Levels and mobility of Cu, Pb and Cd in citrus orchards of two contrasting ages in north-central region of Nigeria

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Summary

Widespread contamination of cultivated lands with heavy metals increase human exposure to contaminated agricultural products. The study investigated soil levels and transfer of Cu, Pb and Cd into leaves and barks of different citrus species in 35- and 11-year old orchards in north-central region of Nigeria. Heavy metals concentrations in soils were below permissible limits but greater than background concentrations (Cu = 2.27 mg kg^{-1}, Pb = 0.25 mg kg^{-1} and Cd = 3.9 mg kg^{-1}). Geo-accumulation index (I_{geo}) indicated that the 35-year orchard (orchard I) soil was uncontaminated with Cu (Cu_{-Igeo} = -0.43), uncontaminated to moderately contaminated with Pb (Pb_{-Igeo} < 0.39) and moderately contaminated with Cd (Cd_{-Igeo} = 1.65). The 11-year old orchard (orchard II) soil was uncontaminated to moderately contaminated (I_{geo} for Cu, Pb and Cd was ≤ 1). Bioavailability of the heavy metals soil of orchards I and II were ≥ 30%. Levels of Cd and Pb in bark and leaf tissues were above the maximum permissible limits whereas Cu concentration was within the recommended nutritional status (10 - 50 mg kg^{-1}) for fruit trees. Soil-to-tissue transfer (bioaccumulation) of all heavy metals into the citrus species was ≥ 1.0 and higher in orchard II. High concentration of heavy metals in the leaves and barks of the different citrus species is related to their high bioaccumulation factors despite the relatively low soil concentration of heavy metals in the orchards.

Key words

bioavailability, citrus, metal pollution, orchard, heavy metals

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Introduction

Heavy metals (HMs) constitute toxic components of particulate matter (Ite et al., 2014), that arise from either natural sources or from human-induced activities of industrialization, urbanization, inappropriate waste management and other local and/or regional human sources (Ite et al., 2016). The after-effects of HMs on soil systems and the immediate environment cannot be estimated simply by measuring the total concentration because only the dissolvable, mobile and bioavailable fractions have the potential to leach or to be absorbed by plants and enter the food chain (Tchounwou et al., 2012). The solubility, mobility and accumulation of trace elements in biological tissues depend on a number of soil, microbial and plant factors, as well as the physico-chemical properties of the target trace element (Bradl, 2005; Robinson et al., 2005).

Two major channels of human exposure to metals are the consumption of polluted crops and the ingestion or inhalation of metal particles in dusts. Therefore, it is important to have information on pollution status of soil and plant for assessing their threat to human health (Li et al., 2014). Heavy metals in agricultural soils such as orchard soils may be transferred to agricultural products. Consumption of agricultural products such as fruits, which is an important component of the human diet (Fang and Zhu, 2014), has been noted to be a major human exposure route to soil metal contamination (Fang and Zhu, 2014). Furthermore, exposure to HMs contamination has been implicated in several diseases such as gastrointestinal symptoms, coronary heart malady, diversity of cancers, and even death has also been associated with persistent and/or severe exposures to metals (Lamas et al., 2016). Therefore, an understanding of the transfer of HMs from orchard soils to fruit trees and subsequently to fruits (though this study did not cover this aspect) is very important. In addition, monitoring and assessment of HMs concentrations in the environment contribute effectively to the understanding of biogeochemical processes and gauging ecosystem health (Ite et al., 2014).

Premised on the aforementioned, the study was carried out in two orchards established at different times in north-central Nigeria in order to understand the influence of age on level and/or mobility of some HMs in citrus species as well as possible sources of contamination. The objectives of this present study are: (i) to determine the soil level and phyto-availability of Cu, Cd and Pb, and (ii) their mobility in tissues of some citrus species from 11- and 35-year old orchards.

Materials and methods

Description of orchards

The present study was carried out in two contiguous orchards, located in the expansive premises of the Lower Niger River Basin Development Authority (LNRBA), Ilorin, north-central Nigeria (Lat 8° 30° N; Long 4° 35° E) (Fig. 1). The city (Ilorin) and its soil-type is a ferruginous tropical soil type (Ogunkunle et al., 2016a). The LNRBA was established to support investment programme for public irrigation, and provides low cost techniques for tapping shallow groundwater to promote high yield and quality agricultural productions in drier northern parts and the middle belt of Nigeria. The LNRBA has ensured sustainable development and improvement of significant area through irrigated agriculture, rural and urban water supply, construction of dams and erosion control in the region (LNRBDA, 2002). The first studied site (orchard I) is a 35-year old orchard of an area of 12,544 m^2, and the second site (orchard II) is a 11-year old orchard of an area of 6,300 m^2. Agronomic practices in the orchards include mechanical weeding, irrigation by sprinkler, fertilization (in the early growth stage of the orchards) and phytosanitary control.

Method of sampling

In April 2015, systematic sampling was carried out in the orchards by demarcating the 35- and 11-year old orchards (orchards I and II) into 12 and 10 plots, respectively. In each plot, soil samples of approximately 200 g (≥ 4 subsamples) within 0 - 30 cm depth were collected using a stainless soil auger and bulked to give a single composite sample before being tagged. Composite soil sample was air-dried in the laboratory to constant weight, ground and sieved through a < 2 mm sieve to remove pebbles and larger particles.

In order to evaluate level of metal concentrations and metal mobility in tissues of citrus species from the orchards, citrus species in each orchard were enumerated to species level. Five Citrus species namely - sweet orange (*Citrus sinensis* (L.) Osbeck), mandarin orange (*Citrus reticulata* Blanco), grapefruit (*Citrus paradisi* Macfad), rough lemon (*Citrus jambhiri* Lush.) and lime (*Citrus aurantifolia* (Cristm.) Swingle) were identified in the 35-year old orchard (orchard I) while only three (3) species (*C. sinensis*, *C. reticulata*, and *C. jambhiri*) were recorded in the 11-year old orchard (orchard II). Six (6) heathy stands of each citrus species in the orchards were selected for sample collection. Samples of tree bark were collected in a circular pattern at 1 m height above the ground level from each citrus species with a stainless scalpel. Accordingly, same citrus stand was also sampled for matured but not senescing leaves (≥ 10 leaves) and properly tagged according to citrus species stands. The same sampling procedure was repeated for all the three species in the 11-year old orchard. Bark and leaf tissues from sampled citrus stand were dried in an oven at 45°C for 24 hours, ground into powder in a ceramic mortar and bulked to obtain a composite sample.

Physico-chemical analyses of soil and plant tissues

Soil pH was determined in water suspension (2.5:1 v/w) and electrical conductivity was measured using digital multi-parameter meter (HI-98129 model, Hanna Instruments). The soil organic matter content (SOM) in soil sample was measure by Loss-on-ignition method. This was done by measuring weight loss after weighing 5 g of soil sample in a crucible and heating the soil in a muffle furnace at 450°C for 24 h (Sutherland, 1998). Soil textural analysis was performed using hydrometer method by estimating various soil particle sizes (Ashworth et al., 2001).

Concentration of HMs (Cu, Cd and Pb) in the soil was determined according to the method of Intawangse and Dean (2006). Potential bioavailable metal fraction of soil sample was extracted by continuous shaking of 5 g of soil sample in 25 ml of 0.05 M EDTA for 1 hr (Quevauviller et al., 1997). Quantification of metal concentrations was carried out by Flame Atomic Absorption Spectrophotometry (FAAS-Perkin Elmer A Analyst 200).

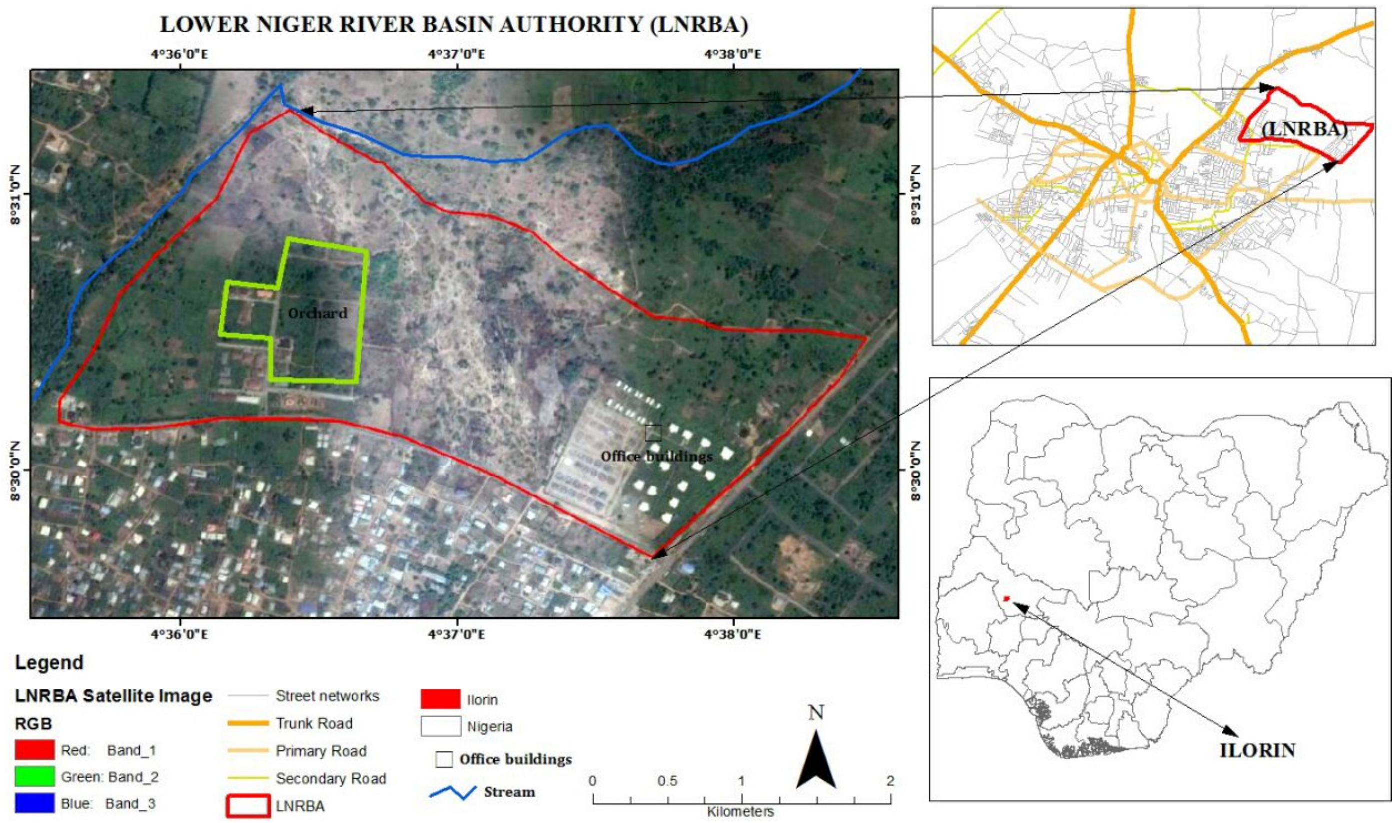


Figure 1. Map of Ilorin, Nigeria showing the location of the orchards

Leaf or bark tissues were digested according to the method of Intawangse and Dean (2006). Determination of metal concentrations (Cu, Cd and Pb) was carried out by Flame Atomic Absorption Spectrophotometry (FAAS-Perkin Elmer A Analyst 200). Quality assurance and quality control (QA/QC) for HMs concentrations in soil and citrus tissues was carried out using certified reference materials (CRMs) IAEA-SL-1 (soil) and IAEA-359 (cabbage leaves).

Soil contamination assessment

The degree of metal contamination in the orchard soils was determined by the geo-accumulation index (I_{geo}) as proposed by Muller (1969):

$$I_{geo} = log_2 [Cn / 1.5xBn] \quad \text{(Eq. 1)}$$

where Cn represents total metal concentration in orchard soil, Bn is the natural background soil HM concentration in the area (Botanical garden of University of Ilorin, approximately 15 kilometers from study area) and 1.5 is a correction factor. The background area is relatively uncontaminated. Based on I_{geo} categories, the contamination levels of soil are classified into seven grades: $I_{geo} = 0$ represents uncontaminated; $0 < I_{geo} \leq 1$, uncontaminated to moderately contaminated; $1 < I_{geo} \leq 2$, moderately contaminated; $2 < I_{geo} \leq 3$, moderately to heavily contaminated; $3 < I_{geo} \leq 4$, heavily contaminated; $4 < I_{geo} \leq 5$, heavily to extremely contaminated; $5 < I_{geo}$, extremely contaminated (Muller, 1969).

Mobility assessment of heavy metals in the citrus species

The ratio of concentrations between the plant parts (bark and leaf) and soil was estimated in the study by applying the bioaccumulation factor (BF) using following formula:

$$BF = C_p / C_s \quad \text{(Eq. 2)}$$

where C_p is the HM concentration in bark and leaf parts, and C_s is the bioavailable concentration of the same metal in the soil sample. If BF > 1, the citrus species is regarded as an accumulator. If BF = 1, there are no influences of the soil on HMs by the plant and if BF < 1, the plant can be regarded as potential excluder (Radulescu et al., 2013).

Data processing

Soil data obtained in the study were subjected to Normality test by Kolmogorov-Smirnov prior to further statistical analysis. Data were found to be normally distributed. Student t-test was computed using IBM SPSS software version 20.0 and figures were presented using Origin software version 8.0. Leaf and bark data were analyzed by analysis of variance (ANOVA) and significant means were subjected to Tukey HSD Post Hoc test at P≤0.05 using SPSS statistical software for Windows.

Results and discussion

Total metal contents and bioavailability of the orchard soils

The statistics for physico-chemical parameters and metal concentrations of the orchard and background soils are presented in Table 1. The orchard soils are slightly acidic in nature but more acidic (6.1±0.1) in the 11-year-old orchard soil (orchard II), probably due to recent applications of fertilizer at growing stage. Electrical conductivity (EC) were relatively low in the orchard soils. For the soil texture, a strong predominance of sand fractions (81.7% and 75.5%, respectively) were present in soils (loamy sand) with soil fertility at relatively low level in both 35-year old and 11-year old orchards (SOM_I = 1.71; SOM_{II} = 1.17).

Soil concentrations of Cu, Cd and Pb in orchard I ranged from 1.25 - 3.75 mg kg^{-1}, 0.75 - 1.10 mg kg^{-1} and 4.25 - 11.75 mg kg^{-1}, respectively. In orchard II, Cu ranged from 2.50 - 4.25 mg kg^{-1}, Cd ranged from 0.00-1.50 mg kg^{-1} and Pb from 5.75 - 17.25 mg kg^{-1} (Table 1). The average values of Cu in orchard soils were comparable with the background value obtained in a remotely distant location, whereas the average Pb and Cd contents in the two orchards were several folds greater than the background values (Table 1). Heavy metal contents of orchard soils displayed heterogeneity as shown by the coefficient of variation (CV). The CV is the most discriminating factor describing variability (Zhan et al., 2014). The CVs of Cu, Cd and Pb in the 35- and 11-year old orchards were 31.2%, 19.2%, 28.9% and 16.2%, 74.8%. 34.2%, respectively. The relatively low variability of Cu and Pb in the orchard soils and the comparability to background values are reflections of interplay of natural factors. Variability of Cd (CV=74.8%) in the 11-year old orchard (orchard II) was moderate, indicating several hotspots of Cd that may have been affected by agronomic activities such as recent fertilization.

Average concentration of metals in the orchard soils were below critical values of soil quality guideline for agricultural soils (Kabata-Pendias and Pendias, 2001; CCME, 2007). This suggests no metal pollution (Cu, Cd and Pb) of orchard soils, and this is important based on the reported relationships among nutrients of plant and mutual effects on plant uptake (Boskovic-Rakocevic et al., 2014). In fact, the Cu content of the orchard soils falls below the range of values reported for agricultural soils affected by application of Cu-based fungicides (Fernández-Calviño et al., 2008; Fan et al., 2011). Cu has been reported as a typical marker of anthropogenic inputs in citrus cultivated soils (Kabata-Pendias and Pendias, 2001; Fernández-Calviño et al., 2008), hence the relatively low Cu contents of the orchard soils thus indicate less anthropic disturbance of the soil. The Pb contents from the orchard soils in this study are comparable with results obtained for a Ferrasol soil of a vineyard of 40 years in Brazil (Mirlean et al., 2007). The generally low metal contents of the orchard soils is a good reflection of the level of SOM in the 35-and 11-year old orchards (1.71% and 1.17%, respectively) that has been proved to be a major reservoir of metals in agricultural soils. The I_{geo} values for HMs showed that the pollution index of the orchard soils ranged from no contamination to moderate contamination, and therefore suitable for fruit orchard establishment. Moderate contamination of the 35-year old orchard soils by Cd (I_{geo} = 1.65) is an indication of slight alterations caused by long-term irrigation and fertilization but poses no potential ecological risk to soil.

Metal availability in orchard soils

The use of total metal concentration of soil has been widely disproved as effective for assessment of potential impact of metals on soil (Adriano, 2001). Rather, the use of available fractions is noted to provide useful and concise information of mobility and potential uptake by plants. The amount of EDTA-extractable Cu, Cd and Pb were greatly higher than that of extractable-Zn in the orchard soils (Table 2). The great bioavailability of Cu, Cd and Pb of the total metal contents is an indication of high mobility and phyto-availability of metals for uptake by the citrus species. Similar bioavailability was reported for Cu and Cd by Kelepertzis et al. (2015) in a similar study on citrus farm in Greece. The low SOM contents of the orchard soils may have favoured readily extractability of Pb as complexation of Pb with SOM in most soils is always enhanced when OM is abundant in soil. It has also been previously reported that large organic matter content of orchard and garden soils (> 17%) reduced extractable Pb content to less than 10% of total Pb (McBride, 2016). Also, ample evidence in literature have shown that the solubility, extractability and bioavailability of Pb in soil are reduced by organic matter (Sizmur et al., 2011; Fleming et al., 2013; Attanayake et al., 2015).

Though, the studied orchard soils showed no significant accumulation of HMs; the bioavailable contents were critical, especially for Cu, Cd and Pb, possibly as a result of their presence in much labile solid fractions. In addition, the slight acidic nature of the orchard soils highly influenced metal bioavailability, as soil pH is notably an important factor controlling the availability of metals in soil environment to plants. This scenario may portend ecotoxicological risk due to possible uptake of these metals by the citrus trees in large amount and possible bioaccumulation in fruits.

Metal accumulation in citrus tissues

Heavy metal accumulation in the orchards was also checked by measuring metal contents in barks and leaves (mg kg^{-1}, dry weight) of six stands of individual species enumerated in the orchards. The results are shown in Figures 2a and b for 35- and 11-year old orchards, respectively. The HM contents in bark and leaf tissues differed significantly, indicating heterogeneous accumulation without any specific patterns among citrus species. The varied patterns of accumulation/partitioning in the orchards is not unusual as plants differ in metal uptake and accumulation depending upon the physiological and biochemical processes that exist in the species (Antonious and Kochler, 2009; Ogunkunle et al., 2016b).

Cu concentrations were within the normal levels in plants (Kabata-Pendias and Mukherjee, 2007) in bark and leaf tissues of all citrus species in the orchards (Figs. 2ai and 2bi). However, tissue contents of Cu in the two orchards were lower than the recommended nutritional status ($10 \leq Cu \leq 50$ mg kg^{-1}) for fruit trees (Quaggio, 1996), thereby indicating the fruit trees to be Cu-deficient. On the other hand, previous studies have reported quite high uptake of Cu by plants (Kloke et al., 1984), but the reported low bioaccumulation in this study could be caused by low mobility of Cu due to binding to xylem as observed in trees by Nissen and Lepp (1997).

Table 1. Selected properties of soil in the orchards I and II

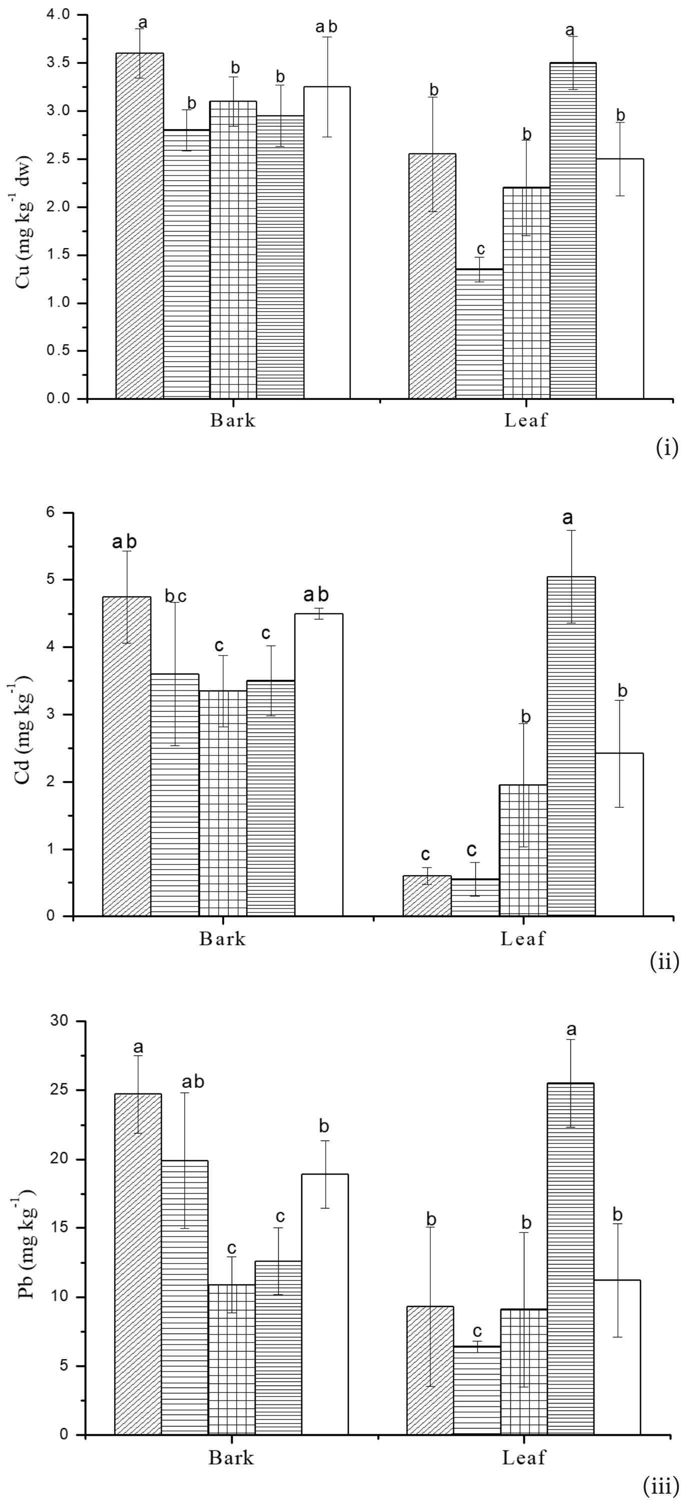
Orchard	**pH**	**EC (dS/m)**	**SOM (%)**	**Sand (%)**	**Silt (%)**	**Clay (%)**
I	6.3±0.40	0.04±0.01	1.71±0.24	81.7	8.7	9.6
II	6.1±0.07	0.05±0.01	1.17±0.60	75.5	11.3	13.2
					Concentration (mg kg^{-1})	
	Orchard			Cu	Cd	Pb
	I		Mean	2.62±0.82	1.10±0.21	8.10±2.35
			Range	1.25-3.75	0.75-1.10	4.25-11.75
			Coefficient of Variation (CV)	31.2%	19.2%	28.9%
	II		Mean	3.65±0.59	0.67±0.50	9.86±3.38
			Range	2.50-4.25	0.00-1.50	5.75-17.25
			Coefficient of Variation (CV)	16.2%	74.8%	34.2%
		t-test (p-value)		0.018*	0.048*	0.025*
Background concentration[a]				2.27±0.41	0.25±0.09	3.90± 0.78
Kabata-Pendias and Pendias (2001)[b]				100	3	100
CCME (2007)[c]				63	1.4	70
I-geo$_{\text{-orchard I}}$				-0.43	1.65	0.39
I-geo$_{\text{-orchard II}}$				0.12	0.88	0.85

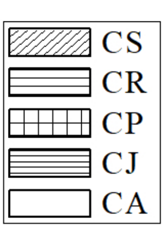
* Denotes significant difference between mean values of the two orchards at $P < 0.05$ (2-tailed). n = 12 and 10 in orchard I (35-year old) and II (11-year old), respectively.
[a] Soil samples (n - 10) collected from the Botanical garden, University of Ilorin, Ilorin (approximetely 15 kilometers from study area)
[b] Maximum permissible concentration for agricultural soils in some European countries
[c] Canadian Council of Ministers of Environment

Table 2. Extractable content (0.05 M EDTA) and bioavailability of metals in orchard soils

Orchard		**Concentration (mg kg^{-1})**					
		Cu	**(%)[a]**	**Cd**	**(%)**	**Pb**	**(%)**
I	Mean	1.34±0.15	51	0.58±0.16	53	4.04±0.48	50
	Range	1.15-1.55		0.40-0.80		3.50-4.50	
II	Mean	1.21±0.38	33	0.40±0.10	60	3.01±0.44	30
	Range	0.75-1.60		0.30-0.50		2.25-3.30	

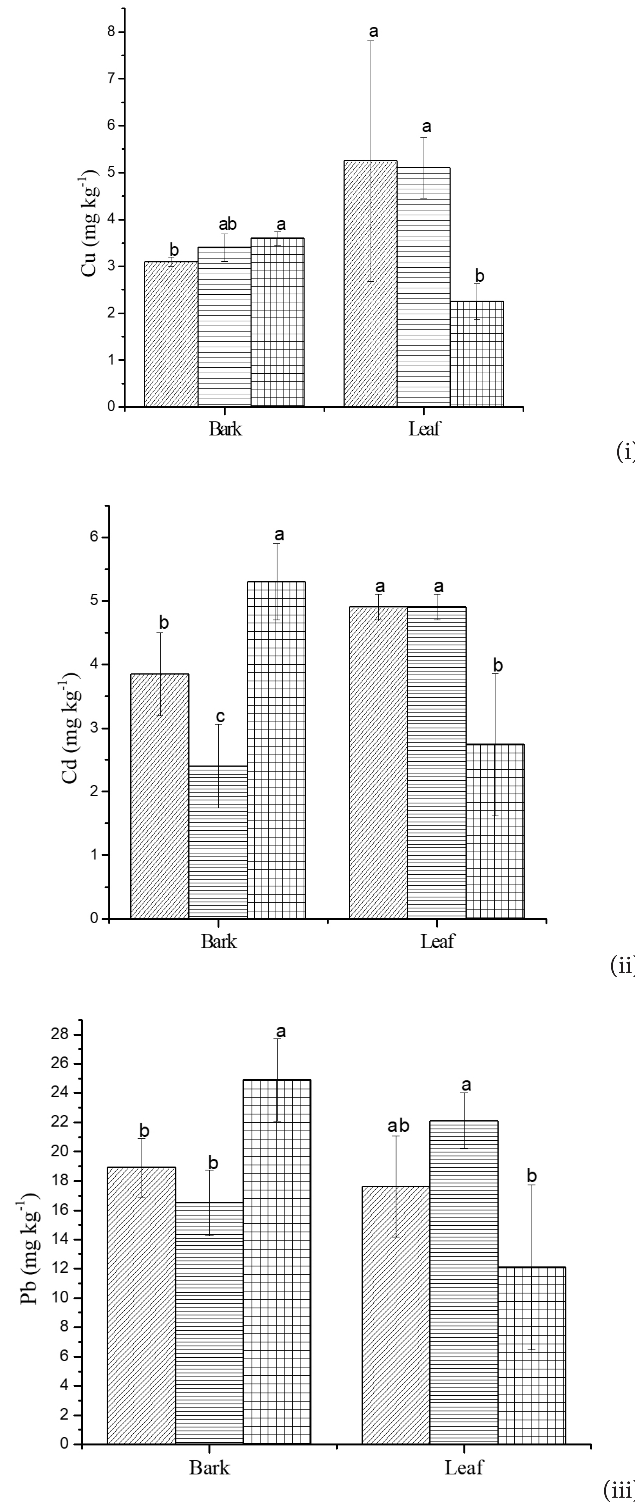
[a] Percentage of bioavailability of metal in soil

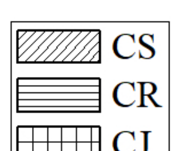




Note: CS - *C. sinensis*; CR - *C. reticulate*; CP - *C. paradise*; CJ - *C. jambhiri* and CA - *C. aurantifolia*; error bars represent standard deviation, columns within the same tissue marked with the same letter are not statistically significant according to Tukey HSD Post Hoc test at $P \leq 0.05$.

Figure 2a. Levels of (i) copper (ii) cadmium and (iii) lead in bark and leaves of citrus trees in orchard I.





Note: CS - *C. sinensis*; CR - *C. reticulate*; CP - *C. paradise*; CJ - *C. jambhiri* and CA - *C. aurantifolia*; error bars represent standard deviation, columns within the same tissue marked with the same letter are not statistically significant according to Tukey HSD Post Hoc test at $P \geq 0.05$.

Figure 2b. Levels of (i) copper (ii) cadmium and (iii) lead in bark and leaves of citrus trees in orchard II

Concentrations of Cd in both bark and leaf tissues of citrus species in the 35- and 11- year old orchards exceeded the permissible limit of 0.2 mg kg^{-1} (FAO/WHO, 1996) in plants. Higher amounts of Cd were accumulated in bark tissues of the citrus species in the 35-year old orchard as compared to the leaf tissue (Figure 2aii). For instance, Cd concentrations in bark of *C. sinensis*, *C. reticulata*, *C. paradisi* and *C. aurantifolia* (≃ 4.7, 3.6, 3.4 and 4.6 mg kg^{-1} dw, respectively) were 9.4, 7.2, 1.7 and 1.8 times higher than leaf concentration, respectively. The partitioning of larger amounts of Cd in barks is likely an adaptive strategy of Cd exclusion from cellular sites where processes such as cell division and respiration occur, thus proving to be an effective protective mechanism (Hall, 2002). Apart from intracellular uptake and extracellular sorption of HMs into barks, atmospheric deposition from dusts (Schelle et al., 2006) and stem flow overtime can contribute significantly to higher levels of HMs in barks compared with leaves. It is interesting to highlight that all citrus species in the 11-year-old orchard (orchard II) exceeded the recommended foliar Cd range (Figure 2bii) in uncontaminated woody plants (0.1 - 2.4 mg kg^{-1}; Alloway, 1995) (Figure 2bii), possibly due to the high demand for nutrients in the growth stage.

For Pb, there were appreciable accumulations (6 mg kg^{-1} < Pb ≤ 25 mg kg^{-1}) in both bark and leaf tissues of all citrus species in the two orchards (Figures 2aiii and 2biii), and these concentrations exceeded the normal levels in plants (Kabata-Pendias and Mukherjee, 2007). High Pb concentrations in plant tissues increase abscission especially in older leaves (Sharma and Dubey, 2005). These concentrations are critical as only about 0.005 to 0.13% of Pb in the soil solution was reported to be available for plant uptake (Kabata-Pendias and Mukherjeee, 2007). However, Pb uptake from soil is generally passive and not directly translocated to the edible parts of fruit tree (Ward and Savage, 1994). So, this remarkable Pb contents in the bark and leaf tissues of the citrus trees are not likely to get into the fruits of this valuable tree crops. Ademoroti (1986) attributed such high Pb contents in plant tissues to traffic-related deposition on rough surfaces such as tree bark and possibly leaf surface.

Transfer of heavy metals in soil-citrus system

In attempt to better understand metal accumulation and transfer in the citrus trees (specifically relationship between Cu, Cd and Pb in citrus tissues and EDTA-extractable contents), bioaccumulation factor (BF) of the bark and leaf are presented in Table 3. Bioaccumulation of metals that was indicated by the BF was generally higher in the 11-year old orchard compared to the 35-year old orchard. For instance, BF_{Cd} for bark in *C. sinensis*, *C. reticulata* and *C. jambhiri* of the 11-year-old orchard were 1.1, 1.0 and 2.0 fold higher than same species in the 35-year-old orchard. This was also consistent with foliar BF_{Cd}. Metal uptake are remarkably affected by soil pH, organic matter, CEC, and plant type (Wang et al., 2013; Zeng et al., 2011). The slight acidity of the 11-year old orchard soil enhanced solubilization of metals and subsequent bioavailability for plant uptake. In addition, uptake and accumulation of HMs in plants follow the root and foliar pathways (Li et al., 2006) and the major driving force for metal transport from root in plants is transpiration through xylem transport (Uraguchi et al., 2009). The high demand for nutrient in vegetative growth stage of the 11-year old orchard may have promoted the high rate of metal translocation in the young citrus trees. Dinelli and Lombini (1996) have observed that metal concentrations were generally higher in the early vegetative growth stage of *Salix*, *Silene* and *Populus* species growing on mine soil, due to a relatively high nutrient uptake compared to growth rate. Metal bioaccumulation in the citrus leaves of the 35-year old orchard was low, compared to the 11-year old orchard. This may be attributed to metal dilution by higher biomass production and accumulation in the litters due to several years of leaf senescence as reported by Pulford and Watson (2003).

Table 3. Bioaccumulation factor (BF) of metals in tissues of *Citrus* species

		Bioaccumulation factor [BF]							
		Orchard I					Orchard II		
Bark		CS	CR	CP	CJ	CA	CS	CR	CJ
	Cu	2.7	2.0	2.3	2.0	2.4	2.5	2.8	3.0
	Cd	8.4	6.0	5.7	6.2	7.9	9.5	6.0	12.7
	Pb	5.9	4.5	2.5	3.0	4.5	6.3	5.0	8.0
Leaf		CS	CR	CP	CJ	CA	CS	CR	CJ
	Cu	1.7	1.0	1.7	2.5	1.9	4.5	4.0	15.0
	Cd	1.0	1.5	3.6	5.7	4.3	12.2	12.0	15.0
	Pb	2.0	1.7	2.0	2.5	3.0	5.3	7.3	4.1

Note: CS - *C. sinensis*; CR - *C. reticulata*; CP - *C. paradise*; CA - *C. auratifolia*; and CJ - *C. jambhiri*

It is also understood that the major portions of Cu and Cd are more associated with the mobile fraction (soluble and exchangeable) in soil because of their high mobility and phytoavailability, and their relatively poor sorption in soil. It is suggested that this behaviour of Cu and Cd could have contributed to the high uptake and bioaccumulation of these metals in the bark and leaf tissues of the citrus species in the orchards. Another factor contributed to the high bioaccumulation of metals in citrus species of the 11-year old orchard is age. Hasselgren (1999) found that concentration of metals in willow trees generally decreased with age. Pb is associated with non-mobile fractions (acid soluble, organic bound, Fe-Mn oxides, and residual) and binds strongly with organic matter, so it has a low mobility from soil to plants. But contrarily in this study, relatively high BF_{Pb} was observed in the citrus trees of the two orchards, and this can be adduced to the relatively low organic matter content. Therefore, soil properties (such as pH and OM), which are the main influencing factors for transfer of metals from soil to plants, played a significant role in the uptake/bioaccumulation of metals in the studied citrus trees.

Conclusion

The agronomic management practices adopted in the orchards only promoted Cd contamination in the orchards soils. Although contamination level has not reached the intervention thresholds, there is the need for monitoring to avert potential ecological perturbation. High bioavailability (≥ 30%) of the HMs (Cu, Cd and Pb) in the orchard soils was possibly influenced by the slightly acidic pH and low organic matter. This accounted for bioaccumulation of metals in citrus bark and leaf tissues, especially Cd and Pb with concentration in several-folds in the leaves and barks compared with the soil concentration. However, further study on the safety level of HMs in the citrus fruits is necessary to assess the mobility potential in the fruits and their safety for consumption.

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References

Ademoroti C. M. A. (1986). Levels of heavy metals on bark and fruit of trees. J. Environ. Pollut. B. 11: 241-253.

Adriano D. C. (2001). Bioavailability of heavy metals. In: Adriano D. C. (ed.): Trace Elements in Terrestrial Environments. New York, USA: Springer.

Alloway B. J. (1995). The origins of heavy metals in soils. In: Alloway B. J. (ed.): Heavy Metals in Soils, pp. 39-57, London: Blackie Academic and Professional.

Antonious G.F., Kochher T.S. (2009). Mobility of heavy metals from soil into hot pepper fruits: a field study. B. Environ. Contam. Tox. 82(1): 59-63.

Ashworth J., Keyes D., Kirk R. and Lessard R. (2001). Standard procedure in the hydrometer method for particle size analysis. Journal Communications in Soil Science and Plant Analysis 32 (5-6): 633-642.

Attanayake C. P., Hettiarachchi G. M., Martin S., Pierzynski G. M. (2015). Potential bioavailability of lead, arsenic, and polycyclic aromatic hydrocarbons in compost-amended urban soils. J. Environ. Qual. 44: 930-944.

Boskovic-Rakocevic L., Milivojevic J., Milosevic T., Paunovic G. (2014). Heavy metal content of soils and plum orchards in an uncontaminated area. Water Air Soil Poll. 225: doi-10.1007s/11270-014-2199-z

Bradl H. B. (2005). Heavy Metals in the Environment: Origin, Interaction and Remediation, Boston: Elsevier Academic Press

Canadian Council of Ministers of the Environment (CCME) (2007). Canadian soil quality guidelines for the protection of environmental and human health: summary tables. Canadian Council of Ministers of the Environment, Winnipeg. Excerpt from Publication No. 1299, ISBN 1-896997-34-1

Dinelli E., Lombini, A. (1996). Metal distributions in plants growing on copper mine spoils in Northern Apennines, Italy: the evaluation of seasonal variations. Appl. Geochem. 11: 375-385.

Fan J., He Z., Ma L. Q., Stoffella P. J. (2011). Accumulation and availability of copper in citrus grove soils as affected by fungicide application. J Soils Sediments. 11: 639-648.

Fang B., Zhu X. Q. (2014). High content of five heavy metals in four fruits: Evidence from a case study of Pujiang County, Zhejiang Province. China, Food Control 39: 62-67.

FAO/WHO (1996). Permissible limit of heavy metals in soil and plants. Geneva, Switzerland: World Health Organization.

Fernández-Calviño D., Rodríguez-Suárez J. A., López-Periago E., Arias-Estévez M., Simal-Gándara J. (2008). Copper content of soils and river sediments in a winegrowing area, and its distribution among soil or sediment components. Geoderma 145: 91-97.

Fleming M., Tai Y., Zhuang P., McBride M. B. (2013). Extractability and bioavailability of Pb and As in historically contaminated orchard soil: effects of compost amendments. Environ. Pollut. 177: 90-97.

Hall L. (2002). Cellular mechanisms for heavy metal detoxification and tolerance. J. Exp. Biol. 53: 1-11.

Hasselgren K. (1999). Utilization of sewage sludge in short-rotation energy forestry: a pilot study. Waste Manag. Res. 17: 251-62.

Intawangse N., Dean J. R. (2006). Uptake of heavy metals in vegetable plants grown on contaminated soil and their bioavailability in the human gastrointestinal tract. Food Addit. Contam. 23: 36-48.

Ite A. E., Ubong U. U., Etesin U. M., Nsi E. W., Ukpong E. J., Ekanem A. N., Ufot U. F., Udo A. I. (2016). Heavy metals in epiphytic lichens and mosses of oil-producing communities of Eket and Ibeno, Akwa Ibom State, Nigeria. Am. J. Environ. Prot. 4: 38-47.

Ite A.E., Udousoro I. I., Ibok U. J. (2014). Distribution of some atmospheric heavy metals in lichen and moss samples collected from Eket and Ibeno local government areas of Akwa Ibom State, Nigeria. Am. J. Environ. Prot. 2: 22-31.

Kabata-Pendias, A. and Mukherjeee, A. (2007). Trace elements from soil to human. Heidelberg: Springer

Kabata-Pendias, A. and Pendias, A. (2001). Trace elements in soils and plants, third edition, Boca Raton, Florida: CRC Press.

Kelepertzis, E., Paraskevopoulou, V., Argyraki, A., Fligos, G. and Chalkiadaki, O. (2015). Evaluation of single extraction procedures for the assessment of heavy metal extractability in citrus agricultural soil of a typical Mediterranean environment (Argolida, Greece). J Soil Sediment. 15: 2265-2275. doi 10.1007/s11368-015-1163-x

Kloke, A., Sauerbeck, D. R. and Vetter, H. (1984). The contamination of plants and soils with heavy metals and the transport of metals in the terrestrial food chains. In: Nriagu J. O. (ed.): Changing Metal Cycles and Human Health, pp. 113–141, Berlin: Springer Verlag.

Lamas, G. A., Navas-Acien, A., Mark, D. B. and Lee, K. L. (2016). Heavy metals, cardiovascular disease, and the unexpected benefits of chelation therapy. J. Am. Coll. Cardiol. 67: 2411-2418.

Li, J. T., Qiu, J. W., Wang, X. W., Zhong, Y., Lan, C. Y., Shu, W. S. (2006). Cadmium contamination in orchard soils and fruits and its potential health risk in Guangzhou, China. Environ. Pollut. 143: 159-165

Li, Z., Ma, Z., Jan van der Kuijp, T., Yuan, Z. and Huang, L. (2014). A review of soil heavy metal pollution from mines in China: pollution and health risk assessment. Sci Total Environ. 468/469: 843-853.

Lower Niger River Basin Development Authority (LNRBDA) (2002). The Journey so Far. Enhancing sustainable water resources development. *LNRBDA News Letter*: 1-2

McBride M. B. (2016). Extractability of Pb in urban gardens and orchards linked to soil properties. Eur. J. Soil Sci. 67: 686-694. doi:0.111/ejss.12372

Mirlean N., Roisenberg A., Chies, J. O. (2007). Metal contamination of vineyard soils in wet subtropics (southern Brazil). Environ. Pollut. 149: 10-17.

Muller, G. (1969). Index of geo-accumulation in sediments of the Rhine River. GeoJournal 2: 108-118.

Nissen L R., Lepp N. W. (1997). Baseline concentrations of copper and zinc in shoot tissues of a range of *Salix* species. Biomass Bioenerg. 12: 115-120.

Ogunkunle C. O., Folarin O. O., Olorunmaiye K. S., Varun M., Fatoba P. O. (2016b). Transfer of metals from crude oil impacted soils to some native wetland species in the Niger-Delta, Nigeria: Implications for phytoremediation potentials. J Agr Sci. 61: 181-199.

Ogunkunle C. O., Varun M., Jimoh M. A., Olorunmaiye K. S., Fatoba, P. O. (2016a). Evaluating the heavy metal pollution of an urban paddy soil and bioaccumulation in rice (*Oryza sativa* L.) with the associated dietary risks to local population: a case study of Ilorin, north-central Nigeria. Environ. Earth Sci. 75: 1-12. doi 10.1007/s12665-016-6203-3

Pulford I. D., Watson C. (2003). Phytoremediation of heavy metal-contaminated land by trees- a review. Environ. Int. 29: 529-540

Quaggio J. A. (1996). Fertilization and liming for hose and fruit quality. In: São José A. R., Souza I. V. B., Martins-Filho J., Morais O. M. (eds.): Mango Production Technology And Market, pp. 106-135, Vitória da Conquista: DBZ / UESB.

Quevauviller P., Rauret R., Rubio G., Lopez-Sanchez J. F., Ure A.M., Bacon H., Muntau, J. R. (1997). Certified reference materials for the quality control of EDTA- and acetic acid-extractable contents of trace elements in sewage sludge amended soils (CRMs 483 and 484). Fresen. J. Anal. Chem. 357: 611–618.

Radulescu C., Stihi C., Popescu I.V., Dulama I.D., Chelarescu E.D., Chilian A. (2013). Heavy metal accumulation and translocation in different parts of *Brassica oleracea* L. Rom. J. Phys. 58: 1337-1354.

Robinson B., Bolan N., Mahimairaja S., Clothie B., 2005. Speciation, mobility and bioavailability of metals in the environment, in: Landner L., Reuther R. (Eds.), Metals in society and in the environment: A critical review of current knowledge on fluxes, speciation, bioavailability and risk for adverse effects of copper, chromium, nickel and zinc, pp. 139-274, Dordrecht: Springer.

Schelle L. M., Gallo M. V., Denham M., Ravenscroft J. (2006). Effects of pollution on human growth and development: an introduction. J Physiol Anthropol. 25: 103-112.

Sharma P. and Dubey R.S. (2005). Lead toxicity in plants. Braz. J. Plant Physiol. 17(1): 35-52.

Sizmur T., Palumbo-Roe B., Hodson M. E. (2011). Impact of earthworms on trace element solubility in contaminated mine soils amended with green waste compost. Environ. Pollut. 159: 1852-1860.

Sutherland R.A. (1998). Loss-on-ignition estimates of organic matter and relationships to organic carbon in fluvial bed sediments. Hydrobiologia 389:153–167

Tchounwou P.B., Yedjou C.G., Patlolla A.K., Sutton D.J. (2012) Heavy Metal Toxicity and the Environment. In: Luch A. (eds) Molecular, Clinical and Environmental Toxicology. Experientia Supplementum, vol 101. Springer, Basel

Uraguchi S., Mori S., Kalamata M., Kawasaki A., Arao T., Ishikawa S. (2009). Root-to-shoot Cd translocation via the xylem is the major process determining shoot and grain cadmium accumulation in rice. J. Exp. Bot. 60: 2677-2688.

Wang C., Yang Z., Yuan X., Browne P., Chen L., Ji, J. (2013). The influences of soil properties on Cu and Zn availability in soil and their transfer to wheat (*Triticum aestivum* L.) in the Yangtze River delta region, China. Geoderma 193/194: 31-139.

Ward N. I., Savage J. M. (1994). Metal dispersion and transportation activities using food crops as biomonitors. Sci. Total Environ. 146/147: 309-319.

Zeng F., Ali S., Zhang H., Ouyang Y., Qiu B., Wu F., Zhang G. (2011). The influence of pH and organic matter content in paddy soil on heavy metal availability and their uptake by rice plants. Environ. Pollut. 159: 84-91.

Zhan H. Y., Jiang Y. F., Yuan J., Hu X. F., Nartey O. D., Wang B. L. (2014). Heavy metal pollution in soil and wild plants from lead-zinc smelting areas in Huixian County, Northwest China. J. Geochem. Explor. 147: 182-188.

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