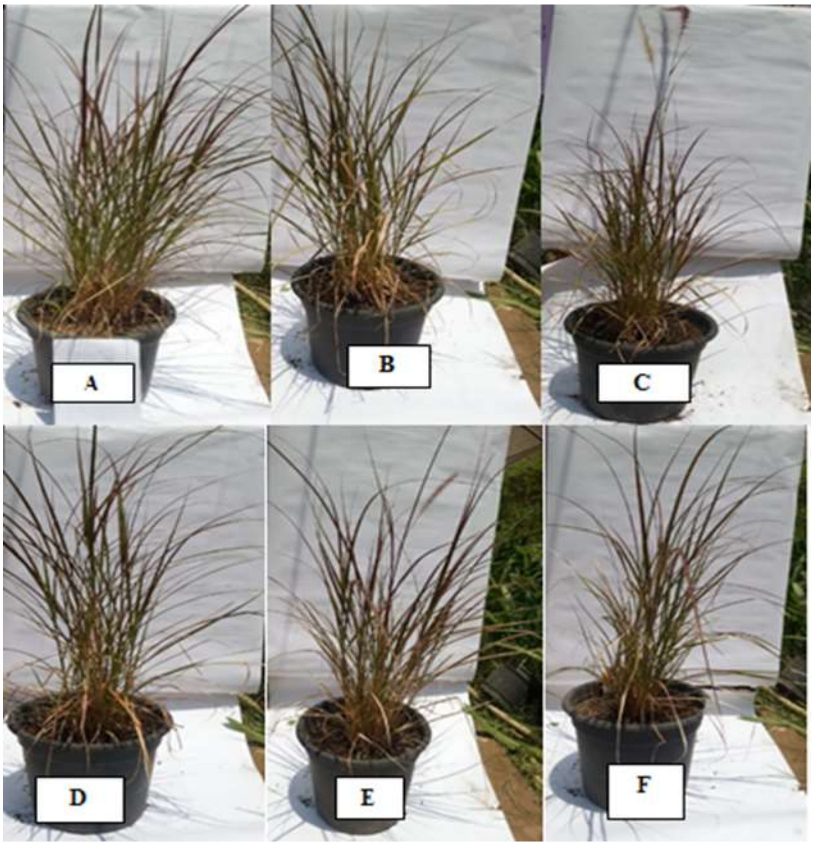

Figure 1. *Pennisetum setaceum* cv. rubrum plants grown in uncontaminated soil (A, B and C) and Cd-contaminated soil (D, E and F) during the second season.

A. Control B. CA1=Citric acid at the dose of 1 mmol/kg C. CA2= Citric acid at the dose of 2 mmol/kg
D. Cd= Cadmium at the dose of 100 mg/kg E. Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg
F. Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg.

Cd accumulation in herb and root

The success of phytoextraction depends on the higher accumulation of the metal, high biomass production and plant resistance (Fan *et al.*, 2011). Results in (Tab. 3) showed that cadmium soil application caused significantly increased in Cd content in herb and roots of *Pennisetum setaceum* cv. rubrum plants compared to plants grown in unpolluted soil.

In both seasons, cadmium content in the herb was higher than in the roots. These results may be due to the ability of the root to transport cadmium to the herb. This is considered a good sign of Red Fountain plant tolerance to cadmium because, despite the movement of cadmium from the root to the herb and accumulation inside it, the plant had good growth parameters as previously shown in (Tab. 2).

Table 2. Effect of citric acid on vegetative characters of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil during 2017 and 2018 seasons

Characters	Plant length [cm]	Herb fresh weight [g]	Herb dry weight [g]	No. of tillers/plant	No. of inflorescences /plant	Root fresh weight [g]	Root dry weight [g]
First season, 2017							
Control	90.00±2.00	114.60±1.15	36.17±0.29	16.67±1.15	15.00±1.00	67.17±2.75	18.20±0.53
CA1	61.00±2.65	82.43±0.55	29.23±2.16	22.67±0.58	10.00±1.00	56.47±2.35	15.67±0.58
CA2	60.67±0.58	69.37±1.60	21.83±0.29	19.67±0.58	6.00±1.00	58.73±2.19	16.87±0.35
Cd	69.00±2.65	91.30±0.40	34.33±1.53	18.67±1.53	13.67±1.53	45.80±2.55	14.33±0.58
Cd-CA1	66.33±3.21	79.80±3.60	28.27±2.00	19.33±0.58	13.33±0.58	36.87±1.55	12.67±0.58
Cd-CA2	61.33±1.53	77.27±2.70	25.67±2.08	19.67±0.58	13.00±1.00	52.07±2.63	15.30±0.61
L.S.D. (0.05)	4.40	3.74	2.99	1.62	2.02	4.63	0.99
Second season, 2018							
Control	93.67±3.21	121.90±1.82	37.17±0.76	14.00±1.00	14.00±1.00	78.30±1.76	20.63±0.55
CA1	62.00±1.73	82.17±0.76	28.50±2.18	20.33±1.53	10.33±0.58	51.87±3.67	15.40±0.66
CA2	61.33±2.31	67.90±3.44	24.33±1.33	19.00±1.00	5.67±0.58	68.80±5.11	18.50±0.46
Cd	66.33±1.15	89.63±1.50	31.67±1.53	20.00±1.00	14.00±1.73	47.00±4.01	14.77±0.68
Cd-CA1	65.00±2.00	77.13±3.60	24.83±2.02	19.67±0.58	13.67±1.15	38.80±1.80	13.50±0.50
Cd-CA2	62.00±3.46	73.93±2.50	24.67±0.58	19.33±1.53	11.67±1.15	48.60±1.22	15.00±0.50
L.S.D. (0.05)	4.18	4.62	2.95	2.17	1.87	6.39	0.95

Control CA1=Citric acid at the dose of 1 mmol/kg CA2= Citric acid at the dose of 2 mmol/kg Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

Table 3. Effect of citric acid on Cd accumulation in the herb and root of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil during 2017 and 2018 seasons

Characters	Cd in herb [mg/kg DW]		Cd in root [mg/kg DW]		(TF) Translocation factor	
Treatments	First season	Second season	First season	Second season	First season	Second season
Control	2.80±0.35	3.20±0.30	3.50±0.36	4.43±0.21	0.81±0.19	0.72±0.04
CA1	4.80±0.20	4.90±0.44	4.26±0.66	3.30±0.10	1.15±0.22	1.49±0.18
CA2	3.33±0.49	4.93±0.46	4.00±0.35	3.70±0.20	0.84±0.18	1.34±0.16
Cd	26.40±0.53	44.27±2.30	16.00±1.44	31.87±1.10	1.66±0.12	1.39±0.05
Cd-CA1	65.83±3.31	67.63±2.85	51.40±0.85	42.13±1.36	1.28±0.08	1.61±0.03
Cd-CA2	62.73±2.65	74.07±2.05	43.33±1.86	63.27±2.61	1.45±0.01	1.17±0.02
L.S.D. (0.05)	3.50	2.87	2.05	2.35	0.27	0.19

TF= Metal concentration in shoots/ Metal concentration in roots. Control CA1=Citric acid at the dose of 1 mmol/kg CA2= Citric acid at the dose of 2 mmol/kg Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

Using citric acid with cadmium application caused a significant increase in cadmium content in the plant tissue compared with plants untreated by citric. The treatment of citric acid at the dose of 1 and 2 mmol/kg combined with cadmium increased the Cd content in the herb (149.3, 137.6%) and (52.76, 67.31%) in the first and second seasons, respectively. Whereas it caused Cd content increment in the root (221.25, 170.81%) and (32.19, 98.52%) in the first and second seasons, respectively, compared to the treatment of Cadmium alone.

The results in (Tab. 3) showed that the translocation factor of Cd was ranged between 0.81 and 1.66 in the first season and between 0.72 and 1.61 in the second season. According to (Mirecki *et al*., 2015) who reported that, if the value of translocation factor bigger than one, the plants have accumulated elements; the translocation factor around 1 pointed out that the plants were not affected by the elements. Despite the medium negative impact on the growth of the Red fountain plant treated with citric acid and grown in Cd-contaminated soil, the positive side was the possibility of using citric acid to reduce the soil contaminated with cadmium. This may be due to the ability of citric acid to facilitate cadmium transport into the root hairs and increasing the translocation to the aerial part of the plant.

Similar results were reported by (Han *et al.,* 2018) they found that Cd and Zn accumulation increased in the vegetative growth of *Iris halophila* plant due to the treatment of 2 mmol kg^{-1} citric acid. Also, (Yang *et al.*, 2011) found that, in the presence of multiple metal contaminants (Cd and Pb), citric acid treatment can significantly enhance the Cd (10-30%) and Pb (10-20%) accumulation in *Solanum nigrum* when compared to control. This enhancement resulted from the increment solubility of the heavy metals in the soil. It could be interpreted as the role of citric acid in defense-related enzymes.

Cd hyper accumulation, defined as the accumulation and tolerance of Cd up to 100 mg/kg in shoots by a plant (Tian *et al*., 2011). Thus from previous results, Red fountain may be a useful for phytoremediation because it had a large biomass and accumulated moderate concentration of Cd in its leaves up to 74.07 mg/kg dry weight in the presence of citric acid at the dose of 2 mmol/kg in the second season

Total Chlorophylls content [mg/g FW]

Data in (Fig. 2) showed that control plants gave the highest content of total chlorophylls (2.028 and 1.058 mg/g FW in the first and second seasons, respectively) followed by the treatment of cadmium at the dose of 100 mg/kg (1.467 and 0.795 mg/g FW in the first and second seasons, respectively). On the other hand, data in (Fig. 2) showed that total chlorophylls content decreased with citric acid application. The treatment of citric acid at the dose of 2 mmol/ kg gave the lowest values of total chlorophylls (0.750 and 0.540 mg/g FW in the first and second seasons, respectively). This result was in agreement with the result reported by (Shah *et al*., 2017) on *Tagetes erecta*.

Eissa and Abeed (2019) mentioned that the high concentration of cadmium in the soil reduced chlorophyll synthesis in *Atriplex lentiformis* plants. Cadmium led to Fe deficiency and reducing the activity of enzymes participates in CO_2 fixation (Asati *et al*., 2016). Kabata-Pendias and Pendias (1992) found that the amount of chlorophyll can be considered an indicator of the Cd concentration in plant tissues. They estimated the phytotoxic concentrations of Cd to be 5–10 mg/kg in sensitive plant species, whereas the range of 10–20 mg/kg as critical Cd levels. The Cd toxicity results in disturbed stomatal conductance and the electron transport system.

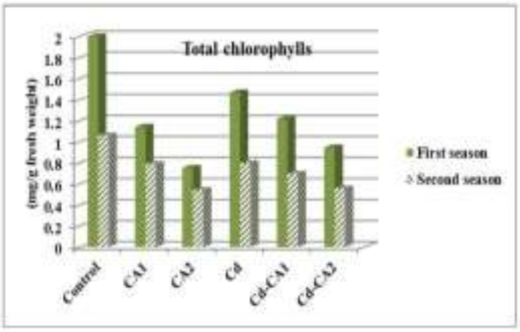

Figure 2. Effect of citric acid on total chlorophylls [mg/g fresh weight] of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil during 2017 and 2018 seasons

Control CA1=Citric acid at the dose of 1 mmol/kg CA2= Citric acid at the dose of 2 mmol/kg Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

Total Carbohydrates % and phenols content [mg GA/g DW]

Generally, it was observed that the total carbohydrates values of control plants were the highest compared with all other treatments (Tab. 4). The obtained results revealed that cadmium in soil (100 mg/kg) caused decrease in carbohydrates percent in Red fountain grass and this result was in agreement with the result reported by (Shah *et al*. 2017) on *Tagetes erecta.* They found that the total sugar, reducing sugar and starch continuously increased till mid-level (Cd-18 mg/kg), and then continuously decreased at higher concentrations (Cd-30 mg/kg). Data represented in (Tab. 4) showed that total carbohydrates percent significantly decreased with citric acid application in the plants grown in unpolluted or polluted soil. In both seasons, the lowest values of carbohydrates percent was observed in plants grown in Cd -polluted soil and treated with citric acid 2 mmol/ kg.

The phenols content of the plants grown in the Cd-polluted soil exceeded those plants grown in unpolluted one. Phenols content increased significantly by 72.5% and 73.5% in plants grown in Cd- polluted soil (100 mg/kg of soil) and without citric acid treatment, whereas the enhancement rate of phenols content reached to 102.3% and 93.0% with citric acid application at the dose of 1 mmol/kg compared to control plants in both seasons, respectively. These results were in agreement with that mentioned by (Eissa and Abeed, 2019) they added seven different levels of Cd ranged between (0, and 240 mg / kg of soil) to *Atriplex lentiformis* plants. Their results revealed that the plants achieved different defense mechanisms included enhancing the synthesizing of phenols. Cd at the dose of 240 mg significantly enhanced phenols leaves content. Marquez-Garcıa *et al.* (2012) reported that phenolic compounds have a serious function in the metabolism of *Erica andevalensis* to overcome the cadmium contaminated soils. Also, phenolic compounds are considered plant defense mechanisms as most of them have potent antioxidant properties, neutralizing the effects of oxidative stress (Kulbat, 2016).

Table 4. Effect of citric acid on total carbohydrates [%] and phenols [mg GAE /g DW] contents in dry herb of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil during 2017 and 2018 seasons

Characters	Total carbohydrates [%]		Phenols [mg GAE/ g DW]	
Treatments	First season	Second season	First season	Second season
Control	44.00±1.00	58.00±2.65	7.33±0.58	7.80±0.72
CA1	38.33±1.53	50.33±1.53	10.58±0.48	11.37±0.40
CA2	36.33±2.08	48.67±1.15	8.67±0.68	9.84±0.15
Cd	42.67±2.52	56.67±1.53	12.65±0.56	13.54±0.50
Cd-CA1	37.33±1.53	46.00±2.65	14.83±0.76	15.06±0.11
Cd-CA2	33.67±1.53	37.67±1.53	13.72±1.02	14.53±0.65
L.S.D. (0.05)	3.09	2.65	1.34	0.94

Control CA1=Citric acid at the dose of 1 mmol/kg CA2= Citric acid at the dose of 2 mmol/kg Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

N, P and K %

It is clear from the results in (Tab. 5) that, the highest N, P and K percentages (1.50, 0.23 and 1.65%, respectively) in the dry herb of *Pennisetum setaceum* cv. rubrum resulted from healthy control plants followed by the treatment of cadmium alone with an insignificant decrease in P % and a significant decreases in N and K %. In most cases, citric acid concentrations applied either alone or in combination with cadmium caused significant decreases in N, P and K % in the dry herb. These results were in harmony with (González-Rodríguez *et al*., 2010) who found that *Pennisetum setaceum* had low nitrogen content with value 0.7%. Whereas, (Williams and Black, 1996) found that the nitrogen content of *Pennisetum setaceum* leaves ranged from 1.61 to 2.86 % and photosynthetic capacity was highly affected by the leaf nitrogen content. Cadmium interferes with the uptake of several elements (Ca, Mg, P, and K) and can affect the plasma membrane permeability; Cd could slow down the nitrate reductase activity, which in turn reduces the absorption and transport of nitrate from roots to shoots. Asati *et al*., 2016 found that cadmium reduces the ATPase activity of the plasma membrane fraction of roots.

Catalase activity [U/g FW]

Catalase activity as affected by citric acid concentration and cadmium accumulation in soil presented in (Tab. 5) showed that the highest catalase activity (0.93 U/g fresh weights) was recorded with the treatment of citric acid 1 mmol/kg combined with cadmium. The data showed that the control plants had the lowest catalase activity (0.75 U/g fresh weights).

Table 5. Effect of citric acid on N, P and K contents in dry herb and Catalase activity in fresh herb of *Pennisetum setaceum* cv. rubrum plants grown in Cd-contaminated soil during the second season

Treatments	N%	P%	K%	Catalase U/g
Control	1.50±0.20	0.23±0.01	1.65±0.15	0.75±0.10
CA1	1.10±0.10	0.22±0.03	1.03±0.05	0.82±0.07
CA2	0.40±0.05	0.20±0.00	1.03±0.06	0.79±0.03
Cd	1.20±0.10	0.22±0.02	1.25±0.05	0.86±0.05
Cd-CA1	0.63±0.06	0.20±0.03	1.03±0.05	0.93±0.01
Cd-CA2	0.50±0.09	0.18±0.02	0.85±0.17	0.88±0.03
L.S.D. (0.05)	0.18	0.03	0.21	0.097

Control CA1=Citric acid at the dose of 1 mmol/kg CA2= Citric acid at the dose of 2 mmol/kg Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

The exposure to toxic concentrations of heavy metals can defeat the systems protecting the plants and building more reactive oxygen species (ROS) in plants. The balance between ROS production and activities of the antioxidative enzyme (such as catalase) determines whether the damage will occur or not (Anjum et. al., 2011). Kamel et al. (2012) reported that heavy metals induce oxidative stress in plants and increase the antioxidant enzymes such as catalase which act as an adaptive response to heavy metals.

Soil content of Cd [mg/kg]

At the beginning of the experiment, the soil of control treatment contained 0.1 mg/kg of cadmium, whereas, at the end of the experiment, cadmium reduced to 0.06 and 0.04 mg/kg in the first and second seasons, respectively as shown in (Tab. 6). That means the *Pennisetum setaceum* plants had extracted 40 and 60% of the initial cadmium concentration in the soil. The extraction percent by *Pennisetum* plants was increased in the Cd-polluted soil. The percent of extractable cadmium by the Red fountain plant reached 96.95% and 97.05% of the initial cadmium concentration in the polluted soil (100mg Cd/kg soil) with the treatment of citric acid at the dose of 1 mmol/kg in the first and second seasons, respectively. Coakley et. al., 2019 evaluated *Impatiens glandulifera* plants for cadmium phytoremediation. The results revealed that the total Cd removed by all parts of the plant exceeds that of the soil.

Table 6. Determination of Cd [mg/kg] in soil at the end of the experiment during 2017 and 2018 seasons

Treatments	First season	Second season
Control	0.06±0.01	0.04±0.02
Cd	14.78±1.55	25.21±0.53
Cd-C1	3.05±0.23	2.95±0.20
Cd-C2	22.90±1.01	22.26±0.38
L.S.D. (0.05)	1.70	0.57

Cd= Cadmium at the dose of 100 mg/kg Cd-CA1= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 1 mmol/kg Cd-CA2= Cadmium at the dose of 100 mg/kg + Citric acid at the dose of 2 mmol/kg

CONCLUSION

Acquired results revealed that *Pennisetum setaceum* plants are capable of extracting cadmium from the soil, then accumulating it in their tissues. Also, citric acid had the possibility to reduce the soil contaminated with cadmium, due to facilitate and increase cadmium transport from the roots to the shoots of fast-growing, high biomass-producing plants such as Red fountain grass.

From the obtained results it could be concluded that *Pennisetum setaceum* plant had the potential to grow well in Cd-contaminated soil (100 mg/kg) and can be effectively used for phytoremediation.

REFERENCES

ABOU EL-ANWAR, E.A. - SAMY, Y.M. - SALMAN, S.A. 2018. Heavy metals hazard in Rosetta Branch sediments. *Egypt. J. Mater. Environ. Sci.*, vol. *9*, no. 7, pp. 2142-2152.

AL NAGGAR, Y. - NAIEM, E. - MOHAMED, M. - GIESY, J.P. - SEIF, A. 2014. Metals in agricultural soils and plants in Egypt. *Toxicological and Environmental chemistry*, vol. *96*, no. 5, pp. 730-742. DOI:10.1080/02772248.2014.984496

ANJUM S. A. - XIE X. - WANG L. - SALEEM M. F. - MAN C. - LEI W. 2011.Morphological, physiological and biochemical responses of plants to drought stress. *African Journal of Agricultural Research*, vol. *6*, no.9, pp. 2026-2032.

ASATI, A. - PICHHODE, M. - NIKHIL, K. 2016. Effect of heavy metals on plants: An Overview. *International Journal of Application or Innovation in Engineering & Management*, vol.*5*, no. 3, pp. 1-11.

BEYER, W.F. - FRIDOVICH, I. 1987. Assaying for superoxide dismutase activity: Some large consequences of minor changes in conditions. *Analytical Biochemistry*, vol.*161*, no. 2, pp. 559-566.

COAKLEY, S. - CAHILL, G. - ENRIGHT, A. - O'ROURKE, B. - PETTI, C. 2019. Cadmium Hyperaccumulation and Translocation in *Impatiens glandulifera*: from Foe to Friend? *Sustainability*, vol.*11*, *no.18*, pp.1-17. DOI: 10.3390/su11185018.

COTTENIE, A. - VERLOO, M. - KIEKENS, L. - VELGHE, G. - CAMERLYNCK, R. 1982. Chemical Analysis of Plant and Soil. *Laboratory of Analytical and Agrochemistry*, *State University*. Ghent, Belgium. PP. 100 – 129.

EISSA, M. A. - ABEED, A.H.A. 2019. Growth and biochemical changes in quail bush (*Atriplex lentiformis* (Torr.) S. Wats) under Cd stress. *Environmental Science and Pollution Research*, vol.*26*, no. 1, pp. 628–635

FAN, K.C. – HIS, H.C. – CHEN, C.W. – LEE, H.L. – HSEU, Z.Y. 2011. Cadmium accumulation and tolerance of mahogany (*Swietenia macrophylla*) seedlings for phytoextraction applications. *Journal of Environmental Management*, vol. *92*, no10, pp. 2818-22. DOI:10.1016/j.jenvman.2011.06.032

FARID, M. - ALI, S. - SAEED, R. - RIZWAN, M. - BUKHARI, S.A.H. - ABBASI, G.H. - HUSSAIN, A. - ALI, B. - ZAMIR, M.S. - AHMAD, I. 2019. Combined application of citric acid and 5-aminolevulinic acid improved biomass, photosynthesis, and gas exchange attributes of sunflower (*Helianthus annuus* L.) grown on chromium contaminated soil. *International Journal of Phytoremediation*, vol.*18*, pp. 1-8. DOI:10.1080/ 15226514. 2018.1556595

GONZÁLEZ-RODRÍGUEZ, A. - BARUCH, Z. - PALOMO, D. - CRUZ-TRUJILLO, G. - JIMÉNEZ, S. - MORALES, D. 2010. Eco physiology of the invader *Pennisetum setaceum* and three native grasses in the Canary Islands. *Acta cologica.*, vol.*36*, pp. 248-254.

HAN, Y. - ZHANG, L. - GU, J. - ZHAO, J. - FU, J. 2018. Citric acid and EDTA on the growth, photosynthetic properties and heavy metal accumulation of *Iris halophile* Pall. cultivated in Pb mine tailings. *International Biodeterioration and Biodegradation*, vol.*128*, pp. 15-21.

HASHIM, T.A. - ABBAS, H.H. - FARID, I. M. - EL-HUSSEINY, O. H. M. - ABBAS, M. H. H. 2017. Accumulation of some heavy metals in plants and soils adjacent to Cairo – Alexandria Agricultural Highway. *Egypt. J. Soil Sci.*, vol.*57*, no. 2, pp. 215-232.

HELRICH, K.1990. *Official methods of analysis of the Association of Official Analytical Chemists*, 15^{th} Ed., Association of Official Analytical Chemists, Arlington, Virginia, pp. 84-85.

HERBERT, D. – PHILIPS, P.J. –STRANGE, R.E. 1971. Determination of Total Carbohydrates. *Methods in Microbiology*, vol. *5* pp.204-244.

JACKSON, M.I. 1973. *Soil Chemical Analysis*. Pientic-Hall, Inc.Limited, New York.

JOSANGELA DO CARMO - DE ARAUJO, T. - WILLIAMS C. DO NASCIMENTO. 2010. Phytoextraction of lead from soil from a battery recycling site: The use of citric acid and NTA. *Water Air Soil Pollution*, vol.*211*, 113-120.

KABATA-PENDIAS, A. – PENDIAS, H. 1992. *Trace Elements in Soils and Plants*. Boca Raton, FLA: CRC Press. pp. 158-172.

KAMEL, M.M. - KHATER, H.A. - SOLIMAN, A. SH. – SHANAN, N. T. 2012. Effect of microbial inoculation and EDTA on the physiology and phytoremediation efficiency of *Delonix regia* plants growing in polluted soil. *American-Eurasian J. Agric. & Environ. Sci.*, vol.*12*, no. 6, pp. 719-728.

KAMEL, M.M. - SAKR, W.R. 2009. Response of *Senna occidentalis*, Link plants to fertilization as well as citric acid and their role in remediating soil polluted with Cu and Pb. *World Journal of Agricultural Sciences*, vol.*5*, no. 6, pp. 784-798.

KING, E. J. 1951. *Micro-Analysis in Medical Biochemistry*, 2nd Ed., Churchill Publishing Co., London. 260p.

KLUTE, A. 1986. *Methods of Soil Analysis*. Part 1" Physical and Mineralogical Methods". 2nd Ed., Argon. Madison, Wisconsin, USA.

KULBAT, K. 2016. The role of phenolic compounds in plant resistance. *Biotechnol Food Sci.*, vol.*80*, no. 2, pp. 97-108.

MARQUEZ-GARCIA, B. - FERNANDEZ-RECAMALES, M.A. - CORDOBA, F. 2012. Effects of cadmium on phenolic composition and antioxidant activities of *Erica andevalensis*. *Journal of Botany*, vol.*12*, 1-6. https://doi.org/10.1155/2012/936950.

MIRECKI, N. - AGIČ, R. - ŠUNIĆ, L. - MILENKOVIĆ, L. - ILIĆ, Z.S. 2015. Transfer factor as indicator of heavy metals content in plants. *Fresenius Environmental Bulletin*, vol.*24*, no. 11c, pp. 4212-4219.

MORAN, R. 1982. Formula for determination of chlorophyll pigments extracted with N.N. dimethyl formamide. *Plant Physiol.*, vol.*69*, pp.1371-1381.

PREGL, P. 1945. *Quantitative Organic Microanalysis*. 4th Ed., Churchill Publ. Co. London. 200.

SHAH, K. - MANKAD, A.U. - REDDY, M.N. 2017. Cadmium accumulation and its effects on growth and biochemical parameters in *Tagetes erecta* L. *Journal of Pharmacognosy and Phytochemistry,* vol.*6* , no. 3, pp. 111-115.

SINGLETON, V.L. - ROSSI, J.A. 1965. Colorimetry of total phenolic with phosphomolybdic phosphotungstic acid reagents. *In American Journal of Enology and Viticulture*, vol. *16*, pp. 144–158.

SNEDECOR, G.W. - COCHRAN, W.C. 1982. *Statistical Methods*. 6th Ed., Iowa State Univ., Press Ames, Iowa, USA: 953.

TIAN, S. - LU, L. - LABAVITCH, J. - YANG, X. HE, Z. - HU, H. - SARANGI, R. - NEWVILLE, M. - COMMISSO, J. - BROWN, P. 2011. Cellular sequestration of cadmium in the hyperaccumulator plant species *Sedum alfredii*. *Plant Physiology*, vol.*157*, no. 4, pp. 1914–1925.

WAHID, A. - ARSHAD, M. - FAROOQ, M. 2009. Cadmium phytotoxicity: Responses, mechanisms, and mitigation strategies. Organic Farming Pest control and Remediation of soil pollutants. *Sustainable Agriculture Reviews*, no. 1, pp. 371-403.

WILLIAMS, D. G. - BLACK, R. A. 1996. Effects of nutrient amendment and environment on growth and gas exchange for introduced *Pennisetum setaceum* in Hawaii. *Canadian Journal of Botany*, vol.*74*, no. 2, pp. 268-275.

YANG, G. - MIAO, C. - XIA, J. - LUO, C. - MAO, L. - ZHOU, P. - SHI, W. 2011. Effect of citric acid on phytoextraction and antioxidative defense in *Solanum nigrum* L.as a hyperaccumulator under Cd and Pb combined pollution. *Environmental Earth Sciences*, 65, pp. 1923-1932.

قدرة عشب النافورة الحمراء على النمو في التربة الملوثة بالكادميوم

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تم اجراء تجربة اصص خلال الموسمين المتعاقبين 2017 و 2018 في المزرعة التجريبية بكلية الزراعة ، جامعة القاهرة ، الجيزة ، مصر. الهدف من هذه الدراسة هو تحديد مقدرة نبات *Pennisetum setaceum* cv. rubrum على النمو في التربة الملوثة بالكادميوم (100 ملجم / كجم). وأيضا ، التحقق من تأثير استخدام حامض الستريك (1 و 2 مللي مول / كجم) على قدرة النبات على استخلاص الكادميوم من التربة الملوثة . أوضحت النتائج أن وجود الكادميوم (100 ملجم / كجم) في التربة تسبب في انخفاض طفيف في نمو الأعشاب مقارنة بالتربة غير الملوثة. كانت معدلات الانخفاض في كلا الموسمين 20.33 و 26.47 ٪ للوزن الطازج للعشب ، 5.08 و 14.79 ٪ للوزن الجاف للعشب ، 8.86 و 0.0 ٪ لعدد النورات / النبات ، على التوالي. ادت المعاملة بحامض الستريك (1 مللي مول / كجم) الى زيادة محتوى الكادميوم في العشب بنسبة 149.3 ، 137.5٪ ، و داخل جذر النبات بنسبة 221.25 ، و 170.81٪ في كلا الموسمين ، على التوالي مقارنةً بالكنترول. و تسببت هذه المعاملة أيضا في ارتفاع محتوى الفينولات في العشب إلى 102.3 و 93.0 ٪ مقارنة مع الكنترول في كلا الموسمين ، على التوالي. أظهرت النتائج أن عامل الانتقال (TF) كانت قيمته أكبر من واحد صحيح مما يعني قدرة النباتات على تجميع الكادميوم في أنسجتها ؛ ومع ذلك ، تحمل النبات النمو في التربة الملوثة بالكادميوم مصحوبة بمحتوى عالٍ من الفينولات ونشاط إنزيم الكتاليز. يمكن أن نستخلص من ذلك أن *P. setaceum* cv. rubrum يعد من النباتات الواعدة للمعالجة النباتية للملوثات.