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Heavy metal contamination from mining site in marrakech-Morocco: Diffusional transport of trace elements from contaminated soil to crop plants

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ABSTRACT

Couch grass, Tamarix Galica and wheat, grown in industrially polluted region, were included in the present research. The experimental plots were situated at two different villages near a mine area(district of DARÄA LASFAR) who is located in North-West of the Mrabtine zone, located at approximately 10 km in the west of Marrakech-MORROCO. The aim of this work was to estimate whether a given soil is suitable for cultivation of plants used as food or feed. The paper discusses the transfer of metal ions(Al, Cd, Cu, Pb and Zn) from contaminated soils to plants. We investigated the level of soils pollution and the way heavy metals enter the fibre crops, by taking soil and plant samples. The contents of heavy metals in plant materials(roots and leaves) were determined after the method of the dry mineralization. The quantitative measurements were carried out with atomic absorption spectroscopy (AAS). A clearly distinguished species peculiarity exists in the accumulation of heavy metals in the vegetative and reproductive organs of the three corps. Couch grass seems to be the most strongly absorber and accumulator of heavy metals from the soil; it removes considerable quantities of heavy metals from the soil with its root system. Such a relationship is generally observed between the total soil content of a given metal and that of the plant. For this reason, it was necessary to develop a mathematic expression to get easy the comprehension of the way heavy metals enter the fibre crops and their transfer from contaminated soils, based on diffusional transport of the soil solute towards the roots.

KEYWORDS

Plants;
Soil;
Trace elements;
Transfer.

INTRODUCTION

Heavy metals are the main group of inorganic contaminants and a considerable large area of land is contaminated with them due to use of sludge or municipal compost, pesticides, fertilizers, and emissions from municipal waste incinerators, car exhausts, residues from metalliferous mines, and smelting industries[21,24,61].

Irrespective of the origin of the metals in the soil, excessive levels of many metals can result in soil quality degradation, crop yield reduction, and poor quality of agricultural products[35], pose significant hazards to human, animal, and ecosystem health[4,61]. It is the case of the district of Daraa Lasfar who is located in North-West of the Mrabtine zone, located at approximately 10km in the west of Marrakech.

Although metals are present naturally in the Earth's crust at various levels and many metals are essential for cells(e.g. copper(Cu), iron(Fe), manganese(Mn), nickel (Ni), zinc(Zn)), all metals are toxic at higher concen trations[8,40,61].

The accumulation of heavy metals and metalloids in agricultural soils is of increasing concern due to the food safety issues and potential health risks as well as its detrimental effects on soil ecosystems[19,18,8,40].

The threat of heavy metals to human and animal health is aggravated by their long-term persistence in the environment[23,61], they may be transferred and accumulated in the bodies of animals or human beings through food chain, which will probably cause DNA damage and carcinogenic effects by their mutagenic ability[11,31,40,61].

Soil-to-plant transfer of heavy metals is the major pathway of human exposure to soil contamination. Health risk due to soil contamination with single heavy metal has been widely studied. For example, lifetime exposure to low level soil contamination with cadmium(Cd) has shown to cause renal dysfunction in residents living near the contamination sites in Japan[52] and China [6,28,44,57,58,10].

The bioavailability of metallic elements to plants is controlled by many factors associated with soil and climatic conditions, plant genotype and agronomic management, including: active/passive transfer processes, sequestration and speciation, redox states, the type of plant root system and the response of plants to elements in relation to seasonal cycles[29,8], the nature of the soil on which the plant is grown and the degree of maturity of the plant at the time of harvesting[32,54,56]. The nature of the soil is one of the most important factors in determining the heavy metal content of food plants [27,37]. However, the heavy metal content in plants can also be affected by other factors such as the application of fertilisers, sewage sludge or irrigation with wastewater [38,14,20,41].

The aim of this work was to estimate whether a given soil is suitable for cultivation of plants used as food or feed on the basis of the composition of soil extract by studying metal ions transfer from soil to plant and to predict before sowing(based on soil composition) whether the concentration of a given contaminant in plants would be below the acceptable level.

An important part of this investigation was the elaboration of a mathematic expression (simple model) that enabled estimation of the content of metals in soil transferred to plants during the growing season.

A significant problem is the choice of a variable that has the largest impact on the level of concentration of a given element in plants. For many years, such a variable chosen was the total content of a given trace element in soil. But it was difficult to find a clear dependence, since not all the forms of metals present in soil are available to plants[8].

As is well known, a major fraction of a trace element present in the rooting-zone may be fixed to soil constituents. This has led to the development of the concept of 'bioavailability' of trace substances in soil[13,53].

Procedures which are simple and reliable unfortu nately do not seem to be available for determining the bioavailable fractions of metallic elements traces in soils. Commonly, sequential extraction techniques are applied which use a sequence of progressively aggressive reagents to selectively leach the fractions of trace substances bound to specific soil components[3,19,47,48,49], they provide a scale of availability of these pollutants. Both selectivity and reproducibility of the extraction steps, however, have been debated[3,9,30,39,43,60]. Specifically, a relationship between the operationally-defined 'bioavailable' fraction determined by these techniques and plant root uptake remains to be established[17].

MATERIEL AND METHOD

In order to investigate the level of pollution and the way heavy metals enter the plants, soil and plant samples were taken.

Soil samples were taken at depth from 0 to 20cm, they were air-dried and crushed to pass through a 2mm nylon screen and were then used to determine total metallic elements traces (Al, Cd, Cu, Pb, Se and Zn).

A1.000g sample of the <2mm soil fraction mineralised into a Muffle furnace(T=400^{0}C) for 4 h, then was digested in an open system in Teflon crucibles with a solution of concentrated HF at 150^{0}C to assure complete dissolution of silica. General soil properties (OM, pH, texture) were analyzed using standard methods[8]. Soil chemical and physical properties of the samples are listed in TABLE 1.

Plant samples were washed in deionised water[16], transferred to paper bags, and dried in an air-forced oven at 60^{0}C for 48h. Dry plant material was ground in a stainless steel blender to pass through a 2-mm screen. All samples were stored at ambient temperature and humidity.

The contents of heavy metals in the plant material were determined after the method of the dry mineralization. The results are summarised in TABLES 2, 3 and 4.

TABLE 1: Organic and mineralogical proprieties of soils(n=3)

	S1 (village 1)	S2 (village 2)
Clay	21.,4±2,1	25.3±3.1
Fine silt	16.1±1,7	13.5±2.7
Coarse slit	9.2? 1.4	8.7±1.5
Fine sand	25.2? 2.9	23.4±3.1
Coarse sand	27.7±2.5	28.3±2.8
	Chemical properties	
pH	8.12? 0.57	7.93±0.64
C.E (ms/cm)	1.51±0.43	1.67±0.36
CEC (meq/100g)	31.61±3.06	35.24±2.67
MO (%)	4.72? 1.09	5.54±0.68
COT (%)	2.74±0.63	3.22±0.40
Al (μg/g)	4762.71±1210.02	2577.12±916.35
Cd (μg/g)	1.11±0.66	2.21±0.15
Cu (μg/g)	227.83±225.27	3 Long 53±22.76
Pb (μg/g)	221.36±40.70	288.61±44.24
Se (μg/g)	9.23±3.42	34.05±9.93
Zn (μg/g)	648.04±174 Long	890.45±101.03

TABLE 2 : Evolution of heavy metals concentration in couch grass vegetative and reproductive organs during corps flowering stage

		Couch Grass									
Months		Al (mg/kg dry weight)		Cd (mg/kg dry weight)		Cu (mg/kg dry weight)		Pb (mg/kg dry weight)		Zn (mg/kg dry weight)	
		roots	leaves	roots	leaves	roots	leaves	roots	leaves	roots	leaves
1	V1	134.44±8.80	3.53±0.11	0.41±0.04	0.28±0.02	7.86±0.30	1.07±0.19	5.73±0.32	0.20±0.01	370.99±38.36	121.43±7.16
	V2	664.24±34.13	10.23±1.71	0.50±0.01	0.25±0.01	10.74±0.27	1.73±0.04	8.43±0.17	0.29±0.01	589.40±20.07	162.86±1.65
2	V1	1246.48±119.71	32.77±1.54	0.66±0.02	0.46±0.01	10.16±0.44	1.38±0.28	6.00±0.18	0.21±0.01	534.60±8.85	174.99±1.65
	V2	2204.67±44.87	33.96±0.93	0.83±0.05	0.42±0.01	13.64±0.34	2.20±0.05	9.74±0.14	0.34±0.02	1098.44±34.58	303.51±2.84
3	V1	1477.45±91.25	38.84±1.17	0.67±0.04	0.47±0.02	12.04±0.20	1.63±0.13	16.01±0.23	0.56±0.03	722.22±23.24	236.40±4.34
	V2	2897.34±65.54	44.63±1.36	1.02±0.09	0.52±0.01	14.35±0.44	2.31±0.07	19.70±0.29	0.69±0.06	1370.01±162.76	378.55±13.38
4	V1	1977.47±77.36	56.50±1.22	0.75±0.05	0.61±0.02	13.00±0.50	3.40±0.36	37.88±0.69	1.32±0.04	978.02±45.89	336.48±11.69
	V2	3900.21±303.95	66.32±1.10	1.58±0.15	0.93±0.03	15.11±0.57	5.37±0.29	38.17±1.66	1.98±0.03	1558.02±25.77	539.40±73.60
5	V1	3302.46±39.13	63.51±2.23	1.08±0.04	0.62±0.02	14.47±0.48	5.51±0.43	61.84±0.97	1.37±0.03	1281.88±67.03	245.31±11.79
	V2	4115.33±237.21	86.69±1.71	1.83±0.03	0.71±0.01	17.32±1.03	7.99±0.42	67.06±1.39	2.02±0.01	2016.88±33.24	549.42±135.67
6	V1	4021.99±117.57	80.76±3.29	1.19±0.08	0.68±0.04	16.01±0.22	6.09±0.20	61.84±0.97	2.03±0.01	1443.68±81.72	276.27±14.38
	V2	6263.10±53.99	96.65±3.05	2.06±0.05	0.80±0.03	19.25±1.04	8.88±0.43	78.17±1.66	2.74±0.04	2097.67±18.70	571.43±76.34
7	V1	4210.68±281.11	90.76±3.26	1.33±0.03	0.76±0.02	24.04±0.45	9.15±0.41	92.59±1.99	3.23±0.07	2266.97±54.69	540.56±94.98
	V2	8921.25±11.89	115.04±3.11	2.73±0.05	1.07±0.04	27.49±1.72	12.69±0.71	100.42±2.16	5.56±0.13	2824.74±539.79	617.55±19.13
8	V1	4925.63±144.58	113.06±4.27	1.55±0.81	0.88±0.36	28.19±0.38	10.73±0.34	94.65±1.20	3.30±0.04	2754.27±533.01	589.85±41.56
	V2	13653.36±22.44	138.90±4.33	3.07±0.21	1.20±0.10	29.65? 0.30	13.68±0.26	102.64±2.21	6.24±0.09	3082.27±236.18	750.29±26.58

TABLE 3: Evolution of heavy metals concentration in wheat vegetative and reproductive organs during corps flowering stage

		Wheat									
Months		Al (mg/kg dry weight)		Cd (mg/kg dry weight)		Cu (mg/kg dry weight)		Pb (mg/kg dry weight)		Zn (mg/kg dry weight)	
		roots	leaves	roots	leaves	roots	leaves	roots	leaves	roots	leaves
1	V1	197.0.3±42.37	72.75±2.93	0.28±0.03	0.18±0.01	5.35±0.23	1.54±0.02	2.01±0.03	0.03±0.01	163.75±27.28	448.63±21.40
	V2	245.10±16.01	86.30±3.42	0.44±0.01	0.26±0.01	8.59±1.07	3.12±0.23	2.99±0.03	0.45±0.01	398.56±86.45	937.50 ± 45.23
2	V1	253.99±56.54	93.79±3.91	0.66±0.03	0.42±0.01	6.96±0.74	2.01±0.05	3.20±0.08	0.35±0.03	319.57±59.77	288.83±46.88
	V2	302.77±9.73	106.61±2.08	0.80±0.07	0.47±0.06	10.18±1.00	3.70±0.22	4.97±0.36	0.85±0.05	548.72±65.42	253.32±15.26
3	V1	297.01±4.98	122.38±5.08	1.05±0.03	0.44±0.01	8.75±0.06	2.84±0.02	3.23±0.16	0.52±0.01	457.27±19.58	271.41±28.39
	V2	404.63±14.37	135.59±1.35	1.17±0.06	0.58±0.17	12.62±0.24	4.62±0.07	6.94±0.08	0.84±0.08	812.27±27.37	365.33±44.51
4	V1	342.25±2.29	141.96±1.69	1.22±0.02	0.47±0.07	15.81±0.40	3.02±0.01	7.81±0.25	1.03±0.01	492.27±34.00	366.78±17.15
	V2	448.54±13.04	193.68±2.28	1.60±0.06	0.79±0.06	18.51±0.62	5.64±0.01	9.83±0.68	2.12±0.08	780.17±23.14	932.95±129.35
5	V1	383.06±4.51	177.51±3.52	1.45±0.03	0.58±0.17	17.01±0.33	3.09±0.09	8.54±0.14	1.11±0.01	591.99±53.07	224.15±9.21
	V2	496.73±100.34	210.70±2.27	1.75±0.04	0.93±0.05	21.50±0.75	6.17±0.06	11.93±0.13	2.60±0.32	918.32±68.10	1422.61±155.88
6	V1	413.04±20.48	195.21±5.84	1.70±0.03	0.60±0.06	17.74±0.86	4.15±0.03	9.36±0.09	2.00±0.01	658.02±7.21	490.00±42.43
	V2	569.30±5.88	242.51±3.85	1.93±0.04	1.18±0.04	21.93±0.82	6.58±0.06	11.60±0.11	2.62±0.01	970.19±18.97	598.11±34.85
7	V1	468.54±45.17	204.10±3.43	1.78±0.04	0.83±0.04	18.95±0.48	5.23±0.36	10.89±0.39	2.94±0.06	693.99±85.22	332.62±38.87
	V2	626.39±47.29	331.45±2.01	2.19±0.05	1.40±0.02	23.48±0.31	6.69±0.12	12.51±0.29	3.87±0.53	1016.22±31.14	424.21±20.28
8	V1	673.33±37.29	394.30±5.39	2.23±0.19	0.88±0.05	23.79±0.46	5.56±1.30	11.22±0.11	3.68±0.59	725.54±31.14	418.90±5.45
	V2	833.17±159.04	422.53±69.57	2.79±0.06	1.70±0.08	28.72±0.97	7.10±0.54	13.92±0.13	4.82±0.58	1085.47±133.29	470.72±44.88

TABLE 4 : Evolution of heavy metals concentration in Tamarix Galica vegetative and reproductive organs during corps flowering stage

Month	Tamarix Galica									
	Al (mg/kg dry weight)		Cd (mg/kg dryweight)		Cu (mg/kg dry weight)		Pb (mg/kg dryweight)		Zn (mg/kg dry weight)	
	roots	leaves	roots	leaves	roots	leaves	roots	leaves	roots	leaves
1	771.8±19.9	941.9±43.4	0.28±0.02	0.92±0.01	5.92±0.17	12.64±0.19	1.38±0.03	3.83±0.14	168.19±8.87	474.33±5.06
2	805.5±43.4	1091.0±47.0	0.29±0.02	0.89±0.05	6.74±0.06	14.65±0.17	1.75±0.07	5.73±0.13	380.61±41.76	508.31±22.49
3	812.2±53.7	1153.4±78.2	0.33±0.03	0.78±0.06	10.83±0.37	13.05±0.62	1.82±0.09	6.35±0.21	428.21±31.74	446.60±10.04
4	1130.5±165.1	1484.5±219.3	0.48±0.03	0.54±0.01	14.82±0.45	13.75±0.13	2.75±0.12	7.19±0.30	486.36±39.48	818.68±48.56

A sample was weighed in a quartz crucible to 1g and put into a Muffle furnace (T=400^{0}C) for 4h until ashing. After cooling to room temperature, 1ml HNO3 (1:1) was added, evaporated in a sand bath and put again into the Muffle furnace (T=400^{0}C). The procedures were repeated until the ash was white. It was finally dissolved in 5ml 5% HCl, transferred in a scaled 10ml flask and brought to volume with bi-distilled water[1].

RESULTS

Hyperaccumulation of heavy metals by higher plants is a complex phenomenon. It involves several steps, such as: (a) transport of metals across the plasma membrane of root cells; (b) xylem loading and translocation; and(c) detoxification and sequestration of metals at the whole plant and cellular levels[34,61].

Heavy metals had no influence on the crops' development and productivity. Anthropogenic increase of heavy metal concentration leads to increase uptake of heavy metals by Couch grass, Tamarix Galica and wheat without evident yield depression or decrease of quality of harvested products[1].

Our results, presented in TABLES 2, 3 and 4, show that all plants used in this study have an ability to absorb metals from the soil and accumulate them in the shoots under low and high metal levels[29,61]. The plant response to heavy metals in soil depends on the plant species, the total soil metal concentration, and on the bioavailability of the metal itself depending on physico-chemical properties of soils[5].

The results for the heavy metals contents in the studied plants given in TABLES 2, 3 and 4 show considerable differences in the metals distribution in the separate plant parts. For all metallic trace elements, the main concentrations were accumulated in the roots and their quantity increased as their time of growing increased.

This report indicated a clearly expressed tendency: For the same heavy metals contents in the soil, as the duration of the corps flowering stage increased, as strong tendency towards increase of heavy metals contents in the roots of studied corps was observed.

The results obtained proved that heavy metals movement and accumulation in the vegetative organs of the studied crops differed considerably.

The highest values were obtained after eighth month in couch grass roots, where Pb reached 94,7mg/kg in village 1 VS 102.6mg/kg in village 2, Cd 1,6mg/kg in village 1 VS 3,1 mg/kg in village 2,Cu 28,2 mg/kg in village 1 VS 29.7mg/kg in village 2, Zn 2754.27 mg/kg in village 1 VS Long82.3mg/kg in village 2 et al. 4925,6mg/kg in village 1 VS 13653,4 mg/kg in village 2(TABLE 2). Lower values were established in Tamarix Galiva roots-2,8mg/kg Pb, 14,8 mg/kg Cu, 486,4mg/kg Zn and 0.5mg/kg Cd (TABLE 4). The results obtained could be explained with the anatomic and biological features of the plants[1]. A bigger part of the heavy metals that had entered the soil were fixed and accumulated in the couch grass roots, as couch grass formed weakly developed root system. The lower values, obtained in Tamarix Galiva, were correlated to its more deeply penetrating root system.

The results for the heavy metals contents in the roots wheat(TABLE 3), grown in the studied region, were analogous, but the obtained values were far lower compared to those obtained for couch grass. Pb contents varied from 2mg/kg in village and 3.0 mg/kg in village 2 in the first month to 11.2mg/kg and 13.9mg/kg in the eighth one, Cd from 0,3 and 0.4 mg/kg to 2,2 and 2.8mg/kg, Cu from 5,4 and 8.6mg/kg to 23.8 and 28,7mg/kg, Al from 197,0 and 245.1mg/kg to 673.3 and 833,2mg/kg and Zn from 163,7 and 398.6mg/kg to 725.5 and 1085,5mg/kg.

Our results show that the trace elements quantities

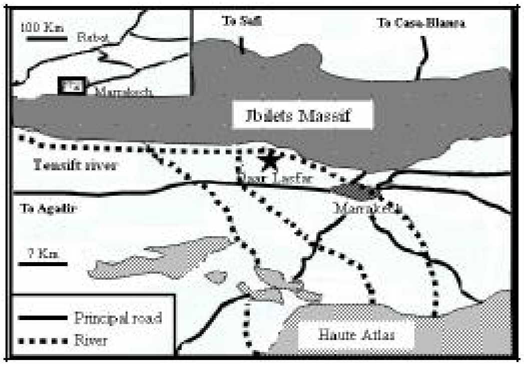

Figure 1: Draa Lasfar mine localisation in Marrakech region

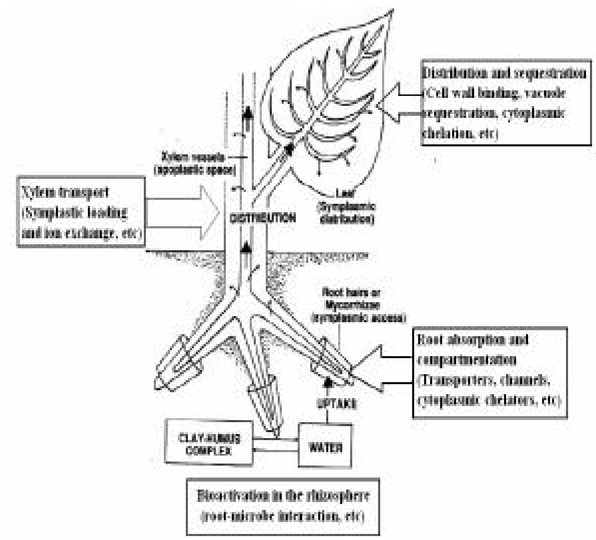

Figure 2 : Major processes proposed to be involved in heavy metal hyperaccumulation by plants

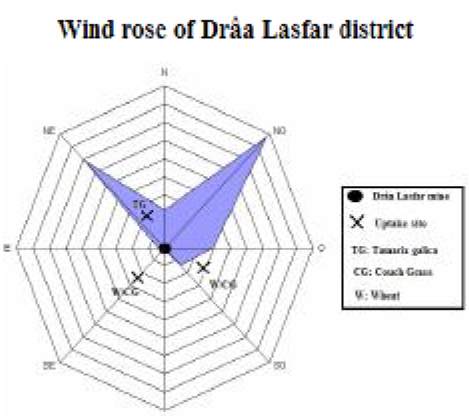

Figure 3 : The wind rose of Draa Lasfar district

in the fibre crops' leaves were considerably lower compared to the root system, which proved that heavy metals movement along the plants conductive system was strongly limited[1].

These results show a clearly distinguished species peculiarity existed in the accumulation of heavy metals in Tamarix Galica, couch grass and wheat vegetative and reproductive organs. Couch grass was the crop that most strongly extracted and accumulated heavy metals from the soil, followed by wheat and Tamarix Galica. This was probably in connection with the plats anatomic and biological features, as well as with the presence of protective mechanisms in plants[61].

Finally, the results show that heavy metals contents in Tamarix Galica leaves were higher compared to root system, while the opposite tendency was observed in couch grass and wheat. Pb in Tamarix Galica leaves reached 7.2mg/kg (TABLE 4), while the obtained values in couch grass and wheat were very close-6.2 and 4.8 mg/kg respectively(TABLEs 2 and 3). The results obtained for Al, Cu and Zn were analogous. This higher heavy metals accumulation was probably due to the fact that the Tamarix Galica leaves were exposed to the dominant wind directions which enabled the transfer and embedding of aerosol contaminators on their surface and their absorption into the leaves. Our results corresponded to the ones obtained from[33], according to which, under conditions of soil and air pollution from pollution(from a factory producing amorphous), considerable quantities of Pb and Zn were deposited and absorbed in the leaves of cotton plants[33,61]. The obtained results matched well with those of Watson et al.[59] and Mullins and Burmester[42], who found that Cd, Cu, Fe, Mn and Zn contents were the highest in leaves of some plants.

The study of the wind rose of the city of Marrakech-Morocco(figure 3) can be used to justify the increase of concentrations of metallic elements traces obtained in Tamarix galica leaves. Strictly defined, the wind rose denotes a class of diagrams designed to display the distribution of wind direction experienced at a given location over a period of time.

The wind rose shows that northwest-southeast wind directions are dominant and that the relative frequency of the wind speed covers the samples site of Tamarix Galica, witch explain the role of wind as a principal transportation and dispersion factor of metallic elements.

Diffusional transport

Solutes are transported to plant roots by mass flow and diffusion[2]. Mass flow occurs with the convective flow of water which is created by root water uptake in response to transpiration. If, however, root uptake rates of a solute exceed mass flow rates, depletion of the solute at the root-soil interface creates a concentration gradient which initiates additional diffusional transport of the solute towards the roots. As a consequence, a depletion zone around the absorbing root develops, which in the long term reduces uptake rates of the solute[45,17].

The passive diffusion tends to establish a balance between the concentrations exist on both sides of a biological membrane. The toxic cell accumulation is comparable to the oil-water partition.

At the equilibrium state, the following equation will be satisfied[12]:

$$Co \underset{K_2}{\overset{K_1}{\rightleftharpoons}} C_i$$

When an organism is exposed to a poison, the movements of entry per unit of time can be described by [47]:

$$\begin{aligned} dCt/dt &= K_1Co - K_2C_i \\ dCt/dt &= Co(K_1 - K_2C_i/Co) \\ dCt/Co &= (K_1 - K_2C_i/Co)dt \\ Ln(Ct/Co) &= (K_1 - K_2C_i/Co)t + cst \end{aligned} \quad (1)$$

At- t=teq, Ct/Co=1, K_1 Co=K_2 C_i ; So- cst = 0

(1) Becomes

$$\begin{aligned} Ln(St/Co) &= (K_1 - K_2C_i/Co)t \\ Ct/Co &= e^{(K_1\ K_2C_i/Co)t} \\ Ct &= Co\, e^{(K_1\ K_2C_i/Co)\, t} \end{aligned} \quad (2)$$

To characterize quantitatively the transfer of an element from soil to plant, the soil–plant Partition Coefficient[8] or Transfer Factor(TF) or Concentration Ratio or Biological Accumulation Coefficient(BAC) that expresses the ratio of contaminant concentration in plant parts to concentration in dry soil can be used[7,15,22,25,8,50,17,51,55].

TF = CPlant/CTotal_Soil.

(2) Becomes

$$Ct = Co\, e^{(K_1 - K_2TF)t}$$

CONCLUSION

High correlation between heavy metal content in soil and in plants was found for all studied soils and crops. Species of plants, type of soil and physico chemical properties of heavy metals determine the most important parameters of this dependence.

Couch grass is a crop, suitable for growing in industrially polluted regions, as they remove considerable quantities of heavy metals from the soil with their root system and can be used as potential crops for cleaning soil from heavy metals.

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REFERENCES

[1] V.Angelova, R.Ivanova, V.Delibaltova, K.Ivanov; Industrial Crops and Products, **19**, 197-205 **(2004)**.
[2] S.A.Barber; Soil Science, **93**, 39-49 **(1962)**.
[3] P.H.T.Beckett; Advances in Soil Science, **9**, 143-176 **(1989)**.
[4] M.J.Blaylock, J.W.Huang; 'Phytoextraction of Metals', In: I.Raskin, B.D.Ensley editors; 'Phytoremediation of Toxic Metals: using Plants to Clean-up the Environment', New York, Wiley, 53-70 **(2000)**.
[5] A.Boularbah, C.Schwartz, G.Bitton, W.Aboudrar, A.Ouhammou, J.L.Morel; Chemosphere, **5**, 811-7 **(2005)**.
[6] S.Cai, L.Yue, Z.N.Hu, X.Z.Zhong, Z.L.Ye, H.D.Xu; Science of the Total Environment. **90**, 67-73 **(1990)**.
[7] A.C.Chamberlain; Atmospheric Environment, **17**, 693-706 **(1983)**.
[8] K.Chojnacka, A.Chojnacki, H.Gorecka, H.Gorecki; Science of the Total Environment, **337**, 175-182 **(2005)**.
[9] S.B.Clark, W.H.Johnson, M.A.Malek, S.M.Serkiz, T.G.Hinton; Radiochimica Acta, **74**, 173-179 **(1996)**.
[10] Y.J.Cui, Y.G.Zhu, R.H.Zhai, D.Y.Chen, Y.Z.Huang, Q.YI, J.Z.Liang; China, Environment International Long, **30(6)**, 785-791 **(2004)**.
[11] P.Das, S.Samantaray, G.R.Rout; Environ.Pollut., **98**,

29-36 **(1997)**.

[12] P.Derache ; Toxicologie & Securite Des Aliments: Mecanisme d'Action des Toxiques, Lavoisier, Tec & Doc **(1986)**, deuxireme triage, 39-64 **(1989)**.

[13] G.M.Desmet, L.R.Van Loon, B.J.Howard; Science of the Total Environment, **100**, 105-124 **(1991)**.

[14] B.Devkota, G.H.Schmidt; Greece.Agric.Ecosyst. Environ., **78**, 85-91 **(2000)**.

[15] E.Dinelli, A.Lombini; Appl.Geochem., **11**, 375- 85 **(1996)**.

[16] S.Dudka, M.Piotrowska, H.Terelak; Environmentul Pollurion, **94(2)**, 181-188 **(1996)**.

[17] S.Ehlken, G.Kirchner; Radioactivity., **58**, 97-112 **(2002)**.

[18] EPA; 'Drinking Water Standards for Arsenic', United States Environmental Protection Agency, **815**, 00-015 **(2001)**.

[19] A.O.Fayiga, L.Q.Ma, X.Cao, B.Rathinasabapathi L.Environmental Pollution. **132**, 289-296 **(2004)**.

[20] H.L.Frost, L.H.Ketchum; Adv.Environ.Res., **4**, 347-355.

[21] C.Garbisu, I.Etalkorta; Eur.J.Min.Proc.Environ. Protect., **13**, 58-66 **(2000)**.

[22] C.H.Gast, E.Jansen, J.Bierling, L.Haanstra; Chemosphere, **17**, 789-799 **(1988)**.

[23] C.Gisbert, R.Ros, A.De Haro, D.J.Walker, M.P. Bernal, R.Serrano, J.Navarro-Avino; Biochem. Biophys.Res.Commun., 4: **303(2)**, 440-5 **(2003)**.

[24] M.Halim, P.Conte, A.Piccolo; Chemosphere, **52(1)**, 265-275 **(2003)**.

[25] M.D.Ho, G.J.Evans; Environmental Science and Technology, **34(6)**, 1030-1035 **(2000)**.

[26] B.K.Hope; Chemosphere, **30(12)**, 2267-2287 **(1995)**.

[27] F.Itanna; Ethiopian.J.Health Dev., **16(3)**, 295-302 **(2002)**.

[28] T.Jin, M.Nordberg, W.Frech, X.Dumont, A. Bernard, T.Ye; Bio.Metals., **15**, 397– 410 **(2002)**.

[29] A.Kabata-Pendias, H.Pendias; 'Trace Elements in Soils and Plants', Boca Raton, FL7 CRC Press, **(1984)**.

[30] C.Kheboian, C.F.Bauer; Analytical Chemistry, **59**, 1417-1423 **(1987)**.

[31] S.Knasmuller, E.Gottmann, H.Steinkellner, A.Fomin, C.Pickl, A.Paschke, R.God, M.Kundi; Mutat.Res., **420**, 37-48 **(1998)**.

[32] D.L.Lake, P.W.W.Kirk, J.N.Lester; J.Environ. Qual., **13**, 175-183 **(1984)**.

[33] A.V.Litvinovich, O.Yu.Pavlova; Agrochimia, **12**, 105-110 **(1995)**.

[34] E.Lombi, K.L.Tearall, J.R.Howarth, F.J.Zhao, M.J. Hawkesford, S.P.McGrath; Plant.Physiol., **128**, 1359-1367 **(2002)**.

[35] X.X.Long, X.E.Yang, W.Z.Ni; J.Appl.Ecol., **13**, 757-762 **(2002)**.

[36] L.Q.Ma, K.M.Komar, C.Tu, W.Zhang, Y.Cai, E.D.Kennelly; Nature, **409**, 579 **(2001)**.

[37] S.Madyiwa, M.Chimbari, J.Nyamangara, C.Bangira; Phys.Chem.Earth Parts, **27**, 747-753 **(2002)**.

[38] E.S.Mangwayana; 'Heavy Metals Pollution from Sewage Sludge and Effluent of Soil and Grasses at Crowborough Farm', B.Sc.Thesis. University of Zimbabwe, **63** **(1995)**.

[39] J.M.Martin, P.Nirel, A.J.Thomas; Marine Chemistry, **22**, 313-341 **(1987)**.

[40] M.J.Mc Laughlin, D.R.Parker, J.M.Clark; Field.Crops.Res., **60**, 143-63 **(1999)**.

[41] M.Muchuweti, J.W.Birkett, E.Chinyanga, R.Zvauya, M.D.Scrimshaw, J.N.Lester; Agriculture, Ecosystems and Environment **112(1)**, 41-48 **(2005)**.

[42] G.L.Mullins, C.H.Burmester; Better.Crops.Plant Food, **4**, 6-8 **(1991)**.

[43] P.M.V.Nirel, F.M.M.Morel; Water Research, **24**, 1055-1056 **(1990)**.

[44] G.F.Nordberg, T.Jin, Q.Kong Q, T.Ye, S.Cai, Z.Wang; Science of the total Environment, **199**, 111-114 **(1997)**.

[45] P.H.Nye, P.B.Tinker; The Journal of Ecology, **66(3)**, 1057 **(1977)**.

[46] A.L.Pag, R.H.Miller, D.R.Keeney; 'Methods of Soil Analysis-II', Chemical and Microbiological Properties, 2nd edn. ASA, SSSA, Madison, WI, USA, 1159 **(1982)**.

[47] W.F.Pickering; 'Selective Chemical Extraction of Soil Components and Bound Metal Species', CRC Critical Reviews in Analytical Chemistry, **12**, 233-266 **(1981)**.

[48] P.Quevauviller; The Science of the Total Environment, **178**, 1-145 **(1996)**.

[49] G.Riise, H.E.Bjornstad, H.N.Lien, D.H.Oughton, B.Salbu; Journal of Radioanalytical and Nuclear Chemistry, **142**, 531-538 **(1990)**.

[50] M.C.Roca, V.R.Vallejo; J.Environ Radioact, **28(2)**, 141-59 **(1995)**.

[51] P.B.Rodriguez, F.V.Tome, J.C.Lozano; Sci.Total. Environ, **284**, 167-175 **(2002)**.

[52] J.A.Ryan, H.R.Pahren, J.B.Lucas; Environmental Research, **28**, 251-302 **(1982)**.

[53] J.L.Schnoor; Environmental Modeling: Fate and Transport of Pollutants in Water, Air, and Soil. New

York, Wiley **(1996)**.

[54] D.Scott, J.Keoghan, B.E.Metallan; N.Z.J.Agric. Res., **39**, 499-512 **(1996)**.

[55] F.V.Tome, M.P.B.Rodriguez, J.C.Lozano; J.Environ.Radioact, **65**,161-75 **(2003)**.

[56] D.Voutsa, A.Grimanis, C.Samara; Environ.Pollut., **94**, 325-335 **(1996)**.

[57] T.Watanab, Z.W.Zhang, J.B.Qu, W.P.Gao, Z.K.Jian, S.Shimbo; Science of the total Environment., **247**, 1-13 **(2000)**.

[58] T.Watanabe, Z.W.Zhang, J.B.Qu, G.F.Xu, J.J.Wang, S.Shimbo; Science of the Total Environment. **217**, 1-8 **(1998)**.

[59] J.E.Watson, I.L.Pepper, M.Unger, W.H.Fuller; J. Environ.Qual., **14**, 174-177 **(1985)**.

[60] S.Xiao-Quan, C.Bin; Analytical Chemistry, **65**, 802-807 **(1993)**.

[61] E.Yang Xiao, Xiao-Fen Jin, Ying Feng, Ejazul Islam; Journal of Trace Elements in Medicine and Biology, **18**, 339-353 **(2005)**.

[62] X.E.Yang, X.X.Long, W.Z.Ni; J.Plant.Nutr.Fert, **8**, 8-15 **(2002)**.