Journal of Science and Sustainable Development (JSSD), 2019, 7(1), 1-9

ISSN: 2304-2702 (print); 2414-4479 (online)
DOI: https://doi.org/10.20372/au.jssd.7.1.2019.091

Biochemical Composition of Green Coffee Beans as Geographic Indication in Ethiopian Specialty Coffees

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Abstract

Biochemical composition of green coffee beans can be used as reliable geographical indicators to characterize quality. No efforts have been made in Ethiopia to elucidate the biochemical composition of green coffees based on geographical origin. The objective of this research was to determine the biochemical composition of green coffee beans in Ethiopia based on location. Green coffee beans were obtained from three replicated sites per farm from a total of 24 farms in south-western (SW), western (W), southern (S), north-western (NW) and eastern (E) major coffee growing regions of Ethiopia. Handpicked coffee cherries at peak maturity were used for the study during the 2010/2011 growing season. The study followed a multistage purposive random sampling method in a nested design. The results of the study revealed that location significantly ($P<0.05$) affected the values of caffeine, 5-CQA (5-Caffeoylquinic acid), Feruloylquinic acids (FQA), 3,4-Dicaffeoylquinic acids (3,4-DCQA) and 4,5-Dicaffeoylquinic acids (4,5-DCQA). Green coffee bean composition of 4-CQA (4-Caffeoylquinic acid), 5-CQA, 4,5-DCQA, caffeine, FQA, and 3-CQA in a descending order of importance contributed much to separate coffees originating from the eastern region distinctly from those originating from both the western and southern regions. However, 4-CQA, 3-CQA (3-Caffeoylquinic acid), FQA, 4,5-DCQA, 5-CQA, and caffeine in their descending order of importance contributed much to separate coffees originating from the south-western region from those originating from both the southern and north-western regions. However, a repeated and an integrated study is sought to come to a comprehensive conclusion and recommendation.

Keywords: Bean chemistry, Coffee appellations, Coffee origins, Flavour precursors, Location

1. Introduction

Coffee is one of the most popular beverages in the world. Economically, coffee is the second most exported commodity after oil, and employs over 100 million people worldwide (Gray *et al.,* 2013). In Ethiopia, coffee is the most important export commodity, with a share of 20-25% of the total foreign exchange earnings. At least 15 million people also directly or indirectly rely on coffee for their livelihood (MoT, 2012).

As the county of origin for crop, Ethiopia produces premium quality coffee. It is the leading producer in Africa, and the 5th in the world, following Brazil, Vietnam, Colombia and Indonesia. However, consider Arabica alone, Ethiopia is the 3rd largest producer after Brazil and Colombia (ICO, 2015). Ethiopia also has the largest highland area suitable for Arabica coffee production and, hence has the potential to be a leading producer in both quality and quantity. The trends in quantity and quality of coffee produced for export is increasing in Ethiopia for over the past decade (MoT, 2012). The production has increased from around 160,000 tons in 2001 to over 400,000 in 2014, while areas under coffee has increased from 250,000 ha in 2000 to around 800,000 ha in 2014 (FAOSTAT, 2015).

Tesfaye (2006) reported high genetic variability within and between different wild populations in Ethiopia. The presence of high genetic variation in natural coffee populations in the forest and semi-forest systems can partly be attributed to the presence of a wide ecological variation, ranging from 1000 m to 2000 m in altitude, with highly dissected and rolling topography exhibited by ranges of temperature and humidity gradients as well as

soil variability (Adugnaw *et al*., 2015). In Ethiopia, there are different coffee types recognized by their origin and quality, and used as trade names. These include Bebeka, Harar, Jimma, Kaffa, Lekemti, Limmu, Sidama, Teppi, and Yirgacheffe. Under each coffee type, 2-5 different local types are recognized (Tesfaye, 2006). Such a high level of diversity is partly attributed to not only due to genetic and agro ecological diversity but also due to the presence of indigenous traditional production systems of coffee in the country.

Description of coffee relates not only to the species or varieties but also to the geographical origin and the environmental conditions (Rubayiza and Marco, 2005; Petit, 2007). Moreover, it is often reported that Arabica coffee trees at high elevation are known to affect the final quality of the beverage favourably (Petit, 2007). Hence, discrimination methods of coffee origins may fetch premium prices for quality coffees. An increasing interest in geographical indications of origin (GIs) as a tool of product differentiation is sought to establish coffee appellation systems in the specialty coffee sector (Osario, 2002).

Several attempts have been made to determine the origin of green and roasted coffee beans. Analytical methods such as gas chromatography-mass spectrometry (GC-MS) (Costa Freitas *et al.*, 2001) and near infrared spectroscopy (NIR spectroscopy) (Bertrand *et al.*, 2005) were applied for the determination of organic compounds such as fatty acids profiles (Martín *et al.*, 2001), and tocopherols and triglycerides (González *et al.*, 2007). Stable isotope ratios of carbon, nitrogen and oxygen of specific compounds extracted from green coffee beans were studied with promising results (Weckerle *et al.*, 2002).

Certain biochemical constituents, such as trigonelline, sucrose and chlorogenic acids, are flavour precursors, whereas others such as caffeine play a role in the bitterness of the roasted coffee, and fat helps in fixing the flavour compounds formed during roasting (Perrone *et al*., 2008). Hence, biochemical composition of green coffees can be used to characterize quality. Similarly, post-harvest treatments of the beans are known to affect the generation of flavour (Joët, *et al*., 2010). Furthermore, it is reported that caffeine, most chlorogenic acid (CGA) isomers, and fatty acid (FA) were unaffected with post-harvest treatment but sorbitol content after wet processing depend on the glucose content of fresh beans (Joët, *et al*., 2010).

Even though the biochemical composition of green coffee beans has been employed for origin discrimination, neither the composition of Ethiopian specialty coffees, nor origin discrimination, based on these constituents has been done in the country. Hence, the objective of this study was to elucidate the biochemical composition of green coffee beans and to discriminate different coffee origins in Ethiopia.

2. Materials and methods

2.1. Site selection and sample preparation

Twenty-four coffee farms (Figure 1, and Table 1 and 2) were purposively selected from the area spanning 3^0 30' to 14^0 55' North latitude, and 33^0 to 48^0 East longitude in Ethiopia. The experiment was laid out in nested Design with three replications. Each farm was stratified into top (replication 1), middle (replication 2) and bottom (replication 3) position of slope inclination. Ripe coffee cherries (6 kg/replication) were handpicked at their fresh red stage during the 2010/11 cropping season.

The fresh cherries were pulped the same date they were picked to process in wet processing method. When the fermentation was complete the parchment coffees were sundried and parchments mechanically removed. Green coffee beans were subjected to freeze drying at pressures and temperature below triple point (eutectic) i.e. 4.579 mm of Hg and 0.0099 degree Celsius, respectively just before grinding to fine powder using a hand-held electrical blade coffee grinder (Bosch MKM 6003 UC, Robert Bosch Hausgeräte GmbH, Germany). Grinding was assumed to be sufficient when the powder escaped to the ceiling of the cap of the grinder, and the

powder was immediately packed in a plastic cup with a tight stopper, and kept at room temperature until laboratory analysis.

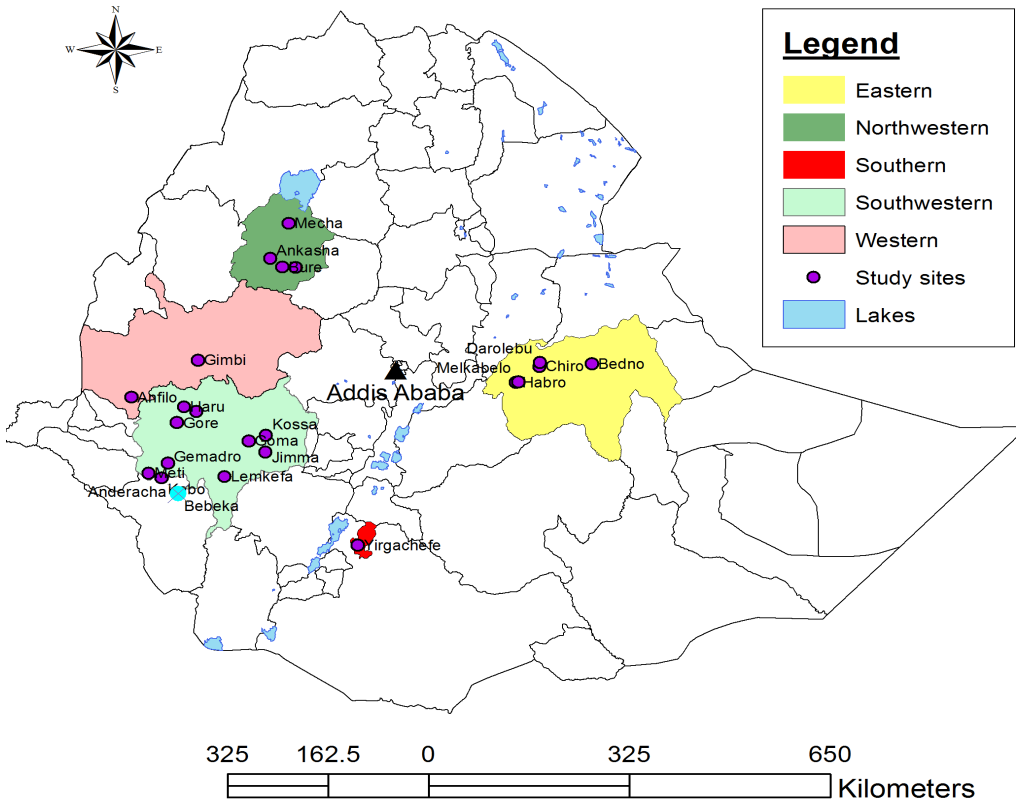


Figure 1. Green coffee bean sample collection sites.

Table 1. Mean annual weather data and altitudinal range of the locations

Locations	Climatic Factors					Altitude (m a.s.l.)
	RF (mm)	MAX (ºC)	MIN (ºC)	RH (%)	Sunshine (HRS/day)	
Eastern (26 yrs)	643.7	27.8	12.8	NA	NA	1874 - 2266
North-western (23 yrs)	1140.5	NA	NA	NA	NA	1774 - 2000
Southern (30 yrs)	1345.7	26.6	11.7	69	5.7	2091
South-western (30 yrs)	1564.9	26.1	13.2	73.3	5.4	1150 - 1820
Western (28 yrs)	1385.2	25.6	13.9	70.5	6.5	1800 - 1907

Source: NMSA, Ethiopia (2010); NMSA = National Meteorological Services Agency; NA = data not available; RF mm= rainfall in mm; MAX (^{0}C) = Maximum temperature; MIN (^{0}C) = Minimum temperature; RH (%) = Relative humidity; HRS = hours; m a. s. l. = metres above sea level; yrs = years.

Table 2. Origins of unwashed green coffee beans used for the study in Ethiopia

Farm	N	Location	Administrative Region	Administrative Zone	Latitude	Longitude
Bebeka	3	Southwest	SNNP	Benchmaji	6.99442	35.5684
Anderacha	3	Southwest	SNNP	Godere	7.23987	35.3169
Kabo	3	Southwest	SNNP	Godere	7.24124	35.3197
Meti	3	Southwest	SNNP	Godere	7.32297	35.1288
Gemadro	3	Southwest	SNNP	Sheka	7.48639	35.4131
Lemkefa	3	Southwest	SNNP	Kafa	7.27274	36.2427
Jimma	3	Southwest	Oromia	Jimma	7.67884	36.8385
Kossa	3	Southwest	Oromia	Jimma	7.95452	36.8468
Goma	3	Southwest	Oromia	Jimma	7.85752	36.5885
Yayo	3	Southwest	Oromia	Illuababora	8.33601	35.8226
Gore	3	Southwest	Oromia	Illuababora	8.14905	35.5369
Gimbi	3	West	Oromia	West Wollega	9.17125	35.8359
Haru	3	West	Oromia	West Wollega	8.40717	35.6396
Anfilo	3	West	Oromia	Kelem Wollega	8.55386	34.8651
Yirgacheffe	3	South	SNNP	Gedeo	6.15848	38.1958
Jabi	3	Northwest	Amhara	West Gojam	10.6918	37.2665
Bure	3	Northwest	Amhara	West Gojam	10.7003	37.0668
Ankasha	3	Northwest	Amhara	West Gojam	10.8436	36.8914
Mecha	3	Northwest	Amhara	West Gojam	11.417	37.1557
Chiro	2	East	Oromia	West Hararghe	9.06989	40.8646
Habro	3	East	Oromia	West Hararghe	8.81697	40.5167
Darolebu	3	East	Oromia	West Hararghe	9.14549	40.8691
Melkabelo	2	East	Oromia	East Hararghe	8.82672	40.5499
Bedeno	3	East	Oromia	East Hararghe	9.11447	41.6335
Total	70					

2.2. Chemical analyses: Caffeine and Chlorogenic acids

The caffeine and chlorogenic acid contents were determined using HPLC/THERMO following the method of Alonso-Salces et al. (2009) and sucrose measurement using GC (VARIAN 3800, Varian, Inc. USA) following the standard method (Adugnaw, 2014).

2.3. Statistical analysis

One-way analysis of variance and canonical discriminant function analysis were applied to study the significant differences in the biochemical composition of green coffee beans from the different locations (provenance). Moreover, percentage contribution of predictors to variation of biochemical composition was done using SPSS, version 16.2. Mean separation was done by Tukey HSD.

3. Results

Location had a significant ($P<0.05$) effect on caffeine, 5-CQA (5-Caffeoylquinic acid), FQA (Feruloylquinic acids), 3,4-DCQA (3,4-

Dicaffeoylquinic acids), and 4,5-DCQA (4,5-Dicaffeoylquinic acid; CFQA), but it had non-significant effects on CFQA (Caffeoylferuloylquinic acids), TCGA (total chlorogenic acid) and sucrose contents of green coffee beans (Table 3). With regard to caffeine content of green coffee beans, both the eastern and southern regions showed significant differences from all others except with each other. The highest caffeine content (14.6±0.2) followed by North-western (14.5±0.4) and South-western (13.2±0.2), which are statistically in parry. The mean value of caffeine content from coffees originating from the south-western region was significantly higher than the mean values from the southern and eastern regions while the later were statistically in parry. The southern region could also be separated by its significantly higher mean value from East and southwest, whereas the rest remain inseparable with respect to 5-CQA content of green coffee beans (Table 3).

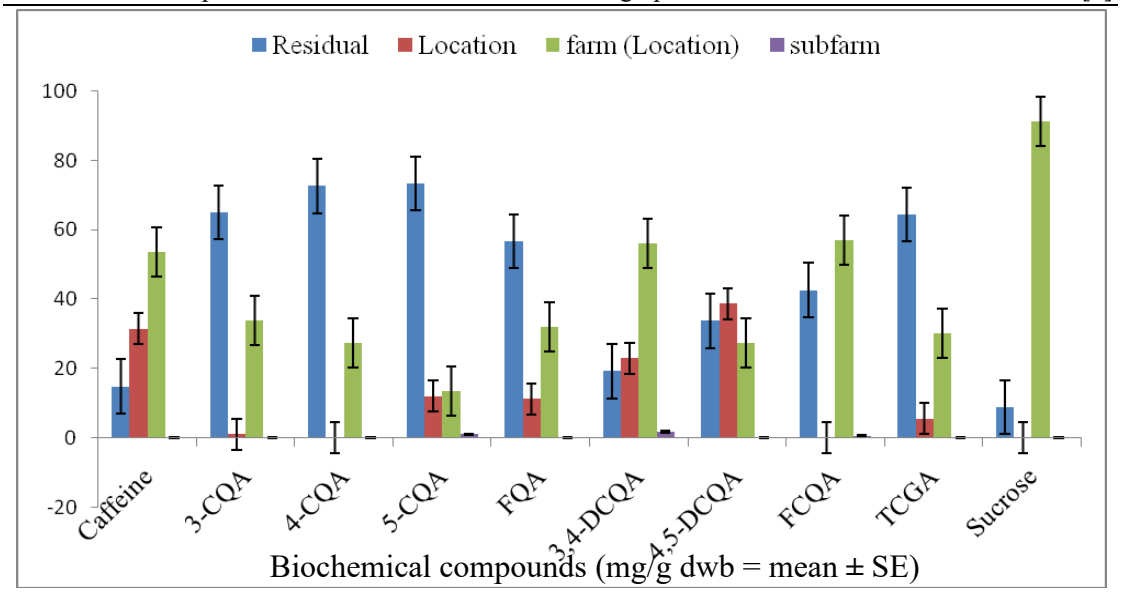
The southern region showed significantly smaller mean value (3.4±0.1) as compared to eastern (4.3±0.1) and south-western regions (4.1±0.1) for FQA content of green coffee beans. On the other hand, eastern could be separated based on its highest value of 3,4 - DCQA content of green coffee beans from both southern and western locations. However, eastern and north-western regions were inseparable based on the mean value of 4, 5-DCQA content of green coffee beans, with both of them being significantly higher as compared to that of western region. Moreover, eastern region could be further separated from the southern and south-western region with regard to 4,5-DCQA content of green coffee beans (Table 3).

Table 3. Biochemical composition of unwashed green coffee beans by geographical origin (mg/g dwb)

Location (N)	Caffeine	3-CQA	4- CQA	5- CQA	FQA	3,4-DCQA	4,5-DCQA	CFQA	TCGA	Sucrose
E (13)	12.0±0.4 b	3.7±0.17 a	5.8±0.2 a	27.0±0.2 b	4.3±0.1 a	3.4±0.1 a	9.0±0.6 a	3.9±0.3 a	57.1±0.8 a	47.9±1.3 a
NW (12)	14.5±0.4 a	3.3±0.08 a	5.8±0.1 a	28.7±0.4 ab	3.7±0.1 ab	2.8±0.1 ab	8.4±0.3 ab	3.7±0.3 a	56.5±0.6 a	50.3±2.8 a
S (3)	12.0±0.3b	3.6±0.10 a	6.0±0.1 a	29.8±0.1 a	3.4±0.1 b	2.4±0.1 b	6.4±0.3 bc	3.4±0.1 a	54.8±0.3 a	45.9±0.7 a
SW (33)	13.2±0.2 ab	3.8±0.12 a	5.7±0.1 a	27.2±0.5 b	4.1±0.1 a	2.8±0.1 ab	6.4±0.3 bc	3.8±0.2 a	53.8±1.0 a	46.6±1.4 a
W (9)	14.6±0.2 a	3.5±0.22 a	5.7±0.2 a	28.9±0.4 ab	4.0±0.1 ab	2.2±0.1 b	5.7±0.3 c	3.4±0.1 a	53.4±0.5 a	43.3±2.1 a
P	<0.0001	0.262	0.974	0.017	0.005	<0.0001	<0.0001	0.644	0.094	0.286
SD	1.54	0.62	0.66	2.14	0.48	0.70	1.92	0.92	4.44	7.40
CV (%)	11.61	17	11.56	7.71	11.90	24.74	26.95	24.64	8.09	15.73

Means with the same letter within a column are not significantly different ($P < 0.05$) SPSS (Tukey HSD）; Where, dwb = dry weight basis;; 3-CQA = Caffeoylquinic acid; 4-CQA = Caffeoylquinic acid; 5-CQA = 5-Caffeoylquinic acid; FQA = Feruloylquinic acids; 3,4-DCQA = 3,4-Dicaffeoylquinic acids; 4,5-DCQA = Dicaffeoylquinic acid; CFQA = Caffeoylferuloylquinic acids; TCGA = total chlorogenic acid; E = eastern; NW = north-western; S = southern; SW = south-western; W = western.

Figure 2 shows the percentage variation in the biochemical composition of green coffee beans attributed to various origins. For caffeine, the largest variation was due to farm (location) followed by location. Again, farm (location) contributed the largest variation to the green coffee bean content of 3,4-DCQA followed by location; CFQA followed by sub farm. Most of the variations in 3-CQA contents of green coffee beans were attributed to farm (location) (33.97%) followed by location. With the exception of the fact that the unexplained error (residual) component played the highest role for 4-CQA, 5-CQA, FQA, TCGA and Sucrose, farm (location) contributed higher than by location. The variation of 4, 5-DCQA green coffee content was largely influenced by location, followed by farm (location).



3-CQA = Caffeoylquinic acid; 4-CQA = Caffeoylquinic acid; 5-CQA = 5-Caffeoylquinic acid; FQA = Feruloylquinic acids; 3,4-DCQA = 3,4-Dicaffeoylquinic acids; 4,5-DCQA = Dicaffeoylquinic acid; CFQA = Caffeoylferuloylquinic acids; TCGA = total chlorogenic acid.

Figure 2. Percentage variation in biochemical composition of green coffee beans by origin

A significant difference for biochemical composition of beans, a canonical discriminant function was conducted to examine the capacity of the compounds to separate green coffee bean origins. Accordingly, four canonical discriminant functions were employed in the analysis, where the first three showed significant contribution to discrimination of the groups. Moreover, Eigen values indicate that function1, function 2 and function 3 gave 52.5%, 38.6% and 7.9% of the variation with their respective Eigen values of 1.874, 1.377 and 0.283 associated with canonical correlation coefficients of 0.807, 0.761 and 0.470, respectively. It was noted that group centroids revealed that function 1 significantly separated eastern from both western and southern regions, while function 2 separated both southern and north-western regions from south-western region (Figure 3).

Green coffee bean constituents including 4, 5-DCQA, caffeine, 3,4-DCQA, FQA, 5-CQA with respective wilk's lambda of 0.581, 0.647, 0.735, 0.799, 0.81, 0.833 contributed significant capacity to separate the locations. The contribution of the remaining compounds, namely TCGA, 3-CQA, sucrose, CFQA and 4-CQA to origin discrimination was not found significant. The standardized discriminant function coefficients indicate the relative importance of the independent variables in predicting the dependent groups allowing the comparison of variables measured on different scales. Coefficients with large absolute values correspond to variables with greater discriminating ability. In this regard, 4-CQA, 5-CQA, 4, 5-DCQA, caffeine, FQA and 3-CQA in descending order contributed much to function 1 to separate eastern from both western and southern regions, whereas 4-CQA, 3-CQA, FQA, 4,5-DCQA, 5-CQA and caffeine in their descending order of importance contributed much to function 2 to separate south-western from both southern and north-western regions. The classification results indicate that 77.1% of original and 72.9% of cross-validated grouped cases were correctly classified. With regard to the grouping variables, 78.8% of south-western, 66.7% of western, 100% of southern, 75% of north-western, and 76.9% of eastern regions were correctly classified.

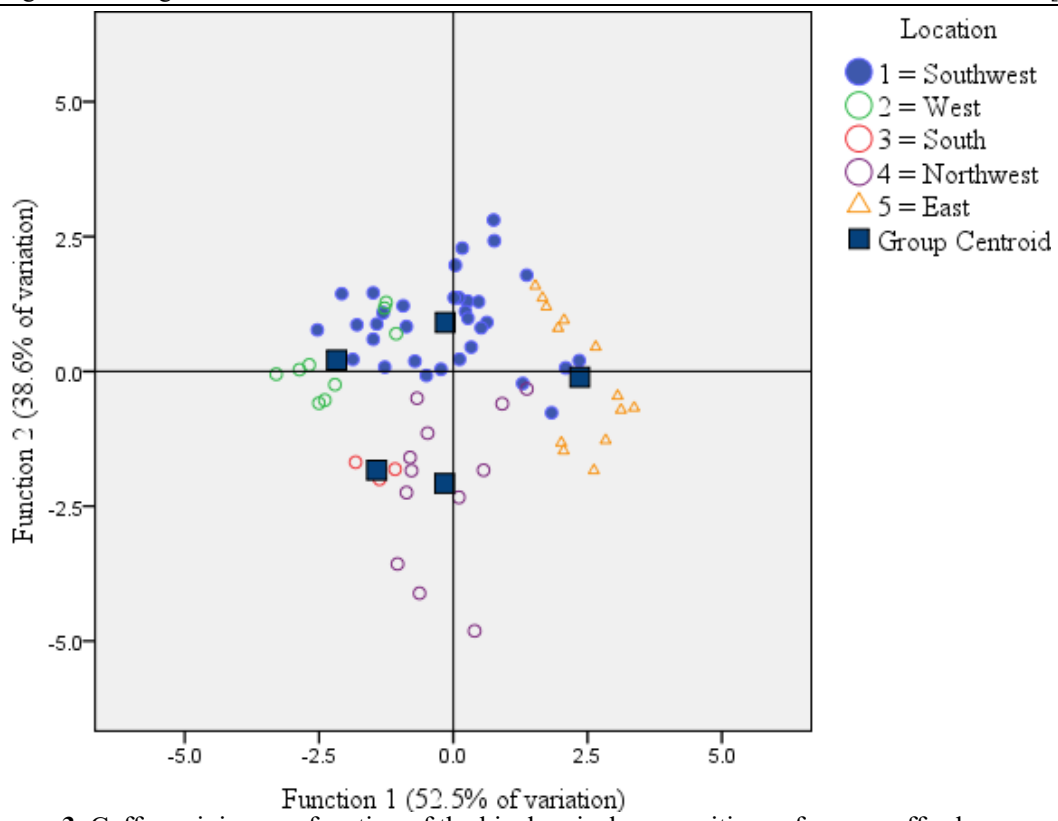


Figure 3. Coffee origins as a function of the biochemical compositions of green coffee beans.

4. Discussion

The present work determined the effect of location on the biochemical composition of green coffee beans in Ethiopian specialty coffees from 24 farms. The average trend values showed an increasing content of the biochemical compounds in green coffee beans in the order of eastern, north-western, southern, south-western and western regions for caffeine and 5-CQA, and a decreasing trend for the other biochemical compounds following the humidity gradient, which increases in that order (Table 1). Most variables indicated the highest potential of the country to deliver several specialty coffee appellations to the World coffee market which could be likely due to actual differences on agronomic management, the age of coffee trees and genetic composition, microclimate such as slope orientation, soil, and others. The work of Joët, *et al*. (2010) in Reunion Island showed that chlorogenic acids and fatty acids in the bean were controlled by the mean air temperature during bean development. By contrast, their findings revealed that total lipid, total soluble sugar, total polysaccharide and total chlorogenic acid contents were not influenced by climate, while glucose content was positively affected by altitude.

In this study, it was understood that green coffee bean origins could be well identified based on the factor loadings of canonical discriminant functions. The classification results indicate that 77.1% of original and 72.9% of cross-validated grouped cases were correctly classified. However, 21.1 % of south-western, 33.3% of western, 16.7% of north-western and 23.1% of eastern regions were misclassified likely due to the practical situation of genetic material exchange prevailed among locations. A report also showed that chlorogenic acid, fatty acids, certain elements, and caffeine were found to be indicators of certain geographical and/or varietal origins of coffees (Perrone *et al*.,

2008). The phenol and methylxanthine profiles of green coffee beans are affected by several factors, including genetic properties of the cultivars, maturity of the beans, harvesting method and postharvest processing conditions, agricultural practices, and environmental and climatic factors (Perrone *et al.*, 2008).

In this study, it was observed that the caffeine content ranges from 1.2% in south-western and southern to 1.46% in eastern region. An evaluation of the caffeine content of beans from 99 Ethiopian progenies by Silvarolla, *et al*., (2000) revealed the presences of intra- and inter-progeny variability. They found caffeine values in the range 0.46-2.82% (mean 1.18%), in 68 progenies from the Kaffa region and from 0.42 to 2.90% (mean 1.10%) in 22 progenies from Illuababora region. The concentrations of these biochemical descriptors are considered to be reliable geographical indicators as well as chemo taxonomical markers.

5. Conclusion and Recommendation

An increasing interest in geographical indications of origin (GIs) as a tool of product differentiation was sought in the specialty coffee sector to establish appellation systems for coffees. This study has demonstrated the existence of a great variability in the biochemical composition of Ethiopian green coffee beans, which is a precursor of organoleptic quality of coffee origins. The results revealed that there is a great potential for Ethiopia to supply several distinct coffee origins for the world market. The study has also indicated that method has the potential for complementary coffee quality evaluation and prediction. The method enabled us to discriminate about 70% the green coffee origins, signifying a wider application for Ethiopian specialty coffee mapping. In conclusion, the coffee origins were able to be clearly discriminated and labelled based on the biochemical composition to countercheck deliberate or accidental adulterations of lower quality coffees with higher quality coffees. This is a promising indicator of the genetic and agro ecological diversity of the crop, which is a potential resource for niche market and its genetic improvement in the country. This variability could be exploited for low-caffeine content breeding program. However, a repeated and an integrated study is sought to come to a comprehensive conclusion and recommendation.

Acknowledgements

The authors are grateful to the Ministry of Education of Federal Democratic Republic of Ethiopia as well as IUC-JU Soil Fertility Project for funding the research. Special thanks are for the laboratory facilities of Ghent University, Belgium for providing all facilities required to analyse the coffee bean samples. Prof Dr. Ir. Pascal Boeckx and Professor Jan Diels are thanked for the immense support in facilitating the laboratories in Ghent, Belgium.

References

Adugnaw Mintesnot (2014). Association of Arabica Coffee Quality with Geographic Origins in Ethiopia. PhD Dissertation, Haramaya University, Ethiopia.

Adugnaw Mintesnot, Nigussie Dechassa, and Ali Mohammed (2015). Association of Arabica Coffee Quality Attributes with Selected Soil Chemical Properties, East African Journal of Sciences Volume 9 (2) 121-130.

Alonso-Salces, RM, Serra, F, Reniero, F, He´Berger, KR (2009). Botanical and geographical characterization of green coffee (*Coffea arabica* and *Coffea canephora*): Chemometric Evaluation of Phenolic and Methyl xanthine Contents. J. Agric. Food Chem., 57, 4224–4235, DOI:10.1021/jf8037117.

Bertrand, B, Etienne, H, Lashermes, P, Guyot, B, Davrieux, F. (2005). Can near-infrared reflectance of green coffee be used to detect introgression in *Coffea arabica* cultivars? J. Sci. Food Agric. 85: 955-962.

Costa Freitas, AM, Parreira, C, Vilas-Boas, L. (2001). The use of an electronic aroma-sensing device to assess coffee differentiation-comparison with SPME gas chromatography-mass

spectrometry aroma patterns. J. Food Comp. Anal. 14: 513-522.

FAO, 2015. Statistical Pocketbook, World Food and Agriculture, accessed in September (2015).

González-Rios, M, Suarez-Quiroz, ML, Boulanger, R, Barel, M, Guyot, B, Guirard, JP, Schrorr-Galindo, S. (2007). Impact of 'ecological' post-harvest processing on coffee aroma: II. Roasted coffee. J. Food Comp. Anal. 20: 297-307.

Gray, Q, Tefera, A, Tefera, T. (2013). Ethiopia: Coffee Annual Report. GAIN Report No. ET 1302.

ICO. (2015). Coffee Production Data. www.ico.org. Accessed on August 8, 2015: International Coffee Organization.

Joët, T, Andréina, L, Frédéric, D, Sylvie, D, Bertrand, B, Alexandre de, k, Stéphane, D. (2010). Influence of environmental factors, wet processing and their interactions on the biochemical composition of green Arabica coffee beans. Food Chem. 118: 693–701.

Martín, M, Pablos, F, González, A, Valdenebro, M, León-Camacho, M. (2001). Fatty acid profiles as discriminant parameters for coffee varieties differentiation. Talanta, 54: 291-297.

Ministry of Trade (MoT), FDRE. (2012). Coffee opportunities in Ethiopia. Presentation of the Ministry of Trade of the FDRE. February 2012, Addis Ababa.

Osorio, N. (2002). The Global Coffee Crisis: A Threat to Sustainable Development, Submission to the World Summit on Sustainable Development, Johannesburg, 30 August 2002 International Coffee Organization. p 5.

Perrone, D, Donangelo, CM, Farah, A. (2008). Fast simultaneous analysis of caffeine, trigonelline, nicotinic acid and sucrose in coffee by liquid chromatography–mass spectrometry. Food Chem. 110: 1030–1035.

Petit, N. (2007). Ethiopia's Coffee Sector: A Bitter or Better Future?. J. Agrarian Change, 7(2): 225–263.

Rubayiza, AB, Marco, M. (2005). Chemical discrimination of arabica and robusta Coffees by fourier transform raman spectroscopy, J. Agric. Food Chem. 53(12): 4654–4659.

Silvarolla, MB, Mazzefera, P, Alves de Lima, MM. (2000). Caffeine Content of Ethiopian *Coffea arabica* Beans. Genet. Mol. Biol. 23 (1): 213 – 215.

Tesfaye, K. (2006). Genetic diversity of wild Coffea arabica populations in Ethiopia as a contribution for conservation and use planning. Ecology and Development Series, No.44, Centre for Development Research, University of Bonn.

Weckerle, B, Richling, E, Heinrich, S, Schreier, P. (2002). Origin assessment of green coffee (Coffea arabica) by multi-element stable isotope analysis of caffeine. Anal. Bioanal. Chem. 374: 886-890.