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Determination of Some Heavy Metals in Selected Edible Vegetables Grown Along River Owade in Oyugis Area, Homa Bay County, Kenya

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Abstract
Harmful materials such as heavy metals, which may be itoxic are sometimes found in vegetables. The levels of some of these heavy metals were investigated in selected vegetables, *kale* and *cabbage*, which are grown in Oyugis along Owade River in Homa Bay County, Kenya. All samples were randomly collected from two gardens along the river. The levels of the selected heavy metals (Copper, Iron, Cadmium, Chromium, and Zinc) were analysed using an Atomic Absorption Spectrophotometer. In the samples analysed, Cd and Cr were not detected. In kale, iron was found to be the highest in farm A, with an average of 46.68, and copper in farm B (32.73). Zinc was the lowest in both farms, with an average of 12.00 and 7.70, respectively. In cabbage, iron was highest in farm A (43.00) while copper was highest in farm B (37.00). Lowest in both farms was zinc (10.53 and 11.50, respectively). The data were analysed with t-test and analysis of variance (ANOVA). There were significant differences ($p < 0.05$) between the levels of the heavy metals in the vegetables obtained from Farms A and B. The concentration levels of metals in vegetables in the two gardens could be attributed to excessive usage of fertilisers and other agro-chemicals. The results were, however, lower than the values considered toxic, except for Fe, with higher values, which may be due to soil composition. From the results of this study, the consumption of these vegetables as food may not pose health hazards to humans.

Introduction

A vegetable is a plant or part of a plant used as food. There are different types of vegetables, namely leafy vegetables, stem vegetables, root vegetables, and fruit vegetables, among others. (Galadima et al., 2015). Eating vegetables regularly in the diet has many health benefits (Martha et al., 2018). Vegetables contain large quantities of minerals, vitamins, carbohydrates, essential amino acids and dietary fibres, and are therefore referred to as "Protective food" (Okunlola & Adunola, 2017; Okunlola, 2019).

Among the benefits is the reduction of many health-related diseases, and it is used to convert fats and carbohydrates into energy (Hanna et al., 2021). Eating vegetables is one of the most essential ways for the human body to absorb minerals, which are necessary for its healthy development. Unfortunately, harmful materials such as heavy metals, which may lead to intoxication, are sometimes found in these vegetables (Gergen & Harmanescu, 2012; Govind et al., 2022). As human activities intensify, particularly with the widespread application of modern technology, pollution and contamination of the human food chain have increased (Ling & Wahab, 2020).

Heavy metal concentrations in soil are influenced by anthropogenic activities, including agricultural practices, industrial activities, and waste disposal methods (João et al., 2019). The long-term use of metal-containing fertilisers and pesticides leads to an increase in the formation of toxic metal complexes in agricultural soil, which then enters crops and plants in general.

Heavy metal contamination occurs when their concentrations exceed the recommended limits in soil, water, or plant resources, resulting in negative effects on humans or animals (Manyara et al., 2024). Contamination and subsequent pollution of the environment by heavy metals have become a global concern due to their effects on the ecosystem (Saikat et al., 2022). Due to their cumulative behaviour and toxicity, heavy metals have a potentially hazardous effect on both plants and human health (Ata et al., 2021; Saikat et al., 2022; Anjan et al., 2024). The distribution of heavy metals in plants depends upon their availability and concentration as well as particular plant species (Misbah et al., 2023). Studies have shown that some common vegetables are capable of accumulating high levels of metals from the soil (Maurya et al., 2018; Okunlola, 2019; Misbah et al., 2023).

Neurological disorders, central nervous system destruction and cancers of various body organs are some of the heavy metal poisoning (Dimple et al., 2020; Klaudia et al., 2024). Low birth weight and severe mental retardation of newly born children have been reported in some cases where pregnant women ingest toxic heavy metals (Misbah et al., 2023).

Heavy metals like Fe, Cu, Zn, and Ni are essential for the proper functioning of biological systems, and their deficiency or excess could lead to a number of disorders (Pratush et al., 2018; Klaudia et al., 2024).

Industrial and agricultural wastes result in an increased concentration of heavy metals, which pollute both soil and plants. It is therefore necessary to examine the state of the polluted soil and plant and establish the influence of heavy metals on both. Heavy metals have significant importance due to their toxicity and accumulative behaviour, and are not biodegradable (Sarubbo et al., 2015; Rahman, 2020). Surface soil may act as a carrier and a potential source of pollution, as the mobility of these metals allows them to remain in the upper layers of the soil.

The use of polluted water in the immediate surroundings for growing vegetables is a common practice in towns and cities. Although this water is considered a rich source of organic matter and plant nutrients, it also contains some soluble salts and metals like Fe, Mn, Cu, Zn, Pb, Ni, Sn, Hg, and Cr. When such water is used for crop irrigation over a long period, these heavy metals may accumulate in the soil and become toxic to the plants (Abdur et al., 2023).

Wastewater is commonly used in agricultural irrigation. The long-term use of wastewater on agricultural lands often results in the accumulation of high levels of heavy metals in soils (Abdur et al., 2023). Crops grown on such soils accumulate these metals in excessive quantities that are sufficient to cause health problems in both animals and humans.

The study aims to investigate the levels of selected heavy metals (Cu, Cd, Fe, Cr, and Zn) in the leaves of kale and cabbage in the Oyugis area to determine the suitability of these vegetables for human consumption.

Materials and Methods

Analytical reagent (AnalaR) grade chemicals and distilled water were used throughout the study. All glassware and plastic containers used in this work were washed with a detergent solution, followed by 20% (v/v) nitric acid, and then rinsed with tap water and finally with distilled water.

Study Area

Oyugis region geographically is located in the Western part of Kenya, Homa Bay County. Its geographical coordinates are 0° 30' South and 34° 43' East of the Greenwich meridian.

Sampling and sample treatment

The samples analysed include *kale and cabbage* leaves. Samples were collected from March to May 2024 from two different gardens (A and B) located along the River Owade in the Oyugis area. Leaves of the fresh samples of *kale and cabbage* were randomly collected (handpicked) from two different vegetable gardens (A and B). The samples were wrapped in big brown envelopes and labelled. A total of ten samples, each of *kale and cabbage, were collected* from each of the vegetable farms along River Owade in Oyugis. Samples from each of the two gardens were combined to obtain two homogeneous samples.

In the laboratory, the vegetable samples were washed first with tap water and then with distilled water. The water was allowed to drip out and then sliced into smaller portions, which were dried in an oven at 80°C for 24 hours. At the end of the drying process, the oven was turned off and left overnight to allow the sample to cool to room temperature. Each sample was ground into a fine powder, sieved and finally stored in a 250 cm^3 screw capped plastic jar appropriately labelled.

Digestion procedure

1.0 g of each powdered leaf was weighed out into a Kjeldahl digestion flask, mixed with 10 cm^3 of concentrated sulphuric acid, concentrated perchloric acid and concentrated nitric acid in the ratio 1:2:20 by volume, respectively and left to stand overnight. The flask was then heated to 70°C for 40 minutes, and subsequently the temperature was increased to 120°C. The digestion was completed when the solution became clear and white fumes appeared. The solution was diluted with 20 cm^3 of distilled water and boiled for 15 min. This was then allowed to cool, transferred into 100 cm^3 volumetric flasks and diluted to the mark with distilled water. The sample solution was then filtered through a filter paper into a screw capped polyethene bottle.

Determination of heavy metals

Levels of Cd, Cu, Fe, Zn and Cr in the vegetable samples were determined using a Buck 210 model Atomic absorption spectrophotometer (AAS). For the AAS, a hollow cathode lamp with discharge lamps containing the respective metals was used, and a flame atomiser was used as the atomiser. The detector used was a photomultiplier tube (PMT). Working standards were prepared by further dilution of a 1000 ppm stock solution of each of the metals. The mean values of six determinations per sample were recorded.

Statistical analysis

All analysis was performed in triplicate. Results were expressed by means of ± SD. Statistical significance was established using one-way analysis of variance (ANOVA). Means were separated according to Duncan's multiple range analysis ($p < 0.05$) using software SPSS 16.0.

Limitations

Several limitations should be acknowledged in this study. The study did not include a parallel analysis of soil and water, which limited the ability to trace the source of contamination. The vegetable samples were collected from a limited number of locations, which may not represent broader regional contamination levels. Common chemical forms of these metals, which they take when absorbed by plants, were also not analysed. Examples of these chemical forms are: trivalent and hexavalent chromium (Cr^{3+} and Cr^{6+}); Fe^{2+} (ferrous) and Fe^{3+} (ferric); $Cu^{2+}$and Cu^{+}, among others.

Results and Discussion

The levels of heavy metals (Cu, Cd, Cr, Fe and Zn) in *kale and cabbage* are as shown in Tables 1 and 2 of Farm A and B.

Table 1: Concentrations of some heavy metals in kale and cabbage from Farm A (mg/kg)

Vegetables/Sampling sites	Cd	Cr	Cu	Fe	Zn
kale					
Location 1	ND	ND	30.00 ± 0.18	46.68 ± 0.1	12.00 ± 0.01
Location 2	ND	ND	29.10 ± 14.00	35.35 ± 0.02	24.50± 0.48
cabbage					
Location 1	ND	ND	33.33 ± 0.41	43.00 ± 0.03	10.53 ± 0.16
Location 2	ND	ND	33.50 ± 0.06	31.52 ± 0.70	21.07 ± 0.12
WHO	0.3	0.3	73	18	100

All values represent mean ± standard deviation of triplicate determination. **ND =** Not detected**, WHO =** World Health Organisation.

Table 2: Concentration of some heavy metals in kale and cabbage from Farm B (mg/kg)

Vegetables/Sampling sites	Cd	Cr	Cu	Fe	Zn
kale					
Location 1	ND	ND	30.30 ± 0.23	27.47 ± 0.08	8.30 ± 0.03
Location 2	ND	ND	32.73 ± 0.04	24.06 ± 0.23	7.70 ± 0.02
cabbage					
Location 1	ND	ND	37.00 ± 0.11	29.40 ± 0.10	24.00 ± 0.45
Location 2	ND	ND	34.03 ± 0.40	28.55 ± 0.11	11.50 ± 0.10
WHO	0.3	0.3	73	18	100

All values represent mean ± standard deviation of triplicate determination. **ND** = Not detected**, WHO** = World Health Organisation.

In kale obtained from Farm A, the metal levels from locations 1 and 2, respectively, were: Cu, 30.00 ± 0.18 mg/kg and 30.30 ± 0.23; Fe, 46.68 ± 0.1 and 35.35 ± 0.02 mg/kg; and Zn, 12.00 ± 0.01 and 24.50± 0.48 mg/kg. In kale obtained from Farm B, the metal levels from locations 1 and 2, respectively, were: Cu, 30.30 ± 0.23 and 32.73 ± 0.04 mg/kg; Fe, 27.47 ± 0.08 and 24.06 ± 0.23 mg/kg; and Zn, 8.30 ± 0.03 and 7.70 ± 0.02 mg/kg. In cabbage obtained from Farm A, the metal levels from locations 1 and 2, respectively, were: Cu, 33.33 ± 0.41 and 33.50 ± 0.06 mg/kg; Fe, 43.00 ± 0.03 and 31.52 ± 0.70 mg/kg; and Zn, 10.53 ± 0.16 and 21.07 ± 0.12 mg/kg. In cabbage obtained from Farm B, the metal levels at locations 1 and 2, respectively, were: Cu, 37.00 ± 0.11 and 34.03 ± 0.40 mg/kg; Fe, 29.40 ± 0.10 and 28.55 ± 0.11 mg/kg; and Zn, 24.00 ± 0.45 and 11.50 ± 0.10 mg/kg.

In both Farms A and B, chromium and cadmium were not detected. This could be attributed to the fact that Cd and Cr tend to bind tightly to soil particles, which makes them less mobile and hence less absorbed by plants. A study by Govind et al. (2022) also found a similar trend where Cd in samples were found to be below detectable levels. Mutune et al. (2014), while studying heavy metal concentration of chosen leafy vegetables of Africa planted in peri-urban as well as urban Nairobi city in Kenya, reported that all the vegetables' metal content, except Cu, were in some sites above the acceptable limits. Guerra et al., (2012) reported that Cd levels were from 0.01 to 0.18 mg/kg, Cr concentrations ranged from 0.01 to 0.60 mg/kg while Pb levels vary from 0.02 to 2.50 mg/kg. Similar

studies were done by Shirkhanloo et al. (2015). In their study, they investigated the levels of Co, V, Hg, As, and Ni in leafy edible vegetables. They found that the levels of V, Ni, and Co in the investigated vegetables exceeded the acceptable limit for heavy metals set by WHO guidelines.
The analysis revealed that kale contained a lower concentration of copper than cabbage, although the maximum values recorded in both the vegetables are within the WHO/FAO maximum tolerable Cu concentration of 73 mg/kg in fresh vegetables (WHO, 2016). On the other hand, the results were also lower than the recommended threshold values for vegetables, except Fe, with a higher level. Cabbage forms a dense head with tightly packed leaves, which may retain more micronutrients or metals due to reduced transpiration loss. Kale has more open, leafy growth, which may lead to lower accumulation of certain metals. This agrees with Anjan et al. (2024), who indicated that cabbage is a good accumulator of metals.

The recommended values are: Fe, 18 mg/kg; Cu, 73 mg/kg; and Zn, 100 mg/kg. The order of the metals contamination in the vegetables was Fe > Cu > Zn in Farm A, and Cu > Fe > Zn in Farm B. This indicates that farm A has more iron in the vegetables than farm B. Farm A had more organic matter, which enhances micronutrient retention, potentially improving iron absorption. Since iron uptake is better in slightly acidic soil, there is a possibility that farm A could have more acidic soil than farm B. Statistical test of significance using the Student t-test and ANOVA showed significant differences ($p < 0.05$) between the levels of the heavy metals in vegetables obtained from the sample sites in Farm A and those from Farm B, except for Cu, which showed no significant differences ($p > 0.05$). The elevated level of Fe in vegetables in the two gardens could be attributed to excessive usage of fertilisers and other agrochemicals, and other environmental factors in the area. High levels of Fe in vegetables can also be a result of soil conditions, farming practices, and plant characteristics. Iron is more available in acidic soils. Organic matter in the form of compost and humus improves iron retention and availability.

Similarly, the concentration levels of the metals in the vegetables obtained from gardens A and B could be due to possible pollution resulting from the extensive agricultural activities in the area, as well as downstream deposition of fertilisers and other agrochemicals as the River flows into the area. The consumption of these vegetables as food may pose potential health hazards to humans due to their high iron content. While iron is an essential nutrient, it can pose health risks when consumed in excess over time. Some of the potential risks are liver damage (cirrhosis), joint pain and fatigue, among others

Conclusion

Considering the health risks encountered in diets as a result of high levels of heavy metals in vegetables, the levels of these metals in vegetables should not exceed the maximum allowable levels. Farmers should be educated on the problems associated with excessive usage of fertilisers and other chemicals, as well as irrigating the crops with waste and polluted water. The vegetables contained variable levels of heavy metals (Cd, Cr, Cu, Fe, and Zn). Except for Fe, the metal levels were within the allowable values considered safe for the consumption of vegetables. Agricultural practices, such as the over-application of fertilisers and the use of wastewater, can affect accumulations of heavy metals.

Although the concentration of most heavy metals in vegetables was within the allowable levels, remediation for contaminated soil using plants to absorb metals or adding amendments that bind heavy metals is recommended. Using raised beds with clean soil also helps in isolating crops from contaminated ground. Irrigation using clean water is essential, and thoroughly washing the vegetables before cooking.

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