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Original Research Article

Elemental and Polyphenol Contents of Green Coffee Beans from Central Gondar Zone, Ethiopia

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ABSTRACT

Essential metals (Ca, Mg, Cu, Zn, Fe, Mn) and total polyphenols were determined in green coffee beans from four districts of Central Gondar Zone, Ethiopia (Gondar Zuria, Takusa, Tach Armachiho, Chilga). Elemental contents of samples were analyzed using flame AAS, after optimized wet digestion with spike recoveries of 85–116%, and polyphenols using UV–Vis spectroscopy. Coffee beans contained high levels of Ca and notable amounts of other elements, with average concentrations (mg/kg) of 2876–3515 (Ca), 839–877 (Mg), 71.8–140 (Fe), 31.6–100 (Cu), 21.0–24.2 (Mn), and 10.0–13.3 (Zn). Takusa beans showed significantly higher Fe and Ca. The polyphenol contents ranged from 43.1±0.7–46.7±10 mgGAE/g, without significant difference ($p < 0.05$) across districts.

Keywords: Green coffee beans, Metals, Total polyphenol, Central Gondar, Ethiopia

INTRODUCTION

Coffee is the most commercialized food product and one of the most widely consumed beverages worldwide. It ranks second only to petroleum among globally traded commodities (Mehari et al., 2016a). Of the numerous coffee species, only Arabica (Coffea arabica) and Robusta (Coffea canephora) are of major commercial importance, accounting for approximately 75% and 25% of global production, respectively (Asfaw et al., 2019). Arabica is predominantly cultivated in upland and highland regions, particularly in East Africa, whereas Robusta thrives in the lowlands of Central and West Africa (Grzeskowiak et al., 2014). Owing to its superior sensory qualities, Arabica coffee is generally preferred by consumers over Robusta (Sabah et al., 2019).

The word coffee is believed to originate from the Ethiopian province of Keffa, where legend attributes its discovery to a shepherd in the 6th century (Amamo, 2014). In Ethiopia, coffee remains a popular beverage, with annual domestic consumption estimated at over 9 million tons (Sabah et al., 2019).

Coffee contains a wide range of chemical constituents, among which key bioactive compounds are phenolics (e.g., chlorogenic acids), methylxanthines (caffeine, theophylline, theobromine), diterpenes (cafestol, kahweol), trigonelline (a precursor of vitamin B3), as well as minerals such as magnesium and potassium (Mehari et al., 2016a, 2016b). Compounds like caffeine, trigonelline, and chlorogenic acids contribute to acidity, bitterness, and astringency, thereby shaping coffee flavor (Mehari et al., 2016b). Caffeine, for instance, acts as a stimulant of the central nervous system, heart rate, and respiration, while chlorogenic acids, esters of caffeic and quinic acids, display antioxidant, antibacterial, and anti-carcinogenic activities (Arai et al., 2015). The flavor and aroma of coffee are further influenced by volatile and nonvolatile components, including proteins, amino acids, fatty acids, and additional phenolic compounds (Dechassa et al., 2018). In addition to genetic factors, the chemical profile of green coffee beans is strongly shaped by environmental growing conditions (Mehari et al., 2016c).

Coffee is widely regarded for its diverse health benefits, largely linked to its strong antioxidant activity. This activity is primarily attributed to polyphenolic acids such as chlorogenic, ferulic, caffeic, and p-coumaric acids (Yashin et al., 2013). Increasing evidence suggests that diets rich in antioxidants from plant-based foods can help prevent oxidative stress and related diseases by functioning as free radical scavengers, chain reaction inhibitors, metal chelators, oxidative enzyme inhibitors, and enzyme cofactors (Karadag et al., 2009). Accordingly, coffee serves as an important dietary source of polyphenols capable of reducing free radical formation, thereby protecting DNA integrity and preventing genetic damage (Yashin et al., 2013).

Coffee is also a source of essential mineral elements, including K, Ca, Mg and Fe, which contribute to its nutritional value (Mehari et al., 2016c). These elements are absorbed from the soil and accumulated in varying amounts, depending on factors such as climate, elevation, geology, soil composition, and agricultural practices, including fertilizer and pesticide use (Habte et al., 2016). Determining their concentrations is therefore important for evaluating the nutritional quality of coffee and assessing potential health risks when present at elevated levels (Sabah et al., 2019).

Coffees from different areas of the Central Gondar Zone, Ethiopia, are known for their distinct flavors, with some areas particularly reputed for producing beans of high quality. Since beverage quality is closely linked to the biochemical composition of green beans, coffees with unique flavor profiles are expected to exhibit distinct chemical characteristics. However, no prior studies have examined the biochemical composition of coffee beans from this zone. This study, therefore, aimed to determine the elemental and total polyphenol contents of green coffee beans from major producing areas of Central Gondar Zone and to assess the influence of geographic origin on their chemical composition.

MATERIALS AND METHODS

Coffee Samples

Coffee samples were collected from four districts of the Central Gondar Zone (Figure 1). These districts were selected for their relatively high coffee productivity. In total, 23 samples were collected from the 2019/2020 crop season, between December 2019 and February 2020. Each sample consisted of 250 g of coffee cherries, stored in polyethylene bags at room temperature until analysis.

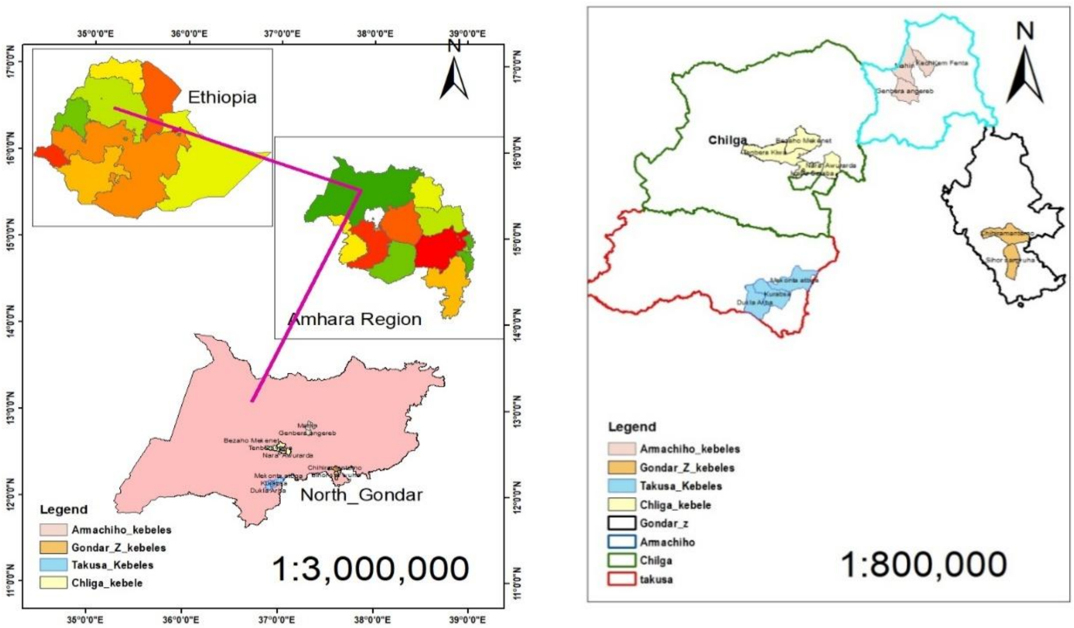

Figure 1: Map of Ethiopia showing the green coffee sample collection districts.

Coffee Sample Preparation

Coffee cherries were dried, husked with a mortar and pestle, washed with tap and distilled water, then 50 g of beans were ground and sieved through a 200 μm mesh.

Chemicals and Equipment

Folin-Ciocalteu reagent, gallic acid, and methanol (Fine Chemicals, India); sulfuric acid (98%), potassium dichromate, and nitric acid (65%) (BDH, India); standard solutions of Ca, Mg, Zn, and Mn, perchloric acid, and sodium carbonate (Blulux Laboratory, India); and standard solutions of Fe and Cu (Loba Chemie, India) were used in this study. A UV–Vis spectrophotometer (Abron Instruments, China) and a flame atomic absorption spectrophotometer (Buck Scientific 210 VGP, USA) were employed for the analyses.

Extraction of Polyphenols

Extraction of polyphenols followed a modified reported procedure (Geremu et al., 2016). In brief, 0.5 g of powdered green coffee beans was macerated with 10 mL of 80% methanol for 24 h, stirred for 30 min, and filtered through Whatman No. 1 paper.

Determination of Total Polyphenols

Total polyphenol content was determined spectrophotometrically using Folin–Ciocalteu's reagent (Asfaw et al., 2019). A 0.5 mL extract was mixed with 0.25 mL reagent and 3 mL water, followed by 1 mL of 7.5% sodium carbonate after 5 min. The mixture was incubated for 1.5 h in the dark, and absorbance was measured at 760 nm. Quantification used a Gallic acid calibration curve, and results were expressed as mg GAE/g of dry coffee beans.

Sample Digestion for Metal Analysis

Green coffee beans were digested using an optimized procedure before FAAS analysis. A 0.5 g sample was treated with 4 mL HNO_3 (69%) and 4 mL $HClO_4$ (70%) at 240 °C for 3 h 50 min. A 1% lanthanum chloride solution was added to release calcium from phosphate. The digest was filtered into a 50 mL volumetric flask and diluted to volume with distilled water. Blanks were prepared similarly using only the acid mixture.

FAAS Analysis of Metals

For FAAS analysis, the instrument was calibrated using five-point standard curves for each metal. Standard concentrations were: Ca (0.5–5.5 mg/L), Mg (0.5–6.5 mg/L), Mn (0.1–3.5 mg/L), Fe (15–35 mg/L), Cu (0.5–4.5 mg/L), and Zn (0.5–4.5 mg/L). Standards were aspirated in increasing order, and absorbance was recorded. Instrumental operating conditions are presented in Table 1.

Table 1: Instrumental operating conditions used for the determination of metals with FAAS.

Metals	Wavelength (nm)	Slit width (nm)	Lamp current (mA)	Energy (erg)
Ca	422.7	0.7	2.0	3.96
Mg	285.2	0.7	1.0	3.726
Cu	342.7	0.7	1.5	3.728
Zn	213.9	0.7	2.0	3.022
Fe	248.3	0.2	7.0	3.093
Mn	279.5	0.7	3.0	4.045

Method Validation

Method accuracy was verified by a spiking experiment, with acceptable recoveries set at 80–120%. Known volumes of standard metal solutions were added to 0.5 g of coffee powder, prepared in triplicate, digested, and analyzed by FAAS. Precision was expressed as RSD, with RSD ≤ 15% indicating good precision. Detection limits were calculated as three times the standard deviation of the blank.

Data Analysis

All measurements were conducted in triplicate, and the results are presented as mean ± standard deviation. Statistical analysis was performed using Microsoft Excel 2013, and one-way ANOVA was applied to determine significant differences among means across the coffee-growing districts.

RESULTS AND DISCUSSION

Analytical Characteristics of the Method

Linearity and Sensitivity

The calibration curves for all analyzed metals exhibited excellent linearity, with coefficients of determination (R^2) ranging from 0.9960 to 0.9996, indicating a strong correlation between absorbance and concentration. Method sensitivity, determined from the slope of the calibration curves, followed the order: Zn > Mg > Cu > Mn > Ca > Fe.

Table 2: The regression equation, correlation coefficient (R2) and limits of detection (LOD, mg/kg) of the FAAS method.

Metal	Regression equation	r^2	LOD	%Recovery
Ca	A = 0.0045C + 0.0024	0.9980	31	85
Mg	A = 0.0893C + 0.8061	0.9960	55	116
Cu	A = 0.0466C - 0.0089	0.9996	0.7	108
Zn	A = 0.1032C + 0.0116	0.9969	0.06	95
Fe	A = 0.0013C + 0.0054	0.9963	1.8	92
Mn	A = 0.0367C - 0.0031	0.9972	0.4	93

*A is absorbance, C is concentration

Precision

The precision of the method was assessed from the relative standard deviation (RSD) of triplicate analysis for each sample. The RSD values were in the range of 0.2–12%, across the different samples and metals.

Limits of detection

The LOD of the method ranged from 0.06 mg/kg for Zn to 55 mg/kg for Mg, which were low enough to enable reliable detection and quantification of the elements in green coffee bean samples.

Accuracy

The optimized digestion procedure was validated by spiking samples with standard solutions, and recoveries were in the range of 85–116% (Table 2), demonstrating that the method was reliable for all metals.

The concentrations of metals in the green coffee beans

All analyzed metals were detected in green coffee beans from all sampling districts. Calcium was the most abundant, followed by magnesium, while among trace metals, iron was highest, followed by copper. Metal concentrations (mg/kg) ranged as follows: Ca 1910–4465, Mg 791–910, Fe 40.5–251, Cu 23.4–46.3, Mn 18.4–28.4, and Zn 3.0–30.1 (Table 3).

The highest mean Ca concentration was found in coffee beans from Genbera (4465 mg/kg) in Tach Armachiho, followed by Nara-Awudarda (4273 mg/kg) in Chilga district. Magnesium was highest in beans from Dikularba (910 mg/kg) in Takusa and Eyaho (875.2 mg/kg) in Chilga. In contrast, the lowest Zn (3.0 mg/kg) and Mn (18.4 mg/kg) levels were observed in samples from Mahin and Kanfenta in Tach Armachiho.

Table 3: The concentration (mg/kg, mean±SD) of metals and total polyphenols (TPC) (mgGAE/g) in the green coffee beans.

District	Sample code	Ca	Mg	Cu	Zn	Mn	Fe	TPC
Tach Armachiho	Arm-1	2791±39	822±2.2	34.8±2.5	8.1±0.7	18.4±1.6	46.2±0.1	50.5±4.6
	Arm-2	2154±34	791±2.3	37.0±2.5	12.0±1	18.4±1.6	71.8±4.4	42.9±2.8
	Arm-3	3228±34	852±1.1	37.7±1.2	8.8±1.2	19.3±0.1	71.8±4.4	47.3±3.2
	Arm-4	3176±90	830±2.3	37.7±1.2	11.4±1.1	21.2±1.6	251±10.3	49.0±4.8
	Arm-5	3324±200	864±14	38.4±2.1	6.5±0.9	22.1±0.1	71.8±9.8	42.2±3.4
	Arm-6	3532±181	872±2.6	43.4±1.2	13.0±1.7	23.9±3.1	123±6.9	48.2±4.4
	Arm-7	4465±105	838±5.6	34.8±2.5	16.9±1.0	22.1±0.1	46.2±0.5	43.7±3.8
	Arm-8	3324±267	847±1.7	46.3±1.2	3±0.4	22.2±1.6	46.2±0.5	48.5±3.1
Chilga	Chi-1	1910±136	808±3.2	29.8±2.1	3.3±0.4	20.3±1.6	46.2±0.5	47.2±3.6
	Chi-2	2569±22	842±3.0	36.0±0.1	9.4±1.3	22.1±0.1	46.2±0.5	43.7±3.0
	Chi-3	2999±13	875±3.4	37.0±1.2	5.9±0.7	26.6±1.6	71.8±2.4	39.6±2.2
	Chi-4	3495±34	857±3.4	33.4±3.3	10.4±1.5	23±1.6	46.2±0.5	51.9±4.3
	Chi-5	3110±90	845±4.0	33.4±3.3	9.1±0.7	20.3±2.1	46.2±0.5	51.5±4.5
	Chi-6	2495±90	849±4.0	34.8±1.2	6.2±0.9	19.3±2.7	46.2±0.5	47.8±3.3
	Chi-7	3228±46	871±1.1	34.1±0.1	11.0±1.0	23.0±1.6	46.2±0.5	48.5±4.1
	Chi-8	2984±13	867±1.7	38.4±3.7	11.0±1.0	21.2±1.6	97.4±8.8	46.8±3.7
	Chi-9	3147±44	849±2.2	34.1±2.1	10.4±1.5	23.0±3.1	46.2±0.5	45.4±2.8
	Chi-10	4273±71	858±3.9	36.3±0.1	30.1±0.6	22.1±0.5	405±44.4	44.9±2.9
Takusa	Tak-1	2910±51	852±0.6	34.1±2.1	10.1±1.0	23.9±1.6	46.2±0.5	31.5±1.9
	Tak-2	3636±102	910±1.9	234±3.7	14.6±2.0	28.4±1.6	185±13.3	47.0±3.7
	Tak-3	3999±46	870±1.9	32.7±1.2	13.6±0.6	20.3±1.6	190±17.8	52.5±5.0
Gondar Zuria	Gon-1	2873±22	842±2.3	29.1±1.2	9.1±1.1	20.3±1.6	71.8±8.1	43.6±3.1
	Gon-2	2880±22	849±1.9	34.1±2.1	17.5±0.6	23.0±1.6	71.8±8.5	42.6±2.8

The overall mean concentrations of metals in green coffee beans from the four districts are shown in Table 4. Mean concentration (mg/kg) ranged from Ca, 2876 (Gondar Zuria) to 3515 (Takusa); Mg, 839.4 (Tach Armachiho) to 877 (Takusa); Cu, 31.6 (Gondar Zuria) to 100 (Takusa); Zn, 10.0 (Tach Armachiho) to 13.3 (Gondar Zuria); Mn, 21 (Tach Armachiho) to 24.2 (Takusa); and Fe, 71.8 (Gondar Zuria) to 140 (Takusa). Iron and Ca levels in beans from Takusa were significantly higher than in the other districts. These variations likely reflect differences in environmental conditions and the genetic characteristics of coffee beans across growing regions (Endaye et al., 2019).

Table 4: The concentration (mg/kg, mean±SD) of elements and total polyphenols (mgGAE/g) in the coffee beans.

District	Ca	Mg	Cu	Zn	Fe	Mn	Polyphenol
Tach Arimachiho	3249±42	839±1.4	38.8±0.64	10.0 ±0.35	91.1±1.6	21.0±0.42	46.5±3.1
Chilga	3021±15	852±0.95	34.7±0.54	10.7±0.28	53.3±1.8	22.1±0.50	46.7±3.6
Takusa	3515±38	877±1.2	30.1±1.3	12.8±0.69	140±5.8	24.2±0.92	43.7±11
Gondar zuria	2876±16	845±1.4	31.6±1.2	13.3±0.56	71.8±5.9	21.6±1.1	43.1±0.7

Comparison of Elemental Contents in Green Coffee with Literature Data

Several studies have documented the concentrations of metals in green coffee beans from various regions worldwide. For example, in Vietnam, the reported concentrations (mg/kg) were Ca 768, Mg 683, Cu 17.4, Zn 5.97, Fe 42.98, and Mn 10.4 (van Cuong et al., 2014), which are generally lower than the levels found in this study for the Central Gondar Zone of Ethiopia. Similarly, in the Sidama region of Ethiopia, green coffee beans contained Mg 1670 ± 20, Ca 880 ± 10, Mn 19.0 ± 1.0, Fe 26.2 ± 1.5, Cu 22.9 ± 2.2, and Zn 21.1 ± 0.1 mg/kg (Gure et al., 2017). These values are also mostly lower than those observed in Central Gondar, except for Mg and Zn. Overall, these comparisons suggest that green coffee beans from the Central Gondar Zone are relatively rich sources of both major and trace metals.

Tootal Polyphenols

The calibration curve for total polyphenol determination was linear over the concentration range of 25–300 mg/L of gallic acid (Figure 2), with a correlation coefficient (R^2) of 0.9989.

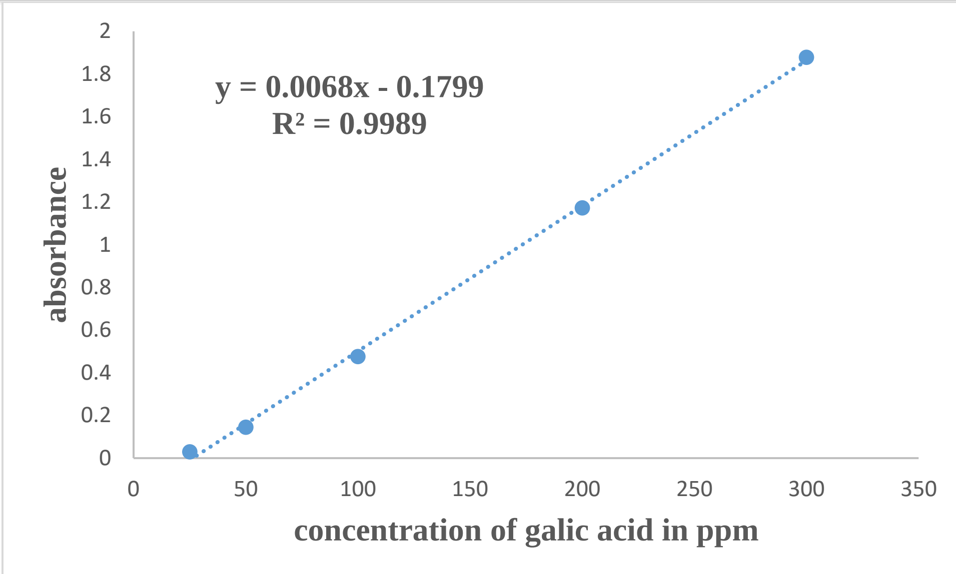

Figure 2: The calibration curve of Gallic acid.

Total Polyphenol Contents of Green Coffee beans

The total polyphenol content (TPC) of green coffee beans ranged from 31.5 to 52.5 mg GAE/g (Table 3), with the highest value observed in Takusa and the lowest in Dembia. Mean TPC values by district (Table 4) varied from 43.1 mg GAE/g in Gondar Zuria to 46.7 mg GAE/g in Chilga, following the order Chilga > Tach Armachiho > Takusa > Gondar Zuria. One-way ANOVA results (Table 5) indicated that sampling site had no statistically significant effect ($p > 0.05$) on mean TPC among the four districts.

Table 5: ANOVA table obtained from the analysis of the variation of mean total polyphenol contents among green coffee beans from the four districts studied. Difference is significant when $p < 0.05$.

Source of Variation	SS	df	MS	F	*P*	F_{crit}
Between Groups	40.73097	3	13.57699	0.605765	0.619317	3.12735
Within Groups	425.8464	19	22.41297			

The total polyphenol contents (TPCs) determined in the present study were compared with previously reported values. The highest mean TPC obtained in this study (46.76 ± 3.44 mg GAE/g) was high than those reported for other Ethiopian green coffee beans from different production regions (29.4–38.5 mg GAE/g) (Mehari et al., 2020), while lower than that reported for Sidama (49.2±0.70 mg GAE/g) and Yirgacheffe (54.5±1.6 mg GAE/g) by Bobkova et al. (2020). Comparatively, much higher TPC values were documented for Brazilian green coffee (746 ± 10.5 mg GAE/g) (Nassar et al., 2019). Conversely, lower TPC values were reported for Indonesian green coffees, including Jember (33.2 mg GAE/g), Malang (3.9 mg GAE/g), Bondowoso (21.9 mg GAE/g), and Banyuwangi (7.7 mg GAE/g) (Perdani et al., 2019). These variations in TPC may be attributed to differences in geographical and environmental conditions, cultivation practices, and post-harvest handling. Previous research has also shown that the phenolic content of coffee beans is influenced by their origin and the type of solvent used during extraction (Daniel et al., 2017).

CONCLUSION

In this study, the levels of some major (Ca and Mg) and minor (Fe, Cu, Zn and Mn) metals and total polyphenols in green coffee beans from the major production areas of Central Gondar Zone were determined. Despite the initial hypothesis, no significant variation was observed in the determined concentrations of total polyphenols and metals, except Fe, with the different growing areas.

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