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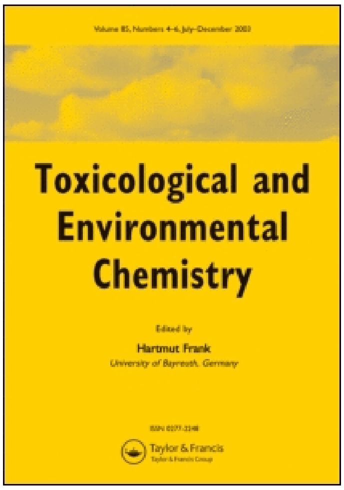

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Effect of inorganic and organo-mineral fertilizers on the uptake of selected heavy metals by *Helianthus annuus* L. and *Tithonia diversifolia* (Hemsl.) under greenhouse conditions

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A greenhouse experiment was conducted to assess the removal of lead (Pb), copper (Cu), and cadmium (Cd) from polluted soils using *Helianthus annuus* and *Tithonia diversifolia*. There were three pot trials consisting of different levels of Pb and Cu at 0, 125, 250, 500, 1000, 2000 $mg\,kg^{-1}$; and Cd at 0, 20, 40, 60, 80, 100 $mg\,kg^{-1}$ rates using soluble compounds of $(CH_3COO)_2Pb{\cdot}3H_2O$, $Cu(NO_3)_2{\cdot}3H_2O$, and $CdSO_4{\cdot}8H_2O$, respectively. Each heavy metal was replicated three times in Completely Randomized Design (CRD) with four different fertility application levels, namely: 300 $kg\,ha^{-1}$ of NPK 20-10-10, (Inorganic Fertilizer); 10 $t\,ha^{-1}$ of Grade A organo-mineral fertilizer (compost amended with additional N and P), control 1, without fertilizer application and control 2, where no fertilizer and no crop but heavy metals were applied. Pre- and post-cropping analyses of soils used were done. Data were analyzed using ANOVA. From the greenhouse experiment, highest remediated values of 0.83, 0.68, and 0.26 $mg\,kg^{-1}$ in the shoots of *H. annuus* for Cu, Pb, and Cd, respectively, were obtained when organo-mineral fertilizer was applied. Significantly ($p < 0.05$) lower values of 0.50, 0.43, and 0.25 $mg\,kg^{-1}$ of Cu, Pb, and Cd, respectively, were obtained with inorganic fertilizer application. *Tithonia diversifolia* shoots uptake were 0.20, 1.37, and 0.30 $mg\,kg^{-1}$ for Cu, Pb, and Cd, respectively, when OMF was applied. Significantly ($p < 0.05$) lower values were obtained with inorganic fertilizer applications in the shoot of *T. diversifolia*. *Helianthus annuus* remediated Cu and Cd better with organo-mineral fertilizer application while *T. diversifolia* remediated Pb better also with organo-mineral fertilizer application.

Keywords: phytoremediation; polluted soils; organo-mineral fertilizer; *Helianthus annuus*; *Tithonia diversifolia*

Introduction

Many industrial sites, especially in the sub-Saharan Africa are being contaminated with a variety of pollutants. Awareness of factory workers on the hazards these contaminated soil could cause on their health if cultivated for edible crops is very low, especially in Nigeria. Premises of a paint industry in Ibadan, southwestern Nigeria, which has been

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receiving factory's wastes and effluents continuously for over 40 years called for examination, more so when part of this polluted land is being used for cultivating yam, sugar cane, and some vegetables.

Also, many areas in the south-south and southwest geopolitical zones of Nigeria have large deposits of crude oil. Large deposit of organic pollutants in these areas is therefore inevitable. Between 1976 and 1990 alone, 784 spills involving 1.4 million barrels of oil in 2796 incidents were recorded in Niger Delta of Nigeria (Atoyebi 2000). In major cities like Lagos, Port-Harcourt and Kaduna, having over 80% of Nigeria industries, no attention is paid to soil and water ecosystems as the factories' effluents are diverted to the nearby lands and streams.

Heavy metals considered as environmental pollutants include: mercury (Hg), cadmium (Cd), lead (Pb), arsenium (As), nickel (Ni), copper (Cu), molybdenum (Mo), manganese (Mn), and boron (B) (Brady 1984). Environmental pollution with toxic metals has drastically increased since the beginning of the industrial revolution in Europe (Nriagu 1979). The toxicity levels of these heavy metals can be enhanced through the increasing use of pesticides (Graham et al. 1992), chemical fertilizers (Zhu and Alva 1993), and untreated sewage sludge (Preer et al. 1995). Soil pollution is also increasing daily as a result of human activities in mining, oil exploration, and agriculture. Therefore, the transfer of these heavy metals from the soil into the food chain is possible (Petrzzelli, Lubrano, and Guidi 1989). Presently in Nigeria, there is dearth of information on the phytoremediation technique using sunflower to remediate soils polluted with heavy metals.

Raskin (2003) defined phytoremediation as the use of green plants to remove pollutants from environment or render them harmless. Various crops, which include maize (Gigliotti, Businelli, and Ginsquiani 1996), okro and bitter leaf (Ademoroti 1996), vetch and barley (Chernykh 1991), *Brassica juncea* L. (Dushenkov et al. 1995), winter wheat (Yanshan et al. 2004), and *Helianthus annuus* and *Tithonia diversifolia* (Adewole 2006) have been used at various times to phytoremediate polluted soils.

Different elements in the soil vary in their availability to plants because of the difference in their solubilities and mobilities. Bioavailability depends on metal solubility in soil solution (Lasat 2002). In heavy metal phytoremediation, chelating agents are used to 'loosen' metals from the soil structure and make them more available (Blaylock et al. 1997). Prasad and Freitas (2003) suggested the use of fertilizer, acidifying agents, and chelating materials to enhance the bioavailability of heavy metals for their uptake from the soil. Yanshan et al. (2004) reported the enhanced uptake of Pb and Zn by Indian mustard (*B. juncea* L.) and winter wheat following combined soil application of elemental sulphur and EDTA.

The study therefore attempts to phytoremediate contaminated soils of variable concentrations using *H. annuus* L. and *T. diversifolia* (Hemsl.) when inorganic and organo-mineral fertilizers are used as ameliorating agents under greenhouse conditions.

Materials and methods

The study was conducted at Saki. Saki is located in the northern part of Oyo State in southwestern Nigeria. Bulk surface soil samples (0–15 cm) from an exhaustively cropped portion of Oyo State Agricultural Development Programme (OYSADEP) Seed farm at Saki was used. The sampled soil was air-dried for seven days; stones and debris were

Table 1. Results of soil analysis before planting.

Property	Value
pH (1 : 1 soil – water)	6.7
pH (1 : 1 soil – 1 M KCl)	6.3
Sand ($g\,kg^{-1}$)	558
Clay ($g\,kg^{-1}$)	82
Silt ($g\,kg^{-1}$)	360
Available phosphorus ($mg\,kg^{-1}$)	10.38
Organic carbon ($g\,kg^{-1}$)	1.148
Total nitrogen ($g\,kg^{-1}$)	0.238
Exchangeable cations ($cmol\,kg^{-1}$)	
Ca	0.87
Mg	0.86
Na	0.65
K	0.26
Exchangeable acidity ($cmol\,kg^{-1}$)	0.05
Effective CEC ($cmol\,kg^{-1}$)	2.69
Base saturation (%)	98.12
Extractable micronutrients ($mg\,kg^{-1}$)	
Fe	132.89
Mn	103.91
Cu	2.73
Zn	4.78
Heavy metals ($mg\,kg^{-1}$)	
Pb	0.03
Cd	0.03

removed and passed through 2 mm sieve. A pre-soil test was carried out to know the physicochemical properties of the soil.

Heavy metal solutions of Pb, Cu, and Cd of known concentrations were prepared using soluble compounds of $(CH_3COO)_2Pb \cdot 3H_2O$, $Cu(NO_3)_2 \cdot 3H_2O$, and $CdSO_4 \cdot 8H_2O$. The metal levels used were: Pb – 0, 125, 250, 500, 1000, 2000 $mg\,kg^{-1}$; Cu – 0, 125, 250, 500, 1000, 2000 $mg\,kg^{-1}$, and Cd – 0, 20, 40, 60, 80, 100 $mg\,kg^{-1}$. A total of 384 polythene pots with 5 kg of soil with drainage holes at the bottom were placed on the tables in the greenhouse randomly and the heavy metals doses were applied. *Helianthus annuus* (Funtua sunflower variety) and *T. diversifolia* (Mexican sunflower, which grows wildly in Nigeria) were used in the experiments. It was a factorial combination of Pb, Cu, and Cd of which each treatment was replicated thrice in completely randomized design (CRD) with four fertilizer application levels.

The heavy metal solutions were applied to degraded soil in the pots to field moisture capacity and left for four days to equilibrate. The main treatments applied in the experiment were:

(i) NPK 20-10-10 fertilizer (labeled as IOF) – this was purchased in the open market and applied at 0.75 g per pot.

(ii) Organo-mineral fertilizer (OMF Grade A) – this was obtained from Oyo State Government Pace-setter organic fertilizer plant, Bodija, Ibadan and has the composition 44.2 $g\,N\,kg^{-1}$, 5.0 $g\,P\,kg^{-1}$ and 1.5 $g\,K\,kg^{-1}$. This was applied at 25 g per pot.

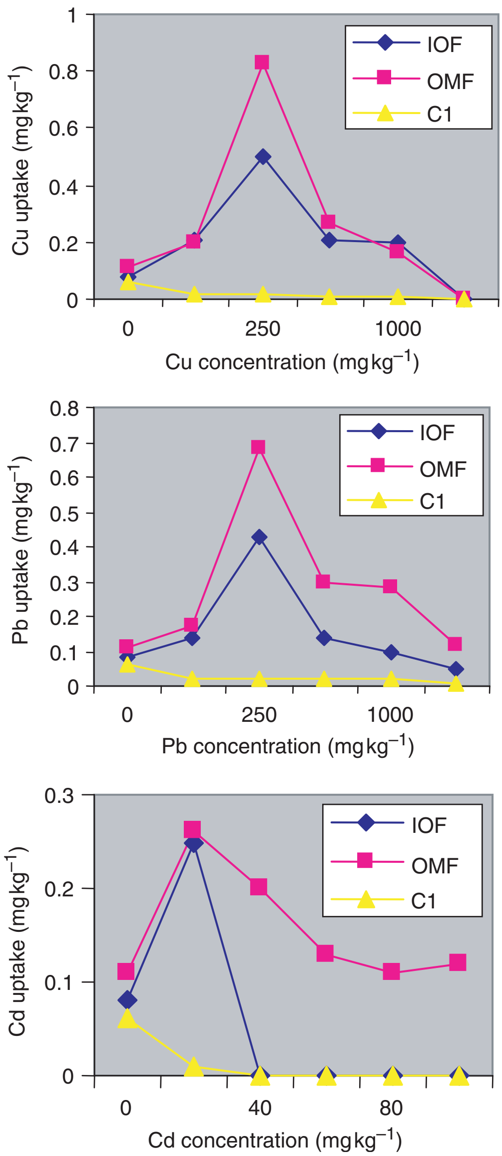

Figure 1. Effects of fertilizer treatments on the uptake of Cu, Pb, and Cd by Funtua sunflower in greenhouse experiment. Note: IOF = inorganic fertilizer; OMF = organo-mineral fertilizer; C1 = control 1 (no fertilizer was applied).

(iii) Control 1, soil with neither mineral nor organo-mineral fertilizer was applied (C1).
(iv) Control 2, soil with no planting and no fertilizer (C2).

The fertilized pots were equilibrated for three days before viable seeds of *H. annuus* (HA) and *T. diversifolia* (TD) were planted at the rate of four seeds per pot. After germination, the seedlings were thinned to two plants per pot at two weeks after planting (WAP). The thinned stands were put back in their respective pots. The plants

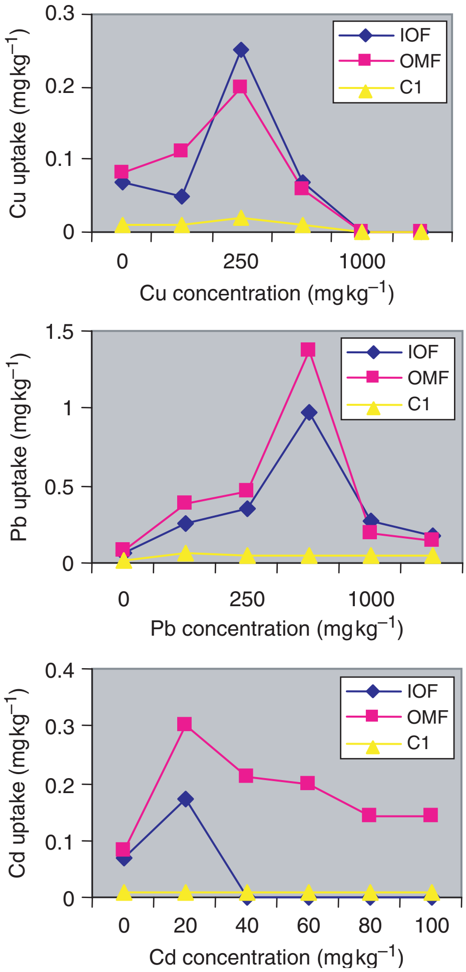

Figure 2. Effects of fertilizer treatments on the uptake of Cu, Pb, and Cd by wild sunflower in greenhouse experiment. Note: IOF = inorganic fertilizer; OMF = organo-mineral fertilizer; C1 = control 1 (no fertilizer was applied).

were regularly watered throughout the growing stage. At 8 WAP, plant samples (stem and leaves) were taken from all the levels of the treatments of Funtua sunflower and Mexican sunflower. The plant samples were washed, oven-dried for 48 h at 70°C, weighed, ground, and analyzed for nutrients and heavy metal uptake. The post-soil analysis was carried out to assess the heavy metal status of the soil samples used in the greenhouse.

Results and discussion

Soil analysis before planting

The pre-soil test of the degraded soil for the greenhouse experiment was presented in Table 1. The texture of the soil was sandy-loam and very slightly acidic with soil pH of 6.7 in 1:1 soil–water ratio. The base line Cu, Pb, and Cd values were 2.73, 0.03, and 0.03 mg kg^{-1}, respectively.

Table 2. Post-harvest levels of heavy metals in the experimental soils.

Heavy metal	Level of heavy metal (mg kg^{-1})	Fertilizer treatment X/Y (mg kg^{-1})			
		IOF	OMF	C1	C2
Cu	0	1.45/1.06	1.07/1.06	1.47/1.80	2.68/2.91
	125	108. 56/110.90	100.42/105.45	112.56/115.70	118.45/120.40
	250	183.25/220.20	170.20/183.83	220.83/200.06	235.70/235.57
	500	444.45/448.22	366.08/416.83	440.20/490.45	494.70/494.94
	1000	595.95/646.43	476.38/560.70	804.02/804.02	948.02/952.25
	2000	1228.07/1772.43	1080.70/1458.67	1628.07/1782.48	1880.45/1804.76
Pb	0	*	*	*	*
	125	106.40/110.55	100.25/102.25	116.35/120.50	121.07/122.00
	250	218.50/227.85	160.37/170.75	230.51/231.27	236.15/236.30
	500	247.55/295.45	244.50/372.27	360.27/392.50	490.15/485.86
	1000	564.23/593.88	535.00/572.27	623.13/735.17	962.62/970.86
	2000	1148.70/1150.65	1120.65/1190.89	1221.18/1352.06	1870.25/1871.00
Cd	0	*	*	*	*
	20	11.42/11.74	10.02/10.80	14.41/12.50	18.12/18.25
	40	32.51/33.60	30.42/30.75	35.75/36.85	37.00/38.51
	60	40.60/42.52	43.65/44.52	45.90/51.00	57.10/56.70
	80	60.50/63.50	46.20/55.60	62.20/62.48	70.33/73.00
	100	63.60/66.00	61.50/63.00	89.00/98.90	98.90/98.86

Note: *Values could not be detected; $X/Y = X$ stood for soil values for Funtua sunflower; Y stood for soil values for Mexican sunflower. C1 = Control 1; C2 = Control 2.

Removal of heavy metals by test crops

Application of increased concentrations of Cu, Pb, and Cd resulted in the increased uptake by the two test crops (HA and TD, Figures 1 and 2). When the soils were treated with OMF, the levels of uptake of Cu, Pb, and Cd (when applied at 250 mg kg^{-1}, 250 mg kg^{-1}, and 20 mg kg^{-1}) by *Helianthus* were 0.83 mg kg^{-1}, 0.68 mg kg^{-1}, and 0.26 mg kg^{-1}, respectively. With the application of IOF, *Helianthus* picked up much lower levels: 0.50 mg kg^{-1} Cu, 0.43 mg kg^{-1} Pb, and 0.25 mg kg^{-1} at 20 mg Cd. On the contrary, *Tithonia* removed these metals significantly less ($p < 0.05$) except for Pb, which was 1.37 mg kg^{-1} indicating a two-fold increase ($p < 0.05$). Addition of OMF enhanced metal solubility and mobility in the soil. As a result, more of Cu, Pb, and Cd were remediated when compared with the application of IOF. It was observed that as soil becomes more acidic, from a pH of 6.30 to pH 5.77 in case of OMF, and a pH of 5.87 in case of IOF, the binding forces of heavy metals into soil particles are presumably broken thereby enhancing their mobility. Previous studies of Johnson, Campbell, and Foy (1997), Lasat (2002), and Adewole (2006) had shown an increase in heavy metals uptake by plants with increase in soil acidity.

Table 2 shows the mean levels of Cu, Pb, and Cd remained in the soil after the test crops have been harvested and the experiment was terminated (after 16 weeks). Application of increased concentrations of heavy metals to the soil resulted in retention of higher levels in the soils once the test crops picked up optimally. This is evident from the controls where there were no test crops: the soil concentrations of heavy metals remained very high (Table 2). Absence of roots of growing plants that stimulate the growth of consortium (Lasat, Baker, and Kochian, 1998), and some soil microorganisms that help to increase heavy metals bioavailability and facilitate plant uptake of these heavy metals are the contributory factors. All the pots were maintained weed-free during the period of the experiment.

Conclusions

The inorganic and organic fertilizers enhanced the uptake of Cu, Pb, and Cd using *H. annuus* L. and *T. diversifolia* (Hemsl.) as phytoremediators. However, organo-mineral fertilizer performed better in the uptake of Cu and Cd using *H. annuus* and in Pb uptake using *T. diversifolia*.

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