Analysis of Some Toxic Heavy Metal Levels in Selected Edible Leafy Vegetables in Sheka Zone, Southwest Ethiopia

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Abstract: There is a risk to human health associated with dietary exposure to the majority of hazardous heavy metals, including As, Cd, Cr, Pb, and Hg. This study looked at the levels of the five most dangerous heavy metals in four types of edible leafy vegetables that are popular in Sheka Zone, southwest Ethiopia. The heavy metals found in a few leafy vegetables, including Brassica carinata, Lactuca sativa var. capital, Beta vulgaris, and Cucurbita Leaf, which were taken from four different districts of the Sheka zone, were determined using the atomic absorption spectroscopy (AAS) technique. The selection of green vegetables was made in consideration of local human nutrition. Compared to other vegetables, those cultivated in Mizan had the highest concentration of heavy metals. Since all obtained values fell below the permissible limits set by the FAO/WHO for edible leafy vegetables, the results demonstrate that consuming these leafy vegetables as part of a healthy diet will not cause heavy metal toxicity and may even be advantageous to users in cases of micronutrient deficiency.

Keywords: AAS, Leaf Vegetable, Heavy Metal, Permissible Limit

1. Introduction

Vegetables are key diet components because they include elements that are vital for preserving health and avoiding disease, including iron, calcium, vitamins, carbs, and proteins. Vegetables had not previously made up a significant portion of the Ethiopian diet. There is also an annual cycle of food scarcity in some parts of Ethiopia, which results in families running out of grain before the subsequent harvest. As a result, they significantly add green vegetables to their diets [1, 2]. However, their consumption has been gradually rising recently, especially in metropolitan areas. This is because people are becoming more aware of the nutritional significance of veggies. Vegetables now play a significant part in Ethiopian agriculture by supplying nourishing nourishment. In Ethiopia, considerable amounts of vegetables are consumed as a staple diet by the middle- and low-income classes. They are employed as supplies of vitamins, minerals, salts, water, calcium, iron, and potash because of their high nutritional value [3]. They also fall under the category of essential protective foods and are excellent for preserving

health as well as the prevention and treatment of many ailments [4]. A lower risk of cancer, cardiovascular disease, stroke, Alzheimer's disease, cataracts, and other aging-related functional deficits is associated with a diet high in vegetables [5, 6]. In order to improve their health, people are advised to eat more veggies, which are a fantastic source of fiber, vitamins, and minerals.

Although vegetables are quite healthful, due to a variety of transit techniques, they also contain both essential and non-essential (toxic) heavy metals in varying concentrations. It is quite concerning when these harmful heavy metals build up in people's bodies over time since it can lead to serious health problems [6]. Heavy metals can accumulate in the body through biological chains, are tenacious and non-biodegradable, and have extended biological half-lives. When eaten over an extended period, they can be extremely hazardous even at low concentrations [7, 8]. In order to protect public health, it is crucial to ascertain the amounts of heavy metals in eatable leafy vegetables and to declare any potential contamination.

In several nations, the contributions of vegetables to a daily intake of heavy metals from eatable green vegetables

have been examined, but no research has been done in the Shaka zone of southwest Ethiopia. However, it is crucial for health that local researchers pay attention to evaluating the concentrations of heavy metals in edible green vegetables, which are often eaten in the Sheka Zone. In light of this, this study project was created to determine the levels of several hazardous heavy metals in a variety of edible leafy vegetables, compare the findings to the WHO standard, and offer a scientific data baseline for regular consumers.

2. Method and Materials

2.1. Chemical and Instrument

In the investigation, standard solutions (1000 mg L^{-1}) of Cr, As, Hg, Pb, and Cd (Merck, Germany), as well as HNO_3, HCl, and H_2O_2, were used. The instrument was a flame atomic absorption spectrophotometer (210VGP, Buck Scientific, USA).

2.2. Collection of Plant Material

Fresh leafy vegetables of four different varieties, including Brassica cainata, Lactuca sativa, Cucurbita leaf, and Beta vulgaris sub. Vulgaris were gathered in the Shaka zone of South Western Ethiopia at markets in Mizan, Fide, Tepi, and Meti. Utilizing the common morphological distinctive traits, the collected vegetables were botanically recognized, authenticated, and prepared for analysis.

2.3. Sample Preparation and Treatment

Before being washed with double-distilled water, the samples were thoroughly cleaned with tap water to get rid of any dust, debris, and parasites. In order to ensure that the pieces dried at the same rate, the samples were subsequently chopped and baked for 24 hours at 90°C. The dried samples were then crushed in a crucible into a fine powder. For identification purposes, each sample was given a label, and it was kept in a bottle that was tightly closed for testing.

2.4. Sample Digestion

Wet digestion has been used to break down leafy plants using various mineral acid combinations. Each sample of powdered vegetables was measured out at 0.5g and put into a conical flask with a volume of 100 mL. This flask was filled with a freshly made 10ml HNO_3, HCl, and H_2O_2 combination in the proportions 4:3:2. To blend the acids with the sample of vegetables, the flask was slightly shaken. The flask was then put on the hot plate, and the temperature was raised gradually, starting at 30°C and reaching 113°C. Bright red color first appeared after 4 minutes, becoming deep yellow after 7 minutes. It took the mixture three minutes to cool. After heating for 5 minutes, 2 mL of HCl was added very little. A bright yellow tint then emerged. The solution was heated until it lost its color. After 18 minutes of digestion, the mixture was allowed to cool to room temperature for around 10 minutes. Then, 10 ml of deionized water was added to the

100 ml volumetric flask. Deionized water was used to rinse the conical flask until the 100mL capacity was attained. Following the same digestive process, the blank solution was made [9].

2.5. Determination of Heavy Metals

The quantities of Cr, Cd, Pb, As, and Hg in the vegetable samples were measured using air acetylene arc atomic absorption spectroscopy (AAS) with a deuterium arc background corrector and an air-acetylene flame under various working conditions (Table 1). The atomic absorption spectrometer was calibrated using a calibration blank and five operating calibration standard solutions of each metal to be studied. By plotting absorbance vs concentration, a calibration curve was created, and working standards were also made by further diluting 1000 ppm stock solutions of each element. Using interpolation, the metal concentration in sample digests was determined. Results were collected after averaging three measurements for each sample [10].

Table 1. *Standard operating procedures for the analysis of the most prevalent heavy metals found in plant materials.*

Elements	λ_{max}(nm)	Slit size (nm)	Flame type
As	193.7	0.8	Air/Acetylene
Cd	228.9	0.7	Air/Acetylene
Cr	357.9	0.5	Air/Acetylene
Hg	253.7	0.7	Air/Acetylene
Pb	283.3	1.3	Air/Acetylene

2.6. Statistical Analysis

The full analysis was carried out three times. The mean SD of the results was displayed. Statistical significance was established using a one-way analysis of variance (ANOVA). Means were separated using Duncan's multiple range analysis using the SPSS 16.0 software (p 0.05).

3. Results and Discussion

The most harmful heavy metals, including As, Cd, Cr, Pb, and Hg, were measured in this study's green vegetables, which are popular foods in the Sheka region in southwest Ethiopia. Table 1 lists the typical concentrations of heavy metals found in the green vegetables that were studied that are popular in the Sheka region in southwest Ethiopia. The results, which are presented as mean SD, represent the averages obtained from three replicates.

3.1. Arsenic (As)

Arsenic is a highly dangerous substance. Because of its release into the air during the smelting of as-containing ores, coal burning, and the usage of arsenic compounds in a variety of products like fungicides, insecticides, herbicides, pesticides, and preservatives, it is polluted. When arsenic levels in food exceed the maximum allowable limit (0.03 mg/kg), both immediate and long-term health effects can occur, including headaches, nausea, diarrhea, weakness, and lack of appetite. Bone marrow and blood cells are also

impacted by arsenic exposure. Arsenicals are known to cause lung cancer, and early skin lesions, they may also cause skin cancer.

Brassica cainata, a sample from the Mizan market, had the highest concentration of arsenic (0.09–0.02 ppm), whereas samples of Lactuca Satk, Cucurbita Leaf, and Beta vulgaris, from the Tepi, Meti, and Fide markets, respectively, had the lowest concentrations (BDL) as shown Table 2, and Figure 1a. Every sample showed results that were below the WHO-recommended threshold, proving that the herbs are safe to consume and utilize medicinally.

3.2. Cadmium (Cd)

Cadmium is a non-essential element that can be found in foods and natural water sources. It mainly builds up in the liver and kidneys [11, 12]. Both acute and chronic poisoning from cadmium can have negative effects on the kidney, liver, vascular, and immunological systems [13]. The use of agricultural inputs such fertilizers, herbicides, and biosolids (sewage sludge) as well as the dumping of industrial waste or the deposition of air pollutants all raise the total concentration of Cd in the soil [14]. Cadmium levels in the current study vary from 0.03 to 0.09 milligrams per kilogram (Table 2, and Figure 1b). The cadmium concentration of every vegetable from the four locations was under the safe WHO standard of 0.2 mg/kg.

3.3. Chromium (Cr)

The mineral chromium (Cr) is both a necessary nutrient and a health risk. Chromium comes in two different forms: trivalent and hexavalent. The first is secure for humans and can be found in foods and supplements. The second substance is a known carcinogen that can lead to lung cancer and skin issues. Due to its high solubility, capacity to penetrate cell membranes, and potent oxidizing ability, hexavalent chromium is regarded as the greatest hazard. Because of its high rate of absorption on living surfaces, Cr (VI) is therefore more hazardous than Cr (III) [15]. The present study revealed that the highest level of chromium was found to be 0.98 0.03 ppm in Beta vulgaris from Mizan market, while the lowest concentration (0.29 0.02) was seen in Lactuca Sat from Fide market, as shown in Table 2, and Figure 1c. The FAO (1984) defined an acceptable limit of 2.3 mg/Kg for Cr in edible leafy greens. All vegetables accumulated Cr below the FAO recommendations when the metal limit in the researched medicinal plants was compared

to these values (1984).

3.4. Lead (Pb)

Various morphological, physiological, and biochemical activities of plants are impaired either directly or indirectly as a result of excessive lead buildup in plant tissue. More lead is likely to be absorbed by leafy vegetables, potatoes, and beans than by fruiting plants like tomatoes. Lead prevents the creation of life-sustaining energy by preventing the use of oxygen and glucose. When the blood-lead level exceeds 0.3 ppm, interference with regular metabolic processes begins. Anemia symptoms will be present when the blood-lead level reaches about 0.8 ppm due to a hemoglobin deficiency. Since lead is harmful to the central and peripheral neurological systems, higher blood levels of lead can harm the kidneys and the brain.

The concentration of lead (Pb) in the current inquiry ranged from BDL to 0.0160.01 ppm, as shown in Table 2 and Figure 1d. Cucurbita Leaf from the Mizan market had the highest concentration found. All of the samples had lead concentrations that were just below the legal limit, enhancing the risk of harm from continuing use of these plants.

3.5. Mercury (Cu)

Mercury is extremely bioaccumulative and poisonous [16]. Although it is a significant trace metal, food does not require it. The fact that it is found in food suggests contamination [17]. There are three major types of it: metallic elements, inorganic salts, and organic molecules. Each type has a varied level of toxicity and bioavailability. Although mercury is naturally present in the soil, it can also enter the environment through the burning of fossil fuels, industrial processes, pesticides, herbicides, and liquid industrial waste. Excessive mercury exposure has been linked to a variety of harmful health effects, including immune system and pituitary gland damage that can occasionally result in death [18].

The mercury concentration of vegetables from this study's four market locations ranges from 0.002 0.00 mg/kg to 0.016 0.01 mg/kg, as shown in Table 2 and Figure 1e. Mercury was occasionally below the detection threshold. All of the vegetables from the four locations had mercury contents that were within the permissible limit established when comparing the measured value to WHO standards. This shows that damage from heavy metals from eating these vegetables is unlikely.

Table 2. Average Concentration of Heavy Metals (Mean ± SD, n = 3, mg/kg Dry Weight) in vegetables Collected from four different Markets.

Heavy Metals	Site	Vegetable sample (ppm)			
		Brassica cainata (Cabbage)	*Lactuca Sat* var (lettuce)	*Cucurbita Leaf* (leaf of gourd)	*Beta vulgaris* (Chard)
As	A	0.09± 0.02	0.01± 0.03	0.05± 0.02	0.03± 0.02
	B	0.05± 0.02	0.08± 0.02	0.03± 0.02	0.02± 0.02
	C	0.03± 0.03	0.07± 0.02	BDL	BDL
	D	0.01± 0.03	BDL	0.02± 0.002	0.02± 0.03
Cd	A	0.08± 0.00	0.09± 0.01	0.09± 0.00	0.07± 0.02
	B	0.05± 0.00	0.05± 0.01	0.08± 0.00	0.07± 0.02
	C	0.03± 0.00	0.05± 0.02	0.06± 0.01	0.03± 0.02
	D	0.04± 0.00	0.03± 0.02	0.04± 0.01	0.06± 0.02

| Heavy Metals	Site	Vegetable sample (ppm)			
		Brassica cainata (Cabbage)	*Lactuca Sat* var (lettuce)	*Cucurbita Leaf* (leaf of gourd)	*Beta vulgaris* (Chard)
Cr	A	0.66± 0.04	0.83± 0.03	0.96± 0.00	0.98± 0.03
	B	0.53± 0.04	0.74± 0.02	0.73± 0.01	0.74± 0.00
	C	0.47± 0.01	0.50± 0.03	0.52± 0.01	0.48± 0.01
	D	0.38± 0.01	0.29± 0.02	0.33± 0.00	0.41± 0.03
Hg	A	0.009± 0.02	0.007± 0.02	0.002± 0.02	0.010± 0.00
	B	0.005± 0.02	0.001± 0.02	0.006± 0.02	0.008± 0.02
	C	0.002± 0.03	0.003± 0.01	BDL	0.001± 0.02
	D	BDL	0.009± 0.02	0.001± 0.01	BDL
Pb	A	0.012± 0.02	0.011± 0.01	0.016± 0.01	0.013± 0.00
	B	0.01± 0.02	0.01± 0.02	0.005± 0.01	0.003± 0.00
	C	0.008± 0.03	BDL	0.06	BDL
	D	BDL	0.002± 0.00	BDL	0.005± 0.01

A = Mizan, B = Tepi, C = Meti, D = Fide, BDL = Below Detectable Level, ppm = parts per million

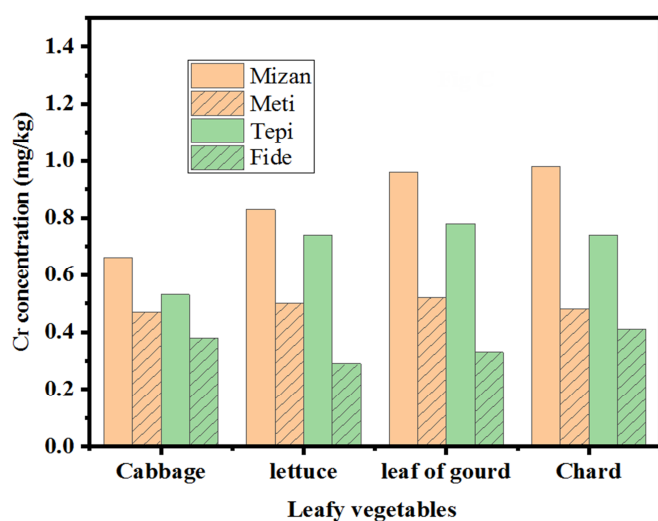***Table 3.** Permissible Guideline for Heavy Metals in Fruits and Vegetables.*

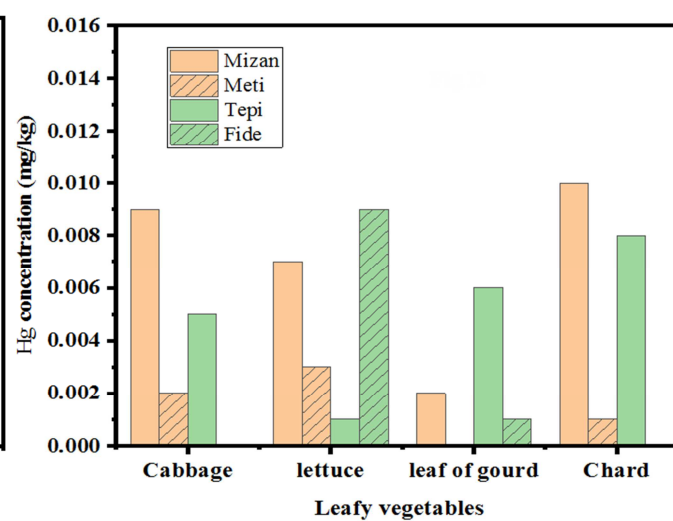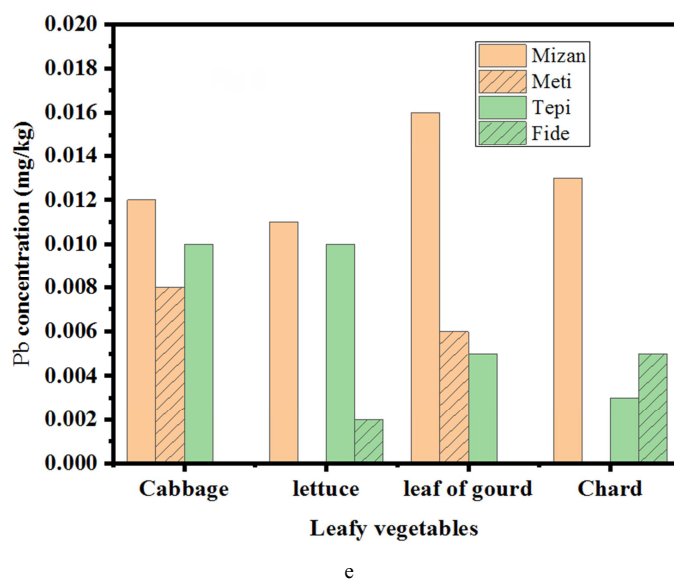| Heavy Metals	Values in mg/kg	Source
Lead	0.1	FAO/WHO (JECFA, 2015)
Mercury	0.1	FAO/WHO (JECFA, 2015)
Cadmium	0.1	FAO/WHO (JECFA, 2015)
Arsenic	0.2	FAO/WHO (JECFA, 2015)
Chromium	4	FAO/WHO 1999
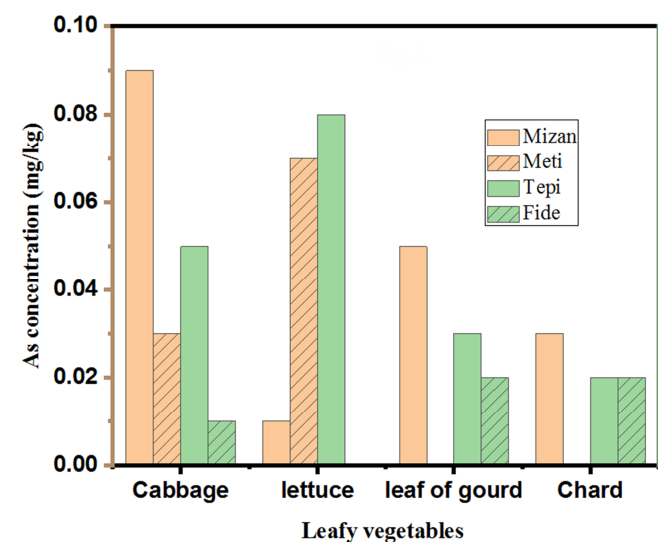
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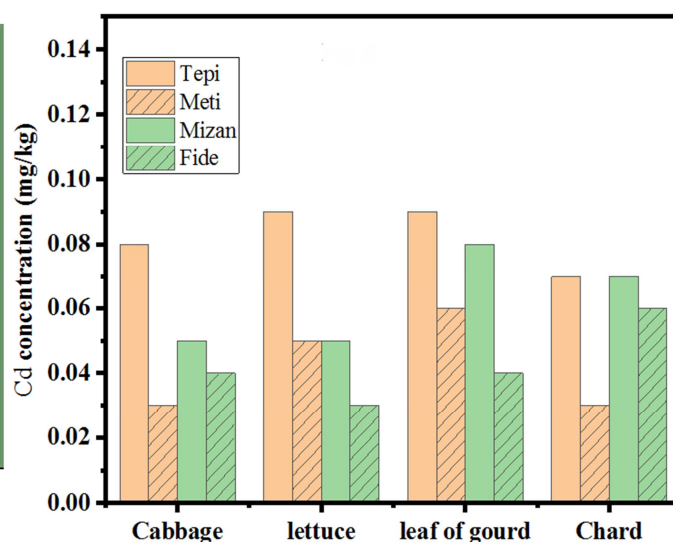
b

c

d

Figure 1. Concentration of heavy metal levels in selected edible leafy vegetables in the sheka zone.

4. Conclusion

Since humans were exposed to vegetable contamination, it was vital to analyze the quantity of hazardous heavy metals in vegetables. In the current investigation, four distinct leafy vegetables that were harvested from four separate sites in the Sheka zone of southwest Ethiopia were tested for five heavy metals, including As, Cd, Cr, Pb, and Hg. The investigation's findings demonstrate that the sampling site's heavy metal level varies. The prevailing soil and other growth circumstances, as the crops' varying capacities for uptake and for transporting heavy metals to the edible region of the vegetable, may all play a role in this diversity. The sample taken from the Mizan district shows higher accumulations of heavy metals, while the sample taken from the Fide district shows lower accumulations. The amounts of heavy metals were found to be within acceptable limits when the results were compared to the FAO/WHO standard permitted limit. Therefore, it can be said that these veggies won't cause heavy metal toxicity and may even help people who are lacking in certain micronutrients.

Data Availability

The corresponding author, who had complete access to all the study's data and who is responsible for the data's integrity and accuracy in the data analysis, will provide the datasets that were gathered and used for analysis upon request.

Conflicts of Interest

The author declare that there are no conficts of interest.

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