

Biochemical and Mineral Composition of Anchote (*Coccinia abyssinica* (Lam.) Cogn.) Accessions from Ethiopia

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Abstract

Anchote (*Coccinia abyssinica* (Lam.) Cogn.) is among few indigenous crops in Ethiopia that has a great potential to be a food and nutritional security and socioeconomically important crop, so far given lower attention in research and development in Ethiopian national research system considering it as a minor crop. In order to fill the knowledge gaps about the nutritional potentials of this crop, the present study was conducted to assess the biochemical composition and anti nutritional profile of 100 anchote accessions collected from western and southwestern parts of Ethiopia. Flour from storage roots of 100 anchote accessions were collected and the samples run in three replications. Data on 20 biochemical, nutritional and root flesh color traits were generated in laboratory and subjected to analysis. Analysis of variance indicated significant variation ($p \leq 0.01$) among the accessions for the 20 traits and root flesh color. The calcium contents ranged from 1.45 to 6.79 mg/g with a mean of 4.16mg/g. The ranges and means of iron (0.041 to 1.12 and 0.15), zinc (0.02 to 0.47 and 0.06), dry matter (%) range and mean (7.71 to 13.73 and 11.79), crude fiber (0.2 to 8.07% and 3.55%), protein (7.21 to 15.93% and 11.81%), carbohydrate (66.56 to 78.14% and 72.13%) and total energy (3.24 to 3.54 kcal/g and 3.34 kcal/g dry matter). The cluster analysis of biochemical traits and root flesh color showed the existence of six divergent groups. According to PCA analysis all the accessions were grouped into seven components based on root flesh color, and biochemical traits of which flesh color, Ca, K, Mg, S, P and Na contents were significant (eigenvalue > 1) and explained 78 % of the total variability. The total genetic variability explained was 84.30% and some traits were negatively correlated; root flesh color, Ca, B, Mn, Zn, crude protein, and crude fiber and the remaining mineral and biochemical compositions correlated positively. The variation exhibited in this experiment could be attributed to environmental and genetic factors. Owing to the potential of the plant nutritionally, further identification of its active compounds is recommended.

Keywords: Anchote, accession, root crop, biochemical, Ethiopia

Introduction

In many developing countries, food insecure people and subsistence farmers depend highly on root and tuber crops as food source and possibly principal source of nutrition, and cash income (Scott *et al.*, 2000). For millions of people in the tropical humid regions of Africa root and tuber crops occupy a position of prestige among the staple foods (Lebot, 2009). Root and tuber crops provide an estimated average of 20% of the daily per capita calorie intake for the 640 million inhabitants of sub-Saharan Africa, where with the growing population there is increasing demand for these crops both for food and feed (Kenyon *et al.*, 2006).

In Ethiopia, root and tuber crops have a great contribution as part of the traditional food system and income generation specifically in Southern, South Western and Western part of Ethiopia (Andargachew *et al.*, 2011; Fantaw *et al.*, 2014). Root and tuber crops are mainly used as food security crops during food shortage since they are drought-tolerant and high yielding (Wheatly, Scoot, and Wiersema, 1995).

Several million people rely on enset (*Ensete ventricosum*) as a staple or co-staple food; as many as 7 to 10 million (Alemayehu *et al.* 2011; Pijls *et al.*, 1995) and other minor root and tuber crops such as Anchote (*Coccinia abyssinica*), Taro (*Colocasia*

esculenta), Yam (*Dioscorea alata*), Cassava (*Manihot esculenta*) and 'Oromo Dinich' (*Plectranthus edulis*) are also important in some areas, mainly in South and South Western Ethiopia (Abdissa 2000; FAO 2007). Although root and tuber crops are drought resistant and food security crops in drought prone areas, their potential as a human food and medicine has not yet been fully recognized and utilized (Gebremedhin, *et al.*, 2008).

Anchote (*Coccinia abyssinica* (Lam.) Cogn.) is one of the tuber crops belongs to the *cucurbitaceae* family and *conccinia* genus having over 30 species, about eight of which are believed to occur in Ethiopia (Desta *et al.*, 2011; Bekele *et al.*, 2013; Yambo and Feyissa, 2013; Yassin *et al.*, 2013). Despite its food and nutritional security and other functional potentials, anchote has been given lower attention in research and development, and not well developed and popularized (Habtamu *et al.*, 2013).

Anchote [*Coccinea abyssinica* (Lam.) Cogn.] is an endemic root and tuber crop of Ethiopia cultivated for human consumption (Abera, 1995; Beruk and Fikre, 2015; Habtamu, 2014; Tilahun *et al.*, 2014). Anchote is a vine like cucurbit with a high yield and short crop cycle used as an important dietary and medicinal plant, grown widely in the Western and South-Western parts of the Ethiopia, encompassing about

115 genera and 960 species better known for its tuberous root and tender leaves for food (Schaefer *et al.*, 2009; Girma and Hailu 2007; Yassin *et al.*, 2013 and Abera, 1995).

Studies indicate that Anchote has relatively higher crude protein, utilizable carbohydrate, crude fiber, energy, and ash content as compared to sweet potato, potato and cassava (Habtamu *et al.*, 2013). The calcium content of Anchote is also reportedly high (Desta, 2011; Habtamu *et al.*, 2013). Traditionally, Anchote is consumed to heal broken or fractured bones which could be attributed to its high calcium content (Abera, 1995; Habtamu and Kelbessa, 1997).

Anchote has a significant contribution for the cultural and social values of the Oromo's (the largest tribe in Ethiopia) since long ago (Abera, 1995; Desta *et al.*, 2011; Daba *et al.*, 2012). The plant has been grown over a wide range of environments for a long time, and its cultivation and utilization have been passed from generation to generation through oral tradition, with very little recorded information mainly by women farmers (Abera, 1995; Girma and Hailu, 2007). The unique characteristics of the plant is the edibility of its different parts such as its tuber, leaf, and fruit which makes the plant ideal as potential food security crop (Amare 1973; Endashaw 2007; Desta *et al.*, 2011). The root yield ranges of anchote varies as reported by different authors; 42 - 76 t/ha (Daba *et al.*, 2012), 10-20t/ha

(Weyesa *et al.*, 2008), 22.71 - 130.83t/ha (Desta *et al.*, 2011)

The high prevalence of malnutrition and persistent food insecurity in Ethiopia is due to the highly selective and restricted food consumption habit of the population as well as less exposure to the important wild and indigenous food plants (Getachew, 2001). According to Dandena (2010), malnutrition in Ethiopia still exists due to poor dietary habit regardless of the favorable highly diversified agro-ecological conditions of the country, which are suitable for the production of various types of fruit and vegetables.

There are a number of studies on genotypes (Abreham, Tileye, and Kassahun, 2014; Tilahun *et al.*, 2014) and agronomic characteristics (Girma and Hailu 2007; Daba *et al.*, 2012; Folla *et al.*, 2013; Yambo and Feyissa 2013; Yassin *et al.*, 2013) of Anchote but limited studies on its nutritional value. The few existing studies indicate its potential for human nutrition and medicine (Desta *et al.*, 2011; Habtamu *et al.*, 2013; Habtamu, 2014; Habtamu and Kelbessa, 1997). For instance, the protein content of Anchote tuber ranges from 4.6-16.4% (Desta *et al.*, 2011) which is high and with wide range compared to other root and tuber crops commonly consumed in Ethiopia, with protein values ranging from 1-2% (Gebremedhin *et al.*, 2008). Hence, understanding of the nutritional importance of root crops particularly anchote has potentially far-reaching

implication in solving the nutritional need of children and women and for further research and development. Therefore, the objective of this study was to characterize genetic diversity in terms of biochemical and nutritional composition of anchote accessions for further breeding, conservation and utilization.

Materials and Methods

Description of Sampling Site

A total of 100 anchote accessions collected from major anchote growing areas of west and south western parts of Ethiopia; East and West Wollega, Horro Guduru Wollega, Jimma and Buno Bedelle zones of Oromia region having altitudes ranging from 1427 to 3025 m.a.s.l (Table 1 and Figure 1) and were planted at Debre Zeit Agricultural Research Center experimental field , located at 08°44'N latitude and longitude of 38°58' E with 1860 m.a.s.l. elevation, mean annual rainfall and temperature of 851mm and 19 °C, respectively. The major soil type in the area is vertisols and alfisols/mollisols and the experiment

was conducted on vertisols in 2017 and 2018 under irrigated system.

Sampling and sample preparation

Five anchote tuber samples per accession were randomly selected per replications, replicated three times, were prepared for laboratory analysis. All laboratory procedures; washing, peeling, slicing to small pieces and mixing thoroughly in order to prepare 400g of each samples and put them in a paper bag and dried to a constant weight in a hot air oven (DHG-9055A, Memment, Germany) at 105 °C for 24 hours. The oven dried anchote tuber chips, were milled using an electric grinder to obtain fine powder of anchote flour. The flour was sieved through in 1 mm sieve, measured and packed into air tight plastic bag and stored in refrigerator until it was used for analysis. Mineral and proximate analysis was conducted at Horticoop Ethiopia (Horticultural) PLC, Soil, Plant and Water Analysis Laboratory and the National Veterinary Institute (NVI), at Bishoftu.

Table 1. List of 100 anchote accessions with areas of collection

Accessions	Zone*	Woreda	Kebele	Longitude	Latitude	Altitude
002/09	E.W	Sibu Sire	Burka Talo	9°01'49.00"N	36°50'52.04"E	1791
003/09	E.W	Wayu Tuka	Warra Migna Babbo	9°03'52.98"N	36°52'05.77"E	1889
004/09	E.W	Wayu Tuka	Warra Migna Babbo	9°03'37.54"N	36°52'34.88"E	1840
005/09	E.W	Wayu Tuka	Warra Migna Babbo	9°03'47.65"N	36°52'56.35"E	1877
008/09	E.W	Wayu Tuka	Kura Migna	9°02'15.02"N	36°51'10.74"E	1830
009/09	E.W	Chingi	Maja Ale	9°02'43.31"N	36°43'10.25"E	1823
010/09	E.W	Wayu Tuka	Gute	9°02'04.84"N	36°40'22.64"E	1884
011/09	E.W	Chingi	Gobu	9°02'55.33"N	36°43'18.10"E	1843
014/09	E.W	Chingi	Gordommo	9°03'06.28"N	36°42'10.24"E	1935
016/09	E.W	Chingi	Jarte/Birbo Gibbi	9°01'55.46"N	36°40'40.97"E	1857
024/09	E.W	Leka Dullecha	Horda Qawusa	9°01'47.65"N	36°25'39.68"E	2110
029/09	E.W	Leka Dullecha	Horda Qawusa	9°01'46.27"N	36°25'06.82"E	2103
032/09	E.W	Leka Dullecha	Horda Qawusa	9°02'01.91"N	36°25'20.10"E	2056
040/09	E.W	Leka Dullecha	Horda Qawusa	8°59'55.44"N	36°30'31.60"E	2209
041/09	E.W	Leka Dullecha	Horda Qawusa	8°59'21.87"N	36°30'25.81"E	2227
043/09	W.W	Gimbi	Jogir	9°10'10.87"N	35°46'31.44"E	1727
049/09	W.W	Gimbi	Chuta Goch	9°12'06.03"N	35°44'03.15"E	1870
050/09	W.W	Gimbi	Inango Dambali	9°10'08.88"N	35°41'45.15"E	1888
051/09	E.W	Digga	Demeksa	9°02'26.20"N	36°27'17.84"E	2199
053/09	W.W	Gimbi	Aba Sena	9°02'00.08"N	35°58'42.00"E	1637
056/09	W.W	Gimbi	Choli	9°12'55.57"N	35°49'27.30"E	1864
058/09	W.W	Gimbi	Lalisa yesus	9°24'43.28"N	35°35'50.16"E	1892
062/09	W.W	Ialo asabi	Haroggi Harowwa	9°27'57.57"N	35°32'26.10"E	1798
065/09	W.W	Mana Sibu Mandi	Gombo Kiltu Jale	9°50'50.05"N	35°04'29.09"E	1753
066/09	W.W	Mana Sibu Mandi	Gombo Kiltu Jale	9°50'45.38"N	35°03'35.58"E	1651
067/09	W.W	Mana Sibu Mandi	Gombo Kiltu Jale	9°50'16.38"N	35°02'38.49"E	1616
075/09	E.W	Gobbu Sayyo	Tibbe Hara	9°05'11.09"N	36°21'50.67"E	1441
080/09	E.W	Leka Dullecha	Shakko	8°57'26.16"N	36°32'30.67"E	1986
081/09	E.W	Nunnu Kumba	Kumba	8°46'10.08"N	36°37'51.14"E	2313
087/09	E.W	Leka Dullecha	Bal'o	8°57'53.82"N	36°27'57.20"E	2248
089/09	E.W	Sibu Sire	Home Baro	9°02'12.41"N	36°53'05.23"E	1842
090/09	E.W	Sibu Sire	Burka Talo	9°01'52.57"N	36°51'01.05"E	1800
091/09	E.W	Bonaya Boshe	Ejersa Gute	8°57'33.64"N	36°39'52.77"E	1759
092/09	E.W	Gudeya Bila	Gonka Ija	9°14'53.50"N	36°57'56.26"E	1943

093/09	E.W	Gudeya Bila	Gonka Ija	9°14'55.42"N	36°57'40.82"E	1934
094/09	E.W	Gudeya Bila	Gonka Ija	9°14'48.15"N	36°57'27.68"E	1949
095/09	E.W	Gudeya Bila	Kalala	9°15'49.79"N	36°59'41.92"E	1989
096/09	E.W	Gudeya Bila	Gonka Ija	9°14'41.02"N	37°00'26.13"E	1910
097/09	E.W	Gudeya Bila	Gonka Ija	9°15'01.93"N	37°00'31.92"E	1943
112/09	W.W	Gimbi	Kombo Mikael	9°05'36.96"N	35°48'32.01"E	1938
113/09	W.W	Gimbi	Kombo Mikael	9°05'20.43"N	35°48'38.52"E	1911
126/09	W.W	Gimbi	Garjo Bikilal	9°14'02.64"N	35°55'19.48"E	1729
131/09	W.W	Gimbi	Garjo Bikilal	9°14'34.03"N	35°54'11.09"E	1704
132/09	W.W	Gimbi	Garjo Bikilal	9°14'57.66"N	35°54'30.61"E	1661
134/09	W.W	Gimbi	Garjo Bikilal	9°12'46.20"N	35°54'39.48"E	1842
140/09	W.W	Gimbi	Garjo Bikilal	9°16'40.84"N	35°51'39.96"E	2052
148/09	W.W	Gimbi	Lalo Choli	9°14'13.95"N	35°48'38.07"E	1793
151/09	W.W	Gimbi	Lalo Choli	9°13'41.94"N	35°48'02.11"E	1838
153/09	W.W	Gimbi	Lalo Choli	9°13'02.91"N	35°48'13.15"E	1827
155/09	W.W	Gimbi	Lalo Choli	9°13'02.03"N	35°47'55.28"E	1814
156/09	W.W	Gimbi	Lalo Choli	9°13'29.23"N	35°47'57.03"E	1845
157/09	W.W	Gimbi	Lalo Choli	9°13'16.14"N	35°48'16.95"E	1846
158/09	W.W	Gimbi	Lalo Choli	9°13'40.52"N	35°48'15.06"E	1826
161/09	W.W	Gimbi	Lalo Choli	9°12'59.74"N	35°47'27.49"E	1811
169/09	W.W	Gimbi	Didisa Bikilal	9°16'07.72"N	35°44'03.88"E	1839
183/09	W.W	Gimbi	Didisa Bikilal	9°17'48.43"N	35°44'29.76"E	1766
206/09	W.W	Mana Sibumandi	Wajitu Mandi	9°48'07.68"N	35°04'40.67"E	1610
209/09	W.W	Mana Sibumandi	Wajitu Mandi 01	9°48'22.76"N	35°04'32.51"E	1610
211/09	W.W	Mana Sibumandi	Wajitu Kiltu Lubo	9°45'36.34"N	35°05'34.92"E	1622
214/09	W.W	Mana Sibumandi	Wajitu Kiltu Lubo	9°45'56.74"N	35°00'29.85"E	1543
218/09	W.W	Boji Dirmaji	Gumbo Boji	9°23'14.75"N	35°36'03.09"E	2004
219/09	W.W	Boji Dirmaji	Lata Bobine	9°23'23.60"N	35°35'57.02"E	2000
222/09	H.G.W	Horro	Doyyo Bariso	9°36'09.82"N	37°11'33.91"E	2399
223/09	H.G.W	Horro	Doyyo Bariso	9°36'12.85"N	37°11'33.27"E	2392
227/09	W.W	Boji Dirmaji	Lata Bobine	9°23'47.77"N	35°35'42.86"E	1964
256/09	Jimma	Dedo	Dedo	7°30'11.60"N	36°52'12.49"E	2294
262/09	E.W	Digga Leqa	Digga	9°02'25.58"N	36°29'03.58"E	2214
271/09	E.W	Gudeya Bila	Hena Jawaja	9°14'09.72"N	37°01'31.09"E	1883
280/09	W.W	Kiltu Kara	Dandi Gudi	9°32'04.39"N	35°22'30.93"E	1817
281/09	W.W	Kiltu Kara	Dandi Gudi	9°32'09.40"N	35°22'09.30"E	1793

286/09	W.W	Boji Dirmaji	Lata Bobine	9°24'20.62"N	35°36'18.70"E	1918
288/09	W.W	Najo	Humna Wakayyo	9°30'32.40"N	35°31'10.33"E	1901
290/09	W.W	Boji Dirmaji	Lata Bobine	9°27'13.65"N	35°33'15.48"E	1853
291/09	W.W	Boji Dirmaji	Gumbo Boji	9°28'42.70"N	35°32'21.80"E	1805
292/09	W.W	Najo	Humna Wakayyo	9°31'20.80"N	35°31'32.35"E	1914
296/09	W.W	Najo	Humna Wakayyo	9°31'09.60"N	35°31'28.18"E	1913
297/09	W.W	Najo	Humna Wakayyo	9°31'36.07"N	35°31'31.17"E	1890
300/09	W.W	Mana Sibumandi	Guyo Sachi	9°50'29.80"N	35°03'50.10"E	1655
302/09	E.W	Limmu	Sakata Kiltu Babbo	9°51'04.95"N	36°29'27.52"E	2181
304/09	E.W	Limmu	Sapera	9°51'49.78"N	36°31'30.58"E	2192
305/09	E.W	Limmu	Degem Silassie	9°51'46.82"N	36°30'46.08"E	2133
308/09	E.W	Limmu	Sapera	9°51'34.09"N	36°29'36.91"E	2115
309/09	E.W	Limmu	Muka Arba Dima	9°51'29.62"N	36°29'03.18"E	2149
313/09	E.W	Gida Ayana	Gute Gudina	9°53'13.79"N	36°37'13.93"E	2060
316/09	E.W	Gida Ayana	Gaba Jimata	9°53'07.17"N	36°39'51.96"E	2098
328/09	E.W	Leka Dullecha	Badh'o	8°55'28.47"N	36°35'39.29"E	1855
376/09	B.B	Didessa	Sasso	8°37'45.79"N	36°22'04.73"E	1459
377/09	B.B	Didessa	Sasso	8°37'46.82"N	36°22'02.86"E	1463
381/09	B.B	Didessa	Yembero	8°37'51.56"N	36°21'28.53"E	1427
385/09	H.G.W	Jimma Geneti	Bikila Nagaro	9°25'53.98"N	37°04'20.75"E	2945
388/09	H.G.W	Jimma Geneti	Gidami Dabsho	9°24'25.81"N	37°04'13.78"E	2703
393/09	E.W	Gudeya Bila	Lanfaji	9°18'10.48"N	37°02'19.92"E	2063
394/09	E.W	Gudeya Bila	Lanfaji	9°18'02.31"N	37°02'51.32"E	2060
397/09	E.W	Gudeya Bila	Walane Lemu	9°20'53.47"N	37°02'16.31"E	2289
400/09	E.W	Gudeya Bila	Walane Lemu	9°20'14.80"N	37°02'55.30"E	2238
402/09	E.W	Gudeya Bila	Walane Lemu	9°20'10.43"N	37°02'11.83"E	2252
406/09	E.W	Gudeya Bila	Walane Lemu	9°19'55.93"N	37°02'15.88"E	2216
407/09	H.G.W	Jimma Geneti	Gamo Nagaro	9°22'45.81"N	37°04'18.96"E	2509
413/09	E.W	Gudeya Bila	Gute Chacho	9°19'11.72"N	37°03'24.73"E	2237
416/09	H.G.W	Horro	Burkitu Oborra	9°27'08.97"N	37°03'43.07"E	3025

*E.W- East Wollega;W.W-West Wollega; H.G.W-Horro Guduru Wollega ;B.B-Buno Bedelle Zones

the sample was burnt on a heater inside a fume cupboard to get rid of the smoke. The samples were moved to preheated muffle furnace (SM 9080) maintained at 550°C until such a time when a light grey ash was noticed. The crucibles were cooled in desiccators and weighed. The ash content was calculated as:

$$\% \text{Ash} = \frac{(\text{weight of crucible} + \text{Ash}) - \text{weight of empty crucible}}{\text{Ash weight of sample} \times 100}$$

$$\text{Ash weight of sample} \times 100$$

The organic matter content was determined by subtract the percent ash from percent total dry matter and the value was expressed in percentage. The % organic matter content was calculated as:

$$\text{Organic matter content (\%)} = \% \text{ dry matter} - \% \text{ ash.}$$

The amount of organic carbon is determined as;

$$\text{Organic carbon (g)} = \frac{\text{weight of ash}}{\text{Sample weight}}$$

Crude fiber of the sample was determined according to AOAC, 2000. Two gram of the sample was crushed with petroleum ether. The crushed sample was boiled in reflux for 30 minutes with 200 ml of a solution contain 1.25 g of H₂SO₄ per 100 ml of solution. The solution was then filtered through linen on a fluted funnel. Then the sample was washed with hot water, using a two-food muslin cloth to trap the particles, the washed sample was transferred quantitatively back to the flask and boiled again in 200 ml of 1.25 g of

carbonate free NaOH per 100 ml for 30 minutes and washed before it transferred to a weighed Gooch crucible and dried in the oven at 105°C for three hours. After cooling in desiccators it was re-weighed. Then the percentage crude fiber was calculated as:

$$\% \text{ CF} = \frac{(\text{weight of sample} + \text{Crucible}) - (\text{weight of crucible} + \text{Ash})}{\text{weight of sample} \times 100}$$

$$\text{weight of sample}$$

The fat contents were determined by using fat extractor with automated control unit (FOSS Soxtec 2055) according to AOAC, 2000. The equipment has six extraction units with each unit accommodates the samples and aluminum cups for collection of the extracted fat. These units enable six samples to be analyzed at a time; within 75 minutes. Percentage of fat was considered as, the difference between weight of the pre-weighed cups and after extraction. One gram of the sample was weighed into the thimble and its mouth plugged with defatted cotton wool, after which it was inserted into the extraction unit. 18 ml of petroleum ether was dropped into each cup and maintained at 135°C aligning each cup with its corresponding thimble. The extraction and rinsing were done for 30 minutes each, after which the sample was aerated for 15 minutes and crude fat calculated as:

$$\% \text{ Fat} = \frac{(W3 - W2)}{W1} \times 100$$

Where: W1=weight of sample, W2=weight of empty cup and W3= weight of cup with the extracted oil.

The analysis of crude protein content was conducted with an aid of micro Kjeldhal system in accordance with AOAC, 2000. A small quantity of the yam flour sample (1 g) was introduced into the digestion tube (Kjeltec 2200 FOSS) and, a catalyst (2 tablets of 5g K₂SO₄ and 5 mg of Se) and 12 ml of concentrated tetra oxo-sulphate VI acid (H₂SO₄) were added. The digestion was run for one hour at 42°C. Eighty (80 ml) and 40 ml of water and sodium hydroxide (NaOH) respectively were used in the distillation using 2200 FOSS distillation unit and the distillate was collected in 4% Boric acid. The percentage nitrogen (N) was calculated as:

$$\% N = \frac{(a - b) \times n \times 0.014 \times mcf}{s}$$

Where; a=ml of H₂SO₄ required for titration of sample, b=ml of H₂SO₄ required for titration of blank, S=air-dry sample weight in mg, n=normality of H₂SO₄ (0.1 N), 0.014= weight of nitrogen in g and mcf=moisture correction factor. Then the protein content of the sample was estimated by percent nitrogen multiplied protein coefficient (6.25). The protein content calculated as 'N' × 6.25 (Bressani, 1994 and Udosen, 1995).

Mineral composition analysis

The phosphorus content of each sample was determined by the dry ash

extraction method following specific mineral element (AOAC, 1990). Five grams of the sample was burnt to ashes in a muffle furnace (SM9080) at 500°C. After complete ashing, the ash was diluted with 1% Hydrochloric (HCl) acid, then filtered into a 100 ml standard flask, and made up to the mark with deionized water. The solution was read with UV-visible spectrophotometer machine (model No: UV-1600, Shimadzu Corporation, Japan) for the determination of phosphorus in mg/100 g. The carbohydrate content of the sample was determined by estimation using arithmetic difference. The energy value was calculated by application of the thermal coefficients of Atwater and Rosa (Atwater and Rosa, 1899) with 4 calories for 1 g of carbohydrates; 4 calories for 1 g of protein and 9 calories for 1 g of crude fat. The available carbohydrate (CHO) and energy value were determined by using the formula as given below; CHO = [100 - (% moisture+% crude protein+% crude fat+crude fibre+% ash)].

Total energy (kcal)=[(% CHO × 4)+(% CP × 4)+(% CF × 9)]. Where; CHO, CP and CF are; carbohydrate, crude protein and crude fat, respectively (Hassan et al., 2008; Elinge et al., 2012; Tawheed and Monika, 2014).

Proximate Analysis

Tubers were peeled with a high-grade stainless steel handheld peeler, and were again washed with distilled water and dried using a paper towel before

they were sliced using a high-grade stainless steel knife. The peeled and washed roots were cut into pieces and oven dried at 105 °C for 24 hours. The dried samples were milled using a stainless steel mill and packed and stored at room temperature into sealed plastic bags for proximate and mineral analysis.

Proximate composition (in dry matter basis) of the oven-dried samples was conducted following the AOAC (2010) standard procedures at NVI and Horticoop. The micro-Kjeldahl procedure was used for the crude protein ($N \times 6.25$) analysis following method 954.01, AOAC (2010). The moisture content (MC) was determined by the hot air oven method as described by 925.10, AOAC (2010). Fat content was determined using the Soxhlet extraction with hexane according to method number 2003.06, AOAC (2010). The method 923.03, AOAC (2010) was used to determine the total ash content. Method number 962.09, AOAC (2010), was used for crude fibre determination. Carbohydrate content was calculated by difference as shown: Total carbohydrate by difference = 100 – (water, protein, total lipid, ash in g/100 g). Standard energy conversion factors (protein 4 kcal/g, fat 9 kcal/g, and carbohydrate 4 kcal/g) were used to estimate energy (in kcal).

Mineral Analysis

Macro (Ca, Mg, P, K, Na, S) and micro (Cu, Co, Fe, Mn, Se, Zn) minerals analysis of anchote root were conducted at the Horticoop

Ethiopia(Horticultural) PLC; Soil, Water and Plant Analysis Laboratory with the standard method, EPA (United States Environmental Protection Agency) 6010A of Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-AES) (VARIAN VISTA RL CCD Simultaneous ICP-AES Spectrometer). Microwave digestion (100 g dry plant material using conc. HNO₃ and 30% H₂O₂) with wet ashing under pressure (for 75 min, 200 °C and 15 bar) was used to prepare samples prior to ICP-AES analysis. Mineral content was recorded in mg/kg of flour and later converted into mg/100 g of dry matter or fresh weight basis.

Data analysis

The data was subjected to analysis of variance (ANOVA), in three replications with Complete Random Design(CRD) using SAS statistical software package, SAS 9.3(32) (SAS Institute, 2000). The entire dataset was standardized by dividing each variable with its respective range, and was subjected to clustering based on Un-weighted Pair Group Method of Arithmetic mean (UPGMA) and cluster distance and principal component analysis (PCA) was analyzed to assess correlations between components and the parameters measured.

Results and Discussion

The result showed there were significant difference among the accessions on biochemical, mineral,

and root flesh color of Anchote in macro and micro minerals; calcium, potassium, magnesium, sulphur, potassium, sodium, boron, copper, iron, manganese, molbedinium, zinc, and other biochemical analysis; dry matter, moisture, ash, crude fiber, protein, fat, carbohydrate and total energy showed that the existence of sufficient genetic variability of these traits within anchote landraces from Ethiopia (Table 2). The variability among landraces also revealed wide chance of developing anchote varieties possessing desirable biochemical traits.

Crude protein: The range of protein content of anchote roots was 7.21 to 15.93% and a mean of $11.81 \pm 2.19\%$ which is higher than protein contents of other common edible roots such as sweet potato (1.4%), cassava (0.5%), yam (7.82%) (Muluaem *et al.*, 2018) and taro (1.1%)(Bradbury and Holloway, 1988). According to Yenenesh *et al.*, (2016), the protein content of the anchote tubers ranged from 5.82 to 13.72 % from 44 accessions collected from western parts of Ethiopia. On contrary, Habtamu *et al.*, (1997), reported that peeled anchote root contains 3.9g of protein.

The crude protein content of anchote root in the present study was stuck between the range value (4.6- 16.4 %) reported by Desta *et al.*, (2011), but higher than the values (3.00-3.90 %) reported by other scholars (EHNRI, 1997; Habtamu *et al.*, 2013; Habtamu *et al.*, 1997). The variations in

genotype, geographical sources and agronomic practices could affect the protein content in root and tuber crops. The high crude protein content in anchote tubers are however comparable to those reported in yam (*Dioscorea alata*) (7.82 and 10.27%) (Muluaem *et al.*, 2018; Ezeocha and Ojimelukwe, 2012); taro (*Colocasia esculenta*) (11.00%) (Temesgen and Negussie, 2015) but in contrary to Otegbayo *et al.*, 2018 on yam collections from Nigeria (4.32%).

Crude fat: The crude fat content of anchote accessions was significantly influenced by the accessions and ranged from 0.01 to 2.25 % with the mean value of 0.59. Comparatively, this result was higher than the report of Habtamu *et al.*, 1997 on anchote (0.12%) but comparable and in the range with fat contents of yam (0.32%), potato (0.09%), and cassava (0.03 to 0.05%), (Muluaem *et al.*, 2018; Montagnac *et al.*, 2009; Yirmaga, 2013).

Ash: The ash contents of anchote accessions were in the range of 3.82 to 6.98% with the mean value of 5.04%. This result is higher than ash contents of potato, guinea yam, and cocoyam (Montagnac *et al.*, 2009; Yirmaga, 2013; Ihediohanma *et al.*, 2014; Kavitha and Parimalavalli, 2014). But it is comparable with enset (7.47 to 8.17%) (Mohammed *et al.*, 2013). The ash content of the anchote accessions in this study (5.04%) is higher than yam (4.41 and 2.61%), Otegbayo *et al.*, 2018 and Muluaem *et al.*, 2018,

and 3.09% in cassava(Christopher *et al.*, 2016).

The difference in ash contents among the accessions might be due to differences in the genotypes and collection areas' agro-ecology.

Carbohydrate: The carbohydrate contents of anchote accessions were in the range of 66.56 to 78.14% and with mean value of 72.13%. This result is comparable with carbohydrate contents of cocoyam (79.14-79.75%), but it is higher than that of sweet potato (25.74%), *enset* (32.75-35.53%), guinea yam (69.50%), yam (*Discorrea spp.*)(21.84%) and cassava (25.3-35.7%) (Montagnac *et al.*, 2009; Yirmaga, 2013; Ihediohanma *et al.*, 2014; Muluaem *et al.*, 2018). In contrary, the mean carbohydrate content presented in this study is by far higher than the reported value (25.5%) by Habtamu *et al.*, 2018 and lower than (80.52%) as reported by Yenenesh *et al.*, 2016. This might be associated to higher moisture contents, which decrease the proportions of other proximate values, the genetic factor, maturity and management factors.

Energy: The gross energy of *anchote* accessions was in the range of 323.71 to 353.53 kcal /100g with mean value of 334.42 kcal. On contrary, the mean energy content presented in this study is by far higher than the reported value (117.50kcal) by Habtamu *et al.*, 1997. On the other hand anchote's energy content is comparable with cereals such as corn (365kcal) and mung bean

(342. kcal) (Montagnac *et al.*, 2009; Blessing and Gregory, 2010).

Crude fiber: The crude fiber from this study was ranged from 0.20 to 8.07 % with the mean value of 3.55% which was higher than the reported value of 1.5% by Habtamu *et al.*, 1997, 1.28% by Muluaem *et al.*, 2018 and 0.68% in cassava(Christopher *et al.*, 2016) but lower than 5.05% by Yenenesh *et al.*, 2016. The crude fiber content of anchote accessions in this study is generally higher than yam(1.28%), potato (2.2%), carrot (2.8%), cassava(0.68%) and is comparable with other fibrous crops like kocho (*enset*) (3.37%), cassava (1.5%), and taro (3.9%) (Muluaem *et al.*, 2018; Bradbury and Holloway, 1988; Yirmaga, 2013). The difference might be due to genotype differences and the maturity stage when they were harvested for analysis.

Moisture and Dry matter: The moisture and dry matter contents ranged from 7.71 to 13.73% and 13.73 to 7.70% with a mean of 11.52% and 11.79%, respectively. The mean dry matter content of 11.79% found in this study is the lowest with the value of 24.10% reported by Yenenesh *et al.*; (2016) might be due to the maturity stage of the roots and the genotype differences.

Mineral Elements Concentration in Anchote Tubers

The range and mean values of major mineral elements (Ca, P, K, Na and Mg) for 100 anchote root samples are

compiled in Table 2. The mean contents of Ca, P, K, Na and Mg in the roots were 416.15, 465.42, 1558.37, 71.1 and 191.36 mg/100g and the ranges were 144.62 - 679.89, 247.52-704.26, 999.83- 2732.02, 35.47-150.20, and 98.32-292.64 mg/100g on dry matter basis respectively. The mean value of trace elements (B, Fe, Zn, Co, Mo and Mn), were 1.72, 15.33, 5.74, 0.79, 0.72, and 0.95 mg/100g on dry basis. The macro mineral elements concentration in anchote roots in this study was by far better than Yenenesh *et al.*, (2016) may due to the inclusion of limited number accessions in the study. On contrary, the mean Ca content

presented in this study is comparable with the reported value, 327 mg/100g by Habtamu *et al.*, 1997 but the value of other mineral elements is by far higher may be due to genetic factors and the environmental conditions where the tubers were grown and the large number of accessions incorporated in this study. The calcium to phosphorous ratio is 0.89 to 1 and the potassium to sodium ratio is 22 to 1.

Correlation analysis was used to look into the patterns of variations of the traits associations and their relation patterns.

Table 2. Variance analysis of 20 mineral and biochemical traits of anchote

Qualitative character	Mean + SE	Range	CV(%)	LSD(0.01)	P value	R ²
Calcium	416.15 + 101.38	144.62- 679.89	5.65	498.18	<.0001	96.37
Potassium	1558.37 + 292.85	999.83 - 2732.02	1.23	406.70	<.0001	99.72
Magnesium	191.36 + 36.31	98.32 - 292.64	1.06	42.90	<.0001	99.79
Sulfur	119.79 + 31.92	66.51 - 231.67	2.15	53.81	<.0001	99.23
Phosphorus	465.42 + 75.11	247.52 -704.26	1.21	119.89	<.0001	99.59
Sodium	71.10 + 20.46	35.47 - 150.20	0.87	13.21	<.0001	99.94
Boron	1.72 + 0.71	0.43- 5.74	10.74	3.91	<.0001	95.46
Copper	0.79 + 0.67	0.09 - 1.51	8.11	1.29	<.0001	95.89
Iron	15.33 + 13.17	4.13 - 111.52	2.62	8.50	<.0001	99.94
Manganese	0.95 + 0.33	0.28 - 1.99	8.52	1.71	<.0001	96.11
Molybdenum	0.72 + 0.48	0.04 - 1.68	12.29	1.81	<.0001	96.50
Zinc	5.74 + 0.52	1.57 - 46.73	3.1	3.76	<.0001	99.92
Dry matter	88.21 + 4.60	86.27 - 92.30	0.14	0.25	<.0001	99.40
Moisture	11.52 + 1.34	7.71 - 13.73	0.03	0.01	<.0001	99.99
Ash	5.04 + 0.61	3.82 - 6.98	0.2	0.02	<.0001	99.98
Crude fiber	3.55 + 1.58	0.20 - 8.07	0.03	0.002	<.0001	1.00
Protein	11.81 + 2.19	7.21 - 15.93	0.06	0.02	<.0001	99.99
Fat	0.59 + 4.24	0.01 - 2.25	0.29	0.002	<.0001	99.99
Carbohydrate	72.13 + 15.50	66.56 - 78.14	0.08	0.12	<.0001	99.97
Energy	334.42 + 17.32	323.71 - 353.53	0.05	0.38	<.0001	99.94

Cluster analysis

Grouping accessions based on their similarity is important for subsequent improvement and selection works. In this study 100 anchote accessions were clustered into six different groups based on 20 mineral and biochemical traits (**Figure 2** and Table 3). Each of the six clusters contributed differently with cluster number IV was the largest, containing 57 accessions and 57% of overall genetic similarity. The remaining clusters, I, II, III, V and VI totally contributed about 43% variation combined having 13, 18, 2, 9 and 1 accessions, respectively. The cluster mean for all the traits revealed that considerable differences were noticed between the cluster means

(Table 4). Accessions from all clusters except for cluster II and IV have produced the highest Ca content than others. Accessions from cluster VI have produced the highest Fe content (0.3844 mg/g) and Zn was highest in clusters I and IV; 0.006 and 0.006mg/g, respectively. Generally, accessions from clusters I, III and VI produced the highest contents of most of the mineral elements than the remaining clusters. Accessions collected from west Wollega (Boji Dirmaji and Gimbi) and east Wollega (Gudeya Bila, Limmu and Wayu Tuka) exhibited wider distribution in four clusters than others (Table 5).

Table 3. Distribution of 100 anchote accessions in to six clusters based on mineral and biochemical traits

Clusters	No. of accessions in each cluster	Accession numbers	% of contribution
I	13	2, 153, 397, 29, 40, 308, 161, 291, 126, 313, 9, 222, 328	13
II	18	8, 406, 51, 11, 58, 183, 223, 131, 43, 75, 280, 416, 158, 16, 132, 93, 286, 155	18
III	2	3, 400	2
IV	57	5, 157, 209, 297, 10, 80, 262, 53, 81, 140, 227, 41, 151, 316, 56, 67, 148, 281, 62, 87, 305, 66, 91, 113, 96, 290, 393, 292, 95, 304, 402, 407, 97, 112, 24, 302, 377, 50, 32, 385, 388, 206, 256, 14, 134, 288, 94, 211, 394, 65, 300, 271, 92, 214, 376, 296, 169	57
V	9	4, 219, 49, 218, 90, 309, 89, 156, 413	9
VI	1	381	1

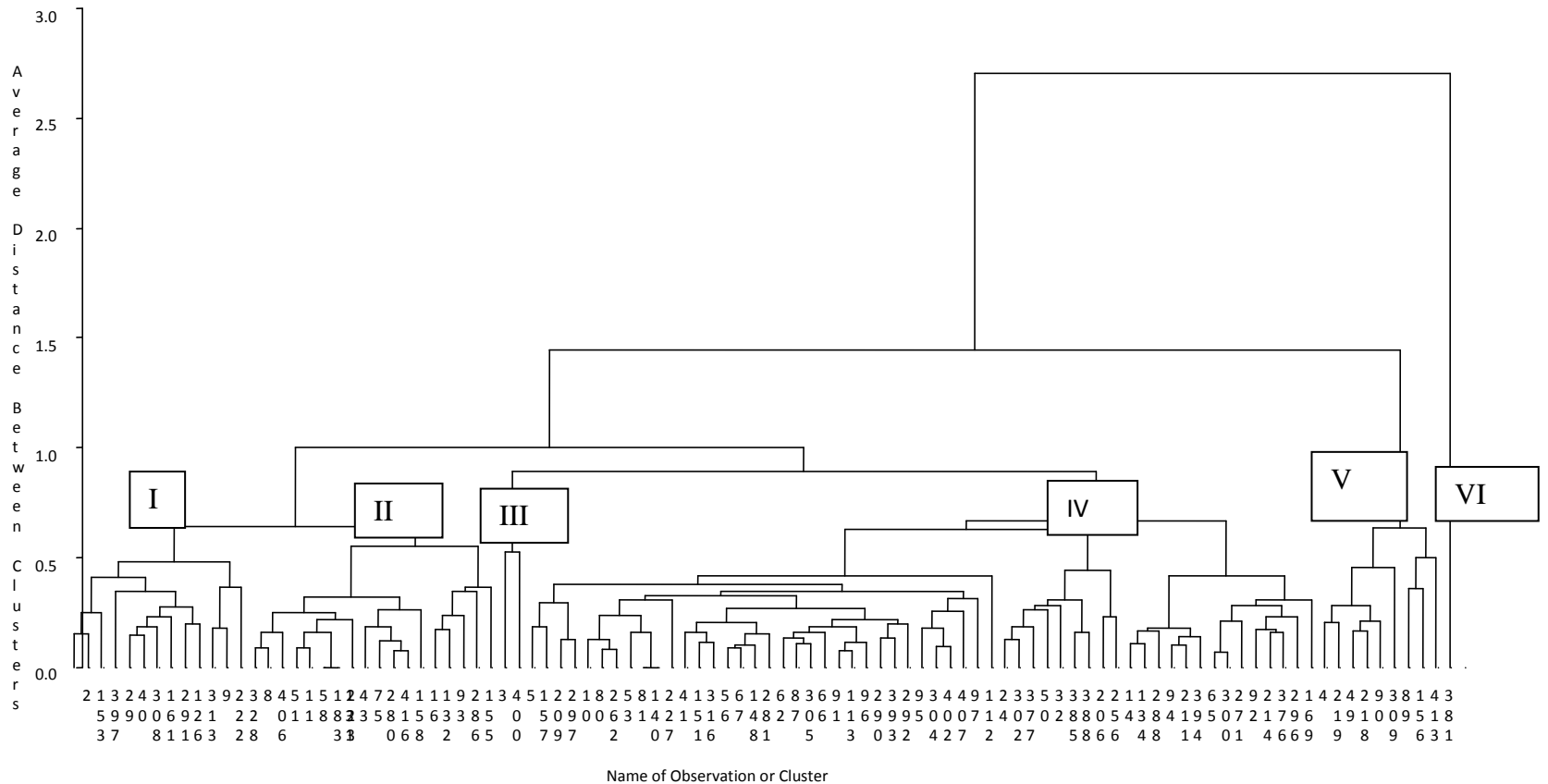


Figure 2. Dendrogram showing hierarchical clustering patterns of 100 anchote accessions (UPGMA) based on 20 qualitative characters

Table 4. Distribution of cluster means of mineral elements content of the 100 anchote accessions

Cluster	Ca	K	Mg	S	P	Na	B	Cu	Fe	Mn	Mo	Zn
I	552.90	1284.16	215.06	123.32	462.94	78.35	2.12	0.09	16.28	1.21	0.90	0.57
II	350.58	1214.62	172.68	105.38	402.03	56.24	1.50	0.09	11.87	0.76	0.52	0.51
III	598.18	1467.34	277.24	178.05	671.10	78.93	2.45	0.09	18.24	1.63	0.82	0.53
IV	397.99	1625.47	185.62	116.66	464.16	73.78	1.67	0.09	15.64	0.91	0.68	0.60
V	407.85	2070.16	207.58	129.74	530.53	75.00	1.48	0.09	15.13	0.95	0.77	0.55
VI	464.26	2707.08	204.62	136.18	700.75	60.26	2.88	0.09	38.44	1.10	1.08	0.53
MEAN	461.96	1728.14	210.47	131.55	538.59	70.43	2.02	0.09	19.27	1.10	0.79	0.55
S.deviation	87.81	518.76	33.08	22.95	110.93	8.87	0.52	0.00	8.78	0.28	0.17	0.03

Table 5. The distribution of anchote accessions in areas of collection based on cluster groups

Zone	District	Clusters						Total
		I	II	III	IV	V	VI	
West Wollega	Boji Dirmaji	1	1		2	2		6
	Gimbi	3	7		11	2		23
	Kiltu Kara		1		1			2
	Lalo Asabi				1			1
	Mana Sibumandi			1	7			8
	Nejo				4			4
East Wollega	Bonaya Boshe				1			1
	Digga	1						1
	Digga Leqa				1			1
	Gida Ayana	1			1			2
	Gobu Sayyo		1					1
	Gudeya Bila	1	2		10	1		14
	Leka Dulecha	3			5			8
	Limmu	1			3	1		5
	Nunu Kumba				1			1
	Sibu Sire	1				1		2
	Wayu Tuka	1		1	2	1		5
	Chingi	2	1		1			4
Jimma	Dedo				1			1
Buno Bedelle	Didesa				3		1	4
	Horro	1	2					3
Horro Guduru Wollega	Jimma Geneti				3			3
Total		16	15	2	58	8	1	100

Accessions grouped under clusters I, II and IV produced higher dry matter, clusters I and VI were superior in crude fiber, clusters II and VI in crude protein, clusters III and V better in carbohydrate and total energy (Table 6). Variability among clusters would be due to genetic factors than environmental influences as most of the clustered accessions exhibited variability in biochemical contents even though differ in collection areas except cluster VI which is accessed from Buno Bedelle, Didesa area. Accessions grouped under clusters III and V produced the lowest

biochemical contents except for carbohydrate and total energy and cluster IV was better in dry matter but moderate in all other biochemical contents. Thus, utilization accessions grouped under clusters I for high dry matter and crude fiber; cluster II for dry matter and crude protein; cluster III and V for carbohydrate and total energy; cluster IV only for higher dry matter, and cluster VI for crude fiber and crude protein in the subsequent anchote improvement programs for better nutritional contents.

Table 6. Cluster means for seven biochemical traits of anchote accessions

Cluster	DM	Ash	CF	CP	Cfat	CHO	Energy
I	11.67	5.15	4.13	11.46	0.49	71.24	335.18
II	11.74	4.89	3.35	12.31	0.31	70.75	335.01
III	10.06	4.16	2.99	11.06	0.32	74.41	344.72
IV	11.66	5.08	3.42	11.86	0.33	71.09	334.71
V	10.83	4.93	3.35	11.29	0.28	72.68	338.36
VI	10.48	6.98	6.15	14.08	0.34	68.13	331.89
MEAN	11.07	5.20	3.90	12.01	0.35	71.38	336.64
S.Dev.	0.66	0.86	1.06	1.01	0.07	1.91	4.07

Principal component analysis (PCA)

The patterns of variation and the relative importance of each trait in explaining the observed variability assessed through principal component analysis. Principal component analysis was executed to characterize 100 anchote accessions based on their mineral and biochemical composition to explain the observed variability to be assessed. The accessions were grouped into seven principal components based on root flesh color, mineral elements and biochemical traits of which flesh color, Ca, K, Mg, S, P and Na contents were significant (eigenvalue > 1) and explained 78 % of the total variability (Table 7).

The first principal component (PC1) alone accounted for 23.79% of the

total variation. Root flesh color, crude fat, crude protein and total energy had the highest loadings across all the seven components. The second principal component (PC 2), explaining 15.98% of the total variation and highly correlated with Ca, K, B, Fe, Zn, dry matter, crude fiber and carbohydrate while PC3 associated with root flesh color, Ca, K, Na, B, and crude protein explained 9.87% of the total variation. The remaining PCs (PC4-PC7) accounted 28.05% of the total variation, and mainly associated with all the variables except the crude fiber which was loaded lesser.

The variation exhibited in this experiment could be attributed to genetic and environmental factors.

Table 7. Variant component scores of root flesh color, minerals and biochemical traits of the first seven principal components for 100 anchote accessions

Variables	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Root flesh color	0.3006	0.2957	0.4375	0.3414	0.4642	0.3549	0.24
Ca	0.2754	0.3404	-0.4309	0.3346	0.3389	0.2903	0.2545
K	0.2409	-0.3793	0.5072	0.3946	0.3188	0.359	0.2525
Mg	0.3195	-0.2134	0.3269	0.5583	-0.4325	0.3539	-0.1506
S	0.3179	0.2323	-0.3782	0.2429	0.3166	-0.4064	0.4735
P	0.2697	0.4091	0.2465	0.178	-0.1586	-0.6478	-0.3115
Na	0.3005	0.2901	0.4123	-0.3081	-0.3282	0.3713	0.3773
B	0.3479	-0.3178	-0.4878	-0.5402	-0.2584	-0.2739	0.5447
Cu	0.2604	0.2899	-0.3221	-0.7028	0.2518	0.1665	0.1423
Fe	0.2959	-0.4118	0.1719	0.1482	-0.4895	0.1679	-0.5913
Mn	0.3607	-0.1789	-0.1902	0.1626	-0.8099	0.2542	0.1251
Mo	0.2794	0.1393	-0.1839	-0.1752	0.5848	0.2247	-0.6213
Zn	0.2493	-0.4429	-0.2003	0.2599	0.2177	0.4028	0.5304
DM	0.1583	-0.4578	0.1344	-0.2027	-0.1897	0.7794	0.1404
Moisture	-0.1576	0.4602	-0.1376	0.2031	0.1821	0.3428	0.7175
Ash	0.2345	-0.3678	0.2677	-0.5651	-0.2769	0.2607	0.2376
Crude fiber	0.6939	-0.5651	-0.2769	0.1484	0.1494	0.1565	0.1291
Crude protein	0.4899	-0.3221	-0.3564	-0.2852	0.1541	0.1584	0.5708
Crude fat	0.4216	0.2332	0.3305	0.2831	-0.3017	0.3166	-0.4946
Carbohydrate	-0.3394	-0.4151	0.2219	-0.1819	0.1923	0.6938	0.1477
Energy	-0.4971	0.1507	-0.1339	-0.1195	-0.4125	0.5692	-0.4052
Eigen value	4.9957	3.3556	2.0735	2.0174	1.5936	1.1978	1.0825
proportion	23.79	15.98	9.87	9.61	7.59	5.7	5.15
Cumulative	23.79	39.77	49.64	59.25	66.84	72.54	77.7

Conclusion and Recommendation

Anchote [*Coccinia abyssinica*] cultivation is quite common in western and southwestern parts of Ethiopia with its full utilization and production practices even though it is an underutilized root crop in other parts of Ethiopia and unknown in world's production and marketing. The result of analysis of variance indicated significant variations ($p \leq 0.01$) among the 100 anchote accessions for all the twenty biochemical and mineral traits considered in this study. There was a wide genetic variability among the anchote accessions tested in this study.

According to the results of this study, anchote could be the best alternative source of protein (11.81%) and carbohydrate (72.13%) than any other root and tuber crops. The variations in biochemical and mineral contents among the tested 100 anchote accessions could be due to genotype, geographical sources and agronomic practices. Anchote's fiber content (3.55%) is the highest to other root and tuber crops even though *kocho* (enset) and taro are comparable. Among the most important contents of anchote, Ca (416.15), Fe (15.33), Zn (5.74) and Mn (0.95), in mg/100g, are the most dependable potentials which are found abundantly and could help in the efforts to solve nutrition insecurity of

Ethiopian population. The higher calcium content of anchote could be an alternative to milk allergic infants and adults and the higher Mn and Zn contents of anchote could be an alternative to control dehydration problems due to diarrhea.

The cluster analysis of biochemical and mineral traits revealed the existence of six divergent groups, 88% of the accessions represented by clusters I, II and IV, with the highest biochemical and mineral traits, regardless of the geographical locations where the accessions were collected, showing the reduced influence of environments on the genetic factor. All the variables were grouped in one principal component of which root flesh color, Ca, K, Mg, S,P and Na were significant (Eigen value > 1) explaining 78% of the total variability. All the traits except for fat, carbohydrate and total energy, contributed maximally to the principal component with calcium the most important contributor (20%). The variation is attributed to genetic and environmental factors. The results obtained from this study confirmed the existence of variability among the accessions which indicates huge potentials for selection and crossing of nutritionally superior anchote genotypes. Anchote could be used as a high potential crop to fight protein and calcium deficiency in countries such as Ethiopia. Accessions numbers 400,328,206,222 and 308 were superior in Ca content. Accession 304 was superior in crude protein and energy content. For dry matter content,

accessions 62, 305,169,402, 58 and 262 were superior among the 100 accessions.

Acknowledgements

This work was supported by AGP II project (Agricultural Growth Programme phase II), Ethiopian Institute of Agricultural Research, farmers who gave us the seed, technical assistants and field assistances of DZARC and we are grateful for the support we got.

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