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Full Length ResearchArticle

Determination and comparisons of Heavy Metals in Moringa Stenopetala Leaf, Seed and Root: using Atomic Absorption Spectroscopy

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Abstract:

This study is aimed to determine and compare the concentration level of heavy metals (Cu, Fe, Pb and Cr) using Atomic Absorption Spectroscopy (AAS) of samples of fresh leaf, seed and root of Moringa stenopetala from Arbaminch area and Konso districts . Wet digestion method involving use of mixture of ($HClO_4$ and HNO_3, ratio 3:1) and 2ml of 30% H_2O_2 at an optimum temperature and time duration was deployed during sample preparations. Results show that the mean concentration level of copper in leaf, seed and root were 2.05 ± 0.05 mg/kg ,0.789 ± 0.003 mg/kg and 1.068 ± 0.023 mg/kg respectively in the Arbaminch area and 1.491 ± 0.02 mg/kg, 0.736 ± 0.04 mg/kg and 1.365 ± 0.017 mg/kg respectively in Konso. As similarly, the mean concentration level of iron metal in leaf, seed and root were 21.6 ± 0.19 mg/kg 12.44 ± 0.051mg/kg and 22.817 ± 0.29 mg/kg respectively in Arbaminch area and also the mean concentration level of iron was 20.321 ± 0.21 mg/kg,12.762 ± 0.0022mg/kg and 21.714 ± 0.22mg/kg respectively in Konso. The level of toxic metal lead was not detected in edible part of Moringa stenopetala and it was detected only in root part 0.347 ± 0.001mg/kg, 0.326 ± 0.005mg/kg in Arbaminch area and Konso districts respectively. Chromium was not detected in all Moringa stenopetala part of (leaf, seed and root). The results of this study indicate that the concentrations of heavy metals are below the range and iron medicinal level in root was closely related to WHO/FAO limits.

1. Introduction

In Africa, many studies have indicated that a vast number of indigenous wild plants play a significant role in the diet of the population (Muhammad *et al.,* 2011). Vegetables are the cheapest and the most available sources of important nutrients, supplying the body with minerals, salts, vitamins and certain hormone precursors, protein, energy and essential amino acids (Amaechi, 2009)
Moringa is one such group whose various species belonging to the family *Moringaceae* and it consists of 13 species, distributed in the Indian subcontinent (*M. oleifera* and *M. concanensis*), Kenya (*M. longituba* and *M. rivae*), northeastern and southwestern Africa (*M. stenopetala*), Arabia, and Madagascar (*M. drouhardii* and *M. hildebrandtii*) (Padayachee and Baijnath, 2012; Saini, 2015).*Moringa oleifera* Lam. is a tropical deciduous perennial dicotyledonous tree and native of the sub-Himalayan mountains of northern India; is now cultivated for a variety of purposes in the whole tropical and sub-tropical regions of the world (Leone *et al*. 2015).

Among different types of Moringa tree, Moringa stenopetala is a multipurpose tree as significant

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economic importance, vital nutritional value as food for human beings and feed for livestock, industrially used for dye, perfume skin lotion, lubricant and medicinal applications (Jahn, 2005).

It is a native plant to the horn of Africa, particularly in southern Ethiopia, north Kenya and Eastern Somalia. In Ethiopia, it is found in many arid zones of the southern Ethiopia most extensively by different vernacular names such as Shiferaw (Amharic), Aleko ,Aluko, Haleko (Gamo Gofa and wolaita), Telahu(Tsemay), Shelkhata (Konso), Haleko (Derashe), Mawe (somali) and others..

It is widely distributed at an altitude range of about 1100 to 1600 m grow small (up to 12 m), with a much branched crown and some time with multi-trunks. The leaves are bi-pinnate or tripinnate with about five pairs of pinnate and three to nine elliptic or ovate leaves lets on each pinna. The fruits are long reddish pods with a grayish bloom Different parts of the Moringa stenopetala tree including roots, bark, leaves, flowers, fruits and seeds are traditionally used in various therapeutic applications including, abdominal tumors, hysteria (a psychological disorder), scurvy, paralysis, helminthic bladder, prostate problems, sores and other skin infection (Farooq *et al*. 2012; Mbikay; 2012). Almost each and every part of *Moringa* tree is useful for medicinal and food preparations, nutraceautical, water purification, and biodiesel production; including roots, leaves, flowers, green pods, and seeds (Saini.,2015). Local people use the plant parts to treat malaria, leishmaniasis and hypertension as a traditional medicine. In general, *Moringa stenopetala* is miraculous tree as the multipurpose importance for the humans and animals.

Vegetables cultivated in soils polluted with toxic and heavy metals take up such metals and accumulate them in their edible and non edible parts in quantities high enough to cause clinical problems both to animals and human beings consuming these metal-rich plants as there is no good mechanism for their elimination from the human body. The uptake of metal by plants roots depends on the form the metals exist in the soil and the nature of the soil and the plant species. Thus, metal mobility and plant availability are very important when assessing the effect of soil contamination, plant metal uptake, and toxicity (Bhuiyan, 2011).

Moringa stenopetala contains several elements which are the basic building blood matter such as calcium, magnesium, potassium, sodium and the minor elements are iron, zinc, copper and manganese (Melesse and Berihun, 2013). Since, it is a source of vitamins and lot of mineral constituents which are necessary for proper development and functioning of different tissues and organs. However, overdose of these vitamins or mineral constituents can be harmful (Farid *et al*., 2004). Determination of minerals and trace elements in foodstuffs is an important part of nutritional and toxicological analyses. Copper, Chromium, Iron and zinc are some of essential micronutrients for human health. In addition, these elements play an important role in human's and animal's body metabolism. The interest in these elements is increasing due to the available reports of relationships between heavy metals status in food and drinking water and the prevalent oxidative diseases in living beings. Lead, Cadmium,, Nickel, Arsenic and Mercury are the most common toxic metals of concern due to the reports of their contamination in various herbal preparations and herbal ingredients and Chromium is associated with diabetes and cardiovascular diseases (Nathani *et al*., 2010).Cadmium is recognized as a metal posing threat to agricultural food quality due to its mobility in the soil– plant system added to soil from the metal-working industries, waste incinerators, urban traffic, cement factories, and as a by product of phosphate fertilizers. In areas with low anthropogenic activity, Cadmium can be released as a result of rock mineralization processes. It enters into plant cells due to its similar chemical and physical characteristics to plant nutrients. It can cause many toxic symptoms, such as inhibition of growth and photosynthesis, deactivation or inhibition of enzymes, disturbances in plant water relationships, ion metabolism, and formation of free radicals (Kumars *et al*., 2008).

The alarming contaminations of heavy metals in soils and plants have become an important issue worldwide because of their adverse effects on human health and serious threat to food safety. Metal pollutants can easily enter the food chain if heavy metal-contaminated soils are used for the production of food crops. Some elements as essential micro-nutrients have a variety of biochemical functions in all living organisms and form an integral part of several enzymes (Sommers, 2003). Although they are essential, they can be toxic when taken in excess; both toxicity and necessity vary from element to element and from species to species (Tripathi, 2008). Thus, information on the intake of heavy metals through food chain is important in assessing risk to human health.

1.1 Statement of problem

There has been a sharp increase in nutrition related diseases and conditions such as diabetes, high blood pressure and cancer. Child mortality rate has also increased due to Malnutrition (Kaburina, 2004). There are over two billion people in the world that suffer from Micronutrient deficiencies due to poor diets (UNSCO, 2004). Micronutrient deficient Diets lead to reduced mental and physical development, performance in school and loss of Productivity in the workplace (Branca, 2002).Balanced nutrition helps to minimize or even eliminate nutrition related diseases in humans.

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Heavy metals are potential environmental contaminants with the capability of causing human health problems if present to excess in the food we eat. They are given special attention throughout the world due to their toxic effects even at very low concentrations (Das, 1990). Several cases of human disease, disorders, malfunction and malformation of organs due to metal toxicity have been reported (Jarup, 2003).

Some heavy metals are very essential for life needed in human and animal diets in order to maintain good health (WHO and FAO, 2002).It mostly known for nutritious food and medicinal utilization that play a vital role in tackling disease protection and mineral micronutrients deficiencies. However, both deficiency and excesses of these heavy give very serious problems in the human and animal body. Because, there is lack of clearly studied elemental composition of these plants to attain the permissible limit for heavy metals in plant leaves, seeds and root as recommended by World Health Organization (WHO).

Studies conducted in Nigeria have assessed concentration of heavy metals in *Moringa oleifera* and found lead, iron, copper and zinc with mean(mg/kg) of 0.9471±0.0173, 55.60±0.012, 0.1762±0.0230and 3.225±0.022, respectively though atomic absorption spectrometer analysis (Abdulkadir et al., 2016).Most of the data reported in research account for *M.oleifera*. However, all the plant parts of *M. stenopetala,*which is highly cultivated in southern part of Ethiopia,have not been analyzed for their metal concentration and nutritive values. The specific objectives of the current study are: to determine the levels of heavy metals such as: Copper, iron, lead and chromium in *Moringa stenopetala* leaf, seed and root collected from different sites Arbaminch and Konso, to compare and contrast the different level of heavy metals in the leaf, seed and root of Stenopetala in Arbaminch area and Konso and with permissible limit for heavy metals in Moringa plant leaf, seed and root as it is recommended by WHO and to compare and contrast the concentration level of heavy metals in M. *Stenopetala* leaf, seed and root of Arbaminch area and Konso with literature review.

2. MATERIALS AND METHODS

The study was carried out in two selected Zones of southern Ethiopia from November
2019 to April 2019 in Arbaminch area and Konso districts of SNNPR, Ethiopia. Arbaminch Zuria district is located at 6° 01'59" N and 37° 32'59" E, altitude of 1269 m above sea level and 505 km away from the capital city, Addis Ababa and Konso district is located at 5°15'00" N and 37°28'59" E and altitude of 1031 m above sea level. It is 536 Km far from Addis Ababa.

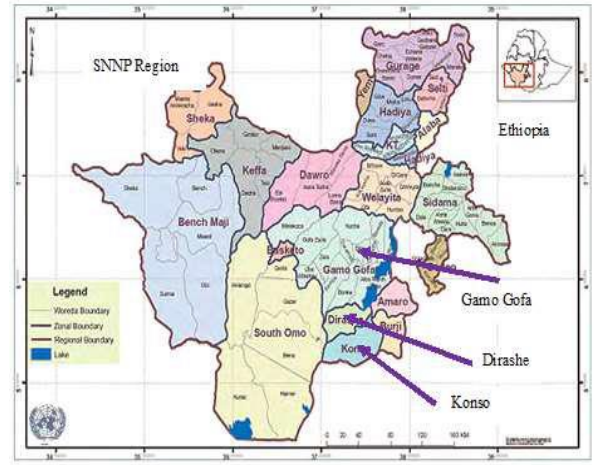

Figure 1. Map of study area (retrieved at: www.rippleethiopia.org/page/snnpr).

2.1 Sampling protocol

The study areaswere selected purposefully based on the productivity and traditionaluse of roots parts of *M. stenopetala* for medicinal purpose. Tocollect the representative sample from each sampling sites, twosubsamples were taken from each site. In total, six samples werecollected and put in clean polyethylene plastic bags, labeled and brought to laboratory for further pre-treatment.

2.2 Preparation of calibration curves

Diluted standard solutions were prepared from the stock standard solutions (1000ppm) which were already made by dissolving amount of their respective metal salts of analytical grade (purity ,99.9%) in $HOCl_4$ and $HN0_3$,diluting with ultra pure distilled water and strong as stock solutions in quartz flask. Standard diluted solutions were prepared for each metal depending up on the linear working range of the absorbance was measured .Triplicate measurements were carried out for each metals and the mean value were reported .A calibration curve of absorbance against concentration of each metal was constructed and interpolated .

2.3 Method performance and validation (Spiking of samples)

The optimized procedure was validated by determining the following method of validation parameters such as; precision (in terms of repeatability), accuracy (in terms of recovery), method detection limit, limit of quantization (LOQ), analysis of laboratory control sample, matrix spike and matrix spike duplicate were carried out (Chauhan, A. *et al*., 2015). The metal concentration of a sample spiked with a known amount of each metal. Then, the percent recovery (%R) of the method was evaluated. The procedures followed in

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the spiking of all leaf, seed and root samples, known concentration of the metal standard solutions were added using a pipette .then, the spiked samples were digested in optimized procedure and analyzed by AAS

2.4 Statistical Analysis

The levels of metals concentration in leaf, seed and root samples were compared. The null hypothesis being tested in the study, that there is no significant difference between samples. Metals concentration of the all leaf, seed and root samples were expressed as mean + SD of triplicate measurements (n=3). Mean and SD of each sample were used to compute the calculated t-value. Thus ,differences between the critical t-value and calculated value of the metals concentration of the leaf, seed and root samples were analyzed by One-way analysis of variance (ANOVA) at $P < 0.05$ was used to determine statistically significant differences in the mean concentrations of metals among groups of leaf, seed and root samples.

2.5 Sample Collections

The immature (medium young), healthy and fresh Moringa leaf, seed and root were collected from southern region, Konso (Gato, Segen and Karat) and Arbaminch area (Secha, Shara and Chano) zones, SNNPR, Ethiopia. The collected samples were carried out by the identification of Arbaminch university botanist and PhD student Mrs Eyasu Chama, the collected plant part samples were kept separately in polyethylene bags in Arbaminch University Chemical engineering laboratory for further pre-treatment.

2.6 Sample preparation

The collected samples of leaf were washed gently and thoroughly with distilled water in order to make free from dust particles and other unwanted materials that may adhere to it. Then, the leaf part was exposed to dry in the room temperature at $37C^o$ for 24 hours, after drying the woody and rotten portions of leaf were removed. The collected samples of seed part, the outer skin or kernel was removed by hand was exposed to drying at room temp of $37C^o$ for 24 hours to constant weight. Other collected samples of root were washed with a tap water to remove attached soil particles and then rinsed with de _ionized water. After cleaning the root was chopped into small pieces using a stainless steel knife, dried under shade to avoid direct sun shine that could degrade or loss of bioactive compounds present in the plant for metal analysis, the sample was taken to oven dried at $90C^o$ for 24 hours until a constant weight obtained. All dried samples were grinded or powdered by using a electronic coffee mortar blender and sieved at 2mm size to prepare uniform particle size .know all samples such as leaf, seed and root powders were weighted and stored in clean, dried plastic bottles at room temperature for further analysis.

Table 1: during sample preparation processes

Plant part	Amount of weight	Temperature	Sieve
Leaf	1200mg	Room ($37C^0$)	2mm
Seed	1200mg	Room ($37C^0$)	2mm
Root	1200mg	Oven ($90C^0$)	2mm

2.7 Wet digestion of samples

The wet digestion was carried out by using Kjeldahl digestion apparatus with a reflux condenser for all samples leaf, seed and root .The four parameters, temperature, time and reagent volume were optimized. The powders of all sieved samples were weighted 1 gram separately and put into 100ml conical flask of each sample. Then, the samples were digested in a separate Kjeldahl digestion apparatus by the addition of 2ml of mixture of $HClO_4$ and HNO_3, ratio 3:1and 2ml of 30% H_2O_2. H_2O_2 was added by a small portion, to avoid any possible overflow of loss of material from the conical flask.

The Samples were digested for 2hr in 100ml conical flask covered with watch a glass, and reflex over a hot plate at $75C^o$ for 2 hours to minimize the vigorous reaction that may occurs during digestion in hot plate. But there were no clear sample solutions obtained in each digestion apparatus. Then, digestions cooled at room temperature and then, 2ml of mixture of $HClO_4$ and $HN0_3$, 3:1, were added to milky (precipitated) solutions and heated for additional 40 min at $150C^o$. There were also no clear solutions were obtained and again for additional 20 min at temperature increased to $245C^o$ until the clear pale yellow solutions were obtained.

Therefore, optimum conditions for digestion of all samples leaf, seed and root were at the temperature of $245C^o$ with the time of 3 hours and reagent 2ml of mixture of $HClO_4$ and $HN0_3$, 3:1. Then, 5ml of distilled water was added and then, the solutions were shaken to dissolve precipitates remained on the wall of the digestion apparatus. Then, the solutions were filtered out into 100ml conical flasks through a Whitman No.42 filter paper with a 0.45micrometer to remove the insoluble solid from the supernatant liquid. The volume was adjusted to 3ml with distilled water and kept in room temperature until to carry out AAS for determination analysis.

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A gram of sieved powder of the samples was weighed out into acid washed glass beaker. Thereafter, the powder was digested with addition of 20 cm^3 of aquaregia (mixture of HCl andHNO_4, ratio 3:1). 10 ml of 30% H_2O_2 was added to avoid any possible overflow leading to loss of material from the 100-ml conical flask. Hydrogen peroxide is also used to enhance the reaction speed and to ensure complete digestion. The analyte was digested for 2 h in 100-ml conical flask covered with watch glass and reflex over a hot plate at 90°C for 2 h. The volume was adjusted to 100-ml with distilled water. Blank solution was handled as detailed for the samples. Working standard solutions of all metals were prepared from stock

Table 2:.Instrumental working conditions for analysing metals using AAS

S.N.	Metals	Wavelength (nm)	Slitwidth (nm)	Lamp current (mA)	Energy
1.	Cu	357.9	0.7	2.0	3.623
2.	Fe	248.3	0.2	5.0	3.331
3.	Pb	324.9	0.7	1.5	3.775
4.	Cr	283.2	0.2	4.5	3.338

3. RESULTS AND DISCUSSION

The quality of results obtained for Cu, Fe, Pb and Cr metals analysis using AAS are affected by the calibration and standard solution preparation procedure. The instrument was calibrated using five series of standards. The calibration graphs of the four metal standard solutions were drawn using the standard solution data and the unknown concentrations of each Metal was determined using the slope equation y=mx+b from the calibration graph. The unknown concentration of the metals was determined from the graphs and the metal Fe has the highest concentration and Cr metal has the lowest concentration in the sample.

Table 3: Concentration of standards samples and absorbance of each metal

Standard	Metals			
	Copper (Cu)	Iron (Fe)	Lead (Pb)	Chromium (Cr)
	Conc. .(mg/L)	Conc. (mg/L)	Conc. .(mg/L)	Conc. (mg/L)
1	0.15	2	0.11	0.1
2	0.25	4	0.12	0.2
3	0.35	6	0.13	0.3
4	0.45	8	0.14	0.4
5	0.55	10	0.15	0.5

Table 4: Summary of Table (8 and 9) for the concentration level of heavy metals in Mean±SD, (N=18)

Parts of plant	Metals	Arbaminch Conc. mean ±SD (mg/kg)	Konso Conc. mean ±SD (mg/kg)	WHO/ FAO
Leaf	Cu	2.05±0.523	1.491±0.021	3mg/kg
	Fe	21.6±0.194	20.32±0.206	28mg/kg
	Pb	ND	ND	
	Cr	ND	ND	
Seed	Cu	0.789±0.031	0.736±0.042	3mg/kg
	Fe	12.44±0.056	12.762±0.02	28mg/kg
	Pb	ND	ND	
	Cr	ND	ND	
Root	Cu	1.068±0.023	1.365±0.017	10mg/kg
	Fe	22.817±0.29	21.714±0.02	20mg/kg
	Pb	0.347±0.001	0.3263±0.05	10mg/kg
	Cr	ND	ND	

All metal levels obtained were analyzed for leaf, seed and root samples were expressed in mg/l (Table8) As in the dry weight expression, the mean concentration level of copper metal in leaf, seed and rootwas2.05±0.523mg/kg, 0.789±0.0311mg/kg and 1.068±0.023mg/kg respectively. As similarly, the mean concentration level of iron metal level in leaf, seed and root were 21.6±0.194 mg/kg, 12.44±0.0516mg/kg and 22.817±0.294mg/kg respectively. Whereas the mean concentration level of Pb was detected only root as 0.347±0.001mg/kg. But, the concentration of Cr was not detected in all parts of plants such as leaf, seed and root respectively. The amounts of analyzed metal levels in the leaf, seed and root samples were expressed in mg/L or ppm on (Table 9). As in dry weight basis, the mean concentration level of copper metal in (Table 9) showed that1.491±0.021mg/kg,0.736 ± 0.042mg/kg,1.365 ± 0.017mg/kg in leaf, seed and root samples respectively.

As similarly, the mean concentration level of iron metal in the above (Table4) indicates that 20.32 ± 0.206mg/kg,12.762±0.0022mg/kg, 21.714±0.0.22mg/kg in leaf, seed and root samples respectively. Whereas the concentration of lead metal was detected only in the root part was 0.3263 ± 0.005mg/kg .However, chromium was not detected in the all plant parts such as leaf, seed and root respectively.

In this study, the concentration levels of metals in leaf, seed and root sampled from the same sample size and from the same plant parts were determined. The above tables (8 and 9), results shows that, the concentration level of iron metal was greater than copper in the leaf part and similarly iron metal was greater than copper in the seed part. As the same manner iron was greatest comparing with copper and lead in the root part of the moringa *stenopetala* plant and lowest lead metal concentration was occurred. However, chromium does not detected in all moringa stenopetala plant parts such as leaf, seed and root

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respectively. Therefore, the concentration level of metals in leaf, seed and root samples were in the order: Fe>Cu, Fe> Cu and Fe>Cu>Pb respectively, in the both sample sites (Arbaminch area and Konso).
In general, comparing the concentration level of metals in leaf, seed and root, the concentration level copper was highest in leaf part comparing with seed and root, whereas lowest concentration was in the seed part. As similarly, the concentration level of iron metal was highest in the root part comparing with the other two parts such as leaf and seed. Whereas, lowest concentration level of iron was found on the seed part. However, the concentration of lead was detected only in the root part only and chromium does not detected in all parts of plants. In general, the concentration level of heavy metals copper and iron were ordered as; leaf>root>seed and root>leaf>seed respectively.

3.1 Comparison of heavy metal concentration in plant parts obtained present study with standard values

The concentration level of copper in leaf, seed and root samples in Arbaminch area were 2.05 ± 0.5mg/kg, 0.789 ± 0.0311mg/kg and 1.068 ± 0.023mg/kg (Table 10) respectively and the concentration level of copper in leaf, seed and root samples in Konso were 1.491±0.021mg/kg, 0.736 ± 0.042mg/kg, 1.365 ± 0.017mg/kg (Table 10) respectively.
In this result the minimum value of copper was 0.67±0.03mg/kg and 0.644±0.942mg/kg in seed part; maximum value was 1.08±0.05mg/kg and 1.409±0.021mg/kg in the leaf part of Arbamincharea and Konso respectively. The maximum limit set by FAO/WHO in for medicinal plant is 10mg/kg, while its intake in edible plant is 3mg/kg (Shagal *et al*., 2012) and the permissible limit in medicinal plants for Cu set by China and Singapore were 20mg/l and 150mg/l respectively (Mao *et al* .,2012). As compare to the FAO/WHO current study which agrees with WHO of below permissible limit in all parts of Moringa s*tenopetala* so as it advisable to eat.
The concentration level of copper in all leaf, seed and root samples in the present study lower than permissible level. Although the copper mobility within tissues strongly depends on the level of Cu supply, it has low mobility relative to iron and other elements in plants and uptake of copper in plants is usually low (Ondo *et al.,* 2012).
The iron concentration obtained in this study as 21.6±0.194mg/kg, 12.44±0.0516mg/kg and 22.87±0.294mg/kg respectively in leaf, seed and root samples respectively (Table 10) in Arbaminch area and the mean concentration levels of iron metals in leaf, seed and root obtained as 20.32±0.206mg/kg, 12.762±0.0022mg/kg, 21.714±0.0022mg/kg respectively (Table 10) in Konso sample sites. This result indicated that, the minimum value of iron was found 12.44±0.0516mg/kg and 12.762±0.0022mg/kg in the seed part of moringa stenopetala Arbaminch area and Konso respectively. Whereas the maximum concentration of iron was 22.817±0.294mg/kg and 21.714.89±0.022mg/kg in the root part of moringa stenopetala part in Arbaminch area and Konso sample sites respectively.
This high concentration of iron is to great advantage for animals that require proper growth and development. In human it plays the vital role for metabolic process such as DNA synthesis and oxygen transport to cells (Ogbe *et al.,* 2012). Daily intake of iron supplements reduces the risk of anemia; and its deficiency can cause various types of diseases (Upadhyay *et al*., 2001).
This is within the recommended permissible limits for WHO/FAO (Table10). Therefore, this makes the plants samples suitable for consumption with regard to Fe (FAO/WHO, 2007). The results showed that the Moringa *stenopetala* root to be a good source of iron. The values are in accordance to recommended daily allowance of iron 100 to 130 mg/kg for children; 70 mg/kg for men and 120 to 160 mg/kg for women and feeding mothers (Ijarotimi *et al*., 2013). In those three selected areas.
Therefore, the concentration of iron metal in root was below the maximum permitted level and it was reported by (Maobe *et al*., (2012) that the range of iron in selective medicinal Herbs of Egypt was between 261 to 1239mg/l. The recommendedpermissible limit of iron level in medicinal plant is 20mg/kg; its dietary intake is 28mg/kg. In present investigation, Fe was found highest concentration in root part and lower in both leaf and seed, which is probably due to iron rich soil of study area.
Lead and Chromium concentrations were not detected in both plant parts leaf and seed in Arbaminch Zuria and Konso respectively (Table 10). These result may be as the result of passive ability of lead and chromium (un-reactive, because of a thin layer of oxide) when treated with nitric acid just as Fe (during digestion).it might also be due to absence of emission from vehicles engines that uses gasoline, which is in combination with lead containing compound as additive and corrosion of lead from vehicle parts (Al-shayeb & Seaward, 2001). These result could indicates the concentration of toxic heavy metals does not present in moringa stenopetala leaf and seed in the small amount.

But the concentration of lead was detected only in the root part of moringa *stenopetala* as 0.33 ± 0.005mg/kg at the lowest value of Konso and 0.3474 ± 0.001 mg/kg as the highest value of Arbaminch area, which is below the permissible limit of FAO/WHO for medicinal plant. Therefore, the plant

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parts have no risk on human being health problem. Chromium was not detected the both selected sample sites in all moringa stenopetala plant parts.

4. DISCUSSION

The concentration level of some metals in moringa *stenopetala* leaf (edible) part obtained in the present work were compared with other common leaf of moringa plants and similar samples reported in literature was summarized in Table1.

The concentration level of copper metal varied from 1.084 – 1.4388 mg/kg, 0.6708 – 0.8778 mg/kg and 0.099 – 1.1439 mg/kg in moringa *stenopetala* leaf, seed and root respectively in Arbaminch area and 1.4097 – 1.536 mg/kg, 0.6411 – 0.906 mg/kg and 1.302 – 1.426 mg/kg in leaf, seed and root respectively in Konso.

The mean concentration level of copper in moringa stenopetala leaf, seed and root in this study were 2.05 ±0.5mg/kg, 0.789±0.0311mg/kg and 1.068±0.023mg/kg respectively in the Arbaminch area and 1.491±0.021mg/kg, 0.736±0.042mg/kg, 1.365±0.017mg/kg respectively in Konso. The mean concentration of copper in this study leaf, seed and root was higher than the levels reported from Mohammed and Masood *et al*., 2015, Leonid and Najat *et al*., 2017, in leaf part and Mohammed and Mommed *et al*., 2015 in seed and also Leonid and Najat *et al*., 2017 and Tsegaye and Kussie *et al*., 2019 in root part. As similarly, the concentration level of iron metal varied from 20.959 – 22.2207 mg/kg, 10.152—13.412 mg/kg and 21.608 -23.613 mg/kg in leaf, seed and root respectively in Arbaminch area and the iron concentration varied 20.955 -20.9577 mg/kg, 11.42 - 13.5936 mg/kg and 20.83 - 22.24 mg/ kg respectively in leaf, seed and root in Konso. The concentration level of iron metal in the moringa stenopetala leaf, seed and root in this study were 21.6 ±0.19 mg/kg, 12.44±0.051mg/kg and 22.817 ±0.29 mg/kg respectively in leaf, seed and root of Arbaminch area and also the mean concentration level of iron in this study was 20.32±0.21 mg/kg 12.762±0.002mg/kg and 21.714±0.22mg/kg in leaf, seed and root respectively in Konso. The mean concentration level of iron in this study was higher than previous review reported from Mohammed and Masood *et al*., 2015 and but much lower than Leonid and Najat *et al*., 2017, in previous report of leaf part.

But the previous reported value from Mohammed and Mommed *et al.,* 2015 , Leonid and Najat et *al.,* 2017, were higher than from the present study leaf and root respectively .

In all analysed plant part samples lead was detected only in root part it concentration range were from 0.3369 - 0.3405 mg/kg and 0.336 - 0.3406 mg/kg in Arbaminch area and Konso respectively.

The mean concentration level of lead in root part was 0.3477±0.03 mg/kg and 0.363±0.01 mg/kg in Arbaminch area and Konso respectively. The previous reported value from Leonid and Najat Mohamed *et al.,* 2017 was higher than the present study in the root part and Tsegaye and Kussie et al .,2019 reported as it does not detected in root part.

However, in the present study the concentration level of chromium does not detected in all moringa stenopetala plants such as leaf, seed and root in both sample sites but the previous reported value from Leonid and Najat et al., 2017 and Mohammed Mommed *et al.,*2015, were 0.44mg/kg and 0.02mg/kg in leaf and seed respectively.

In general, the marked variation in the concentration of metals in the vegetables and medicinal plants might be due to the level of metals in the soils in which they were cultivated (Mohammed and Sharif ,2011), geographical locations ,genetic diversity**,** different farming practices**,** preferential up take of metals by plants and its maturity level , different composition , amount of sampling weight, samples were analyzed by different parameters such as temperature, volume of reagents and time until complete digestion, and different type of instrumental techniques etc.

CONCLUSION

A present investigation the concentration level of heavy metals in the moringa stenopetala leaf, seed and root were determined and compared. The analyses of heavy metals were determined by using Atomic Absorption Spectroscopy in wet digestion method. This investigation helps to know different heavy metal levels found in the *Moringa* Plant leaf, seed, and root

Then, in this result the minimum value of copper was 0.789 ± 0.0311 mg/kg and 0.736 ± 0.042 mg/kg in seed part Arbaminch Zuria and Konso districts respectively.

The maximum value was 1.08±0.05mg/kg and 1.409±0.021mg/kg in the leaf part of Arbaminch Zuria and Konso respectively.

The minimum value of iron was found 12.44 ± 0.0516mg/kg and 12.762±0.0022mg/kg in the seed part Arbaminch Zuria and Konso respectively.

Whereas the maximum concentration of iron was 22.817±0.294mg/kg and 21.714.89±0.022mg/kg in root part Arbaminch Zuria and Konso respectively.

The concentration of lead was detected only in the root part as 0.33 ± 0.005mg/kg at the lowest value of Konso and 0.3474 ± 0.001 mg/kg as the highest value of Arbaminch Zuria,

This analysis showed that there is concentration of essential heavy metals in all selected plant parts and the concentration level of iron metal in root part was higher than copper in the leaf and seed parts.

As the manner iron was the highest one root part comparing with copper and lead, but the concentration of

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chromium was not detected in all plant parts. The presence of cupper and iron in the plant parts justified it is used for nutritional purposes and play an important role to maintain a good human health.

In general, all determined heavy metals concentration were below the permissible limits edible part of plant and closely related in root part iron level with in medicinal plants recommended by WHO.

Therefore, the plant parts have no risk on human being health problem.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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