

*Full Length Research Paper*

# Effects of processing methods on fatty acid profiles and biochemical compounds of Arabica coffee cultivars

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Coffee cherries were processed traditionally by the wet method that uses large quantities of water and eco-friendly methods that utilizes less water and operate mechanically to remove mucilage. The study is aimed at determining the effects of traditional and newly developed coffee processing methods on fatty acid profiles and biochemical components of two coffee cultivars. A complete randomized design was used for the study. Fresh coffee cherries for two cultivars commonly grown in Kenya, Ruiru 11 and SL 28, were processed using three different processing methods. The methods varied on the mode of mucilage removal and pulping techniques. The parchment obtained from the three processes, wet pulper, hand pulper and eco-pulper methods, were sundried and subjected to chemical analysis. Fatty acids profiles were analyzed by the use of a gas chromatography method and biochemical content; caffeine, trigonelline and chlorogenic acid were determined by HPLC analysis. The processing methods showed significant variations in the fatty acids concentrations but did not significantly affect the levels of biochemical compounds. The concentration of fatty acids ranges from 1.16 to 16.88%, with linoleic acid being dominant. The trigonelline level ranges from 1.24 to 1.36%, caffeine ranges from 1.36 to 1.45% and chlorogenic acid from 5.34 to 5.46% in the samples from the different processing methods.

**Key words:** Processing methods, coffee cultivars, fatty acids, biochemical compounds.

## INTRODUCTION

Coffee is one of the most widely used nonalcoholic drinks and its consumption is spreading globally. It is the second most important commodity exchanged in world markets, next to crude oil (Haile and Kang, 2019a). The coffee bean is obtained from the fruit of the coffee plant, a small evergreen shrub belonging to the genus *Coffea*, family *Rubiaceae*. Kenya produces mainly Arabica coffee (*Coffea arabica* L.) (Kathurima et al., 2012). The old

cultivars grown in Kenya are K7 for low altitude areas prone to leaf rust and the SL28 and SL34 for low to medium altitude areas with good rainfall (Mwangi, 1983). The other cultivars are Ruiru 11 and Batian which are suitable for all coffee growing areas in Kenya because of their resistance to Coffee Berry Disease (CBD) and Coffee Leaf Rust (CLR) (Opile and Agwanda, 1993; Kathurima et al., 2012). After harvesting of the fruits, green

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coffee beans are obtained through processing by use of either the dry or wet methods (Murthy and Naidu, 2011) and semidried (Haile and Kang, 2019a). In the dry method, the whole cherry is dried under the sun or mechanical dryer, followed by mechanical removal of the dried outer parts (Duarte et al., 2010). The wet method requires the use of specific equipment and substantial amounts of water, in which the pulp is eliminated by a pulper, followed by natural fermentation (Gonzalez-Rios et al., 2007). Semidry processing is a combination of both dry and wet methods, in which the coffee fruits are depulped but the fermentation process occurs directly under the sun on a platform (Vilela et al., 2010; Haile and Kang, 2019a). At the end of fermentation, the wet processed seeds are washed and dried (Duarte et al., 2010). There are different categories of coffee pulping methods which vary depending on the type of equipment and the mucilage removal processes. The pulping method may be done as continuous processing operation with the use of a mechanized disc pulping equipment or with the use of manually operated equipment. The mucilage removal could be done through natural fermentation and washing with excess water and the process called fully washed method. The other method involves a mechanical operation where the mucilage is scraped by a specialized unit of the pulping machine called an eco-pulper. The natural fermentation and washing of coffee entails the traditional method of removing mucilage while the mechanized process is a new technology considered to be economical and fast in its processing of coffee berries (Roa et al., 2019).

Variations in the quality of coffee obtained by the use of the different processing methods have been reported in literature (Gonzalez-Rios et al., 2007; Bytof et al., 2005; Haile and Kang, 2019a). However there is scanty information on the effects of different processing methods on the chemical components such as fatty acids and other biochemical compounds. The lipid content in coffee grounds ranges from 10 to 17%. However, compared to *Coffea canephora*, higher lipid contents are found in Arabica coffees (Figueiredo et al., 2015). For most of the lipids, the coffee oil, are located in the endosperm of green coffee beans and a small amount, the coffee wax, is located on the outer layer of the bean. The coffee oil fraction is mainly composed of triacylglycerols, which have fatty acid proportions similar to those found in edible vegetable oils (Speer and Kölling-Speer, 2006). Triacylglycerols are the major carriers of aroma in the roasted bean. Their fatty acid composition determines the generation of thermally-induced oxidation products, in particular aldehydes, which react readily with Maillard intermediates, giving rise to additional aroma compounds. Biochemical compounds in coffee such as chlorogenic acid and caffeine are responsible for bitterness (Joet et al., 2010). Trigonelline is a pyridine derivative known to contribute indirectly to the formation of appreciated flavor products including furans, pyrazine,

alkyl-pyridines and pyrroles during coffee roasting (Ky et al., 2001; Perrone et al., 2008). Therefore, coffee processing methods influence on the levels of these components may affect the quality characteristics of roasted coffee beans. There may be variations also in the levels of chemical components among different coffee cultivars such as Ruiru 11 and SL 28. Hence there is need to determine the levels of fatty acids and biochemical components in this cultivars which are among the major determinants of coffee quality. This research aimed at determining the effects of different processing methods on the concentrations of fatty acids and biochemical components in two coffee cultivars commonly grown in Kenya.

## MATERIALS AND METHODS

### Preparation of coffee samples

The red ripe coffee cherries were harvested and processed by three processing methods classified as wet pulper, hand pulper and eco-pulper methods. Wet pulper method was done by continuous pulping operation. The parchments were fermented in plastic containers by dry fermentation for 18 h. It was then washed and graded and the heavier grade (P1) dried and used for analysis. Hand pulper method was done by use of a motorized manual pulping machine. The parchments were subjected to fermentation in plastic containers with dry method for 18 h. After fermentation the parchment were washed and graded and heavier grade dried and used for analysis. The eco-pulper method was done by use of ecological pulping equipment. The machine removed mucilage and cleaned the parchment with little water without fermentation of the coffee parchment. The coffee parchments from the three methods were dried in the sun to a moisture content of  $10 \pm 1\%$ . The dry parchments were then packed and sealed in polythene bags and stored in a freezer at  $-18^{\circ}\text{C}$  until time for analysis.

### Analysis of fatty acids

Lipids were extracted according to the method of Bligh and Dye (1959). The fatty acids in coffee samples were converted to Fatty Acid Methyl Esters (FAME) according to the method described by Ogara (2013). Fatty acid profile analysis was done using gas chromatograph (Shimadzu GC-9A) fitted with capillary column (15%, Diethylene glycol-succinate) and flame ionization detector temperature of  $220^{\circ}\text{C}$  and injector temperature of  $170^{\circ}\text{C}$ . Nitrogen was used as carrier gas. Fatty acid methyl esters were identified by comparison of retention times of the samples with standards and their concentrations expressed as mg/100 gdw.

### Determination of Biochemical compounds

The analysis of chlorogenic acid, caffeine and trigonelline were done according to the method by Ky et al. (2001) and described by Gichimu et al. (2014). The HPLC equipment (Knauer, Japan) was used with a column (YMC\_Pack polyamine \_\_,  $250 \times 4.6$  mm, I.d.S\_5  $\mu\text{m}$ , 12 nm) and detector (knauer K2600A UV). The mobile phase used was acetonitrile (40%) and formic acid (5%) and the pumps operated in isocratic mode with solvent flow of A (37%) and B (63%). The flow rate was set at 1 ml/min. The peaks were

**Table 1.** Concentrations of fatty acids profiles in green coffee for SL 28 cultivar processed by different pulping methods (mg/100 gdw).

Fatty acids	Wet pulper	Hand pulper	Eco-pulper
Palmitic acid	8.14 + 2.53 <sup>a</sup>	7.29 + 2.12 <sup>b</sup>	10.37 + 2.31 <sup>a</sup>
Stearic acid	4.14 + 1.6 <sup>b</sup>	3.43 + 1.94 <sup>b</sup>	5.06 + 0.57 <sup>a</sup>
Oleic	2.50 + 1.06 <sup>ab</sup>	1.93 + 0.80 <sup>b</sup>	3.79 + 0.71 <sup>a</sup>
Linoleic acid	19.30 + 2.39 <sup>a</sup>	13.82 + 1.40 <sup>b</sup>	19.82 + 3.64 <sup>a</sup>
Linolenic acid	1.76 + 0.37 <sup>b</sup>	1.80 + 0.65 <sup>b</sup>	2.52 + 0.76 <sup>a</sup>

<sup>1</sup>values are means ( $\pm$  SD) of triplicate determinations. <sup>2</sup>means designated by different letters in a row are significantly different at (P <0.05).

**Table 2.** Concentrations of fatty acids profiles in green coffee samples for Ruiru 11 cultivar processed by different pulping methods (mg/100 gdw).

Fatty acids	Wet pulper	Hand pulper	Eco-pulper
Palmitic acid	7.82 + 1.54 <sup>a</sup>	7.07 + 0.78 <sup>a</sup>	7.25 + 1.68 <sup>a</sup>
Stearic acid	2.44 + 1.35 <sup>a</sup>	2.16 + 0.72 <sup>a</sup>	2.73 + 0.48 <sup>a</sup>
Oleic	1.59 + 0.47 <sup>a</sup>	1.69 + 0.27 <sup>a</sup>	1.79 + 0.27 <sup>a</sup>
Linoleic acid	16.87 + 4.29 <sup>a</sup>	15.67 + 2.25 <sup>a</sup>	16.88 + 3.20 <sup>a</sup>
Linolenic acid	1.27 + 0.34 <sup>a</sup>	1.07 + 0.28 <sup>a</sup>	1.27 + 0.38 <sup>a</sup>

<sup>1</sup>values are means ( $\pm$  SD) of triplicate determinations. <sup>2</sup>means designated by different letters in a row are significantly different at (P <0.05).

detected at 324 nm for chlorogenic acid, and 263 nm wavelengths for caffeine and trigonelline. The elution time was 5 min and 6 min for trigonelline and caffeine respectively. Identification was achieved by comparison of retention times (Rt) of samples with those of the standards.

### Statistical analysis

All treatments were done in triplicates and analysis of data evaluated using the Statistical Package for Social Scientist (SPSS version 18). Analysis of variance (ANOVA) was conducted, and the differences between group means analyzed using the Least Significant Difference (LSD). Statistical significance was established at  $p \leq 0.05$ .

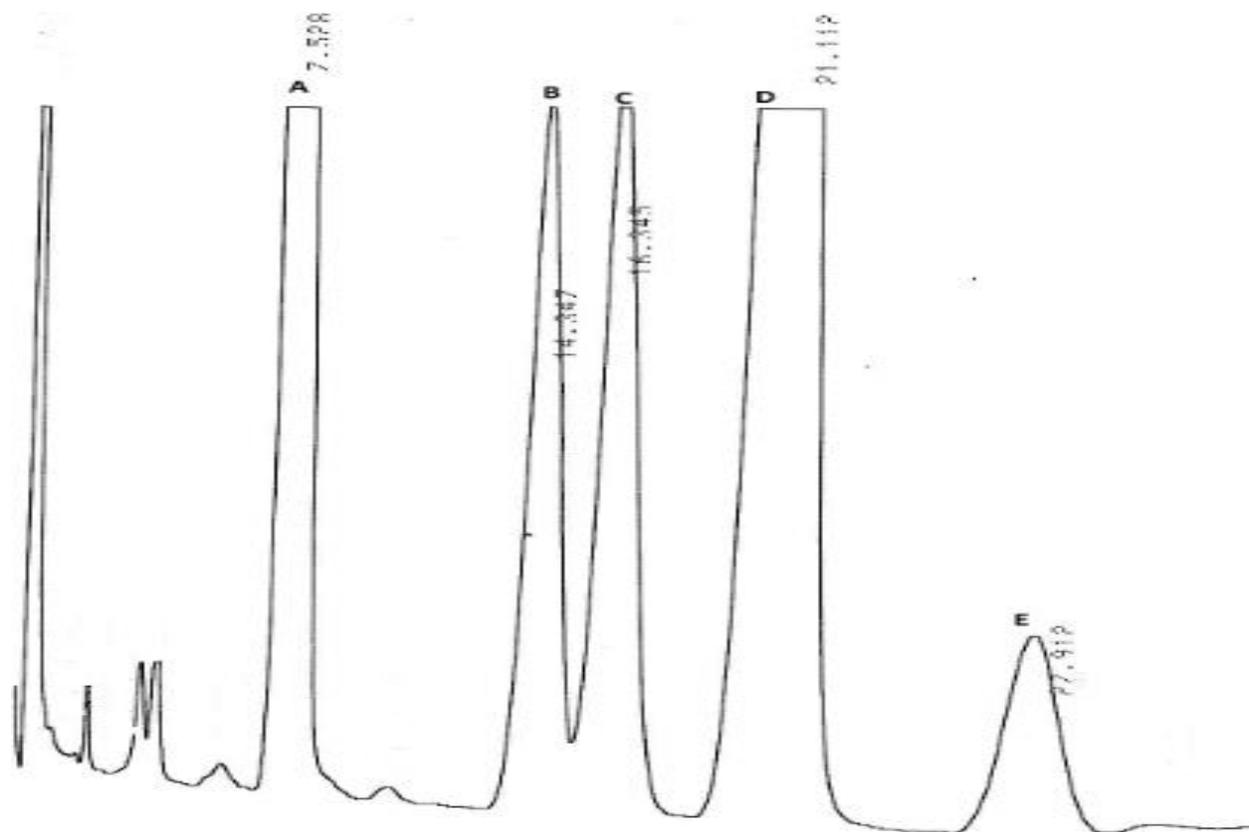
## RESULTS AND DISCUSSION

### Effects of processing methods on the fatty acid concentrations of green coffee

The green coffee beans samples for two main cultivars namely SL 28 and Ruiru 11 were used in the analysis. The results for the fatty acid concentrations are presented on Tables 1 and 2. A sample chromatograph for fatty acids profiles is shown on Figure 1. The samples were processed using three different pulping methods. The fatty acids detected include: palmitic, stearic acid, oleic acid, linoleic acid and linolenic acid. Linoleic acid showed the highest concentration with a range of 13-19

mg/100 g, followed by palmitic acid (7-10 mg/100 g), stearic acid (2-5 mg/100 g), oleic acid (2-4 mg/100 g) and linolenic acid (1-4 mg/100 g) for green coffee samples analysed. Similar trends in the concentration of fatty acids have also been reported by other authors (Martin et al., 2001; Figueiredo et al., 2015; Hung et al., 2018). The coffee samples were processed by three pulping methods named as wet pulper, hand pulper and eco-pulper methods. The pulping methods varied in terms of whether the method involved fermentation process or not and the level of water used during processing. The results indicate that there were significant differences ( $p < 0.05$ ) between the processing methods on the concentrations of some fatty acids content especially for the SL 28 samples. For Ruiru 11 samples, there were no significant variations between the processing methods on the levels of the fatty acids contents.

The hand pulper method showed slightly lower significant levels for some fatty acids content such as palmitic acid, stearic acid, oleic acid and linoleic acid. This variation in the levels of fatty acids could be attributed to the differences in the processing conditions. The eco pulper method which operates without fermentation showed slightly higher trends for the fatty acids contents compared to the wet and hand pulper methods which use fermentation and excess water during processing. Haile and Kang (2019a) indicated reduction of lipids content after fermentation of mucilage. The low levels of fatty acids in the wet and hand pulper methods



**Figure 1.** Chromatograph for the fatty acids profiles in green coffee samples: A-Palmitic acid, B-stearic acid, C-Oleic acid, D-Linoleic acid, E-Linolenic acid.

could be attributed to loss of materials from the coffee due to fermentation and washing processes. Joet et al., (2010) reported occurrence of metabolic processes during wet processing of coffee affecting their chemical composition. In this study it is suggested that the methods with fermentation could expose the coffee beans to microbial and enzymatic activities which may influence the degradation of chemical components. The chemical components in the coffee beans may then be reduced or loss due to processing. Variations in the levels of chemical composition due to the influence of metabolic activities in coffee beans have also been reported by other authors such as Selmar et al. (2006) and Patui et al. (2014). The lipase activity has been reported to be present in the coffee seed which can catalyze the hydrolysis of ester bonds in monoacylglycerol, diacylglycerol and triacylglycerols into free fatty acids and glycerol (Toci et al., 2013; Patui et al., 2014). It is reported that majority of lipids are found in the oil fraction of the coffee bean endosperm and a small amount, the coffee wax, is located on the outer layer of the bean. Hence those on the outer layers may be affected by processing or metabolic activities. The coffee oil fraction is mainly composed of triacylglycerols, which

have fatty acid proportions similar to those found in edible vegetable oils (Speer and Kölling-Speer, 2006; Figueiredo et al., 2015).

#### **Effects of processing methods on biochemical content of coffee.**

Caffeine, trigonelline and chlorogenic acid are the common biochemical components of coffee. Table 3 shows the results of the effects of processing methods on these components in coffee samples processed by the three processing methods. Trigonelline in the SL 28 samples range from 1.24 to 1.29% while in Ruiru 11 the range was between 1.28 to 1.36%. Processing methods did not show any significant differences ( $p < 0.05$ ) on the levels of trigonelline for the coffee samples analysed. The levels for caffeine in SL 28 ranged from 1.26 to 1.36% and 1.29 to 1.45% in the Ruiru 11 samples. The processing methods did not show significant variation on the caffeine content for both the SL 28 and Ruiru 11 samples. The chlorogenic acid content in the SL 28 samples ranged from 5.34 to 5.44% and 5.36 to 5.46% for Ruiru 11 samples. The level of trigonelline, caffeine

**Table 3.** Biochemical contents for SL 28 and Ruiru 11 samples processed by three different processing methods.

	Cultivar	Wet pulper	Hand pulper	Eco-pulper
Trigonelline %	SL 28	1.26 + 0.10 <sup>a</sup>	1.24 + 0.06 <sup>a</sup>	1.29 + 0.10 <sup>a</sup>
	Ruiru 11	1.28 + 0.10 <sup>a</sup>	1.36 + 0.12 <sup>a</sup>	1.36 + 0.19 <sup>a</sup>
Caffeine %	SL28	1.36 + 0.05 <sup>a</sup>	1.35 + 0.02 <sup>a</sup>	1.29 + 0.04 <sup>a</sup>
	Ruiru 11	1.30 + 0.09 <sup>a</sup>	1.45 + 0.18 <sup>a</sup>	1.34 + 0.13 <sup>a</sup>
Chlorogenic acid %	SL28	5.34 + 0.14 <sup>a</sup>	5.34 + 0.14 <sup>a</sup>	5.44 + 0.28 <sup>a</sup>
	Ruiru 11	5.31 + 0.23 <sup>a</sup>	5.34 + 0.07 <sup>a</sup>	5.46 + 0.09 <sup>a</sup>

<sup>1</sup>values are means ( $\pm$  SD) of triplicate determinations. <sup>2</sup>means designated by different letters in a row are significantly different at (P <0.05).

and chlorogenic acid in the coffee samples were within the levels reported by other authors (Mussatto et al., 2011; Gichimu et al., 2014). Though the compounds such as trigonelline and chlorogenic acid are reported as water soluble components which could be lost by squeezing of coffee (Nigam and Singh, 2014), the wet pulper and hand pulper methods which use fermentation did not show any significant reduction for the tested compounds. This could be because the compound are strongly bound within the endosperm of coffee beans and cannot easily be lost through washing and squeezing of coffee parchment during mucilage removal. Similarly the compounds could not be affected by fermentation process.

However Haile and Kang (2019b) reported an increase of total polyphenol content of green coffee beans after fermentation with different strains of yeast. Other authors reported no changes on the level of these compounds due to different processing methods. Duarte et al. (2010) did not find any significant variations on the level of caffeine between wet processing and semi dry processing methods. Ferreira et al. (2013) studied the effects of wet and dry processing of coffee on chemical composition and did not find any significant variations on the levels of caffeine. However, Nigam and Singh (2014) reported loss of chlorogenic acid from coffee during wet processing due to the effect of leaching into the processing water. The eco-pulper using less water compared to the wet and hand pulper methods did not show significant variations between these methods on the level of chlorogenic acid. The level of chlorogenic acid was within the levels reported in the literature. Farah et al. (2006) reported a range of 4 to 14% in green coffee beans. The stability of these biochemical compounds during processing of coffee is important for determination of the quality of coffee beverage. These compounds are important in influencing the aroma and flavor of coffee. The thermal degradation of chlorogenic acids during roasting results in formation of phenolic substances that contribute to bitterness and aromatic compounds which are undesirable to cup quality (Toci and Farah, 2008). Trigonelline is a pyridine derivative known to contribute

indirectly to the formation of appreciated flavor products including furans, pyrazine, alkyl-pyridines and pyrroles during coffee roasting (Ky et al., 2001). Caffeine with its characteristic bitter taste is an important determinant of coffee flavor (Farah et al., 2006).

## Conclusion

From the study, it can be deduced that the processing methods showed variations on the level of some fatty acid components of coffee. SL 28 samples showed more significant variations than the Ruiru 11 samples. The methods with fermentation process displayed less fatty acid levels than the method without fermentation process. The processing methods that use excess water and fermentation process did not vary from the methods with little water and no fermentation in regard to the levels of biochemical compounds. SL 28 cultivar showed slightly higher concentration of fatty acids than the Ruiru 11 cultivar. There was no significant variation in the level of biochemical compounds for both cultivars when processed by different methods.

## CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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