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Comparison of metal bioaccumulation in crop types and consumable parts between two growth periods

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Abstract

A high proportion of populations in most developing countries live below the poverty line and those near refuse grounds resort to dumpsite farming to grow food. Consequently, high levels of waste-derived contaminants are found in crops consumed by these people. This study investigates the extent to which crops cultivated on the Mbale dumpsite (Uganda) were contaminated by 11 metals and 2 non-metals: iron (Fe), aluminum (Al), zinc (Zn), manganese (Mn), copper (Cu), mercury (Hg), lead (Pb), nickel (Ni), cobalt (Co), cadmium (Cd), selenium (Se), chromium (Cr), and arsenic (As). We investigated how element bioaccumulation in crops was influenced by the growth period (short- and long-term crop maturity). The short-term crops were *Zea mays* and *Amaranthus cruentus*, whereas the long-term crops were *Manihot esculenta*, *Colocasia esculenta*, *Musa acuminata*, *Carica papaya*, *Coffea arabica*, and *Saccharum officinarum*. Results showed that nine metals were present at concentrations above World Health Organization/Food and Agriculture Organization (WHO/FAO) food safety recommendations and hence may pose health risks to consumers. In this study, leaves contained higher metal concentrations than other analyzed consumable parts. Pb and Co were found at higher concentrations in leaves of short-term crops than in long-term crops. Among short-term crops, only *Z. mays* seeds contained permissible metal concentrations by WHO/FAO standards. The growth period was also found to influence metal bioaccumulation in crop types. Pb, Co, Fe, Al, and Cu concentrations were significantly higher in the short-term crops than in long-term crops, while Mn, Ni, and Cr concentrations were higher in long-term crops than in short-term crops. Overall, public awareness about the health risks associated with consuming short-term leafy crops grown on dumpsites should be improved to reduce toxic metal exposure. While implementing such a campaign, the food supply of individuals whose survival depends on such crops should not be jeopardized. Therefore, farmers need alternative farming areas outside dumpsites. *Integr Environ Assess Manag* 2022;18:1056–1071. © 2021 The Authors. *Integrated Environmental Assessment and Management* published by Wiley Periodicals LLC on behalf of Society of Environmental Toxicology & Chemistry (SETAC).

KEYWORDS: Growth period, Health risks, Metal accumulation, Solid waste dumpsite, Urban agriculture

INTRODUCTION

The presence of metals in food crops and drinks at toxic levels is hazardous to animals and humans alike (Ali et al., 2014; Maher et al., 2018; Otim et al., 2019). Several pathways have been shown to account for most of the metal intake by humans. These include contaminated dust settling on food crops (Nabulo et al., 2006), the soil upon which food crops are grown (Szolnoki et al., 2013; Yabe et al., 2010), wastewater used to irrigate cultivated fields of food crops (Ngweme et al., 2020), municipal compost soil upon which some urban dwellers rely on to cultivate food (Adefila et al., 2010; Suruchi & Pankaj, 2011), surface floods from urban centers (Hasenmueller & Criss, 2013), and groundwater sources (Teta & Hikwa, 2017). Food crops and drinking water contamination are, therefore, the major routes by which humans are exposed to toxic metal levels worldwide (Lion & Olowoyo, 2013; Lu et al., 2015). This conclusion is supported by several comparative studies showing higher metal concentrations in urban soils and crops than in those from rural environments (Nabulo et al., 2011; Oluyemi et al., 2008; Warming et al., 2015).

This article includes online-only Supporting Information.

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Urban garden soils have been mainly used to cultivate short-term crops such as vegetables, fruits, and cereals both for domestic and commercial uses (Ebong et al., 2008; Nabulo et al., 2010; Saumel et al., 2012). Nevertheless, the urban soils and the leafy crops grown on these soils are known to have elevated zinc, lead, iron, copper, cobalt, and chromium concentrations (Lu et al., 2015; Nabulo et al., 2006). The uptake of these metals by crops depends on the specific metal species, metal source, local soil conditions, metal concentrations in the soil, and crop types and parts (Saumel et al., 2012; Szolnoki et al., 2013). The term "crop type" is used here to represent crop species or genus, while the term "crop part" represents a plant part (i.e., leaf, grain, stem, flower, and roots).

Humans need to consume plenty of vegetables and fruits to derive vitamins, minerals, fiber, and essential elements required for good health (Fung et al., 2018; Hadayat et al., 2018). The World Health Organization (WHO) recommends an average daily dietary vegetable intake of 200 g for children under 6 years of age, 400 g for adults, and intake of one to two fruits a day to prevent diseases associated with dietary deficiencies (WHO, 2003). There is a need, however, to be vigilant over the risks associated with ingesting food crops contaminated with toxic metals (Ali & Al-Qahtani, 2012; Gebeyehu & Bayissa, 2020). Metals in plant tissues are transferred to animal tissues via the food chain or food web. They are deposited into blood, milk, flesh, and other parts of the human body (Musa & Ifatimehin, 2013). These deposits are known to cause hormonal disruption, abnormalities, and diseases (El-Kady & Abdel-Wahhab, 2018). Therefore, crop safety studies are crucial and vital for public health safety, especially for protecting vulnerable populations such as the elderly, children, and pregnant and lactating women from the risks associated with consuming food contaminated with high levels of metals (Lu et al., 2015). At a solid waste dumpsite in Mbale (Uganda), a recent study found high metal concentrations in leaves of commonly consumed crops grown at the dumpsite and the surrounding slope (Awino et al., 2019). This situation is not unique to Uganda (e.g., Cortez & Ching, 2014; Ebong et al., 2008; Gebeyehu & Bayissa, 2020; Mahmood & Malik, 2014) and demands not only the documentation of metal concentrations in leafy crops but also an investigation of crop species and their parts consumed as biomonitors for assessing their safety for human consumption. Furthermore, data would yield a much-needed early-warning documentation of spatial and temporal variabilities of contamination within crop parts and across crop species. The present study sought to determine the differential uptake of metals in eight food crops with varied growth periods (i.e., how long the crops take to mature) cultivated on two potentially contaminated major zones of the Mbale dumpsite (Figure 1). The objectives were (i) to determine differences in the metal concentrations between short-term and long-term crop types (species) as a whole (i.e., whether growth period influences metal uptake) and (ii) to detect the differences in metal concentrations in edible parts to determine whether parts of crops accumulate metals differently. The metal concentrations in crop parts were then checked against international consumer food safety limits for human consumption.

MATERIALS AND METHODS

Study site

This study was conducted at a municipal solid waste dumpsite in Mbale municipality located in eastern Uganda (1°04′50.0″N, 34°10′30.0″E) (Awino et al., 2019). Mbale is Uganda's fourth-largest municipality, covering about 24 km^2 (OPEP, 2007; UBoS, 2014). The municipality is served by a solid waste dumpsite that covers an area of about 4.05 ha (Awino et al., 2019).

The climate in Mbale is tropical, with significant rainfall throughout the year (7140 mm), and a mean annual temperature of about 20 °C. Humidity and wind speed vary by season in Mbale. The most humid period is between the months of March and June, whereas the windiest period is between November and March. The average wind speed over a typical year is about 8 km/h.

According to the Food and Agriculture Organization (FAO) classification, Mbale soil is mostly *lixic ferralsols* (Minai, 2015; Semalulu & Kaizzi, 2012; van Wambeke, 1974). Across this study site, the soil pH was earlier determined to range from 6.4 to 8.7 for the dump center zone and from 7.4 to 8.0 for the slope. From previous studies, food crops cultivated at this site are reported to contain significantly elevated concentrations of Al, Zn, Fe, Cr, Co, Zn, and Cr (Awino et al., 2019).

Study design

The Mbale dumpsite was divided into two major zones: dump center and slope (Figure 1). The dump center was itself subdivided into northeast (N-E), southeast (E-S), southwest (S-W), and northwest (W-N) (Awino et al., 2019). Similarly, the slope was subdivided into areas with a high water table (SHWT) and those with a low water table (SLWT) (Awino, 2020). Due to insufficient samples to support statistical analysis, these subdivisions were recombined into only two major zones (Figure 1).

Eight crop types cultivated at the dump center and six on the slope were used for assessing bioaccumulation of metals in food crops cultivated at the uncontrolled solid waste dumpsite. Forty-one samples in total were collected from the dump center and 39 from the slope. Sampling occurred between November 2016 and January 2017, a period that coincided with the end of a long and extreme drought (by Mbale standard) that lasted for over 6 months. Consequently, the meteorological condition limited the availability of crops to sample (and hence random sampling) in some major sections of the dumpsite (Table 1). The crops sampled with a short-term growth period of 6 months or lesser were *Zea mays* (*Z. mays*) and *Amaranthus cruentus* (*A. cruentus*). The crops sampled with long-term growth periods that need more than 6 months to reach maturity were *Manihot esculenta* (*M. esculenta*), *Colocasia esculenta* (*C. esculenta*), *Musa acuminate* (*M. acuminata*), *Carica papaya* (*C. papaya*), *Coffea*

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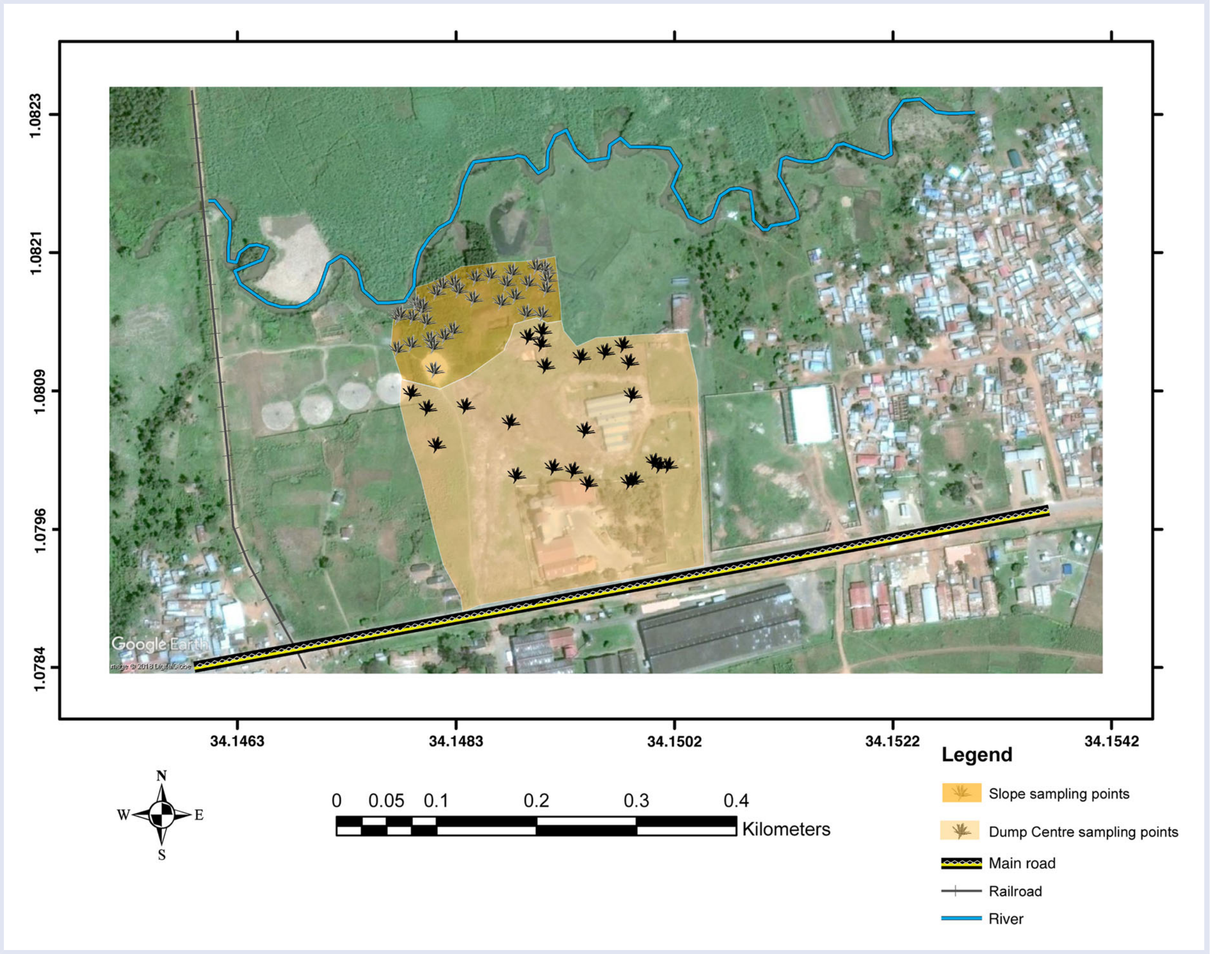

FIGURE 1 Crop sampling points at the Mbale Municipal Council Waste Dump Center and Hill Slope (adapted from Awino et al., 2019)

arabica (*C. arabica*), and *Saccharum officinarum* (*S. officinarum*), and these crops hence grew during both dry and rainy seasons (Table 1). Where applicable, different parts of each crop (i.e., leaves, seeds, fruits, flowers, tubers, or stems) were individually analyzed (Table 1).

Sample collection and preparation

Mature crops of the same type were randomly selected and collected from each of the two zones for analysis. The numbers of crops and plant parts collected are presented in Table 1. In the field, (i) *Z. mays* samples were removed from husks, (ii) leafy vegetables (including with mature flowers containing seeds) and fruits were plucked from plant stems, (iii) tubers were dug out from the ground and packed without peeling or washing, and (iv) seed crops were collected with the seed shells intact. One stem sample of *S. officinarum* included was cut whole and peeled. All the samples were packaged in paper envelopes and transported to the preparatory laboratory in Uganda for processing. Crops of similar type, for example, *Z. mays* or parts (*Z. mays* seeds), but from different subzones (N-E, E-S, S-W, and W-N) within the same major zone (Dump Center), were combined to obtain composite samples comprising a total of six for the final instrumental analysis.

Upon arrival in the preparatory laboratory, *A. cruentus*, *Z. mays*, *M. acuminata*, *M. esculenta*, *C. esculenta*, and *C. papaya* leaves were washed with tap water, followed by de-ionized water, air-dried, oven-dried, ground, sieved, and packaged. Tuber samples (*M. esculenta*, *C. esculenta*.) and fruits (*C. papaya*, *M. acuminata)* were peeled using a ceramic knife to expose their pulp, washed serially with tap water, and de-ionized water to remove dust, air-dried, oven-dried, ground, sieved, and packaged. *Z. mays* and *C. arabica* seeds, covered in pods, were oven-dried without washing. The peeled *S. officinarum* stem and *A. cruentus* flowers were oven-dried (the latter without washing to prevent loss of seeds) at 60–80 °C for 24–48 h (Mwesigye et al., 2019; Ogundele et al., 2014). The samples were separately ground into a fine powder using a porcelain mortar and pestle and sieved using a plastic sieve mesh (0.2 mm pore size). The fine powder was then packaged in clean sterile 50-ml plastic polypropylene tubes with high-density polyethylene screw caps and stored at room temperature, ready for shipment to Australia, where the samples were measured and analyzed.

Sample digestion and analysis

Approximately 0.1 g of each sample measured into a 10-ml plastic centrifuge tube was mixed with 1 ml of

TABLE 1 Sampled short-term and long-term crop parts ($n = 80$) across the Mbale Dumpsite Center and Hill Slope

Crop			Number of samples								
			Dump center					Slope			
Local Name (part)	Scientific name	Growth period (months)	N-E	E-S	S-W	W-N	Total	SHWT	SLWT	Total	Total crop samples
Maize (seeds)	*Zea mays*	≤6	2	2	1	1	6	3	3	6	12
Maize (leaves)		≤6	2	2	1	1	6	3	3	6	12
Red Dodo (leaves)	*Amaranthus cruentus*	≤6	2	1	1	2	6	3	3	6	12
Red Dodo (flowers + seeds)		≤6	2	1	1	2	6	1	2	3	9
Bananas (fruits)	*Musa acuminata*	>6	3				3	3		3	6
Bananas (leaves)		>6	3				3	3		3	6
Cassava tuber		>6			2		2	3	1	4	6
Cassava leaves	*Manihot esculenta*	>6			2		2	3	1	4	6
Balugu (tuber)		>6	1				1	1		1	2
Balugu (leaves)	*Colocasia esculenta*	>6	1				1	1		1	2
Pawpaw (fruits)		>6	1				1	1		1	2
Pawpaw (seeds)	*Carica papaya*	>6	1				1				1
Pawpaw (leaves)		>6	1				1	1		1	2
Sugarcane (stem)	*Saccharum officinarum*	>6	1				1				1
Coffee (seeds)	*Coffea arabica*	>6	1				1				1
Total crop samples per subzone			21	6	8	6	41	26	13	39	80

Abbreviations: E-S, East toward South; N-E, North toward East; S-W, South toward West; SHWT, slope high water table; SLWT, slope low water table; W-N, West toward North.

concentrated nitric acid (Aristar ACS; VWR Chemicals BDH) and incubated for 24 h in a fume cupboard to extract metals associated with sulfates, sulfides, oxides, and carbonates, and then heated with the centrifuge cap loosely closed for 3 h in a shaker water bath maintained at 90–95 °C. After adding 1 ml of 30% (v/v) hydrogen peroxide, cooled overnight, and made up to 10 ml with deionized water, each sample was reheated one more time between 90 °C and 95 °C for 1 h to extract metals associated with organic matter. The digests were then diluted in a ratio of 1:10 (v/v) with deionized water spiked with a known concentration of Li^6, Sc, Y, RH, In, TB, and Ho internal standards for inductively coupled mass spectrometry (ICP-MS) analysis using a Perkin–Elmer Elan DRC-e. The method detection limit (MDL) for all metals was 0.1 mg/kg. Where a concentration was lower than this, the concentration was replaced by 1/2 MDL for statistical analysis.

To ensure that recoveries were acceptable and that our analytical steps were not introducing the targeted elements into the samples, a certified reference material (apple leaves CRM No. 1515, National Institute of Standards and Technology) and acid blanks were similarly processed as crop samples.

Data processing

In this study, (i) metals and metalloids are collectively referred to as metals and (ii) depending on the context, "crop type" may also imply the mean metal concentrations from all parts of the same crop combined for assessment. To determine the temporal and spatial variations in the uptake of the 11 metals (Fe, Al, Zn, Mn, Cu, Pb, Cr, Hg, Co, Ni, Cd) and two metalloids (Se, As) by crop types, or parts of crops from across the dump center and slope areas, data from the two zones were separately analyzed. This approach was based on our previously published study in which significant metal concentration differences in plants were detected in the two study zones (Awino et al., 2019). The order of performing the analysis of the temporal and spatial variations was as follows: first (Strategy 1), all the mean metal concentrations of all crop types in the category of a growth period of 6 months or lesser were grouped and compared with the mean metal concentrations in the category of a growth period of over 6 months in each of the two zones (dump center, slope). That is, the metal concentrations in crops of the same types, which neatly fall into the two study growth periods (≤6 or >6 months to maturity), were

TABLE 2 Strategies for data processing to assess the effects of growth period

Strategies	Crop location	Strategies	Crop types and parts
1	Dump center or slope	Crop types were combined into 2 growth periods and metal concentrations were compared	≤6 months (*Zea mays*, *Amaranthus cruentus*) and >6 months
			– Dump center: *Carica papaya*, *Manihot esculenta*, *Colocasia esculenta*, *Musa acuminata*, *Saccharum officinarum*, *Coffea arabica*. – Slope: *C. papaya*, *M. esculenta*, *C. esculenta*, *M. acuminata*.
2	Dump center and slope	Leaves were combined into two growth periods and metal concentrations were compared	Leaves of crops with a growth period of 6 months or lesser (*Zea mays*, *Amaranthus cruentus*) compared against leaves of crops with a growth period of over 6 months (*Musa acuminata*, *Manihot esculenta*, *Colocasia esculenta*, *Carica papaya*).

combined and compared. Then (Strategy 2), the mean metal concentrations in leaves of crop types with a growth period of 6 months or lesser growing at the dump center were grouped and compared with those of leaves of crop types with a growth period of over 6 months for the same zone. Similarly, the same comparisons were made for leaves of crop types with a growth period from 6 months or lesser and over 6 months (Table 2). In Strategy 2, the leaves were from similar crop types that existed at both the dump center and slope.

A few nontraditional data analysis approaches were also adopted in attempts to understand variability. One approach was to average metal concentrations across the ≤6-month or younger crops and the over 6-month crops and compare the averages. In this case, averaging and combining across the different growth periods were performed to harmonize the data, given variations across samples, crop species, and plant parts. The basis for this choice was metal concentration similarities across the plant parts in the 6-month or younger group (with one exception, *Z. mays* seeds).

Also, note that consumers of dumpsite crops do not pick and choose what to eat from where and when. For example, they will consume either any and/or both 2- to 6-month group crops as a combined meal. A case in point: *Z. mays* seeds powder can be made into a paste commonly referred to as *posho*, which is eaten with *A. cruentus*. Consumption could also be in isolation (e.g., roasted *Z. mays*) or across groups (e.g., *M. acuminata* fruit with *A. cruentus*; *M. esculenta* with *A. cruentus*).

Metal accumulation estimation

We determined the metal accumulation index (MAI) for four of the eight crops for which parts were sampled multiple times. Multiple sampling is necessary for calculating the standard deviation needed for the MAI estimation (Liu et al., 2007). Translocation factors (TF), used largely to evaluate plants for potential uses as phytoremediators (Blaylock & Huang, 2000), were calculated as the ratio: [concentration in leaf]/concentration in root]. In addition, we introduced the concept of the Relative Accumulation Factor (RAF) as a possible indirect measure of the ability of different parts of a crop to accumulate metals relative to each other. RAF is a relative entrenchment factor calculated as the ratio of metal concentrations in a pair of plant parts. A higher RAF value indicates a higher relative enrichment and perhaps a higher translocation factor when a metal concentration in the earliest plant part to develop is placed in the denominator (e.g., RAF = [seed]/[leaf] or [fruit]/[leaf]). To compensate for vast different metal concentrations in different parts of plants, RAF could be expressed as a percentage. Both these approaches are demonstrated here. Note that RAF is used here in lieu of bioconcentration factors ([concentration in plants]/[concentration in soil]), which we were unable to estimate because not all types of samples, including soil samples at the sites of interest, could be collected due to logistical reasons and the physical risks associated with sample collection.

Statistical analysis

Our data are provided as Supporting Information Table 1.A. The data are presented in the text as mean ± standard deviation where applicable. Seven statistical factors were used to test for data distribution, whether normal or not, across 80 crop samples. SPSS 23 software was used for these purposes following the vendor's instructions. Numerical factors covered skewness, kurtosis, Shapiro–Wilk tests/sigma, and *z*-score; visual attributes were normal Q–Q plots, box plots, and histograms. Each of these statistical measures was used to determine whether a data set qualified for either parametric or nonparametric tests. The compliance criteria for each were as follows: normally distributed data would have (i) a skewness close to zero, (ii) kurtosis close to 3, (iii) a *Z*-score of 1 standard deviation, and (iv) symmetrical Q–Q plot, box plot, and histogram. A data set with more than 50% compliance to the seven factors were considered normally distributed and hence qualified for parametric testing (one-way analysis of variance [ANOVA]); a data set with lower than 50% compliance qualified for a nonparametric test (Mann–Whitney test or Kruskal–Wallis test). One-way ANOVA was used for data analysis because (i) the different crops or crop parts were combined for analysis rather than doing so with individual species, (ii) not all the measured parameters/factors (metals, parts, crops, growth periods) qualified for parametric tests, and (iii) in cases where three jointly qualified for nonparametric tests, the Kruskal–Wallis tests were used.

Statistically significant differences between groups in the two strategies listed in Table 2 were assessed at the α level of 0.05. Graphs were plotted using Sigma Plot V13 and Microsoft Excel.

To assess metal concentration differences between the two growth periods, an independent-samples Mann–Whitney *U* test and an independent-samples *T* test (2-factors) were conducted for nonparametric and parametric tests, respectively. For normally distributed data or where the equality of variances was met, an independent *T* test coupled with Shapiro–Wilk's and Levene tests were applied. For samples without normally distributed data, the Welch *T* test was used. The nonparametric test was further followed by pairwise comparisons to estimate the significant mean rank metal concentration differences between growth periods, crop types, and parts. The mean concentrations of Cu, Zn, Ni, Mn, Pb, Cr, Hg, As, and Cd were compared against internationally accepted consumer food safety standard limits. Where applicable, the Codex Alimentarius International Food Standards (WHO/FAO, 2021), the Food Standards Australian/New Zealand (FSANZ, 2019), the Chinese Food Guidelines (M. H. H. Ali & Al-Qahtani, 2012), and the European Food Safety Authority (EFSA, 2021) were used for comparison. The reported limits in mg/kg are as follows: Al: 2.0–3.0, Cr: 0.2, Fe: 20–100, Mn: 5.5, Ni: 2.0, Cu: 15, Zn: 60, Co: 5, As: 0.1, Se: 0.05, Cd: 0.1, Hg: 0.001, and Pb: 0.1–0.3 (Ngweme et al., 2020; Olowoyo et al., 2012; Otim et al., 2019).

RESULTS

Metal concentration variability across samples

In this study, we found high metal concentration variability across samples irrespective of the consumable parts studied or their sources. To determine the correct statistical approach to use for analysis, our data were first tested for normality as described above. The results showed that only Cu concentration data obtained in crops from the dump center and Ni data from the slope had a normal distribution; hence, parametric tools were used to analyze the data (Strategy 1, Table 2). Elsewhere in our data set, Al, Cr, Fe, Mn, Co, Zn, and Pb concentrations qualified for nonparametric tests. In Strategy 2 (Table 2), again, only Cu and Ni concentration data, obtained in crops from the dump center, passed the normality test for data distribution. The rest of the metal concentration data did not pass the test.

To visualize metal concentration variability across samples in a bar chart format, the total sum of each metal in each crop was separately plotted (Figure 2) and (ii) the extent of metal accumulation across the different parts of the eight test crops was similarly plotted (Figure 3).

Overall, it can be seen from Figure 2 that Fe, Al, Zn, Mn, and Cu were present in samples at much higher concentrations than the rest of the targeted metals in these crops, followed by Cr and Ni and then Pb and Co. The total mean concentration of Fe was found to range from 158.4 to 2671.8 mg/kg, Al ranged from 108 to 807.4 mg/kg, Zn ranged from 14.7 to 339 mg/kg, Mn ranged from 27 to 242.7 mg/kg, Cu ranged from 6.7 to 48.8 mg/kg, Cr ranged from 1.2 to 2.5 mg/kg, Ni ranged from 0.8 to 13.6 mg/kg, Pb ranged from 0.1 to 2.2 mg/kg, and Co ranged from 0.1 to 0.5 mg/kg. As, Se, Cd, and Hg were only found in trace amounts ($\leq$0.1 mg/kg). Nevertheless, Hg concentrations were above the acceptable safety limit (0.001–0.03 mg/kg; Ngweme et al., 2020).

Within the same crop, metal concentrations varied widely. In *Z. mays*, for example, the Fe, Al, and Zn concentrations were at least one order of magnitude higher than the Cu concentrations, two orders of magnitude higher than the Cr and Ni concentrations, and three orders of magnitude higher than the Co, As, Se, Cd, and Hg concentrations (Figure 2).

Differential bioaccumulation of metals in the various parts of each crop type was equally observed (Figure 3). The mean Al concentration in *Z. mays* leaves was 13 times higher than that in *Z. mays* seeds. Similarly, the mean Al concentration in *C. papaya* leaves was nine times higher than that in *C. papaya* seeds or fruits. In *M. esculenta* leaves, the mean Al concentration was over 16 times higher than that in *M. esculenta* tuber. In both *Z. mays* and *M. esculenta*, all metal concentrations were higher in the leaves than in the seeds or tubers. In *A. cruentus*, the metal concentrations in the leaves, flowers, and seeds were comparable and in *C. papaya*, only Al, Fe, Cr, and Ni concentrations were the highest in the leaves, while Mn, Zn, Cu, and Pb concentrations were higher in seeds. The metal concentrations in *C. esculenta* tubers and leaves were generally similar, except for Fe and Al, which were higher in leaves. Comparatively, *C. esculenta* tubers contained higher metal concentrations (except Ni) than *M. esculenta* tubers.

Overall, a linear relationship was observed between metal concentrations in the leaves and other crop parts, as higher metal concentrations in the leaves correlated with higher concentrations in seeds, fruits, flowers, and tubers (Figure 4). From these results, it is evident that differential metal uptake does exist between different crop types and parts, the distribution pattern of which is similar in all crops from the two study dumpsite zones (dump center and the slope) (Awino, 2020).

These results are confirmed by the MAI values obtained for four of the eight crops studied (Table 3). *M. esculenta*

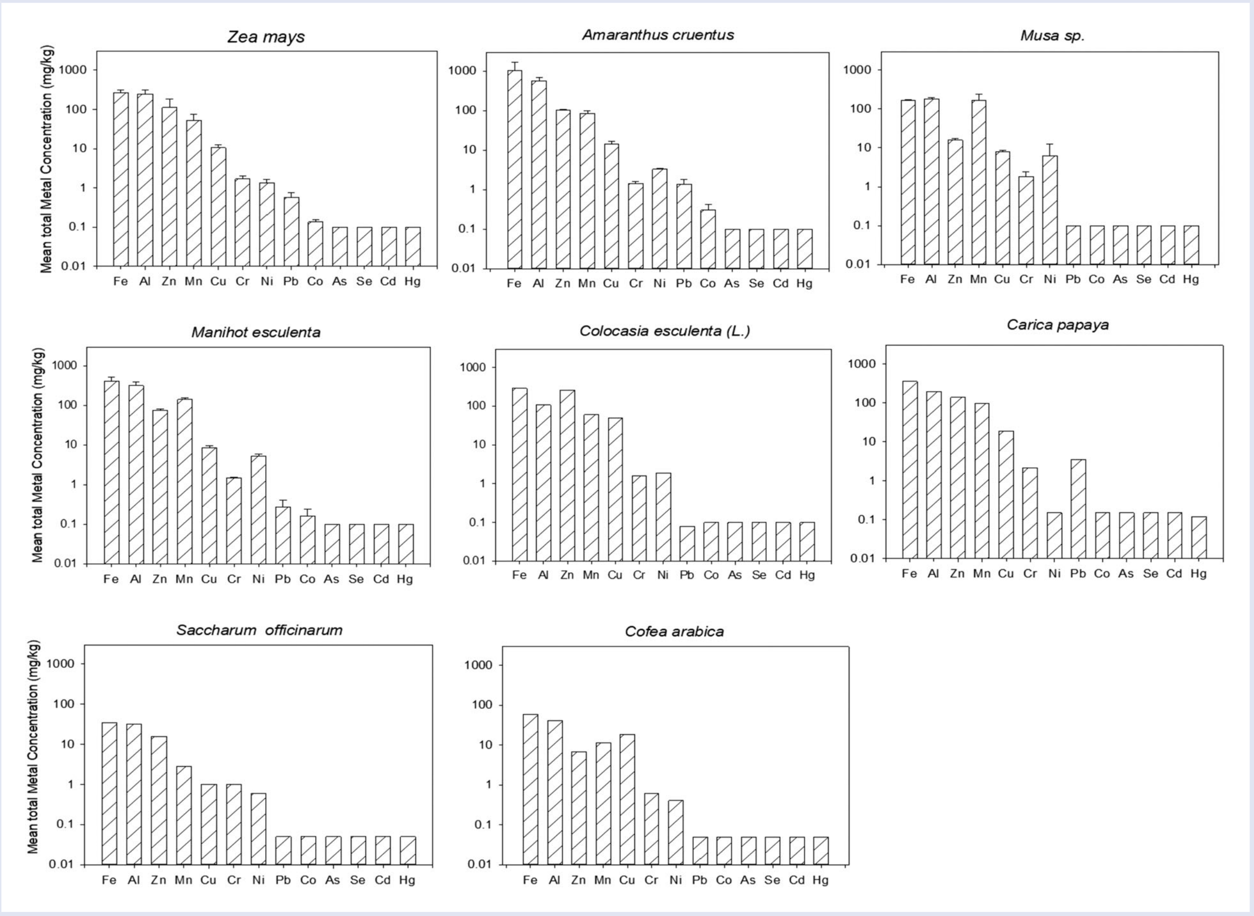

FIGURE 2 A bar graph of the mean metal concentrations (mg/kg) in various crop types at the dump center (the slope equivalent is shown in Figure A.1)

had the highest MAI values for both root and leaf (9.1 and 12.7, respectively), followed by *M. acuminata* leaf (7.6). The lowest MAI value was found for *A. cruentus* flower (2.6). *A. cruentus* leaf, *Z. mays* seed, and leaf had similar values (~3.5). These results showed that these four crops accumulate high levels of metals from the solid waste dump.

To determine whether leaves accumulate more metals than roots, TF were calculated for *M. esculenta* and *C. esculenta*, the two crops for which root/tuber samples were available here (Table 4). A TF value > 1 implies the ability of a metal to be transferred from the root to the other parts of a crop but assumes that aerial deposition on leaves was not a factor. On average, *M. esculenta* hyperaccumulated Mn, Fe, Al, and Zn (56.8, 26.1, 17.2, and 10.7, respectively) and, to some extent, accumulated Pb, Ni, Cr, Co, and Cu in the leaf relative to the root material (4.5, 3.4, 2.7, 2.2, and 2.2, respectively). That is, all these metals were transferred efficiently from *M. esculenta* root to the leaf, especially the hyperaccumulators. In contrast, *C. esculenta* barely accumulated Al (3.1) and Fe (2.2). Little root-to-leaf accumulation was observed for Mn, Co, and Ni for *C. esculenta* (1.3, 1.0, and 1.2, respectively). We also observed that Cr, Cu, Zn, and Pb were translocated from the leaf to the root in *C. esculenta* (0.8, 0.5, 0.6, and 0.6, respectively).

To assess the ability of fruit or seed to accumulate metals relative to their leaves in a leaf-to-fruit or leaf-to-seed direction, the proposed relative enrichment/accumulative factor (RAF) was calculated for all the metals detected above MDLs in *C. papaya*, *Z. mays*, *A. cruentus,* and *M. acuminata* (Table 4). The mean RAF value for *C. papaya* fruit was 0.32; this implies that, on average, only 23% of metals were detected in *C. papaya* fruit. Similarly, only 15% of metals were detected in *Z. mays* seed compared to leaf, 27% in *M. acuminata* fruit, and 47% in *C. papaya* seeds (all RAF values were <1). The exception was for *A. cruentus,* for which RAF = 1 (50:50, flower/leaf), meaning that there was no evidence of the relative enrichment of these metals in this crop. Note that, for individual metals, *C. papaya* seed had very high RAF values for Mn, Cu, and Zn. Similarly, *M. acuminata* fruit had a very high RAF value for Ni. This shows the potential of these metals to transfer from *C. papaya* or *M. acuminata* leaves and accumulate in seeds or fruits, respectively.

Metal concentration variability as a function of the crop growth period

To assess metal concentration variability with the growth period, we regrouped data into two growth periods:

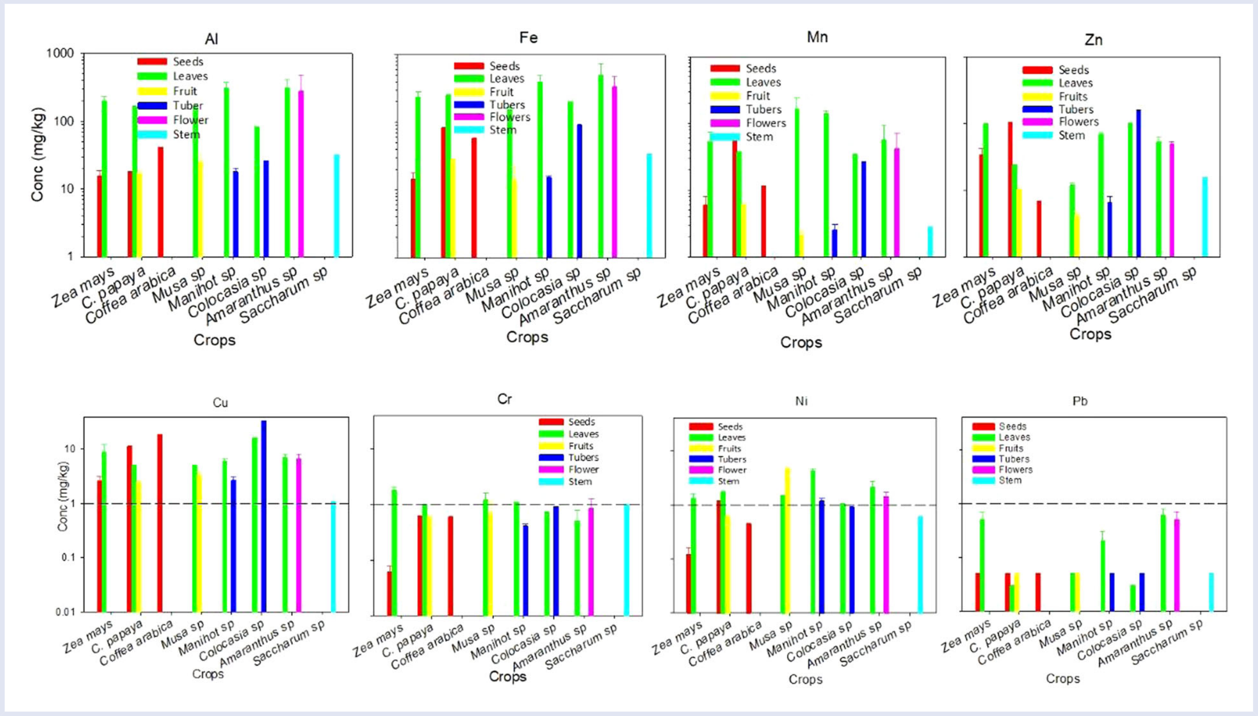

FIGURE 3 Mean metal concentrations in various crop parts by crop type at the dump center

6 months or lesser and over 6 months (Tables 1 and 2) for a pairwise comparison of the mean metal concentrations of nine metals (As, Se, Cd, and Hg concentrations were not included in the pairwise comparison as their concentrations were at or below the detection limits in all crops and crop parts). There were significant differences between the two crop categories, particularly at the dump center. For example, Fe, Al, Pb, and Co concentrations were significantly higher in the short-term crops, while Mn, Ni, and Cr concentrations were significantly higher in the long-term crops (Figure 5A). There was no statistical difference for Zn concentrations.

In the slope area (Figure 5B), significant differences were observed between metal concentrations found in short-term and long-term crops. For example, Fe, Al, Pb, and Co concentrations were consistently higher in all short-term crop samples, while Mn, Ni, and Cr concentrations were consistently higher in long-term crops. When similar comparisons of the mean metal concentrations in leaves of short-term and long-term plants were carried out and ranked, Pb and Co concentrations were significantly higher in the leaves of short-term crops from the two zones (Figure 6).

DISCUSSION

General variation of metal concentrations in crop types and crop parts

The mean metal concentrations were generally found to decrease in the order Fe, Al, Zn, Mn, Cu, Ni, Cr, Pb to Co, with Se, As, Cd, and Hg occurring in trace amounts. Other studies have found similar enrichment of Zn, Pb, Fe, Cu, Co, Cd, and Cr in urban crop types, leaves, and flowers that are probably correlated to the contaminated urban garden soils in which the crops were grown (Lu et al., 2015; Nabulo et al., 2006; Szolnoki et al., 2013). In particular, Zn, Cu, Fe, Pb, Cd, and Cr were reported to bioaccumulate in the leaves of such crops.

The mechanisms by which metals accumulate in various crop parts are varied. Sources here include aerial deposition on leaves from industrial and vehicular particulate emissions (Nabulo et al., 2012; ćPivić et al., 2013; Saumel et al., 2012; Ukpong et al., 2013), the burning of motor tires at dumpsites (Ogundele et al., 2014), and wet or dry deposition (Edelstein & Ben-Hur, 2018; Shahid et al., 2017). The differential metal uptake by crop types could be attributed to their varying potential to absorb metals from the soil and bio-accumulate in different plant parts or to retain contaminated atmospheric particulates from polluted air. In general, metals are available in soils for uptake by plants and as metals are persistent, an increased uptake would lead to metal accumulation in crops. The uptake of metals such as Zn, Mn, and Cu by plants has been shown to correspond to increasing soil metal concentrations (Intawongse & Dean, 2006).

Aerial deposition as a source of metal contamination

Leaves and flowers have higher metal concentrations than seeds, fruits, or tuber (Table 1.A). In this study, unwashed *A. cruentus* flowers were found to contain much higher

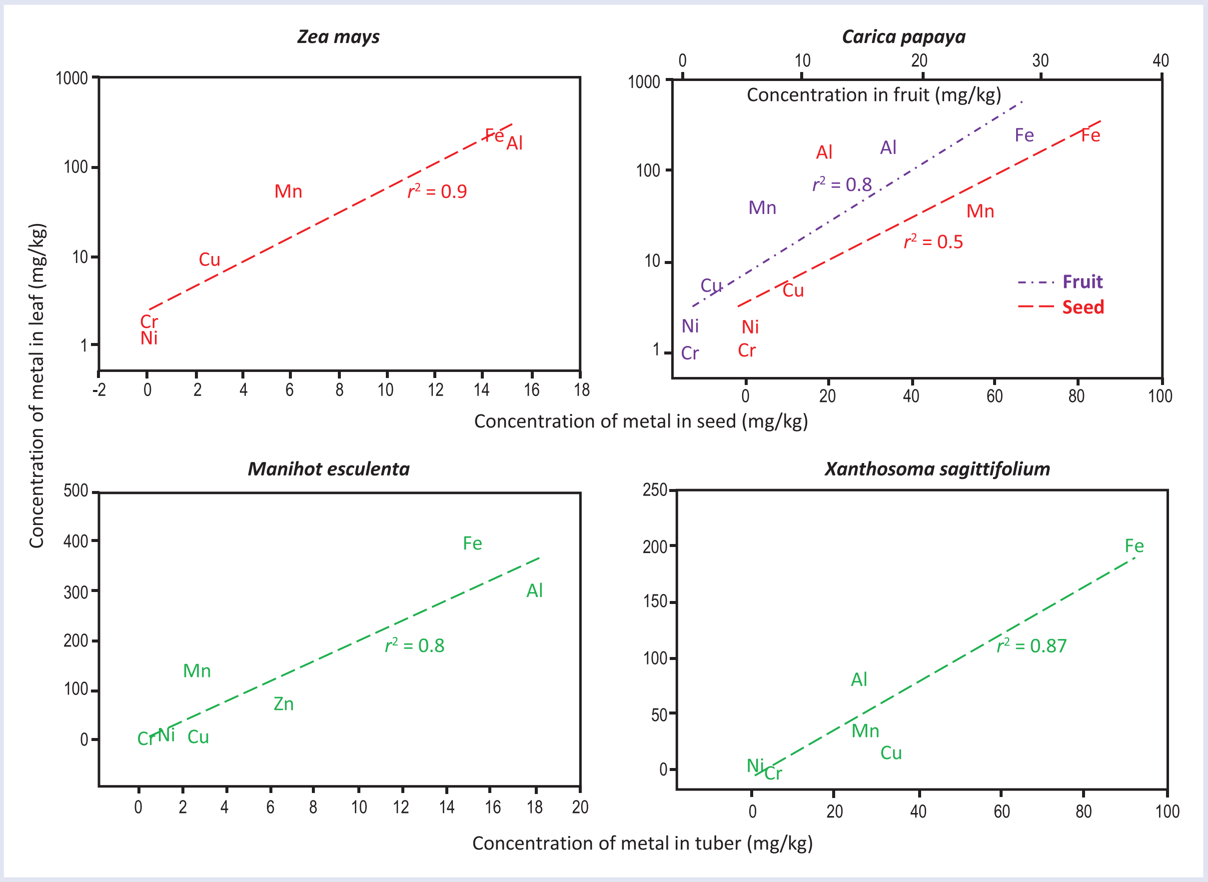

FIGURE 4 Linear relationship between the concentration of metals in leaves and other crop parts

metal concentrations than other washed parts, and this is in agreement with reports from other studies (Nabulo et al., 2006). Given the location of the dumpsite near industrial-scale facilities that include metal scrap mining and rice winnowing, and alongside a major highway (Mbale-Tirinyi) and near a railway line, these results suggest that flowers accumulate aerial deposits of wind-borne particles containing metals. Furthermore, within the dumpsite, solid waste including medical waste is preferentially burnt as an alternative to waste management. These collectively support aerial deposition of metals in particulates and are hence a potential source of crop contamination (Rahman et al., 2014). The Namatala River also passes close to the dumpsite (Figure 1), but more studies are needed to evaluate whether the river delivers metals to the soil–plant system at the dumpsite.

Role of leaves in the differential metal distribution in crops

Metals measured at higher concentrations in leaves also had higher concentrations in other parts of the same crop. A linear relationship was detected between the metal concentrations in leaves and in other parts of the crops studied. This observation could be explained by the role that leaves play as the sites of photosynthetic activity during the growth and development of leafy crops (Madejon et al., 2011; Mahmood & Malik, 2014). In plants, leaves promote the continuous flow of nutrients and metals upward from the soil for metabolic activities associated with photosynthesis

TABLE 3 Metal accumulation index (MAI) for *Zea mays*, *Amaranthus cruentus*, *Musa acuminata*, and *Manihot esculenta* seeds, leaf, flower, fruit, and root

	Z. mays		*A. cruentus*		*M. acuminata*		*M. esculenta*	
	seed	leaf	flower	leaf	fruit	leaf	root	leaf
Metal count	7	9	9	9	7	7	7	9
Sample replicates	14	14	11	17	3	3	2	2
MAI	3.2	3.4	2.6	3.5	3.8	7.6	9.1	12.7

TABLE 4 Translocation factors of metals from the roots to the leaves in *Manihot esculenta* and *Colocasia esculenta* along with the proposed binary relative accumulation factors of the metals in *Carica papaya*, *Zea mays*, *Amaranthus cruentus*, and *Musa acuminata*

	Translocation factor		Relative accumulation factor									
	M. esculenta	*C. esculenta*	*C. papaya*				*Z. mays*		*A. cruentus*		*M. acuminata*	
	L/R	L/R	Fr/L	%	Sd/L	%	Sd/L	%	Fl/L	%	Fr/L	%
Al	17.2	3.1	0.11	10	0.11	10	0.07	7	0.81	45	0.16	14
Cr	2.7	0.8	0.56	36	0.64	39	0.04	3	0.47	32	0.59	37
Fe	26.1	2.2	0.11	10	0.33	25	0.06	5	1.95	66	0.09	8
Mn	56.8	1.3	0.17	14	1.54	61	0.12	10	1.07	52	0.01	1
Co	2.2	1.0	-	-	-	-	0.58	37	0.87	46	-	-
Ni	3.4	1.2	0.36	27	0.67	40	0.11	10	1.36	58	3.19	76
Cu	2.2	0.5	0.49	33	2.21	69	0.32	24	1.05	51	0.68	40
Zn	10.7	0.6	0.44	30	4.49	82	0.40	29	1.05	51	0.35	11
Pb	4.5	0.6	-	-	-	-	0.10	9	0.97	49	-	-
Mean	-	-	0.32	23	1.43	47	0.20	15	1.07	50	0.72	27
SD	-	-	0.19	11	1.54	25	0.19	12	0.41	9	1.12	26

Abbreviations: Fl, flower; Fr, fruit; L, leaf; R, root/tuber; SD, standard deviation; Sd, seed.

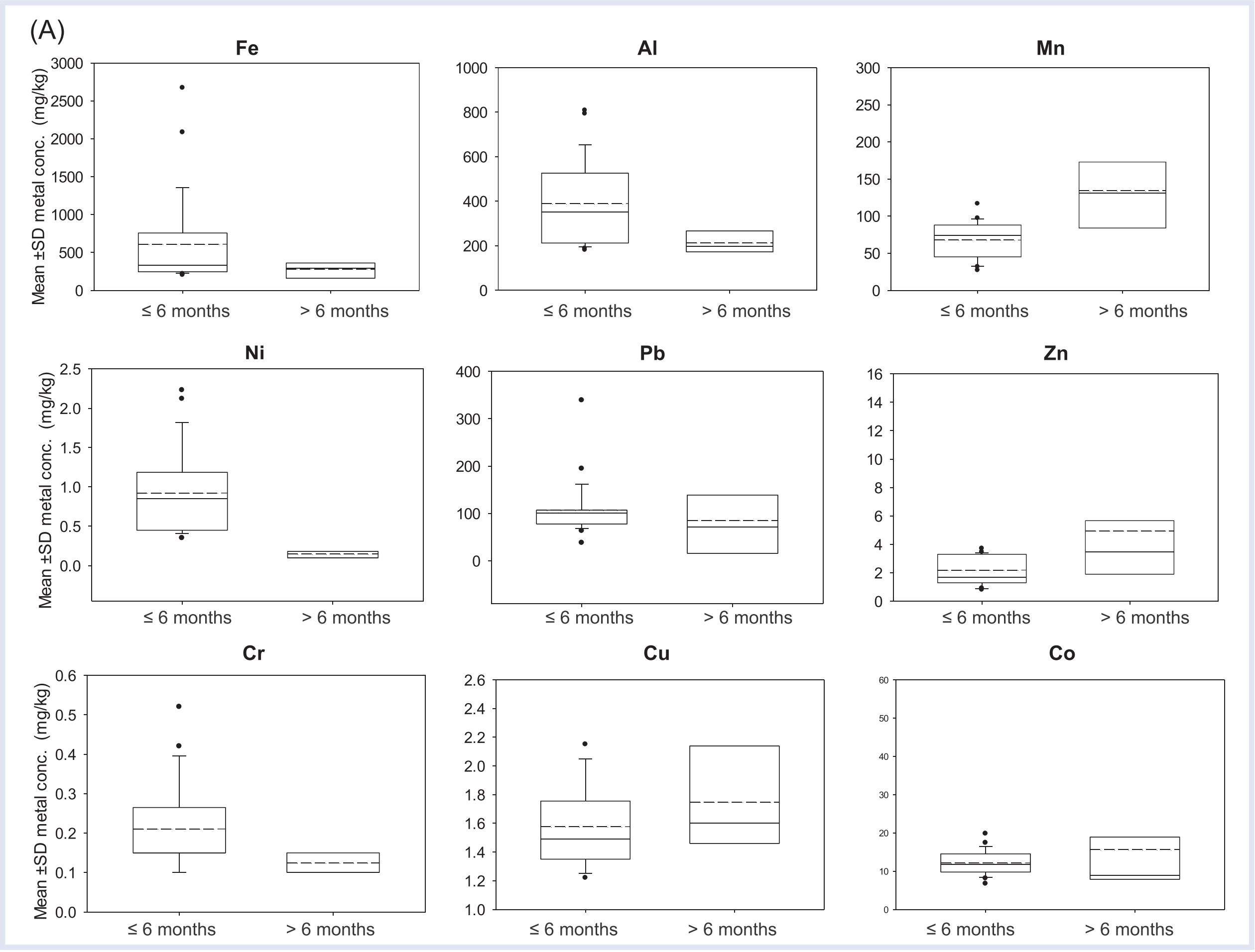

FIGURE 5 (A) Comparison of the mean metal concentrations (±SD) at the dump center in short-term crop types (≤6 months) and long-term (>6 months) crop types. (B) Comparison of the mean metal concentrations (±SD) at the slope in short-term crop types (≤6 months) and long-term (>6 months) crop types

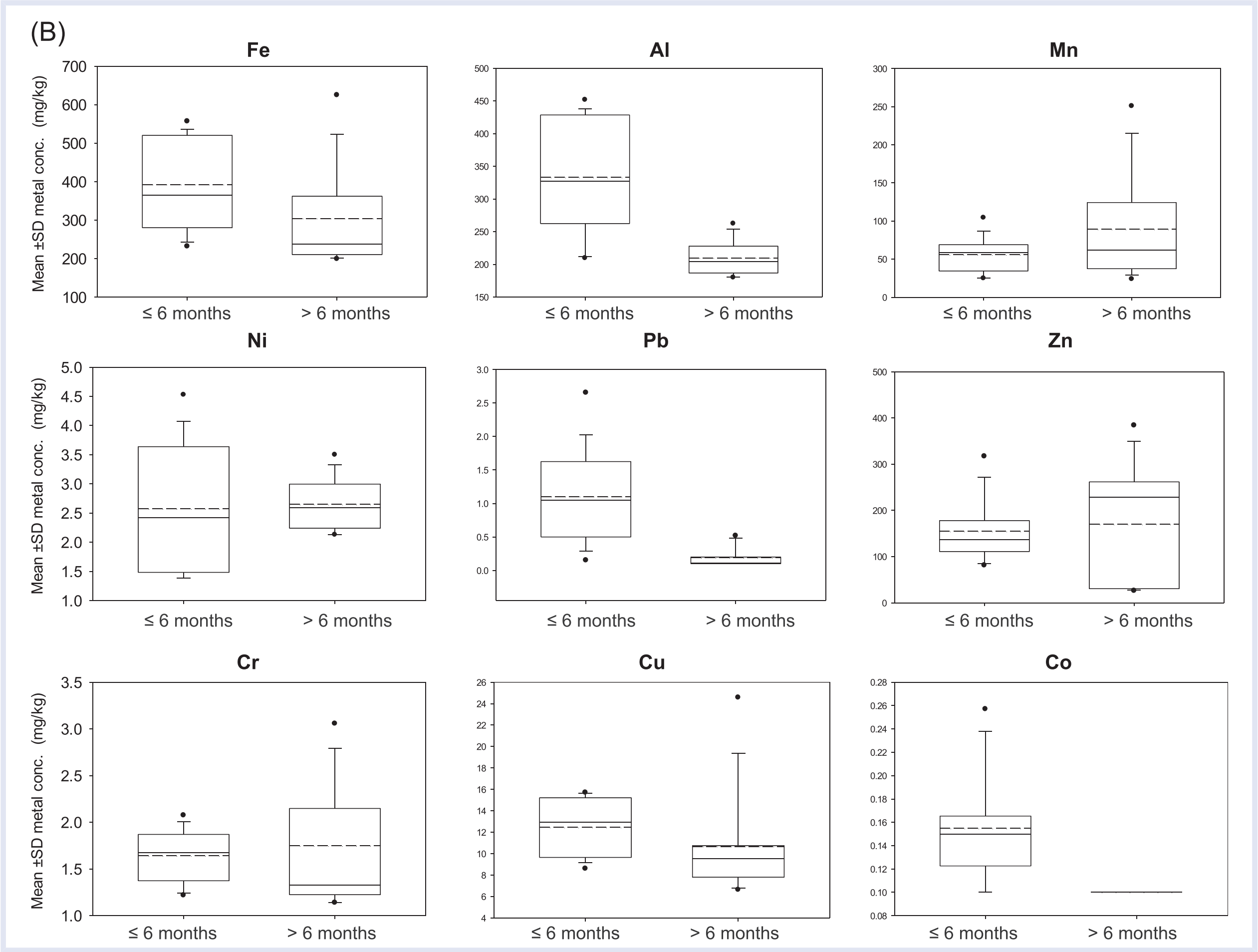

FIGURE 5 Continued.

(Nabulo et al., 2012; ćPivić et al., 2013). Thus, metal accumulation in leaves at higher levels relative to other plant parts was expected (Mwesigye et al., 2019; Szolnoki et al., 2013; Ukpong et al., 2013). This, however, is not always the case since roots are the first points of contact between plants and metals and could also be expected to have higher metal concentrations. Furthermore, plants generally follow an exclusion strategy by keeping metals away from physiologically active organs, such as leaves (DalCorso et al., 2013; Manara, 2012).

Influence of soil pH on metal uptake

The pH of the soil upon which plants grow is an important factor in plant–metal uptake from the soil. For example, a lower soil pH promotes metal uptake in crop types and parts (Ebong et al., 2008; Rahmdel et al., 2018). This probably explains the varied metal uptake in crops at some sections of the dumpsite, given the soil pH fluctuations between the sections, that is, 6.4–8.7 at the dump center and 7.4–8.0 at the slope (Awino et al., 2019). These are dumpsite areas where waste burning and scouring for metal scrap among others are practiced. Within the solid waste compost, the pH was 9.0 (Awino et al., 2019). The overall pH variation from slightly acidic soil at the dump center to alkaline soil along the slope may have contributed to the higher metal concentrations in crops collected from the dump center (discussed below). A comparable pH gradient was observed in a study on metal concentrations in Kampala roadside crops (Nabulo et al., 2006). At a low soil pH of 6.0, Cd concentrations (~1 mg/kg) in the roadside crops were 20 times that in crops (0.05 mg/kg) from the Mbale waste dumpsite crops. The low concentrations of Pb, Co, and Cr in crops here may be attributed to their lower mobility in transportation from roots to shoots because of the higher soil pH in Mbale dumpsite soils (Kumar et al., 2019).

Role of the soil-to-plant concentration gradient in metal uptake

The selective absorption of metals by plants from soil through their roots and the subsequent translocation to different plant parts occur partly in response to a metal concentration gradient from the soil to the extremities of plants (Arif et al., 2016). In this context, the lower Pb, Cd, and Cu concentrations measured in crops may be

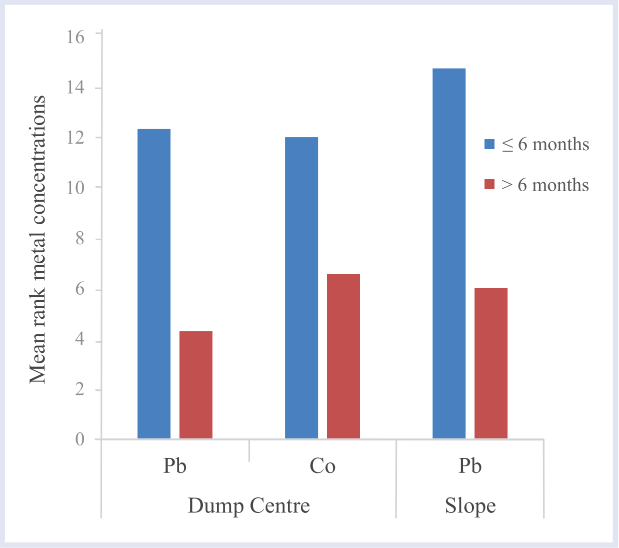

FIGURE 6 Comparison of the mean rank metal concentration variations in leaves of crops that were similar across the Mbale dump center and hill slope

associated with lower soil metal concentrations in the study area as reported earlier by other researchers (OPEP, 2007). Further evidence of this is clear from the higher Fe and Al concentrations found in our crop samples that correlate to higher Fe and Al concentrations in soils as these are the most abundant metals in the earth's crust (Kumar et al., 2019). A higher soil metal concentration would result in an increased uptake (Griffiths & York, 2020), although the availability of Fe in the soil is limited by the low solubility in water and by the antagonistic effects of some nutrients toward Fe uptake in plants (Zhang et al., 2019). Despite the earlier reports on low Zn concentrations in Mbale dumpsite soils (OPEP, 2007), this study measured a high Zn concentration in Mbale crops. This implies that factors other than pH and soil metal concentration are influencing metal uptake in crops at the Mbale dumpsite.

Potential evidence of antagonistic relationships during metal uptake

Metal species and crop types potentially played important roles in the extent to which metals were taken up by crops in the study areas (Cruz et al., 2014; Zwolak et al., 2019). It has been shown that excess Cd and/or Cr in soils suppress the uptake by plants of several essential elements, including Fe and Mn (Kumar et al., 2019). Thus, the high Fe and Mn concentrations found in the Mbale dumpsite crops may have been enhanced because Cd and Cr concentrations in the dumpsite soil were low. An excess of Zn in the roots is also known to hinder the transfer of essential elements such as Mn and Cu to plant shoots/leaves (Kumar et al., 2019). We believe that the Zn concentrations in the Mbale dumpsite crops were not sufficient to inhibit the uptake of Mn and Cu. The low Pb, Co, and Cr concentrations in crops here may be explained by the high soil pH and their lower mobility resulting in less transportation from roots to shoots (Kumar et al., 2019).

Influence of the growth period on metal concentrations in crops

Our comparison of short-term and long-term crop types showed that there were significant differences in the metal concentrations ($p \leq 0.05$), potentially attributable to metabolic differences between crop species (Mwesigye et al., 2019; Saumel et al., 2012). Differences may also arise from root depth and the variation in root uptake kinetics among plant species caused in turn by the variation in the type and number of transporters, assimilation apparatus, and anatomical features (Griffiths & York, 2020). Specifically, most short-term crops have shallow roots and higher transpiration rates, thus promoting greater metal uptake and accumulation from saturated topsoils (Cortez & Ching, 2014; Olowoyo et al., 2012; Ukpong et al., 2013). Short-term crops, therefore, grow faster and have higher transpiration and translocation rates to sustain growth and moisture content. In our context, the latter means increased metal accumulation relative to long-term crops (Madejon et al., 2011; Mahmood & Malik, 2014), hence the higher Fe, Al, Co, and Cu concentrations found in short-term crops and the higher Mn, Ni, and Cr concentrations measured in long-term crops from both the dump center (Figure 5A) and slope (Figure 5B). Thus, short-term crops in the present study probably have a higher density of or a higher affinity for Fe, Al, Co, Cu, and Pb transporters, while the long-term crops had more of or a greater affinity for Mn, Ni, and Cr transporters, resulting in a greater uptake of these metals in the respective crop types. It is clear that the growth period of a crop type influences its metal accumulation.

Comparison of metal concentrations with international food safety limits

In some of the crops, Al, Fe, Zn, Mn, Cu, Ni, Cr, Pb, and Hg concentrations were found to be above internationally accepted food safety standards (compare the first and last columns of Table 5). Whereas Fe, Zn, Mn, Cu, and Co are essential elements required by plants at low concentrations for optimum growth and development, elevated metal concentrations are potentially harmful to both plants and humans alike (Zwolak et al., 2019). Al, Cd, Hg, As, Ni, and Pb are non-essential elements, have *no* safe levels and are potential carcinogens and health hazards. Cr is a mutagen (IRIS, 2019). Their presence in crops, therefore, may pose health risks to consumers. By international food health standards, there is evidence from this study pointing to a disparity in safety among crops cultivated at the Mbale dumpsite and/or their edible parts (Table 5). From the data, Al, Fe, Zn, Mn, Pb, Cr, and Hg concentrations in short-term crops pose health concerns, particularly the consumption of leaves, where Zn, Mn, Fe, and Pb concentrations were found to be mostly above food safety limits. Similarly, Al, Fe, Zn, Mn, Cu, Ni, Cr, Pb, and Hg concentrations in long-term crops are of concern here because they were found to be above food safety levels. Thus, for short-term and long-term crops cultivated at the Mbale dumpsite, Al, Fe, Zn, Mn, Cu,

TABLE 5 Safety of consuming metals in Mbale dumpsite crops against international food safety limits

Crop	Crop part	Safety limits Passing	 Failing
Amaranthus cruentus (Dodo)	Seed, leaf, flower	Cu, Ni, Se, As, Cd, Co	
	Leaf, flower	Zn	Al, Fe, Mn, Pb, Hg, Cr
Carica papaya (Pawpaw)	Fruit, seed, leaf	Ni, Cu, Co, As, Cd, Se, Pb	Mn, Cr, Hg, Al
	Fruit, leaf	Zn	
	Fruit, seed	Fe	
	Seed		Zn
	Leaf		Fe
Coffea arabica (Coffee)	Seed	Ni, Fe, Cu, Zn, Mn, Pb, As, Se, Co, Cd	Al, Cr, Hg
Colocasia esculenta (Balugu)	Leaf	Cu	
	Root, leaf	Fe, Co, As, Se, Cd, Ni, Pb	Al, Cr, Mn, Zn, Hg
	Root		Cu
Manihot esculenta (Cassava)	Leaf, root	Cu, Co, As, Se, Cd	Al, Cr, Hg
	Root	Fe, Mn, Ni, Zn, Pb	
	Leaf		Fe, Mn, Ni, Zn, Pb
Musa acuminata (Banana)	Leaf		Fe
	Fruits, leaf		Al, Cr, Mn, Ni, Hg
Saccharum officinarum (Sugarcane)	Stem	Ni, Fe, Cu, Zn, Mn, Pb, As, Se, Co, Cd	Al, Cr, Hg
Zea mays (Maize)	Seed, leaf	Cu, Ni, Se, As, Cd, Co	
	Seed	Pb, Zn, Fe, Cr	Hg, Al, Mn
	Leaf		Al, Fe, Zn, Mn, Cr, Pb, Hg

Cr, and Pb (and perhaps Ni) concentrations in crops should be monitored. Furthermore, these findings are not unique to the Mbale dumpsite. These metal contaminants in crops cultivated under similar circumstances have been reported elsewhere in Uganda. In particular, short-term crops like *Amaranthus* were reported to be mostly contaminated by Fe, Zn, Cu, Pb, and Co (Awino et al., 2019; Kasozi et al., 2021; Nabulo et al., 2006). Our Mbale study confirms that plant species and the different parts of the same plant vary in their metal uptake due to varied maturity periods. The study also shows that (i) short-term crops and (ii) leafy vegetables have a higher potential to pose health hazards to consumers.

Study limitations

We are aware that metal concentrations in plants are largely dependent on relevant physiochemical soil properties. We were unable to collect soil samples from the study sites for detailed analysis due to logistical challenges and relied mostly on previous soil studies and on the measured soil pH for our discussion. The missing information would have allowed us to perform a correlation analysis as a means of determining the sources of the metals and similarities among the samples. In addition, samples from sites other than the dumpsite were not collected for analysis. This would have allowed a comparative assessment of whether metal contamination was confined to the dumpsite and the extent to which aerial contamination contributed to metal contamination observed in leaves and flowers. The pool of sampling in this study should have been larger than those in current strategies, given the likely high heterogeneity of the distribution of soil metal concentrations. This would have enhanced the reliability of the data. The failure here is, again, due to logistical reasons.

CONCLUSIONS

This study at the Mbale dumpsite demonstrates the importance of monitoring metal concentrations in crops cultivated on contaminated urban soils. The results show that whereas Se, As, and Cd may not have been of health concern in crops in this area, Fe, Al, Zn, Mn, Cu, Ni, Cr, Pb, Co, and Hg were at levels potentially hazardous to consumers of such crops. In particular, leafy vegetables were found to have higher metal concentrations compared to seeds, flowers, stems, fruits, and tubers. We attribute these differences in metal uptake to the specificity of crop types, crop phytotomy, and root depth and/or growth period, and those leaves are the ultimate destination for nutrients including metals.

Here, we also show the need to pay attention to the crop growth period when monitoring metal contamination of food crops grown at dumpsites. A comparison of short-term and long-term crop data to assess the influence of the growth period on the metal concentrations showed significant variability in metal concentrations across the eight commonly consumed crops from both the dumpsite center and slope. Fe, Al, Co, Cu, and Pb concentrations were higher in short-term crops than in long-term crops, while Mn, Ni, and Cr concentrations were higher in long-term crops than in short-term crops. In both cases, Fe, Al, Cu, Pb, Mn, Cr, and Ni concentrations were present above the recommended food safety limits. Overall, all consumable parts of short-term crops and leaves of long-term crops were found to contain high metal concentrations. We cannot, however, recommend discontinuing the consumption of any part of short-term crops or leaves of long-term crops because of a current lack of viable alternatives for the local communities supported by agriculture at the Mbale dumpsite. We, nevertheless, encourage the Mbale municipal authorities to increase awareness of the identified metal hazard in this study. In general, urban farmers could be empowered with knowledge such that they become selective about the crops they grow on dumpsites and the parts of such crops to consume—and when. For example, perhaps more fruits, stems, and root tubers should be grown than short-term leafy vegetable crops. In addition, local communities could also be encouraged to farm on the more alkaline sections of the dumpsite as acidic soils increase the bioavailability of metals, or where possible, to farm in alternative areas outside the urban settings.

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CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

DISCLAIMER

No decision was made by sponsors toward the study design, data collection, analysis, interpretation, report write-up, or submission of the article for publication.

AUTHOR CONTRIBUTION

Funding acquisition, project administration, study concept design, fieldwork, sample preparation, instrumental analysis, statistical analysis, data interpretation, original manuscript drafting, and final write-up, and all other technical and administrative roles: Florence B. Awino. *Study concept design supervision and fieldwork approval, supervision of instrumental and statistical analysis, final manuscript editing, and approval*: William Maher. *Study concept design supervision and fieldwork approval, supervision of statistical analysis, and final manuscript editing*: A. Jasmyn J. Lynch. *Statistical analysis, data interpretation, and manuscript editing*: Patricia B. Asanga Fai. *Re-wrote the results, discussion and conclusion sections, and edited the materials and methods, abstract, and introduction sections*: Ochan Otim.

DATA AVAILABILITY STATEMENT

Data, associated metadata, and calculation tools are available from the corresponding author Florence B. Awino (florence.awino@canberra.edu.au).

SUPPORTING INFORMATION

Figure 1.A. A bar graph of mean (±) metal concentrations (mg/kg) in various crop types at the slope.

Table 1.A. The concentration of metals (mg/kg) in crops grown at the Mbale dumpsite and hillslope. Values less than method detection limits (MDL, 0.1 mg/kg) are replaced by 1/2 MDL.

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