

## Influence of using treated wastewater with moringa seeds and chitosan on wheat growth and uptake of nutrients and heavy metals

Sayed A. Abdeen

Soils and Water Department, Faculty of Agriculture, Al-Azhar Univ., Cairo, Egypt

[sayed\\_abdrahman@yahoo.com](mailto:sayed_abdrahman@yahoo.com)

### ABSTRACT

The present study was carried out to treat wastewater and investigate its impact on wheat growth, uptake of nutrients and heavy metals. To achieve this task, two experiments were conducted at the Faculty of Agriculture, Al-Azhar University-Cairo-Egypt, during winter season of 2014. The 1<sup>st</sup> (lab experiment) was for treatment wastewater by using moringa seeds and chitosan with different doses (0.5, 1 and 2g/l as powder and 1, 2 and 3 % as extract). The 2<sup>nd</sup> (pot experiment) was carried out to investigate the effect of the previously treated wastewater on wheat growth, uptake of nutrients and heavy metals.

The main results from the first experiment showed that heavy metals concentration in wastewater decreased significantly by increasing the addition of the added materials. Moringa seeds and chitosan as powders were more effective than its extracts. Results from the second experiment indicated that there was an increase in both fresh and dry weight of wheat plant irrigated with treated wastewater compared with control. Also, the highest values were observed at the highest dose of chitosan extract (3%). On the other hand, the uptake of N, P and K by wheat plants increased significantly by increasing the added materials. Moreover, the heavy metals uptake by wheat plant ( $\mu\text{g}/\text{plant}$ ) was decreased by addition of the treated materials.

**Key words:** Wastewater, wheat growth, nutrients, heavy metals uptake.

### INTRODUCTION

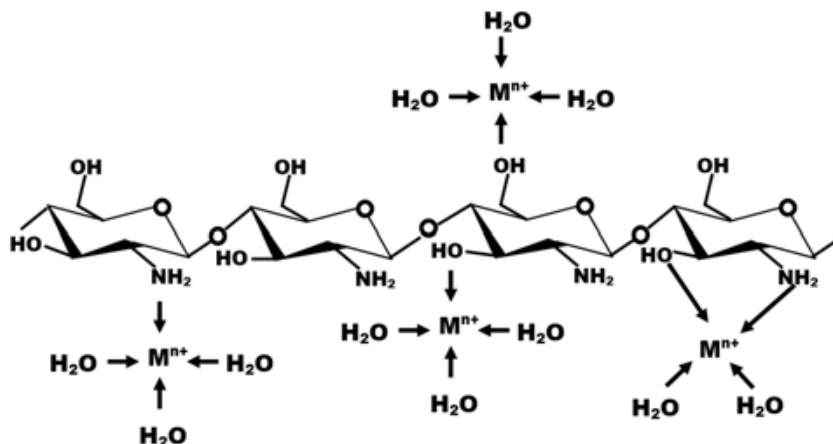
Heavy metals have come to be one of the main contaminants of wastewater in recent years. They arise from different sources/activities carried out by industries. Water pollution has contributed to negative environmental and human health impacts (Sharma *et al.*, 2006). Chemical precipitation, physical treatment such as ion exchange and adsorption are some of the processes that have been reported to be the most effective in the removal of heavy metals (Singh *et al.*, 2001).

The previous studies reported that *Moringa oleifera* seeds (MOS) are effective sorbents for removal of heavy metals and volatile organic compounds in the aqueous system (Sharma *et al.*, 2006). Biological materials such as MOS have been recognized as cheap substitutes for wastewater treatment and are safe for human health (Hsu *et al.*, 2006). Moringa polyelectrolytes contain potential nitrogen and oxygen ligands that are known to have an affinity for coordinating to heavy metals. The chemical composition of MOS indicated that this plant has more powerful antioxidants which reduce the heavy metal toxicity (Bhatti *et al.*, (2007).

On the other hand, chitosan is a biopolymer that can be derived from shrimp shells that previously have displayed a high capacity to fix a great variety of metals. The amine groups present in chitosan biopolymer are weak enough to deprotonate water according to the following expression  $\text{Chitosan}-\text{NH}_2+\text{H}_2\text{O}=\text{Chitosan}-\text{NH}_3^++\text{OH}^-$  (Guibal 1999).

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Chitosan biopolymer has a possess electro- static properties to adsorb metals according to ion exchange mechanism. The adsorption properties of chitosan biopolymer are attributed to its functional group including increase of hydrophilicity by hydroxyl groups of the polymer. This helps to enhance the diffusion of polymer networks and allows adsorbing metals from aqueous solutions. Amino groups of chitosan are very important for adsorption processes compared to hydroxyl group, for which degree of deacetylation is an important parameter to evaluate the quality of the chitosan biopolymer (Crini 2005) .The mechanism of metal ions removal by chitosan is shown in Figure (1).



**Fig. 1. Mechanism of removal of metal ions by chitosan,** Haritma and Gazala (2016).

In this study, adsorption of heavy metals from wastewater was investigated by using *Moringa oleifera* seeds (MOS) and chitosan(Ch) as natural materials, then the impact of this treated wastewater on wheat growth, uptake of nutrients and heavy metals were investigated.

### MATERIALS AND METHODS

Two experiments were conducted at the Faculty of Agriculture, Al-Azhar University-Cairo- Egypt, during winter season of 2014. The 1<sup>st</sup> (lab experiment) was for treating wastewater by using moringa seeds and chitosan with different doses i.e. 0.5, 1 and 2g/l as powder and 1, 2 and 3 % as extract. The studied materials prepared as follows:

#### *Moringa oleifera* Seeds

Dried seeds of *Moringa* were obtained from The National Research Centre (NRC) in Dokki-Giza,Cairo. Seed coats were removed and the white kernel was crushed to a powder, using an electric grinder and sieved through 0.8 mm sieve. The powder was used directly to water samples as a powder with doses 0.5, 1 and 2 g/l., and agitated by shaker for 5 min. Also, moringa seed extract (MSE) was prepared by addition of moringa seed powder on distilled water (2%) as described by Ndabigengesere *et al.*, (1995). The suspension was given vigorous shaking for 5 min and filtered before using directly. The extract was used with doses 1, 2 and 3 %.

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### Chitosan

It was obtained from the Egyptian Petroleum research institute, it was prepared from shrimp shells with 110 kDa of molecular weight and 85 % of degree of deacetylation (Hussien *et al.*, 2012). Chitosan was crushed to a powder, using an electric grinder and sieved through 0.8 mm sieve. The powder was used directly to water samples as a powder with doses 0.5, 1 and 2 g/l., and agitated by shaker for 5 min. Also, chitosan extract (2%) was prepared by adding chitosan powder in 0.1 M HCl (Divakaran and Pillai 2002). The suspension was given vigorous shaking for 5 min and filtered before the using directly. The extract was applied with doses 1, 2 and 3 %. After treating wastewater samples by the studied materials, the resulted samples were filtered by passing throw sandy filter (2- 0.2 mm) to be used in the cultivation process. Analysis of moringa seeds and chitosan are presented in Table (1).

**Table (1). Analysis of moringa seeds and chitosan**

parameter	%			mg.kg <sup>-1</sup>			Heavy metals mg.kg <sup>-1</sup>						
	N	P	K	Ca	Mg	Na	Fe	Zn	Mn	Cu	Cd	Pb	Ni
Moringa seeds	6.2	1.35	1.15	123	46.5	643	27.8	18.5	13.6	14.1	0.3	0.1	0.6
Chitosan	7.1	2.2	1.90	365	433	2100	67	4.3	28.7	34.3	0.0	0.3	16.5

The 2<sup>nd</sup> (pot experiment) was carried out to investigate the effect of the treated wastewater by the investigated aforementioned application rates on wheat growth, uptake of nutrients and heavy metals by wheat plant (*Triticum aestivum*, cv. Gemmiza). The experiment was laid out in a randomized complete block design in factorial arrangement with three replications. Ten seeds of wheat were planted in every pot and moisture content of pots was kept approximately at field capacity. The cultivated plants were fertilized with ammonium sulfate, super phosphate and K- sulfate according to the general recommendation dose of Ministry of Agriculture. After 45 days from planting, wheat shoots of each treatment were cut just one cm above the soil surface and prepared for analysis.

The characteristics of the investigated soil, i.e. Particle size distribution, Soil pH, EC, soluble cations and anions, OM, CEC, available N, P, K were determined according to Page *et al.*, (1982) and Klute 1986). Soil samples were extracted for available heavy metals in DTPA extract according to Lindsay and Norvell (1978).

Wheat plants were washed with distilled water, dried at 70°C and ground, then the samples were wet digested using both HClO<sub>4</sub> and H<sub>2</sub>SO<sub>4</sub> acid mixture to determine NPK and heavy metals in the plant digest according to Cottenie *et al.*, (1982).

Wastewater samples used in this study were collected from El-Rahawy drain, Giza, Egypt. Some drops of toluene were added to suppress microbial activity. Wastewater analysis pH, EC, cations and anions, and heavy metals were determined as described by Standard Method (APHA 1995). All extracts of heavy metals were determined using Perkin Elmer Inductivity Coupled Spectrophotometer Plasma (ICP 400).

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The statistical analysis for all data, which obtained was carried out and differences between means were calculated using L.S.D test according to Steel and Torrie (1980). Analysis of the investigated soil and water samples are presented in in Table (2).

**Table (2). Analysis of the investigated soil and water.**

Parameter	Soil	Wastewater
	Value	Value
<b>Some physical properties</b>		
Sand %	75.6	-
Silt %	17.5	-
Clay %	6.9	-
Textural class	Loamy sand	-
Field capacity (F C) %	14.00	-
<b>Chemical properties</b>		
pH	7.78	7.52
EC dS/m (1:2.5 soil extract)	1.39	0.98
OM g.kg <sup>-1</sup>	3.8	-
CEC cmol <sub>c</sub> kg <sup>-1</sup> soil	3.20	-
<b>Soluble ions cmol<sub>c</sub> kg<sup>-1</sup>soil</b>		<b>Soluble ions cmol<sub>c</sub> L<sup>-1</sup></b>
Ca <sup>++</sup>	0.80	2.8
Mg <sup>++</sup>	0.55	1.65
Na <sup>+</sup>	1.63	5.5
K <sup>+</sup>	0.20	0.9
CO <sub>3</sub> <sup>=</sup>	0.00	0.0
HCO <sub>3</sub> <sup>=</sup>	0.78	3.2
Cl <sup>-</sup>	1.88	4.5
SO <sub>4</sub> <sup>=</sup>	0.52	3.15
SAR	-	3.69
<b>Available macro nutrients mg/kg</b>		<b>Macronutrients mg/L</b>
N	42.00	17.9
P	13.40	9.3
K	67.90	1.43
<b>Available heavy metals mg/kg</b>		<b>Heavy metals mg/L</b>
Fe	5.6	3.87
Zn	1.6	2.60
Mn	1.9	1.40
Cu	0.8	0.76
Pb	0.40	1.90
Cd	0.0	0.05
Ni	0.0	0.31

## RESULTS AND DISCUSSION

### Heavy metals (Fe, Zn, Mn and Cu) as affected by treated materials

Moringa seeds and chitosan act as natural adsorbent to remove the heavy metals from water samples. It was obvious from Table (3) that heavy metals concentration in wastewater (Fe, Zn, Mn and Cu) decreased significantly by increasing the addition of the investigated treated materials i.e. moringa seeds and chitosan either powder or extract. Moringa seeds powder was more effective than seeds extract. The highest removal

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percentages were observed at the highest addition rate (2g/l). This is may be due to the adsorptive capacity of *Moringa oleifera* seeds which contain considerable quantities of cellulosic interlinked with lignin in their structure.

**Table (3). Heavy metals (Fe,Zn,Mn and Cu) as affected by treated materials.**

Materials	Application Rates	Parameters								
		Fe		Zn		Mn		Cu		
		Con. mg/L	Removal** %	mg/L	Removal %	mg/L	Removal %	mg/L	Removal %	
Permissible limits for irrigation water*		5.00	-	2.00	-	0.20	-	0.20	-	
Control		3.87	0.00	2.60	0.0	1.40	0.0	0.76	0.0	
Moringa seeds	Powder (g/l)	0.5	3.55	8.27	2.20	15.38	1.10	21.43	0.50	34.21
		1	2.83	26.87	1.76	32.31	0.76	45.71	0.50	34.21
		2	2.10	45.74	1.40	46.15	0.50	64.29	0.40	47.37
	mean		2.83	26.96	1.79	31.28	0.79	43.81	0.47	38.60
	Extract (%)	1	3.60	6.98	2.50	3.85	1.24	11.43	0.60	21.05
		2	3.10	19.90	2.20	15.38	0.90	35.71	0.50	34.21
		3	2.60	32.82	1.90	26.92	0.65	53.57	0.44	42.11
	mean		3.10	19.90	2.10	15.38	0.93	33.57	0.51	32.46
	Chitosan	Powder (g/l)	0.5	3.20	17.31	2.20	15.38	1.32	5.71	0.45
1			2.40	37.98	1.85	28.85	0.92	34.29	0.34	55.26
2			2.00	48.32	1.35	48.08	0.40	71.43	0.16	78.95
mean		2.53	34.54	1.80	30.77	0.88	37.14	0.32	58.33	
Extract (%)		1	3.40	12.14	2.10	19.23	1.20	14.29	0.46	39.47
		2	2.85	26.36	1.53	41.15	1.20	14.29	0.32	57.89
		3	1.80	53.49	1.00	61.54	0.90	35.71	0.18	76.32
mean		2.68	30.66	1.54	40.64	1.10	21.43	0.32	57.89	
LSD at 5%		A	0.19	-	0.20	-	0.15	-	0.13	-
	B	0.18	-	0.19	-	0.14	-	0.12	-	
	A B	N. S	-	N. S	-	N. S	-	N. S	-	

A = Materials and B= Application rates. \*Permissible limits (Egyptian Chemical Standard 1982) and Radojevic, M. and Bashkin, V. (2010)

\*\*Removal percentage % = 100 x [1 - (heavy metal in treatment/ heavy metal in control)]

Lignin is a complex biopolymeric heterogeneous molecule which is endowed with many different functional groups, such as methoxy, hydroxyl-aliphatic, carboxyl and phenolic Shin and Rowell (2005). The highest removal percentage of Fe, Zn, Mn and Cu by using 2% extract of MOS were 45.74, 46.15, 64.29 and 47.37%, respectively, compared with that observed at 3% MOS extract which recorded 32.82, 26.92, 53.57 and 42.11 %, respectively. This result agreed with that given by Sajidu *et al.*, (2005) and Subramanium *et al.* (2011) who reported that Moringa seeds have been able to remove heavy metals from water samples. Adsorption describes attachment of ions and molecules from seed protein by means of specific mechanisms. Heavy metals and solids that have high charges than Moringa colloidal surface will remove high percentage of metals. The mechanism that brings about the adsorption of heavy metals is through the positive metal ion that forms a bridge among the anionic polyelectrolyte and negatively charged protein functional groups on the colloidal particle surface (Benes and Steinnes 1995). Generally, the affinity of moringa seeds for cations adsorbed selectivity in the following order: Mn >Cu >Zn >Fe. Also, chitosan as powder was more effective than extract for removal of Mn and Cu. The highest percentage (71.43 and 78.95 %, respectively) at 2g /l was recorded, compared with that observed at 3% chitosan extract which recorded 35.71 and 76.32%, respectively. While, the highest recorded percentage of Fe and Zn (53.49 and 61.54%, respectively) was at 3% of chitosan extract compared with that obtained by 2g/l chitosan powder which recorded 48.32 and 48.08%,

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respectively. This indicated that the metal uptake capacity of the adsorbent increases with the increase in dosage as the number of active sites available for metal increases with the increase in the amount of adsorbent. These findings were in agreement with Guibal (2004).

Generally, the affinity of chitosan powder for cations selectivity was in the following order: Cu > Mn > Fe > Zn. While, the affinity of chitosan extract for cations selectivity was Cu > Zn > Fe > Mn. In this concern, Varma *et al.*, (2004) reported that chitosan has a tendency to bind/chelate with heavy metal like cadmium, copper, lead etc. The amine groups present along the polymer chain are strongly active with metal ions. The affinity of chitosan for cations adsorbed on film presents selectivity in the following order: Cu > Zn > Cd > Ni (Rinaudo 2006).

#### Heavy metals (Cd, Pb and Ni) as affected by the treated materials

The removal percentage of Cd, Pb, and Ni metals from the contaminated wastewater by the studied sorbents is given in Table (4). The highest rates of chitosan powder or extract removed 100% of Cd. Whereas, moringa powder removed 80% and the extract removed 40% of Cd. Similarly, Mataka *et al.*, (2010) found that the increasing of moringa seeds powder dosage increased Cd removal percentage from about 20% to about 58%. Also, in the present study removal of Lead (Pb) decreased with the increase of the investigated treated materials and chitosan was more effective than moringa seeds. The lowest value (1.11 mg/L) at 2g /l chitosan powder was recorded compared with control which recorded (1.9 mg/L).

**Table (4): Heavy metals (Cd, Pb and Ni) concentration in wastewater as affected by treated materials.**

Materials		Application Rates	Parameters					
			Cd		Pb		Ni	
			mg/L	Removal** %	mg/L	Removal %	mg/L	Removal %
Permissible limits for irrigation water *			0.01	-	5.00	-	0.20	-
Control			0.05	100.0	1.90	100.0	0.31	100.0
Moringa seeds	Powder (g/l)	0.5	0.05	0.00	1.70	10.53	0.28	9.68
		1	0.05	0.00	1.54	18.95	0.22	29.03
		2	0.03	40.0	1.35	28.95	0.15	51.62
	Mean		0.04	13.33	1.53	19.48	0.22	30.11
	Extract (%)	1	0.04	20.0	1.90	0.00	0.26	16.13
		2	0.03	40.0	1.80	5.26	0.20	35.48
		3	0.01	80.0	1.45	23.68	0.18	41.94
Mean		0.03	46.67	1.72	9.65	0.21	31.18	
Chitosan	Powder (g/l)	0.5	0.04	20.0	1.60	15.79	0.20	35.48
		1	0.04	20.0	1.34	29.47	0.13	58.06
		2	0.00	100.0	1.11	41.58	0.13	58.06
	Mean		0.03	46.67	1.35	28.95	0.15	50.53
	Extract (%)	1	0.03	40.0	1.76	7.37	0.22	29.03
		2	0.02	60.0	1.44	24.21	0.18	41.94
		3	0.00	100.0	1.20	36.84	0.18	41.94
Mean		0.02	66.67	1.47	22.81	0.19	37.64	
LSD at 5%		A	0.007	-	0.15	-	N. S	-
		B	0.006	-	0.14	-	0.05	-
		A B	0.020	-	N. S	-	N. S	-

A = Materials and B = Application rates. \*Permissible limits (Egyptian Chemical Standard 1982) and Radojevic, M. and Bashkin, V. (2010)

\*\*Removal percentage % = 100 x [1 - (heavy metal in treatment / heavy metal in control)]

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Also, the highest removal percentage (41.58%) was recorded at 2 g/l of chitosan powder. On the other hand, chitosan or moringa seeds powder and extract removed 41%, 29% and 36.84%, 23.68% of Pb, respectively. These results are in agreement with those obtained by Subramaniam *et al.*, (2011) who found that moringa seeds reduced Pb concentration up to 70%. Affonso *et al.*, (2012) reported that the metal removal efficiency of Cd and Pb using *Moringa oleifera* seeds, increased with the mass increase, a fact that could be attributed to the increased surface area for adsorption and to the availability of active adsorption sites. Olayinka *et al.*, (2016) found that the maximum removal of Pb and Ni were 82.1% and 68.28%, respectively by using chitosan powder. This was due to the adsorption properties of chitosan biopolymer which was attributed to its functional groups including the increase of hydrophilicity by hydroxyl groups of the polymer. The highest removal percentage of Ni recorded in the current study was 58.06 %, 51.62 % and 41.94, 41.94 at the highest doses of chitosan, moringa seeds powder and extract, respectively. It was observed that the chitosan selectivity for Cadmium, Nickel and Lead in the studied wastewater was as follows: C d > N i > P b .

#### Effect of using the treated wastewater on total biomass, NP and K uptake of wheat plant

The total biomass (fresh and dry weight) of wheat plant is presented in Table (5). Generally, it was obvious that there was an increase in both fresh and dry weights of wheat plant irrigated with the treated wastewater compared with control. In this respect, Sheikh and AL-Malki (2011) indicated that chitosan enhanced bean shoot and root length, fresh and dry weights of shoots, root and leaf area as well as the level of chlorophylls. The effect of chitosan on plant growth may be attributed to an increase in the key enzyme activities of nitrogen metabolism (nitrate reductase, glutamine synthetase and protease) and increased photosynthesis which enhanced the plant growth (Mondal *et al.*, 2012).

**Table (5) Effect of using the treated wastewater on fresh, dry weight, NP and K uptake by wheat plant.**

Materials		Application Rates	FW g/pot	DW g/pot	Uptake mg/plant		
					N	P	K
		Control	36.4	7.10	10.60	1.92	5.54
Moringa seeds	Powder(g/l)	0.5	36.4	7.20	10.80	2.02	5.76
		1	37.2	7.23	10.85	2.02	6.29
		2	37.5	7.22	11.19	2.17	7.94
	mean		37.03	7.22	10.95	2.07	6.66
	Extract (%)	1	37.4	7.11	10.24	1.92	7.11
		2	37.9	7.31	11.78	1.97	8.77
		3	38.2	7.40	12.14	2.15	8.88
mean		37.83	7.27	11.39	2.01	8.25	
Chitosan	Powder(g/l)	0.5	36.7	7.11	10.24	1.99	6.40
		1	37.2	7.25	10.73	2.18	6.53
		2	37.7	7.42	11.87	2.23	8.16
	mean		37.20	7.26	10.95	2.13	7.03
	Extract (%)	1	37.3	7.33	11.14	2.13	8.06
		2	38.5	7.50	11.70	2.25	9.00
		3	39.2	7.72	12.82	2.47	9.50
mean		38.33	7.52	11.89	2.28	8.85	
LSD at 5%		A	N. S	0.10	0.14	0.11	0.19
		B	N. S	0.09	0.13	0.10	0.90
		A B	N. S	0.19	0.27	N. S	1.81

A = Materials and B= Application rates

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The highest recorded values of fresh and dry weights were 39.2 and 7.72 g/pot, respectively compared with control which recorded 36.4 and 7.1 g/pot, respectively. The order of increasing fresh and dry weights were: chitosan extract > moringa seeds extract > chitosan powder > moringa seeds powder. Hilal *et al.*, (2006) found that chitosan was able to enhance the growth of many crops.

Uptake values of macronutrients (NP and K) by wheat plant are shown in Table (5). Generally, NP and K uptake by wheat plant increased significantly by increasing the addition rates of chitosan and moringa seeds. It can be seen that the values of N, P and K uptake ranged from 10.24 - 12.82, 1.92 - 2.47 and 5.54 - 9.50 mg/plant, respectively. Generally, the lowest values were observed at control, while the highest values were observed at the highest dose of chitosan extract (3%). This is may be attributed to the increase in the availability and uptake of water and essential nutrients through adjusting cell osmotic pressure and reducing the accumulation of harmful free radicals by increasing antioxidants and enzyme activities (Guan *et al.*, 2009). Also, the order of increasing uptake of macronutrients were: chitosan extract > moringa seeds extract > chitosan powder > moringa seeds powder. The underlying mechanisms for this plant growth promoting action may be attributed to effects on plant physiological processes such as nutrient uptake, cell elongation, cell division, enzymatic activation and protein synthesis (Amin *et al.*, 2007).

#### Heavy metals uptake by wheat plant as affected by the treated wastewater

It was obvious from Table (6) that heavy metals uptake by wheat plant ( $\mu\text{g/plant}$ ) was decreased significantly by treated materials and its application rates. Concentrations of Fe, Zn Mn, and Cu ranged from 30.05-42.48, 6.22-10.80, 7.71-11.93 and 1.27-2.25  $\mu\text{g/plant}$ , respectively; compared with control which recorded 47.93, 11.86, 17.54 and 2.75  $\mu\text{g/plant}$ . The lowest values for Fe, Zn and Mn were observed at 2g/l of chitosan powder. However, it was found that for Cu the lowest value was observed at 3% chitosan extract. Also, the order of decreasing uptake of Fe and Zn were: chitosan powder > chitosan extract > moringa seeds powder > moringa seeds extract. While, the order of decreasing Mn and Cu were: chitosan extract > chitosan powder > moringa seeds extract > moringa seeds powder. In this concern, Henry (2002) showed that *Moringa oleifera* press cake can be used for heavy metals removal from water and as coagulant to remove turbidity from water, but more work need to be done to improve the quality of *Moringa oleifera* press cake as bio sorbent and coagulant.

Also, the data showed that concentration of Cd uptake by wheat plant had not been detected in the highest doses of moringa seeds and chitosan extracts. The concentration of Pb uptake by wheat plant ranged from 0.52 – 0.62  $\mu\text{g/plant}$ . The lowest value was observed at 2g/l chitosan powder, while that of Ni ranged from 0.15 – 0.28  $\mu\text{g/plant}$ . The lowest value was observed at 3% chitosan extract. Generally, the order of decreased uptake of Cd, Pb and Ni were: chitosan extract > moringa seeds extract > chitosan powder > moringa seeds powder. These data are in agreement with those obtained by Olayinka *et al.*, (2016) and Mataka *et al.*, (2010) who found that heavy metals uptake reduced by addition of chitosan and moringa seeds as resulted in reducing in the content of plant.



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**Table (6). Effect of the treated wastewater on heavy metals uptake ( $\mu\text{g}/\text{plant}$ ) by wheat plant.**

Materials		Application Rates	Heavy metals uptake( $\mu\text{g}/\text{plant}$ )						
			Fe	Zn	Mn	Cu	Cd	Pb	Ni
		Control	47.93	11.86	17.54	2.75	0.040	0.85	0.28
Moringa seeds	Powder(g/l)	0.5	46.08	12.24	17.78	2.66	0.040	0.86	0.29
		1	45.69	10.48	14.10	2.53	0.020	0.65	0.26
		2	39.40	8.89	11.93	2.25	0.007	0.58	0.22
	mean		43.72	10.54	14.60	2.48	0.022	0.70	0.26
	Extract (%)	1	46.78	11.38	13.03	2.28	0.007	0.57	0.21
		2	46.49	11.70	13.16	2.19	0.007	0.55	0.28
		3	42.48	10.80	11.32	1.96	0.000	0.52	0.28
mean		45.25	11.29	12.50	2.14	0.005	0.55	0.26	
Chitosan	Powder(g/l)	0.5	32.00	9.74	10.10	2.03	0.014	0.64	0.25
		1	30.74	7.40	8.77	1.85	0.007	0.65	0.25
		2	30.05	6.22	7.71	1.78	0.000	0.56	0.22
	mean		30.93	7.79	8.86	1.89	0.007	0.62	0.24
	Extract (%)	1	40.24	9.16	8.65	1.55	0.007	0.59	0.24
		2	34.20	9.00	8.25	1.50	0.008	0.60	0.15
		3	31.81	7.49	8.49	1.27	0.000	0.62	0.15
mean		35.42	8.55	8.46	1.44	0.005	0.60	0.18	
LSD at 5%		A	1.55	0.16	0.76	0.14	0.009	N. S	0.024
		B	1.53	0.14	0.74	0.12	0.007	0.083	0.022
		A B	3.08	0.32	1.51	0.27	N. S	N. S	0.048

A = Materials and B= Application rates

### Conclusion

In conclusion, the study has shown that *Moringa oleifera* seed powder and chitosan act as natural adsorbents to remove the heavy metals from wastewater. The maximum removal of metals ions i.e. Fe, Zn, Mn, Cu, Cd, Pb and Ni from wastewater were: 53.49, 61.54, 64.29, 76.32, 100, 41.58 and 58.06%, respectively. Generally, the affinity of moringa seeds for cations adsorbed selectivity was in the following order: Cd > Mn > Cu > Ni > Zn > Fe > Pb. While, the affinity of chitosan for cations adsorbed selectivity was in the following order: Cd > Cu > Ni > Zn > Mn > Fe > Pb. On the other hand, it was obvious that there was an increase in both fresh and dry weight of wheat plant irrigated with treated wastewater compared with control. The order of increasing fresh and dry weights were: chitosan extract > moringa seeds extract > chitosan powder > moringa seeds powder. Also, the uptake of N, P and K by wheat increased by increasing the added removal materials. Heavy metals uptake by wheat plant ( $\mu\text{g}/\text{plant}$ ) i.e. Fe, Zn, Mn Cu, Cd, Pb and Ni decreased significantly by addition of treated materials. Finally, the present results indicated that the treated wastewater by using the investigated materials can be used for irrigation of wheat plant, but it must be considering the impact of long term application of treated wastewater on human health and environment in terms of heavy metals and pathogens.

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### تأثير استخدام مياه الصرف الصحي المعالجة ببذور المورنجا والشيتوزان على نمو القمح وامتصاص العناصر المغذية والمعادن الثقيلة

سيد عبد الرحمن عابدين

قسم الاراضى والمياه – كلية الزراعة - جامعة الازهر بالقاهرة – مصر

#### المستخلص

أجريت هذه الدراسة بهدف معالجة مياه الصرف الصحي ودراسة تأثيرها على نمو نبات القمح وامتصاصه للعناصر المغذية والعناصر الثقيلة. ولتحقيق هذا الهدف تم إجراء تجربتين في كلية الزراعة - جامعة الأزهر - القاهرة - مصر خلال موسم الشتاء من عام 2014. تم زراعة نبات القمح كمؤشر لتحقيق الغرض من الدراسة. كانت التجربة الأولى (تجربة معملية) بهدف معالجة مياه الصرف الصحي باستخدام بذور المورنجا والشيتوزان بمعدلات مختلفة (0.5 ، 1 ، 2 جم/ لتر كمسحوق و 1 ، 2 ، 3٪ كمستخلص). تم إجراء التجربة الثانية (تجربة اصص) لدراسة تأثير مياه الصرف المعالجة ببذور المورنجا والشيتوزان على نمو نبات القمح ، وامتصاص العناصر المغذية والعناصر الثقيلة .

اظهرت نتائج الدراسة ما يلي :

#### التجربة الاولى

- انخفاض معنوي لتركيز المعادن الثقيلة (الحديد، الزنك، المنجنيز، النحاس، كاديوم ، الرصاص والنيكل) في مياه الصرف الصحي مع زيادة إضافة المواد المعالجة سواء على هيئة مسحوق او مستخلص ، حيث كان لمسحوق بذور المورنجا أكثر فعالية في ازالة العناصر الثقيلة اكثر من مستخلص البذور. عموماً، كانت قدرة المورنجا على ازالة العناصر الثقيلة مرتبة كالتالى : كاديوم < منجنيز < نحاس < نيكل < الزنك < الحديد < رصاص.
- أيضاً، كان لمسحوق الشيتوزان فاعلية اكثر من المستخلص في ازالة المنجنيز والنحاس. بينما كان لمستخلص الشيتوزان قدرة اعلى في ازالة الحديد والزنك والكاديوم والرصاص والنيكل. عموماً لوحظ أن قدرة الشيتوزان على ازالة العناصر الثقيلة مرتبة كالتالى : كاديوم < نحاس < نيكل < زنك < منجنيز < حديد < رصاص.

#### التجربة الثانية :

- ادى استخدام مياه الصرف المعالجة تحسناً معنويًا في نمو النبات وامتصاص المغذيات مقارنة بالكنترول حيث لوحظ ان هناك زيادة في الوزن الطازج والجاف لنباتات القمح المروية بمياه الصرف المعالجة مقارنة بالكنترول. حيث كانت الزيادة في الأوزان الطازجة والجافة طبقاً لترتيب المعاملات كالتالى : مستخلص الشيتوزان < مستخلص بذور المورنجا < مسحوق الشيتوزان < مسحوق بذور المورنجا.
- زيادة معنوية في امتصاص النبات للعناصر المغذية ( نيتروجين – فوسفور – بوتاسيوم) حيث كانت اعلى قيم عند اعلى معدلات من المواد المدروسة وخصوصاً الشيتوزان (3٪).
- على الجانب الاخر لوحظ انخفاض معنوي في امتصاص المعادن الثقيلة ( حديد – منجنيز – نحاس- زنك- كاديوم - رصاص – نيكل) بواسطة نبات القمح (ميكروجرام / نبات) بزيادة اضافة المواد المعالجة. حيث كان ترتيب انخفاض الحديد والزنك وفقاً للمعاملات كالتالى : مسحوق الشيتوزان < مستخلص الشيتوزان < مسحوق بذور المورنجا < مستخلص بذور المورنجا. في حين كان ترتيب تناقص المنجنيز والنحاس كالتالى: مستخلص الشيتوزان < مسحوق الشيتوزان < مستخلص بذور المورنجا < مسحوق بذور المورنجا. بينما كان انخفاض الكاديوم، الرصاص و النيكل كالتالى: مستخلص الشيتوزان < مستخلص بذور المورنجا < مسحوق الشيتوزان < مسحوق بذور المورنجا.
- عموماً اظهرت الدراسة ان لبذور المورنجا والشيتوزان قدرة عالية على ازالة العناصر الثقيلة من مياه الصرف مما يؤدي الى تحسن في خصائص هذه المياه فى الاستخدام الزراعى ، ولكن يجب الاستمرار فى عمل دراسات على تأثير هذه النوعية من المياه على المدى الطويل على صحة الإنسان والبيئة من حيث المعادن الثقيلة ومسببات الأمراض.