Journal of Applied Botany and Food Quality 87, 196 - 205 (2014), DOI:10.5073/JABFQ.2014.087.028

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Use of sesame and sorghum crops to verify the remediation of contaminated sewaged soils

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(Received November 2, 2013)

Summary

At the time being, treated sewage effluent is repeatedly used in farming, particularly in newly reclaimed sandy soils, as it fortify their content of nutrients and organic matter as time goes on, despite there are significant concerns about the long-term accumulation of PTEs in the soil ecosystem. Pot experiment was conducted to verify the effectiveness of bioremediation of two sewaged soils, highly and marginally contaminated, with either canola (*Brassica napus*) or Indian mustard (*Brassica juncea*) plants in the absence and presence of AM inoculation, by growing sesame (*Sesamum indicum*) and sorghum (*Sorghum bicolor*) plants. Results indicated that the vegetative parameters of both test crops did not show any sign of adverse symptoms when grown in either bioremediated sewaged soils. Phytoremediation with either canola or Indian mustard obviously reduced Cu and Zn contents in both sesame and sorghum plants and completely removed Ni from soil. Both sesame and sorghum plants grown in both bioremediated sewaged soils contained lower PTEs, however, at varying degrees. Zinc equivalent value in marginally and highly decontaminated soils after harvesting either sorghum or sesame obviously decreased compared to control soil. After phytoremediation, the dehydrogenase activity increased in all soils. Generally, the efficiency of phytoremediation with canola far exceeded that with Indian mustard, despite both were effective tools.

Abbreviation: AM: Arbiscular Mycorrhiza, PTEs: Potential toxic elements, CFU: Colony forming unit

Introduction

Reuse of sewage effluent in farming is well recognized as a workable environmental prospect. Abouzeina et al. (2013) stated that Cairo is now served by six large wastewater treatment plants which produce significant quantities of sewage effluent. The preferred option is to use it in farming, particularly on newly reclaimed desert land which is inherently deficient in organic matter, nutrients and trace elements. Sewage effluent has been used in farming, since distant past, however at a restricted magnitude. Early in 1531 sewage farming started in Germany and thereafter progressively distributed in other parts in the world. He added that in Egypt the first sewage farm was installed in 1930 at El-Gabal Al-Asfer near Cairo and sewage farms gradually disseminated later on in other regions. At the time being, sewage effluent represents one of the substantial natural water resources in Egypt that should be well managed, rather than discarded. Sewage effluent, however, is contaminated with enormous amounts of contaminants, for the most part PTEs that mount up in sewaged soils and could effortlessly invade the food chain, with subsequent decisive adverse environmental and public health impacts. Indubitably safe and sustainable reuse of sewage effluent in farming necessitates the removal of soil contaminants.

Abouziena et al. (2013) stated that Salt et al. (1995) highlighted the concepts of phytoremediation using certain hyperaccumulator plants capable to remove PTEs from soil. Metal hyperaccumulators are commonly defined as plant species that can approximately accumulate 100-fold higher metal levels than common plant species (Brooks, 1998). The advantages of bioremediation represent maintaining characters of the soil ecosystem after phytoremediation, visually being unobtrusive and offers the possibility the bio-recovery of PTEs (Sun et al., 2006; Fawzi, 2008). Recently it was noticed that coupling phytoremediation with other techniques such as microbial, physical and/or chemical treatment could be viable in many cases. Throughout the world, over 400 species of terrestrial plants have been identified as hyperaccumulators of various PTEs (Baker et al., 2000; Sun et al., 2006).

The possibility to clean heavy metal contaminated soils with hyperaccumulator plants has shown great potential. At present, phytoremediation of metals may be approaching commercialization (Midarkodi, 2007). Two of the most recently studied species used in phytoremediation applications as hyper-accumulators plants are canola (Marchiol et al., 2004; Podar et al., 2004; Ashour et al., 2006) and Indian mustard (Banuelos et al., 2005; Hamlina and Barkera, 2006; Chauhan and Rai, 2009; Panwar et al., 2011; Abouziena et al., 2012). In a pot experiment, Purakayastha et al. (2008) grew five different species of Indian mustard as possible accumulators of PTEs in soils sewaged for more than two decades. In general, they found that the highest concentration and uptake of PTEs was observed in shoots compared to roots or seeds of the different species of Indian mustard. Correlation analysis showed negative relation between total uptake of PTEs and their available forms as well as the total concentrations of PTEs. In addition, root length emerged as a powerful parameter indicating the uptake of PTEs by Indian mustard compared to other root parameters.

Microbial remediation of sewaged soils is a well welcomed technique as microorganisms perform an urgent mode in the devastation of PTEs in agricultural ecosystems, and several researchers had been attempted to develop means of using microorganisms and microbial enzymes for decontaminate sewaged soils. However, as far as PTEs are not biologically degradable and persist in indefinitely soil ecosystem, Glick (1995) stated that once they accumulated in the soil, they inversely affect the microbial compositions, particularly plant growth promoting rhizobacteria in the rhizosphere, and their metabolic activities. He added that the elevated concentration of PTEs in soils and their uptake adversely affect plant growth, symbiosis and consequently crop yields through disrupting photosynthesis, inactivating respiration, protein synthesis and/or carbohydrate metabolism.

Phytoremediation efficiency is often limited by many factors such as bioavailable forms of contaminants in soil, plant-root development, and the level of tolerance of the plant to each particular contaminant (Pilon-Smits, 2005). Given the fact that there were a large number of variables in the current experiment, it is not possible to distill this information and come up with one ideal set of conditions for all PTEs contaminant phytoremediation experiments. These variables include plant type, soil composition, endogenous bacteria, concentration and range of the contaminants, temperature range, and type and physiological state of bio-additives. This complexity notwithstanding, careful examination of the data suggests that certain con-

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siderations might facilitate the phytoremediation of soil contaminants. RAMASAMY et al. (2000) concluded that the application of chelate ion exchange resin, cation – exchange resin, ferrous sulfate and silica gel, lime, gypsum, and naturally occurring clay minerals such as bentonite, zeolite and green sand had been found to immobilize PTEs in contaminated soil ecosystems.

In this context, the present work had been planned, to investigate the growth of sesame and sorghum in two sewaged soils, highly and marginally contaminated, previously phytoremediated with either canola or Indian mustard in the absence and presence of AM inoculation.

Materials and methods

Soil Sampling and soil preparation

In this work, two surface sewaged soil samples collected from Abu-Rawash farm previously bioremediated by both canola and Indian mustard were used. Some chemical and physical properties of the original soils and the kinetic study applied were previously documented in SOAD EL-ASHERY et al. (2011) as shown in Tab. 1.

The concentrations of PTEs, the distribution of these pollutants after remediation and Zn equivalent as a parameter used for contamination or remediation were documented in SABER et al. (2012). The two groups of soils i.e. after Indian mustard and canola were separately remixed well and repacked to be used in this work.

Experimental

In a completely randomized plot design, greenhouse experiments were conducted with three replicates and each replicate consists of five pots for growing sesame (*Sesamum indicum* L) and sorghum (*Sorghum bicolor* L. Moench) as PTEs sensitive (SUMATHI and UMAGOWRIEB, 2012) and tolerant (MARCHIOL et al., 2007) crops, respectively, in the two remediated sewaged soils after either Indian mustard or canola during 2011 winter season and summer 2012.

Seeds from each tested plants were sown on 6th June 2011 at the rate of 8 seeds in plastic pots (30 x 30 cm) filled with 4 kg of either the two contaminated sewaged soils. Fifty percent of the cultivated pots were sown with seeds inoculated with 20 ml of a suspension of mycorrhyzal conidia (10^6 CFU) while the rest were left without mycorrhyzal inoculation. It is worthy to mention that control pots represented by the prior bioremediated soils without neither cultivation nor inoculation with AM to understand rizosphere effects on the rate of PTEs uptake under such conditions. All treatments including control did not receive any mineral fertilizers to eliminate reactions could be take place between PTEs and applied fertilizers. The moisture content of all pots was initially adjusted to 50 % of the soil water holding capacity and kept at the same level during the experimental periods. After three weeks, the seedlings were thinned to 5 and 3 plants per pot. The plants from each pot were harvested at maturity seed production stage (harvest). Soil samples were collected at initially and maturity stages, analyzed for their PTEs concentrations and dehydrogenase activity. Plant and its parts dry weight as well as yield and its components characteristics were determined before their content of PTEs was estimated.

Methods

Microbiological Methods

Mycorrhyzal (AM) conidia: AM fungi spores were extracted from soil by wet sieving and sucrose density gradient centrifugation (SYLIVA et al., 1993).

Dehydrogenase activity: Dehydrogenase activity in the sewaged soils was estimated initially and at harvest according the method given by SKUJINS (1973) as µl H_2 g^{-1} soil 24^{-h}.

Chemical Methods

Potential toxic elements (PTEs): Potential toxic elements in soil and plant parts were extracted by the ammonium bicarbonate DPTA as described by COTTENIE et al. (1982) and analyzed using Atomic Absorption according to A.O.A.C (1984).

Soil quality criteria: Zn equivalent model was numerically expressed for the levels of PTEs toxicity in used soils according to (CHUMBLEY, 1971) using the following equation:

Zn concentration*1+ Cu concentration *2+Ni concentration*8.

Statistical Analysis: All data were processed by Microsoft Excel (MICROSOFT, 2000) and the regression of linear and nonlinear and other statistical analyses were conducted using the programs of SAS Release 6.12 (SAS INSTITUTE, 1996).

Results

Plant dry weight and seed yield/plant: Dry matter biomass and seed yield of sorghum and sesame plants grown in highly or marginally contaminated sewaged soils previously bioremediated by either canola or Indian mustard hyperaccumulators in the presence or absent of AM inoculation are given in Tab. 2 and 3. Results indicated higher dry weight of roots, straw, panicle or capsules and total plant dry weight for both sorghum and sesame plants grown in contaminated sewaged soils previously bioremediated with canola plants compared to those grown in contaminated sewaged soil previously bioremediated with Indian mustard either in the presence or absence of AM inoculation.

Sorghum plants performance: The dry matter of sorghum plants and its parts were higher when grown in the highly than in the marginally contaminated sewaged soil (Tab. 2). The role of AM inoculation in increasing the dry matter of sorghum plants was more noticeable in the soil previously bioremediated with Indian mustard than that in the soil previously bioremediated with canola plants both in marginally or highly contaminated sewaged soil.

Tab. 1: Total PTE contents in Abou-Rawash farm soils * (ppm oven dry basis)

Soil depth (cm)	Sewage farming started since	Landscape	Cd	Cu	Fe	Mn	Pb	Zn	Ni	Zn equivalent
0-30	30	Sandy soil was cultivated with artichoke	-	35.65	596.0	45.19	57.7	400.6	18.0	633.9
30-60			-	33.50	850.0	73.34	63.2	94.55	14.0	287.6
0-30	30	Loamy sand soil was cultivated with artichoke	14.95	111.7	758.5	160.0	48.2	73.05	18.8	458.4
30-60			13.45	112.1	761.0	227.0	49.3	75.05	10.0	389.2

Tab. 2: Plant criteria of sorghum grown in remediated sewaged soil by Indian mustard or canola in the presences and absence of inoculation with AM.

Treatments		Dry weight (g plant^{-1})				Grains yield (g plant^{-1})
		Root	Straw	Panicle	Plant	
Marginally contaminated sewaged soil	Mustard	4.8	44.5	18.0	67.3	123.04
	Mustard+ AM	4.8	67.0	21.0	98.0	152.6
Highly contaminated sewaged soil	Mustard	10.0	110.3	17.0	137.3	121.7
	Mustard+AM	15.3	112.2	23.5	151.0	164.0
Mean		8.7	83.5	19.9	113.4	129.0
Marginally contaminated sewaged soil	Canola	5.8	87.7	18.5	84.3	138.1
	Canola+ AM	12.8	115.9	28.3	157.0	210.0
Highly contaminated sewaged soil	Canola	14.7	128.5	22.0	165.2	160.0
	Canola + AM	12.5	97.6	17.9	128.0	228.7
Mean		11.5	107.4	21.7	133.6	163.2
LSD at 0.05		1.6	30.2	4.0	12.9	21.4

Inoculation of sorghum plants with AM caused a significant increase in plant dry weight reaching 45.6 % and 10.0 % in the marginally and highly contaminated sewaged soil, respectively relative to that without AM inoculation.

After bioremediation with canola in association with AM inoculation as shown in Tab. 2, the dry weight of sorghum plants increased by 52.2 % in the marginally contaminated sewaged soil compared to that without AM inoculation. It was noticed that seed yield of sorghum plants was increased by 23.6 % and 35.5 % in the marginally and highly contaminated sewaged soil respectively; this result might be ascribed to the higher concentrations of nutrients in highly contaminated soil (Soad El-Ashry et al. (2011). The highest grain yield of sorghum plant, which is our main concern, was harvested from the marginally contaminated sewaged soil previously bioremediated with canola without AM inoculation, which was insignificantly different from plants grown in highly contaminated sewaged soil previously bioremediated with canola with AM inoculation (Tab. 1).

Sesame plants performance: Results in Tab. 3 indicated that phytoremediation of the sewaged soils either by canola or Indian mustard exhibited a significant effect on sesame plant criteria at harvest. In most cases, inoculation with AM increased dry weight of sesame plant and its parts in both marginally and highly contaminated sewaged soils.

Data indicated that growing sesame plants in the marginally contaminated sewaged soil previously bioremediated with Indian mustard in association with AM inoculation increased their yield more than three folds in terms of the dry matter of both total and separate plant organs weights. These increments were also recorded when sesame was grown in the highly contaminated sewaged soil, but at a lower level (Tab. 3).

Seed yield of sesame plants grown after bioremediation with canola plants without AM inoculation was lower by 191.7 and 29.6 % than when associated with AM inoculation in the marginally and highly contaminated sewaged soils, respectively.

PTEs content in sorghum and sesame plants and their parts

Sorghum plants: Sorghum plant and its parts had more or less similar Cu contents whether grown in the marginally or highly contaminated sewaged soil previously bioremediated with either of the hyper-accumulators canola or Indian mustard in the presence or absent of AM inoculation (Tab. 4). The lowest value of Cu in sorghum

Tab. 3: Plant criteria of sesame grown in remediated sewaged soil by Indian mustard or canola in the presences and absence of inoculation with AM.

Treatments		Dry weight (g plant^{-1})				Seeds weight (g plant^{-1})
		Root	Straw	Capsules	Plant	
Marginally contaminated sewaged soil	Mustard	1.2	25.0	1.1	27.3	1.5
	Mustard+ AM	7.6	78.9	11.6	98.1	12.5
Highly contaminated sewaged soil	Mustard	7.0	55.2	8.9	71.1	19.8
	Mustard+ AM	5.1	60.0	15.0	80.1	33.5
Mean		5.2	54.8	6.7	66.7	14.3
Marginally contaminated sewaged soil	Canola	2.0	19.2	1.2	22.4	1.2
	Canola + AM	5.4	27.2	2.5	11.7	3.5
Highly contaminated sewaged soil	Canola	5.0	30.0	2.9	37.9	4.4
	Canola + AM	3.4	48.4	3.1	54.9	5.7
Mean		4.0	31.2	2.4	31.7	3.7
LSD at 0.05		1.1	7.6	1.9	8.1	3.4

seeds, however, was found in plants grown in the highly contaminated sewaged soil previously bioremediated with canola plants, as it was 75.9 % less than that of the same plants grown in soil previously bioremediated with Indian mustard. This might be ascribed to that canola plants accumulated more Cu in straw as previously indicated by ANTONIOUS et al. (2011).

Results also indicated that canola hyper-phytoremediator was much more efficient in decontaminating the sewaged soil from Cu than Indian mustard, where the concentration of Cu in sorghum grains after canola and Indian mustard were 9.0 and 12.3 μg g^{-1} $plant^{-1}$ in marginally and 3.2 and 13.3 3.2 μg g^{-1} $plant^{-1}$ in highly contaminated sewaged soil, respectively (Tab. 4). Sorghum plants grown in the marginally and highly contaminated sewaged soils previously bioremediated with canola had a lower Cu content in their seeds compared to those grown in soil previously bioremediated with Indian mustard by 26.8 % and 74.6 %, respectively.

No differences were recorded in the Zn content of sorghum plants grown in the highly contaminated sewaged soil previously bioremediated with either canola or Indian mustard (Tab. 3). In the marginally contaminated sewaged soils, however, sorghum plants contained lower levels of Zn when grown after bioremediation with canola compared to Indian mustard.

It is worthy to state that Ni was not detected in sorghum plant or its organs after being grown in either marginally or highly contaminated sewaged soils previously bioremediated with either canola or Indian mustard (Tab. 4).

Sesame plants: Data presented in Tab. 5 indicated that sesame plants grown in the sewaged soil previously bioremediated with canola had lower Cu contents in their stem compared to plants grown in the sewaged soil previously bioremediated with Indian mustard, while a visa versa trend was noticeable in the Cu content in the plant roots. The lowest Cu content (6 μg/g dry weight) in sesame seeds was found in plants grown in the marginally contaminated sewaged soil previously bioremediated with canola plants, where the Cu content in the seeds was less than 53.9 % compared to those plants grown in the sewaged soil previously bioremediated with Indian mustard. In contrast, in the highly contaminated sewaged soils, the lowest Cu content in sesame seeds was found in plants grown in the sewaged soil previously bioremediated with Indian mustard causing a reduction in the Cu content of seeds up to 21.% compared to plants grown in the sewaged soil previously bioremediated with canola plants.

It’s worthy to mention that the concentrations of PTEs studied in the roots, shoot and seeds of Indian mustard plant were found to be lower than the Permissible Maximum Limits of the recommendation of World Health Organization (WHO, 1996).

Data given in Tab. 5 indicated that using canola to decontaminate the highly contaminated sewaged soil resulted in a decrement in the Zn content of sesame seeds, which is a sensitive plant to Zn, by 13.8 % compared to plants grown in the sewaged soil previously bioremediated with Indian mustard. In the marginally contaminated sewaged soil canola caused a reduction in the Zn content of seeds by 34.6 % compared to plants grown in the sewaged soils previously bioremediated with Indian mustard.

The Ni content in sesame seeds, as shown in Tab. 5 exhibited the same trend as sorghum plants where Ni was not detected in sesame plant or its organs grown in either marginally or highly contaminated sewaged soils previously bioremediated with either canola or Indian mustard.

This finding means that canola and Indian mustard might be considered efficient hyper-accumulators in phytoremediating sewaged soils contaminated with Ni and the Ni-bioremediated sewaged soil could be safely used for a sustainable growing for even sensitive crops such sesame.

Tab. 4: Cu, Zn and Ni content (μg/g DM plant) in sorghum plants and its parts grown in previously bioremediated sewaged soils at harvest.

Treatments		Cu			Zn			Ni		
Level of soil contamination	Bioremediated crops	Root	Stem	Seed	Root	Stem	Seed	Root	Stem	Seed
Marginally	Mustard	9.7	12.6	12.3	156.5	184.8	197.0	ND*	ND	ND
	Canola	11.7	12.8	9.0	178.9	152.3	166.6	ND	ND	ND
High	Mustard	9.7	12.6	13.3	184.8	212.2	162.3	ND	ND	ND
	Canola	11.7	25.0	3.2	152.3	171.5	164.9	ND	ND	ND

*ND: Non detected (less than 0.001 μg/g).

Tab. 5: Cu, Zn and Ni content (μg/g DM plant) in sesame plants and its parts grown in previously bioremediated sewaged soils at harvest.

Treatments		Cu			Zn			Ni		
Level of soil contamination	Bio-remediated crops	Root	Stem	Seed	Root	Stem	Seed	Root	Stem	Seed
Marginally	Mustard	9.5	10.5	13.0	182.0	153.0	275.0	ND*	ND	ND
	Canola	16.5	7.4	6.0	106.0	128.0	180.0	ND	ND	ND
High	Mustard	9.9	15.9	12.2	236.8	306.4	187.1	ND	ND	ND
	Canola	18.6	2.9	15.6	180.4	111.4	161.3	ND	ND	ND

*ND: Non detected (less than 0.001 μg/g).

Zinc equivalent

Despite Zn equivalent index is usually used in estimating the pollution of sewage sludge, in the current it was applied as a promising tools to follow the fate of PTEs in sewaged soil ecosystems as recommended by Hilal (1987), Soad El-Ashry, et al. (2011); Saber et al. (2012). The Zn equivalent index at harvesting of both sesame and sorghum grown in contaminated sewaged soils previously bioremediated with either canola or Indian mustard is shown in Tab. 6.

Both Zn equivalent value and Ni content in both marginally and highly contaminated sewaged soils after harvesting of sorghum or sesame obviously decreased compared to control soil. This finding indicated that canola and Indian mustard were efficient in phytoremediating the contaminated sewaged soil from PTEs, particularly Ni, and hence it is recommended that the bioremediated sewaged soil might be safely used for the sustainable growing of sensitive crops in such as sorghum and sesame.

In conclusion, results indicated that in the highly contaminated sewaged soil canola was significantly more effective in minimizing Zn equivalent index compared to Indian mustard which also exhibited a significant effectiveness. Worth to mention, the same trend was observed in the marginally contaminated sewaged soil.

PTEs contents in the sewaged soils

Data in given Tab. 7 indicated that bioremediation of the sewaged soil with either canola or Indian mustard plants followed by growing of sesame reduced Cu content in soil by more than 51 % and 40 % in the marginally contaminated sewaged soil and by 49 % and 52 % in

Tab. 6: Zn equivalent index in remediated soils in the presence and absence of AM inoculation after harvesting of sorghum and sesame crops.

Treatments	Control	T1 Mustard + Sesame	T2 Canola + Sesame	T3 Mustard + Sorghum	T4 Canola + Sorghum
Control	630*				
Marginally contaminated sewaged soils	210	49.0	43.6	52.2	44.9
Highly contaminated sewaged soils	275	61.8	54.8	66.3	56.4

*More details are documented in Soad El-Ashry et al. (2011)

Tab. 7: Contents (µg/g DW) and reduction percent of PTEs in sewaged soils previously bioremediated with either canola or Indian mustard at sesame harvest.

Treatments		Cu		Zn		Ni	
Level of soil contamination	Bioremediator crops	Conc.	Red. %	Conc.	Red. %	Conc.	Red. %
Marginally	Control*	9.5	-	159.0	-	4.07	-
	Canola	4.8	49.5	33.6	78.9	0.05	100
	Mustard	3.5	63.2	41.6	73.8	0.05	100
Highly	Control*	12.0	-	218.0	-	4.07	-
	Canola	6.0	50.0	42.0	80.7	0.1	100
	Mustard	4.5	62.5	52.0	76.1	0.1	100

*Without cultivation (no sesame/sorghum), Conc.: concentration Red.: Reduction

Tab. 8: Contents (µg/g DW) and reduction percent of PTEs in sewaged soils previously bioremediated with either canola or Indian mustard at sorghum harvest.

Treatments		Cu		Zn		Ni	
Level of soil contamination	Bioremediator crops	Conc.** ppm	Red. %	Conc. **	Red. %	Conc. **	Red. %
Marginally	Control*	9.50	-	159.0	-	4.07	-
	Canola	4.64	51.2	35.2	77.9	0.05	100
	Mustard	5.40	43.2	41.0	74.2	0.05	100
Highly	Control*	12.0	-	218.0	-	4.07	-
	Canola	5.80	51.7	44.0	79.8	0.1	100
	Mustard	6.73	43.9	52.0	76.1	0.1	100

*Without cultivation, Conc.: concentration Red.: Reduction

** Concentration after canola or Indian mustard and before sowing sorghum the highly contaminated sewaged soil, respectively. The same trend of results was achieved in both Zn and Ni pollutants

Results in Tab. 8 also indicated that bioremediation of the sewaged soil with canola followed by growing sorghum reduced Zn content in both marginally and highly contaminated sewaged soil by more than 77 %. Bioremediation with Indian mustard followed by growing sorghum or sesame, however, caused more than 73 % reduction in the Zn content in both marginally and highly contaminated sewaged soils.

Sorghum sowing after canola plants removed 59.4 % and 18.4 % of Cu and Zn, respectively, from the marginally contaminated sewaged soil. The removed percent of Cu and Zn were much higher in the highly contaminated sewaged soil. In the highly contaminated sewaged soil the decontamination role of canola plants was more pronounced, compared to that exhibited by plants grown in the marginally contaminated sewaged soil. More or less growing sesame plants after canola or mustard plants displayed the same trend as sorghum.

Results indicated that Ni reached a non-detectable level in the contaminated sewaged soils at sorghum and sesame harvest from both marginally and highly contaminated sewaged soils bioremediated by the two hyperaccumulators plants canola and Indian mustard.

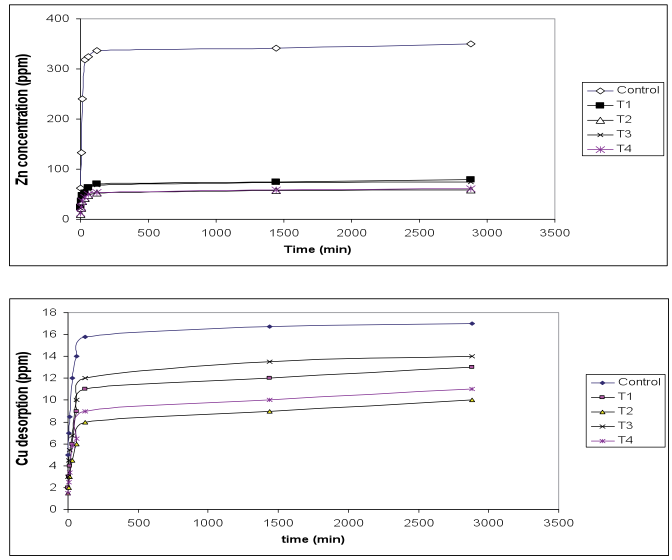

Fig. 1: Kinetics of Zn and Cu desorption from highly contaminated soil as affected by different treatments (T1: Sesame after Indian mustard; T2: Sesame after Canola; T3: Sorghum after Indian mustard; and T4: Sorghum after Canola).

Kinetics of PTE's release after phytoremediation of the highly contaminated soil

Kinetic study used in this work to understand the rate of potential toxic elements after different remediation treatments applied as an indicator for fat of pollutants released. Worth to mention that the succession in remediation for both contaminated sewaged soils was achieved through the experimental work as Ni was not detected directly after phytoremediation either in soil or plant compared to control treatment.

Concerning Zn equivalent, result showed significant minimizing in this parameter to a save level. Furthermore, in all treatment the rate of Cu and Zn desorption from the highly contaminated soil, as shown in Fig. 1, the rate processes started with speed rate trend of PTEs released, followed by a decreasing order to almost steady state conditions even in control treatment. Unquestionably, the rate of PTEs desorption was influenced by soil type, and the concentration of sorbents found in different treatment which were already limited according to remediation treatments applied except control. According to the same parameter, it could be emphasized that phytoremediation in removing both Zn and Cu from the contaminated sewaged soils with canola far exceeded compared to Indian mustard. Their existence in both contaminated sewaged soils was noticeably diminished at sesame and sorghum harvest.

The kinetic constants given in Tab. 9 represents the rate constants i.e. intensity and capacity factors of Cu and Zn desorbed from the highly contaminated sewaged soil previously phytoremediated with either Indian mustard or canola plants. In this study, Elovich, MFE and Horel's models were the best fitted models in describing the kinetic data according to highly coefficient of determination (R^2) and low standard error (SE) compared to other models tested i.e. diffusion and 1st order models (data not shown), those models showed less conformity in describing the kinetic data. It should be mention that high values for R^2 in more than one model mean that different mechanisms take place in desorption phenomena and subsequently the uptake of such PTEs by growing plants used as indicators after phytoremediation. As far as the intensity factor represented by ***a*** constant of Elovich equation, results pointed out that all treatments significantly decreased the rate of Zn desorption from both contaminated sewaged soils.

The Numerical values of **a**, the rate of Zn release as describe by Elovich model, indicated that, in highly contaminated soil, growing sesame after Indian mustard decreased the rate of Zn desorption from 34.4 in control to 6.76 mg kg^{-1} min^{-1}, the corresponding values in case of growing sesame after canola or sorghum after canola were 6.61 and 6.05 kg^{-1} min^{-1} and the best treatment under this condition was growing sesame after canola, which gave the lowest value i.e. 5.96 kg^{-1} min^{-1}. Results in Tab. (9) also indicated that the capacity factor represents by ***b*** constant values decreased in all treatments compared to the control. Furthermore, data pointed out that the lowest value, 16.93 mg kg^{-1}, was found in sesame grow after canola, the same value was 128.10 mg kg^{-1} and it should be mention that the same trend was observed in both control and all other treatments applied.

In MFE, data indicated that ***b***, the capacity factor of Zn desorption, decreased from 2.05 to 1.22 when growing sesame after canola. The same respective values were 1.49, 1.46 and 1.28 in cases of growing sesame after Indian mustard, sorghum after Indian mustard or sorghum after canola; all are significantly less than control, the same trend was observed in the same constant of Cu desorption from highly contaminated soil.

Before viewing the results of the Hoerl model, it should be mention that PTEs in remediated soil move through different pathways. For example, pollutants could be transferred to cultivated plants, distributed into hardly available form etc.. Horel model applied in this work describe transferring of PTEs from different treated soils into cultivated plants and according to design of model increasing the rate constant value, meaning increase the rate of pollutant absorbed by plants and vice versa. This model showed high conformity to describe PTEs release from the contaminated sewaged soils for the entire reaction time by having high significant R^2 ranged between 0.91**-0.96** for different treatments.

In zinc pollutant, increasing the intensity factor to 0.32 in T2 to be almost near the control, ensured the phytoremediation efficiency of this treatment compared to other treatments which gave higher values reached to 0.20, 0.21 and 0.29 in sesame grow after Indian mustard, sorghum after Indian mustard or sorghum after canola respectively. The same trend was also observed in Cu desorption from the highly contaminated soil.

Fig. 2 represents the kinetic parameters of Cu and Zn desorption from marginally contaminated sewaged soil. The kinetics of all PTEs

Tab. 9: Kinetic parameters of PTE's desorption from highly contaminated sewaged soil previously phytoremediated before growing sesame or sorghum.

Elovich									
	Zn					Cu			
Treatment	a	b	R^2	SE	Treatment	a	b	R^2	SE
Control	34.40	128.10	0.86**	0.69	Control	1.62	5.63	0.95**	1.59
T1	6.76	29.69	0.96**	5.63	T1	1.52	0.71	0.98**	0.77
T2	5.96	16.93	0.94**	6.31	T2	1.20	1.53	0.96**	1.28
T3	6.61	28.03	0.96**	5.79	T3	1.52	2.67	0.97**	1.18
T4	6.05	19.28	0.94**	6.14	T4	1.28	1.20	0.97**	0.87
MFE									
	a	*b*	R^2	SE		a	*b*	R^2	SE
Control	0.18	2.05	0.81**	0.17	Control	0.51	0.78	0.92**	0.08
T1	0.13	1.49	0.93**	0.07	T1	0.25	0.38	0.95**	0.10
T2	0.19	1.22	0.87**	0.14	T2	0.24	0.23	0.94**	0.11
T3	0.14	1.46	0.91**	0.08	T3	0.20	0.54	0.95**	0.08
T4	0.18	1.28	0.87**	0.13	T4	0.25	0.29	0.94**	0.11
Horel's model									
	a	b	R^2	SE		a	b	R^2	SE
Control	0.34	4.39	0.86**	0.37	Control	0.44	1.62	0.97**	0.04
T1	0.20	0.23	0.96**	0.04	T1	0.26	0.09	0.97**	0.06
T2	0.32	2.55	0.91**	0.24	T2	0.41	0.65	0.96**	0.11
T3	0.21	3.20	0.95**	0.06	T3	0.35	0.08	0.97**	0.05
T4	0.29	2.71	0.91**	0.18	T4	0.35	0.42	0.97**	0.09

T1: Sesame after Indian mustard T2: Sesame after Canola
T3: Sorghum after Indian mustard T4: Sorghum after Canola

desorbed were divided into three reaction periods; the 1st period was characterized by rapid reaction started from the beginning of ESFU working and reached to about 30 min. The 2nd period showed a decline in PTEs adsorption on different clay minerals and this period was about 90 min. The 3rd one, however, stayed for the rest of the reaction period, about 60 min, and was almost characterized by stability or slight increase in rate of adsorption (slow step) regardless the type of PTEs.

Again, data indicated that the concentration of PTEs in the marginally contaminated sewaged soil previously phytoremediated with either canola or Indian mustard before growing of sesame and sorghum were decreased and subsequently the rate of desorption of these pollutants from the soil were took place. Results also confirmed that growing of sesame after canola was the best effective treatment in diminishing PTEs desorption rate. Through the kinetic study it could be mention that although using of Indian mustard as a phytoremediator plant significantly decreased Cu and Zn desorption compared to control, data indicated that canola was much more preferable to be applied under the current experimental conditions.

The comparison between sesame and sorghum in their efficiency in adsorbing Cu and Zn from sewaged soils, results indicated that sesame after phytoremediation treatment was more effective compared to sorghum. Numerically, as shown in Zn desorption, the maximum value was 63 ppm when growing sesame after Indian mustard, the corresponding value was 60 ppm when sorghum grow after Indian mustard, the same trend was observed in the cases of growing sesame after canola or sorghum after canola.

The rate constants of used models described the kinetics of Zn release from the marginally sewaged soil are given in Tab. 10. All used

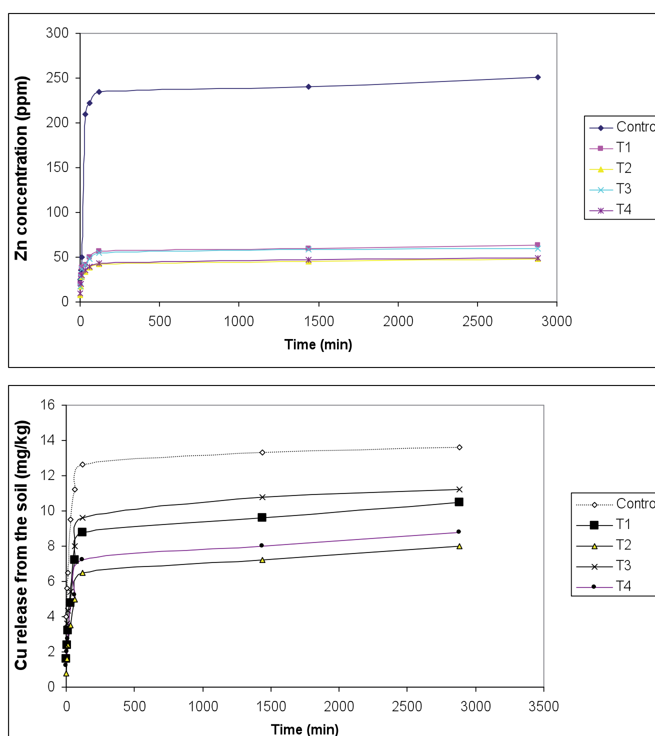

Fig. 2: Kinetics of Zn and Cu desorption from the marginally contaminated sewaged after phytoremediation.

models describe the kinetics of PTEs studied were suitable to describe the kinetic data according to high and significant R^2 and low of SE.

In the marginally contaminated sewaged soil, again data implied that the rate constants of Horel model showed significant decrease in control followed by sesame after canola treatment and sorghum after canola or sorghum after Indian mustard respectively.

The lowest value was recorded when growing sesame after Indian mustard means that growing sesame after canola was the best in minimizing the hazards of such contaminates and hence ensures sustainability.

In MFE represented in the same table, although the same trend was observed when sesame grow after canola, data indicated that the rate of Cu desorption was decreased in almost all treatments by about 50 % compared to control. The corresponding percentage values for different treatments in Zn were about 70 % in sesame grown after canola and sorghum after canola. The lowest values in were found when sorghum or sesame were grown after Indian mustard.

The impacts of phytoremediation either alone or in combination with AM inoculation, on the biological characteristics of both highly and marginally contaminated sewaged soils at sowing and maturity stages under sesame or sorghum plantation are drawn Fig. 3 and 4.

After sole phytoremediation in the highly contaminated sewaged soil, the dehydrogenase activity significantly increased from 22.84 to 60.23 µg g^{-1} soil after 24 hours under sesame and from 22.84 to 61.48 µg H_2 g^{-1} soil after 24 hours under sorghum.

Phytoremediation associated with AM inoculation led to slightly higher significant dehydrogenase activity that increased from 22.84 to 65.79 µg H_2 g^{-1} soil after 24 hours and from 22.84to 63.26 µg H_2 g^{-1} soil after 24 hours after phytoremediation, respectively with sesame or sorghum.

After sole phytoremediation in the marginally contaminated sewaged soil, the dehydrogenase activity showed slight increase from 24.68 to59.45 µg H_2 g^{-1} soil after 24 hours under sesame and from 24.68 to 60.61 mg H_2/g soil after 24 hour under sorghum.

Phytoremediation associated with AM inoculation led to slightly higher dehydrogenase activity that increased from 24.68 to 67.32 µg H_2 g^{-1} soil after 24 hours and from 24.68 to 61.78 µg H_2 g^{-1} soil after 24 hour after phytoremediation respectively with sesame or sorghum.

It seems reasonable to conclude that during the cultivation season, indigenous rhizosphere soil microorganism flourished in the control

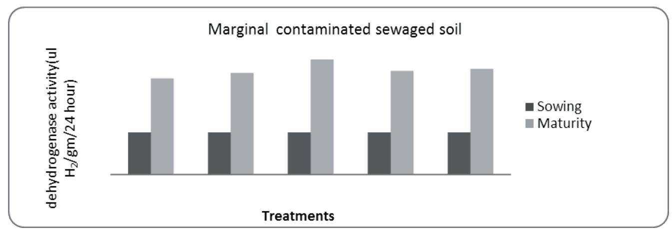

Fig. 3: Sowing and final dehydrogenase activity the marginally contaminated sewaged soil under different phytoremediation treatments (µl H_2 g^{-1} 24^{-h})

Tab. 10: Kinetic parameters of PTEs desorption from the marginally contaminated sewaged previously phytoremediated before growing sesame or sorghum**.**

Elovich									
	Zn					Cu			
Treatment	*a*	*b*	R2	SE	Treatment	*a*	*b*	R2	SE
Control	32.35	31.45	0.87**	0.71	Control	1.30	4.42	0.95**	1.30
T1	5.41	23.75	0.96**	4.50	T1	1.20	0.67	0.97**	0.69
T2	4.88	13.54	0.94**	5.05	T2	095	1.20	0.96**	1.02
T3	5.32	22.21	0.96**	4.70	T3	1.22	2.14	0.97**	0.94
T4	4.77	15.43	0.95**	4.80	T4	1.02	0.96	0.97**	0.69
MFE									
Treatment	*a*	*b*	R2	SE	Treatment	*a*	*b*	R2	SE
Control	0.31	1.53	0.85**	0.24	Control	0.52	0.68	0.92**	0.08
T1	0.19	1.40	0.93**	0.07	T1	0.26	0.10	0.95**	0.12
T2	0.13	1.13	0.87**	0.14	T2	0.20	0.28	0.94**	0.11
T3	0.18	1.36	0.91**	0.08	T3	0.24	0.45	0.95**	0.08
T4	0.14	1.19	0.87**	0.13	T4	0.23	0.19	0.94**	0.11
Horel's									
Treatment	*a*	*b*	R2	SE	Treatment	*a*	*b*	R2	SE
Control	0.52	3.09	0.88**	0.81	Control	0.23	1.42	0.97**	0.04
T1	0.19	0.07	0.96**	0.04	T1	0.37	3.92	0.97**	0.11
T2	0.32	2.32	0.91**	0.24	T2	0.35	0.42	0.96**	0.11
T3	0.21	0.97	0.95**	0.05	T3	0.27	2.20	0.97**	0.05
T4	0.29	2.49	0.91**	0.19	T4	0.35	0.20	0.97**	0.09

T1: Sesame after Indian mustard T2: Sesame after Canola
T3: Sorghum after Indian mustard T4: Sorghum after Canola

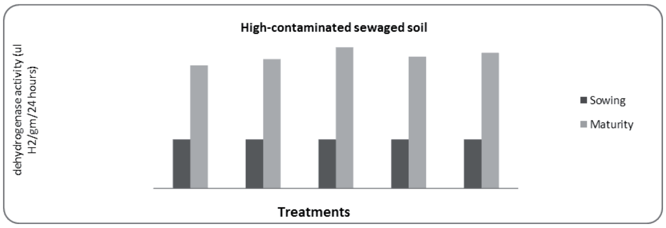

Fig. 4: Sowing and final dehydrogenase activity the highly contaminated sewaged soil under different phytoremediation treatments (µl H_2 g^{-1} 24^{-h}).

and phytoremediate treatments of both contaminated sewaged soils due to the new well-furnished soil ecosystems and hence increased the dehydrogenase activity therein.

Recorded changes in dehydrogenase activity under the different phytoremediation treatments confirmed slight increases in the dehydrogenase activity in both soils due to cultivation in the presence and absence of AM inoculation despite being slightly higher under the later treatments. It seems reasonable to conclude that phytoremediation did not show any adverse impacts on the biological characteristics of both highly and marginally contaminated soils.

Discussion

The use of both hyperaccumulators plants to remove PTEs from sewaged soils (phytoremediation) is expanding due to its cost-effectiveness compared to conventional methods besides its great potential. Plants that absorb highly levels of a given element from soil called hyperaccumulators are now being closely investigated, both by molecular techniques and by soil/plant analyses, at the sites where they occur. The term phytoremediation generally refers to phytostabilization and phytoextraction. In phytostabilisation, soil amendments and plants are used to alter the chemical and physical state of the PTEs in the soil. In phytoextraction, plants are used to remove contaminants from the soil and are then harvested for processing.

Plants suitable for phytoremediation should possess a highly ability to accumulate the targeted PTEs, preferably in the aerial parts, can tolerate the highly accumulated PTEs concentrations, characterized with a fast growth of the PTEs accumulating biomass as well as with ease cultivation and harvesting (Baker and Brooks, 1989). However, Chaney et al. (1997) argued that PTEs tolerance and hyperaccumulators are more important factors than highly biomass production.

Recently, hyper-accumulating plants had been a subject of several phytochemical studies because unlike most other plants, milligram rather than microgram quantities of organo-metallic complexes could be separated from their plant tissues. Hence, great attention is now being paid to the potential use of hyperaccumulators to bioremediate contaminated sewage soils.

Worthy to mention that both Indian mustard and canola used as hyperaccumulators plants, before sowing sesame and sorghum in the current study, almost absorbed almost all Ni found in the both contaminated sewaged soils which directly decreased the value of the Zn equivalent parameter that is frequently used to define the level of PTEs contamination in soils.

Reeves and Brooks (1983) reported that Indian mustard is a suitable hyperaccumulator plant in minimizing the hazards of Zn in contaminated soils compared to other PTEs that might be found in soil ecosystem. On the other hand, under the current experimental conditions, canola was more effective than Indian mustard because it significantly accumulated higher amounts of Ni, Zn and Cu from the sewaged soil and resulted in minimizing their amounts in the following crops (sesame and sorghum) as previously shown by Turan and Esringü (2007) that canola was more effective than Indian mustard in the uptake of Cu, Cd, Pb and Zn. In a lysimeter experiment, Agoea et al. (2009) investigated two bioremediation amendments; either compost or clay inoculated with AM and *Streptomyces* sp. followed by growing a mixture of two plant species. They found that leaching and plant uptake of PTEs from the amended plots were higher than in the control, mainly because of the higher plant biomass. The inoculation of control soil with AM decreased the leaching of PTEs, but the effect on their uptake by plants was PTEs and species dependent. The extra inoculation with *Streptomyces* sp. increased the leaching of many PTES, and also in many cases their uptake by plants. The cumulative of PTEs by plants and leaching was higher in treatments with expanded clay in the case of Mn, Ca, Ni and Pb, smaller in the case of Cu.

The dehydrogenase activity is repeatedly used to follow up the biological activities in the soil ecosystem. In the current work the final level of dehydrogenase activity slightly increased compared to its initial value indicating that the trailed materials have a simulative rather than inhibitive impacts on soil biomass.

Conclusion

It might be concluded that using canola as phytoremedator was an effective tool to bioremediated contaminated sewaged soil rendering it proper for growing economic crops.

Acknowledgment

The authors would like to express their appreciations and gratitude to the authorities of Science and Technology Development Fund (STDF) for financing the present work through the project number 1425 contracted with the National Research Center on Bioremediation of Sewaged Soils.

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