



Heavy metal concentration in amaranthus, cowpea, black nightshade and kale in response to wastewater irrigation in Kitui County, Kenya

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ABSTRACT

Low quality water, treated and non-treated wastewater, is used for irrigation and production of leafy vegetables in developing countries. The water contains crucial plant nutrients but also toxic substances including heavy metals such as Cadmium, Copper, Zinc, Lead and Iron. By consuming plants grown with contaminated water containing elevated metal concentrations puts human life at risk. Field experiments were conducted at South Eastern Kenya University to evaluate the heavy metal concentration of amaranthus, cowpea, black nightshade and kale irrigated with wastewater. To test the performance of the leafy vegetables, treatments were laid in a Randomized Complete Block Design (RCBD) with three replications. The vegetables were subjected to two irrigation systems, foliar irrigation (S) and root irrigation (R) with two water treatments, wastewater (WW) and Clean / fresh water (CW) applied to the vegetables. The vegetables, soil and water samples were analyzed for heavy metal concentrations using Atomic Absorption Spectroscopy (AAS). The vegetables were harvested fortnightly upon attaining their horticultural maturity. They differed in heavy metal accumulation with highest concentration of heavy metals found in Brassica oleraceae with a concentration of 3.50 mg/kg and 20.33 mg/kg of Cadmium and Lead respectively while Amaranthus, cowpea and black nightshade had relatively lower concentrations. Some of the vegetables grown with wastewater exceeded the maximum permissible limits of heavy metals in vegetables. This study recommends consistent monitoring of heavy metal concentrations in agricultural soil and water used for irrigation to avoid excessive accumulation in vegetables.

Key words: Cadmium, irrigation, lead, permissible limit, wastewater

RÉSUMÉ

L'eau de faible qualité, usée traitée et non traitée, est utilisée pour l'irrigation et la production de légumes à feuilles dans les pays en développement. L'eau contient des éléments nutritifs essentiels pour les plantes, mais aussi des substances toxiques comme le cadmium, le cuivre, le zinc, le plomb et le fer. La vie humaine est mise en danger en consommant des plantes cultivées avec de l'eau contaminée contenant des concentrations élevées de métaux. Des expériences sur le terrain ont été menées à South Eastern Kenya University pour évaluer

la concentration en métaux lourds d'amarante, de niébé, de noctuelle noire et de chou frisé irrigué avec des eaux usées. Pour tester la performance des légumes à feuilles, des traitements ont été appliqués dans un dispositif de bloc aléatoire complet (BAC) avec trois répétitions. Les légumes ont été soumis à deux systèmes d'irrigation, l'irrigation foliaire (S) et l'irrigation radiculaire (R) avec deux traitements d'eau, les eaux usées (WW) et l'eau propre/douce (CW) appliquées aux légumes. Les échantillons de légumes, de sol et d'eau ont été analysés pour déterminer les concentrations de métaux lourds à l'aide de la spectroscopie d'absorption atomique (SAA). Les légumes étaient récoltés tous les quinze jours après avoir atteint leur maturité horticole. Ils se distinguaient par l'accumulation de métaux lourds avec la concentration la plus élevée de métaux lourds trouvée dans Brassica oleracea avec une concentration de 3,50 mg/kg et de 20,33 mg/kg de Cadmium et de plomb respectivement, tandis que l'Amaranthe, le niébé et la morelle noire présentaient des concentrations relativement plus faibles. Certains légumes cultivés avec des eaux usées dépassaient les limites maximales permises pour les métaux lourds dans les légumes. Cette étude recommande une surveillance cohérente des concentrations de métaux lourds dans le sol agricole et l'eau utilisée pour l'irrigation afin d'éviter une accumulation excessive dans les légumes.

Mots clés : Cadmium, irrigation, plomb, limite admissible, eaux usées

INTRODUCTION

The supply of wastewater to soil dates back to 400 years and is a usual practice around the globe. It is projected that, 20 million hectares of arable land internationally are watered with some sort of wastewater originating from leaching of dumpsites, industrial emissions, sewage sludge (bio solids), heavy traffic using petroleum fuels, paints that are lead based, municipal waters, fertilizers and metal based pesticides (Sardar *et al.*, 2013). The volume of wastewater has been increasing with the increasing population and urbanisation, improved living standards, urbanization and economic development (Khaled and Muhammad, 2015). The continued disposal of wastewater into land and water courses reduces the quality of water available for crop growth. Domestic sewage and refuse find their way into the water from settlements, municipal wastes or institution drainages through leaching, direct discharge and runoff. Human faeces contain high concentrations of toxic elements from the normal dietary

matter presenting principal input of toxic metals to domestic wastewater and sludge of domestic origin (Mohammed and Folorunsho, 2015). Although wastewater is a source of plant nutrients and organic matter, it also contains heavy metals in soluble, chelated and exchangeable forms available to plants (Chiroma *et al.*, 2014; National Research Council, 1996). When applied to crops it becomes a threat to both the soil and plants. Soils have therefore become polluted with heavy metals when such water is used for cultivation of crops. The plants growing in such soil have reduced growth rate, poor performance and low yield (Chibuke and Obiora, 2014). According to Mohammed *et al.* (2014), the use of sewage water for irrigation improves soil fertility and the chemical properties of the soil. This is attributed to the fact that wastewater contains essential elements for plant growth besides the heavy metals.

Contamination by heavy metals is one of

most common determinants of food quality (Mohamed and Khairia, 2012). Anthropogenic activities such as manure and fertilizer application, use of farm chemicals, Lead based fuels or sewage sludge application affects soil heavy metal content. Heavy metal availability in the soil contributes to the concentration of heavy metals in vegetables. Accumulation of heavy metals in vegetables follows two major routes: foliar and root uptake. Unlike root uptake, little information is available on the potentiality of foliar plant parts to take up heavy metals, water and plant nutrients. Foliar plant parts act as a pathway for contamination when extra-fine particles come into contact with the leaves and enter the plant system via the lenticels, ectodesmata, aqueous pores, and stomata enhancing heavy metal concentration (Muhammad *et al.*, 2017). Some of the heavy metals such as lead, chromium, copper, nickel and cadmium are cumulative poisons (Nazemi, 2012), and often cannot be removed or separated from waste water even after treatment. Heavy metal accumulation is a function of the amount of pollutants and depends on the physiochemical properties of the soil, concentration in waste water, efficiency of the crop to remove the metals from soil, the climatic factors and time of exposure (Shah, 2010; Sardar *et al.*, 2013). In vegetables, contamination with heavy metals is a great health concern (Hassan and Mohammad, 2013). According to Sajjad *et al.* (2009), the most toxic and abundant heavy metals in plants are lead and cadmium.

Vegetables constitute important diet components such as vitamins, proteins, carbohydrates, iron and calcium, among other nutrients (Wubishet *et al.*, 2017). The use of African Indigenous Leafy Vegetables (AILVs) was part of cultural heritage on customs and traditions but recently interest on them has grown significantly. They are often produced in backyard gardens, and possess several advantages and potentials that have not yet been exploited. The AIVs have a short

growth period and are better adapted to harsh climatic conditions and diseases than exotic vegetables (Abukutsa-Onyango, 2010). They are an important commodity to many African Communities (Kimaru *et al.*, 2014). Indigenous vegetables play a great role in food and nutritional security especially in the dry areas of Kenya, therefore the country seeks to improve its production in order to improve the nutrition and health of its population because they are more nutritive than Kale and cabbage. Exotic vegetables mainly kale and cabbage account for 90 percent of the vegetables consumed in Kenya compared to 3.7 percent of indigenous vegetables at any given time (Otieno, 2013). Amaranthus (*Amaranthus hybridus*), cowpea (*Vigna unguiculata*) and black nightshade (*Solanum nigrum*) are African indigenous vegetables exhibiting better growth at higher temperatures. Amaranthus and black nightshade occur as annual and casual weeds on cultivated land and in waste places. After the stomata close down to conserve moisture during hot and dry conditions, they continue photosynthesizing while the cool weather leaf crops such as kale have their production severely limited during hot weather (IPCC, 2001).

Wastewater is continuously used in irrigating the agricultural fields in developing countries (Madhvi *et al.*, 2014). For that reason, most of the human activities that use water generally produce wastewater. As demand for the water increases, the pollution load and the quantity of the wastewater continuously increase. Eighty percent of the total world's wastewater and 95% of the wastewater in some less developed countries is released to the environment without being treated (WWDR, 2017). Continuous use of this untreated wastewater for irrigation to provide macro and micro nutrient requirements of the plants affects soil and plant quality as higher quantities become harmful to the plants (Madhvi *et al.*, 2014).

In African and Asian towns, studies have shown that 50 percent of vegetable production for urban areas relies on agriculture with wastewater with the farmers not worried about the environmental dangers associated with this water. They are only concerned with maximizing production and their profits (Sardar *et al.*, 2013). The citizens dwelling in towns have to deal with poor diets and limited incomes; thus have resorted to farming in backyards, open public spaces such as industrial areas, under power lines and along river banks (Carin *et al.*, 2012). There is need for new and alternative approaches to ensuring food and nutrition security. Such solutions should be sustainable, resilient and practical. This need has led to renewed focus on identifying and improving underutilized indigenous and traditional crops. Wastewater use provides an alternative to improving the productivity of indigenous vegetables (Mabhaudhi, 2009).

Soil pollution with heavy metals due to use of untreated wastewater is a threat to the ecological integrity (Mahmood and Malik, 2014). Naturally, varying levels of heavy metals do exist in domestic or municipal sewage waters that depend on lifestyle of the population and mix-up of industrial waste effluents with the domestic or municipal sewage waters. Aziz (2006) reported that the uptake of heavy metals by vegetables, and particularly leafy vegetables, is more than for cereals. Due to the high transpiration rate, leafy vegetables have the potential for accumulating heavy metals compared to fruit crops and grains (Mohammed and Folorunsho, 2015). Therefore, it is understandable that certain amounts of heavy metals accumulate in edible parts of plants grown on sewage-irrigated fields which affects both the underground and above ground plant parts (Shah, 2010). This facilitates easy transfer of heavy metals from vegetables to consumers leading to health risks (Nabulo, 2009).

One of the Millennium Development Goals

(MDGs), which is to eradicate extreme poverty and hunger is being hindered by water scarcity. Like in many other countries, Kenya is also below the international water scarcity threshold (1 000 m³ per person per year) with only 935 m³ available per person per year. Unfortunately, population growth is forecast to reduce this figure to 359 m³ by 2020 (WWDR2, 2006).

The climate of Kitui County is classified as semi-arid and characterized by frequent drought occurrences. Most areas are generally hot and dry having high rate of evapotranspiration in many seasons. Currently the water harvesting measures have had a limited impact. Water is the primary medium through which impacts of climate change and variability can be experienced resulting in negative effects on food production. The decreasing rainfall in areas that are already water-short is likely to impact on cultivation of indigenous or traditional crops. The IPCC 4th African Assessment Report estimates that by 2020, 75 to 250 million people are likely to be exposed to water stress and that rain fed agricultural production could reduce by up to 50% (Nkomo *et al.*, 2006). Under these conditions, food, nutritional and income insecurity, which are already a challenge, may be intensified (Schulze, 2011).

Vegetables form an essential part of the human diet although there are limited studies on their irrigation using wastewater. The use of wastewater in greenhouse production is also lacking (Richard, 2007).

In the arid and semi-arid areas characterized by high temperatures, water use is fairly low at approximately 90 litres/day per person and the concentration of elements in the wastewater tend to be high (FAO, 2002). Anthropogenic activities such as use of sewage sludge, fertilizers, pesticide use in agriculture, waste disposal and burning of fossil fuels have resulted in water pollution (Chibuike and Obiora, 2014).

This study examines the effect of wastewater utilization in production of *Amaranthus* (*Amaranthus hybridus*), cowpea (*Vigna unguiculata*), black nightshade (*Solanum nigrum*) and Kale (*Brassica oleraceae* var. *acephala*) by assessing the concentration levels of lead and cadmium in the vegetables. These vegetables are among those grown with wastewater within Kitui County of Kenya.

MATERIALS AND METHODS

Four types of vegetables were grown following standard practices at South Eastern Kenya University in two independent field experiments, i.e., September to November 2017 and January to March 2018. Two water treatments, wastewater (WW) and clean /fresh water (CW) were applied to four vegetables: (*Amaranthus hybridus*, *Brassica oleraceae* var. *acephala*, *Solanum nigrum* and *Vigna unguiculata* named A, Kale, N and C in the layout, respectively). The vegetables were chosen due to their popularity in production within the County and similar growth rates, based on the field survey analysis. The eight treatments (2 water treatments $\times$ 4 vegetable types) were subjected to two irrigation systems: Shoot/foliar irrigation (S) and root irrigation (R), resulting into sixteen treatments (Kale WW Root, Kale WW Shoot, Kale CW Shoot, Kale CW Root, Amaranthus CW Shoot, Amaranthus CW Root, Amaranthus WW Shoot, Amaranthus WW Root, Cowpeas CW Shoot, Cowpeas WW Root, Cowpeas CW Root, Cowpeas WW shoot, Black nightshade CW Shoot, Black nightshade CW Root, Black nightshade WW Shoot, Black nightshade WW Root). To test the performance of the leafy vegetables, treatments were laid in a Randomized Complete Block Design (RCBD) on 5 m² experimental plots with three replications.

Collection and preparation of soil samples for analytical procedures. Soil samples (0-15 cm depth) were collected from vegetable fields

where wastewater is used using a 2.5 cm hand auger and homogenized. Each field sample consisted of three sub-samples collected from the edges and middle of the farm. These samples were dried at 95°C in the oven and ground into a fine powder using mortar and pestle. Fifteen millilitres of 1 M HNO₃ were added 15 g of the finely ground soil in a conical flask. This was followed by addition of 30 ml into the mixture then the solution was kept for 24 hours. The solution was centrifuged and filtered using Whatman's filter paper (size 41).

Collection and preparation of water samples for analytical procedures. Three representative samples were collected into sealed containers. A 2-point pH calibration was done with pH 4/pH 7 and pH7/pH10 buffer solutions to accommodate both acidic and basic water pH measurements. The electrode and the probe were rinsed with DI water and blot dried. The samples were thoroughly stirred before placing the ATC probe in water to allow the measurements to stabilize. The physico-chemical parameters that influence plant growth and potentially toxic elements such as cadmium, copper, lead and zinc which might be at phytotoxic levels were identified. Water samples were filtered to remove suspended particulate matter and suspended solids by use of solid-Phase extraction (SPE) method. Nitrogen (as N), Phosphorus (as P), potassium (as K), pH (as pH –H₂O 1:2.5 v/v) and Ec (mS/cm) were the quantified (Steven *et al.*, 2007).

Determination of heavy metal concentration in vegetable and soil samples. Atomic Absorption Spectroscopy [AAS Shimadzu Model: 6200] was used for quantifying heavy metals. Elements Specific wavelengths were provided by passing a lamp light beam through a flame whose cathode is made from the element. A photon multiplier detected the amount of reduction in light intensity due to analyte absorption relating to the amount of element in the sample. Determining heavy metals

concentration was based on soil and vegetable dry weight and expressed in milligrams per kilogram of soil (mg kg^{-1}).

RESULTS AND DISCUSSION

Lead and cadmium are the most lethal heavy metal contaminants in human diet and health and among the smallest fine particles ($<1\mu\text{m}$). Different crops exhibit different heavy metal accumulation abilities in various morphological organs and therefore, the timing of production, the locality and cultivar determine the transportation and absorption of lead and cadmium (Sekara *et al.*, 2005). The Standard Regulatory Bodies such as FAO, WHO and Ewers U, however, have established the maximum permissible limits of heavy metal concentration in soils, irrigation water and vegetables for cadmium and lead. For agricultural soil, the ideal concentration should be $3\mu\text{g/g}$ and $100\mu\text{g/g}$, irrigation water should measure $0.013\mu\text{g/ml}$ and $0.0653\mu\text{g/g}$ while vegetables should have $0.13\mu\text{g/g}$ and $0.33\mu\text{g/g}$ for cadmium and lead respectively (Chiroma *et al.*, 2014). These limits were exceeded in this

study where wastewater had 1.91 ppm and 6.05 ppm and the soil had 3.543 mg/kg and 76.26 mg/kg of cadmium and lead, respectively. The chemical composition of soil plays an important role in the overall growth and development of vegetables. Heavy metal availability in the soil contributes to the concentration of heavy metals in vegetables (Nazemi, 2012). The heavy metal concentrations of the vegetables followed the same pattern in both the first and second season.

Heavy metal accumulation on leafy and edible portion of vegetables depends on morphology, surface area, cuticle characteristics, stomatal density, texture of the leaves and physico-chemical properties of the particular heavy metal and its concentration in irrigation source. However, barely no documented data exists on foliar uptake and metal speciation in plants. Different vegetable species vary in heavy metal uptake while even when growing in similar environmental conditions (Muhammad *et al.*, 2017). For instance, vegetables of the

Table 1. Mean Cadmium concentration in the edible leafy portion and interaction effect between vegetable type (V), water quality (W) and irrigation type (Foliar and Root)

Variety	Cadmium Concentration (mg/kg)			
	Week 6	Week 8	Week 10	Week 12
Cowpeas	0.26b	1.18b	2.20a	2.18b
Black nightshade	0.27b	0.85b	2.15a	2.23b
Kale	0.34b	2.15a	2.73a	3.50a
Amaranthus	0.60a	1.30b	2.03a	2.68ab
LSD (0.05)	0.11	0.39	1.03	0.71
Water Quality				
Clean Water	0.17b	1.5a	2.17a	2.32b
Waste Water	0.56a	1.24a	2.39a	2.98a
LSD (0.05)	0.14	0.52	0.25	0.41
Irrigation Type				
Foliar	0.18b	1.29a	2.12a	2.44b
Root	0.56a	1.45a	2.44a	2.85a
LSD (0.05)	0.12	0.26	0.36	0.33
V*W	*	*	*	*
V*I	*	*	*	*

of the Brassicaceae family are highly preferred among farmers because of quick leaf regeneration (Bere, 2014). *Brassica oleraceae* var. *acephala* (kale) is a locally grown vegetable and one of the hyper accumulators of heavy metal ions. In a similar study, it has been observed that the metal pollution index of spinach was found to be maximum when compared with other vegetables irrigated with wastewater (Ashita *et al.*, 2016). As reported by Muhammad *et al.* (2017), Pb has particularly been found to accumulate in vegetables of Brassicaceae family (Brassicaoleraceae and Spinaciaoleraceae) at Toulouse – France which is in line with the findings of this study where kale attained the highest Pb at concentration of 20.33mg/kg at the 10th week compared to the other vegetables. This concentration was above the maximum permissible level by WHO.

The increase in lead concentration attributed

to wastewater irrigation in this study is in agreement with that reported by Mohamed and Khairia (2012), at 17.54 to 25 mg/kg. In a study carried out on heavy metal accumulation of vegetables irrigated with wastewater, Khaled and Muhammad (2015) observed that the concentrations of heavy metals in vegetables irrigated with wastewater were higher than those grown in control soil as is evident in this study with clean water irrigation, 0.17mg/kg to 2.32mg/kg and 10.22mg/kg to 17.35mg/kg in comparison with 0.56 to 2.98 and 11.53 to 17.99 for cadmium and lead, respectively. In agreement with results obtained in this study, use of wastewater for irrigation increased the cadmium and lead levels of the four vegetable types.

The vegetable types started showing significant differences ($p < 0.05$) in cadmium concentrations by the sixth week after planting. The variation

Table 2: Mean Lead concentration of the edible leafy portion and interaction effect between vegetable type (V), water quality (W) and irrigation type (Foliar and Root)

Variety	Lead Concentration (mg/kg)			
	Week 6	Week 8	Week 10	Week 12
Cowpeas	9.71a	16.02a	16.05b	16.38a
Amaranthus	10.05a	8.85b	12.20c	16.15a
Black nightshade	11.37a	12.95ab	14.06bc	19.15a
Kale	12.37a	14.22a	20.33a	17.83a
LSD (0.05)	2.85	3.43	2.63	6.89
Water Quality				
Clean water	10.22a	10.93b	13.32b	17.35a
Waste water	11.53a	15.09a	17.99a	17.41a
LSD (0.05)	1.78	2.07	2.56	1.79
Irrigation Type				
Foliar	10.79a	11.08b	12.95b	15.93b
Root	10.97a	14.94a	18.37a	18.82a
LSD (0.05)	2.03	2.16	1.52	1.58
V*W	NS	*	*	NS
V*I	NS	*	*	*

*Values with the same letters in a column are not significantly different at ($p < 0.05$) level of significance

in heavy metal uptake with respect to different vegetables occurred due to differences in morphology, physiological parameters and variation in translocation mechanism of different vegetable species. Similar studies by Ashita *et al.* (2016), along a wastewater drain revealed that there were no significant differences between metals in the study site but concentration of heavy metals in the vegetables irrigated with wastewater was significantly different ($p < 0.01$). Compared to lead, cadmium is mobile and easily transported from the roots to shoots. Greater shoot biomass production can result into lower heavy metal concentration in the shoot.

Foliar heavy metal uptake and root uptake are of similar importance to plant organs such as leaves (Muhammad *et al.*, 2017). Significant differences were observed in cadmium and lead metal uptake between the sixth and tenth week for both foliar and root irrigation. Unlike root irrigation, foliar irrigation hydrates the leaf enhancing cuticle expansion and stomatal opening to favour penetration of the heavy metals present in wastewater. The stage of plant growth and development also determined the demand of certain nutrients thus significant differences in cadmium concentration were observed in week six while significant interaction differences were observed in lead concentration by week eight because different metals vary in their capacity to go through leaves or roots.

CONCLUSIONS

Irrigation of agricultural land with wastewater results in the accumulation of heavy metals in vegetables. Variation in heavy metal content between the African indigenous leafy vegetables and kale show the different uptake capabilities while growing in the same environmental conditions. This depends on the prevailing soil composition, vegetable type and the type of heavy metal. Concentration of heavy metals in some of the vegetables obtained from the field experiment was found to be above the maximum

permissible limits set by either WHO, FAO, SEPA, Indian Safe limit or WAV. Monitoring the levels of heavy metals in vegetables can promote food safety.

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STATEMENT OF NO-CONFLICT OF INTEREST

The authors declare that there is no conflict of interest in this paper.

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