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Tolerance of *Tithonia diversifolia* and *Chromolaena odorata* in heavy metal simulated-polluted soils and three selected dumpsites

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ABSTRACT

Soil heavy metals pollution is of global concern in view of their flow through the food chain. The convectional, physical, and chemical approaches to remediate polluted soils are usually expensive and not eco-friendly. Phytoextraction is a promising alternative because of the cost effectiveness and eco-friendliness. Therefore, this study was designed to determine the abilities of *Chromolaena odorata* (Co) and *Tithonia diversifolia* (Td) to extract heavy metals from polluted soil.

Soil analysis was done in part per million (ppm) before Td and Co were grown separately on sixty soil samples of 6.5 kg each collected from 3 selected dumpsites in Irese, New Stadium, and a control site (Ijare), Akure in Ondo State. Three replicates of the soil samples collected from Ijare were simulated with 10 g/kg each of cadmium (Cd), zinc (Zn), iron (Fe), copper (Cu) and lead (Pb) and used in assessing the phytoextraction capabilities of Co and Td. Two grams per kilogram of each of the heavy metals were mixed together and used in simulating three soil samples collected from Ijare and phytoextraction capabilities of Co and Td assessed. A control experiment using soil sample from Ijare was also set up in three replicates for Co and Td. Soil analyses were also carried out after the experiment in ppm. Plant biomass was assessed. Heavy metal contents in roots, shoots and soil samples were analyzed after harvest to determine the bioaccumulation (BF). Data obtained were subjected to one way Analysis of Variance at $\alpha_{0.05}$.

The most contaminated soil sample before and after the experiment was obtained in soil sample (mg/kg) from OD with Cd = 0.08 and 0.071, Zn = 1.92 and 1.85, Fe = 8.44 and 6.94, Cu = 3.04 and 2.54 and Pb = 1.4 and 0.93 respectively. The highest fresh and dry weights (g) for Td and Co were recorded in the plants grown in the control soil. The fresh weight (g) for Td and Co shoots and roots were 110.58 and 52.90; and 48.41 and 7.18 respectively. The dry weights (mg) for Td and Co shoots and roots were 20.56 and 4.68; and 16.66 and 0.36 respectively. Uptake of heavy metals in *T. diversifolia* and *C. odorata* tissues (ppm) were Cd (0.43 and 0.06), Zn (6.57 and 3.8), Cu (3.93 and 2.21), Pb (2.37 and 1.94) and Fe (55.15 and 32.82) respectively.

This study showed *Tithonia diversifolia* and *C. odorata* were capable of reducing heavy metals in polluted soils. Thus the plants are good candidates for the phytoextraction of heavy metals from polluted soils.

1. Introduction

Heavy metal toxicity is a menace capable of reducing growth and development of plants, and seriously causing injury or hazard to the health of animals and humans [1] through their involvement in the food chain. Heavy metals, unlike organic contaminants, cannot be biodegraded. As a consequence, they accumulate in the environment [2]. In addition, HM from the environment also migrates toward and accumulates in the living organisms by the processes of bioaccumulation and biomagnifications through the trophic levels of the ecosystem [2].

Some Africa countries like Nigeria is rich with coal deposit especially in Enugu State. This coal is also one of the most important pillars of China's economy [2].The coal mining activity produces a large amount of coal gangues, from which toxic substances could be released during the destruction of gangue mineral structure under the combined effects of water, microorganisms, vegetation, sunlight radiation and heat [3].

Plant growth and development is the primary source of energy for stability and functionality for plants which is photosynthesis. Therefore,

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deviation of plant from normal structure in photosynthesis may result in the reduction of plant growth or biomass [4]. The ions of the heavy metals inhibit multiple metabolic processes in plants which are shown by changes in many chlorophyll *a* fluorescence parameters [5]. Some metals such as Fe, Pb, Hg, Cu, Cd, Ni or Zn may substitute the central Mg in chlorophyll molecule, consequently lowering PSII quantum efficiency [4]. Therefore, the formation of these chlorophyll-metal ions complexes is leading to a loss in efficiency in conversion of light energy into photochemical reaction [20]. The influence of particular heavy metal ion on the physiological features of plant may be different but in the case of naturally polluted soils, all pollutants act synergistically so detection is only possible on the cumulative effect of their influence. Our experiment was performed using simulated-polluted soil and selected dumpsites from rural and urban region of Ifedore and Akure south respectively. The experiment was undertaken to study the effect of the concentration of soil heavy metals ions on the growth and biomass *T. diversifolia* and *C. odorata*. The acquired knowledge is to be used in planning of biomass processing systems and, in parallel, phytoextraction of contaminated soils, using hyperaccumulator plants.

2. Materials and methods

The experiment was carried out in Green House of Bethel College of Education, Ijare, Ondo-State. The College lies between latitude 6°20'N and 7°50'E with mean value temperature of 33.5 °C

The treatments followed a complete randomized design (CRD) with three replications.

The seeds of *T. diversifolia* and *C. odorata* were collected from the wild planted in all the contaminated and simulated soils for the phytoextraction of Cd, Pb, Cu, Fe and Zn.

Sixty (60) soil samples were used for this study. Soil samples were collected from four sites [Irese dumpsite (ID), New Stadium Dumpsite (NSD), Onyarugbulem Dumpsite (OD) and Ijare (control site)]. Soil samples collected from each site were further sub-divided to accommodate two plant species (*T. diversifolia* and *C. odorata*) used for this study.

Phytoextraction experiments were carried out in polythene bags in Green House. For this purpose, the soil samples were air-dried and sieved through a 2 mm wire net sieve. *Tithonia diversifolia* and *Chromolaena odorata* were grown separately on sixty soil samples of 6.5 kg each collected from 3 selected dumpsites; Irese dumpsite (ID), New Stadium Dumpsite (NSD) and Onyarugbulem Dumpsite (OD) and a control site, Ijare (IJ). Three replicates from each dumpsite were assessed for phytoextraction using both plant species. Three replicates of the soil samples collected from IJ were simulated with 10 g/kg each of cadmium (Cd), zinc (Zn), iron (Fe), copper (Cu) and lead (Pb) and used in assessing the phytoextraction capabilities of Co and Td. Two grams per kilogram of each of the heavy metals were mixed together and used in simulating three soil samples collected from IJ and phytoextraction capabilities of Co and Td assessed. A control experiment using soil sample from IJ was also set up in three replicates for Co and Td.

Fifteen seeds of *T. diversifolia* and *C. odorata* were broadcast into each polythene bag for each subunit respectively. Each treatment was represented in triplicates.

The plant growth parameters studied and measured included shoot length, root length, stem girth, and plant biomass after maturity. Metre rule was used to measure shoot length and root length, stem girth was measured using venier caliper and weight was carried out using electronic digital weighing balance.

Each experiment was conducted in triplicate (n = 3). Results were shown as mean ± standard error. Experimental data were analyzed using were analyzed using atomic absorption spectrophotometer after digestion with aqua regia. While the descriptive analysis were done using statistical software SPSS version 16.0. Heavy metal concentrations in control and experiment were compared using Duncan Multiple Range Test.

Table 1
Soil analysis before experiment (mg/kg).

Parameters	Ijare	Irese dumpsite	New stadium dumpsite	Onyarubulem dumpsite
pH	6.78	6.23	5.7	5.2
% O.C	0.05	5.21	0.02	0.016
% T.N	0.15	7.24	0.08	0.06
g/kg P	0.03	0.05	0.014	0.011
% Sand	63.74	83.31	93.21	96.11
% Silt	13.98	1.68	2.69	1.89
% Clay	22.28	15.04	4.04	2.03
Cmol/Kg Ca	2.1	0.01	0.009	0.007
Cmol/Kg Mg	1	1.11	0.004	0.002
Cmol/Kg K	0.38	0.25	0.001	0.001
Cmol/Kg Na	0.21	0.56	0.007	0.003
PPM Cd	0.01	0.01	0.07	0.08
PPM Zn	0.54	0.09	0.48	1.92
PPM Fe	354	0.99	10.06	8.44
PPM Cu	0.1	1.11	2.42	3.04
PPM Pb	0.44	0.2	0.4	1.4

3. Results and discussion

The physical and chemical soil properties from the locations are presented in Table 1. Heavy metals concentration in simulated-polluted and the selected dumpsites soil did not exceed WHO and ERS limits for arable soil before simulation. Soils used in this experiment were almost similar in pH value, organic matter contents and different in clay and sand amounts. The above result agrees with the work of Żurek et al. [4]. From the above soil analysis before experiment, the heavy metals were heavily present in the simulated soils than in the already polluted soil samples (S_2, S_3, S_4, S_5, $S_6 > S_1$, S_7, S_8, S_9 and S_{10}). The soil samples had high quantity of Iron (Fe) before simulation except for the dumpsite soil samples (S_8, S_9 and S_{10}). (Figs. 1 and 2)

Simulated-polluted soils of *T. diversifolia* and *C. odorata* had significant reduction in total chlorophyll content. Reduction in chlorophyll concentration in this study could be attributed to the inhibitory effect of accumulated ions.

The germination percentage of a seed may be influenced by the living environment. The present study established the effect of heavy metals on the germination of *T. diversifolia* and *C. odorata* as shown in Table 2 and 3 respectively. The results of this study indicated that the control soil treatment for *T. diversifolia* and *C. odorata* had the highest germination percentage with 93.33% and 86.67% respectively. This may be due to low concentration of heavy metals and high presence of iron (Fe). Germination was not recorded for soil treatment obtained from Onyarubulem dumpsite (OD) for both *T. diversifolia* and *C. odorata* which may be due to the spent engine oil level present in the soil which did not allow the percolation of water into the soil or delayed breaking of seed dormancy. The lowest germination percentage was evaluated in the soil treatment simulated with cadmium (13.33%) for both *T. diversifolia* and *C. odorata.* The decrease in germination of *T. diversifolia* and *C. odorata* seeds may be related to harmful effect of cadmium on water uptake and water movement [6]. The minimal seed germination may also be as a result of available enzyme inside the seed and acid phosphate that delays seed germination [7] which leads to reduced ATP production. The most toxic metals for seed germination was Cd, followed by Cu, and Pb. Delayed germination was also observed in all cases except in the control soil treatment.

The correlation between the mean relative growth rate (mRGR) and the biomass for Td and Co were very strong. However, Co had a better correlation than Td at shoot (0.97 and 0.85) and root (0.91 and 0.79) level respectively. These showed that the two plants experienced no disturbance despite the heavy metals present which contributed immensely to the product of the plant species.

Claire et al [8] obtained similar results in a study using nickel and other heavy metals on cabbage, lettuce, millet, radish, turnip, and

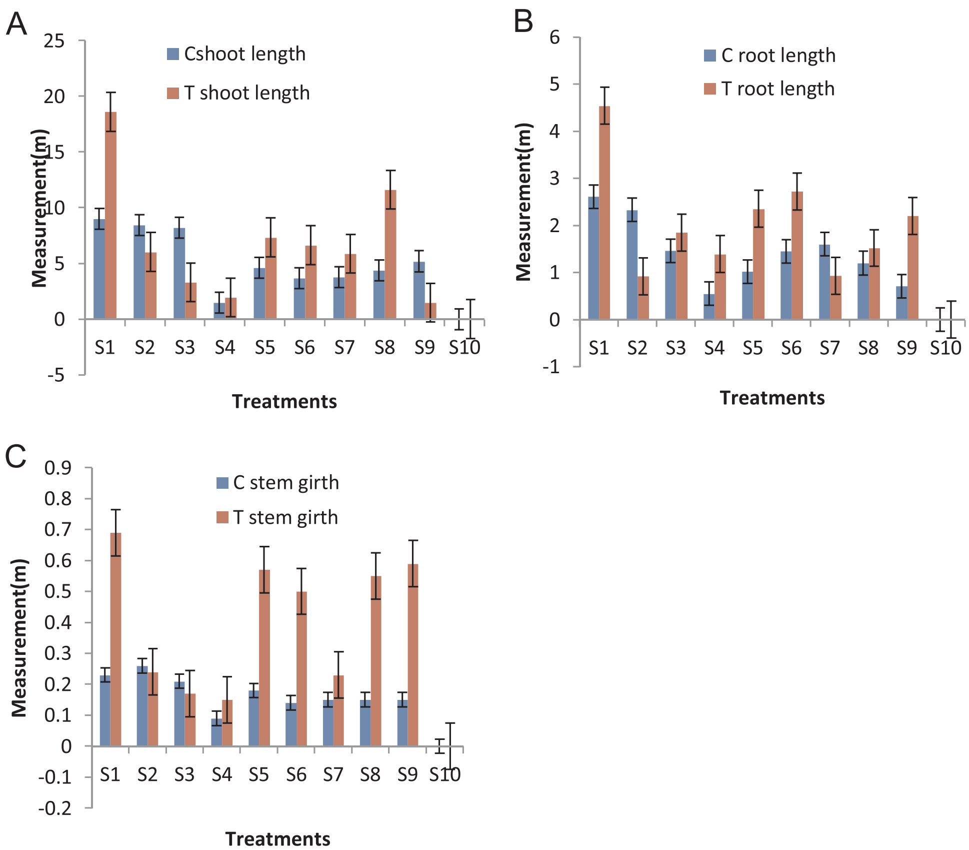

Fig. 1. (a) Comparison of shoot length between *C. odorata* and *T. diversifolia* with Error bars present as the standard deviation of the mean. (b) Comparison of root length between *C. odorata* and *T. diversifolia* with Error bars present as the standard deviation of the mean. (c) Comparison of stem girth between *C. odorata* and *T. diversifolia* with Error bars present as the standard deviation of the mean.
Note: C shoot length = shoot length of *C. odorata*, T shoot length = shoot length of *T. diversifolia*.
C root length = root length of *C. odorata*, T root length = root length of *T. diversifolia*
C stem girth = stem girth of *C. odorata*, T stem girth = stem girth of *T. diversifolia*
S_1 (Control: no simulation), S_2 (soil simulated with 2 g each of all heavy metal), S_3 (soil simulated with Iron-Fe), S_4 (soil simulated with Cadmium-Cd), S_5 (soil simulated with Lead-Pb), S_6 (soil simulated with Copper-Cu), S_7 (soil simulated with Zinc-Zn), and dumpsite soils; S_8 (soil collected from Irese dump site-ID), S_9 (soil collected from New stadium dump site-NSD), S_{10} (soil collected from Onyarubulem dump site-OD).

wheat. The heavy metals (Cd and Ni) at higher concentration inhibit the seed germination and growth of sunflower plant [9]. The failure of the seeds of *T. diversifolia* and *C. odorata* to germinate at soil treatment got from Onyarubulem dumpsite (OD) may be as a result of retarded water uptake, inhibited cell divisions and enlargements in the embryo and or an overall decrease in metabolic activity relevant to these steps. The blockage of any one of the phases may inhibit the germination process [10]. In this respect, Bazzaz et al. [11] demonstrated that interference with stomata function is a primary mode of action of cadmium and several other heavy metals. (Tables 4 and 5)

The highest value of heavy metals (Cd, Zn, Cu and Fe) in the root and shoot was found in *T. diversifolia* except for Pb that had highest value in *C. odorata*. Concentrations of heavy metals (Cd, Zn, Cu, Pb and Fe) in roots of the studied plants were higher than the concentration in the shoots. Özkan [12] also established this fact.

Metal concentrations in the tissue of the plants varies according to the treatment but not above the threshold limit according to National Standard Environmental Regulatory Agency (NSERA), European Regulatory Standard (ERS), World Health Organization (WHO) etc

For a plant to be efficient tool in phytoextraction of heavy metals in a polluted soil, the bioaccumulation factor and translocation factor must be higher than one [13,14] which is in agreement with our

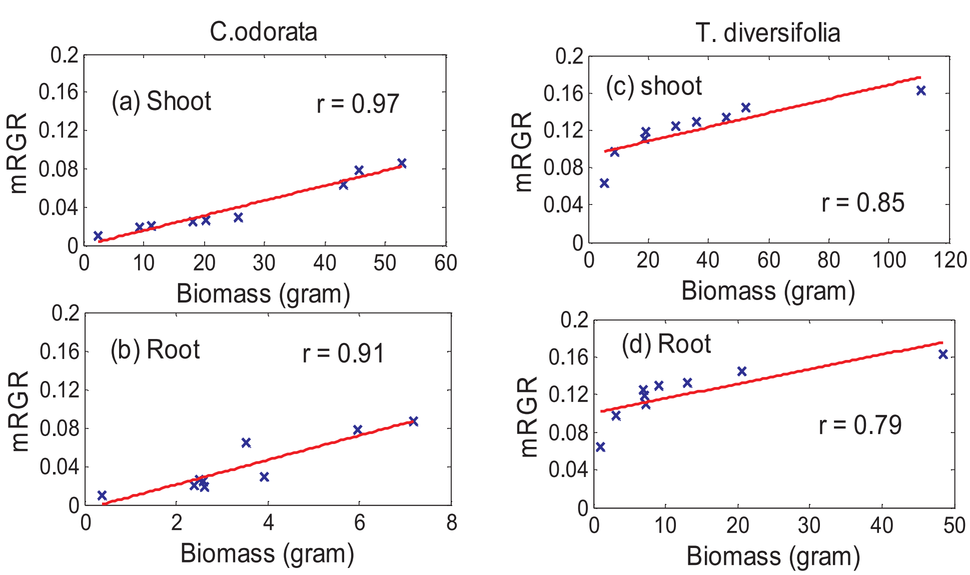

Fig. 2. Correlation between mRGR and Biomass : for *C. odorata* (a and b) and *T.diversifolia* (c and d).

Table 2
Effects of heavy metals concentrations on the biomass of *T. diversifolia*.

Parameters	SL (m)	RL (m)	SG (m)	SW (FW)	RW (FW)	SW (DW)	RW (DW)
T1	1.855^{g}	0.454^{f}	0.069^{c}	110.58^{g}	48.41^{g}	20.56^{g}	16.66^{e}
T2	0.603^{c}	0.092^{a}	0.024^{a}	19.13^{c}	7.03^{c}	3.85 ^{c}	1.18^{bc}
T3	0.330^{b}	0.185^{c}	0.017^{a}	8.82^{ab}	3.06^{b}	1.32^{b}	0.59^{b}
T4	0.194^{a}	0.139^{b}	0.015^{a}	5.58^{a}	1.02^{a}	0.92^{a}	0.22^{a}
T5	0.733^{d}	0.235^{d}	0.057^{b}	36.2^{d}	9.12^{de}	8.72^{e}	1.56 ^{c}
T6	0.663^{cd}	0.272^{e}	0.050^{b}	29.15^{cd}	6.96 ^{c}	5.24^{d}	1.49 ^{c}
T7	0.586^{c}	0.093^{a}	0.023^{a}	18.61^{c}	7.21^{d}	3.55 ^{c}	0.69^{b}
T8	1.160^{e}	0.152^{bc}	0.055^{b}	45.83^{e}	12.99^{e}	10.08^{ef}	3.04^{d}
T9	1.470^{f}	0.220^{d}	0.059^{b}	52.29^{f}	20.64^{f}	11.45^{f}	3.25 ^{d}
T10	–	–	–	–	–	–	–

T_1 (Control: no simulation), T_2 (soil simulated with 2 g each of all heavy metal), T_3 (soil simulated with Iron-Fe), T_4 (soil simulated with Cadmium-Cd), T_5 (soil simulated with Lead-Pb), T_6 (soil simulated with Copper-Cu), T_7 (soil simulated with Zinc-Zn), and dumpsite soils; T_8 (soil collected from Irese dumpsite-ID), T_9 (soil collected from New stadium dumpsite-NSD), T_{10} (soil collected from Onyarubulem dumpsite-OD). SL = shoot length, RL = root length, SG = stem girth, SW = shoot weight, and RW = root weight. Means followed with the same letters down the columns are not significantly different at P = 0.05 according to Duncan's multiple range test.

Table 3
Effects of heavy metals concentrations on biomass of *C. odorata*.

Parameters	SL (m)	RL(m)	SG (m)	SW (FW)	RW (FW)	SW (DW)	RW (DW)
C1	0.900^{g}	0.261^{e}	0.023^{de}	52.9^{f}	7.18^{e}	4.68	0.36^{e}
C2	0.842f	0.233^{d}	0.026^{e}	45.63^{e}	5.98^{d}	0.93^{c}	0.28^{d}
C3	0.821^{f}	0.146^{c}	0.021^{cd}	43.21 ^{e}	3.52^{c}	0.85 ^{c}	0.26 ^{d}
C4	0.146^{a}	0.055^{a}	0.009^{a}	2.41^{a}	0.39^{a}	0.10^{a}	0.05^{a}
C5	0.459^{d}	0.102^{b}	0.018^{bc}	20.43^{cd}	2.51^{b}	0.59^{bc}	0.22^{c}
C6	0.366^{b}	0.145^{c}	0.014^{b}	9.36^{b}	2.63^{bc}	0.17^{a}	0.08^{ab}
C7	0.378^{b}	0.160^{c}	0.015^{b}	11.30^{b}	2.40^{b}	0.38^{b}	0.09 ^{ab}
C8	0.436^{c}	0.120^{b}	0.015^{b}	18.10^{c}	2.60^{b}	0.45 ^{b}	0.12^{b}
C9	0.519^{e}	0.071^{a}	0.015^{b}	25.63^{d}	3.94^{c}	0.62^{bc}	0.22^{c}
C10	–	–	–	–	–	–	–

C_1 (Control: no simulation), C_2 (soil simulated with 2 g each of all heavy metal), C_3 (soil simulated with Iron-Fe), C_4 (soil simulated with Cadmium-Cd), C_5 (soil simulated with Lead-Pb), C_6 (soil simulated with Copper-Cu), C_7 (soil simulated with Zinc-Zn), and dumpsite soils; C_8 (soil collected from Irese dumpsite-ID), C_9 (soil collected from New stadium dumpsite-NSD), C_{10} (soil collected from Onyarubulem dumpsite-OD). SL = shoot length, RL = root length, SG = stem girth, SW = shoot weight, and RW = root weight. Means followed with the same letters down the columns are not significantly different at P = 0.05 according to Duncan's multiple range tests.

Table 4
Concentration of heavy metals in the shoot of *T. diversifolia* and *Chromolaena odorata* (mg/kg).

PARAMETERS	T.Cd	C.Cd	T.Zn	C.Zn	T.Fe	C.Fe	T.Cu	C.Cu	T.Pb	C.Pb
S1	0.05b	ND	0.84 g	0.81f	3.98 g	0.99d	0.53f	0.08bc	0.50c	0.07 cd
S2	0.1a	ND	0.31d	0.03a	1.44c	0.91c	0.11c	0.09 cd	0.05ab	0.03ab
S3	0.01a	ND	0.11b	0.08b	3.42f	0.52a	0.18d	0.03a	0.10b	0.03ab
S4	ND	ND	0.05a	0.07b	0.98a	0.98d	0.02a	0.11d	0.08ab	0.07c
S5	ND	0.01	0.14b	0.09bc	1.49c	2.13e	0.07b	0.16e	0.02a	0.10d
S6	0.01a	ND	0.24c	0.34e	1.25b	1.05d	0.06b	0.16e	0.02a	0.10d
S7	ND	ND	0.22c	0.11c	1.80d	1.02d	0.06b	0.06b	0.32bc	0.04b
S8	0.01a	ND	0.58f	0.15d	2.19e	0.98d	0.32e	0.20f	0.1ab	0.18e
S9	ND	ND	0.39e	0.08b	1.01a	0.77b	0.16d	0.01a	0.03ab	0.01a
S10	–	–	–	–	–	–	–	–	–	–

T = *T. diversifolia;* C = *C. odorata;* Cd = Cadmium; Zn = Zinc; Fe = Iron; Cu = Copper; Pb = Lead.
ND = Not detected.
S_1 (Control: no simulation), S_2 (soil simulated with all heavy metal with 2 mg each), S_3 (soil simulated with Iron-Fe), S_4 (soil simulated with Cadmium-Cd), S_5 (soil simulated with Lead-Pb), S_6 (soil simulated with Copper-Cu), S_7 (soil simulated with Zinc-Zn), and dumpsites; S_8 (Irese dumpsite-ID), S_9 (New stadium dumpsite-NSD), S_{10} (Onyarubulem dumpsite-OD).

findings.

Heavy metals are considered as the most dangerous elemental pollutants and are of particular concern because of their toxicities to human health [15].

This might also contribute to the performance of *T.diversifolia* and *C.odorata* in the phytoextracted soil.

Heavy metals (Fe, Zn, Cu, Cd and Pb) concentrations in plant tissues collected from the polluted soils (Figs. 3 and 4) revealed that the metal contents in the plant tissues differed among species from the heavy metal contaminated soil indicating their different capacities for heavy metal uptake.

The reduction in growth of *T.diversifolia* and *C.odorata* in the simulated-polluted soils could be due to decreased water uptake, toxicity of zinc, cadmium and copper ions as well as reduced photosynthetic rates [16]

Heavy metal accumulation in agricultural soils and water bodies poses a considerable risk to human health by different exposure pathways which could be direct ingestion, dermal absorption, inhalation and food chains [17,18]

Mohammed and Mohammed, [19], reported that the target hazard

Table 5
Concentration of heavy metals in the root of *T. diversifolia* and *Chromolaena odorata* (mg/kg).

PARAMETERS	T.Cd	C.Cd	T.Zn	C.Zn	T.Fe	C.Fe	T.Cu	C.Cu	T.Pb	C.Pb
S1	0.11	ND	0.91	0.26	6.31	2.4	0.66	0.1	0.2	0.1
S2	0.03	ND	0.48	0.09	3.69	2.87	0.27	0.11	0.1	0.1
S3	0.03	ND	0.19	0.31	5.91	1.87	0.25	0.07	0.2	0.1
S4	0.01	0.01	0.12	0.11	7.84	1.42	0.12	0.15	0.2	0.2
S5	ND	0.02	0.22	0.26	2.92	3.66	0.09	0.21	0.05	0.21
S6	0.01	ND	0.37	0.52	2.7	3.2	0.17	0.28	0.1	0.3
S7	0.01	0.01	0.36	0.17	2.77	2.77	0.12	0.09	0.1	0.09
S8	0.04	0.01	0.61	0.18	3.57	3.72	0.49	0.27	0.1	0.2
S9	0.01	ND	0.43	0.14	1.88	1.56	0.25	0.03	0.1	0.01
S10	–	–	–	–	–	–	–	–	–	–

T = *T. diversifolia;* C = *C. odorata;* Cd = Cadmium; Zn = Zinc; Fe = Iron; Cu = Copper; Pb = Lead.
ND = Not detected.
S_1 (Control: no simulation), S_2 (soil simulated with all heavy metal with 2 mg each), S_3 (soil simulated with Iron-Fe), S_4 (soil simulated with Cadmium-Cd), S_5 (soil simulated with Lead-Pb), S_6 (soil simulated with Copper-Cu), S_7 (soil simulated with Zinc-Zn), and dumpsites; S_8 (Irese dumpsite-ID), S_9 (New stadium dumpsite-NSD), S_{10} (Onyarubulem dumpsite-OD).

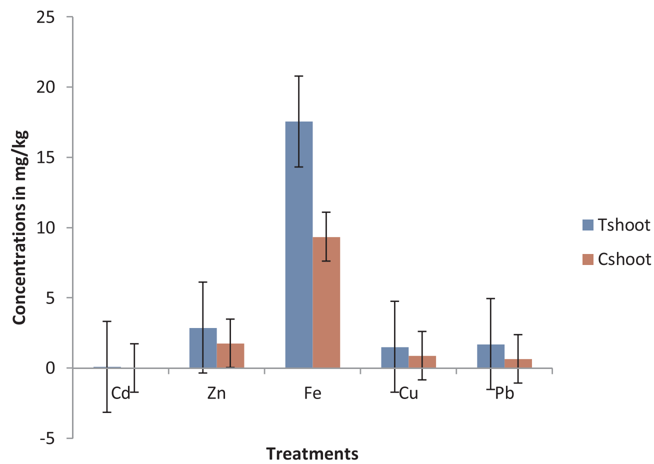

Fig. 3. Concentration of heavy metals in shoots of *T. diversifolia* and *C. odorata* T = *T. diversifolia*; C = *C. odorata.*

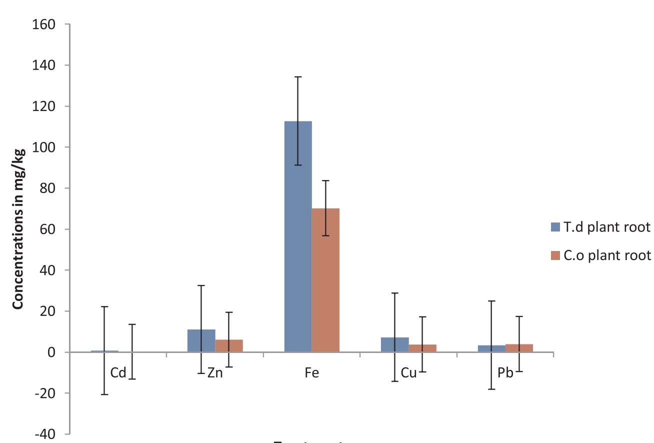

Fig. 4. concentration of heavy metals in roots of *T. diversifolia* and *C. odorata* T.d = *T. diversifolia*; C.o = *C. odorata.*

quotient (THQ) values for heavy metals were above 1 for both species, showing the estimated exposure is potentially of concern. In the context of the present study, THQ of Hg was found to be 2.53 due to the spiny dogfish consumption by the Greek population. So what can be seen is that although the mean concentrations of Hg were below recommended limits, THQ value of this metal was above 1 for the Greek consumer. Thus, it can be concluded that the elevated consumption may increase the risk, and estimated exposure could be considered as a potential concern.

4. Conclusion and recommendation

This study was conducted to screen plants growing on a contaminated site to determine their potential for metal accumulation. The plant species examined in this study grew very well and propagated quickly in substrata with Fe-contaminated, Pb-contaminated, Cu-contaminated soil conditions which would be an advantage in the re-vegetation of polluted soil as cost would be reduced without fertilizer.

The plant species can colonize heavy metal-contaminated sites, because they were able to absorb a wide range of soil heavy metals (Fe, Zn, Cu, Cd and Pb),and were not affected by high concentration of heavy metal contents which implies they have higher tolerance than more sensitive species.

This study showed *Tithonia diversifolia* and *C. odorata* reduced heavy metals in polluted soils. Thus the plants are good candidates for the phytoextraction of heavy metals from polluted soils. However, *Tithonia diversifolia* was considered as a promising species for phytoextraction of heavy metal-contaminated sites because of its performance.

The phytoremediation of *T. diversifolia*, especially for Irese dumpsite, needed to be investigated.

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