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Original Research

Assessment of bio-accessibility of heavy metals (Cd, Pb, and As) through consumption of medicinal plants collected from different regions in Nyamira-Kenya

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Abstract:

Background: In this study, the total and bio-accessible levels of cadmium (Cd), lead (Pb), and arsenic (As) in 19 Kenyan medicinal plants from two regions (Manga and Borabu) are presented.

Methods: Inductively coupled plasma mass spectrometry (ICP-MS) was used to determine the total and bio-accessed heavy metals in plants. The NIST 1647 plant reference material was used to study the performance of the method. The method offered excellent quality parameters in terms of detection and quantification limits of 0.08 and 0.24 µg/kg, 0.5 and 1.5 µg/kg, and 3.1 and 9.5 µg /kg, linearity (r^2 >0.997) and recoveries of 95%, 99% and 93% for Cd, Pb, and As, respectively.

Results: The dry weights of the plants from Manga and Borabu showed low concentrations of Cd (270 ± 20 and 260 ± 20), As (320 ± 20 and 480 ± 40), and Pb (1230 ± 110 and 1160 ± 100) µg/kg. Significantly higher mean concentrations of Cd, Pb, and As (0.45 ± 0.11, 0.46 ± 0.12 and 0.37 ± 0.10 µg/kg) than (0.32 ± 0.07, 0.34 ± 0.11 and 0.26 ± 0.08 µg/kg) were bioaccessible enzymatically than aquatically from dry weight ($p<0.05$). The percentage bioaccessibility of the elements from the plants ranged from 0.08 to 10.66% and 0.02 to 2.56% for the enzymatic and aquatic procedures, respectively.

Conclusion: The low bioaccessible concentrations of heavy (toxic) elements in plants justify their therapeutic use.

Keywords: Bio-accessibility, Metalloids, Toxicity, Medicinal plants

Introduction

Information on medicinal plants used to treat and manage various diseases and conditions based on traditional folk practices has been perpetuated over generations. Most conventional medicines are very expensive for most rural people. This has led to an upsurge in the use of herbal plants, with no proven scientific efficacy on their curative powers or safety. Medicinal plants are considered safe, although many toxic side effects have been reported [1-3]. The toxic effects reported include allergic reactions, microbial contamination, interactions with drugs and other herbs, altered food consumption, altered body and organ weights, visible pathological changes, and altered enzyme levels [3-5].

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Several African medicinal plants are believed to be effective [1,4,6,7]. Some have been reported to be effective in treating and managing diseases associated with the liver, circulatory, skin, and respiratory systems, and diseases affecting children [8,9]. However, few of them are potentially toxic and carcinogenic [10-12]. The liver and kidneys, which are involved in the metabolism and excretion of chemical compounds in plants, are the organs most vulnerable to toxicity [13]. Therefore, the efficacy and safety of medicinal plants must be evaluated before their use to avoid harmful effects [3,14,15].

The toxicities of Cd (cadmium), Pb (lead), and As (arsenic) have been reported in many parts of the world [3,6,11,13,16,17]. Cd, Pb, and As are present in different amounts in soil, water, and certain foods such as leafy vegetables, potatoes, grains, and seeds [3,18,19]. Large populations in different parts of the world have been reported to be exposed to high levels of As in drinking water. They display various clinicopathological conditions such as cardiovascular and peripheral vascular diseases, developmental anomalies, neurological and neurobehavioral disorders, diabetes, hearing loss, hematologic disorders, and carcinoma [4,10,20]. Environmental As originates from human activities and natural weathering [12,21,22]. The background concentrations of arsenic in the soil range from 1000 to 40,000 µg/kg [23], and high concentrations have been related to soils with high sulfides [24]. Other contributors include the combustion of fossil fuels, smelting, use of As-containing pesticides, copper-chrome arsenate as a wood preservative [25], and the use of arsenic-based cattle dip acaricides [26]. Inorganic forms of As are dominant in most soils and are toxic to humans and other mammals, even at low levels when exposed for an extended period [21,27]. Plants growing in soils under oxidizing conditions have been reported to mainly absorb As (V) [21], which is reduced to As (III) via plant metabolism [28], from which it can be available to the human body when the plant is consumed.

Exposure to high concentrations of Pb causes abdominal pain, constipation, loss of appetite, nausea, vomiting, insomnia, headache, irritability, dizziness, and lead encephalopathy [29], whereas exposure to Cd at high concentrations leads to flu-like symptoms (chills, fever, and muscle pain), and prolonged exposure can damage the lungs, kidneys, and bones [30]. Pb is added to complementary medicine to treat diabetes, spleen enlargement, diarrhea, and skin diseases [31]. Pb has been reported to be the most common toxic element in medicinal plants, where the soluble fraction constitutes less than 10% of the total Pb content [32]. Its bioavailable concentration ranges from 20, 130 to 50,230 µg/kg in herbal medicines, and its bioavailability from herbal medicine is higher than that of Ayurvedic medicine [32]. This implies that the intake of medicinal plants can contribute to lead toxicity in humans and that plant products must be assessed thoroughly to ensure their safety.

Yang *et al.* reported that Pb^{2+}-activated GR-5 DNAzyme produced cleaved substrates that can serve as the activator of Csm6, and the Csm6-DNAzyme tandem improved the sensitivity for detecting Pb^{2+} by 6.1 times compared to the original GR-5 DNAzyme [33]. Zhang et al. proposed a dually amplified and homogeneous DNAzyme assay for the sensitive one-pot detection of lead pollution [34]. Azam *et al*. prepared a cost-effective adsorbent based on ajwa date pits to remove Cu (II) ions from aqueous media. They revealed that treated ajwa date pits (TADP) possessed greater adsorption capability than ajwa date pits (A.D.P.)[35] and further developed a method to remove Cd(II) and Cr(III) heavy metals from aqueous solutions using treated date seeds [36]. Yaqoob *et al*. used novel electrode material and natural organic waste material derived from graphene-polyaniline (GO-PANI) composite anode fabrication to improve the electron transfer rate. They showed that the remediation efficiency of GO-PANI was 65.51% for Cd (II) and 60.33% for Pb (II), which was also higher than that of the unmodified graphene anode (UMGA) [37]. Alqadami *et al*. reported the concentration of heavy metals in cosmetic samples to be in the range of 1.0-12.3 (As), 33-7097 bismuth (Bi), 0.20-0.6 (Cd), 0.70-2700 mercury (Hg), 1.20-143 (Pb), and 2.0-1650 titanium (Ti) (µg/g) [38]. Khan *et al*. further reported levels of heavy metals in acrylic color paints commonly used by school children to range from 0.05 372.59 µg/g [39]. This means that heavy metals are present in environmental and plant samples in different amounts and should be monitored by using various techniques to ensure the safety for the plants users.

The recommended maximum permissible limits for Pb, As, and Cd in herbal plants are 10,000, 400, and 300 µg/kg dry weight (D.W.), respectively [40]. These toxic element concentrations should be maintained through regular monitoring. This requirement is not observed in most countries where medicinal plant use is not controlled or regulated by quality assurance parameters [1,4,6,20]. This should be performed by determining the total content of toxic elements in medicinal plants. More information on toxicity can be gathered by determining the bioaccessibility (the amount of ingested nutrients that are available for absorption in the gut after digestion) and bioavailability (the biologically available chemical fraction of an ingested nutrient that reaches the systemic circulation and the specific sites where it can exert its biological action) of the toxic elements from medicinal plants [10,16,17,31,40-45].

This study presents Cd, Pb, and As concentrations in medicinal plants from two Kenyan regions. The total concentration of elements in the dry matter indicates therapeutic plant toxicity. The actual quantities of these elements in the infusions prepared according to the procedures used by the herbalists should be determined to provide a clear picture of the bio-accessed element, and hence, the toxicity as the consumers use most of the plant products in the infusion form. This was achieved by conducting *in vitro* studies to simulate the digestion process in the gastrointestinal system. Medicinal plants with different farm input applications were collected from two regions. The principal farm inputs were animal manure, agrochemicals, and inorganic fertilizers containing Cd, Pb, and As [46]. Borabu sub-county had intense agricultural activities (frequent applications of farm additives), while Manga sub-county had limited agricultural activities (low application of farm additives). The plants had different element absorption abilities owing to

the different environmental conditions and absorption abilities. The amount of elements absorbed into the human body from the plant is affected by interactions with the food matrix and metabolic processes mediated by the intestine, liver, and microbiota. Consumers accessed different amounts of As, Pb, and Cd, which formed the basis for plant selection.

Experimental

Study sites

The study sites were in Borabu and Manga sub-counties of Nyamira County, which lies between latitudes 0°30″ and 0°45″ South and longitudes 34°45″ and 35°00″ East, with an altitude range of between 1,250 m and 2,100 m above sea level [47], (Figure 1). Nyamira County is divided into two major agroecological zones. The highland zone covers 82 percent, the upper midland zone covers 16 percent, and the lower zone covers the remaining [47]. The lower zones comprise swampy wetlands and valley bottoms, while the hills dominate the upper zones. The primary soil type is red volcanic (Nitosols), which is deep, fertile, and well drained, accounting for 75 percent, and the remaining is clay found in the valley bottoms and swampy areas. The two sub-counties have a bimodal pattern of annual rainfall that is well distributed, reliable, and adequate for the growth of a wide range of flora. Annual rainfall ranges between 1200 mm-2100 mm per annum. The long and short rainy seasons start from April to August and September to December, respectively, followed by three dry spells (January to March). The county's tree cover is mainly agroforestry, with only a small fraction of natural forests around the hilltops and uncultivated farm areas. Ten sampling sites in each sub-county were selected, and three experienced herbalists at each sampling site were randomly selected and recruited for the study. The areas chosen for medicinal plant harvesting are located in the neighborhood of agricultural activities.

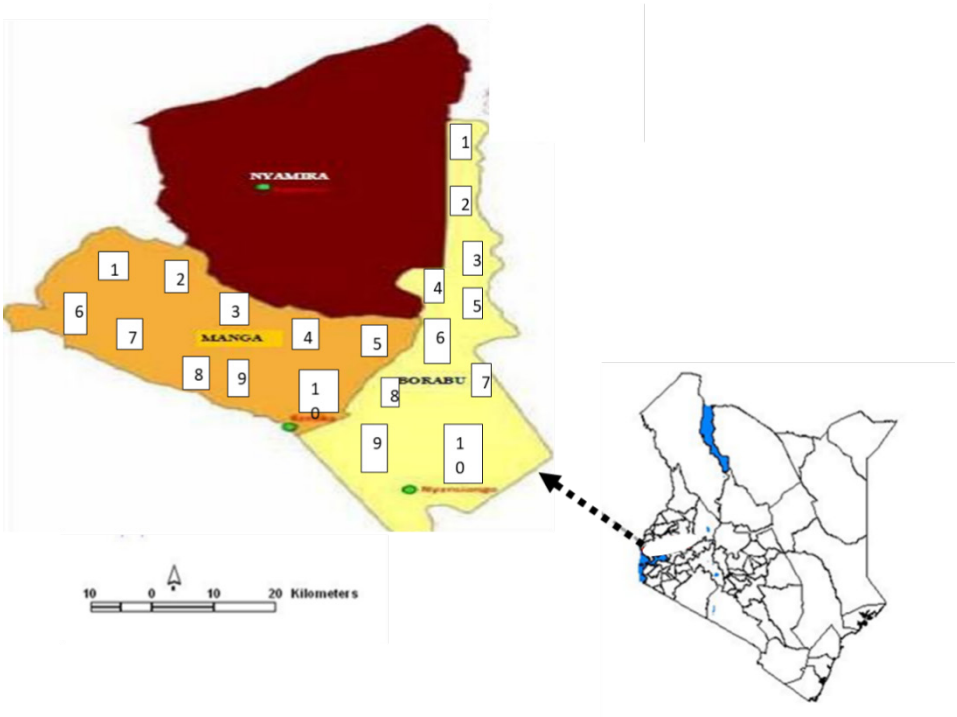

Figure 1 Study area showing sampling points in Manga and Borabu sub-counties

Recruitment of herbalists and sampling of soil and plant materials

Herbalists from the highland agro-ecological zone in Borabu and from the upper midland agro-ecological area in Manga were recruited. The details of herbalist recruitment in the Manga and Borabu regions have been described in our previous studies [43-45]. Based on the herbalists' information on the most commonly used herbal plants and the protocols used in their preparations, the plants used in the study were identified. Thirty herbalists from each study region were recruited and requested to supply one kilogram of dry plant sample for each species. Plant material was collected between February 2022 and April 2022. The 19 plant materials were botanically identified (at Kisii University herbarium and confirmed at the National Museum of Kenya where the voucher plant samples were deposited), the adhering soil particles were removed, washed three times with deionized water and 0.05 M HCl, and then rinsed with distilled deionized water to dislodge

and remove the dust particles. The medicinal plants were then placed in paper bags, separately air-dried under shade, and ground using a different pestle and wooden mortar for each species to avoid exogenous contamination. Care was taken while collecting and storing the plant samples to avoid contamination. Each ground sample was placed in a well-labelled and sealed paper envelope and kept at 4 °C until analysis. The medicinal plants used in the study were *Warburgia ugandensis, Toddalia asiatica, Erythrina abyssinica, Senna didymobotrya, Veronia auriculifera, Croton macrostachyus, Melia azedarach, Mangifera indica, Acacia abyssinica, Tabernae montanastapfiana, Acacia hockii, aloe Aloe vera, Carissa eludes Plecaranthus babatus, Urtica dioica, Bidens pilosa, Solanum indicum, Solanum mauense and Clerodendrum myricoides* (Table 1). Each herbalist was also requested to supply half a kilogram of soil from medicinal plant collection sites. Representative 60 soil samples were collected to determine Cd, Pb, and As contents. The herbalists were requested to collect soil samples from the upper soil horizon (0-15 cm) using a Soil Auger. Fresh soils were dried, crushed, sieved (0.5 mm), and stored at 4 °C in the dark. Before the experiments, the soil samples were maintained at room temperature (20 ± 2 °C) for ten days to equilibrate and ensure comparable conditions in all soils at the start of the experiments. The soil samples were moistened to a water potential of -15 kPa and compacted to a density of 1.3 g cm^{-3} before they were used.

Table 1 Botanical names, common names, local names, plants parts used, and uses of the medicinal plants used in the study

Botanical Name	Common Name	Local Names (Ekegusii)	Part of the plant used	Use of the plant
Warburgia ugandensis Sprague.	Ugandan greenheart	Esoko	Stem bark	Malaria, diabetes, pneumonia, typhoid
Urtica dioica L.	Stinging nettle, California Nettle, Slender Nettle, Tall Nettle	Rise	Leaves	Diabetes, cancer, stomachache, Asthma, anaemia, chest aches, and boils.
Solanum indicum L.	Bush tomato, Indian nightshade, Poisonberry	Omorobo	Roots	Gonorrhea, wounds, boils
Solanum mauense Bitter	nightshade, Purple nightshade, Small flower nightshade, American black nightshade	Ekengenta mbori	Leaves	Diabetes, children's diseases, malaria, cancer
Clerodendrum myricoides Vatke	Blue-flowered tinder wood, Ugandense, Blue Glory Bower, Butterfly Bush	Omonyasese	Root bark	Malaria, gonorrhoea, typhoid, diabetes
Toddalia asiatica Lam.	orange climber, Forest pepper, Wild orange tree	Ekenagwa ekiegarori	Roots	Malaria, gonorrhoea, pneumonia, typhoid, heart disease
Erythrina abyssinica Lam.	Red-hot-poker, Coral tree, Lucky-bean tree	Omotembe	Stem bark	Diabetes, malaria, gonorrhoea, allergy
Senna didymobotrya (Fresen) Irwin &Barneby.	African senna, Popcorn senna, Candelabra tree and Peanut butter cassia	Omobeno	Leaves	Malaria, pneumonia, skin disease, constipation, worms
Vernonia auriculifera Hiern.	Bitterleaf	Omosabakwa	Leaves	Diabetes, pneumonia
Croton macrostachyus Del.	Broad-leaved Croton	Omosocho	Leaves	Malaria, diabetes, typhoid, diarrhoea, bleeding
Melia azedarach L.	Persian lilac, Chinaberry, Bead tree, Syringa, white cedar, mwarubaini (Kiswahili)	Om-warubaine	Leaves	Malaria, STIS, typhoid, anaemia
Magnifera indica L.	Mango	Riembe	Leaves	Diabetes, constipation, burns, and scalds.
Acacia abyssinica Benth.	Umbrella thorn, Flat-top acacia, Nyanga flat-top	Omonyenye	Stem bark	Malaria, gonorrhoea, joints ache
Tabernaemontana stapfiana Britten.	Soccerball fruit	Omobondo	Leaves	Malaria, anemia and fevers
Acacia hockii De wild.	Shittim wood	Omokonge	Stem bark	Diarrhoea, malaria, and fungal infection
Aloe Vera Miller.	Chinese Aloe, Indian Aloe, True Aloe, Barbados Aloe, Burn Aloe, First Aid Plant.	Omogaka	Leaves	Malaria, Asthma, diabetes, cancer, skin diseases, and typhoid
Carissa edulis (Forssk) Vahl.	Simple-spinednum-num, Climbing num-num, Small num-num	Omonyanga-tetia	Roots	Malaria, typhoid, gonorrhoea
Plectranthus barbatus Andr.	Big leave Plectranthus, Indian coleus, and Forskohlii	Omoroka	Leaves	Worms, diarrhoea, diabetes
Biden spilosa L.	Blackjack, Beggarticks, cobbler's pegs, and Spanish needle	Ekemoga-mogia	Leaves	Diabetes, malaria, worms, gonorrhea

Source: Kokwaro, 2009

Reagents, chemicals, and reference materials

Analytical-grade reagents were used in this study. Calibration standards were prepared using Certipur stock solutions (Merck, Darmstadt, Germany) in 3% nitric acid. Deionized water used in the study was prepared using a Millipore system. The blank values of Cd, As, and Pb in deionized water were 0.001, 0.001, and 0.003 µg/kg, respectively. The reagents used for the enzymatic digestion were pepsin, sodium malate, sodium citrate, lactic acid, acetic acid, bile salts, and pancreatin. The reference material was NIST 1647 (peach leaves), purchased from L.G.C. Standards, Germany.

Determination of total content of Cd, Pb, and As by ICP-MS in medicinal plants

The total concentrations of Cd, Pb, and As in the plant and soil samples were determined via closed-vessel microwave digestion with subsequent analysis using Inductively Coupled Plasma Mass Spectrometry (ICP-MS) [43]. Before microwave digestion, the plant and soil samples were finely ground using a ball mill consisting of zirconium oxide vessels and balls (Pulverisette 6, Fritsch, Germany). Aliquots of 50 mg of the ground plant and soil samples were digested in triplicate using 2 mL of 7.9 M nitric acid (Suprapur, Merck, Germany) and 1 mL of 6.29 M hydrogen peroxide (Suprapur, Merck, Germany) in a MARS 5 closed vessel microwave system (C.E.M., Germany) at 160°C using 1150 watts of power. Complete digestion of the organic matrix in the plant was achieved with the occasional addition of silicate residues. The mineralized solution was transferred to calibrated polystyrene sample vials and made up to 10 mL with deionized water. Blank and plant reference materials were processed using the same procedure. The Pb, Cd, and As levels in the plant and soil sample digests were determined using an Agilent 7500 quadrupole ICP-MS with a collision cell in He-Mode (Agilent Technologies, Japan). A micromist nebulizer with a double-pass spray chamber was used for the analysis. The He flow rate of the collision cell was 4 mL/min, the sample uptake rate was 400 µL/min, and an argon (nebulizer gas) flow rate of 0.98 L/min. The mean and standard deviation from triplicate digestion and measurements were calculated (n = 3) (and ICP-MS software was used to determine the amount of each element in each sample in triplicate). The NIST 1647 peach plant reference material was analyzed for quality control. ICP-MS results were backed up with the analysis of plant reference material NIST 1647 (peach leaves), which had recoveries of 95% for Cd, 99% for As, 93% for Pb, and 5.7% moisture content, and the detection and quantification limits for the elements were As, 3.1 and 9.5 µg/kg; Cd, 0.08 and 0.24 µg/kg and Pb, 0.5 and 1.5 µg/Kg. This indicated that the digestion and quantification procedures yielded valid and reliable results. The few outliers for Pb and As were not included, likely because of the homogeneity of the plant samples used for ICP-MS analysis. The separate aliquots of pre-ground material used could have led to mass loss due to the open digestion process and moisture in the plant material.

Determination of bio accessible of Cd, Pb, and As from medicinal plants

An *in vitro* gastrointestinal digestion method was used in this study [44,48]. 0.3 g of accurately weighed plant material was placed in a 50 mL polypropylene tube and treated with 30 mL of gastric solution (1250 µg of pepsin, 5×10^5 µg of sodium malate, 5×10^5 µg of sodium citrate, 420 µL of 11.3 M (molar) lactic acid and 500 µL of 17.4 M acetic acid, made up to 1 L with deionized water, and the pH was adjusted to 2.5 with 11.65 M hydrochloric acid) and an equal amount was placed in a 50 mL polypropylene tube, and deionized water was added to the 40 ml mark. The mixtures were shaken at 100 × g in a thermostatic bath maintained at 37 °C for one hour. The mixtures were then centrifuged at 3000 × g for 10 min, and a 5 mL aliquot was removed, filtered through a 0.45 µm micro filter into a 5 ml polypropylene tube, and stored at 4 °C. The removed solution and water were replaced with the original gastric acid solution and deionized water to retain the actual amount of the solutions.

Small intestinal digestion conditions were created by adding 52.5 mg bile salts and 15 mg pancreatin into the same sample tube. A saturated sodium bicarbonate solution was added to adjust the pH to 7.0. The mixtures were maintained at 37 °C in a thermostatic bath, shaken at 100 g for 2 h, and then a second 5 ml aliquot was removed and filtered. The remaining sample solutions were centrifuged at 3000 × g for 10 min and filtered, and the residues were retained for acid microwave digestion and ICP-MS analysis. The extracted samples were stored at 4 °C and analyzed within 24 h by ICP-MS. Triplicate gastric, intestinal, and water extracts of the plants were performed in every batch and diluted to 1:10 w/w before analysis. The bioaccessible toxic element contents in the plant extracts (gastric and intestinal fractions) were determined by ICP-MS, and the daily element intake (the actual oral dose of the element) was determined using the Bolan *et al*. (2016) equation [32].

$$DI = D.D. \times W \times MC$$

Where DI is the daily intake of the element, D.D. is the daily dose of the medicinal plant, W is the dry weight of the medicinal plant taken daily, and MC is the therapeutic plant element content (mg/kg). The recoveries of the reference material NIST 1647 (peach leaves) for the gastric and intestinal phases were 94% for Cd, 96% for As, 89% for Pb, and 5.7% for moisture content, and 91% for Cd, 98% for As, 92% for Pb, and 4.3%, respectively.This indicated that the digestion and quantification procedures yielded valid and reliable results.The amounts of As, Cd, and Pb extracted in each phase were divided by the total amount of element extracted (extracted by acid digestion) to obtain the elemental extraction percentage (%).

Data analysis

The results obtained from laboratory procedures were analyzed using the I.B.M. SPPS version 23 program. Descriptive analyses were conducted on the concentrations of toxic elements in medicinal plants. The relationships between the different study areas' medicinal plant element concentrations and the independent variables were determined by correlation coefficient and linear regression, where applicable. Linear regression equations were used to determine the total and bioaccessible quantities of toxic elements in medicinal plants. The relationship between the total concentrations of the plant elements in the study areas and the plant species was analyzed by t-test and correlation coefficients to establish significant differences between the same plant species from different study sites, different plant species from the same study area, and different plant species from other study sites. A binomial t-test was carried out to determine whether the plant and the study area from where the plants were collected contributed significantly to the levels of toxic elements in the plants. A parametric statistical test (Student's t-test) was used to test the different hypotheses. An α value of 0.05. was adopted as the critical level for all statistical testing, giving a 95% confidence level.

Results and discussion

Total content of Cd, Pb, and As in the medicinal plants

The mean concentrations of Cd, Pb, and As in the medicinal plant species collected from the two regions (Manga and Borabu) are presented in Table 2. The mean arsenic level in the Soil from Manga was 6,020 ± 10 µg/kg, while that from Borabu was 8,150 ± 30 µg/kg (D.W.). The As levels in the soil were below the global recommended average limit for agricultural soils by the European Union (10000 µg/kg). As concentrations in plants from the Manga region ranged from 80 ±10 µg/kg to 900 ± 70 µg/kg. Twelve plants from the Borabu region had mean arsenic concentrations ranging from 200 ± 10 to 620 ± 70 µg/kg, with *S. didymobotrya* (21,00 ± 200 µg/kg) and *B. pilosa* (940 ± 70 µg/kg) having concentrations above the range, and five plant species having mean levels below this range. The five plant species, *W. ugandensis*, *V. auriculifera*, *M. indica*, *A. abyssinica*, and *A. hockii* from Manga and Borabu, had As concentrations of <200 µg/kg.

The mean As content in plants from the same sampling region was frequently broad, with maximum concentrations approximately 11-fold higher than the minimum concentration in Manga sub-county and 30-fold higher in Borabu sub-county. Borabu sub-county plants had slightly higher mean As concentrations than Manga sub-county plants. *S. didymobotrya*, *C. myricoides*, *S. mauense*, *C. edulis*, *B. pilosa*, and *U. dioica* from the two regions had high concentrations of As. *W. ugandensis* from Borabu and Manga had the lowest arsenic absorption efficiencies from the soil (0.86%, 1.33%), while *S. didymobotrya* had the highest (14.95%, 25.77%), which was calculated as

$$\left(\frac{Total\ plant\ metalloid\ mass}{Mass\ of\ metalloid\ in\ the\ soil}\times 100\%\right)$$

(Table 3). All medicinal plants in the two regions except for *S. didymobotrya* from both study areas and *B. pilosa* from Borabu had less than 10% arsenic absorption efficiency. Because *B. Pilosa* and *S. didymobotrya* species have high As absorption efficiency, they can be used for phytoremediation in arsenic-contaminated soils. The As absorption efficiency of plants from the soil can be affected by soil pH, as it affects its bioavailability through adsorption and desorption reactions [49]. Acidic and basic soils have greater As solubility than neutral soils because of their more significant adsorption to soil colloids [50]. Charges develop on soil particle surfaces during the As adsorption process, affecting desorption [21,51]. The levels of arsenic in most plants from Borabu sub-county resulted from animal manure, agrochemicals, or inorganic fertilizers [52]. Our findings for As are similar to those reported in different flora at less than 10-400 µg/kg [53,54]. As concentrations of not detected to 9 µg/kg reported in other plants collected from the Awash River basin in Ethiopia were lower than our findings, the Awash basin experiences frequent floods as it is flat, and its soil is mainly clay [55]. In contrast, those in the present study were collected from hilly areas with primarily volcanic soil. Clays and oxides control the solubility of As by adsorption to hydroxyl groups, presence of a large number of binding sites, co-precipitation, and precipitation [56-62]. The soils of Borabu had a relatively higher clay content than those of Manga. This reduced As bioaccessibility to the plants from Borabu, even if there were high inputs of farm additives. The plants in the present study grew in different microecological areas with various biotic and abiotic factors, accounting for intraspecies differences in the bioaccessibility of the element under investigation. The maximum allowable limit of As for humans in consumed foods is 200 µg/kg per day [40]. The presence of As, even in trace amounts in plants, is a primary risk to the food safety and human health, as As is classified as a toxic element [40].

The mean concentration of Cd in the Soil from Manga was 4,880 ± 70 µg/kg, while that from Borabu was 7380 ± 40 µg/kg. The Cd concentration in Manga plants ranged from <80 to 980 µg/kg, while that in Borabu ranged from <80 to 980 ± 60 µg/kg, with the highest in *T. stapfiana* and the lowest in *A. vera* (Table 2). Plants from Borabu were collected from areas with high farm additives, and 54% of the plants had higher levels of Cd than those from Manga. Six plants from both regions, *V. auriculifera, C. macrostachyus, M. azedarach, T. stapfiana, B. pilosa,* and *C. myricoides,* had mean Cd concentrations above the

Table 2 The mean total levels (µg /kg) of Cd, Pb, and As in plants from Manga and Borabu Study areas, Kenya

Botanical Name	Arsenic (As) (× 10^3 µg/kg)		Cadmium (Cd) (× 10^3 µg /kg)		Lead (Pb) (× 10^3 µg /kg)	
	MMP	BMP	MMP	BMP	MMP	BMP
W. ugandensis	0.08 ± 0.01	0.07 ± 0.02	0.13 ± 0.03	0.13 ± 0.03	0.43 ± 0.02	0.37 ± 0.06
S. indicum	0.11 ± 0.06	0.50 ± 0.04	<0.08	<0.10	1.00 ± 0.10	1.25 ± 0.05
T. asiatica	0.27 ± 0.02	0.50 ± 0.05	0.17 ± 0.02	0.13 ± 0.01	1.40 ± 0.10	1.25 ± 0.05
E.abyssinica	0.09 ± 0.01	0.50 ± 0.02	<0.09	<0.10	0.17 ± 0.04	0.27 ± 0.03
S. didymobotrya	0.90 ± 0.07	2.10 ± 0.20	0.16 ± 0.05	0.20 ± 0.05	0.65 ± 0.03	0.39 ± 0.03
V.auriculifera	0.40 ± 0.03	<0.20	0.34 ± 0.05	0.22 ± 0.06	0.73 ± 0.03	0.40 ± 0.10
P. barbatus	0.30 ± 0.02	0.35 ± 0.07	0.16 ± 0.05	0.16 ± 0.04	1.00 ± 0.04	1.62 ± 0.10
U.dioica	0.30 ± 0.02	0.62 ± 0.07	0.10 ± 0.02	0.09 ± 0.02	4.20 ± 0.20	2.69 ± 0.09
C. macrostachyus	0.30 ± 0.02	0.45 ± 0.05	0.98 ± 0.03	0.50 ± 0.04	2.20 ± 0.10	0.81 ± 0.08
B. Pilosa	0.39 ± 0.08	0.94 ± 0.07	0.51 ± 0.04	0.33 ± 0.04	2.76 ± 0.10	4.50 ± 0.40
M. azedarach	0.20 ± 0.02	0.30 ± 0.01	0.40 ± 0.02	0.70 ± 0.10	0.24 ± 0.02	1.70 ± 0.30
S. mauense	0.50 ± 0.03	0.5 ± 0.02	0.24 ± 0.02	0.42 ± 0.04	2.90 ± 0.10	1.76 ± 0.10
M.indica	0.35 ± 0.07	<0.20	0.08 ± 0.02	<0.10	0.25 ± 0.02	0.21 ± 0.05
A.hockii	0.24 ± 0.07	<0.20	0.09 ± 0.04	<0.10	0.16 ± 0.02	0.15 ± 0.01
A.abyssinica	0.24 ± 0.05	<0.20	0.14 ± 0.02	<0.10	0.34 ± 0.02	0.72 ± 0.09
C.myricoides	0.60 ± 0.10	0.40 ± 0.03	0.47 ± 0.05	0.16 ± 0.04	1.72 ± 0.09	0.64 ± 0.04
C.edulis	0.39 ± 0.07	0.51 ± 0.08	0.27 ± 0.03	0.32 ± 0.05	1.30 ± 0.10	1.36 ± 0.04
T.stapfiana	0.10 ± 0.05	0.20 ± 0.01	0.53 ± 0.04	0.98 ± 0.06	0.14 ± 0.03	0.22 ± 0.03
A. Vera	0.28 ± 0.07	0.32 ± 0.08	0.12 ± 0.02	<0.09	1.75 ± 0.05	1.80 ± 0.10
R		**0.731**		**0.647**		**0.713**
t-test		**0.163**		**0.441**		**0.370**
Soil	6.02 ± 0.01	8.15 ± 0.03	4.88 ± 0.07	7.38 ± 0.04	7.82 ± 0.03	8.75 ± 0.03

BMP- Borabu plant/ soil; MMP- Manga plant/soil. ± standard deviation

WHO permissible limit [33]. The medicinal plants from the two regions with low Cd levels were *A. vera, M. indica, A. hockii, S. indicum,* and *E. abyssinica*. The lowest Cd absorption efficiencies from the soil were observed in *A. vera* and *U. dioica* (1.20%), while the highest was observed in *C. macrostachyus* (20.08%) (Table 3). The medicinal plants investigated had less than 10% cadmium absorption efficiency, except for *T. stapfiana* from both study areas and *B. pilosa* and *C. macrostachyus* from Manga. Because of their high absorption efficiencies, *T. stapfiana, B. pilosa,* and *C. macrostachyus* can be used for phytoremediation of cadmium-contaminated soils. Cadmium concentrations in plants from the same sampling region ranged widely, with maximum and minimum concentrations approximately 12-fold higher in manga and 11-fold higher in borabu.

Cadmium enters the environment through the burning of fossil fuels and municipal waste. Cd is highly carcinogenic, teratogenic, and mutagenic [63]. When municipal waste and sewage are not disposed of properly, as occurs in the study areas, plants growing in the area absorb Cd. The biological and biochemical transformation of Cd in plant roots leads to differences in bioavailability and toxicity in plants [63]. The inorganic form of free Cd ions in soil has been reported to be more dominant. Its mobility does not depend on its ability to complex with organic ligands and its transfer into the xylem is an active transport process [64]. Zn competitively inhibits Cd uptake through similar metabolic pathways. Cd and Fe exhibit close homeostasis in plants. Soil pH controls dissolution, precipitation, ion exchange adsorption, redox reactions, and other complex reactions within the soil that control Cd bioavailability, solubility, and plant absorption [59,65]. Soil pH is a significant factor influencing Cd and Zn bioavailability [61]; for example, decreases in soil pH increase Cd solubility and bioavailability [66,67,69]. The soil pH at the plant collection sites differed from one region to another, accounting for inter- and intra-regional medicinal plant Cd concentrations. Zn fertilizers can be applied on soils with high Cd levels to facilitate the safety of the plants, as Zn competitively inhibits its absorption.

The mean concentration of Pb in the Manga soils was 7,820 ± 30 µg/kg. At the same time, those of Borabu were 8,750 ± 30 µg/kg (Table 2), which were lower than the European Union's recommended global limit for agricultural soils (10,000 µg/kg). Pb concentrations in plants from Manga ranged from 160 ± 20 (A. hockii) to 4,200 ± 200 µg/kg (*U. dioica*) and Borabu from 150 ± 10 µg/kg (*A. hockii*) to 4,500 ± 400 µg/kg (*B. Pilosa*). *M. indica* and *T. stapfiana*

from the study areas had the lowest Pb absorption efficiencies from the Soil, with 1.16% and 1.17%, respectively, while *B. pilosa* and *U.dioica* had the highest, with 24.79% and 34.94%, respectively (Table 3). The medicinal plants studied had Pb absorption efficiency from the Soil of less than 10% except for *U.dioica* and *B. pilosa* from both study areas, *T. asiatica*, *C. macrostachyus*, *S. mauense*, *C.myricoides*, *C.edulis*, and *A. Vera* from Manga. *U.dioica, B. pilosa, T. asiatica, C. macrostachyus, S. mauense, C.myricoides, C.edulis,* and *A. Vera* can be used for Pb phytoremediation in Pb-contaminated soils. The mean concentrations of Pb in medicinal plants are below the permissible limit set by the WHO [40]. Five plants from Manga and Borabu with low mean concentrations of Pb were *W. ugandensis, T. stapfiana, A. hockii, M. indica,* and *E. abyssinica.* The mean Pb concentrations in plants from the same sampling region were frequently broad, with maximum and minimum concentrations approximately 27-fold higher in manga and 30-fold higher in borabu. Pb concentrations in the medicinal plants reported in the present study were lower than those reported in vegetables grown on Pb-contaminated soils in Thika town, Kenya [46,47] and in vegetation grown on contaminated soils, industrial effluents, or in areas with high vehicular fumes [70]. Plants used in this study were collected from rural areas with low contamination levels. Contaminated soils raise Pb concentrations due to chipping, scraping, sanding, and sandblasting of structures bearing lead-based paint [46]. Soil Pb contamination could also result from the previous use of tetraethyl lead as an anti-knock ingredient in gasoline and lead arsenate used in insecticides for fruit orchards [29,70]. This accounted for the higher Pb levels in plants collected from urban areas than those in the present study. High mean Pb concentrations of 10400 µg/kg were reported in herbs purchased from general stores in Irbid City, Jordan, as most of the samples were collected from contaminated sites [71].

Chemical and biological reactions occurring in the soil where the plants grow affect their elemental absorption ability. Elements interact with each other through several chemical and biological reactions. Soil pH, sorbent nature, organic and inorganic ligands, root exudates, and nutrients have been shown to affect element mobility in soil [72]. The bioavailable fraction of a toxic element is the proportion of the total pool of soil elements that can be extracted with a chemical reagent and potentially absorbed by plants [72]. The solubility of toxic elements is low and they exist in a state unavailable for plant absorption [73]. In natural ecosystems, bioavailable forms of toxic elements are inadequate for inducing plant toxicity. Most are reported as soil solution, exchangeable, organically and colloidal bound, residual, and within the primary phase of the mineral fractions [58,74-76]. The soil properties and processes responsible for the bioavailability of toxic elements in plants include organic matter, soil pH, redox potential, clay, and oxide levels [77-79]. Soil redox conditions also affect the solubility, speciation, and bioaccessibility [80,81].The redox conditions of the harvested areas of the plants (Borabu and Manga) were

Table 3 Percentage absorption efficiency of heavy metal by plants from the Soil

Plant Species	Arsenic		Cadmium		Lead	
	MMP	BMP	MMP	BMP	MMP	BMP
W. ugandensis	1.329	0.859	2.664	1.762	3.577	2.039
S. indicum	1.827	6.135	1.639	1.355	8.319	6.887
T. asiatica	4.485	6.135	3.484	1.762	11.647	6.887
E.abyssinica	1.495	6.135	1.844	1.355	1.414	1.488
S. didymobotrya	14.950	25.767	3.279	2.710	5.408	2.149
V.auriculifera	6.645	2.454	6.967	2.981	6.073	2.204
P. barbatus	4.983	4.294	3.279	2.168	8.319	8.926
U.dioica	4.983	7.607	2.049	1.220	34.942	14.821
C. macrostachyus	4.983	5.521	20.082	6.775	18.303	4.463
B. Pilosa	6.478	11.533	10.451	4.472	22.962	24.793
M. azedarach	3.322	3.681	8.197	9.485	1.997	9.366
S. mauense	8.306	6.135	4.918	5.691	24.126	9.697
M.indica	5.814	2.454	1.639	1.355	2.080	1.157
A.hockii	3.987	2.454	1.844	1.355	1.331	0.826
A.abyssinica	3.987	2.454	2.869	1.355	2.829	3.967
C.myricoides	9.967	4.908	9.631	2.168	14.309	3.526
C.edulis	6.478	6.258	5.533	4.336	10.815	7.493
T.stapfiana	1.661	2.454	10.861	13.279	1.165	1.212
A. Vera	4.651	3.926	2.459	1.220	14.559	9.917

influenced by microbial processes, oxygen concentration, organic matter, pH, and microclimatic conditions. Redox conditions have been shown to impact elemental bioaccessibility [81,82] and could influence their concentrations in plants in our study. Some toxic elements have been reported to be more toxic and mobile in the most reduced form, e.g., As (III) [83]; however, in others, such as Zn, Ni, Cd, and Cu, the redox conditions play a minor role in the solubility and bioavailability [84].

Bio-accessibility of As, Cd, and Pb from plants in gastric and intestinal phases

Lead from S. mauense had the least (0.02%) bio-accessed gastric phase element, whereas As from S. indicum had the highest (18%). The lowest bio-accessible element in the intestinal phase was Pb from P. barbatus and S. mauense (0.06%), whereas As from S. indicum had the highest bio-accessibility (45%) (Table 4). The least total enzymatic bio-accessed element was Pb from P. barbatus (0.08%), whereas the highest was As from S. indicum (63%). Lead from U. dioica was the least aqueous bio-accessed element (0.01%), whereas As from S. mauense was the highest (22%) (Figures 2-4, Tables S1-S2). The bioaccessibility of each enzymatic phase differed from one element to another and among plants. The total aquatically bioaccessed Cd, Pb, and As were statistically significantly different from those accessed enzymatically. The amounts of Cd, Pb, and As in the gastric and intestinal phases were not significantly different (P <0.05) (Supplementary Table S1). There was a positive correlation between the amounts of Cd, Pb, and As in the gastric and intestinal phases, with the highest correlation observed for lead (r = 0.931). The lowest arsenic concentration (r = 0.103), and a similar trend was observed when the total enzymatic and aquatic bio-accessed concentrations were compared.

Table 4 Total, enzymatic and water bioaccessible, % of enzymatic and water bio accessed and daily intake of As, Cd and Pb from Manga plants

Medicinal plant	Metalloid	Enzymatic bio-accessible (µg/kg)	Water bio-accessible (µg/kg)	Total (µg/kg)	% Enzymatic extraction	% Water extraction	Daily intake (µg/day)
S. indicum	As	0.697	0.247	110	0.634	0.225	0.014
	Cd	1.956	0.493	80	2.445	0.616	0.039
	Pb	3.71	0.31	1000	0.371	0.031	0.074
P. barbatus	As	0.579	0.348	300	0.193	0.116	0.012
	Cd	1.557	0.589	160	0.973	0.368	0.031
	Pb	0.490	0.030	1000	0.049	0.003	0.017
U.dioica	As	0.618	0.231	300	0.206	0.077	0.012
	Cd	2.350	0.288	100	2.350	0.288	0.047
	Pb	7.392	0.504	4200	0.176	0.012	0.148
B. Pilosa	As	0.460	0.265	390	0.118	0.068	0.001
	Cd	3.912	0.622	510	0.767	0.122	0.078
	Pb	6.513	0.552	2760	0.236	0.020	0.130
S. mauense	As	0.690	0.330	500	0.138	0.066	0.014
	Cd	0.192	0.034	240	0.080	0.014	0.038
	Pb	2.436	0.493	2900	0.084	0.017	0.049
C. myricoides	As	0.906	0.246	600	0.151	0.041	0.018
	Cd	3.130	0.381	470	0.666	0.081	0.063
	Pb	1.995	0.344	1720	0.116	0.020	0.040
C. edulis	As	0.842	0.187	390	0.216	0.048	0.017
	Cd	1.631	0.308	270	0.604	0.114	0.033
	Pb	1.846	0.208	1300	0.142	0.016	0.037
A. Vera	As	1.064	0.134	280	0.380	0.048	0.021
	Cd	1.302	0.431	120	1.085	0.359	0.026
	Pb	1.453	0.350	1750	0.083	0.020	0.029

Table S1 Size fractionation of Pb, Cd, and As in aqueous plant extracts using sequential filtration (mean and standard deviation). The extraction efficiency of the aqueous extraction compared with the total element extracted (digestion)

Element	Plant Species	0.45-5µm [µg/g]	10kDa-0.45µm [µg/g]	3-10kDa [µg/g]	<3kDa [µg/g]	Extraction Efficiency (%)	Total elemental Concentration (digestion) [mg/kg]
Pb	*S. indicum*	0.124	0.289	0.000	0.000	0.041	1.00 ± 0.10
	P. barbatus	0.096	0.584	0.000	0.000	0.068	1.00 ± 0.04
	U. dioica	0.045	0.321	0.150	0.000	0.012	4.20 ± 0.20
	B. pilosa	0.123	0.373	0.024	0.000	0.019	2.76 ± 0.10
	S. mauense	0.153	0.322	0.000	0.000	0.016	2.90 ± 0.10
	C. myricoides	0.123	0.321	0.082	0.000	0.031	1.72 ± 0.09
	C. edulis	0.084	0.121	0.000	0.000	0.016	1.30 ± 0.10
	A. vera	0.072	0.212	0.102	0.045	0.025	1.75 ± 0.05
	NIST 1647	0.303	0.189	0.081	0.010	0.042	1.38 ± 0.07
Cd	*S. indicum*	0.049	0.101	0.106	0.157	0.516	0.08
	P. barbatus	0.400	0.135	0.000	0.000	0.334	0.16 ± 0.05
	U. dioica	0.099	0.086	0.017	0.008	0.209	0.10 ± 0.02
	B. pilosa	0.014	0.322	0.232	0.009	0.113	0.51 ± 0.04
	S. mauense	0.023	0.149	0.080	0.172	0.176	0.24 ± 0.02
	C. myricoides	0.030	0.173	0.029	0.112	0.073	0.47 ± 0.05
	C. edulis	0.019	0.260	0.072	0.065	0.109	0.27 ± 0.03
	A. vera	0.229	0.169	0.067	0.090	0.395	0.12 ± 0.02
	NIST 1647	0.242	0.288	0.023	0.091	0.225	0.25 ± 0.04
As	*S. indicum*	0.126	0.016	0.031	0.081	0.231	0.11 ± 0.06
	P. barbatus	0.225	0.018	0.059	0.065	0.122	0.30 ± 0.02
	U. dioica	0.046	0.095	0.014	0.104	0.086	0.30 ± 0.02
	B. pilosa	0.108	0.066	0.018	0.087	0.071	0.39 ± 0.08
	S. mauense	0.112	0.119	0.013	0.121	0.073	0.50 ± 0.03
	C. myricoides	0.062	0.101	0.016	0.280	0.076	0.60 ± 0.10
	C. edulis	0.210	0.185	0.061	0.088	0.126	0.39 ± 0.07
	A. vera	0.087	0.096	0.017	0.081	0.101	0.28 ± 0.07
	NIST 1647	0.135	0.179	0.032	0.091	0.079	0.45 ± 0.03

Table S2 Correlation coefficient(r) t test and F test for Pb, Cd and As extracted in the gastric (E_1) and intestinal (E_2) phases and those total enzymatic (E) in aqueous extracts (W) from selected plant species

Plant Species										
						Pb				
	E_1	E_2	r test	t test	F test	Total E	Total W	r test	t test	F test
S. indicum	1.20	2.52				3.71	0.31			
P. barbatus	0.28	0.56				0.83	0.49			
U. dioica	2.78	4.63	0.931	0.128	0.203	7.41	0.50	0.32	0.011	4.22
B. Pilosa	1.89	4.69				6.52	0.56			
S. mauense	0.57	1.88				2.45	0.49			
C. myricoides	0.77	1.22				1.99	0.44			
						Cd				
S. indicum	0.85	1.11				1.96	0.49			
P. barbatus	0.58	0.97				1.56	0.59			
U. dioica	0.38	1.97	0.095	0.672	0.243	2.35	0.29	0.217	0.0003	0.0008
B. Pilosa	2.28	1.64				3.91	0.62			
S. mauense	1.06	0.87				1.92	0.35			
C. myricoides	1.82	1.31				3.13	0.38			
						As				
S. indicum	0.20	0.49				0.70	0.25			
P. barbatus	0.26	0.32				0.58	0.35			
U. dioica	0.33	0.28	0.103	0.525	0.500	0.62	0.23	0.681	0.0005	0.200
B. Pilosa	0.27	0.19				0.46	0.27			
S. mauense	0.36	0.33				0.69	0.33			
C. myricoides	0.44	0.47				0.90	0.44			

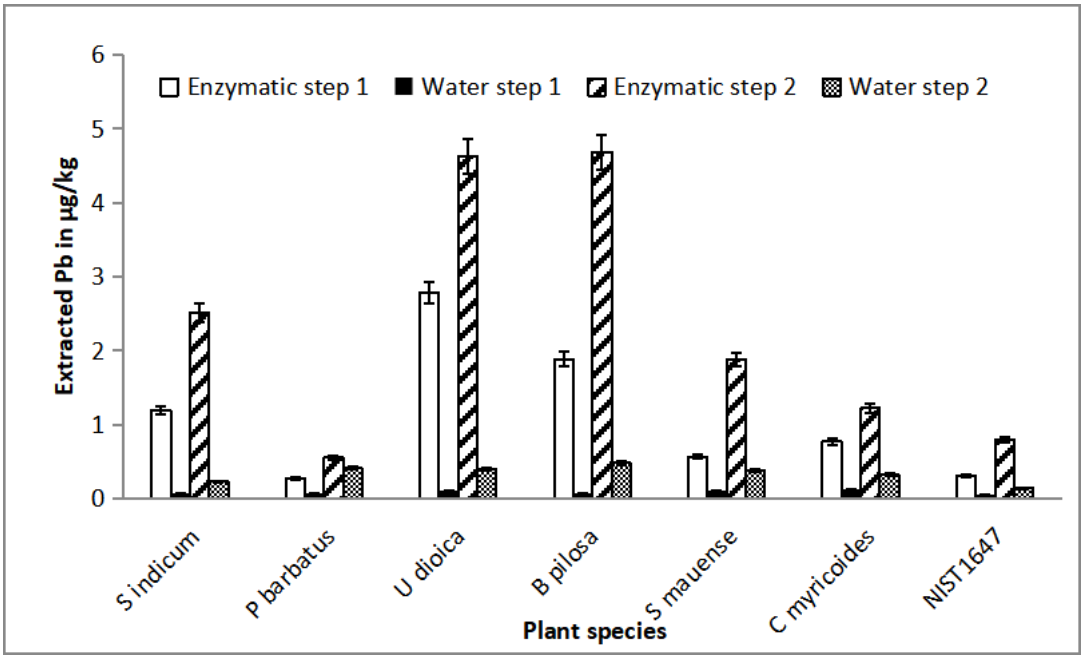

Figure 2 Comparison of 2-step of maximum mass of aqueous and enzymatic extraction for plant Pb in selected plant species

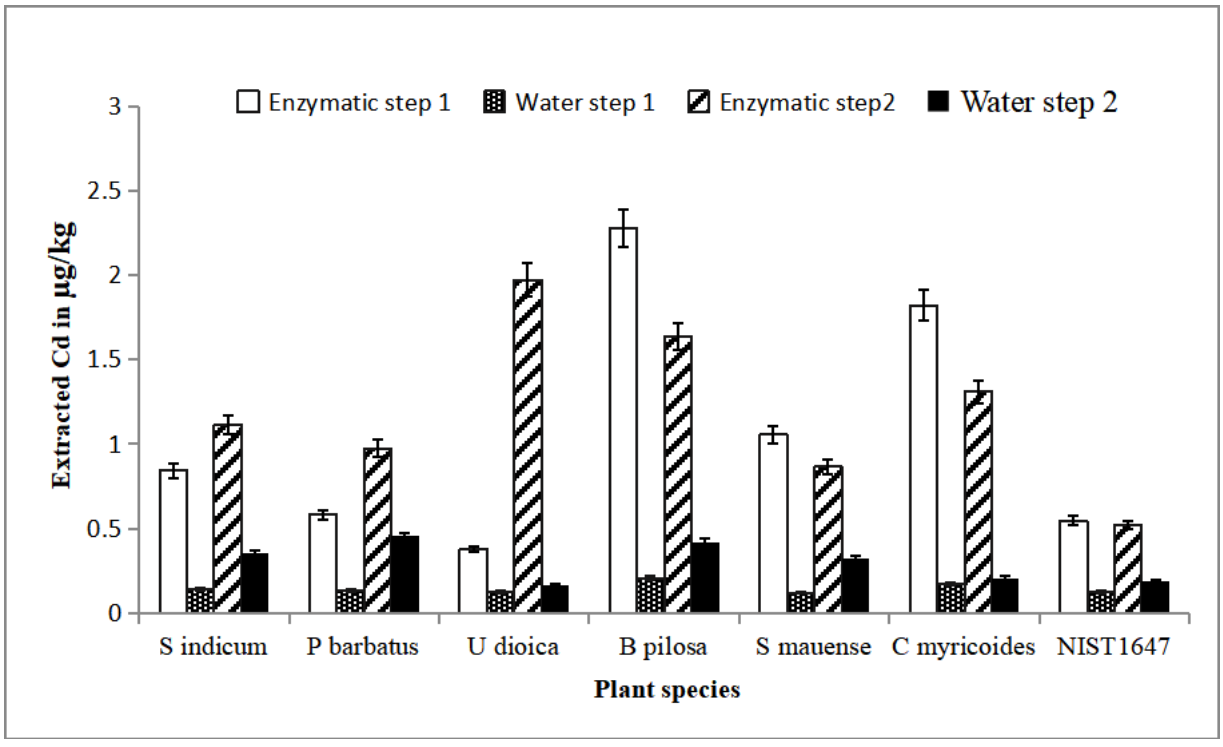

Figure 3 Comparison of 2-step of maximum mass of aqueous and enzymatic extraction for Cd in selected plant species

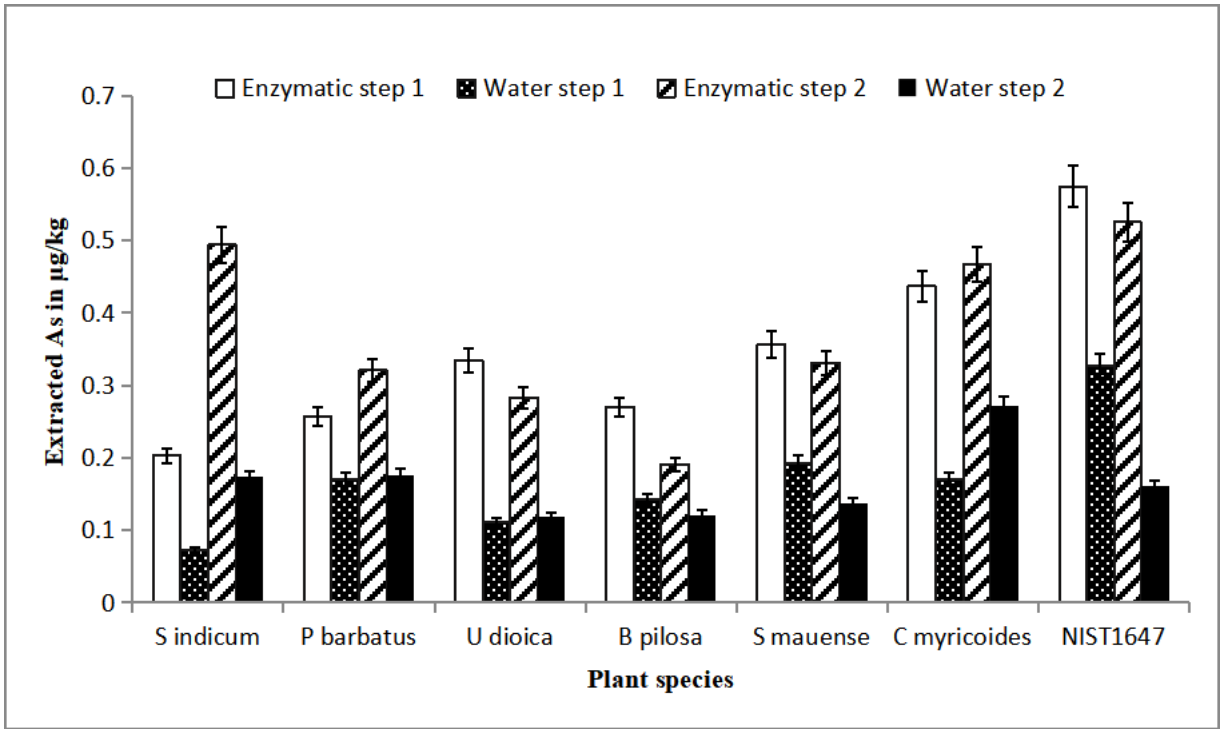

Figure 4 Comparison of 2-step of maximum mass of aqueous and enzymatic extraction for As in selected plant species

The amount of Cd, As, and Pb infused from the plants ranged from 0.29-0.62, 0.13-0.35, and 0.20-0.56 µg/kg, respectively (Figures 2-4). The infused elemental concentrations reported from the plants are similar to those reported for As, Cd, and Pb of not detected to 1.25, not detected to 0.61, and not detected to 5.3 µg/kg, respectively released from traditional medicine and herbal teas [85]. Physiologically based extractions assess the bioaccessibility of an element dissolved during digestion and are available for absorption into the human body. As concentrations in the plant infusions were low, with little transfer from the plants to the infusion. Low As concentrations were reported in the infusions compared to the total elemental concentration, and 29-88% of it was mainly in the inorganic species form [86]. As enters the body through ingestion, inhalation, and dermal absorption, and inorganic As has a half-life of 10 h in humans [87]. Absorbed arsenic undergoes biomethylation in the liver, and approximately 70% is excreted within a few days of ingestion. Most ingested soluble As (III) compounds are absorbed from the gastrointestinal tract [87,88]. After absorption through the lungs or gastrointestinal tract, arsenic is distributed in various body parts via the bloodstream [89,90]. The absorbed As is reduced to arsenite by red blood cells, white blood cells, and other cells in the presence of glutathione [91,92] before methylation occurs in the liver via enzymatic transfer of the methyl group from S-adenosylmethionine (S.A.M.) to methyl arsenate (M.M.A. V), and dimethyl arsenate (D.M.A. V) [89,92]. This ensures that As in the human body is detoxified. Cd concentrations in tea infusions were reported to range from 0.13 to 0.61 μgL^{-1}, with 5-21% being solubilized [85]. Forty-one, 64.46, 18.91, and 81.14% of Pb, As, Hg, and Cd were bio-accessed from C. Sinensis and 8.69% of the total bio-accessed As was in the inorganic form constituted by 560±160 and 290±60 µg/kg of As (III) and As (V), respectively [16], whereas the sum of Cd in the gastrointestinal extractions was 2.00-2.73%. The hazard bioaccessibility quotient target values for Pb, As, Cd, and Hg were 0.0040, 0.5334, 0.0020, and 0.0005, respectively [16], much higher than those reported in the present study (Figures 2-4).

The result of the enzymatic breakdown of the plant as it passes down the human alimentary canal was used to estimate the daily elemental intake. To determine how the intake of Cd, Pb, and As compares with recommended safety values, calculation of the daily intake and total concentrations of the toxic metals are required [11,32]. Total and bioaccessible concentrations were used to evaluate the daily intake of As, Cd, and Pb based on the maximum daily recommended in the product instructions for each complementary medicine, which were then compared with the recommended concentrations [11,32]. The safety guidelines are based on the United States Environmental Protection Agency reference doses and Agency for Toxic Substances and Diseases Registry [11]. The daily intake of toxic elements in the present study was lower than that specified in the standard guidelines. Toxic element absorption in humans depends on their transport through mucosal cells to reach the target organ or the bloodstream [93].Since we did not consider the determinants at the cellular level for toxic element absorption in the daily

calculation, the actual concentrations absorbed may be different. The absorption of toxic elements in the gastrointestinal canal is affected by nutrients such as calcium, phosphates, iron, vitamin D, fats, and fiber [32]. This makes the concentration of the toxic element bioaccessible by an individual depending on his or her diet and nutrition.

If one consumed a 200 mL infusion prepared from 2×10^6 µg of the powdered plant twice a day, as recommended by the herbalist, the daily intake of Pb, As, and Cd was estimated using the mean calculated extraction efficiency (%). The daily intakes of Pb, As, and Cd from the plants investigated were 0.027-0.148, 0.001-0.021, and 0.026-0.078 µg/day, respectively. Our results are similar to those reported for Cd and Pb in Chamomile blossom and Peppermint leaves of 0.07, 0.16, and 0.08, 0.08 µg/day, respectively [12]. The daily intake of lead from young tea leaves was reported to be less than 1.23×10^{-1} µg /kg /day [94], while that of As was approximately 100 folds lower than 2.67×10^{-1} µg/kg /day [94]. These values are far below the tolerable daily intake of Pb (300µg /day) and Cd (1.5 ug/day). Mature tea leaves bioavailed higher Pb, Cd, Hg, and As to the human body than young leaves [94-96]. This means that young plant leaves (for those plants in which the leaves are used for therapeutic purposes) should be used to make infusions to facilitate their safety. The medicinal plants investigated in the present study had low potential to release toxic elements and were safe for nutrient supplementation and therapeutic use. However, the Pb, As, and Cd content of medicinal plants must be monitored frequently to avoid bioaccumulation in consumer bodies and the environment to monitor environmental pollution.

Conclusion

The strategy of total element and physiologically based extraction tests comprehensively determined the total content and bio-accessibility of Cd, As, and Pb to the human body from the commonly used medicinal plants in the two Kenyan regions. Authorities can use the data generated to regulate medicinal plants use to ensure safe consumption and to take appropriate measures to avoid using plants grown in contaminated areas. This approach supports toxicological investigations to provide complementary information on the uptake and metabolism of toxic elements, such as Cd, As, and Pb, from plants. Based on the data collected, it is possible to select the safest plant species among the many available plants in Kenya and other countries for the secure management of diseases. This approach opens a perspective to maximize the benefits from locally available low-cost plant products, thus lowering the healthcare system pressure for countries such as Kenya. However, for the safest age of plants, the organic ligands of Cd, As, and Pb in the plants need to be determined for detailed organometallic compound determination using HPLC-MS/MS. In vivo bioavailability and bioaccessibility studies of Cd, As, and Pb from plants using laboratory animals need to be conducted to understand the mechanism and metabolism of toxic elements in detail.

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Consent to participate

The authors (R.M., K.O., S.O., and E.O.K.) participated in the preparation and edited the manuscript. They consented to the corresponding author to make any necessary adjustments to the manuscript.

Consent to publish

The authors (R.M., K.O., S.O., and E.O.K.) consented to the article's publication.

Authors' contributions

All the authors contributed to the conception and design of the study. Material preparation, data collection, and analysis were performed by R.M., K.O., S.O., and E.O.K.. R.M. wrote the first draft of the manuscript and all authors commented on the previous versions. All authors read and approved the final manuscript."

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Competing interests

The authors have no relevant financial or non-financial interests to disclose.

Data availability statement

The data presented in this paper cannot be used in any form without the authors' permission.

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