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Nutritional and Anti-Nutritional Analysis of Anchote (*Coccinia abyssinica*) Tubers Collected from Local Markets of Jimma City, Gimbi and Dambi Dollo Towns, Ethiopia

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Nutritional and Anti-Nutritional Analysis of Anchote (*Coccinia abyssinica*) Tubers Collected from Local Markets of Jimma City, Gimbi and Dembi Dollo Towns, Ethiopia

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Declaration

This thesis work was conduct under my supervision and fulfills all requirements of research standard of the program. I hereby approve the submission of this thesis/senior research for examination.

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ABSTRACT

Anchote is a valuable food source and according to local farmers, it helps in fast mending of broken/ fracture bones and displaced joints, as it contains high calcium, and proteins than other common and wide spread root and tuber crops. This study was aimed to determine the nutritional, anti-nutritional and mineral compositions of Anchote (Coccinia abyssinica) tubers obtained from local markets of Jimma city, Dembi Dollo, Gimbi towns. Totally, 9 kg samples, 3 kg from each site, were purchased from local markets. Their nutritional compositions such as: moisture, crude protein, total ash, crude fiber, crude fat, total carbohydrate and total energy were determined using the Association of Official Analytical Chemists (AOAC) method. The anti-nutritional components including oxalate and, phytate were determined by titrimetric method. Mineral compositions such as Ca, Mg, Fe and Zn were determined by Flame Atomic Absorption Spectrophotometer. The obtained results for nutritional contents for Dembi Dollo, Gimbi and Jimma samples were (66, 70.2, and 73 g/100 g) moisture content;(1.55, 0.80 and 1.17 g/100 g) crude protein; (4.73, 4.43, and 4.24 g/100 g) total ash; (5.92, 5.69, and 5.73 g/100 g) crude fiber; (0.14, 0.22, and 0.41 g/100 g) crude fat; (21.34, 18.54, and 15.50 g/100 g) carbohydrate content; (92.81, 79.38, and 70.37 g/100 g) gross energy, respectively. The phytate and oxalate content were 0.18, 0.12, 0.07 and 0.81, 0.62, and 0.26 mg/; while the mineral content of raw Anchote samples contained Fe (6.8, 34.95, and 36.9 mg/100 g), Ca (114.95, 49.95, and 64 mg/100 g), Mg (16.35, 21.1, and 19.65 mg/100 g), and Zn (12.7, 11.25, and 9.1 mg/100 g)100g in Dembi Dollo, Gimbi and Jimma samples, respectively. One way ANOVA ($p< 0.05$) showed the presence of significant variation among anchote samples collected from different sites.

Keywords: *Nutritional, Anti-nutritional, Mineral compositions, Anchote (Coccinia abyssinica) tubers*

1. INTRODUCTION

Anchote (*Cocciniaabyssinica*) is a tuber crop, belongs to the order *Cucurbitales,* family *Cucurbitaceae* and it is indigenous to Ethiopia [1]. There are about 10 species of *Coccinia* in Ethiopia; however, only *Coccinia abyssinica* is cultivated for human consumption [2]. The most widely used vernacular name of the crop is "*Anchote*", spelt "*Ancootee*" in Afan Oromo. It is also named as Ushushu (Welayita), UShushe (Dawuro), and Ajjo (Kafigna) [3]. Anchote is widely cultivated and used in Jimma, Illu Abba Bora, Buno Bedelle, West and East Wollega, Qellem Wollega and Horo Guduru Wellega Zones of Oromia Regional State, Ethiopia [4]. It is a subsistence crop commonly produced to fill food security gaps during the hunger months (June to September). The total yield of Anchote is 150-180 quintals/hectare, which is in the same range of the total yield of sweet potato, and potato [5]. Cultivation of Anchote grows at elevation of 1300–2800 m with an annual rainfall of 762–1016 mm [6, 7]. It prefers soil having pH of 4.5 to 7.5, mean minimum and maximum temperature of 12 °C and 28 °C, respectively [8].

Anchote has good nutritional compositions with good supplement of vitamins and minerals compared to other tuber crops, and regarded as a leading protein root crop with high calcium content [8]. It has also potential importance to be used to fight protein deficiency in developing countries such as Ethiopia [9]. Juice prepared from Anchote is used to treat gonorrhea, tuberculosis and tumor cancer among local community in the western part of Ethiopia [10]. Moreover, Anchote is used among to prepare a variety of food items which can be used during traditional ceremonies, as special food for guests and animal fattening [11].It is used to treat bone fractures and displaced joints among traditional communities in the area where it is widely used [12]. It is not only grown for home consumption, but also for sale; apart from tubers as food, Anchote seeds and seedlings for propagation are some of the items which are marketed.

Anchote is prepared by heating raw tubers in hot water until it become soft and easier to eat. Then, after peeling, it is eaten with some salt and ground pepper. In most cases, after pealing boiled Anchote, it is chopped and ground and then, mixed with spices and butter to eat it alone or with bread or Injera [13]. Like other roots and tubers, Anchote tubers also contain anti-nutritional composition in varying quantities and should be adequately processed before consumption. The presence of anti-nutritional components in the foods reduces the bioavailability of nutrients and also affects the food qualities [7].For instance, an anti-nutrient, phytate can decreases the bioavailability of critical nutrients such as Zn, Fe, Ca and Mg in foods suchas whole grains, nuts and legumes [14]. At the same time, phytate may have beneficial roles as an antioxidant and anti-carcinogen [15].

Anchote grows in wide environmental conditions from drier to cooler regions in western and southwestern Ethiopia [5]. This makes the crop to be a potential food security crop. However, adequate attention has not been given to improve its productivity, and hence it is remained underutilized crop in Ethiopia. So far, there has been little effort made to undertake varietal development to identify suitable cultivars with different desirable traits adaptable to the different agro-ecological zone of Ethiopia, which makes its use to be limited to specific regions. Ethiopia has 18 agro-ecological zones which are endowed with suitable climatic and edaphically conditions for quality and quantity production of various kinds of root and tuber crops [16]. On other hand, Anchote tubers which grow in different environment are expected to contain various nutritional, anti-nutritional and mineral compositions. To the best of our knowledge, there only few research works had done on nutritional, anti-nutritional and mineral compositions Anchote tubers in our country in general no research done in the selected study areas. Therefore main objective of this research was to determine nutritional, anti-nutritional and mineral compositions of Anchote (Coccinia abyssinica tubers which are grown in Kellem Wollega, West Wollega and Jimma Zones. The obtained nutritional, anti-nutritional and mineral compositions of the crop were also compared with that of sweet potato, potato and cassava crops.

1.2 Statements of Problems

Anchote is nutritious and drought tolerant crop. It is commonly grown in west and southwest Ormia reginal State such as Jimma, Illu Ababa Bora, West and East Wollaga, Horo Guduru Wollega and Kellem Wollega Zones. Although, it is one of the preferred crops in these areas, the research done on nutritional, anti-nutritional and its mineral compositions on these areas are very few. It is also known that nutritional, anti-nutritional and its mineral compositions of crops can vary based on their growing areas/regions. Thus, in this study the nutritional, anti-nutritional and mineral Anchote tubers which were grown in Jimma, West Wollega and Kellem Wollega zones were investigated to answer the following basic research inquiries.

- Do the nutritional, anti-nutritional and mineral compositions of Anchote tubers of the three zones varied or the same?
- Do the nutritional, anti-nutritional and mineral of the Anchote tubers of the three zones are similar or different from the reported values?
- Do the nutritional, anti-nutritional and mineral compositions of Anchote tubers similar or different from other tubers such as sweet potato, potato and cassava crops?

1.3 Objectives of the Study

1.3.1 General Objective.

- ❖ The main objective of this study was to investigate and compare nutritional, anti-nutritional and mineral contents of raw Anchote tubers grown in different west and southwest Ethiopia.

1.3.2 Specific Objectives.

- ❖ To determine moisture content, total ash, crude protein, crude fiber and crude fat of Anchote tubers of commercialized in Jimma city, Gimbi and Dembi Dollo towns, Ethiopia and compare with other tuber crop.
- ❖ To analyze phytate and oxalate contents of Anchote roots tubes commercialized in the three local markets and compare with other tuber crop.
- ❖ To determine the mineral contents such as Ca, Mg, Fe and Zn in Anchote tubers collected from the three areas and compare with other tuber crop.

1.4. Significances of the study

Anchote is particularly important for food security. The crop has also excellent adaptation and growth performance in different agroecologies with high productivity. The findings of this study could enable consumers to know the nutritional, anti-nutritional, and mineral compositions of Anchote tubers which are grown in different parts of the country. The finding could also be used as background information for researchers who want to undergo further research on nutritional composition, anti-nutritional factors and mineral contents of Anchote or other crops.

2. REVIEW OF RELATED LITERATURE

2.1 Origin and Distribution of Anchote

Ethiopia is the country in the world where crop domestication started, and considered as a primary gene centre for several crop plants [17]. Other scientists reported the existence of many cultivated crops in Ethiopia which show considerable genetic diversity [18].At least 7000 vascular plant species occur in Ethiopia, of which 12% are believed to be endemic [19]. It is also stated that crops such as teff *(Eragrostistef(Zucc.) Trotter*), noog *(Guizotiaabyssinica (L.F.) Cass.),* gesho *(Rhamnusprinoides(L"Hér.),* kosso *(Hageniaabyssinica(Bruce) J. F.Gimbiel),* Ethiopian mustard *(Brassica carinata(A.Br.),* enset *(Enseteventricosum(Welw.)Cheesman*), chat *(Catha edulis(Vahl.) Endl.),* Oromo potato *(Plectranthusedulis(Vatke) Agnew),*Anchote *(Coccinia abyssinica(Lam.) Cogn.)* and Coffee (*Coffeaarabica L.)* have great diversity and believed to have originated in Ethiopia [20]. The genus *Coccinia* is made up of 30 speciesof which eight are reported to occur in Ethiopia. The species recorded in flora of Ethiopia since 1995 include *Coccinia abyssinica (Lam.) Cogn*. Adoensis(*Hochst. Ex. A. Rich.) Cogn.),*grandis(L.)Voigh (*Syn. C. indicawight and Arn.),* C. *megarhiza*, C.Jeffrey and C. *schlie benii* Harms. The remaining three species have not yet been described [21].

Anchote is one of several root and tuberous crops (Yam, Taro, Oromo Potato, Irish Potato, Sweet potato and Enset) grown in west and southwestern parts of the highlands[22]. Anchote is cultivated in areas between 1300-2800 m above sea level where the annual rainfall ranged between 762-1016 mm. Ethiopia has 18 agro-ecological zones which are endowed with suitable climatic and edaphic conditions for quality and quantity production of various kinds of root and tuber crops [23].

2.2. *Anchote* Morphology and Production in Ethiopia

Anchote *(Coccinia abyssinica)* is a perennial trailing vine that belongs to family *Cucurbitaceae* which are fruit bearing plants. However, it is the only tuber bearing crop in the genus *Coccinia* and family *Cucurbitaceous*. The genus *Coccinia* comprises 27 species, all of which are confined to sub-Saharan Africa, where it is diversified into various habitat types [24]. A total of eight species of Coccinia is found in Ethiopia [25]. Among these species, C. *abyssinica* is the only species cultivated for its edible tuberous roots and the young shoots used as leaf vegetables [14].

Anchote is a drought tolerant dry land crop with wider ecological adaptation [13]. In Ethiopia, it is commonly cultivated at an elevation that ranges from 1,300 to 2,800 m a.s.l, with average rainfall 762–1,016 mm. The plant seems to have its center of origin and diversity in the western and southwestern parts of Ethiopia. According to Anchote germplasm collection records of the Ethiopian Institute of Biodiversity, the majority of anchote populations were collected from Oromia Regional State mainly from western Ethiopia (Wallega) that has a long history of cultivation and diversified tradition of consumption. Anchote is distributed in the western and southwestern parts of Ethiopia. Particularly, it is widely cultivated and used in Jimma, Illu-Abba-Bora and Wallega areas of the Oromia Regional State [8]. It was also indicated that it occurs in Gondar, Gojam and Bale areas, although its extent of cultivation and utilization was not pointed out [7]. Anchote has spherical a cone-shaped tuber. The shoots have simple tendrils by which it climbs up support. Tubers vary in shape depending on environmental conditions, but generally spherical or elongated at maturity. Anchote is a dioeciously plant; its pollination and fertilization are limited, thereby, reducing its natural regeneration and population.

Anchote is propagated both vegetative and by seeds, the latter is an easy technique and commonly employed by local farmers. Vegetative propagation is performed by planting either the whole tuber or by slicing it into two or more pieces, each piece having rootlets and an external covering [9, 11]. Figure 1 shows anchote plant (A) and tubers (B).

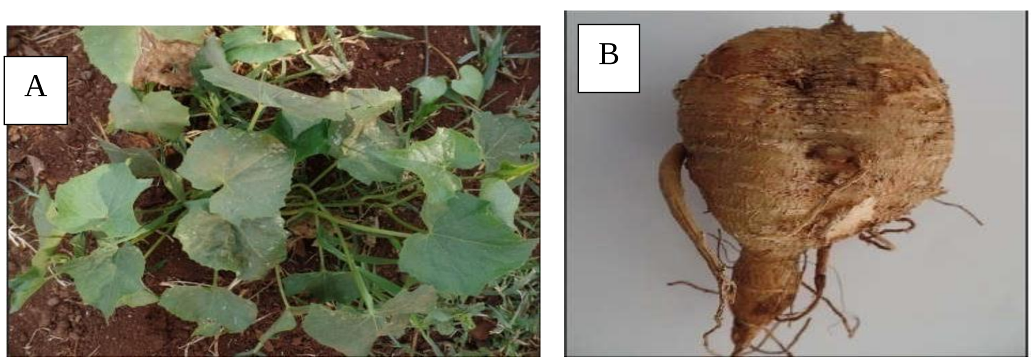

Figure 1: Anchote plant (A) and tuber (B).

2.3 Importance of Anchote

Anchote is endemic root crop of Ethiopia and it is a unique root crop in its uses and the parts consumed. All the three harvestable parts of anchote (i.e. seeds, shoot tips and tubers) are marketable even though the root is the most economic concern in most growing areas of Ethiopia. The consumable parts: root, leaf, and fruit are rich in protein, calcium, iron, and potassium. Few analyses made on the food content of anchote show that anchote is best in protein, utilizable carbohydrate, iron and calcium content as compared to some root crops grown in anchote growing areas and fourth after enset ,sugar beet and potato in energy content. The crop has been contributing much to the diet of the rural societies in its growing areas since its domestication [7]. As its protein, calcium, iron and carbohydrate contents are better than other root crops, it could be an excellent source of macro- and micronutrients. But, its agriculture needs to be modernized and scaled up. As native biological resources are adapted to local conditions, due attention should be given to such crops. Traditional indigenous crops have the potential to diversify and expand the diet of the local societies in particular, and the world, in general.

2.4 Nutritional Composition of *Anchote* and Health Benefits

Anchote is a good source of minerals and fiber content. Its protein content is also by far greater than other root crops, although, root crops are known for their low protein content. The raw Anchote tuber contains organic (carbohydrate, crude protein, crude fiber) and inorganic substances (calcium, magnesium, iron) as well as low levels of antinutrients (Oxalate, tannin, and cyanide) except phytate, when compared to other tuberous crop plants [12]. Anchote is a valuable food source and according to local farmers, it helps in fast mending of broken/ fracture bones and displaced joints, as it contains high calcium, and proteins than other common and wide spread root and tuber crops [4].

Traditionally, it is also believed that Anchote makes lactating mothers healthier and stronger [15]. Root and tuber crops are one of the important sources of minerals that are linked to prevent deficiency diseases such as Anemia and Rickets, and daily consumption of these foods is being encouraged [26]. Anchote is plant endemic in Ethiopia with a high calcium content grown for its edible tuberous roots. Calcium is the major component of bone and assists in tooth development. Calcium concentrations are also necessary for blood coagulation and for the integrity of intracellular cement substances [25]. So a number of edible tubers, roots and corms form an important part of the diet of many people in different parts of the world but their nutrient composition is not fully studied.

3. MATERIALS AND METHODS

3.1 Chemicals and Reagents

Chemicals and reagent used in this study include hydrochloric acid (HCl), Sulpheric acid (H_2SO_4) 98%, Boric acid (H_3BO_4) 4%, Sodium Hydroxide (NaOH) 40%, potassium permanganate ($KMnO_4$) and Nitric acid (HNO_3) 70%, Ferric chloride ($FeCl_3$), and Ammoniumthiocynate (NH_4SCN).

3.2 Instruments and apparatus

Kjeldahlflask, flame Atomic Absorption spectroscopy(Buck scientific Model 210VGPAAS,NORWALK,USA), drying oven (DHG-9070A), fume hood, Quartz digestion tube, Centrifuge, and Muffle furnace(D-6072 Driesch, West Germany) were used in the study.

3.3 The study Area

Anchote samples were collected from Jimma city, Gimbi and Dembi Dollo towns. Jimma city is the capital of Jimma zone south western part of Oromia regional state a at latitude: 7°40'43''N; longitude: 36°50'18''E, 352 km from Addis Ababa,. The altitude of the area is about 1700 m above sea level. The annual rainfall of the area ranges from 100 mm to1500 mm with a mean annual temperature ranging from 24 °C to 28 °C. Dembi Dollo town in the capital of Kellem Wollega zone (latitude: 08°31'60''N; longitude: 34° 47'60''E) which is located in the western part of Oromia regional state, at 624 km from Addis Ababa. The altitude of the area is about 1788 m above sea level. The annual rainfall of the area ranges from 1800mm to 2200 mm with a mean annual temperature range of 20 °C to 25°C.Gimbi town is the capital of west Wollega zone, and it is located in western part of Oromia regional state at latitude: 09°10'13.12''N; longitude: 35°50'5.68''E430 km from Addis Ababa. The altitude of the area is about 1845 to 1930 m above sea level. The annual rainfall ranges from 800 mm to 2000 mm with a mean annual temperature range of 17.8°C to29°C. Figure 2 show map of the study area.

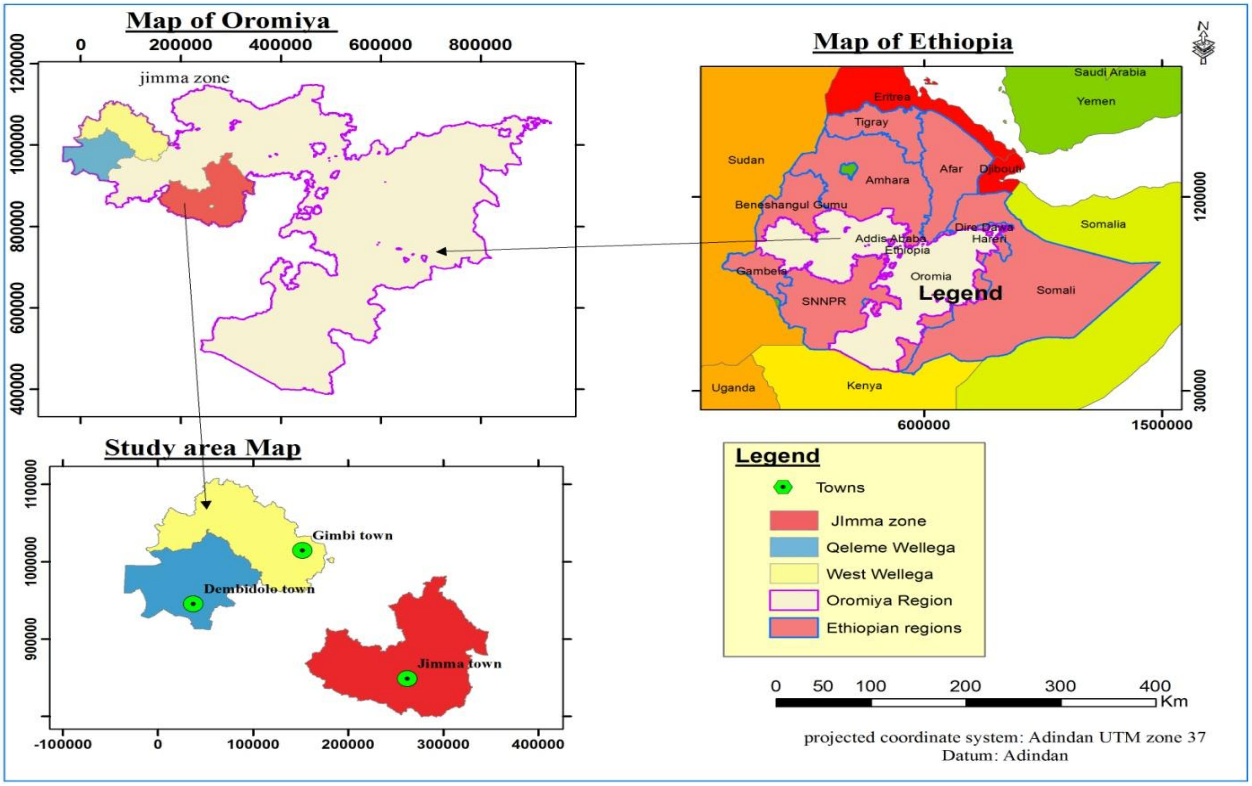

Figure 2: Map of study area

3.4 Sample Collection and Preparation

About 9 kg of the representative Anchote tuber samples, 3 kg samples from each site, were collected from local markets in Jimma city, Dembi Dollo and Gimbi towns, Ethiopia. The samples were collected in new and pre-cleaned polyethylene bags, and then, transported to Jimma University Analytical chemistry laboratory for further analysis. The samples were washed with tap water and the covers were pealed out. They were then chopped /sliced into small pieces. Their moisture contents were determined immediately after they were sliced into pieces. Prior to nutritional, anti-nutrient and mineral content determinations the samples were by dispersing the chopped samples on to plastic pan. The dried samples were then milled into fine powder using coffee grinder. Finally, the samples were stored in dry glass bottles for further analytical study.

3.5. Nutritional Content Analysis

3.5.1. Determination of Moisture Content

Moisture content was determined by AOAC subcomponent 925.09, oven drying method [27].Accordingly, 10 g of well-mixed sample was accurately weighed in dried crucible (W_1). The crucible was placed in an oven at 105 °C for about 6 h until constant weight was obtained. Then the crucible containing the dried sample was taken out from the oven and placed in the desiccators for 30 min until it was cooled. Finally, the sample was weighed (W_2) to determine the percent moisture content of the product by the following formula:

$$\%\text{Moisture} = \frac{W_1 - W_2}{\text{Sample weight}} \times 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots.. (1)$$

Where, $W_1$and W_2 is initial and final weight of the crucible containing the sample, respectively.

3.5.2. Determination of Total Ash Content

A total ash content Anchote sample was determined according to AOAC using a sub component of dry oven method by incineration of known weights of the samples in a muffle furnace at 550 °C until a white ash was obtained [27]. About 2.000 g of Anchote sample (W_S) was added to formerly dried and weighed crucible (W_1). The sample was placed into the Muffle Furnace and ignited at 550 °C for 3 h. The ignited sample, which has white in color, was removed from the furnace and immediately placed in desiccators until it cooled to room temperature. Finally, the crucible containing the ash was weighed (w_2). The %ash of the sample was calculated as follows.

$$\%\text{Ash} = \frac{W_2 - W_1}{W_S} \text{ x } 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (2)$$

Where, W_1, W_2 and W_S are weight of the crucible, weight of the crucible containing the sample after drying and weight of the sample before drying, respectively.

3.5.3. Determination of Crude Protein Contents

The content of crude protein was determined by the AOAC macro-Kjeldahl method, which involve digestion, distillation and titration [28]. Accordingly, about 5.01 g of dried Anchote sample was taken in digestion flask. Then, 15 mL concentrated H_2SO_4 and 8 g of digestion mixture, i.e. K_2SO_4:$CuSO_4$ (8:1) was added. The flask was swirled in order to mix the contents thoroughly, and then placed on heater to start digestion until the mixture become clear (blue green in color), which needed about 2 h. The digest was cooled and transferred to 100 mL volumetric flask and made to the mark by distilled water. A distillation of the digest was performed on Markam Still Distillation Apparatus. 10 mL of digest was introduced into the distillation tube, and then 10 mL 0.5 N NaOH was added. Distillation was continued for at about 10 min and the produced; NH_3 was collected as NH_4OH in a conical flask containing 20 mL of 4% boric acid solution with few drops of modified methyl red indicator. During distillation yellowish color appears due to NH_4OH. The distillate was then titrated against standard 0.2 N HCl solutions until pink color was observed. A blank was also prepared by the same procedure. The percent crude protein content of the sample was calculated by using the following formula:

$$\%\text{Crude protein}=6.25\times\%\text{N} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(3)}$$

$$\%\text{N} = \frac{(\text{S}-\text{B})\times 0.014\times \text{D}\times 100}{\text{Wt.ofsammple}} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{(4)}$$

Where, 0.014 is Millis equivalent weight of nitrogen, S is sample titration reading, B is blank titration reading, N is Normality of HCl, D is dilution of sample after digestion and V is Volume taken for distillation

Sciame1. During digestion step**:** Protein (-N) + H_2SO_4 ⟶ (**N**H_4) $_2SO_4$ + CO_2 + H_2O

Sciame2. During distillation step **:**(NH_4) $_2SO_4$+2NaOH⟶ 2NH_3 (g) +Na_2SO_4+2H_2O

Sciame 1 and 2 show digestion and distillation of anchote root tubes sample.

3.5.4 Determination of Crude Fat Contents

Crude fat content was determined by AOAC subcomponent 920.39 ether extraction method using a Soxhlet apparatus [27].Thus, 2 g dry sample was taken into extraction thimble and then the thimble was placed in previously weighed and petroleum containing beaker. Fat extraction was carried out for 4 h. was water by heating on the water bath. After extraction of fat, the beaker was taken out, dried in hot air oven at about 105ºC for 2 h, cooled in desiccators and weighed. The gain in weight of beakers was calculated as the fat of the Anchote sample [29].

$$\%\text{Crude fat}=\frac{\text{Wt.of ether extracted}\times 100}{\text{Wt.of sample}}\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots..\ (5)$$

3.5.5 Determination of Crude Fiber Contents

Crude fiber content Anchote samples were determined by AOAC method, sub component 962.09 which involve digestion, filtration, washing, drying and combustion [27]. Thus, 0.15 g dried Anchote sample ($W_{\circ}$) was weighed and transferred to a crucible and 150 mL 1.25% hot H_2SO_4 was added. It was heated to boiling for 45 min under condensers. Acid was drained by vacuum suction and the sample was washed with distilled water. Precipitates were again boiled for 45 min with hot 150 mL of 1.25% NaOH. Alkali was drained by vacuum suction and the sample was washed with distilled water. It was dried at 105 ºC to constant weight and recorded as weight after drying (W_1). Ashing was carried out in a muffle furnace at 550 ºC for 3 h. Crucibles were cooled in desiccators and weight recorded as weight after ashes (W_2). Crude fiber was then calculated by the following formula.

$$\%\text{crude fiber}=\frac{W1-W2\times 100}{W0}\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots..\ (6)$$

3.5.6 Determination of Carbohydrates

The carbohydrate content was determined by calculation using the difference method [29]. The mathematical expression is as follows:

%Carbohydrate=100 - (%moisture + %ash + % crude fat + %crude fiber + %crude protein)..(7)

3.5.7 Determination of Total Energy Value

The gross Energy value of Anchote sample was estimated using the Atwater factors [30] for protein (4), fat (9), and carbohydrate (4). The equation is:

$$\%\text{Gross energy} = (\%\text{crude protein x 4}) + (\%\text{fat content x 9}) + (\%\text{carbohydrates x 4}) \ldots (8)$$

3.6 Analysis of Ant-nutritional Factors

Anti-nutritional factors are those substances or chemical compounds found in fruits and food substances in general which are poisonous to humans or in some ways by limiting the nutrient availability to the body. Anti-nutritional factor is present in different food substances in varying amounts depending on the kind of food, mode of its propagation, chemicals used in growing the crops as well as those chemicals used in the storage and preservation of the food substances. Starchy roots and tubers like potato and sweet potato contain anti-nutritional factors like tannin, tryps in inhibitors, phytic acid and oxalic acid. The presence of ant-nutritional factors in the foods (raw or cooked products) reduces the bioavailability of nutrients and also the food qualities [31].

3.6.1 Determination of Oxalate Contents

Oxalates were extracted with acid followed by titrimetric analysis. Which involve sample digestion, filtration and titration [32]. Accordingly, 1 g Anchote sample was added to 75 mL of1.5 N H_2SO_4. The resulting mixture was carefully stirred for 1 h, followed by filtration. The filtrate (25 mL) was then warmed and titrated against 0.1 M $KMnO_4$ solution until a faint pink color was observed, representing the end point of titration. The oxalate content was then estimated using Equation:

$$\text{Oxalate} = (\text{Titer value} \times 0.9004)\ \text{mg/100 g} \ldots\ldots\ldots\ldots (9)$$

3.6.2 Determination of Phytate Content

To determine phytate content, 100 mL 2% HCl was added to 2.5 g ground Anchote sample [33]. The mixture was shaken for 3h. After filtration, 25 mL was taken and 5mL of 0.3% ammonium thiocyanate (NH_4SCN) was added. Then, 50 mL distilled water was added to obtain the desired acidity. There salting solution was titrated against a 0.00195 g/mL $FeCl_3$ solution until a persistent brownish yellow color was observed. The %phytate content was calculated as:

$$\%\text{phytate} = t \times 0.1635 \text{Where t is titrate value} \ldots\ldots\ldots\ldots (10)$$

3.7 Determination of minerals

The standard method of AOAC sub component 923.03 [34] was used for determination of Ca, Fe, Mg, and Zn using a flame Atomic Absorption Spectroscopy (FAAS). Accordingly, 10 g of Anchote sample was weighed into crucible, and placed in a Muffle Furnace at 550 ºC for 3 h. The sample was taken out from the furnace and immediately placed in desiccators until it was cooled to room temperature. Then 1 g ash sample taken into the beaker and12 mL of 1:3 (HNO_3: HCl) was added to completely dissolve the sample and followed by careful dry in at low temperature on hot plate. After addition of 5 mL H_2O_2 the sample was again heated on a hot plate until it just boils, then, it was cooled and filtered through a Whitman no.1 filter paper into a 50 mL volumetric flask. Then, the beaker was washed with water added to the sample in the volumetric flask. Finally, volume was the mark with deionized water and analysed by FAAS. Blank sample were also prepared by same amount procedure

3.8. Method Validation for Metal Analysis

3.8.1. Instrument Calibration

Calibration curves were constructed to determine the concentration of the metals (Ca, Mg, Fe, and Zn) in the sample solutions. Five series of standard solutions of each metal ranging from 1 – 5 mg/L were prepared from stock standard solutions containing 1000 mg/L. Calibration curves were plotted with different points for each metal standard solution using absorbance against concentrations (mg/L). Immediately after calibration using the standard solutions, the sample solutions were put into the AAS instrument and the instrument response becomes recorded.

3.8.2. Limit of Detection

Limit of detection (LOD) is the minimum concentration of analyte that can be detected, but not necessarily quantified with an acceptable uncertainty. LOD for each metal was determined from analysis of five replicates of blank analysis. Blank sample was digested in the same digestion procedure as the actual samples. LOD was determined as $3SD_b$ ($3SD_b$is the standard deviation of the method blank).

3.8.3. Limit of Quantification

The limit of quantification (LOQ) is the lowest concentration of an analyte in a sample which can be quantitatively determined by acceptable uncertainty. Blank sample was digested in the same digestion procedure as the actual samples. LOQ was determined as $10SD_b$.

3.8.4. Precision and Accuracy

Precision and accuracy of the results were assessed by determining repeatability and percent recovery (%R) of spiked sample with known concentration of the analytes. In doing so, 5 samples were spiked with known concentration (i.e., near the mid-range calibration) of the analytes. The spiked samples were digested and analyzed following the same analytical procedure. Precision was reported as SD of replicating analysis. Accuracy of the method was reported as %R, which was calculated as:

$$\%R = \frac{\text{conc.in spiked sample} - \text{conce.unspiked sample}}{\text{actual spiked conce.}} \times 100 \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots.. (11)$$

3.9 Statistical Analysis

In the present study, one-way ANOVA was used to evaluate the variation of results among the samples. Satirical analysis was performed using SPSS software.

4. RESULTS AND DISCUSSION-

4.1 Nutrient Composition of Anchote Tubers

Nutritional value is the main concern when a crop is considered as a food source. Anchote is endemic tuber crop used as a food source in parts of southwestern and western Ethiopia. As shown Table 1, Anchote tubers have good nutrient compositions.

Table 1: Nutritional compositions (Mean ±SD, g/100g) and total energy (kcal) in the studied Anchote samples/

	Current Study			**Reported [26]**	**Report [26]**		
Parameters	**D/Dollo**	**Gimbi**	**Jimma**	**Nekemte**	**Sweet potato**	**Potato**	**Cassava**
Moisture	66.00 ± 2.00	70.33 ± 0.96	73.00 ± 0.82	73.00 ± 0.52	67.43	74.70	62.86
Ash	4.73 ± 0.06	4.43 ± 0.03	4.24 ± 0.06	2.00 ± 0.15	1.10	0.60	0.84
Crude fiber	5.92 ± 0.53	5.67 ± 1.07	5.73 ± 0.20	1.60 ± 0.13	1.10	0.40	1.48
Crude fat	0.14 ± 0.02	0.22 ± 0.01	0.41 ± 0.05	0.17 ± 0.02	2.00	0.10	0.17
Crude protein	1.55 ± 0.04	0.80 ± 0.17	1.17 ± 0.01	3.00 ± 0.14	1.30	1.60	0.53
Total Carbohydrate	21.34 ± 2.01	18.54 ± 1.30	15.44 ± 0.60	22.50 ± 0.45	28.20	22.60	31.0
Total Energy	92.81 ± 7.84	79.38 ± 4.64	70.37 ± 1.73	103.50 ± 0.45	136.00	97.00	NF

NF:Not found

It is known that moisture content determination is an integral part of the proximate composition analysis of food. It is an essential parameter in life maintenance and its analysis is widely used in food processing. It can also affect the processibility, shelf life, usability and quality of food and other products. In this study, the moisture contents of D/Dollo, Gimbi and Jimma Anchote sample were 66 g/100 g,70.33 g/100 g and 73 g/100 g, respectively (Table 1).The obtained results were in agreement with the earlier report [34]. The lowest and highest moisture contents were observed in samples collected from D/Dollo town and Jimma city, respectively. This variation might be attributed to the variation of environmental condition and the variability of the sample collected. Moisture content affects the physical, chemical aspects of food which relates to the freshness and stability for the storage of the food for a long period of time. It also determines the actual quality of the food before consumption and to the subsequent processing in the food sector by the food producers. Moisture rich foods are easily susceptible to the microbial attack and got spoiled and damaged. Low moisture containing foods usually show slower growth of microorganisms [35].

Total ash content is directly proportional to inorganic element content of Anchote. Hence the samples with high ash contents are expected to have high concentrations of various mineral elements, which are advantageous to speed up metabolic processes and improve growth and development [36]. The lowest ash content was observed in Jimma whereas the highest was determined in D/Dollo Anchote samples (Table 1). One way ANOVA ($p < 0.05$) also indicated that there were significant differences in the ash content of the studied Anchote samples. These variations might be related to the soil types, stage of maturity, and agronomic practices.

The food fibers are defined as the sum of non starch polysaccharides (cellulose, hemicelluloses, and pectic substances) and lignins, which are mainly components of plant cell walls. The fiber content of D/Dollo, Gimbi and Jimma Anchote sample were 5.92, 5.67, and 5.73 g/100 g respectively. The obtained fiber contents were higher than the reported values [37, 38]. D/Dollo Anchote sample was shown the highest crude fiber content than Gimbi and Jimma Anchote sample. These variations were probably related to extent storage time and variations in the soils where they were grown. Fibers exhibit beneficial physiological effects to the human body, as they stimulate and accelerate intestinal contraction and transit, and increased feces volume [39].

Proteins are used for growth and replacement of lost tissues in the human body. Comparatively high amount of protein is required for growth. As can be seen from Table 1, the protein content of the Anchote sample collected from D/Dollo has the highest protein content (1.55 g/100 g). This implies that it consists of higher nitrogenous substances than the other samples. The smallest protein content was observed from Gimbi sample; indicating the presence of lower nitrogenous substances in Gimbi Anchote samples. Presence of these differences in the total protein content might be related to the nitrogen content in the soil.

Anchote has low crude fat content. In the current study, the crude fat content of studied Anchote samples were 0.14 g/100 g for D/Dollo; 0.22 g/100 g for Gimbi; and 0.41 for Jimma samples, which were similar to the literature values [37, 38]. Relatively Jimma Anchote sample has exhibited the highest crude fat contents than other samples. The slight differences in the total fat content might be related to the soil types. One way ANOVA ($p < 0.05$) also indicated the presence of significant variations in the content of crude fat among the studied Anchote samples.

Carbohydrates are the main source of energy for our body. Table 1 shows the carbohydrate contents of the Anchote tubers. The carbohydrate contents were 21.34 g/100 g for D/Dollo; 18.54 g/100 g for Gimbi, and 15.44 g/100 g Jimma Anchote samples. The highest carbohydrate content was observed in D/Dollo anchote tubers. The slight differences in the total carbohydrate content might be related to the stage of maturity. The output of this investigation implies that, Anchote is rich carbohydrate content. Thus, consuming Anchote, contain higher carbohydrate content gives more energy [40].

The observed total energy contents were 92.81 kcal/100 g for D/Dollo; 79.38 kcal/100 g for Gimbi and Jimma 70.37 Kcal/100 g Anchote samples . Higher total Energy content was observed in the D/Dollo anchote sample than Jimma and Gimbi sample. The slight differences in the total Energy content might be related to the soil types and stage of maturity. The obtained totally energy is relatively similar to those reported earlier [39]. The total energy values obtained in this study were also exhibited significant differences ($p<0.05$).

4.2 Anti-nutritional Factor Content of Anchote

Anti-nutritional factors are substances, which by themselves, or through their metabolic products arising in living systems, interfere with food utilization and affect the health and production of animals. Anti-nutrients are known to reduce the maximum utilization of nutrients, especially proteins, vitamins, and minerals [36].Some anti-nutritional factors (phytate and oxalate) content of the Anchote tuber is indicated in Table 2.

Table 2: Anti-nutritional factor content (Mean ± SD, n =3) of Anchote roots sample.

Parameter	**Current Study**			**Reported [26]**
	D/Dollo	**Gimbi**	**Jimma**	**Nekemte**
Oxalates content (mg/100g)	0.81±0.02	0.62±0.02	0.26±0.01	7.10 ± 1.20
Phytate content (mg/100g)	0.18±0.02	0.12±0.01	0.07±0.01	0. 20 ±10. 1

Oxalic acid inhibits the absorption of calcium by forming insoluble calcium oxalate [40]. But there may be also the possibility of bacterial degradation which may occur in the intestine, making the calcium available from calcium oxalate [40]. In this study, the observed oxalate contents were D/Dollo for 0.81 mg/100 g, 0.62 mg/100gfor Gimbi, and 0.26 mg/100g for Jimma Anchote tuber sample (Table 2). The oxalate contents of the three sites were also significantly different ($p < 0.05$). The observed differences might be related to the soil types and stage of maturity of Anchote tubers.

Phytate is a salt form of phytic acid. Phytic acid is strong to form protein and mineral-phytic acid complexes chelate; resulting in the net reduction of bio available protein and mineral [41]. The major concern about the presence of phytate in the diet is its negative effect on mineral uptake the phytate contents were 0.18 mg/100 g for D/Dollo, 0.12 mg/100 g for Gimbi, and 0.07 mg/100 g for Jimma Anchote samples. The highest phytate content was observed in D/Dollo Anchote sample, indicating necessity of processing to remove phytate before consuming.

4.3. Mineral Contents of Anchote Tubers

4.3.1 Analytical Performance Study

Before determination of mineral contents including Ca, Mg, Fe and Zn in the Anchote samples, calibration curves were constructed using five concentration points ranging from 1/0 – 5 mg/L. The obtained curves linear dynamic ranges (LDR), coefficient of determination (R^2), LOD and LOQ are presented in Table 3

Table 3: Analytical performance of the method.

Metal	LDR	LOD	LOQ	R^2
Ca	1.0 -5.0	0.08	0.27	0.998
Mg	1.0 - 5.0	0.14	0.47	0.999
Fe	1.0 -5.0	0.18	0.60	0.997
Zn	1.0 -5.0	0.21	0.71	0.998

As can be seen calibration curve of each element exhibited satisfactory R^2, i.e., 0.998 or better, low LOD for analysis of the metals.

4.3.2 Recovery Study

Spiking of known concentration standard is usually used for recovery study in the absence of certified reference material (CRM). The obtained %Rare presented in Table 4.It was observed that the %R of the studied elements ranging from 87.49 – 98.19 %, which are within the acceptable range for metal analysis, indicating good accuracy of the method.

Table.4: Recovery study of the spiked Anchote sample.

Metal	Conc. in sample (mg/L)	Spiked conc. (mg/L)	Conc. in spiked (mg/L	Recovery (%)
Ca	12.79	2	14.54	87.47
Mg	7.37	2	9.24	93.00
Fe	7.90	2	9.77	93.36
Zn	3.94	2	5.90	98.19

4.3.3 Metal Concentration of Anchote Samples

Minerals in the diet are responsible for several existing problems relating to human health. The human body requires more than 22 mineral elements that can be supplied in a diet in varying amounts for proper growth, health maintenance, and general wellbeing [42]. Root and tuber crops are important sources of minerals that are linked to prevent deficiency diseases such as anemia and rickets, and daily consumption of these foods is being encouraged [42]. The mineral contents of the Anchote are presented in (Table 5).

Table 5: Mineral content (Mean ± SD, mg/100 g) of Anchote sample.

Metal	Current Study				Reported [26]		
	D/Dollo	Gimbi	Jimma	Nekemte	Sweet potato	Potato	Cassava
Ca	114.95 ± 1.29	49.55 ± 0.07	64.00 ± 0.13	344.00 ± 13.00	52	10	20
Mg	16.35 ± 0.38	21.10± 0.01	19.65 ± 0.50	80.00 ± 4.00	NF	NF	30
Fe	6.80 ± 0.02	34.95 ± 0.13	36.90 ± 0.02	5.50 ± 0.40	3.40	6.7	0.23
Zn	12.70 ± 0.12	11.25 ± 0.12	9.10 ± 0.02	1.80 ± 0.20	NF	NF	NF

NF: Not found

It is known that Ca is the major component of bone and teeth. It is also necessary for blood coagulation and for the integrity of intracellular cement substances [43]. Anchote may also play a vital role in supplying the Ca requirements for infants and children, in place where milk and milk products are not easy to obtain. The obtained Ca contents were114.95 mg/100 g for D/Dollo, 49.95 mg/100g for Gimbi and 64.00 mg/100g for Jimma Anchote tubers. The lowest and highest Ca contents were detected in Gimbi and D/Dollo Anchote samples respectively. Such variations in Ca contents might be related to the pH of soil, availability of Ca bearing minerals in soil and water. Usually ground and surface waters are rich in Ca, which can easily be absorbed by the plant.

Fe is essential in the body; it is mainly used to transport oxygen to blood cells and thus about two-thirds of the body Fe is found in hemoglobin. It is also used to regulate proper growth of human body, to maintaining robust health and to produce red blood cells. But, at very high concentration it causes vomiting, abdominal pain and liver enlargement. In this study, the determined Fe contents were 6.80 mg/100 g in D/Dollo, 34.95 mg/100 g in Gimbi and 36.9 mg/100 g in Jimma Anchote samples. Anchote sample which was collected from Jimma contained higher concentrations of Fe than D/Dollo and Gimbi Anchote tubers.

The recommended Mg daily intake in most countries is 128 mg for children 1-2 years, 280 mg for women and 350 mg for men [44]. The Mg content of D/Dollo, Gimbi and Jimma Anchote tubers were 16.35 mg/100 g, 21.1 mg/100 g and 19.65 mg/100 g, respectively. Gimbi Anchote sample had the highest concentration of Mg than D/Dollo and Jimma samples. The differences in the Mg contents might be related to the soil types and its mobile in plants. Mg is highly mobile in the plant tissue and translated from old plant tissue to new plant tissue.

Zn is an essential element for human, animal and certain types of plant. It is necessary for a healthy immune system, cell division and synthesis of protein and collagen, which is great for wound healing and healthy skin. However, a higher amount of it can cause anemia, pancreas damage and in production of high density lipoprotein cholesterol [45]. In this study, the contents of Zn were12.70 mg/100 g in D/Dollo, 11.25 mg/100 g in Gimbi and 9.10 mg/100 g in Jimma Anchote tubers. The highest Zn content was detected in D/Dollo Anchote sample. The obtained

Zn contents in all Anchote samples were higher than the reported values. This variation might be related to the nature of the soil, because Zn is readily available in acidic soils. The recommended daily intake for Zn in most countries is 3.9 mg for infants and 7.4 mg for children [46]. The obtained Zn contents were significantly different ($p<0.05$) among the studied samples.

Compared to other tuber crops such sweet potatoes, potato and cassava (Table 5), Anchote is rich in Ca, Fe, and Zn and Mg contents, indicating its high potential for food security.

5. CONCLUSION AND RECOMMENDATION

5.1 Conclusion

In the present study, the nutritional anti-nutritional and mineral compositions of Anchote tubers grown in D Dollo, Gimbi, and Jimma were studied. The finding demonstrated that Anchote samples collected from D/Dollo town content relatively the highest Ash, crude protein, crude fiber, carbohydrate, and Total Energy and Zn than the other samples. D/Dollo anchote sample also contain the highest concentration of phytate and oxalate. Crude Fat, moisture, and Iron content were highest in the Jimma Anchote sample, and the highest concentration of Mg was detected in Gimbi Anchote sample. The finding also has shown as D/Dollo Anchote tubers needs processing such as long boiling, fermentation and soaking before consummation to remove the anti-nutrients. In comparison to other tropical root and tuber crops (cassava, potato and sweet potato) Anchote have high nutritional and mineral components. In general, the finding revealed that regardless of their growing areas, Anchote tubers contain appreciable amounts of nutrients like crude protein, fiber, calcium, iron, and zinc; and the low levels of oxalate and phytate. Therefore, increasing its production and consumption is helpful to supplement/formulate diets and alleviate the problems associated with malnutrition in the country.

5.2 Recommendations

Based on the findings the researched would like to forward the following recommendations.

- To obtain detailed scientific evidences about the variation of nutritional, Antinutritional and mineral contents of Anchote, further study is required by incorporating the soils, and maturity level of Anchote tubers.
- Cultivation and consumption of Anchote needs to be encouraged in all part of the country, because of its high nutrient contents like protein, crude fiber, Ca, Zn, Mg and low anti-nutritional factors.
- Agricultural research institutions and other concerned bodies should give great attention on the genetic modification and productivity of the crop to sustain food scarcity.

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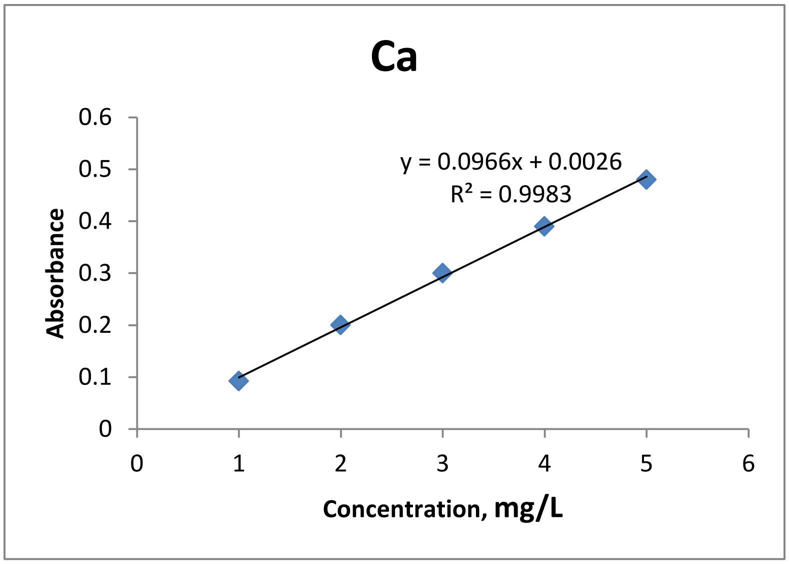
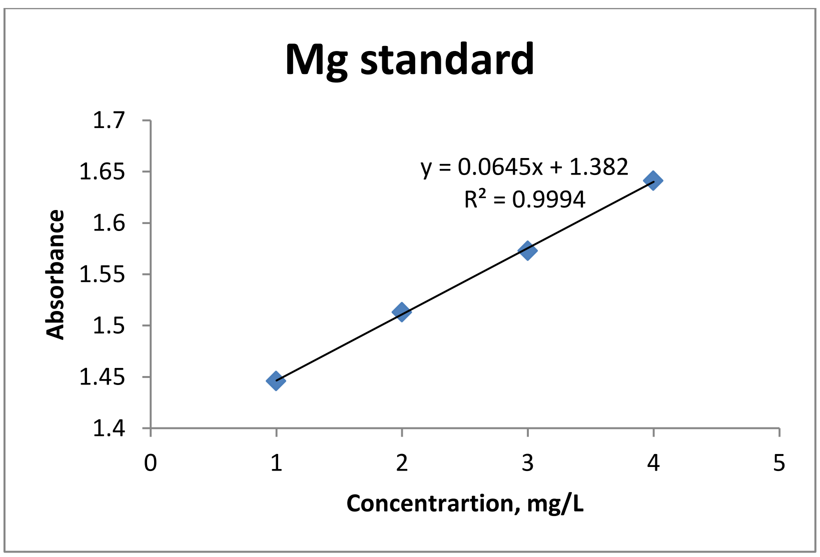
Appendix

Calibration curves


Ca
0.6
0.5
0.4
0.3
0.2
0.1
0
Absorbance
y = 0.0966x + 0.0026
R² = 0.9983
0
1
2
3
4
5
6
Concentration, mg/L

Ca cal. curve


Mg standard
1.7
1.65
1.6
1.55
1.5
1.45
1.4
Absorbance
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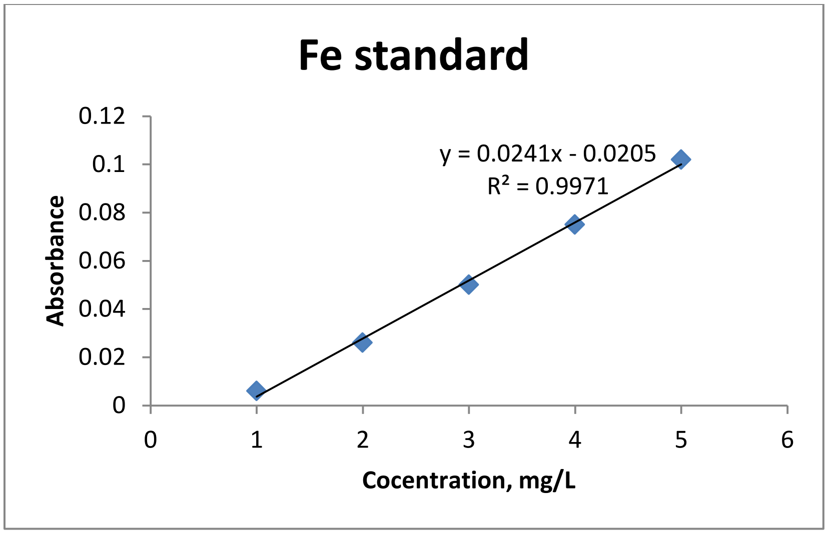
R² = 0.9994
0
1
2
3
4
5
Concentrartion, mg/L


Fe standard
y = 0.0241x - 0.0205
R² = 0.9971
Absorbance
Cocentration, mg/L
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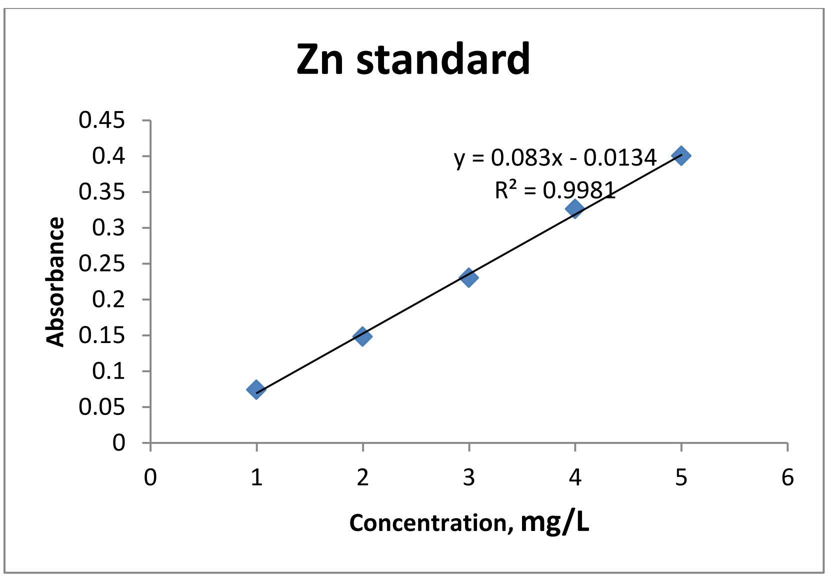
0.1
0.08
0.06
0.04
0.02
0
0 1 2 3 4 5 6


Zn standard
y = 0.083x - 0.0134
R² = 0.9981
Absorbance
Concentration, mg/L
0.45
0.4
0.35
0.3
0.25
0.2
0.15
0.1
0.05
0
0 1 2 3 4 5 6

ANOVA

Moisture

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	74.889	2	37.444	18.965	.003
Within Groups	11.847	6	1.974		
Total	86.736	8			

ANOVA

Ash

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.357	2	.179	56.404	.000
Within Groups	.019	6	.003		
Total	.376	8			

ANOVA

Protein

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.821	2	.411	42.051	.000
Within Groups	.059	6	.010		
Total	.880	8			

ANOVA

Fiber

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4.471	2	2.235	.879	.462
Within Groups	15.252	6	2.542		
Total	19.723	8			

ANOVA

Fatt

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	7.179	2	3.590	1.240	.354
Within Groups	17.375	6	2.896		
Total	24.554	8			

ANOVA

Energy

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3212.881	2	1606.440	2.738	.143
Within Groups	3520.767	6	586.794		
Total	6733.647	8			

ANOVA

carbohydrate

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	376.862	2	188.431	.615	.572
Within Groups	1839.015	6	306.502		
Total	2215.877	8			

ANOVA

Calcium

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	274.390	2	137.195	148.839	.000
Within Groups	5.531	6	.922		
Total	279.921	8			

ANOVA

Magnicium

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	96.805	2	48.402	15.481	.004
Within Groups	18.759	6	3.126		
Total	115.564	8			

ANOVA

Zink

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.166	2	.083	.163	.854
Within Groups	3.069	6	.512		
Total	3.235	8			

ANOVA

Phytate

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.507	2	.253	.753	.511
Within Groups	2.020	6	.337		
Total	2.526	8			

ANOVA

Oxalate

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.598	2	.299	66.656	.000
Within Groups	.027	6	.004		
Total	.625	8			