ISSN: 2754-477X

Journal of Food Technology & Nutrition Sciences

SCIENTIFIC Research and Community

Research Article

Open Access

Determination of Nutritional and Anti-Nutritional Contents of Agew Potato (*Plectranthus Edulis*) Tubers Cultivated in Awi Zone, Amhara Regional State of Ethiopia

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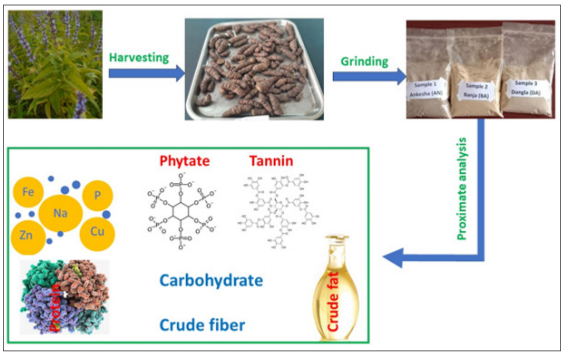
ABSTRACT

Plectranthus edulis is one of the most important tuber plants, and its nutritional value has not been fully investigated. It is also an indigenous plant and grows in mid- and high-altitude areas. This study investigated the proximate, anti-nutritional, anti-oxidant, and mineral compositions of Plectranthus edulis (Agew potato), a plant grown in the Awi zone of the Amhara region. The AOAC method was used for the determination of proximate composition and recovery test used for mineral determinations. In this study, the moisture content, ash content, crude fat, crude protein, crude fiber, and carbohydrate, phytate, tannin, TPC, TFC, and DPPH were obtained in the range of 70.87 - 74.92 g/100g, 3.92 - 4.58 g/100g, 0.36 - 0.61 g/100g, 6.86 - 9.29 g/100g, 3.18-3.56 g/100g, 71.79 - 78.81 g/100g, 61.48 - 108.04 mg/100g, 8.46 - 44.15 mg/100g, 17.53 - 28.42 mg/g, 2.75 - 5.71 mg/g, and 52.94 - 71.49 %, respectively. In addition, the calibration curve, linear range, correlation coefficient, instrumental detection limit, limit of detection, and limit of quantification were determined to validate the determination of analytes in Plectranthus edulis. Based on the calibration curves, the concentrations of Na, P, Fe, Cu, and Zn were found in the range of 261.85–603.90 mg/kg, 82.02–251.82 mg/100g, 23.74–65.36 mg/kg, 9.98–16.27 mg/kg, and 17.68–30.20 mg/kg, respectively. Overall, the output of the study revealed that Agew potato is a good source of nutrients such as carbohydrate, crude protein, crude fiber, crude ash, crude fat, sodium, copper, iron, zinc, and phosphorus. Owing to this, the agew potato has become a substantial variation among the various vegetables cultivated in the region. Generally, Dangila district had high proximate composition, anti-nutrient, anti-oxidant, and mineral contents relative to other sample areas; however, Banja district had the highest levels of tannin, fiber, and carbohydrates.

Graphical Abstract



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Received: July 08, 2024; **Accepted:** July 15, 2024; **Published:** August 30, 2024

Keywords: Agew Potato, Proximate, Anti-Nutrient, Anti-Oxidant Activity, Mineral Content

Abbreviations

AOAC: Association of official Analytical chemists
$CuSO_4$: Copper sulfate
DPPH: Diphenyl-1-Picrylhydrazyl
FAO: Food and Agriculture Organization
IDL: Instrumental Detection Limit
Kg/ha: Kilo Gram Per Hectare
LDL: Low Detection Limit
NaOH: Sodium Hydroxide
H_2SO_4: Sulfuric Acid
KOH: Potassium Hydroxide
H_2O_2: Hydrogen Per Oxide
Edulis: Plectranthus Edulis

HNO_3: Nitric Acid
HCl: Hydrochloric Acid
K_2SO_4: Potassium Sulfate
TFC: Total Flavonoid Compound
TPC: Total Phenolic Compound
UV-Vis: Ultra Violet Visible
RDA: Recommended Daily Allowance
SD: Standard Deviation

Introduction

Vegetables are highly recommended for humans to consume frequently. They are a major source of a healthy diet and excellent sources of antioxidants (e.g., flavonoids, saponins, phenols, and carotenoids), vitamins, carbohydrates, minerals (e.g., calcium, nitrates, potassium, magnesium, and iron), and dietary fiber. Potato (Solanum tuberosum L.) is the fourth most important food crop next to rice, wheat, and maize and has a great contribution to food and nutrition security in the world. Potato is an important staple food consumed daily worldwide. It is considered an essential component of a healthy diet, so nutrition experts recommend eating at least a few servings of it every day [1-3].

Several agricultural crops are known to originate from Ethiopia, and it is also stated that Ethiopia is well known for its diversity of indigenous food plants, of which 27% are cultivated vegetables by traditional farmers in home gardens and about 29% are non-cultivated vegetable species. However, the food insecurity crisis is still persisting and prevalent in many parts of Ethiopia. This is mainly due to the highly selective and restricted food consumption habits of the population, as well as less exposure to important indigenous food plants such as Plectranthus edulis, Coccinia abyssinica, and Dioscorea abyssinica tubers. Plectranthus edulis (Vatke) Agnew (Lamiaceae) is an ancient Ethiopian tuber crop grown in mid- and high-altitude areas in the south and south-west of Ethiopia. It is particularly important as local diets mainly occur in the autumn season, from September to November. It is highly valued for its contribution to food security in these periods since other food crops are not ready for consumption [4].

Anti-nutritional components in potatoes exist naturally in tubers, are created as effects of destabilization of the plant's metabolism, or can be absorbed from a polluted environment. Several types of anti-nutritional factors with toxic potential have been measured in foods and shown to be heat-stable or heat-labile. These factors include saponins, tannins, phytic acid, gossypol, lectins, protease inhibitors, amylase inhibitors, antivitamin factors, metal-binding ingredients, goitrogens, etc. [5-6].

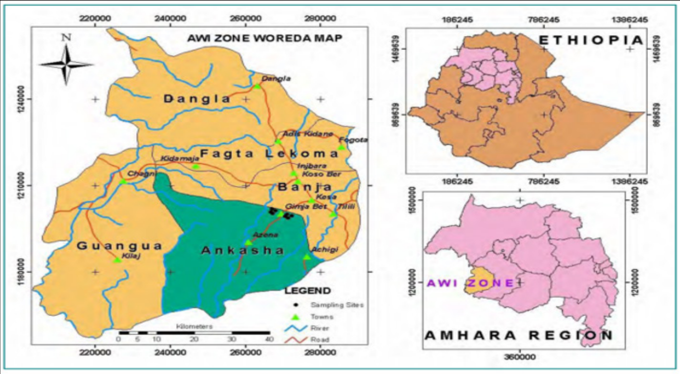
In spite of its nutritional, economic, and industrial benefits, potato's potential is not well studied in terms of utilization at both the industry and household levels. Many scholars have determined the nutritional qualities of potato cultivars that were planted in different areas and indicated significantly different nutrients among different potato cultivars, which might be related to soil, climate, cultivation technologies, etc. Since a long time ago, P. edulis has been a major traditional food crop in several regions of Ethiopia. In some regions, people who ingested this potato have been regarded as poor. This might be due to a lack of knowledge of its nutritional content. Despite its importance for food security and medicinal value, little research has been conducted on the crop [4,7].

The objective of this study was to investigate the nutritional and anti-nutrient content of Agew potato (Plectranthus edulis), commonly cultivated in the Awi zone, Amhara region, Ethiopia. Furthermore, the output of the study has provided additional data on the nutritional and nutraceutical potential of Agew potatoes in Ethiopia.

Material and Methods

Description of the Study Area

The study was conducted at Awi Zone, which is located in the Amhara Region of Northwestern Ethiopia, roughly in the middle between Debre Markos and Bahir Dar towns. In the current administrative structure, the zone has seven main administrative districts: Banja, Ankesha, Fagta lekoma, Dangila, Guangua, Guagusa shekuadad, and Jawi (Figure 1). The last two districts are not indicated on the map because they were recently organized as districts. The zone is known for its sufficient water resources and high rainfall. Awi Zone is located at a latitude of 10.95° N and a longitude of 36.5° E. It lies in an altitude range of 1800–3100 m above sea level, with an average altitude of 2300 m. It is predominantly inhabited by the Awi, an ethnic group that belongs to the central Cushitic sub-family and inhabits Northwestern Ethiopia [8].



(Source: ArcMap version 9.1, Gis by ESRI, 2005)

Figure 1: Map and Location of the Study Area in Amhara National Regional State

Materials and Instruments

UV spectrophotometer (UV-1800, model India Shimadzu), MP-AES (model 4200, made in India Shimadzu), Soxhlet (2055 Soxtec manual extraction unit equipped with time and temperature control), Kjeldahl (UK apparatus, FOSS, S-1712050), electronic balance (OHAUS), muffle furnace (Carbolite, S-302RR), oven (GX-65B), vortex, and hot plate were used for analysis.

Chemicals

All chemicals and reagents were used in analytical grade: NaOH (99.99%, Fisher Scientific), H2SO4 (98%, Merck), H2O2 (30%, Merck), HNO3 (70%, Merck), HCl (37%, Merck), KOH (99.99%, Merck), CuSO4 (Fisher Scientific), and metal stock standard solution (1000 ppm of Cu, Fe, Na, and Zn) were used to prepare the calibration standard.

Sampling and Sample Preparation

Depending on the availability of the tuber crops, representative tuber crops of 1.5 kg of plectranthus edulis were collected systematically from three different districts of the Awi zone, namely, Banja, Ankesha, and Dangila districts. From each district, one sampling site was selected. After collection, the samples were kept in polyethylene plastic bags, labeled properly, and then transported to the laboratory and kept at room temperature (20–30 °C) for further analysis. Once the samples reached the laboratory, various steps were carried out. First, the plectranthus edulis tubers were thoroughly washed with tap water to remove all the dust. Next, the cleaned tubers of plectranthus edulis were peeled and sliced into small pieces with the help of a sharp stainless-steel knife for quick drying. Then the sliced samples were kept in an oven and dried at 105 °C for 24 h to remove the majority of the moisture present. After that, the dried samples were grinded and homogenized by a grinder and sieved using a 0.5 mm sieve. Finally, the dried powdered root samples were kept in polyethylene bags until the analysis (Figure 2).

Figure 2: Agew Dinch (Plectranthus Edulis) Tubers

Proximate Analysis

The AOAC official method of 925.10 (AOAC, 2013), 923.05, 2016 (AOAC, 2016), and the Weende method, which is modified by hum as stated in AOAC (2003), 2003.05 (AOAC, 2016), and AOAC 979.09, 2005, had been used for the determination of moisture content, ash content, crude fiber, crude fat, and crude protein, respectively, and carbohydrates had been determined by the difference of all other proximate values from 100% [9].

Anti-Nutrient Determination

Phytate

For phytate determination, the Latta and Eskin (1980) and modified Latta and Eskin procedures were used by Vaintraub and Lapteva (1988). A 0.1 g of dried sample was extracted with 10 mL of 0.2 N HCl for 1h at ambient temperature and centrifuged at 3000 rpm for 30 min. The clear supernatant was collected for the phytate estimation. Then, 2 mL of wade reagent was added to 3 mL of the supernatant sample solution. The sample solution was immediately homogenized and centrifuged at 3000 rpm for 10 min. After that, the solution was introduced into the cuvette, and the absorbance was measured at 500 nm using a UV-Vis spectrophotometer. Then the phytate concentration was calculated from the difference between the absorbance of the blank (3 mL of 0.2 N HCl + 2 mL of Wade reagent) and that of the assayed sample. Finally, the amount of phytic acid was calculated using the phytic acid standard curve, and the result was expressed as phytic acid in µg/g [10,11].

Tannin

Tannin content was determined by the method of Maxson and Rooney (1972). 1 g of dried sample was measured in a screw-cap test tube, and 10 ml of 1% HCl in methanol was added to the tube containing the sample. Then the tube containing the sample was put on a mechanical shaker for 24 h at room temperature, and then the tube was centrifuged at 1000 rpm for 5 min. After that, 1 mL of supernatant was taken and mixed with 5 mL of vanillin-HCl reagent in another test tube. Finally, the test tubes were kept for 20 min to complete the reaction, and the absorbance was measured at 500 nm [12].

Determination of Total Phenolic, Flavonoid and Free Radical Scavenging Antioxidant \ Activities

Sample Extraction

A method reported by Punia (2018) was used for the extraction and quantitative determination of the total phenolic, flavonoid, and free radical-scavenging antioxidant activities of Agew dinch (p. edulis). 1 g of freeze-dried potato flour was extracted with 10 mL of 80% methanol for the determination of DPPH, total phenolic content, and total flavonoid content. The mixture was centrifuged at 6000 rpm for 15 min at room temperature, and the extracted samples were stored at -20 ºC until analysis [13].

Determination of Total Phenolic Content

The total phenol content of the extract was determined using the method reported by Singleton and Rossi (1965). A sample of methanolic extract (0.1 mL) was mixed with distilled water to make a total of 1.5 mL. Next, 0.5 mL of folin Ciocalteu reagent and 10 ml of 7.5% $Na2CO3$ were added consecutively and then incubated at 37 oC for 60 min. The absorbance of the extract was measured at 760 nm by a spectrophotometer. Finally, the total phenolic content was calculated using gallic acid as a standard [14].

Standard Preparations

The standard gallic acid solution was made by dissolving 10 mg of gallic acid in 10 mL of acidic methanol (80%) (1 mg/mL). Various concentrations of gallic acid solutions in 80% acidic methanol (0, 0.05, 0.1, 0.2, 0.3, and 0.4 mg/mL) were prepared from the standard solution. To each concentration, 0.2 mL of 10% Folin-Ciocalteu reagent, 2 mL of purified water, and 2 mL of 7.5% $Na2CO3$ were added to make a final volume of 4.3 mL. The total phenolic content was expressed as gallic acid equivalents using the linear equation based on the calibration curve (gallic equivalent (mg of GAE/g of sample)).

$$\mathbf{C\ (mg/g,\ in\ GAE) = C_1 x \frac{V}{m}}$$

Where; C = total phenolic content in mg/g, in GAE (Gallic acid equivalent)
$C1$ = concentration of Gallic acid established from the calibration curve in mg/mL,
V = volume of extract in mL, and
m = the weight of the plant extract in g.

Determination of Total Flavonoid Content

The amount of flavonoid content in methanolic extracts was determined by the aluminum chloride colorimetric method. A l mL of the extract was mixed with distilled water and made up to 5 mL with distilled water. After that, 0.5 mL of 5% $NaNO2$ was added. After 5 min, 0.6 mL of 10% $AlCl3$ was added and mixed. After 6 min, 2 mL of 1N NaOH were added and mixed well. Then 2.1 mL of distilled water was added to make the volume reach 10 mL. The absorbance of the resulting pink color was measured at 510 nm. The total flavonoid content was calculated using quercetin as a standard [15].

Standard Preparations

A stock solution (1 mg/mL) of quercetin was prepared by dissolving 10 mg of quercetin in 10 mL of 80% acidic methanol. Then the standard solution was diluted serially to make various concentrations of 0, 0.1, 0.2, 0.4, 0.6, 0.8, and 1 mg/mL solutions. To each concentration, 150 µL of 5% NaNO3, 15 0 µL of 10% AlCl3, 3 mL of deionized water, and 2 mL of 1M NaOH were added, making a final volume of 4.8 mL. The total flavonoid content was expressed as quercetin equivalents using the linear equation based on the calibration curve (quercetin equivalent (mg of QAE/g of sample).

Determination of Free Radical Scavenging Antioxidant Activity Using DPPH Method

The antioxidant activity of the extracts, on the basis of the scavenging activity of the stable DPPH free radical, was determined by the method followed by Brand-Williams. An aliquot of 0.1 mL of the samples was put in a test tube and reconstituted into a volume of 1 mL with methanol, and then 3 mL of DPPH reagent was added and vortexed, then let stand at 370 °C in the dark for 20 min. The absorbance of the resulting oxidized solution was measured at 517 nm against methanol as a blank. The DPPH content was calculated using an ascorbic acid standard [16].

Standard Preparations

The stock standard solution of ascorbic acid was prepared by dissolving 10 mg of ascorbic acid in 10 mL of 99.8% methanol. Then the standard solution was diluted serially to make various concentrations of 0, 5, 10, 15, 20, and 25 µg/mL solutions. To each concentration, 3 mL of 0.004% DPPH solution was added, making a final volume of 4 mL. The capability to scavenge the DPPH radical was expressed as a percent using the linear equation based on the calibration curve.

Mineral Determinations

Phosphorus

An accurately 0.5 mg (± 1mg) homogeneous test portion was measured in the crucible. Add 0.5 g of ZnO to a crucible and mix with a glass rod. Dry for 1-2 h at 110 ºC on a hot plate until the residue is black. After that, the crucible was placed in a muffle furnace at 525 º C for 4 h. The crucible would be removed from the furnace and allowed to cool to room temperature. Then, 5 mL H2O and 5 mL HCl were added to the cooled crucible, and covered with a watch glass, and then the contents were carefully boiled for 5 min on the hotplate. The contents of the crucible were filtered into a 100 mL volumetric flask, and the crucible and inner surface of the watch glass were rinsed with 5 mL of hot water. The rinsing process was repeated four times with 5 mL of hot water, and all rinses were transferred through the filter into the volumetric flask. The solution was neutralized by adding 50% KOH solution until the solution was slightly opalescent [Zn (OH)2]. Then HCl was added drop wise until opalescence disappeared, and the solution was cooled to room temperature and then diluted to 100 mL with H2O. Depending on the expected content of P, an accurately 1.00 mL treated solution was pipetted into a 50 mL volumetric flask and diluted to 5 mL with H2O. A 20 mL molybdate-ascorbic acid solution was added to the test solution in a 50 mL flask and also to the phosphorous standard solution. Swirl the contents carefully. The flasks containing solution were placed in a boiling water bath and kept in the water bath for exactly 15 min. Finally, the flasks were cooled under tap water to 20–30 oC, and then the contents were diluted to 50 mL with deionized water and mixed. The solutions were transferred to 1 cm cuvettes, and the absorbance of each solution against the reagent blank was measured at 823 nm. The measurement must be made within 1 h after the color reaction. The standard curve was constructed by plotting absorbance against the amount of P in P standard solutions.

Determination of Sodium, Copper, Iron, and Zinc

Next to ash digestion methods, the concentration of targeted metals (Fe, Zn, Cu, and Na) in Agew potatoes was determined by using microwave plasma atomic emission spectroscopy (MP-AES).

Ash Digestion

The potato samples were dried at 60–70 ºC in an oven for 24 h to remove moisture. And then the dried samples were ground using a mortar and pestle to obtain a homogenized powder. 1 g of the powdered sample was weighed in a crucible, introduced into a muffle furnace, and heated at 550 oC for 6 h. The crucible containing the ash was cooled down to room temperature. Then, 20 mL of 1:1 HCl was added to the cooled ash and covered with a watch glass. The crucible was heated over a hotplate at a moderate temperature for 4 h and then the crucible was removed from the hotplate and cooled to room temperature. The solution was filtered with filter paper, and the filtrate was introduced into a 100 mL volumetric flask. The remaining volume was reconstituted with distilled water. Finally, the extracted sample solution was analyzed by microwave plasma atomic emission spectroscopy. A blank sample analysis was carried out, similar to the potato sample analysis procedure.

Statistical analysis

Using the one-way analysis of variance (ANOVA) technique from the IBM SPSS version 20 software, all data were statistically processed. The mean and standard deviation (SD) of the data were computed. The Tukey's test (P 0.05) was used to determine the statistical significance of the differences between the means.

Results and Discussion

Proximate Composition

The proximate composition of a cultivated Agew potato (Plectranthus edulis), namely, moisture, crude protein, crude fat, carbohydrate, total ash content, and crude fibers, were assessed. The moisture contents of Agew dinch are shown in Table 1.

Table 1: Moisture Contents of Agew Potato before Sample Preparation (G/100g In Fresh Weight, N = 3)

Parameter	Sample code		
Moisture content(g/100g)	Ankesha (AN)	Banja (BA)	Dangila (DA)
	72.30 ± 0.87^b	70.87 ± 0.15^a	74.92 ± 0.22^c

The findings showed that there were significant differences ($P \leq 0.05$) in the moisture contents of Agew potato (p. edulis) among the three sampling areas. The highest moisture content of the samples was found in the sample site of Dangila (75.81±0.09%), while the lowest amounts were found in Banja (70.87±15%). Our results fell between the range of 65 to 82% reported by.The results of the analysis of the proximate composition of Agew potato are shown in Table 2. As can be seen, the carbohydrate content has the highest value, followed by moisture, crude protein, total ash, crude fiber, and crude fat on a dry basis [17,18].

Table 2: Proximate Composition of Agew Potato (Plectranthus Edulis) (G/100g In Dry Weight, N = 3, Mean ±Sd)

Sample code	Moisture	Ash	Fat	Protein	Fiber	Carbohydrate
AN	9.48 ±0.07[b]	3.93±0.05[a]	0.36±0.03[a]	7.03±0.15[a]	3.39±0.18[a]	75.81±0.09[b]
BA	6.48±0.19[a]	3.92±0.02[a]	0.48±0.11[ab]	6.86±0.25[a]	3.56±0.22[a]	78.81±0.08[c]
DA	10.55±0.40[c]	4.58±0.06[b]	0.61±0.07[b]	9.29±0.03[b]	3.18±0.07[a]	71.79±0.40[a]

AN: Ankesha, BA: Banja, DA: Dangila.

Ash contains some important minerals and trace elements essential to various body structures and functions. Among the chosen potato sampling areas, the total ash content of DA significantly differs ($P \leq 0.05$) from the other sample sites, while the ash contents of AN and BA do not differ ($p > 0.05$). The highest amount of ash was found in Dangila (4.58 ± 0.06 g/100g), and the lowest amount was found in Banja (3.92 ± 0.02 g/100g). Based on the experimental results, our findings were in close agreement with the ash contents (3.98–4.71%) of DWB reported by Gumul et al. Protein contents differed insignificantly among three sampling sites of DA, AN and BA ($p > 0.05$). The highest protein level was found in Dangila (9.29 ± 0.03 g/100g), and the lowest level was found in Banja (6.86 ± 0.25 g/100g). The crude fat content of BA was not significantly different ($p > 0.05$) from that of AN and DA, although there was a significant difference ($P \leq 0.05$) between the sampling areas of AN and DA. BA had the lowest crude fat content (0.36 ± 0.03 g/100g), while DA had the highest crude fat content (0.61 ± 0.07 g/100g). The range of our crude fat concentrations was largely in close proximity to the results reported by Zhou et al., which ranged from 0.10% to 0.73%. The fiber content of the plectranthus edulis (Agew potato) was in the range of 3.18 ± 0.07 g/100g to 3.56 ± 0.22 g/100g. No significant differences ($p > 0.05$) were seen in the proximate analysis of crude fiber content among different sampling sites. The maximum crude fiber content was observed in BA (3.56 ± 0.22 g/100g), while the lowest content was noticed in DA (3.18 ± 0.07 g/100g). The finding fell within the range reported by Burlingame et al., which ranged between 0.3 and 3.67 g/100g. Another crude fiber content reported by Naz. is the range between 3.677± 0.44 and 5.180±1.32 g/100g, which is similar to our finding. The carbohydrate contents were in the range of 71.79 ± 0.04 to 78.81 ± 0.08 g/100g; the highest value was recorded in BA, whereas the minimum value was recorded in DA. In Dangila, the moisture content is the highest, and owing to this, the carbohydrate content is the lowest. The amount of moisture lost during dehydration is inversely related to the amount of carbohydrates attained [7,19-22].

Anti-Nutrient Composition

Based on the output study of the phytate and tannin contents in the sample, their concentrations differed significantly ($P \leq 0.05$) among the samples collected from various areas. The highest and lowest phytate content of the study was found in DA and AN, respectively, while the highest and lowest tannin content was found in BA and AN, respectively. The tannin results of the finding ranged from 8.46 ± 0.98 to 44.15 ± 1.71 mg/100g, and the phytate contents of the finding ranged from 61.48 ± 0.95 to 108.04 ± 0.84 mg/100g. The phytate and tannin contents reported by Williams et al. were 1.325± 0.007% (1325± 0.007 mg/100g) and 0.535± 0.021% (535± 0.021 mg/100g), respectively, which is greater than our finding. Calibration curve equation, linear range and correlation coefficient of anti- nutrient are shown in Table 3. The anti-nutrient content of Agew potato (Plectranthus edulis) is shown in Table 4 [18].

Table 3: Calibration Curve Equation, Linear Range and Correlation Coefficient of Anti- Nutrient

Anti-nutrient	Calibration curve equation	Linear range (µg/mL and mg/L respectively	Correlation coefficient (r^2)
Phytate	Y = -0.0076X + 0.4049	0 – 36	0.9993
Tannin	Y = 0.0051X + 0.0034	0 - 66.67	0.9993

Table 4: Anti-Nutrient Content of Agew Potato (in dry weight, n=3)

Sample code	Phytate (mg/100 g)	Tannin mg/100g)
AN	61.48±0.95[a]	8.46±0.98[a]
BA	89.33 ± 1.27[b]	44.15±1.71[c]
DA	108.04 ± 0.84[c]	18.24±2.09[b]

Different letters within one column denote statistically significant differences (P≤0.05) by ANOVA and Tukey's test

Gemede et al, reported that the problem associated with phytate in food is that it can bind some essential mineral nutrients in the digestive tract and can result in mineral deficiencies. Tannins have been reported to affect protein digestion ability, adversely influencing the bioavailability of non-heme iron, leading to poor iron and calcium absorption [23].

Total Phenolic, Flavonoid Content and Free Radical Scavenging Activities (DPPH)

The total phenolic content ranged from 17.53 ± 0.61 to 28.42 ± 1.36 mg GAE/g. The total phenolic contents of Agew potato in sample DA differed considerably ($P \leq 0.05$) from AN and BA, although AN and BA did not differ significantly ($p > 0.05$) in terms of total phenolic content. The sample area of Dangila had the highest total phenolic content compared to the others. The total phenolic content of potatoes, as reported by Al-Saikhan et al. (11.41–27.47 mg GAE/g), was substantially supported by the findings [24].

Significant differences among the samples may be attributed to genotypes and harvest location, which influence the accumulation of phenolic compounds by synthesizing different quantities and/or types of phenolics. The total flavonoid contents of Agew dinch ranged from 2.75 ± 0.18 to 5.71 ± 0.41 mg/g. All sample locations had significantly different flavonoid contents ($p < 0.05$). DA had the maximum flavonoid contents of the entire sample. Overall, the present study's results were reported by Zubair et al. with a TFC of 2.38 ± 0.73 mg/g DW, which is less than our finding. The general location in which their plant was cultivated may have contributed to the higher production of total flavonoid content. Excessive flavonoids are appreciable because they behave as powerful protective agents against inflammatory disorders. In comparison to the other sampling areas (AN and BA), DA significantly differs ($p >0.05$) in terms of DPPH. In our investigation, the antioxidant activity of plectrantus edulis (Agew potato flour) ranged from 52.94 ±1.06% to 71.49 ±0.64%. According to Al-Saikhan et althea antioxidant activity of yellow and white-fleshed potato types was ranged from 65.2% to 89.2%. When comparing the DPPH results from these data, a little variation is visible. This variation may be caused by harvest conditions, agricultural practices, and environmental changes, such as changes in altitude or soil type, among other things. The calibration curve equation, linear range, and correlation coefficient of the anti-oxidant are shown in Table 5 [25-26].

Table 5: Calibration Curve Equation, Linear Range and Correlation Coefficient of Anti- Oxidant

Anti-oxidant	Calibration curve	Linear range (mg/mL), except DPPH (µg/mL)	Correlation coefficient (r^2)
Phenol	y = 0.6499x – 0.0039	0 – 0.4	0.9984
Flavonoid	y = 0.6137x + 0.0148	0 – 0.8	0.9981
DPPH	y = 3.7054x – 2.1901	0 – 25	0.9967

The value of total phenolic content, total flavonoid content and DPPH of Agew potato were shown in Table 6.

Table 6: Total Phenol, Total Flavonoid and Dpph Contents of Agew Dinch (In Dry Weight, N = 3)

Sample code	TPC (mg GAE/g)	TFC (QE/g)	DPPH (%)
AN	18.22 ± 0.88a	2.75 ± 0.18a	58.61 ± 1.04a
BA	17.53 ± 0.61a	3.12 ± 0.14b	52.94 ± 1.06a
DA	28.42 ± 1.36b	5.71 ± 0.41c	71.49 ± 0.64b

Data are expressed on a dry weight. Data are mean ± SD. Different letters within one column denote statistically significant differences ($P \leq 0.05$) by ANOVA and Tukey's test.

Mineral Composition

Instrument Calibration

The instrument was calibrated using a series of working standards. The working standard solutions for each metal were prepared by diluting the intermediate standard solution. The working standard linear range for each analyte is shown in Table 7.

Evaluation of Method Performance

Calibration curves were revealed for five concentration levels, and each level was extracted in triplicate. The calibration curves were constructed by plotting the absorbance of Na, P, Fe, Zn, and Cu as instrumental responses against the corresponding concentrations of the target analytes. Linearity ranges and other parameters were summarized in Table 7. The LOD and LOQ were determined as the lowest concentrations, yielding an S/N of 3 and 10, respectively.

Table 7: Calibration Curve, Linear Range, Correlation Coefficient, Instrumental Detection Limit, Limit of Detection, And Limit of Quantification for The Determination of Analyte

Analyte	Calibration curve equation	Linear range (in ppm, n = 3)	Correlation coefficient (r^2)	IDL (mg/ L)	LOD (mg/L)	LOQ (mg/L)
Na	Y = 306511 X + 28483	0.081-10.00	0.9996	0.019	0.024	0.081
P	Y = 18.361 X + 0.1662	0.01 - 0.06	0.9998	-	-	-
Fe	Y = 10350 X – 283.55	0.024 -10.00	0.9998	0.0026	0.0072	0.024
Zn	Y = 11439 X + 102.01	0.039 -1.00	0.9995	0.0021	0.012	0.039
Cu	Y = 104346 X + 441.09	0.012 -1.00	0.9994	0.0025	0.0035	0.012

Recovery Tests

The accuracy of the method was evaluated by analyzing the digests of spiked samples. 60 µL of Zn and Cu, 400 µL of Fe, as well as 500 µL of Na were spiked into a round-bottom flask that containing 1g of plectranthus edulis (Agew potato) sample. Then, the spiked samples were digested, similar to the procedure that was used to analyze the proximate composition. After that, the filtrate was transferred to a 100 mL conical flask and reconstituted with distilled water. Finally, MP-AES was used to quantify the concentration of target analytes in blank and spiked samples. The recoveries of metals in the spiked Agew dinch samples were in the range of 90.40 to 103.87 % (Table 8). Percentage recovery is calculated by [27]:

$$\% \text{ Recovery} = \frac{\text{spiked sample} - \text{un spiked sample}}{\text{amount of added}} \text{x}100$$

Table 8: Recovery Test for The Optimized Procedure of Agew Potato Samples

Metal	Conc. in sample (mg/kg)	Amount added (mg/kg)	Conc. in spiked sample (mg/kg)	Recovery (%)
Na	540.16 ± 2.85	463.80	959.45 ± 3.87	90.40
Fe	58.46 ± 0.75	388.85	434.43 ± 6.68	96.69
Cu	14.56 ± 1.32	61.55	73.37 ± 5.50	95.55
Zn	27.01± 2.21	66.45	96.03 ± 9.89	103.87

Mineral concentration of Agew potato (plectranthus edulis)

A wide range of mineral elements occur in fruits and vegetables, which are a primary nutritional source. Potatoes are an important source of different dietary minerals. Potatoes are listed as providing 18% of the RDA of potassium, 6% of iron, phosphorus, and magnesium, and 2% of calcium and zinc [28].

Table 9: Concentration of Metals (N = 3, In Dry Weight) in Agew Potato

Sample code	Na (mg/kg)	P (mg/100g)	Fe (mg/kg)	Cu (mg/kg)	Zn (mg/kg)
AN	261.85 ± 1.29^{a}	88.54 ± 6.15^{a}	23.74 ± 0.60^{a}	12.29 ± 0.05^{b}	19.76 ± 0.22^{a}
BA	319.13 ± 13.76^{b}	82.02 ± 10.42^{a}	42.12 ± 0.39^{b}	9.98 ± 0.59^{a}	17.68 ± 0.79^{a}
DA	603.90 ±5.92^{c}	251.82 ± 4.76^{b}	65.36 ± 1.08^{c}	16.27 ± 1.43^{c}	30.20 ± 2.48^{b}

Data are expressed on a dry weight. Data are mean ± SD. Different letters within one column denote statistically significant differences (P≤0.05) by ANOVA and Tukey's test. Various factors, such as soil pH, organic matter content, plant species and cultivars, fertilizers, herbicides, and agricultural practices, have an impact on the concentration of minerals. The mineral and nutrient contents of Agew dinch samples in three different locations are given in Table 9. The highest Na, P, Fe, Zn, and Cu contents in Agew potato samples were recorded in DA.

The concentration of sodium was significantly different in Agew potatoes (P ≤ 0.05). The range for the concentration was 261.85 ± 1.29 to 603.90 ± 5.92 mg/kg. According to the finding, DA has the highest concentration of sodium levels (603.90 ± 5.92 mg/kg), whereas AN has the lowest concentration (261.85 ± 1.29 mg/kg). The phosphorus content of Agew potato tubers was analyzed and shown in Table 9. The data indicated that the concentration of phosphorus varied significantly. The phosphorus concentration of the selected sample was found to vary between 82.02 ± 10.42 and 251.82 ± 4.76 mg/100g. When compared to samples AN, BA, and DA, the phosphorus level of DA significantly differed (P ≤ 0.05), while sample AN and BA differences were not statistically significant (P > 0.05). DA had the highest phosphorus levels (251.82 ± 4.76 mg/100g), whereas BA had the lowest levels (82.02 ± 10.42 mg/100g). According to research papers, potato phosphorus ranges from 1300-6000 µg/g (130-600 mg/100g). Besides potassium, phosphorus is the main mineral present in the tubers. It has many roles in the human body and is a key player for healthy cells, teeth, and bones [29].

The Fe found in potato tubers is considered to be non-heme iron and, therefore, a valuable source of iron for the human diet. The lack of iron can cause severe health problems, including impaired development in adolescence and reduced work capacity, making potatoes useful sources for such nutrients. Moreover, as potatoes also contain vitamin C, which increases the iron uptake from potatoes, the health benefits are significant against anemia. Based on the results of the study, the iron contents of Agew potato significantly differed (p≤0.005) among the three sample locations, which means the range of iron contents varied between 23.79±0.60 and 65.36±1.08 mg/kg. The highest iron contents were found in DA (65.36 ± 1.08), whereas the lowest iron concentration was found in AN (23.79 ± 0.60 mg/kg). Concentrations of Fe in samples were higher than concentrations of other trace metals (Table 9). Most Ethiopian soil has a pH of 5.6–7.3, which is slightly acidic to slightly basic. Plants growing in these soils accumulate more Fe. The high concentration of these metals in Agew dinch (P. edulis) may be due to Fe being easily transferred from soil to the plant and accumulating in P. edulis tubers. Zinc is needed for the body's immune system to properly work and is involved in cell division, cell growth, and wound healing [30,31].

The results of the zinc contents, which are shown in Table 9, revealed that Agew dinch tubers have significantly different (P≤0.05) values among the sample regions. The concentrations of Zn ranged from 19.76 ± 0.22 to 30.20 ± 2.48 mg/kg. The maximum amount of Zn level was found in Dangila (30.20±2.48 mg/kg), whereas Ankesha had the lowest Zn concentration, which was 19.76 ± 0.22 mg/kg. The finding results were different insignificantly from those of other scholars. This difference is caused by variety, location, genotype, fertilizers, and other factors.

Copper is needed for the synthesis of hemoglobin, proper iron metabolism, and maintenance of blood vessels. Table 9 shows the results of the copper content of Agew potato at three sample locations. The findings revealed that the copper concentration of Agew potato, which ranges from 9.98 ± 0.59 to 16.27 ± 1.43 mg/kg, considerably varies between those sample locations. The sampling site of DA had the maximum amount of copper (16.27 ± 1.43 mg/kg), and the minimum amount of copper was found in the sample location of BA (9.98 ± 0.59 mg/kg). Copper is one of the essential micronutrients, and its adequate supply for growing plants should be ensured through artificial or organic fertilizers. Cu occurs in compounds with no known functions as well as enzymes having vital functions in plant metabolism. The quantification of minerals in potatoes grown in Ethiopia is relevant because various factors affect their concentration in the plant. These facts may include soil pH, cation exchange capacity, organic matter content, types and varieties of plants, fertilizers, pesticides, and agricultural practices [32-34].

Conclusion

Agew dinch (Plectrantus edulis) is one of the local tubers in Ethiopia and has a long history of local usage. This study enables the compilation of basic information regarding the nutritional makeup of Agew potato in the Awi zone. It is possible to draw the following conclusions from nutritional evaluation studies of Agew dinch tubers obtained from AN, BA, and DA: Both sample areas were determined to be a good source of nutrients such as carbohydrate, crude protein, crude fiber, crude ash, crude fat, sodium, copper, iron, zinc, and phosphorus. The nutritious and anti-nutritional elements discovered in both the sample region and the examined tubers had approximative values when compared to data on other potato kinds. The nutritional content of Dangila varied significantly among the samples examined for this investigation. This means Dangila demonstrated outstanding nutritional qualities as it placed best in proximate composition, micronutrient, and macronutrient contents. But the highest levels of tannin, fiber, and carbohydrate were found in Banja. The nutritional content of Agew dinch from the other two sample areas (Ankesha and Banja) had relatively the same nutritional compositions. The findings of this study highlight the important role that Agew dinch can play in enhancing human nutrition. Variety and landraces had an impact on the nutritional value of tubers. A well-defined tendency toward antioxidant activity and total phenolic compounds was observed. Generally, the results of this study may help Ethiopian producers and consumers diversify their potato usage.

Acknowledgements
The authors would like to express their gratitude to the Debre Birhan University's department of chemistry and the Ethiopian Public Health Institute's food science and nutrition laboratory for providing the funding necessary to complete this research. Additionally, the authors extend their acknowledgement to the laboratory staff of the Food Science and Nutrition Department at the Ethiopian Public Health Institute for their valuable technical assistance in successfully conducting this investigation.

Author Contributions
Misganaw Andualem contributed to investigation, Experimental design, data conceptualization, data interpretation, article drafting manuscript, and development of laboratory tool, performed laboratory analysis. Buzuayehu Tadesse contributed to the design, analysis, and interpretation of the findings, reviewed progressive drafts, and proof read the manuscript. Abebe Bitew contributed methods validation, prepared reference samples, performed laboratory analysis, and cleaned data. All authors read and approved the final manuscript

Funding
Ethiopian Public Health Institute's food science and nutrition provide the supply of for the achievements of the study. The authors received no financial support for the study design, lab analysis study, data analysis, publication decision, or manuscript preparation.

Availability of Data and Materials
The data sets analyzed for this study are available with the corresponding author, which can be accessed on reasonable request.

Declarations
Ethics Approval and Consent to Participate
Not applicable

Consent for Publication
Not applicable

Conflicts of Interest
The authors declare no conflicts of interest

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