EFFECTS OF ROASTING DEGREES ON TOTAL PHENOLIC, ANTIOXIDANT ACTIVITY AND PHENOLIC COMPOUNDS IN LOCAL LIBERICA COFFEE BEANS

Aida Hamimi Ibrahim
Food Science and Technology Research Centre,
MARDI Headquarters, Selangor, Malaysia
E-mail: aida@mardi.gov.my

Nur Diyana Alyas
Food Science and Technology Research Centre,
MARDI Headquarters, Selangor, Malaysia

Koh Soo Peng
Food Science and Technology Research Centre,
MARDI Headquarters, Selangor, Malaysia

Norizah Mat Ayob
Food Science and Technology Research Centre,
MARDI Headquarters, Selangor, Malaysia

Muhammad Juma'at Jonid
Food Science and Technology Research Centre,
MARDI Headquarters, Selangor, Malaysia

ABSTRACT

Coffee is a popular roasted beverage with health benefits since it contains antioxidants called phenolics. The effect of roasting on antioxidant activity is still up for debate, but the phenolic content of roasted coffee beans can provide important clues about the health advantages of these compounds. Local Liberica coffee beans were roasted at three different temperatures (200°C, 220°C, and 240°C) and their pH value, total soluble solid content, total phenolic content, antioxidant activity, and phenolic compounds were measured. The coffee extracts showed total soluble solids of 3.23 ± 0.10 °Brix and 5.18 ± 0.01 °Brix, with a pH range of 5.00-5.60. The extract total phenolic content varied from 16.00 ± 0.29 to 28.35 ± 0.38 mg gallic acid equivalents per gramme dry weight of coffee. Antioxidant activity as determined by the FRAP value. The antioxidant activity as determined by the DPPH (radical scavenging activity) and FRAP value (ferric reducing of antioxidant power) declined as the roasting temperature increased, going from 73.85 ± 1.99% to 51.82 ± 3.63% and 97.78 ± 6.31 to 49.28 ± 3.41 mg AAE per gramme of dry coffee, respectively. Chlorogenic acid, caffeic acid, and trigonelline concentration all dramatically decreased ($p<0.05$) as roasting intensity rose. However, unlike chlorogenic acid, caffeine concentrations do not react to roasting temperatures in the same way. Even when the level of roasting is raised, the percentage of caffeine remains unchanged. Thus, roasting may increase the production of caffeine, offsetting the reduction in antioxidant action caused on by the breakdown of chlorogenic acid.

Keywords: Liberica coffee, antioxidant activity, total phenolic, roasting

INTRODUCTION

Liberica coffee beans are a type of coffee bean that comes from the Coffea liberica plant. Coffee from the Coffea liberica species is less common compared to Arabica and Robusta varieties, but it has its own distinct characteristics. Liberica coffee beans taste more like robusta coffee than arabica coffee. Robusta coffee is earthy, pungent, and spicy, whereas arabica coffee has caramel and sweet flavours (Bolka and Emire, 2020). In contrast, Liberica coffee beans are larger than those of the other two varieties and smell like jackfruits. The coffee beans' flavour, colour, and aroma will all change after roasting (Bauer et. a., 2018). The roasting process has a significant impact on the antioxidant activities of coffee beans. While raw or green coffee beans contain certain antioxidants, the roasting process transforms the chemical composition of the beans, leading to the development of new compounds and altering the levels of existing ones. Like other coffee varieties, Liberica coffee contains antioxidants, which are compounds that help neutralize harmful free radicals in the body. While the specific antioxidant content can vary based on factors such as growing conditions and processing methods, coffee, in general, is known to be a rich source of antioxidants. The objective of this study was to examine the antioxidant activity total phenolic and phenolic compunds of three type of roasting temperature of Liberica coffee beans such as light -roasted (200 C) medium-roasted (220 C) and dark-roasted (240 C).

MATERIALS AND METHODS

Raw materials collection

Local Liberica green coffee beans are purchased from MyLiberica Sdn. Bhd., Kluang, Johor. Green coffee beans processed using the honey method were selected in this study because this process is often used by local coffee bean producers.

Coffee Roasting Method
Green coffee beans are roasted at three different roasting temperatures, namely at 200 C (light-roasted), 220 C (medium-roasted) and 240 C (dark-roasted). The roasting process is carried out using a 5 kg scale coffee roaster. The roasted coffee beans are left at room temperature for 24 hours and then ground and stored in an airtight container until the analysis is carried out.

Coffee Brewing Method using French Press
The coffee brewing method was slightly modified based on the study conducted by Janda et al., (2020). Approximately 10 grams of finely ground coffee powder was inserted into the press pot positioned in level surface followed by the addition of 180 ml of boiling water. The water was gently poured into the pot. Subsequently, the plunger was placed into the container on top of the liquid and subsequently released after five minutes. The coffee extract was kept for further analysis once the press plunger was depressed.

Determination of pH value
The final pH value of the coffee extract was measured using a single channel benchtop pH meter (Mettler Toledo, USA).

Determination of total soluble solid
The Brix concentration, which quantifies the total soluble solids was determined by measuring the light refraction through a coffee extract using a refractometer (Atago Co. LTD., Japan).

Determination of Total Phenolic Contents
The total phenolic content was determined using Olechno et al. (2022) with minor adjustment. The coffee extract was mixed with 5 ml Folin-Ciocalteu reagent (Merck, USA) and incubated at room temperature for 5 minutes. Subsequently, about 4 ml of 7.5% of sodium carbonate solution were added before incubated in the dark place for two hours. The absorbent was measured at 765 nm against the blank using UV-Vis spectrophotometer (VARIAN Cary 50, USA). The amount of total phenolic content was expressed as Gallic acid equivalents (mg GAE/g extract) from a calibration curve.

Determination of antioxidant activity

Determination of Ferric Reducing of Antioxidant Power (FRAP)
The aliquot extract 150 ul were mixed with 2850 ul of freshly prepared FRAP working solution. The mixture was incubated in the absence of light for 30 minutes. The FRAP values were measured by comparing the absorbance change of blue coloured ferrous-tripyridyltriazine complex at 593 nm using UV-Vis spectrophotometer (VARIAN Cary 50, USA). The results were determined from the ascorbic acid calibration curve.

Determination of 1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity
The extract of 150 ul was mixed with freshly prepared 2850 ul of 1,1-diphenyl-2-picryl-2hydrazil (DPPH) methanolic solution. The mixture was incubated in dark condition for a duration of 30 minutes at room temperature. The absorbance was read at 515 nm using UV spectrophotometer (VARIAN Cary 50, USA) (Anosike, et al., 2015). The DPPH radical scavenging activity was calculated using the following equation:

$$\text{Percentage of scavenging activity (\%)} = \frac{\text{Abs}_{(\text{Control})} - \text{Abs}_{(\text{Sample})}}{\text{Abs}_{(\text{Control})}} \times 100$$

Determination of phenolic compounds using ultra performance liquid chromatography
The phenolic compounds of coffee samples were separated by injection of 1 µL sample and run through gradient solvent profile into Kinetex C18 100 A column (150 mm × 2.1 mm; 1.7 µm) using Ultra Performance Liquid Chromatography (UPLC) system (Waters, USA) with the oven temperature controlled at 40°C and the flow rate fixed at 0.35 mL/min. The absorbance was read at different UV spectrum of 270 and, 330 nm for particular phenolic compounds determination. The gradient elution consists of mobile phase A (water: acetic acid, 97:3) and mobile phase B (methanol). The gradient elution mode was programmed as follow: from 0 to 0.6 min, 100% A; from 0.6 to 12 min, linear gradients from 100 to 40% A; from 12 to 16 min, linear gradient from 40 to 100% A and remain 2 min at 100%A.. Quantification was performed using calibration curves obtained by injecting known amounts of targeted phenolic acids as external standards with known retention time under a specific UV spectrum.

Statistical Analysis
The experimental data was evaluated using Minitab 18 (Minitab, LLC) to compare the response variables of coffee extracts using different roasting temperatures. The mean values and standard deviations (n=3) were examined using one-way analysis of variance (ANOVA). Turkey's test was used for mean comparisons at a significance level of $p<0.05$ in this investigation. The measurements were determined in three replicates.

RESULTS AND DISCUSSION

This study involved roasting experiments at three distinct temperatures (200°C, 220°C, and 240°C) on *C. liberica* beans. Subsequently, the pH value, total soluble solid, total phenolic content, antioxidant activities (FRAP and DPPH radical scavenging activity) and phenolic compounds quantification were analysed. Table 1 presents the outcomes pertaining to the pH value and total soluble solid content derived from three different roasting temperatures of *C. liberica* extracts. The pH values of the studied coffee extracts ranged from 5.00 to 5.60 which contradicts the trend of total soluble solid. It was observed that the coffee beans

roasted at a lower temperature of 200°C exhibited higher acidity with total soluble solid at 5.18 ± 0.01 and 3.23 ± 0.10 °Brix, whereas those roasted at higher temperatures 240°C showed lower acidity and total soluble solid at 5.60 ± 0.01 and 2.00 ± 0.18 °Brix, respectively. The determination of pH value of the sample involved the assessment of hydrogen-ions activity in water-based coffee extract. This measurement quantify the amount of acid molecules that have undergone deprotonation in the extract. The process of roasting can lead to a decrease in the soluble protonated acidic compounds and soluble solids concentration caused by the high temperatures associated with roasting, which in turn leads to the destruction of the cellular matrix (Ginz et al., 2000). Nevertheless, according to study reported by Rao et al. (2020), the process of pyrolysis also leads to the formation of other compounds, which subsequently increases the feasibility of extracting these compounds in the following analyses.

Total phenolic content showed a significant decrement with increasing roasting temperature, where the highest total phenolic content of the coffee extract was at 200°C roasting temperature at 28.35 ± 0.38 mg GAE/g followed by 220°C and 240°C at 18.04 ± 0.32 and 16.00 ± 0.29 mg GAE/g at $p<0.05$ (Figure 1). This findings are in accordance with study done by Mubarak et al. (2019) and Odzakovic et al. (2016) on different roasting temperature of coffee as the total phenolic content was substantially decrease as the roasting degree progressed from light to dark. According to Wang et al. (2011), the condition of high temperature and low water activity during the roasting process created favourable conditions for the initiation of Maillard reaction. This reaction include several phenolic compounds to formed part of the melanoidins. This could be due to the depletion of the compounds that a more vulnerable resulting in a relative increment of the remaining ones. Melanoidins produced during roasting can react with the Folin-Ciocalteu's reagent (Lopez-Galilea et al., 2007). This explains the increment in the total phenolic content of medium roasted process compared to severe roasting degree.

The study revealed the results of antioxidant activities on the FRAP value and DPPH radical scavenging activity presented in Figure 2 and Figure 3. The antioxidant activity of both determinations displayed a consistent pattern similar to total phenolic content. The findings indicate an increase in the degree of roasting led to a decrease in the antioxidant activity of FRAP value and DPPH radical scavenging activity from 97.78 ± 6.31 to 49.28 ± 3.41 mg AAE per gram of dry coffee and from 73.85 ± 1.99% to 51.82 ± 3.63%, respectively. The antioxidant capacity of coffee is attributed to the presence of polyphenolic chemicals and the process of roasting can influence its antioxidant benefits as a result of the elevated temperatures involved. The findings of our study align with other research, which has demonstrated a decrease in the antioxidant capacity of medium dark roasted to heavy roasted *C. liberica* (Mubarak et al., 2019).

This study involved the quantification of four phenolic compounds, namely chlorogenic acid, caffeic acid, trigonelline, and caffeine (Figure 4). These phenolic compounds of coffee extract are known to have antioxidant effects. The amount of chlorogenic acid (8.33 ± 0.10 to 1.32 ± 0.02 mg/g), caffeic acid (10.52 ± 0.12 to 1.58 ± 0.02 mg/g) and trigonelline (1.94 ± 0.05 to 0.65 ± 0.01 mg/g) concentrations shown to be declining, as the roasting temperatures increased. The drastic decline in chlorogenic acid content throughout the roasting process can be attributed to the phenomenon of polyphenolic compounds being assimilated into melanoidins. Furthermore, the process of roasting might lead to the conversion of chlorogenic acid into volatile molecules, quinic acids, and chlorogenic acid lactones. (Dawidowicz and Typek, 2017). Conversely, caffeine remained consistence at 12.134 to 12.306 mg/g as the roasting degree changes as it is thermostable and shown no significant impact as has been previously recorded by Jung et al. (2017). Hence, it is imperative to employ a suitable roasting technique in order to preserve the optimal beneficial properties of coffee, as varying levels of roasting have the potential to diminish the phenolic concentrations during the roasting process.

Table 1. Effect of final pH and total soluble solid with different *Coffea liberica* roasting temperatures

	Extract	pH value	Brix value (°Brix)
Roasting temperature	**200 °c**	5.18 ± 0.01^{c}	3.23 ± 0.10^{a}
	220 °c	5.55 ± 0.02^{b}	1.90 ± 0.00^{b}
	240 °c	5.60 ± 0.01^{a}	2.00 ± 0.18^{b}

*The data were shown as the mean ± standard deviation of triplicate studies, demonstrating substantial variations among various extracts ($p<0.05$).

Figure 1: Effect of different Coffea liberica roasting temperatures on total phenolic content

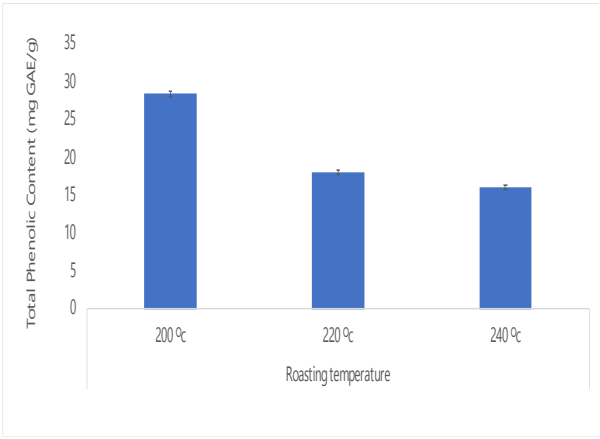


Figure 2: Effect of different *Coffea liberica* temperature on FRAP activity

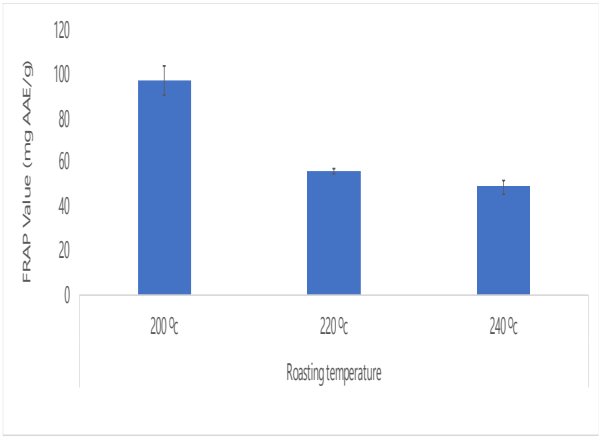


Figure 3: Effect of different *Coffea liberica* roasting temperatures on DPPH free radical scavenging activity

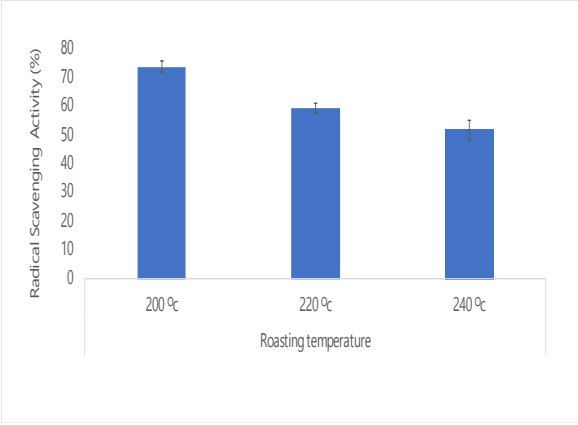
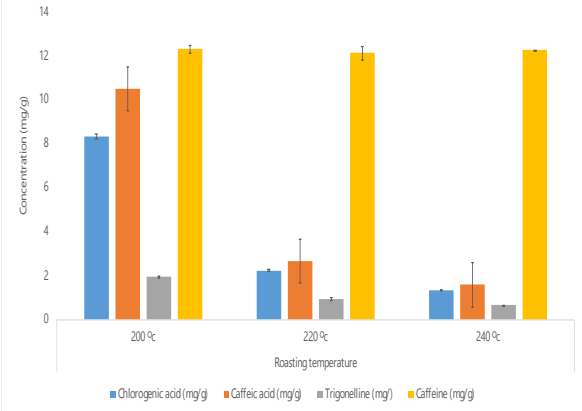


Figure 4: Effect of different Coffea liberica roasting temperatures on phenolic compounds



CONCLUSION

Coffee is a well-liked roasted beverage that offers health advantages due to phenolic antioxidants. Roasting's impact on antioxidant activity is still debatable, although changes in coffee beans' phenolic content brought on by roasting may offer significant indications regarding the health benefits of these substances. Hence, it is imperative to employ a suitable roasting technique in order to preserve the optimal beneficial properties of coffee, as varying levels of roasting have the potential to diminish the phenolic concentrations during the roasting process.

REFERENCES

1. Anosike, C. A., Ogbodo, N. E., Ezugwu, A. I., Uroko, R. I., Ani, C. C. and Abonyi, O. (2015). DPPH (1,1-diphenyl-2-picryl-2hydrazil) radical scavenging activity of some ethnomedicinal plants in Nigeria. American-Eurasian of Toxicological Sciences, 7(2):104-109.
2. Bolka, M.; Emire, S. Effects of coffee roasting technologies on cup quality and bioactive compounds of specialty coffee beans. Food Sci Nutr 2020, 8, 1-11.
3. Bauer, D.; Abreu, J.; Jordao, N.; Rosa, J.S.; Freitas-Silva, O; Teodoro, A. Effect of roasting levels and drying process of Coffea canephora on quality of bioactive compounds and cytotoxicity. Int J Mol Sci 2018, 19, 1-19, https://doi.org/10.3390/ijms19113407.
4. Dawidowicz, A. L. and Typek, R. (2017). Transformation of chlorogenic acids during the coffee beans roasting process. European Food Research and Technology. 243: 379-390.
5. Ginz, M., Balzer, H. H., Bradbury, A. G. W., and Maier, H. G. (2000). Formation of aliphatic acids by carbohydrate degradation during roasting of coffee. European Food Reseach and Technology. 211:404-410.
6. Janda, K., Jakubczyk, K., Baranowska-Bosiacka, I., Kapczuk, P., Kochman, J., Rebaccz-Maron, E. and Gutowska, I. (2020). Mineral composition and antioxidant potential of coffee beverages depending on the brewing method. Foods, 9, 121.
7. Jung, S., Kim, M. H., Park, J. H., Jeong, Y. and Ko, K. S. (2017). Cellular antioxidant and anti-inflammatory effects of coffee extracts with different roasting levels. Journal of Medicinal Food. 20(6): 1-10.
8. Lopez-Galilea, I., Paz de Pena, M. and Cid, C. (2007). Correlation of selected constituents with the total antioxidant capacity of coffee beverages: Influence of the brewing procedure. Journal of Agricultural and Food Chemistry. 55(15):6110-6117.
9. Mubarak, A., Croft, K. D., Bondonno, C. B. and Din, N. S. (2019). Comparison of liberica and arabica coffee: Chlorogenic acid, caffeine, total phenolic and DPPH radical scavenging activity. Asian Journal of Agriculture and Biology. 7(1): 130-136.
10. Odzakovic, B., Dzinic, N., Kurkric, Z. and Grujic, S. (2016). Effecte of roasting degree on the antioxidant activity of different arabica coffee quality classes. Acta Scientiarum Polonorum Technologia Alimentaria. 15(4): 409-417.
11. Olechno, E., Puscion-Jakubik, A., Markiewicz-Zukowska, R. and Socha, K. (2022). Impacts of brewing methods on total phenolic content (TPC) in various types of coffee. Molecules, 25(22):5274.
12. Rao, N. Z., Fuller, M. and Grim, M. D. (2020). Physiochemical characteristics of hot and cold brew coffee chemistry: The effects of roast level and brewing temperature on compound extraction. Foods. 9(7): 902.
13. Wang, H. Y. and Yao, W.R. (2011). Melanoidins produced by the Maillard reaction: Structure and biological activity. Food Chemistry. 128(3):573-584.